SONOMA STATE UNIVERSITY

49th Annual West Coast

BIOLOGICAL SCIENCES UNDERGRADUATE

Research Conference

Saturday, April 11, 2026
at
Sonoma State University
Rohnert Park, California

The Department of Biology of SSU WELCOMES YOU!

Photo credit: Brennan Sparks

Front row: Dr. Shannon Lee, Dr. Joseph Lin, Dr. Brent Hughes, Dr. Mackenzie Zippay, Dr. Richard Whitkus, Elisabeth Kettmann, Dr. Lisa Bentley

Back row: Dr. Sean Place (Chair), Dr. Lisa Hua, Dr. Dan Crocker, Dr. Derek Girman, Dr. Nick Geist, Dr. Nathan Rank, Stephanie Thibault, Rosemary Galten

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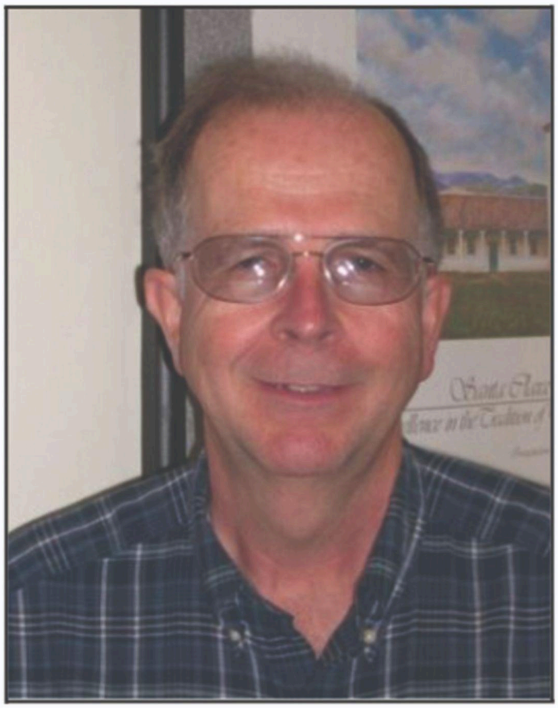
WCBSUR Conference History



Dr. Bill Eisinger

The West Coast Biological Sciences Undergraduate Research Conference (WCBSURC) is the oldest, continuously ongoing, intercollegiate Conference of its kind in the nation. The purposes of the Conference are: 1) to provide a forum for undergraduate researchers to present original data they have generated in the fields of biology and biochemistry; and 2) to foster intercollegiate interactions among students and faculty who share a commitment to undergraduate research in the biological sciences. The WCBSUR Conference was founded in 1976 by Dr. William Eisinger, Professor of Biology at Santa Clara University and two other faculty. It was hosted by Santa Clara every year until 1986, when other institutions began sharing the responsibility, among them Colorado College, Loyola Marymount University, Occidental College, Pacific Lutheran University, Point Loma Nazarene University, the University of California at Irvine and the University of San Francisco.

Participating Institutions (1976 through 2026)

Alabama: University of Alabama at Birmingham; **Alaska**: University of Alaska – Fairbanks, University of Alaska – Juneau; **Arkansas**: University of Arkansas - Fayetteville; **Arizona**: Arizona State University, Northern Arizona University, University of Arizona; **British Columbia (Canada)**: Kwantlen Polytechnic University, University of the Fraser Valley; **California**: Allan Hancock College, American River College, Antelope Valley College, Azusa Pacific University, Biola University, California Baptist University, California Institute of Technology, California Lutheran University, California Polytechnic Institute at Pomona, California Polytechnic Institute at- San Luis Obispo, Chapman University, Charles Drew Medical School, Chico State University, Claremont McKenna College, College of the Canyons, Cosumnes River College, CSU-Bakersfield, CSU-Channel Islands, CSU-Dominguez Hills, CSU -East Bay, CSU–Fresno, CSU–Fullerton, CSU-Hayward, CSU–Humboldt, CSU–Long Beach, CSU-Los Angeles, CSU-Monterey Bay, CSU-Northridge, CSU-Sacramento, CSU-San Bernardino, CSU-San Marcos, CSU-Sonoma, CSU -Stanislaus, Diablo Valley College, Dominican University, Fresno Pacific University, Glendale Community College, Golden West College, Harvey Mudd College, Irvine Valley College, Jet Propulsion Laboratory, LA Mission College, La Sierra University, Loma Linda University, Loyola Marymount University, Mesa College, Mills College, Miracosta College, Miramar College, Mount St. Mary's College, National University, Occidental College, Oxnard College, Pacific University, Palomar College, Pasadena City College, Pepperdine University, Pitzer College, Point Loma Nazarene University, Pomona College, Saint Mary's College, San Diego City College, Saddleback College, San Diego State University, San Francisco State University, San Jose State University, Santa Clara University, Scripps College, Skyline College, Soka University of America, Southwestern College, Stanford University, St. Mary's College of CA, The Master's University, The Scripps Research Institute, UC Berkeley, UC Davis, UC Irvine, UCLA, UC Merced, UC Riverside, UC San Diego, UC San Francisco, UC Santa Barbara, UC Santa Cruz, University of La Verne, University of the Pacific, University of Redlands, University of San Diego, University of San Francisco, University of Southern California, Vanguard University, Victor Valley College, West Los Angeles College, Westmont College, Whittier College; **Colorado**: Adams State University, Colorado College, Fort Lewis College, Regis University, University of Colorado at Boulder, University of Colorado at Denver, University of Northern Colorado, US Air Force Academy; **Connecticut**: Quinnipiac University, University of Connecticut; **Georgia**: Georgia Institute of Technology, University of Georgia; **Hawaii**: BYU Hawaii, Chaminade University, University of Hawaii - Manoa; **Idaho**: College of Idaho, Idaho State University, Northwest Nazarene University, University of Idaho; **Illinois**: Loyola University of Chicago, Northwestern University, Roosevelt University, University of Chicago, Wheaton College; **Indiana**: Earlham College, University of Notre Dame; **Iowa**: Briar Cliff University; **Kansas**: Wichita State University; **Kentucky**: Berea College, Centre College, University of the Cumberlands; **Louisiana**: Louisiana State University, Tulane University, University of Louisiana at Monroe; **Maryland**: Morgan State University; **Massachusetts**: Massachusetts Institute of Technology; **Minnesota**: University of Minnesota-Morris; **Missouri**: Truman State University, Washington University-St. Louis; **Montana**: Montana State University, University of Montana; **Nebraska**: Creighton University, Hastings College, Nebraska Wesleyan University; **Nevada**: Nevada State University, University of Nevada at Reno, UNLV; **New Mexico**: University of New Mexico; **New York**: CUNY Hunter College, New York University, Pace University, Skidmore College; **North Carolina**: Duke University, Elon University; **Ohio**: Case Western Reserve University, Hiram College; **Oklahoma**: East Central University, Oklahoma City University, Oklahoma State University, Oral Roberts University, Southern Nazarene University, University of Oklahoma; **Oregon**: Linfield College, Oregon State University, Portland State University, Reed College, University of Oregon, University of Portland, Willamette University; **Pennsylvania**: Drexel University, Penn State University, Villanova University, Westminster College; **Rhode Island**: Brown University, Rhode Island College; **South Carolina**: Clemson University; **South Dakota**: University of South Dakota; **Tennessee**: Vanderbilt University; **Texas**: St. Edwards' University, St. Mary's University, Texas A&M University-Corpus Christi, Trinity University, University of Texas Austin, University of Texas-Rio Grande Valley; **Utah**: Brigham Young University, Dixie State University, Southern Utah University, University of Utah, Weber State University; **Vermont**: Middlebury College; **Virginia:** University of Virginia; **Washington**: Eastern Washington University, Evergreen State College, Gonzaga University, Pacific Lutheran University, Peninsula College, Seattle Pacific University, Seattle University, University of Puget Sound, University of Washington, Washington State University, Western Washington University, Whitman College, Whitworth College; **Wisconsin**: Edgewood College, Northland College, University of Wisconsin Madison; **Wyoming**: University of Wyoming.

Directions

Parking lot (Parking pass will be included)

From San Francisco (South): Take US-101 North toward Santa Rosa. Exit at Rohnert Park Expressway, turn right, and follow to its end at Petaluma Hill Road. Turn right on Petaluma Hill Road to the stoplight at East Cotati Avenue. Turn right on East Cotati Avenue to the Main Entrance of the campus on your right. The Parking and Information Booth is at the end of the driveway.

From Santa Rosa (North): Take US-101 South. Exit at Rohnert Park Expressway, turn left, and follow to its end at Petaluma Hill Road. Turn right on Petaluma Hill Road to the stoplight at East Cotati Avenue. Turn right on East Cotati Avenue to the Main Entrance of the campus on your right. The Parking and Information Booth is at the end of the driveway.

From Sacramento/East Bay: Take I-80 West to Highway 37 West. Merge onto US-101 North, then exit at Rohnert Park Expressway, turn right, and follow to its end at Petaluma Hill Road. Turn right on Petaluma Hill Road to the stoplight at East Cotati Avenue. Turn right on East Cotati Avenue to the Main Entrance of the campus on your right. The Parking and Information Booth is at the end of the driveway.

Campus Map

Click on the map to open our interactive campus map! (Opens new page)

If you're driving to the conference, please navigate to:
Sonoma State University
1801 East Cotati Avenue
Rohnert Park, CA 94928

Complimentary parking is available for all WCBSURC attendees in Parking Lots D and J—we've covered the cost for you. Look for posted signs on campus with directions to these lots

.

***Parking Attendants: Parking Attendants will hand out printed permits at Lot D.* *Once Lot D is full, Parking Attendants will hand out printed parking permits at Lot J.**

Once parked, follow the signs from the parking areas to the Student Ballroom, located inside the Student Center.

We'll have plenty of signage to guide you along the way!

- Registration will be open at 7:30 AM in the **University Ballroom** to pick up your nametag, tote bag, hang your poster, and check out the room for your talk.
- The Welcome Remarks will be in the University Ballroom starting promptly at 8:30 AM.

Sponsors

The 49th Annual West Coast Biological Sciences Undergraduate Research Conference has been made possible by the generosity and efforts of the following organizations and institutions.

CSUBIOTECH

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Organizing Committee:

Drs. Brent Hughes, Shannon Lee, Nathan Rank, and Lisa Hua

We are especially thankful to:

- Drs. Shannon Lee, Nathan Rank, Brent Hughes and Lisa Hua for reviewing abstracts, organizing the oral presentations and poster sessions
- Dr. Shannon Lee who helped coordinate the talk sessions, and Drs. Nathan Rank, Brent Hughes, and Lisa Hua for coordinating the poster session
- Dr. Shannon Lee, Dr. Derek Girman, Dr. Mackenzie Zippay and Sabrina Piana for organizing the judging
- Dr. Shannon Lee, Dr. Lisa Hua, and Kieran Chiddix for organizing and updating the Digital program, and Alyssa Stadie for editing and formatting
- Emily Porter (ORSP/Provost's Office) was invaluable help for so many things including Infoready support, securing funding for conference gear, foam boards, clips and easels, and other miscellaneous conference planning consultations!
- Brian Wichert, Jenna Hesterberg (Conference and Events Services), Emily Porter, and Alyssa Stadie for helping with the food orders and budget
- Jennifer Haynes, Adriana Vigil, and Jeff Keating for their work with Marketing including designing the Conference logo, and Dr. Shannon Lee for social media
- Brian Orr (Director of Student Financial Services) for helping set up Registration and responding frequently to conference registration related emails
- Emily Porter (Provost's Office) and Tai Russotti (Dean's Office) for administrative help throughout
- Ian Carlson, Sara Marquez, Emily Porter and Rosemary Galten for help with website design
- Dr. Michael McConnell (Point Loma Nazarene University) for guidance and encouragement, and handling conference publicity
- Gabi Chew, Corey Chattey (Campus Prints) for printing of name tags, and flyers
- Our Photographers, Mars Colonga, David Hua & Brennan Chin for their work
- Tiffany O'Neill and Kelley Kaslar for helping answer questions and providing expertise
- Dr. Sean Place for making time at Departmental meetings for conference announcements
- 60+ SSU faculty, staff and student volunteers- worked in a variety of ways, in particular **members of the Hua lab**, mostly behind the scenes and always without fanfare, to ensure the success of the 49th Annual WCBSURC

Internet services:

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Announcing Next Year’s Event

The 50th Annual WCBSURC
Point Loma Nazarene University
April 17th, 2027

Our Keynote Speakers

"Team Beetle", by Drs. Elizabeth Dahlhoff and Nathan Rank


Dr. Elizabeth Dahlhoff, Santa Clara University, and Dr. Nathan Rank, Sonoma State University, have a decades-long collaboration, rooted in their work at SCU and SSU respectively, and the White Mountain Research Station. Their overlapping pursuits and inclusion of undergraduates beautifully exemplify the spirit of this meeting! Both of their research programs have long placed undergraduates at the center of discovery, whether in the laboratory or in the field. Team Beetle has provided generations of students with transformative scientific experiences through collaborative, interdisciplinary work.

Work in the Dahlhoff laboratory focuses on examining physiological, biochemical and molecular mechanisms by which animals respond to environmental change. The fundamental questions asked are: Why do organisms live where they live, and how do they do so? What causes populations to remain stable, grow, fluctuate in size, or go extinct? How does the physical environment, the presence of predators, the genetic properties of a population and the abilities of individuals to adapt to the environment all influence their distribution and abundance? As ecosystems experience rapid climate change, it is imperative to understand and address these questions in vulnerable natural populations. The efforts of undergraduates are the heart of this research program; they work intensively in the laboratory and field as a collaborative, interdisciplinary team.

Dr. Rank's lab studies ecological interactions among plants and their herbivores and pathogens, as well as the adaptive significance of genetic variation in natural populations of insects. Since 1984, research has been focused on populations of the leaf beetle, *Chrysomela aeneicollis*, in the Sierra Nevada mountains of California, including its effects on host plant suitability and on its population and genetic responses to thermal variation in montane populations. Most insects, including leaf beetles, are hosts to bacteria that females transmit to their offspring and which affect the metabolism and reproductive biology of their insect host. The Rank lab investigates how these bacteria contribute to adaptation to local conditions and affect interactions between the beetles and their natural enemies.

Undergraduate Research Mentor Award:

Alison Haupt

Dr. Alison Haupt is an Associate Professor in the Department of Marine Science at California State University, Monterey Bay. Her research lab focuses on answering applied ecological questions that promote improved marine management and conservation by focusing on species and ecosystems confronting anthropogenic stressors such as urbanization, climate change, and fishing pressure. To address questions of applied ecology, her lab uses and applies a wide variety of tools ranging from population genetics to ecological modeling to traditional field ecology. At CSUMB, Dr. Haupt is passionate about creating positive first field experiences by getting students out in the field to pique their curiosity about natural history and ecology. Dr. Haupt has mentored over 60 undergraduate students, many of whom have since pursued careers in the sciences through graduate programs or jobs.

Program Overview AT-A-GLANCE

Schedule (all in University Student Center)

7:30 am	**Check-in & Poster Setup**	3rd floor hallway/Ballroom
8:30 am	**Welcome & Special Guest Appearances**	Ballroom
9:00 am	**Coffee, Juice & Snacks**	3rd floor hallway
9:00-11:15 am	**Morning Concurrent Talk Sessions**	Ballroom, Overlook, & Valley Rooms
	Session 1: **Aquatic/Marine Biology** (T1- T8)	Russian River Valley
	Session 2: **Biological Studies of Disease** (T9- T16)	Sonoma Valley
	Session 3: **Neurobiology** (T17 - T24)	Dry Creek Valley
	Session 4: **Ecology and Evolution** (T25 - T310)	Alexander Valley
	Session 5: **Physiology/Developmental Biology** (T32 - T39)	Bennett Valley
	Session 6: **Biochemistry, Biophysics & Microbiology** (T40 - T44)	Ballroom
	Session 7: **Cell and Molecular Biology** (T45 - T51)	Overlook
12:15 - 1:45 pm	**LUNCH**	The Kitchens
	11:15 -12:45 pm **Poster Session #1** (P1-P126)	Ballroom
	1:30 - 3:00 pm **Poster Session #2** (P127-P216)	Ballroom
3:00-3:30pm	**Coffee, Juice & Snacks**	3rd floor hallway
3:30-4:30 pm	**Keynote Address**	Ballroom
4:45-5:45 pm	**Awards Reception & Closing remarks**	Ballroom

ORAL Presentations AT-A-GLANCE

SEMINAR SESSION 9:00am-11:15pm

lists of concurrent oral presentations

NOTE: abstracts are in the next section of the program

Aquatic/Marine Biology (Russian River Valley Room)

Moderator and assistant: Dr. Brent Hughes and Aiyana Singh

9:00am(T1) **"Plenty of (Rock)Fish in the Sea: Genetic Species ID of Baja California Rockfish Larvae"**, Max Johnston-Gomez, Cal State Monterey Bay, Faculty Advisor: Dr. John Carlos Garza

9:15am(T2) **"Micro-habitat Preference and Stock Origin Assessment of the Striped Bass (*Morone saxatilis*) Mixed Migratory Stock in Southern New Jersey"**, Paige Kohlhase, Stockton University, Faculty Advisor: Dr. Adam Aguiar

9:30am(T3) **"The Zero-Sum Loss: Autotomy Does Not Compromise Burrowing Speed in *Microphiopholis gracillima*"**, Wyatt Lear, University of Tampa, Faculty Advisor: Dr. Kristian Taylor

9:45am(T4) **"Using Microsatellite Makers to Test the Population Genetics of Shrimp Species at Hydrothermal Vent Communities in the Lau Basin"**, Lukas Jimenez, Point Loma Nazarene University, Faculty Advisory: Dr. Walter Cho

10:00am(T5) **"Acoustic Activity of Risso's (*Grampus griseus*) and Pacific white-sided dolphins (*Lagenorhynchus obliquidens*) in the California Current Ecosystem"**, Alexis Hernandez, San Francisco State, Faculty Advisor: Dr. Anne Simonis

10:15am(T6) **"Shining a Light on Ocean Acidification: How European Green Crabs Respond to a Changing Ocean"**, Wendy Escobar*, Siena Fairbanks*, Tanner Ennis, and Diara Spain, Dominican University of California, Faculty Advisor: Dr. Diara Spain

10:30am(T7) **"Is there size assortative mating in wild Costa Rican cichlid fishes?"**, Desiree Thomas, Cal State Sacramento, Faculty Advisor: Dr. Ron Coleman

10:45am(T8) **"The Effects of Predaceous Diving Beetle Larvae Removal on Size and Capture Rate of California Tiger Salamander"**, Abigail Doan, Sonoma State, Faculty Advisor: Dr. Derek Girman

Biological Studies of Disease (Sonoma Valley Room)

Moderator and assistant: Dr. Joseph Lin and Alyssa Stadie

9:00am(T9) **"Wearable Electrochemical Biosensor for Lactate Detection in Sweat for Improved Post-Surgical Outcomes"**, Prisha Bali* and Madeline Kuan, University of California Los Angeles, Faculty Advisor: Dr. Daniel Kamei

9:15am(T10) **"Cellular Mechanisms and Clinical Importance of Fibrosis Regression in Metabolic Dysfunction-Associated Steatohepatitis (MASH)"**, Leonie Sarkissian, Sanford Burnham Prebys, UC San Diego School of Medicine, Faculty Advisor: Dr. Debanjan Dhar

9:30am(T11) **"Synthesis and Structure Activity Relationship of Anticancer Arylsulfonamides with Wnt-1 Modulating Activity"**, Lavernie Chen and Allyson Yu*, Aspiring Scholars Directed Research Program (ASDRP), Faculty Advisor: Dr. Edward Njoo

9:45am(T12) **"Evaluating Drug-Responsive Gene Editing Constructs for In Vivo Gene Therapy of Duchenne Muscular Dystrophy"**, Bianca Druta, University of Washington, Faculty Advisor: Dr. Niclas Bengtsson

10:00am(T13) **"Scaling Laws and Branching Patterns in Tumor Vasculature"**, Josh Mehdian, University of California Los Angeles, Faculty Advisor: Dr. Van Savage

10:15am(T14) **"Preclinical discovery of proscillaridin A glycan analogs for the in vitro and in vivo treatment of solid tumors"**, Shreya Somani, Aspiring Scholars Directed Research Program (ASDRP), Faculty Advisor: Dr. Edward Njoo

10:30am(T15) **"Efficacy and Safety Outcomes in Subjects Undergoing Customized Oral Immunotherapy as a Treatment for Food Allergy"**, Janice Wong, University of California Los Angeles and National University of Singapore, Faculty Advisor: Dr. Elizabeth Huiwen Tham

10:45am(T16) **"CRISPR-Cas9 Mediated Mutagenesis of the CYP2U1 Gene to Generate a Patient-Specific Eye Disease Model"**, Adelin Tulcan* and Gavin Rubin, Point Loma Nazarene University,

Neurobiology (Dry Creek Valley)

Moderator and assistant: Dr. Evan Lintz and Kieran Chiddix

9:00am(T17) **"Computational Neuroscience and Machine Learning Analysis for Understanding Sex-Specific Differences in Microglial Morphology and Cognitive Function in Alzheimer's Disease"**, Avni Sriram, Aspiring Scientists Summer Internship Program (ASSIP) at George Mason University

9:15am(T18) **"Polyaromatic hydrocarbons inhibit neurite outgrowth by binding to the cell-surface laminin receptor"**, Bryce Blaugrund, University of Southern California, Faculty Advisor: Dr. Rayudu Gopalakrishna

9:30am(T19) **"The Effects of Low-Dose Naltrexone on Mice That Have Experienced a Traumatic Stressor"**, Chloe Sy Perez* and Owen Bella, University of California Los Angeles, Faculty Advisor: Catherine M. Cahill

9:45am(T20) **"Investigating the Effect of Noradrenergic μ-Opioid Receptors in Fear Learning via a Fear Contextualization Paradigm"**, Shifa Dhar* and Jeongha (Bryan) Kim, University of California Los Angeles, Faculty Advisor: Dr. Catherine Cahill

10:00am(T21) **"Vagus Nerve Stimulation Reduces Reinstatement of Drug Seeking of Fentanyl in Rats"**, Dylan Cavalheiro, University of California Los Angeles, Faculty Advisor: Catherine M. Cahill

10:15am(T22) **"Characterizing the Cell-Autonomous Role of CHCHD2 in Neural Stem Cell Fate Decisions during Adult Hippocampal Neurogenesis"**, Miguel Angel Sierra-Velazquez, University of North Carolina at Chapel Hill, Faculty Advisor: Dr. Juan Song

10:30am(T23) **"Mechanisms by Which Polyphenols Inhibit Phosphodiesterases and Promote Neuroprotection"**, Anika Seshadri, University of Southern California, Faculty Advisor: Rayudu Gopalakrishna

10:45am(T24) **"CVLT Memory Scores are Predicted by MRI Regional Volumes, Neurite Density, and ERPs Across the Stages of Alzheimer's Disease (AD)"**, Nehal Shahi, University of California Davis

Ecology and Evolution (Alexander Valley Room)

Moderator and assistant: Dr. Matt Clark and Vic Fransen

9:00am(T25) **"The Genetic Architecture of Wisdom: A Genome-Wide Association Study in 131,870 Individuals"**, Fabrizio Malatesta, University of California San Diego, Faculty Advisor: Dr. Sandra Sanchez-Roige

~~**9:15am(T26) "Prevalence of Haemosporidian lineages in a resident songbird across urban-rural habitats"**, Anneliese Roth, Fresno State University~~ CANCELLED

9:30am(T27) **"Global Activity Patterns and Habitat Use of Tapirs (*Tapirus spp.*) as Revealed by Large-Scale Camera Trap Data"**, Elise Kinney and Sophia Peck*, Point Loma Nazarene University, Faculty Advisor: Dr. Michael Mooring

9:45am(T28) **"Conservation Genomics of an Endangered Succulent in Santa Clara County"**, Karina Martinez* and Evan Hackstadt, Santa Clara University, Faculty Advisor: Dr. Justen Whittall

10:00am(T29) **"Protocol Development for Enhanced DNA Visualization in the California Bay Laurel"**, Jillian Gonzalez* and Claudia Tellez*, Sonoma State University, Faculty Advisor: Dr. Lisa Hua

10:15am(T30) **"The impact of parasitism: social distancing in a species from a lineage of gregarious insects"**, Ariana Thompson* and Maya Richter, Santa Clara University, Faculty Advisor: Dr. Janice Edgerly-Rooks

10:30am(T31) **"Going up, tuning down: Tympanum size variation in *Rana boyii*"**, Daniella Mori* and Aziza Corley, University of San Francisco, Faculty Advisor: Dr. Jennifer Dever

Physiology and Developmental Biology (Bennett Valley Room)

Moderator and assistant: Dr. Mackenzie Zippay and Omar Akhtar

9:00am(T32) **"Spatial organization of homologous chromosomes 2 and 16 in mouse embryonic fibroblasts"**, Omoefe Odiase, Sonoma State University, Faculty Advisor: Dr. Lisa Hua

9:15am(T33) **"Voltage-gated Sodium Channel Nav1.1 is Necessary for Muscle Spindle Afferent Function in Adult Mice"**, Mariam Malik, San Jose State University, Faculty Advisor: Dr. Katherine A. Wilkinson

9:30am(T34) **"Investigating the Role of Developmental Signaling Pathways in Early Macrophage Route Choice During Drosophila Embryogenesis"**, Steve Chung, University of California Los Angeles, Faculty Advisor: Dr. Daria Siekhaus

9:45am(T35) **"Dynamics of Gastrulation in *Trachemys scripta* Turtle: Correlating Changing Blastopore Shapes with the Progression of Tissue Internalization"**, Lila Le, Loyola Marymount University, Faculty Advisor: Dr. Maxellende Ezin

10:00am(T36) **"Psilocin Alters Craniofacial Proportions and Accelerates Ossification in Developing Chick Embryos"**, Sarmad Alani, Loyola Marymount University, Faculty Advisor: Dr. Maxellende Ezin

10:15am(T37) **"Cardiovascular Screening in Homeless Outreach: An Operationally Ethical Protocol"**, Sarah Jean Valliant* and Ileana Jade Rodriguez, San Francisco State University

Biochemistry, Biophysics and Microbiology (Ballroom)

Moderator and assistant: Dr. Monica Lares and Jason Romero

9:00am(T38) **"Concentration-Dependent Emergent Cooperativity in Myoglobin Pseudoperoxidase Activity Under Oxidative Stress"**, Kade Sutherland, Utah Valley University, Faculty Advisor: Dr. Daniel Scott

9:15am(T39) **"NanoBRET-based Assay for Monitoring Protein Palmitoylation"**, Andrew Medina* and Dylan Karkainen, University of Southern California, Faculty Advisor: Dr. Chao Zhang

9:30am(T40) **"Identification of potential antibiotic compounds for Gram-positive bacteria via optimization of a high-throughput FRET assay"**, Steven Nguyen, Creighton University

9:45am(T41) **"Structural analysis of *Crassostrea gigas* OAZ-PK RNA via Selective 2'-Hydroxyl Acylation analyzed by Primer Extension (SHAPE)"**, Daniel Cline, Creighton University, Faculty Advisor: Juliane Strauss-Soukup

10:00am(T42) **"Phosphofructokinase mutations in Glycogen Storage Disease Type VII lower ligand affinity and disrupt oligomer assemblies"**, Emily Li, University of Washington, Faculty Advisor: Dr. Justin Kollman

10:15am(T43) **"Color With a Cost: Metabolic Dyes TTC and INT Compromise *Bacillus subtilis* Survival at Common Assay Concentrations"**, Sumaiya Tohorat, South Dakota State University, Faculty Advisor: Dr. Nicholas C Butzin

10:30am(T44) **"Integrating Bioinformatics and Synthetic Biology to Engineer Hypoxia-Resilient Cell Therapies in Jurkat T Cell Models"**, Raymond Nova*, Shardul Singh and Janice Subroto, University of California Los Angeles, Faculty Advisor: Dr. Chase Linsley

Cell and Molecular Biology (Overlook)

Moderator and assistant: Dr. Shona Mookerjee and Sabrina Piana Cervantes

9:00am(T45) **"Assessing the Contribution of Multiciliary Arrays to Unidirectional Fluid Flow"**, Tyler Kang*, Santa Clara University, Faculty Advisor: Dr. Brian Bayless

9:15am(T46) **"Investigating a potential eukaryotic riboswitch in OAZ mRNA from *Agaricus bisporus* via in-line probing"**, Sarah Fowler, Creighton University

9:30am(T47) **"Cowpea Mosaic Virus as a Model for Understanding Viral Evolution"**, Hayden Gielda, Point Loma Nazarene University, Faculty Advisor: Kristopher J. Koudelka

9:45am(T48) **"Major Front-End Migration for GRNsight, a Web Application for Visualizing Gene Regulatory and Protein-Protein Interaction Network Models"**, Cecilia J. Zaragoza, Loyola Marymount University, Faculty Advisor: Faculty Advisor: Dr. Kam D. Dahlquist

10:00am(T49) **"Impact of Oxidized Polyunsaturated Acids on Macrophage Efferocystosis Capacity"**, Victoria Huang, University of California Los Angeles, Faculty Advisor: David Meriwether

10:15am(T50) **"TLR10 Attenuates TLR2-Mediated Inflammatory Signaling in Response to Probiotic Lactobacillus Strains"**, Anya Jaramillo, University of San Francisco, Faculty Advisor: Dr. Sangman Kim

10:30am(T51) **"Magnolia Leaf Silver Nanoparticles Display Dose-Dependent Disruption of Bacterial Biofilms"**, Faith Azer* and Amadeus Nunez, The Master's University, Faculty Advisor: Dr. Haley Smith

SEMINAR SESSION ABSTRACTS

Aquatic/Marine Biology

T1. "Plenty of (Rock)Fish in the Sea: Genetic Species ID of Baja California Rockfish Larvae",
Max Johnston-Gomez, Cal State Monterey Bay, Faculty Advisor: Dr. John Carlos Garza

Rockfishes in the genus Sebastes are highly speciose with over 100 recognized species, most of which inhabit the Northeast Pacific Ocean. Adult rockfishes are highly diverse in both ecological niche and morphology. In contrast, many sympatric rockfish species are morphologically indistinguishable as juveniles. In such cases, genetic species identification provides a valuable alternative to traditional visual methods. Here we evaluate the performance of a genetic species identification tool, originally developed for rockfishes of the central California coast, in accurately identifying the species of a cohort of larval rockfish from Baja California, Mexico. We extracted DNA from 166 larval specimens collected during mid-water trawl surveys, and used high-throughput DNA sequencing to genotype 96 nuclear microhaplotype loci. Then we implemented multiple species assignment tests and compared the results to corroborate identifications for each sample. Our findings indicate largely high confidence assignments with strong concordance across assignment methods. These results demonstrate this tool's effectiveness in identifying larval rockfish outside of the region in which it was originally calibrated. When integrated with traditional visual identification methods, this genetic approach could strengthen research surveys and stock assessments in Baja California marine ecosystems. Ultimately, more accurate species identification can support better-informed management decisions and contribute to the sustainability of both commercial and recreational fisheries in the region.

T2. "Micro-habitat Preference and Stock Origin Assessment of the Striped Bass (*Morone saxatilis*) Mixed Migratory Stock in Southern New Jersey", Paige Kohlhase, Stockton University, Faculty Advisor: Dr. Adam Aguiar

Striped bass (Morone saxatilis) represents the most sought-after inshore game fish on the East Coast of the United States, generating significant tourism, revenue, and industry for the region. Ambiguity remains regarding the micro-habitat preferences, migration behaviors, and stock origins of this vital recreational, commercial, and ecological resource. While the general migration patterns of spawning-size striped bass are somewhat understood, nuances exist across fish size and age. The behavior of smaller bass differs significantly from that of larger spawning-size individuals, and their behavior varies across different geographical areas. Striped bass exhibit high fidelity to their natal spawning grounds. However, the New Jersey coast serves not as a major spawning ground, but as a migratory corridor for mixed-migratory groups during the spring and

fall. The specific contributions from major (and minor local) spawning grounds to this migratory stock remain unclear. Utilizing tagging instrumentation, catch logs, and scale/tissue sampling, followed by DNA sequence comparisons, this study aims to clarify: 1) shifts in migration and location preference, and 2) the extent to which specific spawning grounds contribute to the local mixed-migratory stock—potentially illuminating differences between fall and spring cohorts. Statistical analysis was performed on catch log data to identify behavioral and foraging patterns. Research students utilized Stockton University-specific American Littoral Society circular clip tags during the catch-and-release of striped bass across Ocean and Atlantic counties. Environmental variables (e.g., moon phase, tide, time, and wind direction) were recorded and analyzed for correlation. Fin clippings and scale samples were collected for DNA extraction, purification, and sequence comparison. While tag return data and DNA comparisons are ongoing, preliminary catch log analysis suggests a higher frequency of foraging activity in Barnegat Bay compared to surrounding areas. High slack appears to be the preferred feeding tide, with the average fish size ranging between 24 and 31 inches.

T3. "The Zero-Sum Loss: Autotomy Does Not Compromise Burrowing Speed in *Microphiopholis gracillima*", Wyatt Lear, University of Tampa, Faculty Advisor: Dr. Kristian Taylor

Autotomy is a common defense mechanism seen among various taxa. While different mechanisms are used, the purpose is to sacrifice an appendage in hopes of self-preservation. In many cases, a trade-off occurs whereby autotomy can produce decreased locomotion in arachnids, issues with balance among reptiles, and even hydrodynamic drag among cephalopods. Examining the burrowing speed of Microphiopholis gracillima, a common shallow water ophiuroid found in the western Atlantic, has shown that autotomy does not significantly impact burrowing speed, a common escape mechanism used by members of the Class. Specimens with various numbers of arms (having undergone autotomy before collection) were gathered in Tampa Bay, FL, along with water and local sediment. Individuals were then introduced to separate 20 L tanks and behavior was video recorded. Comparison of burrowing speed among specimens with three, four and five arms revealed no significant differences (one-way ANOVA $p=0.92$). Comparing oral disk diameter, as well as arm length, still resulted in no significant differences. While feeding efficiency might be impacted, this study established that autotomy among M. gracillima does not impact the all-important burrowing behavior.

T4. "Using Microsatellite Makers to Test the Population Genetics of Shrimp Species at Hydrothermal Vent Communities in the Lau Basin", Lukas Jimenez, Point Loma Nazarene University, Faculty Advisory: Dr. Walter Cho

In 2017 during the Underwater Fire expedition cruise, the shrimp species Rimicaris cambonae was collected from hydrothermal vent systems at the underwater West Mata Volcano in the Lau Basin. To examine the genetic structure of this population, DNA was extracted and analyzed from forty-two shrimp samples using seven microsatellite markers. MICRO-CHECKER and a Hardy Weinberg equilibrium test were performed to determine the quality of the markers. Population structure was analyzed using AMOVA, the Mantel Test, and STRUCTURE. The results indicated no population structure and high levels of gene flow across the site, aligning with results from previous studies. These results introduce more information concerning these understudied ecosystems, which may provide insights into the conservation of these locations when it comes to human influence through practices such as deep-sea mining.

T5. "Acoustic Activity of Risso's (*Grampus griseus*) and Pacific white-sided dolphins (*Lagenorhynchus obliquidens*) in the California Current Ecosystem", Alexis Hernandez, San Francisco State, Faculty Advisor: Dr. Anne Simonis

Acoustic Activity of Risso's (Grampus griseus) and Pacific white-sided dolphins (Lagenorhynchus obliquidens) in the California Current Ecosystem Proposed author order: Alexis Hernandez, Lauren Rocheleau, Cory Hom-Weaver, Jeff Moore, Anne Simonis Risso's dolphins (Grampus griseus) and Pacific white-sided dolphins (Lagenorhynchus obliquidens) are two odontocete species commonly found in the California Current Ecosystem. These species can be identified by the spectral characteristics in their echolocation clicks, enabling passive acoustic monitoring as a tool for species identification. The primary objective of this study is to use archived passive acoustic data to evaluate differences in their spatial and temporal habitat use. Acoustic data was collected between July and December 2024 during the NOAA California Current Cetacean and Ecosystem Assessment Survey (CalCurCEAS). Data was collected by drifting acoustic recorders (hydrophones) throughout offshore waters of the California Current. Acoustic data was reviewed manually using long-term spectral averages (LTSAs) to identify species-specific acoustic encounters. The results show that Risso's dolphins and Pacific white-sided dolphins exhibit higher echolocation activity during nighttime and increased acoustic detections during the winter season. Also, data illustrates more Risso detections in the southern region while Pacific white-sided dolphins have more detections in the northern region. Looking further within this data I will evaluate environmental parameters associated with the presence of each species. This study adds to a growing long-term passive acoustic dataset used to assess marine mammal distribution and population dynamics in the California Current. Continued monitoring across broader spatial and temporal scales will improve understanding of habitat use and behavioral ecology for these species.

T6. "Shining a Light on Ocean Acidification: How European Green Crabs Respond to a Changing Ocean", Wendy Escobar*, Siena Fairbanks*, Tanner Ennis, and Diara Spain, Dominican University of California, Faculty Advisor: Dr. Diara Spain

Ocean acidification is an increasing environmental concern that may affect the growth and survival of marine organisms. Popular media such as Moana often portrays marine life as resilient and adaptable, raising questions about how real organisms respond to environmental stress. This study examined how changes in ocean acidity influence the growth and physical development of the European green crab, Carcinus maenas.Ocean acidification is an increasing environmental concern that may affect the growth and survival of marine organisms. Popular media such as Moana often portrays marine life as resilient and adaptable, raising questions about how real organisms respond to environmental stress. This study examined how changes in ocean acidity influence the growth and physical development of the European green crab, Carcinus maenas.Specimens were collected locally from Stinson Beach, California, and maintained in a controlled aquatic laboratory at Dominican University of California. Crabs were divided into control and experimental groups and monitored over an eight-week period. The experimental group was periodically exposed to acidified seawater, while the control group remained in ambient conditions.Growth was assessed through measurements of carapace dimensions and body mass. This experiment resulted in over 75% of specimens in both groups showing an increase in size and mass. Differentiating between the two specimen groups, the control crabs exhibited greater carapace expansion, while the experimental crabs demonstrated a higher increase in body mass. These patterns suggest that exposure to acidified conditions may influence how crabs allocate energy toward structural growth. These findings indicate that European green crabs may maintain physical robustness under the stress of changing ocean chemistry, contributing to their environmental persistence. Much like the shiny crab Tamatoa in Moana, the growth of these crab's exoskeletons is of great importance to their health and well-being.

T7. "Is there size assortative mating in wild Costa Rican cichlid fishes?", Desiree Thomas, Cal State Sacramento, Faculty Advisor: Dr. Ron Coleman

How do individuals choose their mate? Mate choice could be based on many things, including qualities of the mate (color, size, ornamentation, genes, etc.) and/or quality of the territory, spawning location, etc. Previous lab research has shown individuals within a species of cichlid fish appear to mate size-assortatively, i.e., big females mate with big males and small females mate with small males. To investigate this in the field, for 3 weeks in January 2026, I visited ten rivers of northeast Costa Rica to examine breeding pairs of the following cichlid species: Septs (Amatitlania septemfasciata), Myrnae (Amatitlania myrnae), Neets (Neetroplus nematopus), Tubas (Tomocichla tuba), Alfari (Cribroheros alfari), and Convicts (Amatitlania nigrofasciata). I wanted to quantify assortative mating with these species.I wanted to measure the size of the male and

female in each breeding pair of cichlid, however, I could not actually catch the fish to measure them without disturbing their parental care. Instead, I used models of known size to estimate the size of breeding fish. I positioned a model of known length right next to each fish. I got data on 108 pairs of cichlids across six species. For each species, I analyzed the data using a least-squares regression of male length on female length. This tested my hypothesis to see if they are mating size-assortatively. For all species considered together, there was a strong positive relationship between male size and female size (n=108). Furthermore, four of the species analyzed individually shared the same significant relationship, strongly indicating that there was positive size–assortative mating. The results of this study will add to our understanding of mate choice, evolution and sexual selection in neotropical, substrate spawning cichlids.

T8. "The Effects of Predaceous Diving Beetle Larvae Removal on Size and Capture Rate of California Tiger Salamander", Abigail Doan, Sonoma State, Faculty Advisor: Dr. Derek Girman

Vernal pools are seasonal wetland ecosystems that are home to a variety of plant and animal specialist species. Habitat loss has resulted in these specialists becoming threatened or endangered, including the California Tiger Salamander (Ambystoma californiense), which spends its larval stage as a vernal pool predator. In order to properly curate management plans of threatened ecosystems, an understanding of trophic interactions among predators in these ecosystems is vital. In the current study, the impacts of the predaceous diving beetle larvae (Family Dytiscidae), a predator of A. californiense larvae, on the capture rate and size of A. californiense are observed via the removal of Dytiscid larvae from multiple experimental sites. Two vernal pools that are known to contain larvae of both A. californiense and Dytiscidae were divided into a control and experimental side using mesh screening, and Dytiscid larvae were removed from the experimental side via timed dipnet surveys. Timed dipnet surveys were also used to monitor A. californiense larvae on both sides and record their capture rate as well as their body length and gape size. Regular timed dipnetting is performed throughout the season to monitor these factors and routinely remove Dytiscidae from the experimental side.

Biological Studies of Disease

T9. "Wearable Electrochemical Biosensor for Lactate Detection in Sweat for Improved Post-Surgical Outcomes", Prisha Bali* and Madeline Kuan, University of California Los Angeles, Faculty Advisor: Dr. Daniel Kamei

Although recent advancements in medical technology have greatly improved post-surgical outcomes, nearly 40% of surgery patients still experience post-operative complications necessitating reintervention or resulting in mortality. Elevated levels of inflammation at the post-surgical site are considered to be an indicator of potential infection or complications. Recent studies have established a direct correlation between sweat lactate and blood lactate levels, which

serve as a biomarker for inflammation. Continuous and noninvasive monitoring of lactate levels in the postoperative period provides a useful pathway to monitoring localized inflammation. While electrochemical biosensors provide an ideal, cost-effective platform for continuous, wearable lactate monitoring, challenges remain regarding long-term enzyme stability and ensuring reliable current output across varying analyte concentrations. Our designed lactate biosensor utilizes a Prussian blue (PB) and an enzyme layer of immobilized lactate oxidase (LOx) on the working electrode of a screen-printed carbon electrode to maximize low potential current output to enable lactate detection for correlation with tissue inflammation. The PB layer is drop-casted onto the surface rather than being formed via surface deposition. It functions as a catalyst for the reduction of H2O2, produced by the LOx-catalyzed oxidation of lactate, enabling more sensitive sensing of lactate levels at the electrode surface.The enzyme layer is synthesized using a matrix of chitosan, bovine serum albumin (BSA), and trehalose to ensure stabilization and adherence to the electrode. Both catalytic and enzymatic layers are specifically drop-casted onto the working electrode, allowing for current measurements relative to the reference input. Using a potentiostat, the sensor was tested via chronoamperometry at a constant potential of -0.05 V across 10-75 mM lactate solutions, representing physiologically relevant sweat lactate concentrations for inflammation. Linear increases in steady-state current were observed for lactate concentrations between 10 mM and 20 mM, with tests for 40 mM and 75 mM ongoing. The results confirm that the combined PB and LOx sensing layers enable reliable catalytic detection of lactate, demonstrating the device's potential as an affordable, continuous monitor for post-surgical inflammation, aiding in improved patient outcomes.

T10. "Cellular Mechanisms and Clinical Importance of Fibrosis Regression in Metabolic Dysfunction-Associated Steatohepatitis (MASH)", Leonie Sarkissian, Sanford Burnham Prebys, UC San Diego School of Medicine, Faculty Advisor: Dr. Debanjan Dhar

Metabolic dysfunction-associated steatohepatitis (MASH) is the inflammatory and fibrotic form of metabolic dysfunction-associated steatotic liver disease. It develops when excess lipid accumulation in hepatocytes leads to cellular stress, inflammation, ballooning injury, and activation of wound-healing pathways that drive fibrosis. Among all histologic features, fibrosis stage is the strongest predictor of liver-related and overall outcomes, making fibrosis regression a major therapeutic goal. MASH is also a significant global health concern because it can progress to cirrhosis, liver failure, and hepatocellular carcinoma (HCC). It has become one of the fastest-growing causes of HCC and is now the second leading indication for liver transplantation, largely due to the increasing prevalence of obesity and type 2 diabetes.Evidence for MASH regression has been observed in human studies through weight loss, bariatric surgery, and pharmacological therapies such as GLP-1 receptor agonists (e.g., semaglutide and tirzepatide) and pioglitazone. These interventions can reduce the NAFLD Activity Score (NAS) and fibrosis stage. Animal studies have similarly demonstrated regression following high-fat diet withdrawal or genetic modifications. At the cellular level, fibrosis regression is driven largely by hepatic stellate

cell (HSC) inactivation or apoptosis. Activated myofibroblasts may revert to a quiescent-like state through pathways such as PPARγ reactivation and matrix metalloproteinase (MMP)-mediated extracellular matrix degradation.Additional mechanisms include extracellular matrix remodeling, macrophage phenotype switching toward pro-resolution states, hepatocyte regeneration through Wnt/β-catenin and YAP/TAZ signaling, and improved lipid metabolism. However, many aspects of fibrosis regression remain unclear, including why some patients respond to therapy while others do not and how fibrosis remodeling occurs over time. Because MASH is a major driver of advanced liver disease and transplantation, further research is needed to better understand these mechanisms and develop effective therapies that prevent progression to cirrhosis and HCC.

T11. "Synthesis and Structure Activity Relationship of Anticancer Arylsulfonamides with Wnt-1 Modulating Activity", Lavernie Chen and Allyson Yu*, Aspiring Scholars Directed Research Program (ASDRP), Faculty Advisor: Dr. Edward Njoo

The Wnt1/β-catenin signaling pathway plays a critical role in oncogenesis through regulation of key biological processes including embryonic development, organogenesis, and tissue homeostasis. Pathway dysregulation where Axin and APC mutations are introduced leads to abnormal β-catenin accumulation, a known driving factor in the development of several cancers. Previous studies have identified methyl 3-{[(4-methylphenyl)sulfonyl]amino}benzoate (MSAB) as an selective inhibitor of the Wnt1/β-catenin pathway. To further explore the structure activity relationship of this compound, we prepared a systemic library of substituted phenyl, alkyl, and heterocyclic sulfonamides, and their effects were tested in vitro through cell viability assays on HCT116, HT29, CT26, MDA-MB-231, 4T1, HepG2, SK-OV-3, B16F10, A549, Calu1, HEK-293, and 3T3 cell lines. We further deployed a TCF/LEF-dependent fLuc reporter assay to directly interrogate Wnt1 antagonistic activity. We demonstrate that para-substituted aryl systems exhibited the greatest antiproliferative potency, with several clear halogen-substituent effects, while fully saturated systems led to a precipitous loss of potency. Additionally, inspired by similar substructures identified in the patent literature as having STAT3-antagonistic activity, we evaluated the potency of our analogs through a STAT3-dependent reporter cell line assay, and found that trifluoromethyl-substituted arylsulfonamides exhibited moderate STAT3-inhibitory activity. Collectively, these observations provide a robust SAR towards the design and discovery of anticancer arylsulfonamides with selective Wnt1 or STAT3 inhibitory activity.

T12. "Evaluating Drug-Responsive Gene Editing Constructs for In Vivo Gene Therapy of Duchenne Muscular Dystrophy", Bianca Druta, University of Washington, Faculty Advisor: Dr. Niclas Bengtsson

Duchenne Muscular Dystrophy (DMD) is a devastating progressive muscle disease caused by mutations in the dystrophin gene. One function of dystrophin is to stabilize and protect muscle cells from injury during contraction. Therefore, a lack of functional dystrophin causes recurring

muscle damage resulting in muscle necrosis and replacement of fibers with fibrotic tissue and fat. Experimental in vivo gene editing therapies aim to restore functional dystrophin protein expression using viral vector-mediated CRISPR/Cas9 gene editing to correct causative mutations in the dystrophin gene. While these approaches have shown great promise, the continuous expression of bacterially derived editing components can potentially trigger a severe immune response. This project aims to address these immunological complications by developing a drug-responsive and tunable gene editing system that can toggle expression of editing components on and off. Here we present data from preliminary cell culture studies where we screened two classes of RNA elements capable of regulating expression of genes delivered through viral vectors: riboswitches and pA regulators. Our results show that pA regulators clearly outperform riboswitches in the drug-responsive gene activation of a reporter gene, Luciferase. Further in vitro tests helped identify a top pA regulator candidate, exhibiting a favorable dose response curve to the drug Tetracycline at clinically relevant doses. Ongoing in vivo validation studies focus on adapting the delivery of top pA regulators for temporal regulation of dystrophin gene editing activity in a widely used mouse model of DMD. Overall, this tunable system is showing great promise for reducing risks associated with in vivo gene editing for DMD, ultimately presenting a safer approach to gene therapy.

T13. "Scaling Laws and Branching Patterns in Tumor Vasculature", Josh Mehdian, University of California Los Angeles, Faculty Advisor: Dr. Van Savage

Tumors grow by building their own vascular networks, which deliver the oxygen and nutrients needed to survive and expand. In healthy tissue, blood vessels follow well-established biophysical rules, including power-law scaling relationships between vessel size and flow and branching patterns that efficiently fill space. Tumor vessels, by contrast, develop under mechanical stress, chaotic signaling, and uneven nutrient conditions, so they often diverge from these norms. Measuring how tumor vasculature deviates from healthy vasculature principles can reveal how biophysical forces shape their structure. The objective of this study is to test whether tumor vascular networks still obey the power-law scaling and branching principles predicted by optimal network theory. Specifically, we examine two features: (1) how vessel radius scales with vessel length, and (2) how this radius-length relationship and space-filling patterns differ between canonical vessel-labeling and Horton-Strahler labeling. We reconstructed three-dimensional vascular networks from DICOM imaging datasets. For each vessel segment, we measured radius and length, then assigned a Strahler order to capture the branching hierarchy. To assess scaling behavior, we fitted log-log regressions between vessel radius and vessel length, then compared the resulting exponents to theoretical predictions for healthy vasculature, such as Murray's law, which describes how vessel radii relate at branching points to minimize transport costs. Preliminary results indicate that tumor vessels display roughly linear relationships on log-log plots, consistent with power-law behavior. However, the specific scaling exponents deviate from values previously established for healthy vasculature. Additionally, the Horton-Strahler distributions are skewed

towards lower-order branches, implying that tumors favor short, local sprouting rather than deep, space-filling trees. Overall, tumor vasculature exhibits power-law scaling but differs in branching architecture from optimal networks, reflecting the genetic regulation and mechanical constraints inside tumors. Going forward, we plan to expand analysis across tumor types and integrate fluid-dynamic simulations to better understand functional consequences.

T14. "Preclinical discovery of proscillaridin A glycan analogs for the in vitro and in vivo treatment of solid tumors", Shreya Somani, Aspiring Scholars Directed Research Program (ASDRP), Faculty Advisor: Dr. Edward Njoo

Cancer persists as one of the leading causes of mortality in the US, accounting for over 611,000 deaths in 2024. In parallel, several small-molecule drugs are currently in development for the treatment of malignancies. Within those, natural products, their synthetic analogs, or other derivatives, make up about 50% of all FDA-approved anticancer small molecules. Several naturally-derived cardiac glycosides, including digoxin, digitoxin, and proscillaridin A, have been identified as cardiomyocyte modulators and are currently being investigated for their anticancer properties. These cardiac glycosides are generally classified into cardenolides and bufadienolides, which bear butenolide and pyrone D-ring functionality, respectively, and have exhibited remarkable in vitro toxicity in various cancerous cell lines. As simple modifications on steroidal small molecules have demonstrated success in augmenting bioavailability or enhancing downstream biological activities, we synthesized five analogs of proscillaridin A, a bufadienolide isolated from the genus Scilla, bearing acetate esters, silyl ethers, or dimethyl ketals to investigate how ketalization, acetylation, and/or silylation of the A-ring allylic glycoside might alter its anticancer properties. The antiproliferative activity of these compounds was evaluated alongside proscillaridin A, digoxin, and digitoxin across several breast, colorectal, liver, and ovarian cancer cell lines. Through a diverse panel of cell viability and cytotoxicity experiments, reporter assays, and cell cycle and protein marker analysis by flow cytometry, we find that ketalization of the glycan of proscillaridin A provides similar, and in some cases enhanced, in vitro potency. Subsequently, to evaluate the preclinical applicability of these compounds in inhibiting solid tumor growth, we found that direct, intraperitoneal injection of proscillaridin A and its analogs demonstrated reasonably potent antitumor activity in a murine mammary cancer model, albeit with dose-limiting ADMET properties. This study establishes the foundation for further in vitro and in vivo evaluations of cardiac glycosides for the treatment of cancer.

T15. "Efficacy and Safety Outcomes in Subjects Undergoing Customized Oral Immunotherapy as a Treatment for Food Allergy", Janice Wong, University of California Los Angeles and National University of Singapore, Faculty Advisor: Dr. Elizabeth Huiwen Tham

Clinical trials on food oral immunotherapy (OIT) prompt safety concerns due to high rates of adverse reactions (AE) on a strict trial protocol. Clinical OIT, however, allows for tailored

regimens to suit each individual patient's risk profile. This study demonstrates the efficacy and safety of a customized clinical OIT approach in a tertiary allergy centre. Methods: n=97 patients undergoing clinical OIT in a tertiary allergy centre were recruited (n=65 peanut, n=9 milk, n=11 egg, n=12 other). All patients underwent a baseline challenge to determine reaction threshold, and then commenced build-up following a standardized protocol that could be adjusted (by dose, interval or premedication), to reach a maintenance dose chosen in accordance with patients' preferences. Data on AEs, dose adjustments and long-term allergen consumption were collected. Results: Of 97 patients, n=57 (58.7%) achieved desensitization without protocol adjustments, n=29 (29.9%) with protocol adjustments, and n=11 (11.3%) withdrew. During the build-up phase, 5.02 ± 4.73% of doses are associated with AE. Adrenaline use was required in 0.036% of doses at home and none in the hospital. Mean duration of maintenance was 33 ± 23 months with 62.9% of patients experiencing no reactions. Patients achieved mean maintenance doses of milk: 3664 ± 3199 mg, egg: 6250 ± 3489 mg, peanut: 1943 ± 1678 mg. Conclusion: Customized OIT is a safe and effective approach that achieves high rates of desensitization with sustained long-term allergen consumption.

T16. "CRISPR-Cas9 Mediated Mutagenesis of the CYP2U1 Gene to Generate a Patient-Specific Eye Disease Model", Adelin Tulcan* and Gavin Rubin, Point Loma Nazarene University, Faculty Advisor: Michael Dorrell

Macular telangiectasia (MacTel) is a degenerative eye disease that affects the retina and central vision, with progression starting around 50 years of age. The mechanism of disease remains unknown but there is strong evidence of a genetic, inheritable component. Our studies aim to look at genes that may play a role in disease progression. Prior studies have compared genetic information from patients compared with familial controls, identifying CYP2U1 as a gene related to MacTel, however little is known about its function. We suspect that there may be a relationship between a one base pair substitution mutation in two different exons of the CYP2U1 gene. The purpose of this project is to use CRISPR-Cas9 to create patient specific mutations in CYP2U1 to learn more about its disease-related function. Bioinformatics tools were employed to create CRISPR-Cas9 guides or primers capable of targeting and cutting a specific part of the genome. This would then allow us to generate the desired mutation using specific homology directed repair (HDR) guides. The Cas9 plasmid with cutting guides were transformed into bacterial plasmids, produced in E. coli, and then transfected successfully into HEK293 cells using different transfection techniques to optimize transfection. The cut guides were tested for their ability to create insertions and deletions (InDels) at the CYP2U1 mutation site indicating successful targeting and genomic cutting. Currently, we are following the same process using human retinal cells (ARPE19), transfecting with the Cas9, cutting guide plasmids and the HDR guide to create the desired patient-relevant point mutations. Studying ARPE19 cells has the potential to facilitate assays that will provide further insight about the function of these patient-specific CYP2U1 mutations on MacTel and its effects on the retina.

Neurobiology

T17. "Computational Neuroscience and Machine Learning Analysis for Understanding Sex-Specific Differences in Microglial Morphology and Cognitive Function in Alzheimer's Disease", Avni Sriram, Aspiring Scientists Summer Internship Program (ASSIP) at George Mason University

Alzheimer's disease (AD) disproportionately affects women, with females accounting for approximately two-thirds of all dementia cases (Rajan et al., 2021). Despite this striking disparity, the influence of biological sex on AD pathology and behavioral outcomes remains underexplored (Kolahchi et al., 2024). Microglia—the brain's resident immune cells—are critical for synaptic regulation, and their morphology reflects functional state. Therefore, morphological analysis offers critical insight into how microglia influence AD pathology. In this study, we performed a computational investigation quantitatively comparing hippocampal microglial morphology and cognitive performance in female and male 5xFAD mice, a transgenic AD model, and their wild-type controls. Leveraging digitally reconstructed cells from NeuroMorpho.org, we analyzed >6,000 microglia (AD n=4,834; Control n=1,176) using ten morphometrics; we further integrated 5-Choice Serial Reaction Time Task (5CSRTT) data from MouseBytes (Female: AD n=394, Control n=1289; Male: AD n=598, Control n=1028) to examine sex-stratified behavioral outcomes. Except for the number of processes, all morphometrics differed significantly between AD and Control groups (Welch's t-test, $p < 0.0001$). Principal component analysis identified soma size and branching-based metrics as top contributors. Supervised models (random forest, gradient boosting; 5-fold CV) classified cells with 96% accuracy, while SHapley Additive exPlanations (SHAP) identified segments, soma radius, and segment length as key features. Gender stratification revealed stronger AD-related effects in females (~44% decrease in bifurcations and sections), while males showed smaller changes. Behavioral analysis revealed that males had larger differences in threshold accuracy and condition, while females showed greater reductions in perseverative correct responses (~34%). ANOVA revealed that group (AD vs. Control) explained most variance in morphology and behavior. These results indicate that AD alters microglial morphology and cognition in a sex-dependent manner, highlighting the importance of considering sex in AD research and potentially informing the development of targeted, sex-specific interventions.

T18. "Polyaromatic hydrocarbons inhibit neurite outgrowth by binding to the cell-surface laminin receptor", Bryce Blaugrund, University of Southern California, Faculty Advisor: Dr. Rayudu Gopalakrishna

This research focuses on diesel exhaust particles (DEPs), a type of air pollution made up of polyaromatic hydrocarbons (PAHs), such as benzo(a)pyrene (BaP), and their effects on neurite growth. Exposure to DEPs increases the risk of developing neurodegenerative and neurodevelopmental diseases, including Alzheimer's, Parkinson's and autism, but the mechanism

remains unknown.Hypothesis: PAHs, like BaP, bind with high affinity to cell-surface 67-kDa laminin receptors (67LR), inducing downstream generation of hydrogen peroxide, which inhibits neurite outgrowth and collapses already formed neurites.Model system: The hypothesis was tested using Neuroscreen-1 (NS-1) neuronal cells, which rapidly develop neurites in response to nerve growth factor (NGF). A 48-hour treatment with DEPs (2 μg/mL) and BaP (1-5 nM) was conducted, followed by measurement of neurite outgrowth using Metamorph software. These concentrations are environmentally relevant and induce hydrogen peroxide formation without causing cell death. Treatment was performed either concurrently with NGF to assess inhibition of neurite outgrowth or two days after NGF to determine whether DEPs could collapse already formed neurites. Molecular docking and binding studies with synthetic peptides were done. The role of 67LR in the cells was confirmed experimentally using a 67LR-blocking antibody.Results: DEPs, specifically PAHs, exhibited antineuritogenic effects at low concentrations through high-affinity interaction with 67LR. Fluorescence binding assays confirmed strong PAH binding to the peptide G palindrome, with hydrophobic residues critical for ligand interaction. Antioxidants, including N-acetyl-cysteine and glutathione ethyl ester, blocked DEP-induced effects, and the 67LR antibody reduced PAH-induced hydrogen peroxide generation, confirming that 67LR-mediated hydrogen peroxide production drives antineuritogenic activity.Conclusions: These findings demonstrate that DEPs and PAHs exert antineuritogenic effects at low concentrations through high-affinity interaction with 67LR. Binding to the same receptor sites as dietary polyphenols, such as those in green tea, suggests potential strategies to mitigate pollutant-related neurological disorders in the future.

T19. "The Effects of Low-Dose Naltrexone on Mice That Have Experienced a Traumatic Stressor", Chloe Sy Perez* and Owen Bella, University of California Los Angeles, Faculty Advisor: Catherine M. Cahill

Low-dose naltrexone (LDN) is increasingly used off-label to treat chronic pain, providing therapeutic benefits and improving quality of life despite being an opioid receptor antagonist. Previous findings demonstrated that stressed male rats showed conditioned place preference to LDN, implying that LDN is rewarding after experiencing a traumatic stressor. We hypothesize that stressed mice will spend more time in the LDN-paired context, while non-stressed mice will have no preference for either side. First, we conducted conditioned place preference (CPP), and the next day, mice received either 10 footshocks or none for 1 hour in a footshock box. On Days 2-3, conditioning training was conducted using a CPP apparatus, where one context was paired with saline and the other with naltrexone (0.1, 1, or 10 mg/kg, injected subcutaneously), respectively, on separate days. On Day 4, the mice had free exposure to the same CPP apparatus to determine their preferred side. A two-way ANOVA with dose x treatment as independent variables was used for analysis. We found that stressed male mice that received LDN 2 days after the traumatic stressor spent more time in the LDN-paired context than non-stressed mice, indicating LDN is rewarding in stressed males. Furthermore, we found that stressed females spent less time in the LDN-paired

context than non-stressed females, indicating that LDN is not rewarding in stressed females and suggesting a potential sex-dependent divergence in behavioral response. Our future directions are to investigate the mechanism by which LDN exerts its therapeutic effects. We plan to expand this project to transgenic mice to determine the system in which LDN acts. Overall, these results suggest that LDN produces a sex-dependent rewarding effect. Clinically, this slim therapeutic range can be refined to translationally address various conditions implicated in anxiety and stress.

T20. "Investigating the Effect of Noradrenergic μ-Opioid Receptors in Fear Learning via a Fear Contextualization Paradigm", Shifa Dhar* and Jeongha (Bryan) Kim, University of California Los Angeles, Faculty Advisor: Dr. Catherine Cahill

Chronic opioid use leads to hyperactivity of noradrenergic (NA) neurons in the brain, and release of NA contributes to physical withdrawal. Pilot data from our lab revealed that the lack of mu-opioid receptors (MOR) on NA neurons in mice potentiates withdrawal-induced aversion. While noradrenaline is known to strengthen emotional memory via amygdala synaptic plasticity, it remains unclear how MOR-regulated NA signaling can influence fear learning or how this system is perturbed during opioid dependence. This study aims to determine whether acute withdrawal following opioid-dependence enhances fear learning and to what extent MOR expression on NA neurons modulates fear learning. We hypothesize that opioid dependence will enhance fear learning, and this result will be augmented in mice lacking MOR on NA neurons. Opioid dependence was induced in C57B1/6 mice via escalating oral morphine self-administration (0.3 - 0.75 mg/ml) over 13 days; normal drinking water served as control. Male and female C57Bl/6 mice underwent contextual fear conditioning, which consists of fear acquisition (3 foot shocks), retrieval, generalization, extinction, and reinstatement. Freezing behavior was measured to quantify mice's fear response during each phase. Data (means +/- SEM) were analyzed via unpaired student t-tests or 2-way ANOVAs. In non-dependent mice, MOR-knockout mice exhibited freezing behavior comparable to MOR-intact mice during acquisition, retrieval, and generalization. On the 1st day of extinction, MOR-knockout mice displayed reduced fear extinction, though this was not statistically different to MOR-intact mice. MOR-knockout female mice exhibited signs of reduced fear extinction compared to males, suggesting a potential sex-dependent mechanism to fear extinction. While analyses of data from wildtype opioid-dependent mice are still being processed, we expect mice in acute withdrawal following opioid dependence to exhibit enhanced fear learning compared to non-opioid-dependent mice. Future studies will examine opioid-dependent MOR-knockout mice to assess withdrawal-induced modulations in fear contextualization.

T21. "Vagus Nerve Stimulation Reduces Reinstatement of Drug Seeking of Fentanyl in Rats", Dylan Cavalheiro, University of California Los Angeles, Faculty Advisor: Catherine M. Cahill

Fentanyl is a synthetic opioid that is commonly misused and is responsible for the majority of opioid-related deaths. While medication assisted treatments exist, many people relapse within a year of undergoing treatment for opioid use disorder. Recent studies have highlighted the potential of neuromodulation as a promising, non-pharmacological approach to treating substance use disorders. One of these neuromodulatory approaches is vagus nerve stimulation (VNS), which has shown benefits in pre-clinical models modulating brain circuits involved in reward, motivation, and stress. We recently developed a wireless, battery-less VNS device which has the potential to significantly improve clinical use of VNS devices. We hypothesize that VNS treatment would reduce stress-induced reinstatement of drug seeking. METHODS: Male adult Sprague Dawley rats underwent surgery to implant VNS devices and intravenous jugular vein catheters prior to operant training where rats learned to press a lever to receive intravenous fentanyl (2.5µg/kg/infusion). Rats self-administered fentanyl for 10 days prior to undergoing extinction training. During this extinction training, rats were divided into sham or bluetooth VNS stimulation. After 10 days of extinction training, all rats received a stress challenge (yohimbine, 2.5 mg/kg, i.p.) to induce reinstatement of the drug-seeking behavior. RESULTS: All rats learned to self-administered fentanyl. Rats also learned to distinguish between active lever (drug cue) vs the inactive lever (no consequence) within the first 2-3 days of the acquisition phase. All rats extinguished their drug-seeking behavior when fentanyl was not available, but there was no difference in responding between the VNS and sham groups. However, the VNS treatment significantly reduced reinstatement. CONCLUSIONS: These findings support VNS as a potential relapse-prevention therapy for opioid use disorder, and future research can include the variable of chronic pain and the translation into human trials.

T22. "Characterizing the Cell-Autonomous Role of CHCHD2 in Neural Stem Cell Fate Decisions during Adult Hippocampal Neurogenesis", Miguel Angel Sierra-Velazquez, University of North Carolina at Chapel Hill, Faculty Advisor: Dr. Juan Song

Adult Hippocampal Neurogenesis (AHN) is an exciting process wherein an endogenous pool of neural stem cells (NSCs) in the hippocampus makes fate decisions to become adult-born neurons. NSCs primarily rely on glycolysis and fatty acid oxidation. As they shift into active proliferation or differentiation, they increasingly depend on mitochondrial oxidative phosphorylation (OXPHOS). Coiled-coil-helix-coiled-coil-helix domain containing 2 (CHCHD2), a mitochondrial gene that enhances mitochondrial dynamics and OXPHOS, has emerged as a critical mediator of molecular pathways regulating AHN. It is highly expressed in NSCs, and global CHCHD2 knockout impairs AHN. Elucidating CHCHD2's necessity in AHN is key to advancing our understanding of NSC biology and identifying strategies to enhance neurogenesis.
A tamoxifen-inducible triple-transgenic mouse model (Ascl1CreERT2:CHCHD2^fl/fl:R26^tdTomato) was used to selectively excise CHCHD2 in active NSCs and trace their neurogenic lineage progression. Coronal dentate gyrus sections were

collected at neurogenic time points (7-, 14-, and 32-days post-injection) for immunohistochemistry and confocal imaging. Neurogenic lineage progression was assessed using cell-type-specific markers, while cellular morphology was quantified via radial process length, dendritic complexity, and dendritic spine density. While proliferation and immature neuron populations increased, the marked NSC and progenitor pools remained unchanged. CHCHD2 excision increased NSC activity but biased NSCs toward premature neuronal differentiation into immature neurons. CHCHD2-deficient cells exhibited shorter radial processes, reduced dendritic branching, and decreased spine density, indicating impaired morphological maturation as well. These observations indicate that CHCHD2 is necessary to maintain balanced NSC self-renewal and differentiation along with maintaining proper NSC neurogenic lineage progression.

T23. "Mechanisms by Which Polyphenols Inhibit Phosphodiesterases and Promote Neuroprotection", Anika Seshadri, University of Southern California, Faculty Advisor: Rayudu Gopalakrishna

Mechanisms by Which Polyphenols Inhibit Phosphodiesterases and Promote NeuroprotectionDietary polyphenolic compounds have been shown to elicit neuroprotective effects through interactions with the 67kDa laminin receptor, leading to downstream activation of cAMP signalling pathways. Notably, elevation of intracellular cAMP has been shown to inhibit β-amyloid-induced neuronal death through decreasing surface expression of 67LR and the cellular prion protein, which are necessary for β-amyloid uptake (Gopalakrishna et al., 2022). While polyphenols are known to be involved in this pathway, the mechanism remained unclear until recent evidence displayed that polyphenols may additionally modulate intracellular cAMP by competitively inhibiting the phosphodiesterase enzyme. In this study, we investigated if various polyphenols and their glucuronide conjugates can alter enzyme PDE4D or PDE4B activity under varied concentrations of cAMP. Using enzymatic kinetics assays on a panel of polyphenols, we observed isoform-specific inhibition of PDE4D by EGCG, which exhibited stronger potency at high concentrations. Additionally, molecular docking was completed for the polyphenols with PDE4D and other relevant enzymes to determine binding affinities and key amino acid residues present at catalytic and regulatory binding sites to make inferences about competitive or allosteric binding mechanisms. While these biochemical assays do demonstrate inhibitory potential, this mechanism is likely not a realistic possibility for neuroprotection as inhibitory polyphenol concentrations are significantly higher than physiological concentrations. Future directions point towards non-GPCR involved enzymes like soluble Adenylyl Cyclase to provide alternate pathways for intracellular cAMP increases. Elucidating these mechanisms contributes to understanding prevention of β-amyloid induced neuronal death and pioneering neuropharmacology solutions for Alzheimer's and Parkinson's disease.

T24. "CVLT Memory Scores are Predicted by MRI Regional Volumes, Neurite Density, and ERPs Across the Stages of Alzheimer's Disease (AD)", Nehal Shahi, University of California Davis, Faculty Advisor: Dr. John Olichney

The California Verbal Learning Test (CVLT) is a standardized memory test sensitive to memory decline in elderly individuals. Prior research has shown that CVLT memory assessments are useful in diagnosing amnestic mild cognitive impairment (MCI) and mild AD dementia (Silva, 2012). This collaborative study between the UCD and UCSD ADRCs aimed to detect the relationship between memory decline, brain structural changes, and synaptic dysfunction in elderly persons with varying degrees of cognitive impairment. We analyzed baseline event-related brain potential (ERP), MRI cortical gray matter volume, neurite density (ND) ratio, and CVLT memory data from 4 elderly groups: normal old (NO, N=21), pre-clinical AD (pre-AD, N=23), MCI (N=11), and AD dementia (N=7). After conducting multiple linear regression models of CVLT memory scores, we found that ERPs, ND, and MRI measures were each significant predictors. The ND ratio and MRI measures of the entorhinal cortex and MRI hippocampal gray matter volume were consistent predictors of CVLT memory scores. Additional variance in learning (List A) and long delay free recall was explained by the Late Positive Component amplitude (300-550 ms, at Pz). These findings show that multiple medial temporal lobe structures, known to be affected near the onset of AD, are critical for memory and supports how atrophy of brain macrostructures, microstructural changes, and electrophysiology each have additive effects in predicting verbal memory performance.

Ecology and Evolution

T25. "The Genetic Architecture of Wisdom: A Genome-Wide Association Study in 131,870 Individuals", Fabrizio Malatesta, University of California San Diego, Faculty Advisor: Dr. Sandra Sanchez-Roige

Wisdom is a personality trait composed of self-reflection, pro-social behaviors, emotional regulation, acceptance of diverse perspectives, decisiveness, social advising, and spirituality. Greater levels of wisdom are linked to better overall health, well-being, happiness, life satisfaction, and resilience. However, the underlying genetic architecture of wisdom and its genetic relationships with health remain unexplored. We collected responses to SD-WISE-7, a validated survey measure of wisdom, in 131,870 research participants and conducted the first large-scale genome-wide association study (GWAS) of this trait. We identified one genome-wide significant locus (rs1028640, P=3.7e-08) near the MANEA gene on chromosome 6. Wisdom was heritable (h2SNP=0.03±0.004) and genetically correlated with numerous complex traits, including subjective wellbeing (rg=0.34, P=1.52e-09), neuroticism (rg=-0.38, P=1.87e-16), loneliness (rg=-0.27, P=8.45e-07), other personality traits such as openness (rg=0.41, P=1.37e-07) and risk tolerance (rg=0.36, P=6.94e-11). Crucially, wisdom was genetically distinct from intelligence

(rg=0.036, P=0.39) and cognitive educational attainment (rg=-0.01, P=0.83). We calculated polygenic scores (PGS) in All of Us (N≈200,000) and identified significant associations with psychosocial outcomes (e.g., social satisfaction ratings P=1.16e-12 and subjective mental health P=5.93e-10). This study advances our understanding of wisdom as a biologically-grounded trait that could ultimately be used to develop informed interventions that increase wisdom and improve health and well-being.

T26. "Prevalence of Haemosporidian lineages in a resident songbird across urban-rural habitats", Anneliese Roth, Fresno State University

Urbanization can affect ecological dynamics and wildlife health, especially in parasite-host interactions. House finches (Haemorhous mexicanus) inhabit urban, suburban, and rural environments and are susceptible to vector-borne haemosporidian parasites (Plasmodium and Haemoproteus). In our study system, blood smear analysis revealed that house finches in suburban and rural habitats have higher haemosporidian prevalence and parasitemia than urban habitats, but the distribution of specific parasite variants remains unknown. We aim to genetically characterize haemosporidian lineages along three categories of urbanization. We hypothesize that haemosporidian diversity will differ by habitat, with distinct variants emerging in urban, suburban, and rural areas. Specifically, we will amplify the Cyt-b gene from roughly 87 positive samples using nested polymerase chain reaction (PCR), run in triplicate to ensure reliability. Sequencing is currently underway. We will check sequence quality with Geneious and identify variants with BLASTn, followed by haplotype networks and statistical analyses in R. We generated a TCS haplotype network which showed the genetic relationships among parasite lineages across habitats. We found predominantly Haemoproteus lineages, with the highest in the suburban habitats. Plasmodium lineages were predominately in urban habitats. Ultimately, this research will offer insight into whether urbanization shapes the distribution and diversity of avian haemosporidian parasites and to better understand how environmental change influences parasite lineage distribution. This project helps conservation efforts and may help guide urban planning to promote biodiversity and contribute to understanding how parasite-host dynamics are altered at the human-wildlife interface.

T27. "Global Activity Patterns and Habitat Use of Tapirs (*Tapirus spp.*) as Revealed by Large-Scale Camera Trap Data", Elise Kinney and Sophia Peck*, Point Loma Nazarene University, Faculty Advisor: Dr. Michael Mooring

The four species of tapirs – Malayan (Tapirus indicus), Baird's (T. bairdii), Lowland (T. terrestris), and Mountain (T. pinchaque) – are some of the last remaining megafauna species and play an important ecological role as forest gardeners and keystone species in the Old and New World. Because all populations are declining and threatened with extinction, it is important to understand the factors influencing their habitat use and behavior. We hypothesized that each tapir species

would possess different adaptations based on their distinct ecological setting. To this end, we employed a collaborative approach to examine the global Tapirus genus by using >110,000 days of camera-trap data to characterize species-typical activity patterns and habitat use. Activity pattern was analyzed for circadian (24-hour) and lunar (28-day) cycles using 'Overlap' analysis with the 'sunTime' function employed to account for seasonal changes in sunrise/sunset. Additionally, single-season Occupancy models were run in a Bayesian framework with anthropogenic and environmental covariates. Malayan, Baird's, and Lowland tapirs exhibited a nocturnal circadian activity pattern (82-88% activity at night) with activity focused between sunset and sunrise. However, Mountain tapir exhibited a cathemeral activity pattern, being active throughout the day and only 63% of activity at night. The greater daytime activity of Mountain tapir may reflect their colder, higher elevation habitat and their smaller size and thermal capacity, as indicated by their longer fur. However, Mountain tapir diurnal activity makes them more vulnerable to human threats such as hunting and road kills. For all species, nocturnal activity declined during full moon illumination, indicating a lunarphobic response. These results provide important insights to better predict future tapir behavior in time and space under climate change and contribute to conservation decisions and protection for Tapirus.

T28. "Conservation Genomics of an Endangered Succulent in Santa Clara County", Karina Martinez* and Evan Hackstadt, Santa Clara University, Faculty Advisor: Dr. Justen Whittall

Santa Clara Valley Dudleya (Dudleya setchellii) is a federally-listed endangered perennial succulent from southern Santa Clara County. It is restricted to fewer than 20 occurrences on serpentine rocky outcrops in southern Santa Clara County, CA. Therefore, it is critical to understand the distribution of genetic diversity across the landscape, determine the extent of clonal reproduction, and carefully examine its taxonomic classification and the potential for hybridization with closely related species. Previously, we extracted DNA from 833 plant samples and prepared 402 DNA libraries for Illumina short-read sequencing. To analyze genetic diversity in D. setchellii, whole-genome resequencing was conducted leveraging a draft reference genome of ~247 million base pairs. Sequencing data were processed using Santa Clara University's high-performance computing (HPC) cluster, where a bioinformatics workflow was developed that converts raw sequencing reads into population genomic data for conservation genetic analyses. The workflow includes next-generation sequencing analysis steps using Fastp for read filtering, BWA for sequence alignment, SAMtools for alignment processing, ANGSD for genotype likelihoods, and PCAngsd, ngsDist, and FastME to produce results. Genome sequencing data were generated from the sampled individuals with 1-15x coverage. Examining the genetic distances reveals significant isolation by distance within populations (28% of the genetic variation explained by geographic distance) and among populations (42%). Additionally, 12.5-21.9% of samples within a single focal population showed evidence of clonal reproduction at very fine geographic distances (3-18cm). Phylogenetic analyses indicate populations of D. setchellii are monophyletic, but suggest potential hybridization with D. cymosa when growing nearby. Finally, we have very strong evidence against

D. setchellii being classified as a subspecies of D. abramsii. These conservation genomic results indicate that although clonality is limited to fine scales, there are strong genetic limits to dispersal, which can inform conservation management of this endangered species.

T29. "Protocol Development for Enhanced DNA Visualization in the California Bay Laurel", Jillian Gonzalez* and Claudia Tellez*, Sonoma State University, Faculty Advisor: Dr. Lisa Hua

California native plants provide a promising alternative to traditional model systems to study chromosome organization. While observational field studies exist for detecting and quarantining the pathogen Phytophthora ramorum (P. ramorum), there is a lack of molecular tools to understand the pathogen and its foliar host, the California Bay Laurel (Umbellularia californica). P. ramorum is an invasive oomycete eukaryotic microorganism that is a pathogenic agent for the disease, Sudden Oak Death (SOD). This pathogenic agent is a cause for concern as it is responsible for the death and dramatic decline in coastal tanoak tree populations. In coastal rural regions, P. ramorum is a risk against the ongoing efforts of preservation of biodiversity, as 14% of Sonoma County has been affected by SOD. Observing chromosomes in vivo of U. californica is needed to develop molecular protocols for non-traditional host organisms. Our project aims to refine a histological DNA staining protocol for staining U. californica's chromosomes. This will provide the basis for future work to investigate the effects of P. ramorum on chromosomal organization in normal and infected states of the Bay Laurel. By using the Bay Laurel as a new model system to study native host-pathogen interactions, it will allow us to unravel the molecular mechanisms of infection and develop strategies to protect California's threatened coastal forest populations from SOD.

T30. "The impact of parasitism: social distancing in a species from a lineage of gregarious insects", Ariana Thompson* and Maya Richter, Santa Clara University, Faculty Advisor: Dr. Janice Edgerly-Rooks

Parasites often modify host behavior to aid their own transmission. Alternatively, some hosts evolved avoidance behaviors to limit risking infection. This study focuses on insects of the order Embioptera that are parasitized by a gregarine. Previous reports demonstrated that the focal species displayed antisocial behaviors, unusual for embiopterans, but parasitism status was unknown. Here, we ask whether the insects are repelled when encountering parasitized opponents or has antisocial behavior evolved as a behavioral syndrome because of negative pressure exerted by past parasitism. Field-collected individuals of two related sympatric species (both susceptible to the gregarine) were challenged in dyadic interactions of conspecifics or heterospecifics. Parasite presence was confirmed by PCR following behavioral data collection. Acts ranged from gregariousness (e.g. sitting together) to antisocial (e.g. running away when touched). Haploembia tarsalis, displaying a high parasitism rate (54% infection rate vs 14% for H. solieri), consistently displayed antisocial behaviors—parasitized or not. When paired with conspecifics, H. solieri, more resistant to infection, was moderately gregarious—spinning silk

and sitting together. In contrast, they displayed negative reactions to H. tarsalis, including bolting away and attempting to escape the arena. Previous research suggested that the sterilizing effect of the gregarine on male H. solieri likely drove the evolution of the parthenogenetic H. tarsalis lineage. The asexual females, appearing to lack an effective immune response to the parasite, may have evolved social distancing, a generalized strategy to prevent transmission among highly susceptible individuals.

T31. "Going up, tuning down: Tympanum size variation in *Rana boyii*", Daniella Mori* and Aziza Corley, University of San Francisco, Faculty Advisor: Dr. Jennifer Dever

In California, the foothill yellow-legged frog (Rana boylii) is a widely distributed endemic species that has experienced significant declines over the past 50 years for a variety of reasons (Kupferburg 2012; Adams et al. 2017). Limited to rivers and streams, the historical range of this species encompasses a variety of habitats, including differences in temperature, vegetation, stream hydrology, and elevation (Lind 2004). Acoustic communication is closely tied to frog behavior, yet how elevation influences this trait remains poorly understood. Liao and Liu (2008) conducted a literature review testing the relationship between tympanum size and altitude across a wide variety of frog taxa, finding that anuran acoustic organs (vocal sacs and tympana) decline and degenerate as altitude increases. To determine whether a similar relationship exists in R. boylii, we examined museum specimens from the California Academy of Sciences and the Museum of Vertebrate Zoology, collected from two distinct elevational zones: low-elevation (0–300 m) and high-elevation (700–1,000 m). Tympanum and body length measurements were taken from 66 male specimens. We hypothesized that due to very different ecological pressures associated with elevational differences, tympanum size correlates with elevation. We found that frogs in the high-elevation zone have a mean tympanum diameter smaller than that of those collected from the low-elevation zone. Results from the Mann-Whitney U test and the T-Test indicate a highly significant difference between these two groups. These findings suggest that the elevation gradient may be a driving force behind variation in tympanic morphology in R. boylii.

Physiology and Developmental Biology

T32. "Spatial organization of homologous chromosomes 2 and 16 in mouse embryonic fibroblasts", Omoefe Odiase, Sonoma State University, Faculty Advisor: Dr. Lisa Hua

A non-sex cell contains two copies of each chromosome, one from each parent. These homologous chromosomes in human cells separate to opposite sides of the cell. This organization may function to prevent abnormal exchange of genetic material, which has been linked to genomic instability. To test whether other mammalian models also maintain this spatial segregation of homologous chromosomes, we selected mouse fibroblast cells, as mice share approximately 90% genetic similarity to humans. The spatial segregation of chromosomes 17, X and Y in mouse cells were

previously established, suggesting a shared chromosome organization pattern among mice and humans. Our preliminary data of mouse homologous chromosomes show a lack of spatial segregation of chromosomes 1, 2, 17, 19, and X. These results suggest that spatial segregation may be lost. These findings will determine whether spatial segregation is conserved in mice, providing new information on mouse model usage in human biology.

T33. "Voltage-gated Sodium Channel Nav1.1 is Necessary for Muscle Spindle Afferent Function in Adult Mice", Mariam Malik, San Jose State University, Faculty Advisor: Dr. Katherine A. Wilkinson

The sense of body and limb position, or proprioception, is necessary for producing coordinated and balanced movements. Muscle spindle (MS) afferents are responsible for transducing and transmitting muscle stretch information that is necessary for proprioception. The detection and transmission of muscle length information requires multiple isoforms of the voltage-gated sodium channel (NaV). Mutations to genes encoding Nav1.1 or Nav1.6 lead to motor movement disorders, and previous studies in our lab have shown that loss of Nav1.1 from birth results in a reduction in the ability of MS afferents to faithfully encode static stretch length. We hypothesized that Nav1.1 is still necessary for proper function after the MS has developed in adult mice. In order to test our hypothesis, we used a viral injection strategy to knock out the gene encoding Nav1.1, Scn1a. To target proprioceptors, we crossed mice expressing Cre in parvalbumin positive neurons (Jax#017320) with mice expressing Cre-dependent Cas9 under the Rosa 26 locus (Jax#026175). Two retroorbital injections one day apart of either a virus with sgRNAs targeting the Scn1a gene (AAV.PHP.s-sgNa V 1.1-FLEX-TdTomato; Nav1.1 cKO) or a control virus (AAV.PHP.s-sgLacZ-FLEX-TdTomatoa; Nav1.1 WT) were given to mice 21 to 25 days old. We waited until mice were at least 2 months old and then extracted the extensor digitorum muscle with the sciatic nerve and placed it in a tissue bath. An extracellular glass electrode was used to record stretch-sensitive signals and the muscle was stretched to three physiological lengths (2.5, 5, and 7.5% of optimal length). We were able to find stretch sensitive firing from MS afferents in 8 of 9 WT mice but only 4 of 8 cKO mice ($\chi^2(1) = 3.09$, $p = 0.08$). Similarly, all WT animals except 1 exhibited sustained firing over the entire 4 s stretch at all stretch lengths. In contrast, only 1 of 4 cKO mice had at least one afferent that could fire throughout the entire stretch ($\chi^2(1) = 4.69$, $p = 0.03$). This suggests that Nav1.1 is essential for MS afferent static stretch activity in adulthood. Future directions include increasing the sample size to have sufficient statistical power to compare whether the deficit is more severe with loss of Nav1.1 after development. If so, this would suggest that some compensation by the other Nav's is possible if Nav1.1 is lost throughout development.

T34. "Investigating the Role of Developmental Signaling Pathways in Early Macrophage Route Choice During Drosophila Embryogenesis", Steve Chung, University of California Los Angeles, Faculty Advisor: Dr. Daria Siekhaus

During embryogenesis, macrophages disseminate through complex 3D tissue environments to colonize developing tissues. Our lab showed that early macrophage dispersal is driven in part by a "leader" macrophage subpopulation responding to Bone Morphogenetic Protein (BMP), a TGF-β subgroup, to promote germband invasion by guiding "follower" macrophages along this defined invasive route. scRNA-sequencing also revealed that these BMP-responsive macrophages upregulate key regulators of developmental pathways, including EGF, Notch, Hippo, FGF, and Wnt. However, it remains unclear whether these pathways directly influence the earliest migratory decisions of macrophages during Drosophila embryogenesis. To identify which pathways regulate early migration, the GAL4-UAS system was used by crossing UAS-RNAi knockdown lines targeting developmental receptors in macrophages with a macrophage-specific GAL4 driver and marker line. Fixed embryos were imaged using confocal microscopy and analyzed with Imaris to detect changes in macrophage distribution against controls at the same developmental stage. Here, we found that macrophage-specific Wnt, FGF, and Hippo pathway receptor knockdowns affected their migration, while EGF and Notch pathway receptor knockdowns did not. Preliminary spatial analysis revealed that macrophages in Frizzled 2 (Wnt), Heartless (FGF), and Fat (Hippo) Receptor knockdown embryos experienced greater posterior-migration bias compared to controls. Additionally, the Fat Receptor knockdown caused a higher macrophage population and loss of anterior migration. However, the Breathless (FGF) Receptor knockdown caused a loss in both anterior and posterior distribution. These preliminary findings suggest that developmental pathways conserved between Drosophila and vertebrates may regulate early macrophage migratory behavior during embryogenesis. Ongoing analysis and future experiments, including knocking down partner ligands and performing live embryo imaging, will reveal how these knockdowns influence macrophage migration speed, persistence, and directionality. Ultimately, this work will yield a better understanding of the mechanisms influencing the earliest macrophage migratory decisions and provide invaluable insight to support future therapeutic strategies targeting pathological tissues and tumors.

T35. "Dynamics of Gastrulation in *Trachemys scripta* Turtle: Correlating Changing Blastopore Shapes with the Progression of Tissue Internalization", Lila Le, Loyola Marymount University, Faculty Advisor: Dr. Max Ezin

During the crucial process of gastrulation, specific tissues internalize, and germ layers differentiate, setting the foundations of tissue development and organization. Amphibians, reptiles, and birds undergo gastrulation through varying mechanisms. While amphibians gastrulate via involution and birds gastrulate by ingression, reptiles are an evolutionary intermediate, internalizing tissue in both ways. In reptiles, while ingression occurs at the posterior region of the embryo, on the dorsal side, involution occurs at the blastopore lip. As reptilian embryos gastrulate, the morphology of the blastopore lip changes, following 3 main blastopore shapes: upturned, flat, and downturned. The blastopore lip shape can be used as an external marker to evaluate the stage of gastrulation (i.e. early, middle, or late) the embryo has reached. However, there is yet to be a visual sequential correlation of internal, involuting ectodermal tissue with changing blastopore

shapes during gastrulation. We aim to advance reptilian gastrulation understanding by correlating the changes in blastopore shapes with the progression of tissue internalization in turtles, using Trachemys scripta turtle as a model organism. Turtle eggs were collected and incubated for 0-2 days post oviposition, at 6-hour time intervals. Gastrulae were harvested, cryosectioned, and stained with hematoxylin and eosin to visualize involuted tissue. We show that turtle blastopore lips begin upturned, flatten into a thin line after maturing 12-18 hours, and by 24 hours fully invert to a downturned curve. Furthermore, as the blastopore lip shape changed from upturned, to flat, to downturned, we found the following progression of internalized tissue respectively: partial, halfway, and complete. Our results help to further the understanding of chelonian gastrulation and may inform on the evolution of gastrulation mechanisms between amphibians and birds, with reptiles as a key transition.

T36. "Psilocin Alters Craniofacial Proportions and Accelerates Ossification in Developing Chick Embryos", Sarmad Alani, Loyola Marymount University, Faculty Advisor: Dr. Maxellende Ezin

Background Psilocybin has gained increasing medical attention and has received Breakthrough Therapy Designation for depression by the U.S. Food and Drug Administration (FDA). After administration, psilocybin is rapidly converted to psilocin, the active metabolite that acts through serotonergic signaling. While psilocin is being investigated for therapeutic use in adults, little is known about how it may affect embryonic development. Serotonin signaling regulates multiple embryonic processes, but its specific role in shaping neural crest-derived facial skeleton remains incompletely understood. Here, we treat embryos with psilocin to determine the drug's effects on craniofacial morphology and ossification. Methods Chick embryos were exposed to psilocin (10, 20, or 100 μM) or Ringer's saline as a control on embryonic Day 1 and collected at Day 10, a critical stage of craniofacial skeletal development. Following collection, embryos were stained with Alcian Blue and Alizarin Red to visualize cartilage and mineralized bone, respectively. Morphometric measurements were obtained using FIJI. Results Craniofacial morphometric analysis showed that psilocin treatment altered facial proportions relative to controls. Embryos treated with 100 μM PSI showed a significant increase in head length as a percentage of total embryo length compared to controls, indicating altered craniofacial proportions. Alizarin Red analysis further suggested that 100 μM PSI treatment was associated with accelerated ossification in the craniofacial region, which includes the mandible and maxilla. Similar increases were observed in long bones such as the humerus and femur. Conclusion Psilocin-mediated disruption of serotonin signaling may influence embryonic skeletal development by altering craniofacial form and accelerating ossification. Future work will involve staining and measuring Day 10 embryos treated with 10 μM and 20 μM psilocin to assess dose-dependent effects on craniofacial morphology and ossification.

T37. "NO TITLE", Sarah Jean Valliant* and Ileana Jade Rodriguez, San Francisco State University

Unsheltered individuals experience disproportionately high rates of cardiovascular disease (CVD) yet remain underrepresented in formal screening programs. Community outreach organizations frequently provide services but lack standardized, ethical, and replicable frameworks for physiological risk assessment in field settings. This public health survey evaluated cardiovascular risk factors and developed a low-resource screening model adaptable to mobile outreach environments.Objective: To describe the development and implementation of an ethical, field-based cardiovascular screening methodology emphasizing participant safety, consent, and operational feasibility within homeless outreach programs.Methods: A cross-sectional, descriptive operational needs assessment was conducted during mobile outreach missions in San Francisco's Inner Mission, Tenderloin, and Embarcadero neighborhoods (November 2024–October 2025) using convenience sampling. Ethical oversight was provided by the Valliant Foundation Ethics Committee, which determined the project to be Non-Human Subjects Research under 45 CFR 46.102. The screening framework integrated volunteer training, cognitive capacity assessment, verbal consent, vital sign collection, and emergency escalation protocols. Eligible participants were unsheltered adults meeting A&O×4 capacity criteria who voluntarily consented. Data collected included demographics, self-reported cardiovascular risk factors, and point-of-care measures (blood pressure, heart rate, blood glucose). Analyses were descriptive.Results: Emergency protocols were activated in three cases (two hypoglycemia events, one suicidal ideation requiring EMS transport). All analyzed data met verified capacity and consent standards (100%). Nineteen readings were excluded due to missing or implausible values, prompting protocol refinements including dual blood pressure verification. Referrals and educational materials were tailored to the specific population demographics. Conclusion: This study presents a replicable, ethically grounded framework for cardiovascular risk screening in homeless outreach settings. The model enables nonprofit and public health teams to integrate evidence-based physiological assessment into low-resource humanitarian fieldwork.

Biochemistry, Biophysics and Microbiology

T38. "Concentration-Dependent Emergent Cooperativity in Myoglobin Pseudoperoxidase Activity Under Oxidative Stress", Kade Sutherland, Utah Valley University, Faculty Advisor: Dr. Daniel Scott

Enzymes involved in oxidative stress play critical roles in the progression of cardiovascular and inflammatory diseases. Myoglobin, classically recognized for oxygen storage in muscle, also exhibits pseudoperoxidase activity through its heme prosthetic group in the presence of hydrogen peroxide. Previous work has demonstrated substrate inhibition at elevated peroxide concentrations,

consistent with oxidative inactivation of the heme center. In the present study, we investigated how varying myoglobin concentration—while holding hydrogen peroxide constant—affects catalytic behavior under oxidative conditions.Steady-state kinetic assays were performed by increasing myoglobin concentration at fixed hydrogen peroxide levels and monitoring initial reaction velocities spectrophotometrically. At lower peroxide conditions, reaction rates increased modestly and approximately proportionally with increasing myoglobin concentration. In contrast, at higher peroxide levels, increasing myoglobin concentration produced a sharp, sigmoidal transition from minimal activity to rapid acceleration before approaching saturation. This cooperative-like response was unexpected given the monomeric structure of myoglobin. Comparative experiments with Hemoglobin showed a more gradual concentration-dependent increase in activity consistent with its established allosteric properties.Because hydrogen peroxide concentration was held constant, the observed sigmoidal behavior suggests that enzyme concentration itself modulates catalytic efficiency under oxidative stress. We propose that concentration-dependent intermolecular interactions, transient polymerization, or protection from rapid oxidative inactivation at higher protein levels contribute to this emergent kinetic behavior. These findings indicate that myoglobin can display cooperative-like activity in highly oxidative environments, expanding understanding of its redox chemistry and highlighting the importance of enzyme concentration in shaping catalytic responses relevant to disease-associated oxidative stress.

T39. "NanoBRET-based Assay for Monitoring Protein Palmitoylation", Andrew Medina* and Dylan Karkainen, University of Southern California, Faculty Advisor: Dr. Chao Zhang

Protein palmitoylation is a reversible post-translational modification that regulates trafficking, protein-protein interactions, and signaling. However, palmitoylation is still most commonly assayed using gel-based workflows which are low-throughput, semi-quantitative, and labor-intensive, thus limiting mechanistic studies and screening efforts.ObjectivesWe aim to develop a rapid, high-throughput, and quantitative assay for monitoring protein palmitoylation using bioluminescent resonance energy transfer (BRET).MethodsOur strategy uses a chlorinated analog of palmitic acid that can be incorporated into palmitoylated proteins and subsequently undergo covalent capture by a HaloTag protein. HaloTag is labelled with a BODIPY to serve as the NanoBRET acceptor, enabling plate-based detection of palmitoylation through energy transfer from a NanoLuc-tagged target protein.ResultsWe synthesized a 15-carbon palmitic acid analog bearing a terminal chloride and confirmed its structure by 1H and 13C NMR. HaloTag was expressed in E. coli (BL21) and purified by Ni-NTA chromatography, yielding a single band by SDS-PAGE, and was then successfully conjugated to a Py-BODIPY-NHS ester as verified by in-gel fluorescence. Ongoing cell-based experiments in HEK293T cells overexpressing HRAS will assess incorporation of the palmitate analog and quantify palmitoylation via NanoBRET using a microplate reader. ConclusionThese preliminary results establish feasibility of a fluorogenic, plate-based NanoBRET platform for detecting protein palmitoylation. Once fully validated, this assay should provide a streamlined method to quantify palmitoylated HRAS and other targets, with

potential for high-throughput screening for chemical modulators of protein palmitoylation for therapeutic discovery.

T40. "Identification of potential antibiotic compounds for Gram-positive bacteria via optimization of a high-throughput FRET assay", Steven Nguyen, Creighton University

Antibiotic resistance is an increasing threat that necessitates the discovery and identification of novel antibiotics with new molecular targets. Riboswitches are non-coding segments of messenger RNAs (mRNAs) that bind cellular metabolites to induce conformational changes and alter gene expression. Riboswitches control essential metabolic pathways in many bacteria. The glmS riboswitch/ribozyme is present in many Gram-positive bacteria, including the food-borne pathogen Bacillus cereus and the infectious bacteria Staphylococcus aureus. The glmS mRNA is essential as it expresses the enzyme that synthesizes glucosamine-6-phosphate (GlcN6P), a key component in cell wall synthesis. When GlcN6P, the natural ligand, is bound to the glmS RNA, it downregulates gene expression by inducing self-cleavage of the RNA. The glmS riboswitch is mechanistically unique as it is the only known riboswitch for which catalytic activity provides the basis of genetic regulation, and it is the only known ribozyme that depends upon a "coenzyme", the metabolite GlcN6P, to actuate self-cleavage. The function of this ribozyme makes it a clear target for the development of novel antibiotics, possibly utilizing artificial analogs of GlcN6P. In order to evaluate such analogs, we have focused on optimizing a high-throughput fluorescence resonance energy transfer (FRET) assay to test various ligands and libraries of compounds as possible antibiotics against Gram-positive bacteria by targeting the glmS ribozyme. The glmS ribozyme from Bacillus cereus and Staphylococcus aureus has been studied with this method. The goal of this project is to develop the most efficient high-throughput assay in order to evaluate a library of GlcN6P analogs that could potentially be used as antibiotics against B. cereus, S. aureus, and other pathogenic bacteria expressing the glmS ribozyme.

T41. "Structural analysis of *Crassostrea gigas* OAZ-PK RNA via Selective 2'-Hydroxyl Acylation analyzed by Primer Extension (SHAPE)", Daniel Cline, Creighton University, Faculty Advisor: Juliane Strauss-Soukup

Riboswitches are sequences of non-coding RNA present in many bacteria, archaea, plants, and fungi. When a riboswitch binds to its ligand, the RNA undergoes a conformational change, causing modulation in downstream gene expression. Currently, only one riboswitch has been discovered in eukaryotes, however the Ornithine Decarboxylase Antizyme Pseudoknot (OAZ-PK) RNA has been identified as potentially the second eukaryotic riboswitch. Ornithine Decarboxylase Antizyme inhibits synthesis of polyamines, organic molecules aiding in cell growth and proliferation. Identification of this sequence as a riboswitch will allow for novel drug development to treat diseases such as cancer, where overproduction of polyamines has been observed. Using the technique Selective 2´-Hydroxyl Acylation analyzed by Primer Extension (SHAPE), I investigate

structural changes in the OAZ-PK RNA from Crassostrea gigas, a species of oyster, upon binding to natural and synthetic polyamines. The goal of this project is to determine the overall secondary structural changes caused when spermine, the natural ligand for this potential OAZ-PK riboswitch, as well as its analogs, bind to the OAZ-PK RNA. Determination of structural changes will aid in identifying this sequence as a novel eukaryotic riboswitch and will shine light on the potential for eukaryotic riboswitch targeted drug development.

T42. "Phosphofructokinase mutations in Glycogen Storage Disease Type VII lower ligand affinity and disrupt oligomer assemblies", Emily Li, University of Washington, Faculty Advisor: Dr. Justin Kollman

Glycogen Storage Disease VII (GSDVII) is an autosomal recessive genetic disorder that results in multiple muscle symptoms. GSDVII is caused by a deficiency in muscle phosphofructokinase (PFKM), which catalyzes the rate limiting step in glycolysis. PFKM is a tightly regulated, pH-sensitive homotetrameric enzyme that can form higher-order oligomers. The function of these higher order assemblies as it relates to enzyme activity and regulation is unclear. While PFKM deficiency has been implicated in GSDVII, the nature of the deficiency is heterogeneous and several missense mutations in PFKM are associated with GSDVII. We've recently determined a cryo-EM structure of PFKM and found that many of GSDVII mutations occur at or near regulatory hotspots and tetramer interaction sites, suggesting that mechanisms other than disruption of catalytic chemistry contribute to PFKM deficiency in GSDVII. Here, I expressed and purified several naturally occurring disease-associated variants of PFKM in regulatory hotspots and the [JK1] oligomer assembly site. I used negative stain electron microscopy and mass photometry to understand the consequences of these variants on PFKM structure and oligomeric state. These disease variants are structurally intact at the level of functional tetramers but have disrupted higher-order structures. Furthermore, I characterized the catalytic activity of these disease variants using a coupled enzyme assay and examined the influence of pH on regulation. These assays suggest that all disease-causing variants tested have a decreased overall activity[JK2] , but only two have a lower affinity for substrate. One site with reduced affinity is located near an ATP binding regulatory site suggesting that binding in one site may influence the subsequent binding of ligands in other regulatory sites. A variant with reduced substrate affinity is involved in the assembly of oligomers and not in catalytic or regulatory sites, suggesting that higher-order assemblies have a role in increasing PFKM substrate affinity . These findings provide mechanistic insight into PFKM deficiency.

T43. "Color With a Cost: Metabolic Dyes TTC and INT Compromise *Bacillus subtilis* Survival at Common Assay Concentrations", Sumaiya Tohorat, South Dakota State University, Faculty Advisor: Dr. Nicholas C Butzin

Tetrazolium dyes such as triphenyl tetrazolium chloride (TTC) and iodonitrotetrazolium chloride (INT) are often used to measure bacterial metabolic activity through reduction to colored formazans in active cells. Standard assay concentrations in liquid media are ~0.2-0.4 mg/mL for TTC and ~0.1 mg/mL for INT. Despite widespread use, the potential inhibitory effects of these dyes on bacterial physiology are often overlooked, so we tested these effects in Bacillus subtilis using both liquid and solid growth assays. For liquid assays, we monitored growth kinetics in LB broth with varying TTC and INT concentrations using 96-well plates, comparing dye-treated wells with dye-free controls to identify levels that do not alter bacterial growth or physiology. On solid LB agar, disc diffusion and plate assays were conducted to evaluate dye toxicity and visible metabolic color development. In both liquid and solid media, commonly reported dye concentrations inhibited growth and killed substantial fractions of the population. Cells tolerated higher dye levels on solid media; however, concentrations sufficient to generate visible colored colonies strongly inhibited growth and caused extensive cell death in most, but not all, cells. Because both dyes are toxic to B. subtilis at typical assay concentrations, colorimetric results may reflect metabolic stress rather than true metabolic activity, which is particularly relevant for physiological assays such as antimicrobial studies. Thus, the very dyes used to measure metabolism may themselves alter metabolic activity, and conclusions drawn from these assays should be interpreted with caution. Whole-genome sequencing showed that survival is not due to mutation, suggesting a likely epigenetic mechanism. Future work will focus on identifying this mechanism through transcriptional profiling and targeted mutagenesis.

T44. “Integrating Bioinformatics and Synthetic Biology to Engineer Hypoxia-Resilient Cell Therapies in Jurkat T Cell Models”, Raymond Nova*, Shardul Singh and Janice Subroto, University of California Los Angeles, Faculty Advisor: Dr. Chase Linsley

Chimeric antigen receptor-based T (CAR-T) cell therapies have demonstrated remarkable successes in hematologic malignancies but are still largely ineffective against solid tumors. This disparity is driven, in part, by the dense, hypoxic extracellular matrix (ECM) of solid tumor cancers which impairs T cell metabolism and infiltration. Conversely, identifying gene targets that improve hypoxia resilience while preserving cell function remains a significant challenge due to the intricate crosstalk of various biological pathways. Therefore, we propose an integrated computational and cellular engineering approach to identify and enhance key pathways which regulate T cell survival in hypoxic conditions. Using machine learning models, including linear regression, LASSO, ridge regression, elastic net and random forest, we first identified candidate pathways associated with improved cellular fitness under hypoxic conditions. Our preliminary findings motivated us to investigate the role of peroxisome proliferator-activated receptor gamma coactivator-1 alpha (PGC-1a) on T cell function within hypoxic culture. As a transcriptional coactivator that regulates mitochondrial biogenesis and metabolism, previous studies have shown that enhanced PGC-1a expression in CD8+ OT-1 T cells promote metabolic function and antitumor efficacy against B16-OVA melanoma. Therefore, we sought to explore this mechanism and

determine whether its expression correlates to amplified hypoxic resilience without disrupting tumor-killing efficiency. To promote T cell infiltration, we are also studying expression of heparanase: an enzyme which degrades heparan sulfate proteoglycans (HSPGs)— a major component of the ECM. We aim to upregulate the expression of heparanase by inducing a loss-of-function mutation in the TP53 gene in our Jurkat model. This will effectively neutralize the effectiveness of p53, a transcription factor which inhibits HPSE expression and heparanase synthesis. As such, we aim to prolong the persistence of T cells while simultaneously upregulating HPSE expression, thereby facilitating the degradation of the ECM.

Cell and Molecular Biology

T45. "Assessing the Contribution of Multiciliary Arrays to Unidirectional Fluid Flow", Tyler Kang*, Santa Clara University, Faculty Advisor: Dr. Brian Bayless

Motile cilia are extracellular appendages that beat in a whip-like motion to direct fluid flow in the body. Defects in motile cilia can lead to serious human health issues, such as respiratory diseases, hydrocephalus, fertility issues, and cilia-related congenital heart diseases. Aquatic protists Tetrahymena, with their diverse species and clear ciliary responses, offer a valuable model for exploring the mechanisms underlying ciliary movement, which may have implications for understanding and treating ciliary dysfunction in humans. Various Tetrahymena strains have been utilized for these studies, however, there does not seem to be a standard correlation between basal body organization and swim speed. In this study, we examined the basal body organization and swim speed of various Tetrahymena species. Our results show a trend where larger cells swim faster than smaller cells, with close spacing along a ciliary row correlating with faster swim speed. Surprisingly, wider spacing between ciliary rows is also correlated with quicker swim speed. Overall, we are beginning to determine the parameters of ciliary array organization that affect output of directed fluid flow.

T46. "Investigating a potential eukaryotic riboswitch in OAZ mRNA from *Agaricus bisporus* via in-line probing", Sarah Fowler, Creighton University

Riboswitches are gene regulatory elements found in the non-coding region of messenger RNAs (mRNAs). They bind to specific ligands which induces a structural change in the RNA resulting in a change in expression of the gene next door. Whereas most riboswitches have been identified in bacterial species, the goal of this work is to try to demonstrate the presence of a riboswitch in a eukaryotic RNA, the mushroom OAZ RNA. The structural technique in-line probing is being utilized to determine whether there is a change in the OAZ RNA structure that is dependent on the presence of a small molecule, specifically the polyamine spermine and related amine-containing compounds. This technique helps to confirm ligand interaction by showing the presence of

conformational changes when the polyamine ligand is present through the change in RNA cleavage patterns as the concentration of the polyamine ligand is varied. This work, in combination with gene expression assays, will strengthen our proposal that this eukaryotic OAZ RNA has the properties of a riboswitch.

T47. "Cowpea Mosaic Virus as a Model for Understanding Viral Evolution", Hayden Gielda, Point Loma Nazarene University, Faculty Advisor: Kristopher J. Koudelka

Viruses thrive on diversity. In a virus population, there are many variants of a particular virus due to high mutation rates and rapid reproduction. While some of these mutations may be harmful to the virus, others may allow the virus to adapt to host defenses, spread more efficiently, and overcome host resistance genes. Even within a single host, what once was a genetically uniform virus can evolve in a diverse community. This diversity allows for rapid adaptation and evasion of immune responses. Cowpea Mosaic Virus (CPMV) is a single-stranded RNA virus that enters the plant cell via mechanical damage. The viral RNA then acts directly as mRNA while the host's ribosomes translate it and produce viral proteins, including RNA-dependent RNA polymerase (RdRp). The RdRp lacks proofreading, so each replication cycle produces several mutant variants. We aim to quantify CPMV's evolutionary activity by examining replication error rates. Initially, we used nanopore sequencing, which detects every nucleotide from individual RNA molecules, allowing us to most accurately measure error rates in our CPMV samples. Then, we infected black-eyed pea plants by growing until secondary leaves are present, sprinkling carborundum on them to initiate mechanical damage, and then painting the primary leaves with CPMV. In the future, we will once again use nanopore sequencing on our primary and secondary leaf samples to analyze the evolutionary activity of the virus. Although CPMV is a plant virus, it is closely related to poliovirus and coxsackieviruses thus making it a good model system to start with. Our goal is to create a toolset to better understand viral evolution that can then be applied to other viruses.

T48. "Major Front-End Migration for GRNsight, a Web Application for Visualizing Gene Regulatory and Protein-Protein Interaction Network Models", Cecilia J. Zaragoza, Loyola Marymount University, Faculty Advisor: Dr. Kam D. Dahlquist

A gene regulatory network (GRN) consists of genes, transcription factors, and the regulatory connections between them which govern the level of expression of mRNA and protein from genes. A protein-protein physical interaction network (PPI) represents a set of proteins that bind to each other. A specialized software tool would help biologists better view and analyze such networks, so we created GRNsight, an open-source web application for visualizing models of GRNs and PPIs, either uploaded by a user or obtained from our backend budding yeast database. Graph models display rectangular nodes, representing genes/proteins, with edges connecting them. GRNs have directed red edges with pointed end markers or blue edges with blunt end markers to indicate regulatory relationships (activation or repression, respectively), with edge thickness indicating

regulation strength. PPIs have undirected black edges to indicate physical binding. Time-course expression data can be displayed on GRN nodes to facilitate understanding of the regulatory relationships that generated that pattern of expression, creating new insights. GRNsight has been in development for over a decade, during which time both biological datasets and web technologies have advanced significantly. While the original implementation successfully supports the core functionality, it relies on older tools that make long-term maintenance and the addition of new features increasingly difficult. To address this challenge, GRNsight's JavaScript codebase has been migrated from a jQuery/Bootstrap/Pug framework to a modern React-based front-end which improves code readability, state management, and allows for more advanced interactive features. By modernizing the underlying framework, GRNsight is now positioned to continue expanding its functionality as new visualization needs arise for researchers to gain deeper insights into complex molecular systems. GRNsight React is available at https://dondi.github.io/GRNsight/react-thesis-4081.

T49. "Impact of Oxidized Polyunsaturated Acids on Macrophage Efferocystosis Capacity", Victoria Huang, University of California Los Angeles, Faculty Advisor: David Meriwether

Efferocytosis, the process by which macrophages engulf and digest apoptotic cells, is essential for maintaining tissue homeostasis and resolving inflammation. Impairments in efferocytosis are implicated in the progression of chronic inflammatory diseases, including inflammatory bowel disease (IBD) and atherosclerosis. Oxidized polyunsaturated fatty acids (PUFAs), specifically hydroxyeicosatetraenoic acids (HETEs) and hydroxyoctadecadienoic acids (HODEs), accumulate at high levels in inflamed intestinal tissue, though their impact on macrophage efferocytosis remains largely unexplored. In this study, we investigated how exposure to oxidized PUFAs at varying concentrations alters efferocytosis capacity in human macrophages. Across a group of HETEs and HODEs, we found that several oxidized fatty acids including 12-HETE, 15-HETE, and 13-HODE significantly suppressed apoptotic cell binding and uptake by macrophages across a 1-hour timeframe. We further tested whether this effect was mediated by the leukotriene receptor BLT2. To investigate, we performed pharmacologic manipulation using BLT2 agonists and antagonists. Pharmacologic BLT2 agonism reproduced the inhibitory phenotype, while BLT2 antagonism rescued 12-HETE induced defects, supporting a BLT2-dependent mechanism. Efferocytosis capacity was quantified using a fluorescence-based assay in which macrophage–apoptotic cell interactions were imaged after 1 hour, and the ratio of bound/internalized apoptotic neutrophils to macrophages was determined. Together, these findings suggest that oxidized PUFAs impair early macrophage efferocytosis through BLT2 signaling, offering a potential mechanistic link between lipid oxidation and defective inflammation resolution in IBD. By clarifying how specific lipid mediators disrupt efferocytosis, this work contributes to identifying therapeutic targets aimed at restoring macrophage resolution programs in chronic inflammatory disease.

T50. "TLR10 Attenuates TLR2-Mediated Inflammatory Signaling in Response to Probiotic Lactobacillus Strains", Anya Jaramillo, University of San Francisco, Faculty Advisor: Dr. Sangman Kim

Toll-like receptor 10 (TLR10) remains the only human TLR whose ligand specificity, location, and immunological function are not fully understood. Previous studies from our laboratory demonstrated TLR10 can heterodimerize with TLR2 following Mycobacterium tuberculosis stimulation. Because TLR2 primarily recognizes components of Gram-positive bacteria and typically promotes pro-inflammatory signaling, these findings suggest that TLR10 may modulate TLR2-mediated responses to microbial ligands. Emerging evidence also indicates that TLR10 may function as an anti-inflammatory receptor. We therefore hypothesized that TLR10 may recognize probiotic bacteria and attenuate inflammatory signaling triggered by TLR2. To test this, human embryonic kidney reporter (HEK-Blue) cells co-transfected with TLR2 and increasing dosages of TLR10 were stimulated with three probiotic Lactobacillus strains Lactobacillus rhamnosus, Lactobacillus fermentum, and Lactobacillus plantarum. NF-κB activation was quantified using a secreted embryonic alkaline phosphatase (SEAP) reporter assay, and interleukin-8 (IL-8) secretion was measured via enzyme-linked immunosorbent assays. SIncreasing TLR10 expression resulted in progressively reduced NF-κB activation and IL-8 production following Lactobacillus stimulation, demonstrating dose-dependent attenuation of inflammatory signalling. These findings suggest that TLR10 may function as a modulatory receptor that attenuates TLR2-mediated inflammatory responses to probiotic bacteria, and may be involved in maintaining immune tolerance to beneficial microbes. Ongoing studies will elucidate the molecular mechanisms underlying these phenotypes by investigating downstream TLR10 signaling pathways and validating probiotic-induced responses in physiologically relevant immune cell models.

T51. "Magnolia Leaf Silver Nanoparticles Display Dose-Dependent Disruption of Bacterial Biofilms", Faith Azer* and Amadeus Nunez, The Master's University, Faculty Advisor: Dr. Haley Smith

Magnolia leaf silver nanoparticles (MAgNPs) have been shown to display antimicrobial properties against a wide variety of microbes. However, limited research has been done to test their effect on mature bacterial biofilms. This study aims to explore the effects of MAgNPs on sessile bacterial communities. Previous literature has demonstrated that mature biofilms are often resistant to conventional antibiotics. In order for antibiotics to be effective against a biofilm, high dosages and adjunct antibiotics are typically necessary. This limited efficacy of antibiotics lends itself to more research to explore other alternatives to biofilm disruption, such as nanoparticle-based treatment. Previous work has provided evidence that the MAgNPs are effective at inhibiting initial growth on

fresh plates for various species of bacteria, including S. aureus, E. coli, P. aeruginosa, among others. Here these same bacteria were allowed to establish a mature biofilm prior to being challenged with the MAgNPs. To investigate this, the pre-established biofilms were treated with different doses of the MAgNPs. Biofilms were prepared by culturing in broth for 48 hours at 37° C in 16-well plates. Biofilm biomass was assessed using crystal violet staining and analyzed by UV-Vis spectroscopy. A concentration-dependent gradient in biofilm disruption was observed. Ultimately, the results suggest that MAgNPs exhibit a dose-dependent disruptive effect on mature biofilms, suggesting that nanoparticles could represent an alternative strategy for combating biofilm proliferation.

Poster Presentations AT-A-GLANCE

Location: University Ballroom

Discipline	Abstract numbers	Pages
Ecology and Evolution	#P1-55	55 - 89
Biochemistry, Biophysics and Bioinformatics	#P56-89	90 - 111
Neurobiology	#P90-119	112 - 132
Biological studies of disease	#P120-126	133 - 137
Marine Biology	#P127-143	137 - 146
Microbiology	#P144-155	147 - 154
Physiology and Developmental Biology	#P156-171, 217-218	155 - 163, 190-191
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Poster Titles and Abstracts:

WCBSURC Poster Session AM - 11:15am-12:45pm
Poster #’s 1-126

Ecology and Evolution

P1. The Gut-Brain Connection in a Cnidarian Model: Microbiome Rehabilitation and Neural Function in the Sea Anemone Aiptasia,
Sarah McCormack, University of San Diego, Faculty mentor: Dr. Cawa Tran

A crucial determinant of health can be attributed to the significant role of the gut microbiome, which is in constant communication with the brain. Essential to host fitness, but incredibly complex, the connection between the gastrointestinal tract and the brain ignites curiosity in the outcome of manipulating the gut to influence the central nervous system. The sea anemone, Exaiptasia diaphana (Aiptasia), is a cnidarian with a diverse microbiome and one of the first animals to develop a nervous system. These organisms can serve as a model of gut-microbiome depletion and rehabilitation to provide insight into the origins and nuances of this gut-brain relationship. I administered an antibiotic cocktail to sterile, artificial seawater in 12-well plates which contained the antibiotic-treated Aiptasia group, while the non-antibiotic group received only seawater. Then, Aiptasia were homogenized and plated on marine agar to evaluate bacterial growth. Activity of the neuropeptide FMRFamide was also assessed with antibody staining and fluorescence microscopy. Fluorescence intensity decreased in the antibiotic-treated group (suggesting nervous system changes), which also yielded significantly less bacteria than the non-antibiotic group (suggesting successful depletion of the gut microbiome). Following depletion, the gut microbiome was rehabilitated with the beneficial bacterium, Tritonibacter mobilis, which is tagged with Green Fluorescent Protein (McLaren et. al 2025). GFP colonies successfully grew (demonstrating a recovered gut microbiome and uptake of T. mobilis) and the group treated with T. mobilis had the greatest fluorescence intensity. Through these findings, I can conclude that a depletion of the gut microbiome in Aiptasia affects the proficiency of the nervous system, but administering probiotics offers an opportunity to recover and improve host health.

P2. Motor and Feeding Behavior of a Sea Anemone Following Gut Microbiome Depletion, Bridget McFall, University of San Diego, Dr. Cawa Tran

As coral reefs continue to face a challenging climate, the microorganisms within the ecosystem and within corals themselves will continue to shift. Perturbations of the host microbiome may result in modifications to host fitness and physiology via impacts on the nervous system. In complex organisms like humans, the gut microbiome has been linked to brain functioning, but this bidirectional gut-brain axis is not fully understood. Meanwhile, cnidarians like corals and the sea anemone Exaiptasia diaphana (Aiptasia) possess primitive nerve nets and gut microbiomes. Aiptasia consists of a complex core microbiome containing hundreds of microbial partners that contribute to various aspects of host fitness, such as its biomass and asexual reproduction. Aiptasia also features a structurally simple nervous system composed of interconnected nerve nets with sensory and ganglion cells that coordinate motor behaviors, such as muscle contractions. Thus, the gut-"brain" axis of Aiptasia provides a unique opportunity to investigate how disruption of the gut microbiome may alter neural activity and observable motor behaviors. In this study, the Aiptasia microbiome was depleted via direct administration of a broad-spectrum antibiotic concoction (carbenicillin, chloramphenicol, nalidixic acid, and rifampicin) and changes in motor behavior were subsequently observed and quantified. Tilt behavior was significantly reduced in antibiotic-treated Aiptasia. Antibiotic-treated anemones were also less likely to feed on brine-shrimp larvae as there was a remarkable change in the spread of tentacles to capture food. These results suggest that a depleted microbiome can have adverse effects on host motor behavior, possibly linked to nervous-system changes. As Aiptasia has been used as a prominent laboratory model for corals, elucidating the connections between the microbiome, the nervous system, and motor behavior will have implications for the health of corals and other cnidarians, and will become increasingly important as climate change continues to influence these metaorganisms.

P3. Optimizing Long Read Sequencing for the Seagrass Microbiome, Vivian Sapovits, University of California, Davis, Faculty mentor: Jonathan Eisen

Eelgrass (Zostera marina) is a foundational marine plant species found across the Northern Hemisphere that provides food, shelter, and erosion control in shallow areas close to the shoreline. Our lab studies eelgrass to understand how associated microbes in and near the plant affect the host's responses to environmental stressors. My project focused on creating an accessible, reliable protocol for obtaining genomic data from these microorganisms that undergraduates at different levels could complete independently and within their own academic timeline. Using the MinION, a long-read sequencing device that can be run on a lab bench, I compared three DNA extraction techniques and two annotation software packages using 28 bacterial isolates. From this, I created a flexible pipeline that can be completed in as little as a single workday. By extracting DNA directly from freezer stocks and utilizing the free, cloud-based assembly tool KBase, students can extract DNA, sequence, and characterize bacterial genomes quickly without obstacles such as isolate

culturing or coding-based assembly tools. This pipeline removes barriers, allowing more undergraduates to get hands-on experience while generating data that enables us to create a synthetic bacterial community to further test the effect microbes have on Z. marina's tolerance to heat stress.

P4. Characterization and Isolation of Microbes from Unique Serpentine Soil Environments, Santos De La Trinidad Munoz, Architha Dhananjayan, California Polytechnic State University, San Luis Obispo, Faculty mentor: Alejandra Yep

Across California's Central Coast, serpentine soils are characterized by nutrient-poor and metal-rich concentrations, creating strong selective pressure on resident microbial communities. Mining introduces contaminants, disrupting biodiversity, and hosting unique communities with metal-tolerant microorganisms. Baseline comparisons of microbes from disturbed and undisturbed serpentine habitats remains limited. In this work, we developed methods to isolate and characterize serpentine-associated microbes. Serpentine rhizosphere soil samples were collected by our collaborators from three sites: undisturbed North (n=6), mining sites (n=12), and undisturbed South (n=6). Our team conducted phenotypic screening and assessed DNA quality for downstream genotypic analyses. Environmental DNA was extracted from 24 samples using the Qiagen DNeasy PowerLyzer PowerSoil protocol and evaluated by NanoDrop spectrophotometry as a sequencing quality-control checkpoint. DNA recovery varied across samples (0.8–24.2 ng/mL), with total concentrations of 64–1,936 ng, indicating sufficient DNA for microbial sequencing. Purity metrics fell below ideal ranges for some samples (A260/280 = 0.39–1.81; A260/230 = 0.30–1.41), consistent with co-extracted inhibitors and contaminants common in complex environmental samples. Results indicate while many of the extracts contain measurable DNA, additional purification will be required prior to PCR and sequencing. To complement eDNA metrics with phenotypic observations, two mining-site samples (3014 and 3018) were serially diluted and plated to isolate morphologically distinct colonies for staining and microscopy. Isolates displayed diverse colony and cellular phenotypes, including Gram-positive and negative bacteria; many were endospore formers, consistent with stress-tolerant taxa. Ongoing work includes 16S rRNA gene sequencing to compare communities across site types and further characterization of cultured isolates associated with metal-tolerance and environmental persistence. Overall, serpentine rhizosphere samples yielded measurable microbial DNA but often contained inhibitory contaminants. Establishing these quality-control benchmarks strengthens downstream sequencing comparisons and improves confidence that extraction conditions capture microbial diversity.

P5. Seasonal Variability Inhibition of Phytophthora ramorum by California bay laurel Bacterial Isolates, Ishan Shardha, Santa Rosa Junior College, Faculty mentors: Dr. Katy Jamshidi, Dr. Dean Katzung

Inhibition of Phytophthora ramorum by Umbellularia californica bacterial isolates: Implications for Biological Control of Sudden Oak Death Sudden Oak Death, a forest disease caused by the oomycete Phytophthora ramorum, poses a major risk to California ecosystems by causing widespread death of oak and tanoak trees, keystone species in the redwood forests. Due to a lack of effective treatments, researchers and ecologists have turned to naturally occurring microbes to mitigate P. ramorum growth. This study focuses on Umbellularia californica plant bacteria's inhibitive properties against P. ramorum, with a emphasis on investigating inhibitory variation between varying seasonal samples. Six bacterial isolates were cultured on agar plates and incubated for 14 days with a plug from a P. ramorum colony to evaluate effects on vegetative growth and sporulation. Bacillus megaterium, a known inhibitor of P. Ramorum growth, was used as a positive control. Sporulation was induced on day 12 by adding sterile soil water. On day 14, the sporulation plates were quantitatively analyzed for the count of Chlamydospore and Sporangia while vegetative growth was measured in corresponding plates and screened for zones of inhibition. Statistical analysis using ANOVA indicated significant inhibition of both vegetative growth and sporulation of P. Ramorum when exposed to the bacterial isolates. The patterns of inhibition were broadly consistent across seasonal samples, with no individual samples showing significantly greater inhibitory activity. Bacterial samples proved to be more consistent as vegetative growth inhibitors with an average growth reduction of 82.9% relative to the control. These results suggest that U. californica plant bacteria could act as a biological preventative treatment against P. Ramorum, although further testing and development is required to evaluate consistency and ecological applications.

P6. Soil Bacteria Biodiversity in Mipla versus non Milpa Agricultural Systems, Joe Perez, Kenia Cortes-Campos, California State University, Monterey Bay, Faculty mentor: Callie Chappel

Soil microbes play an important role in how agricultural systems function such as; influencing nutrient cycling, soil structure, and long term soil health. However, these microbial communities are often studied without fully considering the cultural and management practices that influence them. The Salinas Soil Project explored how soil microbial communities often differ between milpa and non milpa systems through a community based research approach through collaboration and shared learning in the local community. Milpa systems, which emphasize greater plant diversity and long standing relationships with the land, were compared to non milpa systems including monoculture and unplanted plots. Soil samples were collected during a series of workshops that brought together students, educators, and community members. Using a portable Bento Lab, participants extracted soil DNA and amplified bacterial genetic material using PCR, allowing microbiology to take place in a hands-on setting rather than a traditional laboratory. Initial observations suggest that soils managed under milpa systems support a greater diversity of bacterial communities compared to non milpa soils. These patterns point to the importance of plant diversity and management practices that shape and support healthier soil systems. This project also

demonstrates the value of community science as a way to connect people to both land and research. By combining molecular tools with local knowledge, the Salinas Soil Project shows how community driven microbiology can generate meaningful ecological insights while fostering a deeper understanding of sustainable agriculture. This work contributes to broader conversations about soil health, food systems, and the role of culturally rooted farming practices in supporting resilient ecosystems.

P7. Natividad Creek Soil Biodiversity, Tatiana Chavez Pacheco, California State University, Monterey Bay, Faculty mentor: Callie Chappell

The Salinas Soil Project explored how microbial communities change along the path of Natividad Creek within Natividad Creek Park through a community based research approach that centered local curiosity and collaboration. Guided by the question of whether the creek gains or loses microbial diversity as it flows through the park, community members, students, and educators gathered at three points along the creek to collect soil and sediment samples. This work was rooted in shared observation, meaningful listening, and hands-on learning with portable Bento Lab molecular tools that allowed participants to extract environmental DNA and amplify microbial genetic material in accessible, non-traditional settings. We hypothesized that microbial diversity would increase as the creek moved through areas with slower water flow and more vegetation, conditions that might support a richer set of microbes. The results suggest that while diversity is lower near the start of the park where water enters from agricultural fields, it increases between the middle and end of the park, where vegetation is more abundant and flow is gentler. These patterns point to the ways creek dynamics, plant life, and human influenced landscapes shape living communities beneath the surface. Beyond the biological insights, this project shows that participatory microbiology can open space for people to see themselves as stewards of both land and water. By combining molecular exploration with local knowledge and collective care, the project contributes to broader conversations about watershed health, ecological observation, and community science.

P8. Influence of soil microbial inoculum on the growth and biomass allocation of the invasive grass Bromus diandrus, Ross Hagino, University of California, Davis, Faculty mentor: Tom Buckley

Soil microbes play a critical role in supporting plant performance and ecosystem health, plant productivity, nutrient uptake, defense response, environmental stress alleviation, and plant community composition. We investigated how soil microbial communities may influence the early growth of an invasive grass, ripgut brome (Bromus diandrus Roth), in a Southern California mediterranean ecosystem. Soil and seeds were collected from grassland plots at the Loma Ridge

Global Change Experiment in Irvine, California. We then grew B. diandrus in a greenhouse using a homogenized mixture of sterile soil and soil inoculum. Three treatments of differing inoculated to sterile soil ratios were used: 25%, 50%, and 75% inoculated soil. Upon germination, we employed a serial harvested growth analysis to understand the features of plant growth that mechanistically underline the responses of B. diandrus to microbial exposure. Our findings suggest that larger soil microbial communities have an important impact on growth rate and productivity of B. diandrus. However, these changes in growth rate were environmentally driven rather than by changes in growth strategy such as a redistribution of biomass from belowground to aboveground tissues. Understanding the relationship between soil microbial communities and the mechanistic components of plant growth is essential for predicting and managing invasive plant species such as B. diandrus under changing climate conditions.

P9. Bacterial aerosols' size distribution changes upon oxidative stress, Leslie Mendez, Daisy Matul-Perez, Leslie Mendez Perez, Dominican University of California, Faculty mentor: Christine Koh

Bacterial aerosols' size distribution changes upon oxidative stress Bioaerosols are composed of particulates in the air that contain living, organic substances or are of biological origin. Bacterial aerosols containing Bacillus cereus (B. cereus) or Escherichia coli (E. coli) were studied. Studying bacterial bioaerosols is crucial because they can spread diseases and have a significant impact on our ecosystems. The size distribution of bacterial aerosols under oxidative stress was monitored using Dynamic Light Scattering (DLS). The size of bacterial aerosols is closely linked to their viability, with larger bacterial aerosols showing higher chances of viability. Because B. cereus is a spore-forming bacterium and E. coli is a non-spore-forming bacterium, they exhibit differing patterns in size distribution; however, both show two size distributions upon aerosolization. B. cereus, a sporing bacterium, produces spores that average a size of 914 nm and a larger aggregate with a mean size of 3350 μm. The non-sporing bacterium, E. coli, formed a large aggregate with a mean diameter of 170 μm and small fragments at 30 nm in size. When exposed to oxidative stress, 70% isopropanol or 10% bleach, the two different bacterial aerosols behaved differently. While large aggregates of E.coli remained stable or grew larger, large aggregates of B. cereus were destroyed to form spore clusters by isopropanol or more spores by bleach. Formation of spore clusters or spores upon oxidizer treatment shows how sanitizers can not be effective in spore-forming bacterial aerosols. These results highlight that spore-forming and non-spore-forming bacteria respond differently to oxidative stress when airborne, suggesting that customized strategies are needed to reduce their viability. Such findings are critical for developing practical approaches to limit airborne pathogens from spreading and protect human and environmental health.

P10. Investigating the Effects of Reishi Mushroom Extracts on *Caenorhabditis elegans* Lifespan and Reproduction, Kim Tu, Oregon State University, Faculty mentor: Dee Denver

Caenorhabditis elegans serves as a model organism for studying human health and aging processes. Ganoderma mushroom species, such as G. lingzhi, known as the "mushroom of immortality" in traditional Asian medicines, are prompting investigation into whether G. lingzhi and its Pacific Northwest variant, G. oregonense, increase lifespan in the C. elegans nematode.To examine the effects of Ganoderma mushroom extracts, specifically G. lingzhi and G. oregonense compounds, on C. elegans lifespan and reproductive schedule.C. elegans lifespan and reproductive schedule assays were conducted in laboratory conditions using three distinct treatment groups. One experimental group was exposed to G. lingzhi extract, while a second group received G. oregonense extract, and a third group was treated with the water control. Nematodes were monitored daily throughout their lifecycle to assess both survival rates and reproductive schedule. Data analysis included t-test comparisons to evaluate treatment effects.The study revealed two primary outcomes: differences in survival duration and variations in reproductive schedule across treatment groups. The water control group exhibited the shortest average lifespan, followed by the G. oregonense treated group with intermediate survival times, while the G. lingzhi treatment group demonstrated the longest average lifespan. Statistical analysis revealed significant differences between G. lingzhi and water control groups, with near significant differences observed between G. oregonense and G. lingzhi treatments. Similar patterns were observed for reproductive schedule, with G. lingzhi treated nematodes showing enhanced reproductive performance compared to controls. Statistical analysis indicated that only the G. lingzhi versus water control comparison reached statistical significance, while other group comparisons did not achieve significance thresholds.Results suggest a potential positive effect of Reishi mushroom extracts on C. elegans survivorship, but additional research is needed to provide stronger evidence for the effects of these mushroom compounds on nematode survivorship and reproduction.

P11. Assessing Water Quality and Ecosystem Health in Costa Rica: Impacts of Agricultural and Human Activity, Simon Kovass, Nicolas Folsom, Youngstown State University, Faculty mentor: Carl Johnston

The highly biodiverse freshwater ecosystems of Costa Rica are being increasingly impacted by the country’s widespread agricultural pesticide and fertilizer use. Many of the nearby streams are vulnerable to contamination due to runoff. The study assesses the influence of surrounding land usage in and around La Selva Biological Station in the Sarapiquí Cantón of Costa Rica, where protected forest exists alongside agricultural and developed areas. Over a three-day period, stream sites representing varying levels of anthropogenic influence will be sampled and analyzed for key indicators of water quality. Physical and chemical parameters will include dissolved oxygen, pH, conductivity, temperature, nitrate, phosphate, sulfate, alkalinity, hardness, and chlorine (n=4).

Biological and contaminant analyses will include fecal coliform bacteria, vibrio, and pesticide presence (n=4). Water quality parameters will be compared across sites to evaluate differences associated with land use intensity. Streams influenced by agricultural and human activity are expected to exhibit reduced dissolved oxygen and elevated nutrient, ion, microbial, and pesticide levels relative to protected reference streams within the La Selva reserve. Results will improve understanding of land use impacts on tropical freshwater ecosystems and provide valuable information to support conservation strategies and sustainable water resource management.

P12. Effects of Crop Rotation on Resistance Breaking Root-knot Nematodes, Emma Shigekane Kraft, University of California, Davis, Faculty mentor: Shahid Siddique

The pervasive presence of root-knot nematodes threatens agricultural crop yields and food security around the world. For decades, the only resistance gene against root-knot nematodes in tomatoes has been the Mi-1 resistance gene. Although genetic resistance is present, resistance breaking nematode species for both Meloidogyne incognita and M. javanica are able to infect tomatoes with Mi-1. Preliminary data shows that resistance breaking strains ineffectively reproduce and have a lower number of eggs on crops not containing Mi-1, thus indicating a reduction in eggs when compared to the wildtype strain. We hypothesize this pattern may be due to a fitness cost associated with resistance breaking. This project seeks to investigate this phenomenon by testing field-collected resistance breaking isolates on a variety of rotation crops for processing tomatoes, all of which lack Mi-1 resistance. If the resistance-breaking isolates have a significant reduction in egg counts when compared to the wildtype, this could indicate that crop rotation with plants not harboring Mi-1 is a viable pest management strategy for controlling resistance breaking nematodes.

P13. Using AI to Improve Consumer Recycling Practices and Reduce Waste Contamination, Olga Vasquez De Leon, Dominican University of California, Faculty mentor: Christine Hoffman

In the U.S., recycling systems continue to face challenges related to contamination, inefficiency, and consumer confusion. Although the use of artificial intelligence (AI) has increasingly been used in backend waste management processes (Belyamani, 2025), consumer-facing front-end AI interventions remain limited. Hence, the goal of the study is to evaluate whether an AI-powered, real-time recycling feedback system can reduce consumer confusion and improve sorting accuracy. The focus of this initial study was to identify changes to recycling habits within residence halls at the Dominican University of California. Our preliminary data shows significant recycling sorting improvements after direct feedback interventions. These results are the foundation for a larger behavioral science study evaluating whether AI-powered real-time feedback reduces contamination rates, improves recycling behavior, and supports broader sustainability goals. The

system utilizes a YOLO11-nano computer vision model selected for its balance between classification accuracy, inference speed, and compatibility with resource limited hardware.

P14. The Effects of Sample Size on Ecological Niche Models: Analysis Using the California Ground Squirrel, Paula Ceballos, San Francisco State University, Faculty mentor: Robert Boria

Ecological Niche Models (ENMs) can help us identify potential suitable conditions for species of interest through time and space. They do so by comparing the overall environmental conditions available to the species with the conditions at localities where the species occurs. Identifying the number of occurrence localities needed to model a species potential distribution is often overlooked. Using too many occurrence records may not increase model accuracy and could potentially hinder model performance (i.e., by limiting the amount of available background localities and reducing discriminatory ability). Further, as sample size increases so do associated issues of sampling bias and georeferencing errors, which could potentially lead to overfit models. In this study, we model the effects of varying sample size of the California ground squirrel (Otospermophilus beechyi). Otospermophilus beechyi is an ecologically significant species throughout California. They are a keystone prey species by being a food source for many animals across trophic levels. Furthermore, they are allogenic ecosystem engineers by altering habitats through their burrowing and are key seed dispersers across their geographic ranges. To begin with, we reduced occurrence records of O. beechyi through spatial filtering and generated random localities for differing sample sizes. We used evaluation statistics to identify our model's discriminatory ability. In addition, with only 20% of the data we were able to generate biologically meaningful models. Future work includes comparing our findings with a different species distribution model working with the Merriam’s Chipmunk (Tamias merriami). Tamias merriami are distributed in a smaller geographic range, covering parts of California and Baja California, Mexico. By comparing a widespread species to a geographically constrained species we can assess whether sample size effects differ across geographic distribution in the context of ongoing climate change and anthropogenic disturbance.

P15. Predation risk for insects living in Northern California and Southern Finland: effects of prey position and predator exclusion, Bianca Gaitan, Sonoma State University, Faculty mentor: Nathan Rank

Many factors influence insect abundance, but one of the most important sources of insect mortality is predation by other invertebrates or vertebrates. We lack information about how predation affects insects in different microhabitats and gaining a better understanding of this might help us conserve ecologically important insects. In this study, predation risk was evaluated at 128 sites across the globe with comparisons among prey on foliage versus on the ground or in soil. We focus on two

geographically distinct sites with different seasonal patterns: Fairfield Osborn Preserve (FOP), California, USA and a forest in Kustavi, southern Finland. Standardized live prey beetle larvae (Tenebrio molitor) were placed in the soil, on the ground, or on a branch. To measure predation intensity, we placed a subset of larvae into exclusion cages. Overall, prey survival was higher at FOP than at Kustavi, especially in soil and on the ground. On branches, survival in exclusion cages was greater than that of exposed larvae at FOP while it was similar between exposure and exclusion treatments at Kustavi. Ants were the most frequent predators observed feeding on prey at both sites, and mites, bugs, and a fly were also observed at FOP. The difference between survival on branches at FOP versus Kustavi may have occurred because birds constitute a higher proportion of predators in this part of California compared to southern Finland. These results demonstrate that predation is important in forest habitats across the globe but regional differences cause large differences in insect survival.

P16. Limitations of Gene Flow in an Endangered Succulent *Dudleya setchellii*, Dante Cable, Karina Martinez, Santa Clara University, Faculty mentor: Justen Whittall

Island and island-like habitats are predicted to restrict gene flow among populations. However, when endangered species occupy insular niches, limits to gene flow could threaten them with genetic collapse and eventual extinction. Serpentine outcrops are one such island-like habitat with a high concentration of endemic and often rare species, yet we know very little about gene flow within and among populations occupying these unique soils. Dudleya setchellii is a federally endangered succulent restricted to serpentine outcrops in southern Santa Clara County, California. It is characterized by producing many, showy cream-colored flowers that produce hundreds of 0.7mm long seeds with no dispersal appendages. Gene flow within and among populations of D. setchellii likely occurs through insect-mediated pollination and wind dispersal of the seeds.In order to document gene flow via pollinators, digital video cameras recorded pollinators at two populations from early morning to late afternoon over two seasons. After reviewing approximately 1,000 flower hours, the Yellow-faced Bumblebee (Bombus vosnesenskii) accounted for ~80% of all pollination events with substantial variation in overall visitation rate between the two seasons. To evaluate another source of gene flow, we conducted a seed dispersal experiment under optimal conditions using realistic wind speeds. Seeds traveled an average distance of 2.76m and were normally distributed.These findings suggest that primarily bumblebee pollination and to a lesser extent seed dispersal likely limit gene flow. These results are consistent with a parallel conservation genomic study which revealed isolation by distance within and among populations. Briefly, ~300 genomes were resequenced and pairwise genetic distances accounted for ~30% of all genomic variation within a population, and ~40% among populations. Both genomic investigation and field studies reveal substantial barriers to gene flow which are important when making future conservation decisions and guiding restoration practices for D. setchellii.

P17. Warm Spot Test as a Correlate of Social Dominance in Mice: Evaluating Animal Behavior Tracking Methods, Arjun Prasad, Princeton University, Faculty mentor: Christina Kim

Dominance hierarchies occur naturally in both primate and non-primate species when resources are limited. They allow for organization between groups or populations to minimize conflict, allocate available resources, and ensure survival. Mice are commonly used as a model for social dominance as their behavior can be quantified through assay-specific metrics or by tracking their position and movement. The tube test, in which the dominant mouse advances and the subordinate mouse retreats, is the gold standard for measuring social dominance through numbers of wins. However, other assays are used as correlates of social dominance, often to validate tube test results. One such assay is the warm spot test (WST), in which the most dominant mouse spends the most time in a warm spot amidst a cold floor. Here, we first determined the rank of C57BL6 male mice (n=11 cages) using the tube test assay. The WST was completed after the completion of the tube test assay. Warm spot test results show a moderate correlation with tube test results, with a Spearman's rank correlation coefficient of 0.466 and a Kendall's tau-b coefficient of 0.399. The video data of these assays was manually quantified, potentially introducing multiple sources of error that can compromise reproducibility. Thus, this study aims to train and use deep learning models to track mice during the WST. We employ SLEAP (Social LEAP Estimates Animal Poses), which tracks animal position changes and allows us to assess the validity of the warm spot test in comparison to manual tracking. By quantifying a hierarchy from SLEAP data, performing correlational analysis with tube test data, and comparing it with analysis from manual tracking, we can understand the degree to which SLEAP data can validate the warm spot test and serve as a correlate of social dominance itself.

P18. Bumble bee foraging preference is predicted by community scientist photos, Emeline Huffaker, University of California, Davis, Faculty mentor: Elizabeth Crone

Understanding how different plant species support pollinators is critical for pollinator conservation. One potential source of this information is community scientist observations (such as photo records from iNaturalist). However, there are many reasons why these observations might not accurately reflect resource preference, such as that broad patterns of resource use may simply reflect resource abundance in a landscape. Bombus crotchii, or the golden state bumble bee, has been observed by community scientists as visiting milkweed (Asclepias spp.) more than any other plant genus across California. This is surprising, given that many milkweed species have toxic nectar and that milkweed pollen is entirely unusable for bees. To test if Bombus crotchii actually prefers milkweed over other flowering plants, we placed planter pots with alternative flower species in a field of over 300 stems of A. eriocarpa, or woolypod milkweed, at a site in Northern California. Each planter pot contained the same five plant species: Salvia longispicata x farinacea, Salvia chamaedryoides, Achillea millefolium, Gaillardia pulchella, and Penstemon heterophyllus.

These species were chosen as attractive flowering plants for bumble bees, as well as the general community of pollinators. We measured visitation rates for all visiting pollinator taxa to each flower species. We found that there was a significant difference in B. crotchii visitation between plant species (LRT: $x2=101.18$, $p< 0.0001$), with milkweed visited at a higher frequency than the alternative flowers, suggesting a preference for milkweed. These results suggest that, in this instance, community science records are accurately capturing floral preference for B. crotchii. Additionally, because B. crotchii has been lost from much of its range in the past decade, efforts to support this imperiled species may benefit from promoting milkweed in the landscape.

P19. Cooperation in Juvenile Play in Ground Squirrels, Finn Preston, Ezra Poggio, Ava Eaton, Sadie Kaiser, University of San Francisco, Faculty mentor: Scott Nunes

Play behavior occurs among juveniles in most mammalian species and can provide a range of benefits to young animals, including the development of motor, social, and cognitive skills. During play interactions, juveniles can act as initiators/attackers or targets/defenders. The behaviors and skills associated with being the initiator of a play interaction differ from those associated with being a target. Thus, to maximize the adaptive benefits of play, young animals should express some degree of cooperation and reciprocity during play interactions, taking turns being the initiator vs. target over the course of play with a specific partner. We evaluated cooperation and turn-taking during play in juvenile Belding's ground squirrels (Urocitellus beldingi). Play interactions with a greater number of play bouts were characterized by a significantly higher degree of reciprocity and turn-taking. Juveniles who instigated play interactions were more likely to initiate play bouts during the interaction; however, in interactions with a greater number of play bouts, the number of bouts initiated by the instigator of the interaction and its partner became more equal. These results indicate that reciprocity is a characteristic of multi-bout play interactions, and are consistent with the idea that reciprocity increases the stability and degree of interchange between partners of play interactions in U. beldingi.

P20. Diversity of play partner preferences in juvenile ground squirrels, Ava Eaton, Sadie Kaiser, University of San Francisco, Faculty mentor: Scott Nunes

Juvenile play occurs in a wide range of mammalian species and can provide important benefits to individuals during early life, and in some cases can provide multiple benefits to young animals. The structure of play has been suggested to contribute to the benefits young animals gain from play behavior, in part via influences on early development. If diversity in play allows play to influence multiple elements of development and thus contributes to the multiple benefits of play, it would be expected that juveniles seek a diverse range of play partners, or seek partners who provide a diverse range of challenges during play interactions. We evaluated play partner

preferences in juvenile Belding's ground squirrels (Urocitellus beldingi), a species for which multiple benefits of play have been identified. We assessed partner preferences in litters with > 5 juveniles to ensure that subject animals had a range of partners to choose from when playing. Preliminary data analysis indicates that juveniles play with some littermates more frequently than others and suggests that sex and body mass of littermates may influence partner preferences, providing preliminary support for the idea that juveniles may seek challenge during play interactions by preferentially selecting partners with whom they are evenly matched.

P21. Multi-partner Play Interactions in Juvenile Ground Squirrels, Salean Nguyen, University of San Francisco, Faculty mentor: Scott Nunes

Juvenile play behavior occurs across a range of animal taxa, and is especially common in mammals. Play can provide a range of adaptive benefits to young animals related to development of motor, social, and cognitive skills, and in some species can provide multiple benefits to individuals. The structure of play has been suggested to underlie the benefits it provides, with greater diversity in play contributing to a greater range of benefits provided by play. Juvenile play predominantly occurs in dyadic interactions, but in some cases may involve multiple partners within an interaction. We evaluated the possibility that multi-partner play interactions contribute to the complexity and diversity of play behavior in juvenile Belding's ground squirrels (Urocitellus beldingi). We cataloged the behaviors expressed during dyadic play interactions and multi-partner interactions. We identified behaviors that occur in multi-partner but not dyadic interactions, suggesting that multi-partner interactions may enhance challenges and opportunities for skill development during play.

P22. Impacts of Maternal Care in Phidippus regius, Roxanne Baggott, University of California, Berkeley, Faculty mentor: Damian O. Elias

Spiders exhibit diversity in maternal care in both level of involvement and the form that the care takes. Some behaviors are extreme, such as in matriphagy, where offspring consume their mother, while other species show no care beyond creation of the egg sac (a protective layer of silk around eggs). In jumping spiders (family: Salticidae), a variety of maternal care behaviors have been observed, ranging from some females guarding their egg sac to others feeding offspring after they hatch. Maternal care behavior in most Phidippus, however, are unknown. In this study, we explored maternal care in the spider Phidippus regius, a species distinct for their growing popularity as pets due to social media and their large size which makes them interesting model species. To investigate the impacts of maternal care in P. regius, we collected specimens from a field site in Florida. Pregnant females were placed into one of two treatments: one where females were removed from their egg sac one to three days after laying it, and one where females were left

with their egg sac until spiderlings hatched and fully dispersed; only after that were the females separated from their offspring. Data was collected on the amount of time that it took for the spiderlings to hatch and disperse in both treatments. In the treatment 1 (absent mothers) no spiderlings emerged from any egg sac, despite 23% of the egg sacs hatching. In treatment 2 (present mothers), 63% of egg sacs hatched, and 100% of hatched spiderlings emerged from the eggsac. This research demonstrates that P. regius females engage in maternal care and that females remaining with their egg sac is necessary for successful emergence. This work has important implications for animal husbandry and revealed an underappreciated facet of jumping spider natural history.

P23. Is Seasonal Fattening in Male Squirrel Monkeys (Saimiri collinsi) Associated with Male-Male Competition? Bella Fuentes, California Lutheran University, Faculty mentor: Anita I. Stone

Sexual dimorphism describes the difference in traits between males and females of the same species. Many primates show body size dimorphism, which can help with male-male competition over females. Adult male squirrel monkeys (genus Saimiri) undergo a hormonally-induced "fattening process" in their 8-week breeding season, which increases body size dimorphism. Possibly, male fattening is a sexually-selected trait that allows fatter males to have higher access to mates by being competitively dominant. This field study examined whether the male fattening trait in Saimiri collinsi is linked to advantages acquired during male-male competition events. We hypothesized that more robust males ("fatter") would gain more access to females due to their higher competitive abilities. Specifically, we predicted that more robust males would participate in and win more contests against less robust males. We collected behavioral data on two social groups of squirrel monkeys, during three mating seasons (2023-2025; N=414 contact hours), in Eastern Amazonia, Brazil. Study groups averaged 46 individuals, 12 of which were adult males. Males were classified on a visual scale of 1 to 3 (1=least fat, 3=most fat). Focal animal samples were used to construct nearest neighbor budgets for different males. We also recorded the initiator, recipient and winner of male-male contests, as well as copulation occurrences. Grade 3 males were responsible for 88% of the copulations (N=50). During male-male contests, females were present (within 3m) 73% of the time (N=164 observations). Grade 3 males won 67% of competitive interactions, initiating 71% of these. Grades 2 and 3 males received the most aggression; grade 1 males won few fights. These results show that fatter males are more competitively dominant and gain more access to females, suggesting that the degree of fattening of male S. collinsi affects their

P24. Evidence for Cryptic Female Choice Through Sperm Storage Bias, Elizabeth Zakharyevich, University of California, Riverside, Faculty mentor: David Reznickcompetitive ability and mating success. Funded by NSF (Award no. BCS-2140666).

Fertilization success was long believed to simply depend on sperm competition. However, growing evidence shows that post-copulatory sexual selection is also influenced by female-driven processes known as cryptic female choice (CFC). An example of CFC is female sperm storage biases, which can occur when there is a nonrandom favoring of one male's sperm over another's.Our project goal is to understand the biases that female sperm storage can cause in the live-bearing fish family Poeciliidae. To address this, we plan to work within two populations of the species Poeciliopsis prolifica. Females will be inseminated with two sets of about 40 dyed sperm bundles, originating from a male of each population. By staining sperm from two males with different fluorescent dyes, we can directly quantify whether females store one male's sperm more than the other, providing a measurable test of storage bias and CFC.After insemination, the ovaries will then be dissected, dissociated into a single-cell solution, and analyzed using flow cytometry to determine which fluorescent signal is more present in the ovaries. Data collection and analysis are currently in progress, and we have preliminary results.

P25. Evaluation of cortisol levels in DeBrazza's monkeys with stereotypic stress displays, Alyssa Dennison, Sonoma State University, Faculty mentor: Derek Girman

We conducted a case study of captive DeBrazza's monkeys (Cercopithecus neglectus) where one of the individuals appeared to potentially display stereotypic behaviors associated with stress response (e.g. head rolling, circular locomotion, mouth movements) in captivity. We used a combination of behavioral monitoring, via ethogram, and fecal cortisol measurements to determine 1) whether the male DeBrazza's monkey displayed a significantly higher level of stereotypic behaviors relative to the other monkey in the shared enclosure, and 2) whether the male who displayed these behaviors at a high level showed evidence of chronic stress. We found that the male DeBrazza's monkey did display significantly higher level of stress-associated behaviors, however, we also found that the male did not show evidence of chronic stress or of levels significantly different from the female in the enclosure.

P26. Age Related Differences in Hematology and Serum Biochemistry In Captive Nyala Antelope, Amanda Neuweiler, Sonoma State University, Faculty mentor: Derek Girman

Establishing hematologic and biochemical reference values is essential for accurate health assessment in zoological and wildlife species. This study evaluated age- and sex-related variation in blood parameters of Nyala (Tragelaphus angasii) to establish baseline reference data for clinical interpretation. Under veterinary supervision, Nyala were sedated and blood samples were collected

aseptically and analyzed using a complete blood count (CBC) and serum chemistry panel. Statistical analyses were performed to determine the effects of age and sex on measured variables. Significant age-related differences were observed in mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), lymphocytes, symmetric dimethylarginine (SDMA), phosphorus, calcium, albumin, aspartate aminotransferase (AST), and bilirubin, while hematocrit, hemoglobin, mean corpuscular hemoglobin concentration (MCHC), eosinophils, basophils, monocytes, and red blood cell count showed no significant variation. These trends reflect normal developmental changes in erythropoiesis, immune maturation, renal function, hepatic protein synthesis, mineral metabolism, and muscle growth. Many of the observed patterns were consistent with established bovine hematologic and biochemical findings, supporting the use of comparative bovid physiology as a framework for interpretation. The establishment of baseline blood values in nyala provides a valuable clinical tool for zoological institutions to assess health status, monitor physiological development, and improve disease detection. Additionally, these reference ranges may aid in evaluating wild nyala populations, particularly in regions where tick-borne and other blood-borne diseases are prevalent, contributing to both clinical management and conservation health efforts.

P27. Maternal Stress Does Not Alter White Blood Cell Counts in Western Fence Lizard Offspring, Nathan Tsai, Kate Gutierrez, San Jose State University, Faculty mentor: David Ensminger

As ecological and environmental challenges increase for wildlife, they are experiencing increasing levels of stress. In addition to elevating glucocorticoids, these stress responses can alter white blood cell ratios in mammals and reptiles and change immunological resistance and cellular signaling. While previous work has shown maternal stress during gestation alters offspring physiology, little is known about the impact on offspring white blood cell balance, especially in reptiles. To examine this, we collected gravid fence lizards, gave them either control or glucocorticoid treatments, and then analyzed the white blood cell composition of the offspring. No changes were found between concentrations or ratios of the different white blood cells. While no significant results were found, these results help narrow down the causative agents for the changes in physiology we do find and allows us to better understand western fence lizard physiology and the effects of maternal stress.

P28. Impact of topical acaricide treatment of rodent hosts on ospC genetic diversity in *Borrelia burgdorferi*, Oliver Chiu, San Francisco State University, Faculty mentor: Andrea Swei

Lyme disease, caused by the bacterium Borrelia burgdorferi, is the most commonly reported vector-borne disease in the United States. The pathogen is maintained in the wild through transmission cycles involving ticks and infected reservoir hosts, such as rodents. Previous research has shown that treating rodents with topical acaricides in paired treatment and control plots

reduced tick densities with potential to lower Lyme disease risk in humans. The ospC gene is a major virulence factor in B. burgdorferi, and certain ospC genotypes are associated with more severe and invasive human infections. This study aimed to determine whether acaricide treatment changes ospC genotype diversity in B. burgdorferi-infected ticks. We hypothesized that the acaricide treatment would decrease ospC diversity. Tick samples were collected from three paired acaricide treatment and control plots in two Northern California counties. All ticks were screened for B. burgdorferi using a nested PCR assay. Positive samples underwent an additional nested PCR targeting the ospC gene and sequenced to determine ospC genotype. In 2024-2025, 679 ticks were screened for B. burgdorferi, of which 73 (10.8%) were positive. The ospC gene was amplified from 46 (63%) of 73 samples. Results form 2024 revealed ticks from Acaricide-treatment plots (n = 8) had three ospC genotypes, D (n=1); G (n=6); H (n=1), whereas ticks collected from control plots (n = 13) had four genotypes, D (n=2); G (n=1), H (n=7), H predominant mixed (n=1). These results suggest a decrease in ospC genotype richness in acaricide sites, compared to control sites, and a shift to genotype G in acaracide-treated ticks (p=0.003) by Fisher's Exact Test. Further analysis of 2025 samples is ongoing to confirm the impact of host-targeted acaricide treatment on genotype diversity and disease risk.

P29. Measuring Chronic Stress Through Hair Cortisol in a Latinx Community: A Collaborative Study by The Latina Center and Health Equity Research Lab, Yuliana Caravantes Molina, San Francisco State University, Faculty mentor: Cathy Samayoa

Latinx communities are underrepresented in biomedical research studies, yet disproportionately suffer from chronic health conditions and harmful exposures. These exposures include environmental and chronic stressors, such as racism and discrimination. Chronic stress may lead to dysregulation of the Hypothalamic-Pituitary-Adrenal axis (HPA) which has been associated with cardiovascular disease and type 2 diabetes, possibly driving health disparities. The objective of this study was to use community-engaged research approaches to collect biospecimens and measure hair cortisol concentrations among Latinx individuals. Hair samples were collected in a community setting utilizing hair collection kits assembled and provided by the Health and Equity Research (HER) Lab. The collection kits included an index card with a collection diagram, a small hair rubber band, tape, an envelope, and a label. Approximately 100 strands of hair were collected per participant. For hair cortisol quantification, samples were processed through hair washes and pulverization. About 10 mg of hair powder was used for the extraction of cortisol through an overnight methanol incubation. Next, the methanol was evaporated and samples were resuspended in a buffer. Cortisol quantification was performed using a commercially available Enzyme-Linked Immunosorbent Assay (ELISA). A total of 69 hair samples were collected, with all but 1 sample containing sufficient hair for analysis. We expect hair cortisol concentrations to be elevated in the Latinx samples. In conclusion, we used a community-engaged research approach to collect hair, a non-invasive biospecimen, among Latinx individuals in a community setting. Our work will

examine hair cortisol concentrations and measure their relationship to a variety of exposures relevant to chronic health conditions. This project will help us better understand the role of stress in addressing health inequities among the Latinx community.

P30. Infection and Natural History Impacts on Mammal Movement, Keira Denlinger, Grace Oram, San Francisco State University, Faculty mentor: Andrea Swei

Lyme disease is the most commonly reported vector-borne disease in the United States. It is transmitted by Ixodes species ticks that acquire the pathogen from feeding on infected reservoir hosts, such as rodents. Reservoir-targeted treatments aim to disrupt pathogen transmission between ticks and reservoir hosts by treating hosts with topical acaricides in baited treatment tubes. Such methods have been found to reduce host tick burdens, but it is not known how treatment affects mammal behavior or movement. To address this gap in knowledge, we surveyed three years (2023–2025) of trapping data across six sites in the San Francisco Bay Area to examine if acaricide treatment or other factors influence mammal movement. Animal movement was calculated from mammal trapping data and only included rodents captured at least two days during a field season. We conducted general linear model analyses to determine if the distance a mammal travelled was correlated with species, sex, acaricide treatment, or Lyme disease infection prevalence. We found that acaricide treatment made no significant impact on mammal movement. Additionally, mammal movement was not correlated with tick burden or Lyme disease infection status. Understanding reservoir host behavior can have important implications both for Lyme disease transmission and for the implementation of disease control interventions, such as acaricide or vaccine treatment, and should be better understood to improve the interpretation and application of these methods.

P31. Assessing Neurodevelopmental Risk of Real-World Air Pollution with a Fruit Fly Bioassay, Manh Le, Hartej Dhillon, California State University, Sacramento , Faculty mentor: Kimberly Mulligan

Air pollution is increasingly recognized as a risk factor for neurodevelopmental disorders (NDDs), including attention deficit hyperactivity disorder and autism spectrum disorder. These disorders arise from genetic, environmental, and gene–environment interactions that disrupt brain development, particularly during gestation. In the United States, approximately 18% of children are diagnosed with an NDD, with higher prevalence in low-income communities that also face elevated air pollution levels. This overlap highlights the need for practical tools to directly test whether local air pollution disrupts neurodevelopment and to inform public health policy. We aimed to develop a rapid bioassay using the fruit fly, Drosophila melanogaster, to evaluate whether ambient air exposures impair neurodevelopment. Fruit flies share key neurobiological features with humans and have a short life cycle, enabling rapid data collection.Methods/Results: We used

a larval locomotor assay to assess neurodevelopmental effects. Because larval locomotion depends on proper nervous system development, it serves as a sensitive indicator of disruption. Early- stage larvae were exposed for four hours at two field sites: one adjacent to a major freeway and a lower-pollution control site, based on California Air Resources Board monitoring data. Locomotor behavior was assessed 72 hours later to capture developmental rather than acute effects. Larvae exposed near the freeway showed a significant increase in reorientation events compared to controls, indicating altered sensorimotor processing. Peristaltic contractions and total distance traveled did not differ significantly, suggesting intact basic motor function. Approximately 30 larvae were analyzed per site.Conclusion: These findings support larval locomotor analysis as a sensitive biomonitor of air pollution–related neurodevelopmental effects. Increased reorientation without generalized motor impairment suggests disruption of sensory processing circuits. This scalable, low-cost model enables near-real-time assessment—within three weeks—providing a practical tool to strengthen evidence linking air quality to NDD risk and to inform environmental health decision-making.

P32. Acute Developmental Diesel Exhaust Exposure Alters Neurobehavior in Drosophila, Marisa Gonzalez, Rebekah Cook, California State University Sacramento, Faculty mentor: Kimberley Mulligan

Diesel exhaust (DE) is a widespread air pollutant linked to increased risk of neurodevelopmental disorders (NDDs), which involve impairments in motor, cognitive, and behavioral function. However, the specific concentrations, compositions, and exposure durations of DE that disrupt neurodevelopment remain poorly defined. To address this gap, we conducted a controlled exposure study to determine whether acute developmental exposure to DE alters neurobehavioral outcomes. We used Drosophila melanogaster as a model organism because it shares conserved genes and neural pathways relevant to human neurodevelopment and supports well-established behavioral assays.METHODS/RESULTS: Neurodevelopmental effects were assessed using larval locomotor activity and adult courtship behavior as functional endpoints. In collaboration with the California Air Resources Board (CARB), age-matched larvae were exposed for four hours to a high concentration of DE (900 $\mu g/m^3$), either as whole DE (gaseous and particulate components) or filtered DE (gaseous components only), allowing evaluation of distinct exhaust fractions. Behavioral outcomes were measured during larval and adult stages to assess persistent developmental effects. Larvae exposed to either filtered or whole DE exhibited increased locomotor activity, consistent with hyperactivity and disruption of sensorimotor circuit function. In adulthood, flies exposed to filtered DE, but not whole DE, displayed increased courtship behavior, suggesting that DE components differentially affect neural circuits underlying social behavior.CONCLUSIONS: Acute developmental exposure to DE produces persistent neurobehavioral alterations, with distinct phenotypes depending on exhaust composition. These findings indicate that specific components of diesel exhaust differentially impact developing neural

circuits and that behavioral effects can persist into adulthood. This study establishes Drosophila as a sensitive and scalable model for investigating the neurotoxic effects of air pollution and provides a flexible platform for testing a broader range of DE concentrations and compositions to better define neurodevelopmental risk.

P33. Identifying the Resistance Genes behind Chemotherapy Toxicity, utilizing *Drosophila melanogaster*, Sonia Gallegos, Jaquelyn A. Tafolla, University of California, Irvine, Faculty mentor: Anthony Long

The toxic side effects experienced by patients undergoing chemotherapy treatments vary greatly between individuals. This variation has a genetic component, implying the presence of specific genes that affect susceptibility to chemotherapy toxicity. Antimetabolites are a class of chemotherapeutic agents often used to inhibit cellular proliferation. Two such antimetabolite drugs are methotrexate (MTX) and gemcitabine (GEM). Drosophila melanogaster is an efficient model system for genetic testing due, in part, to the availability of genome-scale data, ease of genetic manipulation, potential for high-throughput assays, and controlled environments. We aim to leverage a high-throughput toxicity assay selecting for chemotoxicity-resistant D. melanogaster as a foundation for genomic analysis of chemotoxicity resistance. This would allow us to identify novel genes that both directly and indirectly mediate the toxic effects of MTX and GEM. We hypothesized that D. melanogaster would show dose-dependent sensitivity to both MTX and GEM, thus making the proposed toxicity assay feasible. We measured resistance to chemotoxicity as a larva's ability to survive into adulthood after exposure to MTX or GEM. Using this toxicity assay, we found a dose-dependent relationship between the concentration of each antimetabolite and the post-treatment survival rate. The preliminary results of our dose titration indicate that, for both MTX and GEM, a 2 mM concentration yields ≤5% survival to adulthood in the population. We plan to perform whole-genome pooled sequencing on the resistant D. melanogaster and utilize an "extreme quantitative trait loci" approach to identify genomic regions mediating this resistance. We hope to use this genomic data from the D. melanogaster model system to identify human orthologs that similarly affect chemotoxicity in humans. Our findings could advance personalized medicine and aid patients in determining the most effective chemotherapy regimen.
Acknowledgements - This work was supported by the NIH MARC U-STAR training grant (T34GM136498) and the NIH award R01-OD034064.

P34. Cholesterol Variation Across Thermal Environments in Grasshoppers, Ashley Brown, University of California, Berkeley, Faculty mentor: Caroline Williams

Variation in environmental temperature alters membrane fluidity and presents a fundamental physiological challenge for ectothermic organisms. Because membrane fluidity increases with

warming and decreases in the cold, organisms must regulate it within their distinct thermal environments. Membrane lipids contribute to membrane fluidity, and cholesterol can help offset increased membrane rigidity at low temperatures. However, the specific role of cholesterol in elevational adaptation in grasshoppers remains unclear. To examine this relationship, wild-caught grasshoppers collected from the Rocky Mountains of Colorado were used to quantify cholesterol concentrations in Melanoplus boulderensis, a cold-adapted alpine species, and Melanoplus sanguinipes, a warm-adapted species. We hypothesized that cholesterol concentrations would be higher in the cold-adapted species to maintain optimal membrane fluidity under colder conditions. Preliminary results support this hypothesis and indicate elevated cholesterol levels in the alpine species. This research suggests that cholesterol may contribute to thermal tolerance in insects and provides insight into physiological responses to changing thermal environments. Future laboratory experiments will rear M. sanguinipes under controlled thermal and dietary conditions to isolate the effects of temperature and nutrition on cholesterol levels.

P35. Adult wing morph in *Gryllus lineaticeps* is shaped by interacting seasonal cues late in juvenile development, Beepashna Bista, Devyn Clayton, University of California, Berkeley, Faculty mentor: Caroline Williams

Seasonality is a major source of environmental variation, shaping morphology, life history and timing of key developmental transitions. Many organisms rely on seasonal cues, such as temperature and photoperiod, to synchronize development with environmental conditions. However, climate change is disrupting these cues, potentially reducing their reliability and altering developmental outcomes. The variable field cricket, Gryllus lineaticeps, is a wing-dimorphic insect that produces two adult morphs with distinct life history strategies: a long-winged morph capable of flight that delays reproduction, and a short-winged morph that is flightless but matures earlier. Wing morph is an environmentally plastic trait, with morph frequencies varying seasonally. However, the timing of juvenile sensitivity to seasonal cues remains poorly understood in hemimetabolous insects, limiting our ability to predict how developmental decisions may shift under increasingly mismatched temperature and photoperiod regimes. Moreover, it remains unclear how these cues interact to determine adult wing morph. We tested two hypotheses: (1) that changes in seasonal cues during the final juvenile stage influence adult wing morph, and (2) that photoperiod has a stronger effect than temperature. To test our first hypothesis, we transferred individually-housed crickets between summer-like and autumn-like rearing environments at different points throughout the final juvenile stage. Each individual was assigned a transfer day, moved to the alternate condition on that day, and scored for wing morph at adulthood. To test our second hypothesis, cohorts of juveniles were reared under seasonally matched or mismatched combinations of temperature and photoperiod, and adult wing morph frequencies were

quantified.Our results indicate that wing morph determination remains sensitive to environmental conditions late into juvenile development, and that photoperiod and temperature interact to shape life history strategy. Our findings suggest that plastic developmental systems may shift as seasonal cues become increasingly decoupled under climate change.

P36. Impacts of wing morphology and sex on iron concentration and protein content in reproductive tissues of Gryllus crickets, Clara Medrano, University of California, Berkeley, Faculty mentor: Caroline Williams

Iron is an essential micronutrient that supports key biological processes, including muscle function and reproduction. In organisms exhibiting life history trade-offs, iron may serve as a limiting resource that constrains allocation between competing traits. The variable field cricket, Gryllus lineaticeps, is wing polymorphic and exhibits a tradeoff between flight and reproduction. Long-winged (LW) adults are flight-capable, investing in long wings and flight muscles, whereas short-winged (SW) adults forgo flight and allocate resources to reproduction. LW individuals later histolyze their flight muscles, potentially repurposing nutrients for reproductive development. I hypothesize that iron recycled from flight muscles supports reproductive investment following muscle breakdown. To test this, I quantified offspring production in LW and SW female crickets that were reared on control or iron-restricted diets. SW females on low iron diets had fewer offspring than SW on control diets, while LW fecundity was unaffected by iron restriction. These results support that flight muscles function as an iron reserve that buffers LW females against dietary iron limitation. Whether similar nutrient reallocation occurs in males remains unknown. Using BCA and Ferrozine iron assays, I am characterizing iron and protein content in reproductive tissues (spermatophore, testes, accessory glands) and flight muscles of LW and SW males throughout flight muscle breakdown. I predict that SW males, which invest in reproduction from early adulthood, will exhibit greater iron and protein content than LW males in their reproductive tissues. Together, this work investigates how nutrient recycling mediates sex-specific allocation strategies and provides mechanistic insight into the physiological basis of life-history trade-offs, especially between antagonistic traits.

P37. Integrating genomic and endocrine mechanisms of threshold trait expression in a wing polymorphic cricket, Ximena Prado, University of California, Berkeley, Faculty mentor: Caroline Williams

Dichotomous traits are often underlain by complex, multigenic architectures. A common framework to explain their expression is the threshold trait model, in which individuals vary in underlying continuous liability that is converted into a discrete phenotype by a threshold. Although this model applies to a wide range of polymorphic traits, including alternative life-history

strategies, our understanding of the genomic and physiological processes that shape threshold traits remains limited. Flight polymorphism in Gryllus crickets provides a classic example of a threshold trait and a system for investigating the genomic and physiological basis of a complex dichotomous phenotype. Adult individuals vary both in their morphology and life-history strategy: either they are long-winged (LW) and flight-capable but delay reproduction, or short-winged (SW) and flight-incapable but reproductively mature sooner. Wing morphology is both a heritable and environmentally plastic trait with an endocrine basis, particularly involving Juvenile Hormone (JH), a key developmental regulator in insect reproduction and wing development. In this study, we used Gryllus lineaticeps, the variable field cricket, to expand on previous genetic work and investigate the mechanistic basis of wing morph. Our lab has conducted an artificial selection experiment to create populations enriched for either wing morph, in parallel to quantitative crosses, all reared to generate a broad range of liabilities. Individuals of extreme liabilities will later be sequenced to identify genomic regions associated with wing morph. To build on the genetics, we will measure JH titers in all populations, testing whether endocrine regulation has responded to selection. We will also measure gene expression on JH biosynthesis genes in the corpus allata, the site of JH biosynthesis, to establish both genetic and regulatory responses to selection. By integrating previous genetic work and our physiological approaches, we aim to reveal the mechanisms that translate underlying liability into discrete morph expression.

P38. Cranking up the Heat: Evaluating Genetic Changes to Melanoplus sanguinipes Grasshoppers Under High-intensity Heat Stress during Juvenile Development, Annie Ta, Kikyo Makino-Siller, University of California, Berkeley, Faculty mentor: Caroline Williams

Heat waves, prolonged periods of excessively high temperatures, are increasing in both frequency and intensity as a consequence of climate change. These extreme thermal events impose substantial physiological stress on locally adapted insect populations and are expected to elevate mortality rates. Given these risks, there is an urgent need to evaluate their biological impact. Although understanding heat stress responses is essential for predicting organismal responses to climate change and informing conservation strategies, the effects of extreme heat on insect gene expression remain incompletely characterized. Here, we examined how stimulated heat waves during juvenile development influences transcriptomic responses to acute heat shock later in life in the low-elevation generalist grasshopper species Melanoplus sanguinipes using RNA sequencing. De novo assembly yielded 194,393 unigenes with a mean length of 737 base pairs; differential expression analysis identified 4,210 genes, representing adult transcriptional response to acute heat shock conditional on juvenile thermal experience. The Gene Ontology (GO) enrichment analysis of these genes revealed significant overrepresentation of genes associated with the biological process of wound healing among the differentially expressed set. The KEGG pathway analysis further identified enrichment of lysine degradation, a pathway linked to energy metabolism and cellular stress-response mechanisms. By integrating developmental thermal history with adult

transcriptional responses to heat stress, this study provides mechanistic insight into how early-life exposure to extreme temperatures shapes molecular strategies of stress resilience. These findings advance our understanding of developmental plasticity as a potential buffer against increasingly frequent heat waves under climate change.

P39. Fight or flight: alternative life-history strategies mediate iron-induced pesticide tolerance in a wing polymorphic cricket, Lily Donzeiser, University of California, Berkeley, Faculty mentor: Caroline Williams

Anthropogenic activities have introduced numerous xenobiotics into natural andagricultural ecosystems, subjecting organisms to co-occurring stressors. These multi-stressorenvironments may facilitate organismal cross-tolerance, where exposure to one stressor increasestolerance to another. Insect pesticide resistance is a complex agricultural challenge, yet is oftencontextualized as a consequence of over-application. Iron-based fertilizers are commonly appliedto address soil deficiencies, but excessive use can expose insects to physiologically disruptiveiron levels. Despite their widespread use, most studies examine these stressors in isolation,leaving the relationship between insect iron stress and pesticide tolerance poorly understood.Though pesticide resistance is also known to incur fitness costs, it remains unclear how cross-tolerance between stressors impacts life-history strategies.Wing-polymorphic insects provide a powerful system for studying alternative life-historystrategies within a single species. Adults emerge as either a long-winged, flight-capable morphthat delays reproduction, or a short-winged morph that matures earlier but has limited dispersalability. These alternative strategies reflect a fundamental trade-off between dispersal andreproduction.We examined the combined effects of iron exposure and bifenthrin, an insecticide, on thelife-history strategies and insecticide tolerance of Gryllus lineaticeps, a wing-polymorphiccricket. Crickets were exposed to iron, bifenthrin, or both stressors sequentially. We assessedmorph-specific effects on flight muscle function and ovary mass, while measuringcarboxylesterase activity as an indicator of a contaminant-induced physiological response.Iron exposure increased bifenthrin tolerance in the short-winged morph but decreasedtolerance in the long-winged morph, despite elevated carboxylesterase activities in both. Thesedifferences corresponded to reduced ovary mass in the short-winged morph and increased flightcapacity in the long-winged morph. Our findings demonstrate that metal exposure can facilitatepesticide tolerance, but cross-tolerance may depend on an insect’s life history strategy. Thishighlights the consequences of dual-agrochemical applications for pest management and insectlife-history, physiology, and population persistence.

P40. [WITHDRAWN] Juvenile Hormone and Morph Determination in *Gryllus lineaticeps*, Devon Konstanzer, University of California, Berkeley, Faculty mentor: Caroline Williams

Flight ability was an important insect adaptation that allowed for immense diversification, but it is an energetically costly process that has often resulted in metabolic trade-offs. Wing polymorphisms have frequently evolved as a way to circumvent the costs of flight by producing individuals that either prioritize flight or reproduction. Gryllus lineaticeps, the variable field cricket, is a wing polymorphic insect where the ability to become long-winged or short-winged depends on a number of environmental, genetic, and epigenetic factors—many of which have yet to be heavily studied. Juvenile Hormone (JH), an important hormone for insect development, regulates growth and is thought to play a role in wing morph determination—though the extent of its involvement in G. lineaticeps' morph determination is not yet known. We hypothesize that JH's temporal regulation plays a significant role in wing morph determination, with high levels of activity in JH signalling and response genes during juvenile development corresponding to short-winged morphs and low levels corresponding to long-winged morphs. To test this hypothesis, a gene expression timeline is being constructed on a suite of JH-related genes across a series of five time points in late juvenile development to gain an understanding of how JH production and signaling change during development and between short- and long-wing destined individuals. Preliminary results support this hypothesis, showing morph-specific and temporal regulation of the JH receptor gene Met, with higher expression present in short-wing destined individuals. This study will provide insight into how morph determination is hormonally regulated in G. lineaticeps.

P41. Sex differences in effects of mitochondrial genotype on response to stress in a montane leaf beetle, Peyton Stevens, Sonoma State University, Faculty mentor: Nathan Rank

Understanding how sex-specific genetic mechanisms influence organismal performance under stress is important for predicting evolutionary responses to environmental change. In this study, we investigated whether females respond to temperature and oxygen-related stress differently than males in the leaf beetle Chrysomela aeneicollis. We also tested whether mitochondrial–nuclear interactions contribute to sex-specific performance differences consistent with the "mother's curse" hypothesis. To reliably identify sex, we used sex-associated transcript expression from a rearing experiment conducted at natural elevation. A principal component analysis (PCA) of transcripts, including doublesex (dsx) and several Vg (vitellogenin) transcripts, clearly separated males and females based on duplicated loci for 87 of 96 beetle larvae. This demonstrated that transcript expression provides a reliable molecular method for determining sex in C. aeneicollis. We next examined the relationship between sex, genotype, and stress using running speed as a measure of performance. Beetles were tested under control conditions (20 °C) and after heat exposure (36 °C). For individuals carrying the southern (A) haplotype, running speed of males

following heat stress was lower than females. While both sexes experienced decreased performance at higher temperature, the decline was greater in males, suggesting sex-specific vulnerability to thermal stress. In contrast, beetles with the northern haplotype (C) exhibited similar reductions in performance for both sexes after heat exposure. This pattern is consistent with predictions of the mother's curse hypothesis, in which mitochondrial mutations inherited maternally can accumulate if they reduce fitness in males while benefiting females. We also detected sex differences in expression of genes related to mitochondrial metabolism among the 87 beetles examined. These results demonstrate that sex-specific responses to environmental stress depend on mitochondrial background.

P42. Heat shock protein evolution in a montane leaf beetle, Jacob Lapier, Sonoma State University, Faculty mentor: Nathan Rank

The willow leaf beetle, Chrysomela aeneicollis, inhabits the west coast of Northern America, particularly in regions at high elevation with substantial variation in temperature over the day and over a growing season. These circumstances, combined with temperature increases brought on by climate change, pose a challenge for the ability of this species to adapt to stressful conditions caused by temperature and limited oxygen supply. Expression of heat shock proteins allows beetles to recover from exposure to stressfully warm temperatures. In this study using Geneious software, we identified heat shock proteins from C. aeneicollis, analyzed them for locations of functional domains, and compared their predicted protein sequences to related taxa in order to understand the history of heat shock gene evolution in beetles, the largest group of insects on the planet. In particular, phylogenies were generated to compare the identified proteins to the well documented heat shock 70 proteins of the butterfly Melitaea cinxia to provide a framework for comparative analysis. We describe a detailed inquiry into the similarities and differences among C. aeneicollis heat shock proteins and quantify their expression pattern for beetle larvae that were examined in a large-scale performance experiment. We discuss the evolutionary implications of the proteins' comparison with other insects, showing a pattern of divergence from common ancestors.

P43. Morphological Comparison of Tarsal Claws & Adhesion Pads of high-mobility Ladybugs and primarily [flight] Bumblebee, Marisabel Perez Moreno, California State Polytechnic University, Humboldt, Faculty mentor: Frank Cappuccio

Claws, adhesion pads, and sensory structures mediate insect interactions with substrates, enabling walking, grasping, and defense while maintaining a grip on hard or smooth surfaces. They determine how insects interact with substrates and mediate transitions between walking and flight. However, structures vary in insects, and many do not share the same roles or specialized structures. Using Scanning Electron Microscopy (SEM), this study compares tarsal morphology in the

arboreal ladybird (Harmonia axyridis) and bumblebee (Bombus sp.) by measuring imaged claw curvature, angle, and the presence of adhesion pads to link form with ecological function. Comparative measurements show both H. axyridis and Bombus sp. possess longer hindclaws than those of their forelimbs, but differ in claw shape and limb geometry. H. axyridis foreclaws are long with moderate curvature, suited for walking and grasping foliage, while its shorter hindclaws are hyper-curved, likely to anchor their body during feeding or prey handling. Bombus sp. displays sharply curved claws with enlarged hindclaws, supporting their rapid transition and grip of the smooth floral surfaces during flight. Claw angles also differed: ladybug forelimbs had wider angles than their hindlimbs, suggesting a role in manipulation. Bees' sharply curved claws showed much larger forelimb angles, likely aiding in grooming and feeding behaviors. These structural adaptations/traits that are influenced by their behavior, environment, and evolution allow them to survive. Future work should examine more specimens, conduct biomechanical tests, look at cuticle structures, and run strength/grip tests to better link form to function.

P44. Antioxidant Response and Ferroptosis Susceptibility during flight muscle breakdown in *Gryllus lineaticeps*, Scott Morford, University of California, Berkeley, Faculty mentor: Caroline Williams

Ferroptosis, an iron-mediated form of cell death, has been a key mechanism in the current understanding of muscle breakdown diseases such as sarcopenia. However, it is not well understood what initiates or protects against ferroptosis in these diseases. The variable field cricket, Gryllus lineaticeps, breaks down its flight muscles as a natural part of its life cycle. During this process, mitochondria are broken down, and iron is released from the flight muscle, which could create a pro-ferroptotic environment in muscle cells. This would result in oxidative damage to the phospholipid bilayer and eventual cell death; however, unpublished HnE blots from our lab showed no evidence of oxidative damage during muscle breakdown. This suggests an underlying protective mechanism against oxidative damage. Using G. lineaticeps, we can help identify mechanisms that protect against oxidative damage caused by iron released during muscle breakdown. Using quantitative PCR, I analyzed the expression of antioxidant genes such as PRDX6, GPX4, Catalase, IDH, and SOD throughout muscle breakdown. Surprisingly, most antioxidant genes showed no significant change during muscle breakdown. However, Nrf2, a transcription factor involved in regulating the oxidative stress response, increased in expression during muscle breakdown. Nrf2 is not well studied in insects, but has been shown to upregulate mechanisms that protect against ferroptosis in addition to antioxidants, such as iron efflux transport proteins. Additionally, Evi5, a gene associated with cellular iron flux, was downregulated in muscle cells as breakdown progressed. This further suggests that the oxidative damage is associated with alterations to iron homeostasis and transport. Currently, treatments that manipulate ferroptosis focus on inducing or inhibiting the antioxidant response. However, the role of iron

transport and homeostasis may be more relevant to protecting against ferroptosis than previously considered, and should be strongly considered in future pharmacological studies.

P45. ddRADseq Genotyping and Analysis of a Hybrid Cross in the Romanian Cave Isopod *Asellus infernus*, Lubna Mulla, University of San Francisco, Faculty mentor: Meredith Protas

Cave animals live in dark environments with no light and limited food resources and they often lack traits such as pigmentation and eyes. The freshwater isopod Asellus aquaticus is present in lakes, streams and groundwater, and exists in both surface dwelling and cave dwelling forms or ecomorphs. This study investigates the genetic basis of the phenotypic differences between the two ecomorphs. Our goals include generating a linkage map of the Romanian cave population, Asellus infernus, mapping regions responsible for different phenotypic traits, and investigating whether the candidate gene efr3 is associated with any phenotypes of interest. An F2 population was generated from a cross between A. infernus and the Romanian surface population resulting in 120 F2 individuals. Double-digest restriction site–associated DNA sequencing (ddRADseq) was performed on parents of the cross and all 120 F2 individuals. Genome-wide single nucleotide polymorphism (SNP) markers were obtained, which will be used to construct a linkage map for the Romanian cave population and to provide a framework for quantitative trait locus (QTL) mapping. In addition, all F2 individuals were genotyped for efr3, a candidate gene which showed higher expression in cave individuals versus surface individuals and the cave allele was more highly expressed than the surface allele in F1 hybrids. Eye and pigmentation phenotypes in the F2 cross have already been measured and additional traits related to head shape and size will also be measured. Using the genotyping data, we will then map regions responsible for all of the measured phenotypes. Our goal is to examine the phenotypic correlation between eye loss and other morphological features of the head, aiming to better understand the developmental and evolutionary relationship among these traits.

P46. Genetic Basis of Eye Pigment Variation in a Croatian Cave Population of the Isopod *Asellus aquaticus*, Kasdin Hamel, University of San Francisco, Faculty mentor: Meredith Protas

The isopod crustacean, Asellus aquaticus, has multiple separate cave-dwelling as well as surface-dwelling populations, and is an emerging model organism for studying the mechanisms of evolution in the cave environment. These populations are widespread across Europe, and, throughout most of the populations, the surface populations are characterized by individuals with pigmentation and fully developed eyes, whereas the cave populations can be unpigmented or have reduced pigment and reduced or absent eyes. Previous work has focused on cave populations with extreme eye and pigment loss from Romania and Slovenia.The Susik Croatian cave population of Asellus aquaticus differs from other populations that have been previously looked at in that most

individuals look like surface individuals, with brown pigmentation and eyes. I compared the genetic basis of pigmentation in this Croatian cave population with what is known from the Slovenian populations to see if the same regions are associated with pigment-loss. A cross was performed between a lightly pigmented Croatian cave individual and a Romanian surface individual. F1s were intercrossed and surprisingly, the F2 had variable phenotypes, including two unpigmented males. One of the unpigmented F2s was crossed to another Romanian surface individual generating another set of F1s and then intercrossed to generate F2s. I genotyped 57 individuals in this cross for a genetic marker associated with the phenotype of no pigmentation in the Slovenian cave populations. I examined whether the genotypes of this gene were associated with presence versus absence of pigmentation in the cross generated with the Croatian Susik cave individual and found a significant association through the Fisher's Exact Test (p-value =6.069e-07). Therefore, it is likely that the same genetic region is responsible for the no pigment phenotype in both populations. These findings help contribute to our understanding of the mechanisms of repeated evolution in cave environments.

P47. The genetic basis of the red-eye phenotype in the Romanian surface population of the Asellus aquaticus species complex, Camila Mouteira, University of San Francisco, Faculty mentor: Meredith Protas

Model organisms are well-studied species used to obtain biological insight that could potentially be applied to other species with similar characteristics. Cave ecosystems have not been greatly analyzed, and their main model organism is Astyanax mexicanus (Mexican tetra), the cave fish. However, a new model organism for invertebrate cave-dwelling species is arising, Asellus aquaticus. We want to understand how changes in the eye and body pigmentation arose in different cave populations of Asellus aquaticus, with the question of whether genetic variation in the ancestral surface population could have been the source of variation now present in caves. A red phenotype, similar to a red phenotype uncovered in crosses with cave populations, was found within the Romanian surface population. Our goal was to investigate whether the region associated with the red phenotype in the cave populations was also associated with the red phenotype in the surface population. To address this, we are working with crosses of surface individuals from Croatia and red surface individuals from Romania, genotyping the F2 population to understand the relationship between the red phenotype and the genotype at pax2, which marks the region responsible for red in previously examined cave populations. We have found a correlation between red versus brown eye pigmentation and the genotype at pax2. Additionally, in another cross between red and brown Romanian surface individuals, we also saw an association between genotype and phenotype, indicating that the same region is associated with the red phenotype in the Romanian surface population and the red phenotype in the previously examined cave populations. Future work will examine whether the same gene or different but linked genes are responsible for the red phenotype in cave and surface populations.

P48. Inferring the evolutionary history of Pitx2 in left-right asymmetry using the threespine stickleback fish, Aja Teng, University of San Francisco, Faculty mentor: Sophie Archambeault

Comparative developmental biology and genetics are key tools that can reveal past evolutionary events. By studying a single gene's role across various organisms, its evolutionary history can be inferred. The bicoid-like family of Pitx transcription factors are present in all chordates, dating back 520 million years. Pitx2 plays several crucial roles during development, including proper left-right asymmetric development of internal organs. Mutations in Pitx2 lead to improper looping of the heart in model organisms like chick, mice, and frogs. However, the model zebrafish exhibited no effect upon Pitx2 knockout, suggesting its role in left-right asymmetry is absent. This raises the question of whether the role of Pitx2 in asymmetry (i) was shared in the common ancestor of mice and bony fishes but lost only in zebrafish or (ii) evolved more recently in the common ancestor of land vertebrates (mice, frogs, and chick). To answer this, we are analyzing the role of Pitx2 in the development of asymmetric organs (heart, liver, and pancreas) in threespine stickleback, a fish that diverged from zebrafish ~250 million years ago. We are using hybridization chain reaction (HCR) to test for Pitx2 mRNA presence in the left lateral plate mesoderm of stickleback, which drives looping of internal organs in chick. Next, we will analyze asymmetric heart looping and placement of liver and pancreas, between mutant Pitx2 knockouts and wild type samples to determine the necessity of Pitx2 in left-right asymmetry in stickleback. If Pitx2 is necessary for left-right asymmetry in stickleback, then this role of Pitx2 must be ancient, supporting hypothesis i. If Pitx2 is not necessary in asymmetry in stickleback, then this role likely arose in the group of land vertebrates, hypothesis ii. Either outcome informs us of the evolutionary history of Pitx2.

P49. Testing the causative role of Bmp6 in tooth number evolution in threespine stickleback, Serenity Isabella, University of San Francisco, Faculty mentor: Sophie Archambeault

Threespined stickleback's repeated invasion and adaptation to freshwater habitats over the past 12,000 years make it an excellent fish model for adaptive evolution. For instance, genetic crosses have identified Bone morphogenetic protein 6 (Bmp6) as a likely causative gene in tooth number evolution in the Paxton benthic (PAXB) stickleback population. Tooth number perfectly correlates with a set of six single-nucleotide polymorphisms (SNPs) in the fourth intron of Bmp6. Bmp6 is differentially expressed in high- and low-tooth stickleback, and mutations in Bmp6 result in fewer teeth. This data suggests that regulatory mutations affect the expression levels of Bmp6, thereby driving tooth gain in PAXB stickleback. However, the causal role of Bmp6 in tooth number

evolution has never been directly tested. We are using a reciprocal hemizygosity test to directly compare the phenotypic effects of the high- and low-toothed versions of Bmp6. We have knocked out Bmp6 in high- and low-toothed fish, and crossed them together, producing siblings that have a single functional copy of Bmp6 – either the high- or low-toothed copy. If Bmp6 is the causal gene, then fish with a single high-toothed copy should have more teeth than their sibling fish with a single low-toothed copy. If Bmp6 is not the causal gene, then these siblings will have the same number of teeth. Each fish is measured for length, genotyped for the functional copy of Bmp6, stained with Alizarin Red to highlight their bone, and dissected for their tooth counts. Tooth numbers are counted twice for accuracy, and additional counts are conducted if initial counts are inconsistent. We will compare the size-adjusted tooth counts between siblings with functional high- and low-toothed copies of Bmp6 to directly test the causal role of Bmp6 in tooth number evolution.

P50. Pitx2 in the Anterior Segment of Stickleback Eyes, Cassandra Raul, University of San Francisco, Faculty mentor: Sophie Archambeault

In humans, reduction of Pitx2 gene expression leads to anterior segment dysgenesis (ASD), characterized by symptoms such as thicker corneas, changes in interocular pressure, and pupil malformation. The role of Pitx2 in anterior segment development is conserved across vertebrates, as mutations in Pitx2 are linked to gross morphological changes in pupil size and placement, and volume of anterior chamber in mice, zebrafish, and threespine stickleback fish. This thesis tests the hypothesis that Pitx2 function is conserved across vertebrates at a cellular level. Specifically, mutations in Pitx2 in threespine stickleback are predicted to drive increases in corneal thickness, disorganization of the cells within the iridiocorneal angle, and smaller eye muscles. To address this hypothesis, eye development will be characterized across key stages using morphological and histological analyses. We will (1) characterize eye muscle development in whole mount embryos using phase contrast microscopy, (2) quantify corneal thickness in hematoxylin and eosin-stained sections (H&E), and (3) characterize the cellular organization of the iridiocorneal angle in H&E sections. These phenotypes will be compared between wild-type and Pitx2 mutant siblings to identify stage-specific deviations in tissue formation. Next, we will investigate the temporal and spatial distribution of Pitx2 during anterior segment morphogenesis in wild-type stickleback using hybridization chain reaction in whole mount and paraffin sections. Together, our results should demonstrate that malformation of the anterior segment in mutant sticklebacks correlates in time and space with expression patterns of Pitx2. This work will facilitate additional studies to understand the downstream genes and specific tissues affected by mutations in Pitx2.

P51. Exploring the Genetic Basis of Plumage Polymorphism in the Newly Described Endemic Galapagos Lava Heron (*Butorides sundevalli*), Kiarra Enberg, Nikita Paulose, San Francisco State University, Faculty mentor: Jaime Chaves

The lava heron (Butorides sundevalli) is a recently described endemic species of the Galápagos Islands characterized by notable variation in plumage coloration, ranging from dark to brown "striated" forms. Most individuals display a striking dark gray plumage, which is hypothesized to function as camouflage while foraging along volcanic dark shorelines. Variation in plumage coloration has been linked to genetic changes in melanocortin-1 receptor (MC1R), a gene involved in melanin production. MC1R regulates the synthesis of eumelanin, the pigment responsible for brown and black coloration, and its expression can vary depending on specific mutations within the gene. Here, we examined sequence variation in MC1R across different plumage variants of the lava heron to investigate the genetic basis of this coloration polymorphism. We first isolated the MC1R sequence from two dark forms and one striated form. Sequences extracted from complete genomes contained the expected 945 bp, including the start and stop codons. Alignment showed that the MC1R sequences of the two dark individuals were identical, while comparison with the striated individual revealed seven single-nucleotide polymorphisms (SNPs). Translation of these sequences indicated that three of these SNPs resulted in non-synonymous amino acid substitutions: D37N, V103M, and V126I. Dark individuals carried nucleotide A at the first position in all three non-synonymous codons. Notably, variation at position c.376 (G→A), resulting in the V126I substitution, has also been reported in the MC1R sequences of Ayam Cemanis and Barn owls' rufous plumage, but the other substitutions have not been documented in avian systems. The remaining four SNPs did not result in amino acid changes. These results suggest a potential association between dark plumage and genetic variation in the MC1R gene in the lava heron. Because plumage coloration is a polygenic trait, further studies with additional pigmentation genes are needed to confirm this relationship.

P52. Identification of a Fragmentary Pterosaur Humerus from the Lance Formation (Upper Cretaceous), Wyoming, Lois Amaya, Rachel Dombrowski, The Master's University, Faculty mentor: Matthew A. McLain

Fragmentary fossils can often obstruct our knowledge of the fossil record, but with careful anatomical comparison, unidentified bone fragments can ultimately contribute to paleontology and taxonomic interpretations. We examined an unidentified hollow bone fragment (HRS 29372) housed at Southwestern Adventist University that was unearthed in the Gar Ridge quarry in the Lance Formation (Upper Cretaceous) on the Hanson Ranch in Niobrara County, Wyoming in 2019. The specimen is a small distal end of a left humerus, measuring a maximum of 4.00 cm in length, 2.20 cm in width, and 1.00 cm in thickness. We observed that the bone contained a distally extended trochlea, a compressed humeral shaft, a well-developed ectepicondyle and entepicondyle,

and a bounded brachial depression. Furthermore, in anterior view, the trochlea has a sub-triangular shape and the capitulum is lozenge-shaped and inclined with the medial side pointing more proximally. We then attempted to determine the taxonomic identity of the specimen. After doing careful systematic comparison with various taxa with similar distal humeri, specifically with birds, non-avian theropods (ornithomimosaurs, caenagnathids, troodontids, and dromaeosaurids), and pterosaurs, we discovered significant differences which aided in the identification of our specimen. Through this process, we concluded that the specimen came from a species of pterosaur because of its prominent expanded epicondyles. Additionally, there is a small ridge along the outer surface of the entepicondyle bounding a depression, similarly seen in some pterosaurs (e.g., Archaeoistiodactylus), which reinforces the pterosaurian identification. Through this identification we can contribute to the diversity and recognition of pterosaurs in the uppermost Cretaceous deposits of Western Northern America. Ultimately, it is very useful to study the comparative anatomy of various species because it gives us a better understanding of the wide range of anatomical structures present among different creatures in the fossil record.

P53. A High-Throughput Luria–Delbrück Framework for Measuring Sequence-Dependent Mutation Rates in Escherichia coli K-12, Xin Cen, Lisa Li, University of Washington, Faculty mentor: Benjamin Kerr

Mutations generate the genetic variation that drives evolution, yet the factors determining why some genomic sites mutate more frequently than others remain poorly understood. Using a next-generation sequencing (NGS) extension of the classical Luria-Delbrück fluctuation assay, our lab previously estimated mutation rates for rifampicin-resistance mutations in the rpoB gene of Escherichia coli K-12. These preliminary data revealed striking heterogeneity in mutation rates: identical base substitutions occurring at nearby positions differed in frequency by nearly two orders of magnitude. Notably, a high-rate A→G transition occurs at a site within the sequence motif GACC. Reanalysis of prior mutation datasets from multiple genes and genome-wide studies shows that this same motif is repeatedly associated with elevated A→G mutation rates, suggesting that local DNA sequence context may directly influence mutational processes. Here, we test the hypothesis that the GACC motif promotes A→G mutations at its central base. To isolate the effect of sequence context from protein function, we engineer synonymous mutations in the rpoB gene that alter the surrounding DNA sequence without changing the encoded amino acid. Specifically, we disrupt the GACC motif at a known high-rate site and introduce this motif at a nearby position where it does not naturally occur. Using our high-throughput NGS-based fluctuation framework, we will quantify mutation rates across these engineered contexts. We expect that introducing or disrupting the GACC motif will significantly alter A→G mutation rates. This work establishes an experimental framework for directly testing sequence-dependent mutation processes and will clarify how local DNA environments shape mutation rate variation in bacterial genomes.

P54. Investigating how host metabolism drives adaptive evolution in bacteriophage T7 using a spectrophotometric assay. Ryan Mun, Institution, Faculty mentor:

The infection dynamics of bacteriophages, viruses that infect bacteria, depend on the metabolic state of their bacterial hosts, which in turn can be altered by nutrient availability. Understanding how host metabolism influences viral infection provides insight into microbial community dynamics with applications in medicine and biotechnology. Here, I investigated how different carbon sources affect the time required for bacteriophage T7 to eradicate its host, Escherichia coli IJ1133. Hosts were grown in minimal media supplemented with either glucose or mannose to induce metabolic differences and infected with T7 across multiple phage-to-bacteria ratios in a 96-well microtiter plate. Bacterial growth was monitored using spectrophotometry at OD600. From growth curves, I quantified the time of maximum bacterial density (tmax) and the time of bacterial extinction (text).In uninfected bacterial-only controls, the OD600 increased exponentially before plateauing as nutrients were depleted. In glucose, the bacteria plateaued at a higher maximum density (OD600 = 0.671 at ~10 hours) than in mannose (OD600 = 0.573 at ~15 hours). Upon infection with T7 bacteriophages, the OD600 initially peaked and then declined as the bacterial population was eradicated. Increasing phage input reduced peak bacterial density in both glucose and mannose. However, extinction times varied across phage-to-host ratios. Glucose-grown cultures generally reached extinction earlier (OD600 = 0.423 at 11.1 hours) than mannose-grown cultures (OD600 = 0.319 at 12.2 hours) across all phage input concentrations, following our hypothesis that text in glucose would be quicker than in mannose.These infection dynamics suggest that host metabolic state plays a dominant role in shaping phage infection outcomes. The high-throughput spectrophotometric assay establishes a baseline for experiments examining how bacteriophages that have evolved in distinct metabolic environments adapt their infection strategies, allowing us to test many genotypic variants simultaneously.

P55. Impact of Circadian Gene ARNTL/BMAL1 Knockdown on Inflammatory Responses in Healthy and Asthmatic Epithelia, Maria Kang, University of Washington, Faculty mentor: Weston Powell

Asthma is an inflammatory disease characterized by airway inflammation, hypersensitivity, and narrowing. Symptoms of asthma and airway resistance increase at night, yet the molecular mechanisms underlying time-of-day differences remain unclear. The differences are hypothesized to be regulated by the circadian clock in airway epithelia and, in animal studies, have been linked to the core circadian gene ARNTL/BMAL1. We induced BMAL1 knockdown in organotypic airway epithelial cells to examine how circadian clock disruption would impact inflammatory responses. We hypothesized that secreted inflammatory protein levels would be reduced after BMAL1 knockdown. Airway epithelial cells from both healthy and asthmatic pediatric donors

were transfected with a lentiviral construct containing a doxycycline-inducible shRNA targeting BMAL1, and differentiated to an organotypic, pseudostratified layer at an air-liquid interface. BMAL1 knockdown was induced 21-28 days after differentiation for 7 days. RT-qPCR was used to measure gene expression of ARNTL, NR1D1, NR1D2, CXCL5, CXCL6, and CXCL10. Western blot analysis was used to quantify BMAL1 protein levels. Measurement of secreted CXCL8 and CXCL10 was performed using ELISA. ARNTL/BMAL1 gene expression and protein levels were reduced 60-80% in 7 primary cell lines. NR1D1 and NR1D2 gene expression was reduced 40-60% after knockdown. CXCL5 and CXCL10 increased gene expression with knockdown, while CXCL6 showed variable expression changes post-knockdown. Our work reveals the circadian role in innate immune gene expression which may contribute to time-of-day changes in airway function. Future work is needed to assess whether BMAL1 knockdown alters responses to infectious stimuli, such as viral infections.

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P56. Installation of electrophilic alkyl amides and sulfonamides modulate the anticancer activity of palbociclib and ribociclib towards achieving isoform-selective CDK inhibition, Alyssa Liu, Srinidhi Venkatesh, Aspiring Scholars Directed Research Program, Faculty mentor: Edward Njoo

Given their central role in aberrant cell cycle regulation found in many cancer types, cyclin dependent kinases (CDK's) have become an increasingly important target in the development of anticancer therapeutics. Recent efforts in this space have led to the discovery and development of palbociclib and ribociclib, two structurally homologous inhibitors of the closely related isoforms CDK 4 and 6. While these compounds have demonstrated remarkable clinical efficacy, potency, and safety tolerability profiles, their limited selectivity between CDK 4 and 6 posed unique target tolerability challenges. To address this challenge, and in efforts towards the development of similar compounds with improved CDK 4 and 6 isoform discrimination, several have elaborated a solvent exposed piperazine handle to either install a covalent warhead or other functional motifs that may imbue greater isoform selectivity. Inspired by these and other approaches, our laboratory has prepared a series of alkyl, alkyl halo, alkynyl amide, and sulfonate derivatives of palbociclib and ribociclib. One lead compound, bearing a brominated alkyl sidechain, exhibits a remarkably different in vitro potency profile compared to palbociclib, its parent compound. Among several analogs prepared, palbociclib exhibited an IC50 of 9.17 μM in HEK293 cells, while its 4-azidobutyrate amide was found to be slightly more potent, with an IC50 of 5.35 μM. This initial hit has prompted the synthesis of palbociclib and ribociclib derivatives in MDA-MBA-231 human breast cancer cells. Here, we disclose the chemical synthesis of over a half dozen palbociclib and ribociclib derivatives with both alkylating and non-alkylating motifs on their shared piperazine handle as well as current progress towards isoform selective biological applications of these compounds as therapeutic leads for the treatment of cancer.

P57. A-ring Analogs of Andrographolide Exhibit Anti-cancer Activity Through the Potentiation of the Wnt1 Pathway in Human and Murine Colon and Breast Cancer Cell Lines, Ruirui Liu, Abigail Yee, Aspiring Scholars Directed Research Program, Faculty mentor: Edward Njoo

Andrographolide, a labdane diterpenoid isolated from the plant Andrographis paniculata, has been previously reported as a natural product lead with unique anticancer activity, putatively through modulation of NF-kB and Wnt1 signaling. Synthetic modifications to its A-ring have been demonstrated to augment the anticancer activity of andrographolide and its analogs, specifically in connection to Wnt1 signaling. We previously reported that andrographolide A-ring trityl and silyl ethers are far more potent than the natural product in inhibition of human colon and breast cancer models (Gu, et al. Bioorganic and Medicinal Chemistry Letters 2025). In analogous murine cancer cell lines, which exhibit fundamentally different mechanisms of regulation of canonical Wnt1 signaling, we observed similar trends in anticancer potency in murine colon carcinoma, breast cancer, and melanoma cell lines as in their human counterparts. Finally, to access a natural co-isolated A-ring oxetane, we developed a five-step sequence with three chromatographic purifications to afford the A-ring oxetane in #% overall yield. This was evaluated alongside a broader library of cyclic A-ring analogs, many of which were explored for the first time in human and murine cancer models.

P58. Flavonoid quercetin and its glucuronide and sulfate conjugates bind to 67-kDa laminin receptor and prevent neuronal cell death induced by serum starvation, Aidan Carillo, University of Southern California, Faculty mentor: Rayudu Gopalakrishna

Quercetin, a common dietary flavonoid, is recognized for its protective properties against various neurodegenerative diseases, yet its precise mechanisms remain elusive. Following oral intake, quercetin is rapidly metabolized into its conjugates, quercetin-3-O-glucuronide (Q-3-G) and quercetin-3-O-sulfate (Q-3-S), which reach only low nanomolar concentrations in the brain. Given their limited antioxidant capacity at such levels, this study investigated whether these metabolites provide neuroprotection by acting as high-affinity ligands for the 67-kDa laminin receptor (67LR), a receptor previously linked to the green tea polyphenol epigallocatechin-3-gallate (EGCG) Methods: Binding affinities of quercetin, Q-3-G, and Q-3-S were evaluated using intrinsic tryptophan fluorescence quenching of "peptide G" (67LR residues 161–180) and molecular docking simulations. Functional neuroprotection was assessed in Neuroscreen-1 cells under serum starvation. To confirm the role of 67LR, a receptor-blocking antibody was utilized during treatment. Results: Spectroscopy and docking confirmed that quercetin and its conjugates bind to the peptide G region with high affinity, comparable to EGCG. In cell culture, Q-3-G and Q-3-S (1–10 nM) demonstrated significantly higher neuroprotective efficacy than the quercetin aglycone or EGCG. This effect was substantially neutralized by the 67LR-blocking antibody, indicating a receptor-dependent mechanism. Furthermore, the use of a β-glucuronidase inhibitor did not abolish Q-3-G's effects, suggesting the intact conjugate is the active mediator. Discussion/Conclusion: These findings indicate that quercetin's neuroprotective benefits are primarily driven by its metabolites binding to 67LR. By triggering survival signaling at physiological concentrations, 67LR serves as a critical high-affinity receptor for dietary flavonoid conjugates.

P59. High-Throughput Identification of Small-Molecules of the FLCN-FNIP2 GAP Complex, Haeyoon Cheong, University of California, Berkeley, Faculty mentor: James H. Hurley

The Folliculin:Folliculin Interacting Protein 1/2 (FLCN:FNIP1/2) complex functions as a GTPase-activating protein (GAP) for Rag GTPase-activating protein on the lysosomal membrane, playing a central role in lysosomal nutrient sensing. Dysregulation of the FLCN:FNIP1/2 complex has been linked to cancer and neurodegenerative diseases. Here, we describe a GAP-assay based screening platform to identify small molecules that target the FLCN:FNIP2 complex and modulate its GAP activity. After optimizing a robust biochemical GAP assay, we performed high-throughput screening of an ~5,000-compound library, yielding ~1.5% hit rate. Confirmed hits with high potency were further validated through orthogonal approaches, including structure-based virtual screening with Boltz2 and thermal shift assays. One compound (UCB-hit1) passed all selection criteria and exhibited an IC_{50} of ~8 μM, and has advanced to cell-based functional studies. Together, this work establishes a FLCN:FNIP2-targeted drug discovery platform and lays the groundwork for developing therapeutics for lysosomal malfunction diseases.

P60. P-gp Inhibition and Anti-Oxidative Effects on Cyclosporine by Hesperidin and Hesperetin, Aviva Iyerkhan, West Valley College, Faculty mentor: Terry Ng

The P-glycoprotein (P-gp) is a transporter protein that plays a crucial role in regulating movement of molecules through the epithelium in organs such as the liver, kidneys, and the small intestine, as well as through the blood-brain barrier. While P-gp has been found to reduce the bioavailability of various therapeutic compounds by actively effluxing them, its effects are speculated to be lessened due to substrate inhibition at higher drug concentrations. In this study, we combine potential P-gp substrates hesperidin, hesperetin, and cyclosporine in a Caenorhabditis elegans (C. elegans) model using MTT and liquid thrashing assays. Hesperidin is a polyphenol, found abundantly in citrus fruits and studied for its antioxidant, anti-inflammatory, antibacterial, and anticancer properties. However, its absorption is limited due to the disaccharide rutinose in its structure, hindering its diffusion through semi-permeable membranes during digestion. Consequently, we studied both hesperidin and its aglycone form hesperetin, which does not contain the rutinose sugar in its structure, in an attempt to enhance their effects. We tested these compounds in combination with cyclosporine, another P-gp substrate derived from the fermentation of the Tolypocladium inflatum fungus known for its immunosuppressive properties. As cyclosporine usage often induces oxidative stress on organs such as kidneys, we utilized hesperidin and hesperetin to attempt to reduce its toxicity. Our liquid thrashing and MTT assays found that both hesperidin and hesperetin reduced cyclosporine's toxicity, with hesperidin inhibiting it to a greater extent. These findings

highlight a possible novel delivery method of cyclosporine that minimizes oxidative stress while maximizing bioavailability.

Iain Davies

P61. Kinetic characterization of the interaction between CAF-1 and PCNA, Iain Davies, Institution, Faculty mentor:

The assembly of nucleosomes on DNA is essential for proper gene regulation, including the maintenance of gene silencing or expression. Nucleosome assembly is facilitated by histone chaperones in either replication-coupled or replication-independent processes. Two proteins crucial for replication-coupled nucleosome assembly are the histone chaperone protein, chromatin assembly factor 1 (CAF-1), and the sliding clamp protein, proliferating cell nuclear antigen (PCNA). PCNA encircles DNA during replication and recruits and regulates the activities of a multitude of proteins. Immediately after DNA synthesis, PCNA recruits CAF-1 to deposit histones on the nascent DNA, which serves as a crucial first step in nucleosome assembly on silenced regions of the genome. Despite the crucial role that CAF-1 and PCNA serve in proper gene silencing, it is unknown how the interaction between these two proteins mediates nucleosome assembly. We are working towards characterizing the binding kinetics of the interaction between CAF-1 and PCNA through surface plasmon resonance. Results show multiple binding regions exist within these proteins that bind with low micromolar affinity. This suggests the interaction between PCNA and CAF-1 is multivalent, and the binding regions have differing functions during nucleosome assembly. Together with structural data, our results provide insights into how PCNA might regulate the activity of CAF-1 during DNA replication and nucleosome assembly to maintain gene expression.

P62. Characterization of Protein Interactions Involved in Gene Silencing Using NanoDSF, Stephanie Limaye, Creighton University, Faculty mentor: Lynne Dieckman

Efficient packaging of DNA in the nucleus is necessary for proper gene expression. First, the DNA double helix is wrapped around histone proteins to form nucleosomes. These nucleosomes then condense further into chromatin. The deposition of these histones occurs immediately following DNA replication via a process called replication-coupled nucleosome assembly. This process requires proliferating cell nuclear antigen (PCNA) and chromatin assembly factor 1 (CAF-1). PCNA is a homotrimeric ring that acts as a sliding clamp by binding DNA and recruiting proteins to the replication fork for multiple DNA-templated processes. CAF-1 is a histone chaperone protein that is recruited to DNA by PCNA and deposits histones on newly replicated DNA. The interaction between PCNA and CAF1 is required for replication-coupled nucleosome assembly;

however, the mechanism of interaction between these proteins is not fully understood. We sought to determine if residues adjacent to a currently known PCNA-interaction site in CAF-1 also contribute to binding PCNA. Using nano-differential scanning fluorimetry, we have observed additional interactions between CAF-1 and PCNA, indicating PCNA-CAF1 interactions occur in multiple locations on CAF-1. These results, combined with structural studies in our lab, will advance our understanding of how PCNA and CAF-1 function together to carry out replication-coupled nucleosome assembly.

P63. Thermodynamic studies of protein-protein interactions involved in nucleosome assembly, Carly Nowoj, Creighton University, Faculty mentor: Lynne Dieckman

Within the nucleus, DNA must be properly compacted into nucleosomes to ensure genetic stability and proper gene silencing. Replication-coupled nucleosome assembly is the process where DNA is packaged into nucleosomes immediately after DNA synthesis. This process requires two proteins: proliferating cell nuclear antigen (PCNA) and chromatin assembly factor 1 (CAF-1). PCNA binds to and encircles DNA, acting as a scaffold to recruit proteins for various DNA replication and repair pathways. CAF-1 deposits histone proteins on newly synthesized DNA and is recruited to the replication fork by PCNA. The direct interaction between PCNA and CAF-1 is required for nucleosome assembly at silenced regions of the genome; however, the mechanism of this interaction is unclear. There is one known site of interaction between PCNA and CAF-1 that our lab has characterized. Additionally, our lab has identified a secondary binding site between these two proteins using a qualitative protein-protein binding assay. The goal of my project is to characterize this novel, secondary interaction at the thermodynamic level using isothermal titration calorimetry (ITC). Thus far, data show the affinity of this secondary interaction is lower than that observed for the known binding interaction between PCNA and CAF-1, suggesting the PCNA-CAF-1 interaction may require multivalent binding sites for high affinity binding. By combining this thermodynamic information with structural and kinetic data, we will fully understand the interaction between PCNA and CAF-1 and gain insight into how PCNA and CAF-1 work together to regulate replication-coupled nucleosome assembly and thereby gene expression.

P64. Cysteine-Scanning and Site-Directed PEGylation of MdtM, a Multidrug Exporter of Escherichia coli: Role of Helix I, Christine Trieu, Nathalie Frontela, California State University, San Bernardino, Faculty mentor: Xiaoxu Jiang

MdtM is an H^+-dependent multidrug antiporter located in the cytoplasmic membrane ofEscherichia coli that exports a wide range of drug molecules from the cytosol and therebyconfers antibiotic resistance [1]. Gene sequence alignment and in silico three-dimensionalstructure prediction

suggest that MdtM consists of twelve transmembrane helices arranged intotwo six-helix bundles [2]. However, to date, structure–function studies of MdtM remain limited,and the mechanism of H^+-coupled drug efflux mediated by this transporter is still unclear. HelixI of MdtM is predicted to line the substrate translocation pathway and contains two highlyconserved residues, Asp22 and Arg30 [3], which may play important roles in drug efflux. In thisstudy, to elucidate the structural and functional role of helix I, cysteine-scanning mutagenesiswas applied to helix I, and the accessibility of each cysteine substitution was assessed in wholecells using site-directed PEGylation in the absence and presence of substrate. This scanningapproach identified solvent-accessible positions in helix I and revealed a substrate-inducedconformational change within this helix. Our data support the hypothesis that helix I lines thesubstrate translocation pathway on the N-terminal bundle side and undergoes a rigid-bodymovement that alternately opens and closes the pathway on the periplasmic side of MdtM. References:[1] K. Nishino, A. Yamaguchi, Analysis of a Complete Library of Putative Drug TransporterGenes in Escherichia coli, J Bacteriol 183 (2001) 5803–5812.https://doi.org/10.1128/JB.183.20.5803-5812.2001.[2] C.J. Law, K.O. Alegre, Clamping down on drugs: the Escherichia coli multidrug effluxprotein MdtM, Research in Microbiology 169 (2018) 461–467.https://doi.org/10.1016/j.resmic.2017.09.006.[3] S.R. Holdsworth, C.J. Law, Functional and biochemical characterisation of the Escherichiacoli major facilitator superfamily multidrug transporter MdtM, Biochimie 94 (2012)1334–1346. https://doi.org/10.1016/j.biochi.2012.03.001.

P65. Substrate-Induced Conformational Change of Helix VII in MdtM, a Multidrug Efflux Pump of Escherichia coli: A Substituted Cysteine Accessibility Study, Francisco Diaz, Notfaza Karim, California State University, San Bernardino, Faculty mentor: Xiaoxu Jiang

Nowadays, multidrug resistance (MDR) has become a serious challenge in the clinical treatmentof bacterial infections. A group of drug efflux pumps embedded in the cytoplasmicmembrane of bacterial cells can actively export drug molecules from the cytosol, therebycontributing to MDR. MdtM is a proton-dependent multidrug efflux pump that belongs to theMajor Facilitator Superfamily (MFS). It is commonly found in the cytoplasmic membrane ofGram-negative bacterial species, such as Escherichia coli. When expressed, MdtM confersresistance to a variety of antibiotics, cytotoxins, and metal ions. In addition, MdtM isinvolved in bacterial tolerance to alkaline conditions. Based on the predicted structural modelof MdtM, the transporter is proposed to contain twelve transmembrane helices. Among these,helix VII is believed to contribute to the formation of the substrate translocation pathway on theperiplasmic side. To assess the solvent accessibility and dynamics of helix VII in MdtM,cysteine-scanning mutagenesis was performed on helix VII, and the accessibility of eachcysteine substitution was tested using the membrane-impermeant reagent methoxy polyethyleneglycol–maleimide-5000 (mPEG-Mal-5K) in the absence and presence of chloramphenicol, ahigh-affinity substrate of MdtM in vivo. Our PEGylation data identified residues in helix VII thatface the hydrophilic substrate translocation

pathway and also revealed a global conformationalchange in helix VII upon substrate binding. These findings support the alternating-accesstransport model proposed for

P66. Substituted cysteine accessibility method (SCAM) analysis of the conformational dynamics of the plug domain in FadL, Evangelina O'Leary, California State University, San Bernardino, Faculty mentor: Xiaoxu Jiang

FadL is a long-chain fatty acid (LCFA) outer membrane transporter that is representative of transporter proteins that transport hydrophobic molecules across the outer membrane (OM) of Gram-negative bacteria. The structure of FadL features a 14-strand transmembrane β-barrel that generates a pore through the outer membrane. Inside the β-barrel, the first 42 N-terminal amino residues form a plug domain that fully blocks the opening of the barrel. The substrate transport mechanism of FadL is generally unclear. The proposed models of FadL transport include the classical transport model and the lateral diffusion model. In this study, we investigate the conformational rearrangement of the plug domain during LCFA transport in Escherichia coli using the substituted cysteine accessibility method (SCAM). We used a GFP-based site-directed PEGylation method to investigate the movement of the plug domain. All 42 amino residues in the plug domain were switched to cysteine individually and the accessibility of each cysteine was evaluated in the presence and absence of the substrate sodium oleate using site-directed PEGylation and in-gel GFP fluorescence. The observed GFP fluorescence reveals a significant increase in solvent accessibility of the plug domain in the presence of substrate, indicating a major conformational rearrangement through the plug domain takes place when the substrate binds FadL.

P67. Investigating the Roles of Platelet- Derived Growth Factor Receptors (PDGFR) Using De Novo Designed Proteins, Mrityunjay Joshi, Alexia Williams, University of Washington, Faculty mentor: Hannele Ruohola-Baker

Precise therapeutic targeting of Platelet-derived growth factor receptor (PDGFR) signaling, a master regulator of embryonic development, angiogenesis, pericyte formation and key driver of fibrosis, remains elusive due to the promiscuity of native ligands. To avoid the constraint of unstable, immunogenic and poor pathway specificity posed by native ligands, we investigate a targeted platform of de novo designed protein antagonists and activators that achieve isoform-specific signaling. Using RFdiffusion-based machine learning, we generated small monomeric binders that competitively inhibit native ligand-mediated receptor dimerization. By assembly onto structured oligomeric scaffolds or connections through flexible linkers, these binders were converted into potent, dimer-specific activators capable of selectively triggering

PDGFRαα, PDGFRββ, or heterodimeric PDGFRα/β signaling as validated in Chinese Hamster Ovary (CHO) cells overexpressing the different human PDGFR isoforms through quantitative western blotting for downstream phospho-effectors.In iPSC-derived endothelial systems (i-ENDO), we show that selective PDGFRβ inhibition reduces pericyte populations, suggesting its targeted activation could offer a route to promote pericyte recovery in microvascular diseases like diabetic retinopathy. Furthermore, we find that selective PDGFRβ inhibition enhances myogenic differentiation from human foreskin fibroblasts (HFFs), which may represent a precise strategy for mitigating muscle degenerative diseases.Our findings demonstrate that synthetic, dimer-specific ligands can enable activation of specific receptor tyrosine kinase (RTK) networks, providing a powerful framework for targeted regenerative medicine.

P68. Depth-Resolved Effects of Myoferlin C2A–C2B on Membrane Packing Using Dipyrene-PC Fluorometry, Jordan Indrawan, James Warner, Oregon State University, Faculty mentor: Colin Johnson

Ferlins are large, multi-C2 domain proteins critical to membrane remodeling: dysferlin mediates skeletal muscle repair, otoferlin drives synaptic vesicle exocytosis in cochlear hair cells, and myoferlin regulates membrane fusion and trafficking. Although several C2 domains bind membranes, their effects on bilayer packing and fluidity remain largely unknown. Building on our previous finding that the otoferlin C2A–C2B linker associates with lipid bilayers, we use dipyrene-phosphatidylcholine (dipyrene-PC) fluorometry to map depth-dependent changes in bilayer organization induced by myoferlin C2AB, with future experiments intended for otoferlin C2AB and dysferlin C2AB. Excimer/monomer (E/M) intensity ratios at defined acyl-chain positions enable depth-resolved mapping of how myoferlin C2AB perturbs bilayer packing.

P69. Membrane Binding and SLiM-Mediated Protein-Protein Interactions of Ferlin C2A-C2B Linkers, Erin Huang, Fakhria Saadat, Oregon State University, Faculty mentor: Colin P. Johnson

Ferlins are a family of multi-domain membrane proteins that utilize C2 domains as Ca^{2+} sensors to regulate diverse vesicle trafficking processes, including vesicle fusion during host cell invasion by single-cell parasites and sperm motility in invertebrates such as C. elegans and Drosophila. In vertebrates, six ferlin orthologues have been identified, including dysferlin, which mediates Ca^{2+}-dependent repair of damaged cellular membranes, and otoferlin, which regulates neurotransmitter release during auditory signal transduction. Despite structural conservation among ferlins and an established calcium-sensing role, the precise functions and molecular mechanisms of ferlins remain unclear. In this study, we examined vertebrate ferlin C2A-C2B linkers for membrane association and short linear motif (SLiM)-mediated protein interactions. In

the otoferlin C2A-C2B linker, sequence analysis identified an arginine-rich region proximal to a putative AP2-binding motif that is predicted to mediate interactions with negatively charged lipid membranes. Linker-membrane interactions were tested using POPC and POPC/POPS liposomes by liposome co-sedimentation and laurdan fluorescence measurements, which showed preferential association of both otoferlin linker isoforms with negatively charged POPC/POPS liposomes. In dysferlin, candidate AP2-, SH3-, WW-, and SH2-binding SLiMs were identified within the C2A-C2B linker, and fluorescence anisotropy titrations showed direct binding to the SH3-containing protein endophilin A1 and the WW domain of kibra protein. Together, these results support a model in which C2A-C2B linkers mediate membrane binding and encode SLiMs for partner recruitment, suggesting that these intrinsically disordered regions function as important interaction hubs that recruit endocytotic proteins to the membrane to initiate endocytosis. Ongoing work is focused on defining how membrane association changes linker structure and SLiM accessibility. We will employ circular dichroism (CD) spectroscopy to investigate the secondary structure of C2A-C2B linkers before and after interaction with liposomes. These experiments will determine whether the intrinsically disordered linker adopts a more ordered conformation upon membrane binding, providing new insight into the molecular basis of ferlin-membrane interactions.

P70. Mutational Analyses of Nucleotide-Binding Sites in the MetNI Methionine ABC Importer, Emiko Uohara, University of San Francisco, Faculty mentor: Janet Yang

ATP-binding cassette (ABC) transporters use the energy from ATP binding and hydrolysis to facilitate the active transport of molecules across lipid bilayers. Regulating the movement of molecules across membranes is crucial for maintaining cellular homeostasis, and ABC transporters facilitate this process by importing essential nutrients and exporting toxins. Malfunctioning ABC transporters are linked to several human diseases, including cystic fibrosis and Alzheimer's disease. This research focuses on the role of ATP binding and hydrolysis in the E. coli methionine importer system, MetNI-Q. The MetNI transporter contains two identical highly conserved nucleotide binding domains (NBDs), and how these two sites coordinate to drive ATP hydrolysis and enable transport remains poorly understood. To explore how the two ATP-binding sites work together, we expressed, purified, and characterized MetNI variants with mutations in key NBD motifs. ATPase measurements showed that an N295A mutation retains wild-type hydrolysis activity, while an E166Q mutation severely diminishes ATP hydrolysis. ITC measurements indicated that N295A and E166Q mutants bind ATP and ADP with similar affinities and that only one nucleotide binds per MetNI dimer. The latter finding is inconsistent with the canonical model of transport which requires two ATP molecules per transport cycle, and further experiments are necessary to confirm the nucleotide binding affinities and stoichiometries. This work advances our understanding of the interdependence between the two nucleotide-binding domains to drive transport, which ultimately may be broadly applicable to the ABC transport family.

P71. Bacterial Growth in the Presence of glmS Riboswitch Analogs, Alessandra Kakish, Creighton University, Faculty mentor:

Antibiotics are becoming more ineffective as bacteria adapt to drugs that were devised to kill them. However, researchers have identified riboswitches, non-coding segments of mRNA that affect the expression of downstream genes, as a new target for antibacterial agents, one of which is the highly extensive glmS riboswitch. The glmS riboswitch controls the gene expression of fructose-6-phosphate amidotransferase, which synthesizes glucosamine-6-phosphate (GlcN6P) in bacterial cells. GlcN6P is a precursor in bacterial cell wall biosynthesis, and therefore, its synthesis is essential. The glmS riboswitch is also a catalytic ribozyme, which self-cleaves upon binding to GlcN6P. This cleavage degrades the mRNA, inhibiting glmS gene expression and preventing bacterial cell wall synthesis. Because the glmS riboswitch can control cell viability, it's a potential target for new antibiotic development. This project focuses on identifying analogs with structural similarity to GlcN6P that can affect the riboswitch as an agonist or antagonist. To determine whether GlcN6P ligand analogs can inhibit bacterial growth, assays are performed to monitor Bacillus subtilis and Staphylococcus aureus growth in the presence or absence of potential GlcN6P analogs. Preliminary data suggests that L-serine decreases bacterial growth at concentrations of ~31.3 mM for B. subtilis and 62.5 mM for S. aureus. Optimization of RT-PCR is also being conducted to verify whether the glmS gene and glmS riboswitch RNAs are downregulated. Future studies will verify that the analogs are decreasing growth via interaction with the glmS riboswitch and will investigate the effects of L-serine on mutant strains of B. subtilis and S. aureus.

P72. Analysis of a Putative Frameshifting RNA Structure from Agaricus bisporus (mushroom), Sulayman Ahmed, Institution, Faculty mentor:

Riboswitches are non-coding segments of messenger RNA that undergo conformational changes when bound to metabolites, resulting in changes to overall gene expression. Riboswitches are involved in many important metabolic pathways in bacteria, but none have been identified in higher organisms. The Ornithine Decarboxylase Antizyme (OAZ) RNA is involved in the synthesis and regulation of polyamines. Polyamines are essential organic molecules that specifically bind to DNA and RNA, and they are heavily involved in many cellular processes. The crucial role that riboswitches play in cell metabolism gives rise to the potential of developing antibiological agents. I have focused on determining whether the OAZ RNA element in the fungus Agaricus bisporus exhibits the characteristics of a riboswitch. A key function of a riboswitch RNA is a change in gene expression upon the RNA binding to a specific ligand, inducing a conformational change in the RNA. To determine if the Agaricus bisporus OAZ RNA supports a change in gene expression, dual luciferase assays are being performed with a construct that contains the OAZ riboswitch upstream of the Photinus (firefly) luciferase gene. After transfection

into HEK-293 cells, gene expression was quantified by measuring the ratio of Photinus luciferase to Renilla luciferase (a transfection control) in the presence versus absence of polyamines (including spermine, spermidine, pentaamine). Results indicate the highest Photinus versus Renilla luciferase activity when exposed to spermine (in comparison to other polyamines), thereby exhibiting a change in gene expression and showing specificity for one polyamine, another key characteristic of riboswitches.

P73. Structural Analysis of OAZ RNA from Homo sapiens, Shawn Ramachandran, Creighton University, Faculty mentor:

Riboswitches are segments of non-coding RNA found in the 5' untranslated region of messenger RNA that influence the expression of a downstream gene when bound to cellular metabolites. Binding of a metabolite induces conformational changes and directly regulates gene expression via alterations in downstream transcription, translation, or RNA processing machinery. While most riboswitch regulatory elements have been identified in bacteria, eukaryotic riboswitch behavior implicated in the biosynthesis of polyamines remains largely uninvestigated. Polyamines are organic compounds that are pervasive in living cells and are vital for many cellular processes, such as growth, survival, and differentiation. Furthermore, they can act as ligands in their interactions with DNA, RNA, and proteins to influence cell maturation. Thus, further research investigating the role of riboswitches as regulatory elements and their specificity for ligands is imperative as it will enhance the ability to utilize riboswitches for applications in medical therapeutics for various diseases, such as cancer. One such potential eukaryotic riboswitch that is highly conserved and preserved in multiple organisms is the Ornithine Decarboxylase Antizyme RNA (OAZ RNA). Recent studies from Dr. Soukup's lab suggest strong evidence for a potential riboswitch in human OAZ RNA through analysis of riboswitch-ligand interactions and conformational changes. The focus of my project is to further investigate the interactions between OAZ RNA from Homo sapiens and varying concentrations of polyamines, specifically with regards to conformational changes and eventual gene expression. In-line probing (ILP) is the central assay used to analyze these structural interactions. Specifically, this assay can elucidate whether the OAZ RNA exhibits one of the three key characteristics of riboswitches—conformational changes induced by ligand binding—by utilizing varying concentrations and types of polyamines. Thus, this research project aims to explore the importance and relevance of interactions between riboswitches and polyamines in humans, thereby contributing to both the scientific and medical communities.

P74. Effects of Phosphorylation of Chromatin Assembly Factor-1 on gene silencing in saccharomyces cerevisiae, Cohen Cisneros, Creighton University, Faculty mentor: Lynne Dieckman

Nucleosomes are the basic structural units of chromatin. They are made of segments of DNA wrapped around histone proteins. The level of compaction of nucleosomes regulates gene expression by altering the accessibility of DNA. Chromatin assembly factor 1 (CAF-1) is a protein that deposits histones onto newly replicated DNA in silenced regions of the genome. CAF-1 is recruited to DNA by proliferating cell nuclear antigen (PCNA). PCNA is a sliding clamp that recruits many other proteins to the replication fork, including CAF-1. CAF-1 is known to be phosphorylated at multiple locations on the protein, but it is not known how these phosphorylation events affect gene expression. We hypothesize that phosphorylation of CAF-1 disrupts its ability to interact with PCNA and/or DNA and therefore causes improper nucleosome assembly. To investigate this, we inserted phosphorylated CAF-1 into yeast cells and observed the effects on gene expression. Thus far, the data suggest individually-phosphorylated residues in CAF-1 do not alter gene silencing. We are currently generating CAF-1 with combinations of phosphorylated sites to observe their combined effect on gene expression. With the completion of these studies, we hope to better understand the relationship between the phosphorylation state of CAF-1 and gene silencing in yeast.

P75. Classifying Canonical and Vacancy-Bearing G-Quadruplexes in Human 5'UTRs and Testing Their Impact on Translation Initiation, Merat Semma, Yale University, Faculty mentor: Wendy Gilbert

G-quadruplexes (G4s) are guanine-rich RNA secondary structures that can inhibit translation initiation by impeding 43S ribosomal scanning along the 5′ untranslated region (5′UTR). Recent studies have proposed a related class of noncanonical structures, guanine-vacancy-bearing quadruplexes (GVBQs), in which one guanine tract contains only two guanines, resulting in reduced intrinsic stability. Although predicted to form under cellular conditions, the role of GVBQs in translational regulation has not been well defined. The objective of this study was to determine whether GVBQ motifs within human 5′UTRs regulate translation initiation in a structure-dependent manner and to compare their effects with those of canonical G4s.We developed a computational pipeline using regular-expression–based pattern matching to identify canonical G4s and non-overlapping GVBQs across annotated human 5′UTRs. Representative candidate sequences were cloned in their wild-type form upstream of a dual-luciferase reporter to measure translation efficiency. To assess whether translational effects depended on RNA secondary structure rather than guanine content alone, corresponding G→A mutations were introduced to disrupt predicted quadruplex formation in both canonical G4- and GVBQ-containing 5′UTRs.Translation efficiency measurements showed that 5′UTRs containing either canonical G4s or predicted GVBQ motifs reduced translation initiation relative to controls. Comparison of wild-type constructs with their G→A mutants revealed relief of translational repression, indicating that reduced translation efficiency depends on quadruplex formation rather than guanine content alone.This work provides a framework for directly comparing canonical and noncanonical G4

variants and supports future high-throughput studies of RNA secondary structure–mediated translational regulation.

P76. Using a Fluorescence Assay to measure the Uptake of glucose in C2C12 mammalian cells in the Presence of Opuntia Ficus (Indica), Rimah El Bayouk, Bronson Early, Sonoma State University, Faculty mentor: Monica Lares

Previous research, including papers such as those of López-Romero (2014), suggests that Nopales have been shown to stabilize blood glucose levels in diabetics when consumed with a high-carbohydrate breakfast (López-Romero, 2014). This raises the question: Do nopales increase glucose uptake via glucose transporters? Do nopales regulate blood glucose levels through a mechanism similar to that of diabetes drugs? These lead to the bigger question: Could Nopales be used as a natural treatment method for Type 2 diabetes? In this study, C2C12 myoblast cells were cultured by feeding and passaging, and were placed in a 24-well plate so that a fluorescence assay could be performed. Eight different conditions were tested a total of three times. The 8 conditions consisted of just cells with media, 50 µg/mL OFI with no NBDG, cells with insulin (Positive Control), Cells with NBDG, and no OFI, 10 µg/mL OFI with NBDG. 50 ug/mL OFI, with NBDG. 100 ug/mL OFI, with NBDG. 200 ug/mL OFI, wiith NBDG. A fluorescence assay was run before and after adding the NBDG. Post-NBDG values and pre-NBDG values were subtracted from one another to obtain the delta fluorescence, showing the intensity of light detected that represents the uptake of glucose. These values were compared with those of insulin as a positive control, and percentages were obtained to measure glucose uptake. The insulin positive control showed 97% glucose uptake, 10 µg/mL OFI, with NBDG showed -79% glucose uptake. 50 ug/mL OFI, with NBDG, showed 217% glucose uptake. 100 ug/mL OFI, with NBDG, showed 918% glucose uptake, and 200 ug/mL OFI, with NBDG, showed 5.41% glucose uptake. These results suggest that Nopales could reduce blood glucose levels and that there may be an optimal concentration for OFI that maximizes glucose uptake.

P77. Kinetic Characterization of Flavonoid Inhibition of Enoyl-ACP Reductase (FabI), Rustin Ghidari, Pepperdine University, Faculty mentor: P. Matthew Joyner

Flavonoids from various plants are an important class of beneficial phytochemicals that have shown interesting antibacterial properties as inhibitors of the enzyme enoyl-acyl carrier protein (ACP) reductase (FabI), which plays a key role in the bacterial fatty acid biosynthetic pathway. The mode of inhibition of FabI by plant-derived flavonoids is still incompletely understood and data from various studies have yielded inconsistent conclusions. This current study utilized enzyme kinetics to characterize the inhibition by various plant flavonoids of FabI from Escherichia

coli, as well as to distinguish their mode of inhibition from the validated FabI inhibitor triclosan. Enzyme kinetics and the oxidation of NADH using spectrophotometric assays were employed to monitor FabI activity as a function of both substrate and inhibitor concentration. Significant inhibition of FabI activity was observed using catechin, while kinetic analysis demonstrated atypical behavior associated with inhibition by quercetin, including the potential for the mode of inhibition to change as a function of concentration. Unlike triclosan, the addition of excess NAD+ did not enhance flavonoid-induced inhibition; therefore, the inhibitory mechanism of flavonoids must differ from that of triclosan, which forms a stable ternary complex with FabI and NAD+. To evaluate whether a cysteine residue near the active site is involved in inhibiting FabI functions, we used cysteine-reactive agents and created a C210A mutant of FabI. The results of our kinetic investigations show a marked decrease of inhibitory activity of flavonoids with the C210A mutant. This supports a model where some flavonoids act as covalent FabI inhibitors using cysteine-dependent mechanisms, rather than via simple reversible binding. Collectively, the results of this study provide insight into novel mechanisms by which natural products may offer potential for the development of new antibacterial agents.

P78. Measuring Enzyme Kinetics Using Electrochemical Detection, Lily Jiang, University of California, Santa Barbara, Faculty mentor: Lior Sepunaru

Enzyme kinetics are traditionally determined using optical assays based on initial-rate measurements, which require multiple experiments and can be limited by absorbance saturation, spectral overlap, and interference from secondary chemical species. We present an electrochemical alternative in which microelectrode chronoamperometry can extract enzyme kinetic parameters from a single, continuous trace, using global analysis of the full catalytic time course. Using horseradish peroxidase as a model system, we monitored catalytic turnover electrochemically by measuring the current of benzoquinone, a product generated during HRP-catalyzed oxidation of hydroquinone by hydrogen peroxide. This current provides a continuous readout of enzymatic activity at the microelectrode surface. By fitting the complete chronoamperometric response, kinetic parameters including Km and Vmax can be obtained without relying solely on initial-rate approximations. This method was validated across a range of substrate concentrations and further tested through inhibition experiments that reduce catalytic turnover. These measurements demonstrate that the method captures the expected decrease in apparent maximal rate within a single electrochemical trace. Additionally, temperature-dependent measurements revealed systematic shifts in the catalytic response consistent with thermally activated turnover, enabling extraction of activation parameters that provide insight into enthalpic and entropic contributions governing the reaction. These results establish electrochemical time-course analysis as a practical and quantitative strategy to characterize enzyme kinetics. This approach has applications in systems where optical methods are unreliable due to limited dynamic range, spectral interference, or opaque media, conditions that are often encountered in complex biological and environmental

samples. HRP serves as a well-characterized benchmark, but this framework is broadly applicable to other enzyme systems of biological interest.

P79. Investigating Electron Transfer in Biological Catalysis Using Electrochemical Noise Analysis, Noya Drori, University of California, Santa Barbara, Faculty mentor: Lior Sepunaru

It is well understood that biological catalysts control reactivity through regulated electron transfer processes. The heterogeneous electron transfer rate constant, k0, measures the rate of electron transfer and is dependent on how easily a redox system reorganizes to energetically support a reaction. Common electrochemical methods, such as Tafel Analysis, quantify kinetics by measuring currents at large overpotentials to ensure that forward and backward currents do not cancel. This poses a risk that the diffusion becomes rate limiting, possibly skewing sensitivity and accuracy. On a biological scale, we understand that the smallest systemic changes can heavily affect reactivity. Thus, we have presented an alternative method of measuring kinetics that has proved more sensitive. Electrochemical noise measures current fluctuations at equilibrium, and we have found that its root-mean-square is directly proportional to k0. Because we perform measurements with no applied potential, we avoid the possibility of being diffusionally constrained. I compared Tafel analysis and Noise derived kinetics across electrolytes to measure their sensitivity, as it is known electrolytes with smaller cation sizes have higher solvent reorganization energy and entropy due to their stronger hydration shells. Noise revealed stronger effects compared to Tafel analysis, and we can infer that diffusion limits the sensitivity of using large current as a source of quantification. By using noise derived kinetics, we believe we can perform further analysis that gives us insight into the roles of thermodynamics and electrolyte effects on catalytic reactivity. For example, we can use our derived k0s in the Arrhenius equation; k0= Ae-Ea/RT by taking measurements across a span of temperatures in order to isolate activation energies, Ea for different systems. In doing such, we can quantify the contributions of entropy and enthalpy. These findings can help explain how catalytic activity is affected by local environments, which can aid in the development of efficient biological catalysts.

P80. Development and Optimization of a DTNB Assay Protocol Kit for CPT1A Activity Inhibition Screening, Himanshu Gupta, Anthony Wong, Aspiring Scholars Directed Research Program, Faculty mentor: Zane Chen

Carnitine palmitoyltransferase 1 (CPT1), which catalyzes the rate-limiting step of fatty acid oxidation, has been implicated in therapeutic approaches to several human diseases characterized by aberrant lipid metabolism. The isoform-specific quantification of CPT1 activity is essential in

the characterization of small molecule inhibitors of CPT1, but several existing means to quantify enzymatic activity, including the use of radioisotope-labeled carnitine, are not amenable to scalable, high throughput screening. Here, we demonstrate that mitochondrial extracts from Expi293 cells transfected with a CPT1a plasmid are a reliable and robust source of catalytically active human CPT1. Moreover, with a source of catalytically active enzyme in hand, we modified a previously reported colorimetric method of coenzyme A (CoA) easily scalable to a 96-well format for the screening of CPT1a inhibitors. This assay platform was validated by two previously reported inhibitors of CPT1a: R-etomoxir and perhexiline. To further demonstrate the applicability of this method in small molecule screening, we prepared and screened a library of 87 known small molecule APIs, validating the inhibitory effect of chlorpromazine on CPT1.

P81. EPR Spectroscopic Characterization of Nanolipoprotein Particles for Drug Delivery Applications, Kush Shah, University of California, Davis, Faculty mentor: John Voss

Treatment of neurodegenerative diseases is challenging due to the limited ability of therapeutic compounds to cross the blood–brain barrier. Nanolipoprotein particles (NLPs) represent a promising platform for targeted drug delivery to the brain. This work investigates the use of NLPs to deliver curcumin, a small-molecule antioxidant, via carrier proteins embedded within the NLP structure to facilitate transport to the brain.To characterize the structural and dynamic properties of these particles (e.g., macromolecular dynamics, stability, distribution, and load capacity), we employ nitroxide spin labels and EPR spectroscopy to probe the lipids, proteins, and curcumin molecules that comprise the NLP. NLPs were formulated, and EPR spectroscopic data were collected and analyzed by fitting experimental spectra to simulated models using the Multicomponent software. From these fits, parameters such as correlation times and tensors were extracted, which are related to various molecular interactions.By collecting EPR data on NLPs varying in factors such as size and lipid composition, this approach enables the identification of structural features that optimize stability and efficiency. These results provide insight into the design principles necessary for developing NLP-based drug delivery systems.

P82. Direct analysis of S-nitrosohemoglobin using a statistical approach on intact, native and top-down mass spectrometry data, Nicole De Felice, Pepperdine University, Faculty mentor: Matthew Joyner

This research project explores improved methods for detecting and analyzing the fragmentation patterns of S-nitrosylated hemoglobin (SNO-Hb), a compound that regulates microvascular vasodilation and tissue oxygen delivery. SNO-Hb has been seen as a compound potentially linked to the performance-enhancing effects of beetroot juice. Beetroot juice is thought to boost athletic performance by increasing nitric oxide (NO) levels in the body, which enhances blood flow, glucose uptake, and exercise efficiency. However, the mechanisms through which nitric oxide is distributed and transported remain uncertain. One leading theory suggests that hemoglobin carries nitric oxide by binding it through nitrosylation, thus forming the SNO-Hb compound.A mass spectrometer was used to obtain intact, native, top-down mass peaks for each sample of interest: five control samples with unaltered hemoglobin and eleven treatment samples with SNO-Hb. All samples were collected with an ion activation energy, ISC, of 20, a data collection resolution of 200k, and a collision cell energy, HCD, of 140. This research project explores the development of new statistical tools to analyze the differentiation between SNO-Hb and normal hemoglobin.Using Principal Component Analysis (PCA), implemented in Python, I aim to differentiate SNO-Hb from unaltered hemoglobin by identifying unique fragmentation patterns to ultimately find ion intensities present in one type of hemoglobin and absent in the other. I firstly began by filtering low-intensity noise and sample outliers to isolate the most significant variance in PC1 and PC2. These components allow for the visualization of distinct clusters and the identification of ion intensities unique to each hemoglobin type. Building on preliminary data that confirms a clear divergence between the two samples, I will ultimately map these specific mass-to-charge ratios back to the mass spectra to pinpoint the exact sites of protein fragmentation.

P83. PEG/DEX Aqueous Phase Separated Emulsions as Model Systems of Intracellular Coacervates, Riya Sandhu, California State University, Sacramento, Faculty mentor: Mikkel Herholdt Jensen

The cytoplasm of eukaryotic cells is a crowded viscoelastic environment with about 20% of the cytoplasmic volume occupied by proteins. In this crowded environment, volume-exclusion effects can lead to aqueous two-phase separation (ATPS). This liquid-liquid phase separation is increasingly recognized as a key process in the cells' internal, membrane-less organization of proteins and metabolites.To study ATPS and the interplay between aqueous phases and cytoskeletal proteins, we developed an in vitro phase-separated model system using two polymers, poly-ethylene glycol (PEG) and dextran (DEX). Though entropically mixed at low concentrations, PEG and DEX are immiscible at high concentrations and form two separate aqueous phases. We emulsify a system of PEG and DEX and stabilize DEX-rich droplets in a PEG-rich phase with the addition of small unilamellar vesicles (SUVs) to form a Pickering emulsion. Using confocal fluorescence microscopy, we confirm that this method generates a stable ATPS system with DEX droplets a few 10s of μm in size. Bulk rheology confirms that both the PEG- and DEX-rich phases

behave as Newtonian fluids.When incorporating skeletal muscle actin into the system, microscopy shows a stronger preference of polymerized actin for the DEX-rich phase, demonstrating ATPS as a potential cellular organizing mechanism of actin. Bulk rheology confirms the formation of a viscoelastic entangled actin network in our systems. We use this model system to probe the dynamic distribution of actin in two-phase aqueous systems and the effects of crowding and confinement on the interactions of actin with other binding constituents. Our results demonstrate that ATPS both influences and is influenced by cytoplasmic protein interactions and their organization.

P84. Engineering DNA Liquid Crystals for Rhythmic Control by a Reconstituted Circadian Clock, Areesha Tariq, Ronald Truong, University of California, Merced, Faculty mentor: Andy LiWang

Biological clocks allow living systems to anticipate and adapt to daily environmental changes. In cyanobacteria, a remarkably simple protein oscillator composed of KaiA, KaiB, and KaiC generates a 24-hour rhythm that ultimately controls the DNA-binding activity of the transcription factor RpaA. We aim to harness this natural timekeeping system to create a new class of biomolecular materials whose optical properties change rhythmically over time.Our central hypothesis is that clock-controlled DNA binding by activated RpaA can perturb the structure of a self-assembled DNA liquid crystal, thereby enabling programmable, time-dependent modulation of optical behavior. If successful, this work would establish a direct link between a reconstituted biological oscillator and mesoscale material properties, laying the foundation for autonomous, rhythmic biomaterials.To test this concept, we focused on a phosphomimetic mutant of RpaA (D53E) that mimics the activated, DNA-binding state. In parallel, we designed a 34-base-pair DNA duplex with complementary sticky ends to promote higher-order self-assembly into liquid crystalline structures. Our approach combines protein purification, DNA self-assembly, and biophysical characterization to determine whether activated RpaA can interact with and modulate these supramolecular DNA assemblies.Preliminary ^{1}H NMR spectra of the DNA construct revealed broad, low-resolution imino proton signals rather than sharp resonances expected for small duplex DNA. Re-annealing with an optimized thermocycler protocol produced similar spectra, suggesting that the broadening does not arise from misfolding but instead from the formation of large multimeric assemblies whose slow tumbling limits high-resolution detection. Size-exclusion chromatography further supported the presence of higher-molecular-weight species, and circular dichroism confirmed preserved helical stacking despite supramolecular assembly. Initial binding assays between RpaA D53E and the DNA assemblies did not show detectable complex formation under standard conditions, motivating ongoing studies to verify DNA composition, optimize binding conditions, and evaluate potential steric effects from purification tags.Together, these findings indicate that we have successfully generated higher-order DNA assemblies consistent with liquid crystalline behavior and established a platform for testing

clock-regulated protein–DNA interactions. Future work will refine binding assays and integrate these assemblies with the reconstituted KaiABC oscillator to assess time-dependent modulation of material properties. This work represents a critical step toward engineering oscillator-controlled DNA liquid crystal systems with programmable optical function.This research is supported by the National Institutes of Health.

P85. Bioinformatics Solutions to Antimicrobial Resistance Diagnostics, Trotter McLemore, Olga Carvello, California Polytechnic State University, San Luis Obispo, Faculty mentor: Jean Davidson

[CANCELLED]

Antimicrobial resistance (AMR) represents one of the most significant threats to modern medicine. As pathogens evolve to evade existing treatments, conventional diagnostics for bacterial infections such as urinary tract infections (UTIs) remain costly and time-consuming, often delaying effective treatment. To address this challenge, we are integrating bioinformatics and wet-lab approaches to develop rapid and accessible diagnostic tools. We implemented a machine learning framework for antibiotic resistance (ABR) prediction using pan-genome data. This pipeline, previously validated in E. coli, was benchmarked for reproducibility and then extended to a much larger dataset, improving accuracy and generalizability. In line with our goal of creating easy-to-access tools, we expanded upon previous work in this space and created a fully pre-trained model capable of predicting on completely new data. By analyzing pan-genome patterns from clinical bacterial isolates, we aim to create a generalizable framework capable of identifying resistance emergence across diverse pathogens. In parallel, we are developing on-site microPAD array assays to enable rapid pathogen detection and guide targeted treatment decisions. We previously designed multiplex qPCR assays targeting common uropathogens, including Esercheria coli, Proteus mirabilis, and Klebsiella pneumoniae. Conserved 16S rRNA regions were used for primer design, while strain-specific probes allow high-resolution pathogen identification and detection of mixed infections. We are currently optimizing multiplex assays for Enterococcus faecalis, Pseudomonas aeruginosa, and Staphylococcus saprophyticus, along with primer–probe sets targeting key β-lactamase resistance genes. Together, these approaches aim to create a comprehensive genomic and diagnostic framework for rapid, patient-specific detection of antimicrobial resistance. These tools could improve antibiotic stewardship, reduce diagnostic turnaround times, and help address the growing public health burden of antibiotic-resistant UTIs.

P86. Deep Learning Classification of Lateral-Incisor Root Resorption from CBCT, Ethan Nusinovich, University of California, Los Angeles, Faculty mentor: Karolina Elżbieta Kaczor-Urbanowicz

Objective: Maxillary impacted canines (MIC) are a common orthodontic finding whose severity guides treatment planning. Manual assessment of cone-beam computed tomography (CBCT) images is time-consuming and subject to inter-examiner variability. This study evaluated multiple 3D convolutional neural network (CNN) approaches for automated MIC severity classification from CBCT scans, systematically comparing input preprocessing strategies to determine what information the model requires for accurate prediction. Materials and Methods: Pre-treatment CBCT scans from 67 patients (134 canines) were obtained from UCLA Dental Clinics (IRB-25-2724). Inclusion criteria required maxillary canine region capture, no prior orthodontic treatment, patient age >12 years, and complete canine apex formation. A standardized 60 × 40 × 50 mm 3D region of interest (ROI) was automatically extracted from the anterior maxilla using template matching. MIC severity was labeled by a calibrated examiner following published guidelines on a 4-class scale (Normal, Mild, Moderate, Severe). Four model architectures were evaluated using a pretrained 3D ResNet-18 with 5-fold stratified cross-validation: (1) a patient-level baseline predicting the worse severity per patient from the full ROI; (2) a per-side model predicting severity for each canine individually from left/right ROI halves; (3) a threshold-masked model retaining only the densest 5% of voxels combined with ordinal loss; and (4) a dual-channel model pairing the original ROI with binary tooth masks generated by a pretrained DentalSegmentator network. Results: The per-side model (Model 2) achieved the best performance with 64.9% accuracy and 58.7% balanced accuracy, improving upon the patient-level baseline (61.2% accuracy, 58.6% balanced accuracy) with substantially higher normal-case precision (94% vs. 58%) and reduced fold-to-fold variance. Threshold masking (Model 3) significantly degraded performance (58.2% accuracy, 43.9% balanced accuracy), with mild-case recall collapsing from 42% to 8%. The segmentation-guided dual-channel model (Model 4) also underperformed (61.2% accuracy, 52.5% balanced accuracy). Normal cases were detected most reliably across all models (76–94% precision), while mild severity was the most challenging class due to limited samples (n = 12).Conclusions: Splitting the ROI to classify each canine individually was the most effective strategy, while density-based masking and segmentation-guided channels both weakened performance. These findings indicate that surrounding bone and soft-tissue context provides critical spatial information for severity assessment, and that the model's primary bottleneck is dataset size and the inherent ambiguity of adjacent severity classes rather than its ability to localize teeth. AI-based CBCT analysis shows promise as a decision-support tool for MIC assessment, though future work should focus on expanding datasets and validating across multiple institutions.

P87. Removal of Fluoride from Drinking Water Using Moringa oleifera Seed-Based Filtration Methods, Alexis Burger, Ella Noecker, Pepperdine University, Faculty mentor: Donna Nofziger

Fluoride, the anion of fluorine, is created from salts that form when fluorine combines with minerals in soil or rocks. Fluoride enters drinking water naturally when groundwater flows through rocks and soils to supply wells, springs, or aquifers. Levels of fluoride in drinking water ranging from 0.5-1.2 mg/L provide health benefits for the maintenance of teeth and bone. The World Health Organization's global permissible limit of fluoride in drinking is 1.5 mg/L and prolonged consumption of fluoride above this limit can cause increased risk of dental and skeletal fluorosis. Analysis of the water at Made in the Streets, a school in Kamulu, Kenya, measured the fluoride concentration in the range of 6.0-12.0 mg/L, which is well above the permissible limit. Moringa oleifera, a tree native to Asia and Africa, is common throughout Kenya. All parts of the tree (leaves, seeds, roots, and flowers) are suitable for human consumption, and have been historically used for traditional medicinal purposes. The seeds within M.oleifera trees contain cationic proteins, which have been implicated in the flocculation and removal of negatively charged particles, such as fluoride. Preliminary analysis performed in Kenya suggested that well water treated with M.oleifera seed powder lowered the levels of fluoride. In this analysis, two different filtration methods using hand-made filters containing powderized M.oleifera seeds were examined for their ability to remove fluoride from water. The control and filtered samples for each trial were examined using a photometer to test for fluoride concentration in mg/L. Using a paired T-test, it was determined that both filter methods significantly lowered the amount of fluoride relative to each trial. In line with the previous field research in Kenya, it was concluded that the hand-made filters containing M.oleifera seeds effectively removed fluoride from drinking water.

P88. Removal of E. Coli from Drinking Water Using Moringa oleifera Seed-Based Filtration Methods, Noah Lee, Julie Yun, Pepperdine University, Faculty mentor: Donna Nofziger

Escherichia coli is a Gram-negative bacteria that commonly colonizes the gastrointestinal tracts of humans and warm-blooded animals. It enters water sources when fecal matter contaminates rivers, lakes, or groundwater, often through agricultural runoff or poor sanitation systems. The presence of E.coli in water indicates recent fecal contamination and the possible presence of harmful pathogens that can cause gastrointestinal illness, diarrhea, and other infections if the water is consumed. An analysis of drinking water at Made in the Streets, a school in Kamulu, Kenya, indicated the presence of E.coli at various locations around campus. Moringa oleifera, a tree found throughout Kenya, is commonly consumed for its nutritious properties and has been historically used for traditional medicinal purposes. The seeds within M.oleifera trees contain a cationic protein, which previous research has shown can result in the flocculation and removal of bacteria,

such as E.coli. This experiment tested two different filtration methods using hand-made filters containing powderized M.oleifera seeds to effectively remove E.Coli from water. The first method, the “pour-over,” consisted of water being poured over a coffee filter containing seed powder. The second method, the “soak,” consisted of seed powder being soaked in water and then the water gets strained through a coffee filter. The control and filtered samples were tested for each trial using E.coli/coliform count petrifilm plates. Using a paired T-test, it was determined that both the pour-over and soak significantly removed E.Coli from the water samples. In line with previous field research in Kenya, it was concluded that the hand-made filters containing M.oleifera seeds effectively removed E.Coli from drinking water.

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P89. Investigating Disordered Domains in the Batten Disease Proteins, Mahnoor Nasir, Bryn Mawr College, Faculty mentor: Adam Williamson

Batten Disease (also called Juvenile Neuronal Ceroid Lipofuscinosis, or JNCL) is a hereditary pediatric onset neurodegenerative disease impacting ~4 in 100,000 births worldwide. It is caused by mutations in one of 13 CLN genes, which lead to a suite of common symptoms that include neuronal loss, accumulation of autofluorescent material, sleep disruptions, seizures, and severe impacts on the visual system. The common nature of the devastating symptoms, despite the vastly different functions of the proteins under normal circumstances, raises the question of whether the mutations converge on similar pathways or whether the proteins have something in common that might explain these similar pathophysiologies. Close inspection of the 13 CLN proteins through structural analysis revealed that they all have one structural component in common: unstructured regions, or tails, on at least one terminus. Because the actual nature of the CLN proteins is poorly understood, we anticipated that these tails, similar to intrinsically disordered regions (IDRs) in other proteins, played a role as binding partners to surrounding lipids and proteins. More specifically, we hypothesized that the tails are molecular "zip codes" directing the proteins to lysosomes for degradation. Our assay tested how well the mutants localize to intracellular puncta or phagosomes surrounding engulfed material. Unexpectedly, the wild type (WT) constructs showed elevated localization around phagosomes as compared to the DT constructs, indicating that the tails may be putative phagolysosome localizing sequences and/or they play key roles as stability determinants of the CLN proteins in RPE cells. Cycloheximide chase assays are in progress in order to determine the half life and degradation rates of the CLN proteins. Future functional studies using CLN knock-out cell lines will also be conducted.

P90. Elucidating the role of MAFB in the songbird circuit system, Keerthi Varadharaj, University of California, Santa Cruz, Faculty mentor: Bradley Colquitt

The brain undergoes complex developmental processes during the acquisition of complex, learned behavior. The molecular and behavioral development underlying motor learning can be studied with the help of the motor system that produces song in songbirds. Birdsong is controlled by several distinct yet interconnected brain regions and learned during a critical period, similar to vocal-learning in humans. The ease of genetic manipulation during the critical timeline of song development makes songbirds a powerful model organism, especially when studying developmental behavior like motor learning. MAFB is a transcription factor whose role in

song-circuit development is our main interest. In non-avian species MAFB is found to play an important role in interneuron development and has been used as a marker for fast-spiking inhibitory neurons. In the avian song system, MAFB is found in fast-spiking excitatory neurons in the robust nucleus of acropallium (RA), which is a song-circuit region that directly projects onto the brainstem regions required for song performance. My research project involves constructing drug-inducible plasmids to overexpress MAFB in the avian arcopallium in order to learn about its role in song-circuit development. First, we will test the efficacy of these constructs in vitro using a finch fibroblast cell line. Then, we will examine the effect of these constructs in vivo using in ovo electroporations of chicken embryos. Overall my project seeks to develop molecular tools to analyze the role of key transcription factors, like MAFB, in song-circuit development in order to provide insights to potential analogs in human motor-circuit development.

P91. Role of μ-Opioid Receptors on Noradrenergic Neurons in Opioid Withdrawal, Nina Bonaventure, University of California, Los Angeles , Faculty mentor: Catherine Cahill

IntroductionWithdrawal is one of the main reasons those with opioid use disorder continue their drug use Noradrenaline release contributes to opioid withdrawal, where drugs such as morphine bind to μ-opioid receptors (MOR) directly on noradrenergic neurons. During withdrawal, noradrenergic neuronal activity is increased and there is increase in noradrenaline release that contributes to both physical and protracted withdrawal states such as insomnia and anxiety. We hypothesize that MOR deletion on noradrenergic neurons will reduce aversion, withdrawal behaviors, and anxiety-like behaviors in animal models. Methods We will be using adult male and female C57 mice and a conditional knockout (cKO) of MORs from all noradrenergic neurons in the whole animal by breedingDbh-cre mice withflMOR/flMOR mice. Cage mate cre- mice will be used as controls. Mice will be made opioid dependent by replacing drinking water with morphine solution (0.25-0.75 mg/mL) over 14 days. Control mice will receive normal drinking water. Affective-like behavioral tests (sucrose preference, elevated plus, social interaction, light dark test and open field) will be performed at baseline, during peak withdrawal (2–5 days after morphine cessation), and after 4 weeks of abstinence. Animal behavior is tracked with AnyMaze. Outcomes include time spent in anxiogenic zones, number of entries, and distance traveled. All data will be analyzed by either a paired-t-test or a repeated measures 2-way ANOVA and graphed in Graphpad Prism. Expected ResultsMice undergoing peak physical withdrawal and protracted abstinence are expected to show reduced exploration of anxiogenic areas compared with baseline. We predict these effects will be attenuated in cKO mice. ConclusionsOur current experiments will provide a protocol that we can use to test our cKO and control mice to gain insight into the role of noradrenaline in withdrawal and negative reinforcement. This may provide the groundwork for withdrawal therapies targeting MORs on noradrenaline neurons.

P92. Evaluating CS1103 as a Treatment for Reversing Opioid Overdose, Shuoshuo Li, Janice Wong, University of California, Los Angeles, Faculty mentor: Catherine M Cahill

CS-1103, a molecular sequestrant, does not cross the blood-brain barrier; it binds to fentanyl in the plasma with high affinity and accelerates its clearance via urine. By rapidly reducing plasma concentrations of fentanyl, CS-1103 can pull ligands off opioid receptors without displacing them with another ligand. The purpose of this study is to determine to what extent CS-1103 produces physical withdrawal in opioid-dependent states and whether it can blunt withdrawal induced by naloxone. We hypothesize that CS-1103 will not elicit withdrawal behaviors and reduce naloxone-induced withdrawal behaviors. Adult male and female C57Bl/6 mice were made dependent by orally-self-administered 0.02 mg/ml fentanyl for 8 or 14 days. On the last day of fentanyl administration, the mice were given IP injections with either CS-1103 (200 mg/kg for 8-day model, 600 mg/kg for 14-day model) or vehicle followed by naloxone (2 mg/kg) 1.5 hrs later. The mice were scored for global withdrawal behaviors immediately after injection of naloxone in both models. Data (means +/- SEM) were analyzed via unpaired student t-tests or 2-way ANOVAs. In the 8-day model, CS-1103 pretreatment in male mice did not significantly alter naloxone-precipitated global withdrawal score compared to vehicle. The ineffectiveness of CS-1103 in this model may be attributed to reduced fentanyl exposure and the dose of CS-1103 administered. In the 14-day model, naloxone-precipitated withdrawal was significantly greater than that seen in vehicle-treated injections ($p<0.001$). Male mice that were scored immediately post-CS1103 injection had scores statistically similar to vehicles. Pre-treatment with CS-1103 did not significantly reduce naloxone-precipitated withdrawal statistically, but withdrawal scores from post-vehicle/CS-1103 injections were not statistically different than the former either, suggesting a partial reduction of withdrawal behaviors. Further studies with larger sample sizes may have the statistical power to elucidate the efficacy of CS-1103 in opioid-dependent states.

P93. Kit Ligand Is Required For Normal Cortical Development, Veronica Ostrin, University of California, Santa Cruz, Faculty mentor: David Feldheim

The cerebral cortex of the brain is integral to human experience, and abnormal development is associated with a number of neurological diseases. Mechanisms that control cortical growth are not well understood. Kit ligand (Kitl), also called stem cell factor or mast cell factor, is a signaling molecule known for its role in melanogenesis, gametogenesis, and hematopoiesis. Kitl is the binding-partner for the cKit receptor, and both are expressed in the developing mouse cortex. To determine if Kitl is required for cortical development, I compared the brains harvested from Emx-Cre; Kitl fl/fl mice (which have Kitl removed from the developing cortex) to those of control littermates. I found that Kitl mutant mice have significantly smaller cortices than their littermates. Furthermore, cryosectioning the brains followed by immunostaining or in situ hybridization allowed me to determine if loss of Kitl leads to altered cerebral architecture or changes in cell

proliferation. This work reveals a dramatic cortical function for this well-studied signaling molecule.

P94. Dysregulation of ER81 and Kv1.1 Underlies Impaired PV Interneuron Homeostatic Plasticity in Neurodevelopmental Disorders, Cecilia Hu, University of California, Berkeley, Faculty mentor: Dan Feldman

Impaired sensory processing is a major feature of autism and other neurodevelopmental disorders, yet the underlying circuit mechanisms remain poorly understood. Our lab has discovered that homeostatic plasticity of PV interneuron intrinsic excitability is absent in autism, a mechanism that could underlie sensory processing differences. The mechanism of this plasticity of intrinsic excitability involves transcription factor ER81, which drives expression of Kv1.1 delayed rectifier channels. In order to test whether this pathway is causally related to PV homeostasis, we need to develop tools to manipulate these pathways. We first optimized a method to pharmacologically inhibit ER81 in vivo. Stereotaxic injection of the small molecule inhibitor BRD32048 into mouse somatosensory cortex, followed by immunohistochemistry for ER81 and PV, revealed a significant decrease in ER81 expression in drug-treated mice relative to controls. This suggests that BRD32048 could be a promising tool for regulating downstream KV1.1 expression and PV cell physiology, however a very high concentration is needed, thus this option requires further optimization. To test the downstream involvement of Kv1.1, we used CRISPRa to deliver a PV-specific viral construct (S5E2-sadCas9-VP64 with sgRNA-Kcna1) to transcriptionally upregulate the Kcna1 gene in the mouse brain. Confocal microscopy confirmed increased Kv1.1 immunofluorescence intensity in mCherry+ cells, indicating successful construct expression and elevated Kv1.1 levels. Together, these tools will allow us to causally interrogate the ER81-Kv1.1 axis bidirectionally in future experiments. We would like to use ER81 inhibition to test whether ER81 is required for plasticity of PV intrinsic excitability. We plan to test if CRISPRa-mediated Kv1.1 upregulation is sufficient to reduce PV excitability and alter sensory processing. These planned experiments will clarify whether the ER81-Kv1.1 pathway is necessary and sufficient for bidirectional homeostatic PV plasticity, and whether its failure underlies sensory processing deficits in autism models. Ultimately, these findings may position the ER81-Kv1.1 pathway as a tractable therapeutic target for restoring homeostatic balance in neurodevelopmental disorders.

P95. Optimization of a Go-NoGo Behavioral Paradigm using Quantitative Performance Metrics, Mamoona Faryal, University of California, Berkeley, Faculty mentor: Dan Feldman

In neuroscience research, fluctuations in an animal's behavioral state can significantly influence neural processing and introduce noise into neural recordings. To adjust for these changes, we use an auditory Go-NoGo behavioral paradigm. The task requires a lick response to a tone (Go

response) and a no-lick response in the absence of the tone (No-Go response) to assess behavioral engagement and impulse control. However, the lack of a standardized training protocol often leads to protracted task training timelines and uncertainty about the reliability of subsequent neural recordings. This study aimed to optimize an auditory Go-NoGo training protocol for mice to ensure efficient training and enable imaging across multiple subjects. The primary goals were to: (1) establish a consistent training framework, (2) optimize the introduction of sensory stimuli, and (3) develop a quantitative analysis to assess learning progress. We show that a structured, nine-session training framework utilizing gradual Go-NoGo ratio shifts and robust quantitative monitoring effectively improves behavioral engagement and task performance. We implemented a protocol that transitioned Go-NoGo ratios from 0.8 down to 0.1, utilized prolonged auto-rewards, and delayed secondary sensory stimuli to allow for stable task acquisition. Performance was evaluated using d-prime to measure discrimination sensitivity, Effective Hit Rate (EHR) for engagement, and lick latency to track reaction times. The quantitative analysis revealed that mice trained with this protocol exhibited later-emerging but more stable improvements in d-prime and steadily increasing EHR compared to mice trained under less gradual protocols. Furthermore, mice trained under this protocol exhibited significantly reduced and more consistent lick latencies as they reached task proficiency. Our findings demonstrate that by using quantitative metrics such as d-prime, EHR, and lick latency, researchers can achieve a more reliable and scalable framework for training mice in complex behavioral tasks.

P96. Synaptic pMad Regulation in Presynaptic Homeostatic Potentiation at the Drosophila Neuromuscular Junction, Kayli Huang, University of California, Berkeley, Faculty mentor: Ehud Isacoff

Neural networks are highly malleable and robust systems composed of diverse neurons with heterogeneous synaptic proteins, allowing for specialized responses to stimuli and adaptable communication between cells. To maintain stable function in changing environments, resilient communication between pre- and post-synaptic cells is essential. Presynaptic homeostatic potentiation (PHP) describes this stabilization where decreased postsynaptic signal leads to an increase in presynaptic neurotransmitter release, restoring signal strength. Prior literature suggests cell-wide PHP, implying no synaptic heterogeneity. However, the Isacoff lab has demonstrated heterogeneity between synapses with the same pre- and post-synaptic cells in short term plasticity and chronic PHP. In acute PHP, it has been shown that presynaptic protein compaction heterogeneously recruits more vesicles for release through Bruchpilot (Brp, a scaffolding protein) and Cacophany (Cac, a presynaptic voltage gated calcium channel). A recently identified PHP synaptic organizer is phosphorylated Mad (pMad). pMad is present in the soma, where it alters transcription to promote neuronal growth, and in the presynapse, where its mechanism is not well understood. To examine synaptic heterogeneity of pMad relative to key synaptic plasticity proteins

– Cac, Brp, and Unc13A – I induced acute PHP at the Drosophila larval neuromuscular junction and used immunohistochemistry and super-resolution Airyscan microscopy to assess colocalization. My research shows that during acute PHP, pMad is heterogeneously affected across synapses, a change that correlates with those of other key proteins. Collectively, studying these highly conserved synaptic proteins during a critical and conserved homeostatic compensation provides insight into potential human diseases and potentially uncovers druggable targets for disorders associated with dysfunctional PHP, such as ASD, anxiety, and epilepsy.

P97. Elucidating Molecular Mechanisms that Diversify and Adjust Synaptic Strength, Luka Perehinets, University of California, Berkeley, Faculty mentor: Ehud Isacoff

Disruptions to nerve cell communication underlie serious neurological conditions, from Alzheimer's disease to epilepsy. At the heart of this communication are synapses — junctions where one neuron releases chemical signals to activate the next. Even along a single nerve fiber, individual release sites vary dramatically in how strongly and reliably they transmit signals. Understanding what drives this diversity, and how it shapes brain output, is a central challenge in neuroscience.To investigate this, we used the fruit fly larval neuromuscular junction, a model system that allows individual synapses to be imaged with exceptional clarity. First, we developed an optical imaging platform that simultaneously measures multiple features of synaptic function across hundreds of individual release sites. Using a fast calcium sensor, SynapGCaMP8f, this system captures synaptic responses across stimulation patterns from single pulses to rapid trains. To isolate spontaneous, activity-independent release, we used genetic knockdown of Complexin — a protein that normally acts as a brake on vesicle fusion. Second, we found that release sites within the same nerve fiber span a 100-fold range in baseline release probability, revealing striking intra-axonal variability. Third, this variation extends to short-term plasticity: facilitating and depressing synapses are interspersed along the same fiber in a pattern that stabilizes overall output across firing frequencies. Fourth, using functional and molecular maps generated by our platform, I aided in performing a multivariate analysis modeling the relationship between molecular composition and synaptic function. Levels of Unc13A, Bruchpilot, and RIM-binding protein together accounted for 35% of variance in baseline release probability, with Unc13A as the strongest predictor. Notably, short-term plasticity was less well predicted, suggesting additional determinants remain unidentified.Connectome projects map how neurons are wired — but wiring alone cannot tell us how strongly or reliably signals flow. By linking molecular composition to functional diversity, this work offers a path toward understanding how synaptic disruptions contribute to neurological disease.

P98. Ethanol-Induced Memory-Like States in DN1a Circadian Neurons, Vanessa Sanchez, University of California, Merced, Faculty mentor: Frederick Wolf

Alcohol Use Disorder (AUD) affects over 29.5 million Americans each year. Investigating early alcohol adaptations, such as ethanol tolerance, may reveal neural mechanisms driving AUD. Ethanol tolerance is an early form of behavioral plasticity and is defined as the acquired resistance to alcohol's pharmacological effects. The fruit fly Drosophila melanogaster develops ethanol tolerance. Rapid ethanol tolerance appears after the first ethanol dose has fully metabolized and mimics binge-like drinking behavior. The first ethanol exposure gives rise to two distinct ethanol memory-like states that mirror classical learning and memory circuits: labile anesthesia-sensitive tolerance (AST) and consolidated anesthesia-resistant tolerance (ART). Current research on ethanol memory-like states is limited to the mushroom body in the fly brain, an important learning and memory center. However, other brain regions have not been explored. Here we examined ethanol encoding memory-like states in the Dorsal Neuron 1 Anterior, or (DN1a). The DN1a's are circadian clock neurons that integrate light and temperature cues to adjust sleep timing and depth. Additionally, the DN1a's require synaptic activity and glutamatergic signaling to promote ethanol tolerance. We used a RNAi knockdown approach using the GAL4/UAS system in conjunction with a behavioral ethanol sedation assay to test whether known AST- and ART-associated genes, kinases, and neurotransmitters function in the DN1a neurons to promote ethanol memory-like states. Preliminary results indicate the DN1a neurons follow an AST trace. Understanding these interactions provides insights into the neurobiology of addiction and how drug memories can persist to influence future behavior.

P99. Ionotropic Receptor Ir25a in Rapid Ethanol Tolerance in Drosophila, Ansley Van Gorkom, California State University, Stanislaus, Faculty mentor: Fred W. Wolf

Our long-term research goal is to uncover the relationship between alcohol and temperature regulation pathways using Drosophila as a model system. Ethanol tolerance is an early form of neural and behavioral plasticity. Following binge-like ethanol intake, humans can develop hypothermia due to disrupted thermoregulation. We recently discovered that the second-order thermosensory projection neurons (TPN-II) and the circadian clock neurons (DN1a) are critical for the development of rapid ethanol tolerance in flies. Synaptic silencing of TPN-II neurons disrupts rapid tolerance. However, it is unknown whether ethanol affects thermosensation, or if it uses the circuit in a temperature signaling-independent manner. To test this, we investigated behavioral effects in mutant flies lacking the thermoreceptor Ionotropic Receptor 25a (Ir25a). Ir25a is expressed in the arista and detects decreases in temperature, generating a signal that is transmitted to higher order brain processing via the TPN-IIs. The goal of our study is to determine whether these primary thermoreceptors known as "cooling cells" are necessary for the development of rapid ethanol tolerance. Rapid tolerance mimics the effects of binge-like drinking, and is defined as the increased resistance to ethanol developed after the first ethanol exposure is metabolized. Tolerance is quantified by measuring the difference in time required for flies to reach 50% sedation

when comparing the first ethanol exposure (E1) and the second ethanol exposure (E2). Preliminary results show that Ir25a does not play a role in tolerance development but does play a role in ethanol sensitivity in heterozygous Ir25a mutant flies. Future work will examine the role of homozygous Ir25a mutants and additional thermoreceptors in rapid ethanol tolerance, helping to define the relationship between temperature regulation and ethanol tolerance.

P100. Using Light to Control Decision Making, Stephanie Linares, Jacki Thacker, California State University Channel Islands, Faculty mentor: Gareth Harris

Our lab uses a variety of behavioral and genetics methods to understand how organisms smell attractive and repulsive odors and make complex decisions during exposure to distinct uni sensory or multisensory conditions. Based on this research focus we have chosen to integrate the use of light, optogenetics, as a stimulus to be able to manipulate and control these behaviors. Optogenetics allows precise control of neural circuits using light, providing an unprecedented way to connect neural activity with behavior. Channelrhodopsin-2 (ChR2), an algal protein, is a light sensitive cation channel that can be used to temporally control neuronal excitement in the brain. C. elegans, with its well-mapped nervous system and flexibility to genetic manipulation, is a powerful model for dissecting how specific neurons drive behaviors. In this study, we aim to use optogenetics methods to control neural function in the worm and manipulate behavior in both normal and genetic mutant worms that lack specific neuron or communication.In this study, we demonstrate that our lab has used a protocol that allows the expression of channel rhodopsin to control a known behavior of the worm, egg laying(Emtage et al. 2012). This provides a platform to further use optogenetics to manipulate select parts of the worm's brain. We show using a worm that expresses a transgenic Channel Rhodopsin in the egg-laying controlling motor neurons of the hermaphrodite worm, was able to stimulate egg laying upon providing blue light exposure as previously shown. We now aim to use optogenetics to, 1) understand further mechanisms involved in egg laying and, 2) use light to manipulate and restore olfactory-based decision-making behaviors in our lab.

P101. Worms are suckers for perfumes too! Identification of molecular pathways that mediate nematode perfume attraction, Joy Gendy, Basia Piekarski, California State University Channel Islands, Faculty mentor: Gareth Harris

The perfume industry reaches over 60 billion dollars in revenue annually, partially due to the numerous chemicals that allow for a cologne's highly attractive aroma for human individuals. Millions of people frequently use a variety of colognes/perfumes that are generally perceived as attractive between individuals. The emphasis on use of colognes continues to grow due to factors

like increasing the awareness of personal grooming, production of luxury and exotic perfumes, and variation in an individual's preference for perfumes. How these perfumes are sensed, processed, and generate attraction and specific preferences is still not understood. Using a population chemotaxis assay that addressed olfaction behavior to cologne odors, we report that the model nematode worm, C. elegans, can smell and perform strong attraction behavior towards various types of oil-based colognes. These colognes, Chanel, Prada, YSL, Cartier, Dior, and Dolce and Gabbana are all attractive to the worm. Using the worm, we have identified worms to be strongly attracted to colognes of various forms. Populations of worms were exposed to colognes on a large agar plate and attraction index was calculated. We have also begun to characterize the neural architecture that drives this sensorimotor behavior. We have identified multiple sensory neuron-expressed ion channels that mediate this odor-guided attraction to cologne stimuli. Ultimately, we provide a platform to thoroughly understand the mechanisms and future neuronal circuits that mediate sensory-guided behavior to a complex human-sensed stimulus, like perfumes. This will also provide an avenue to understand how humans generate preference to complex regularly used perfumes from Chanel to Dolce and Gabbana.

P102. Should I Stay or Should I Go? The Effects of Odor Repellents On Worm Decision-Making, Felix Zendejas, Lily Yered, California State University Channel Islands, Faculty mentor: Gareth Harris

Humans continuously receive and process sensory information from the world around them. Multiple sensory cues are integrated in the brain to shape behavior, interactions, and decision-making in everyday life. Many neurological and psychiatric disorders—including Autism Spectrum Disorder, schizophrenia, and ADHD—show challenges while processing single and, in some cases, multiple simultaneously presented cues. As a result, abnormal coordination of these multisensory pathways can be debilitating for an individual. Despite growing recognition of the significance of multisensory behavior in humans, the underlying mechanisms and neural pathways involved in the brain are poorly understood. Using the invertebrate worms, C. elegans, as a model organism, we assess behavioral differences in the food-leaving escape responses to various repulsive odorants, including octanal, hexyl acetate, and methyl nonyl ketone. We compare these chemical-dependent escape responses to the previously studied food-leaving response of the repulsive odorant, 2-nonanone. Our experiments with wild-type worms showed repulsion to all tested smells, with each chemical inducing food-leaving and distinct escape patterns. The results showed octanal produced immediate food-leaving, whereas methyl nonyl ketone had a decreased leaving rate compared to 2-nonanone. Hexyl acetate showed similar escape patterns to the 2-nonanone dependent food leaving. These findings raise the question of whether structurally similar odorants stimulate the same or different neural circuits. To explore this, we are currently testing genetic mutants and transgenic lines with altered neurons and neurotransmitters. From this

research, we hope to deepen our understanding of how organisms respond to different, yet chemically similar, environmental stimuli.

P103. From Menopause to Neurodegeneration: How Estrogen Decline May Promote Metabolic Stress in Alzheimer's Disease, Sarah Flores, California State Polytechnic University, Pomona, Faculty mentor: Glenn Kageyama

Nearly two-thirds of individuals diagnosed with Alzheimer's disease (AD) are women, a disparity that cannot be explained solely by differences in lifespan. Increasing evidence suggests that metabolic dysfunction plays a central role in AD pathogenesis, leading some researchers to refer to AD as "type 3 diabetes," characterized by brain insulin resistance and impaired glucose metabolism. The menopausal transition represents a major neuroendocrine change unique to women and may interact with these metabolic disturbances to increase vulnerability to AD. Estrogen plays a critical role in maintaining neuronal energy metabolism by promoting glucose uptake, enhancing mitochondrial bioenergetics, regulating insulin signaling, and limiting oxidative stress. During perimenopause and menopause, declining estrogen levels are associated with reduced cerebral glucose metabolism, mitochondrial dysfunction, and increased oxidative stress. Impaired glucose utilization in the brain may lead to glucotoxicity, characterized by chronic metabolic stress, insulin resistance, and increased production of reactive oxygen species. In response to reduced glucose availability, the brain may shift toward increased lipid utilization for energy. This metabolic adaptation may promote lipotoxicity, in which excess lipid metabolites, oxidized fatty acids, and ceramides accumulate and contribute to mitochondrial damage, inflammation, and neuronal dysfunction. Both glucotoxicity and lipotoxicity may increase the burden of cellular damage signals, including misfolded proteins, oxidized lipids, and mitochondrial components. These molecules can function as damage-associated molecular patterns (DAMPs) that activate innate immune pathways in microglia through pattern recognition receptors. Chronic activation of these inflammatory pathways is believed to contribute to amyloid-β accumulation, tau pathology, and neurodegeneration. Together, these findings support a model in which menopause-associated estrogen decline may exacerbate metabolic stress in the brain, promoting glucotoxic and lipotoxic environments that contribute to Alzheimer's disease development. Understanding these mechanisms may help explain the disproportionate burden of AD in women and inform strategies for sex-specific prevention.

P104. Modulation of Neural Stem Cell Fate by Reactive Astrocytes, Ashley Hein, Stanford University, Faculty mentor: Irving Weissman

An accumulation of reactive astrocytes, microglia, and oligodendrocyte progenitor cells around the site of injury is a key structural component of the glial scar. While initially this is beneficial as it

restrains inflammation, chronic glial activation leads to suppression of axonal growth as well as secretion of neurotoxic substances. This project investigates the ways in which neural stem cell differentiation is influenced by cytokines secreted by reactive astrocytes, and how this contributes to the formation of the glial scar. In vitro experiments exposing NSCs to A1-inducing cytokines (IL-1α, TNFα, C1q), A2-inducing cytokines (IL-1β, TNFα), astrocyte-conditioned media, and co-cultures with reactive astrocytes demonstrated that A1-derived signals upregulate Gfap, C3, PDGFRA, and Sox10, promoting differentiation into reactive astrocytes and OPCs. IL-1α drove A1 astrocyte markers, while TNFα favored $C3^+$ OPC formation. With research suggesting that $C3^+$ OPCs are able to differentiate into A1 astrocytes, this could mean reactive astrocytes are not only the receptors of microglia secreted cytokines but also perpetuating a positive feedback loop where they secrete factors themselves that reinforce the negative effects of the scar and impair recovery. Further experiments focus on investigating whether, through use of inhibitors such as dantrolene and salubrinal, which may prevent the metabolic switch in reactive astrocytes, this effect on neural stem cells can be mitigated.

P105. Investigating Developmental Lethality Caused by Pan-Neuronal Expression of Human Amyloid Beta 42 in Drosophila melanogaster, Gabriella Palavicino, Michelle Batbayar, Gabriella Palavicino, University of California, Santa Cruz, Faculty mentor: Jeremy Lee

Alzheimer's disease (AD) is a progressive neurodegenerative disorder and the most common cause of dementia worldwide. One of its pathological characteristics is the accumulation of extracellular amyloid plaques within the brain. These plaques are primarily composed of the peptide amyloid beta (Aβ), which are generated through sequential proteolytic cleavage of the amyloid precursor protein (APP) by β- and γ-secretases. Among the various forms of Aβ, the 42-amino acid species (Aβ42) is believed to play the most significant role in AD pathology. Aβ42 is more prone to aggregation than the shorter Aβ40 isoform, leading to enhanced oligomerization. Familial AD mutations often increase the relative production of Aβ42 or alter the Aβ42/Aβ40 ratio, thereby promoting amyloid deposition and accelerating disease onset. Experimental evidence indicates that even subtle changes in this ratio can have profound effects on neuronal viability and synaptic function[1][2]. We previously generated transgenic fly lines carrying Aβ42 under GAL4-UAS control. We observed that, Drosophila melanogaster lines with two copies of UAS-Aβ42 expressed pan-neuronally under the Appl-Gal4 driver, do not survive to adulthood; flies carrying only a single copy, however, do survive to adulthood. This indicates that elevated levels of Aβ-42 can induce developmental lethality. To investigate this phenomenon further, and to identify juveniles carrying two copies of Aβ42, we generated flies carrying Aβ42 with a Green Fluorescent Protein (GFP) marker and Aβ42 with an mCherry marker, to determine the developmental stage at which homozygous expression of Aβ42 becomes lethal. The validity of the GFP/mCherry construct is analyzed by comparison with a control cross lacking the GFP and mCherry transgenes. As D. melanogaster is a widely used model organism for AD, we hope that identifying the specific stage at which lethality occurs will facilitate the identification of the mechanisms by which Aβ42

induces developmental lethality. This may provide insight into the mechanisms contributing to Aβ42 toxicity.

P106. Investigating whether the human antimicrobial peptide, LL37, can ameliorate the neurodegenerative phenotypes induced in Drosophila by expression of the human amyloid alpha (Aα) peptide, Svasti Kandpal, Daniel Flores, University of California, Santa Cruz, Faculty mentor: Jeremy Lee

To assess the effects of A**α** on neuron function, longevity, and climbing assays were conducted on flies pan-neuronally expressing A**α**, Aβ1-42, or both peptides using the Appl-Gal4 driver line. Scanning electron microscopy (SEM) and RNA sequencing were used to determine A**α**'s structural and molecular effects on the developing eye structure and gene regulation, respectively in flies expressing these peptides under the GMR-Gal4 driver. The results of these assays indicated that Aβ1-42 and A**α** have deleterious effects on locomotive ability, overall lifespan, on ommatidia and bristle structures in the adult eye, and on gene expression, with the most significant degenerative effects seen in flies co-expressing Aβ1-42 and A**α**. In addition, expression of A**α** or Aβ1-42 in developing eye neurons, under the GMR-Gal4 driver, led to increased levels of neuronal apoptosis in eye imaginal discs as compared to controls. All these data suggest that A**α** exacerbates the effects of Aβ1-42 and has similar but less severe neurodegenerative effects when expressed on its own. Our current work expands on these findings by investigating the role of A**α** and its potential interaction with the human antimicrobial peptide LL37. Previous results from our collaborator, Annelise Barron at Stanford, show that the human antimicrobial peptide LL37 interacts with Aβ1-42. In our lab, we have shown that coexpression of LL37 with Aβ1-42 ameliorates the deleterious effects of Aβ1-42. We hypothesize that interaction with LL37 may also modulate A**α** aggregation and, as a result, affect the severity of AD-like pathology and progression induced by A**α**.

P107. Phenotypic Characterization of Synthetic Amyloid Beta (Aβ) Mutations in a Drosophila Model of Alzheimer's Disease, Lucero Vergara, Alicia Jimenez, Kaia Levy-Kanenaga, University of California, Santa Cruz, Faculty mentor: Jeremy Lee

Alzheimer's disease (AD) is a progressive neurodegenerative disorder that affects more than 55 million people worldwide. A central hallmark of AD pathology is the buildup of amyloid beta (Aβ) plaques, which disrupt neuronal signaling and drive neurodegeneration. Using transgenic Drosophila models expressing wild-type (WT) Aβ pan-neuronally, our lab has observed AD-like phenotypes, including reduced lifespan, impaired locomotion, and altered gene expression. Prior in vitro studies from Dr. Jevgenij Raskatov's lab identified two Aβ42 side-chain modifications, Q15E and N27D (deaminations), that reduce Aβ42's cytotoxicity. Based on these results, we generated transgenic Drosophila carrying the corresponding missense mutations. Using the GAL4-UAS system, we compared flies expressing WT Aβ42 with those expressing single and double-mutant

variants to evaluate their effects on AD-related phenotypes in vivo. Longevity assays showed that flies expressing the Q15E variant of Aβ42 survived significantly longer than flies expressing WT Aβ42. A similar but less dramatic increase in longevity was seen for flies expressing the N27D variant and double mutant expressing flies. Morphological analysis with scanning electron microscopy (SEM) of the compound eye revealed distinct differences among WT and mutant genotypes. SEM results supported longevity results, showing a partial rescue effect associated with the missense mutations compared to WT Aβ42. Learning and memory are currently being assessed through the Aversive Phototaxic Suppression assay, and inflammatory cytokine expression is being quantified via qRT-PCR. Together, these approaches will allow us to determine the extent to which these mutations mitigate functional decline and to better understand how altered Aβ42 aggregation kinetics influence toxicity in vivo.Together, these findings suggest that specific Aβ42 modifications can mitigate toxicity in vivo, with differential effects depending on the mutation. With this work, we hope to advance our understanding of the structural basis of Aβ42 toxicity and aggregation and provide insight into potential therapeutic strategies for Alzheimer's disease.

P108. Synaptic Plasticity Alzheimer's Research, Noah Stolier, Subodh Poudyel, University of California, Santa Cruz, Faculty mentor: Jeremy Lee

Alzheimer's disease (AD) is a progressive neurodegenerative disorder characterized by deficits in memory, learning, lifespan and motor skills. A hallmark of the disease is excessive amyloid beta 42 (Aβ42) accumulation, a cleavage product of the amyloid precursor protein (APP) pathway which aggregates and disrupts neuronal functions and circuitry, leading to cell death. Alzheimer's debilitates millions of individuals, yet therapeutic strategies to restore neuronal structure remain limited. The Lee Lab at UCSC has generated a Drosophila melanogaster model that expresses Aβ42 pan-neuronally via the GAL4-UAS system to study Alzheimer's-like degeneration in vivo. Previous projects in our lab have shown phenotypes with deleterious effects on lifespan, eye morphology, behavior, and gene expression—mirroring aspects of AD pathogenesis. In Dr. Yi Zuo's lab at UCSC, psychoplastogens such as tabernanthalog have been shown to reverse dendritic atrophy caused by chronic stress leading to neuronal and behavioral rescues, suggesting a potential for rescuing neuronal circuitry. Additional studies from Dr. Charles Nichols lab at LSU have visualized increased dendritic formation in Drosophila that have been exposed to 2,5-dimethoxy-4-iodoamphetamine (DOI), a 5-HT2A receptor agonist. Given the neurodegenerative phenotypic effects of Aβ42 in Drosophila, we hypothesize that psychoplastogen-induced synaptic plasticity will partially rescue deficits in lifespan and motor function caused by Aβ42 accumulation. Our project aims to test this hypothesis by conducting lifespan and climbing assays on AD model flies either dosed with DOI, ketanserin tartrate, a 5-HT2A receptor antagonist, or an untreated vehicle to measure improvements in longevity and motor function compared to non-expressing controls. Based on the results, we plan to investigate

the molecular mechanism driving the hypothesized phenotype through gene regulation analysis and optogenetic studies. Overall, the long term goal of this research is to advance the understanding of Alzheimer's disease by elucidating the role of synaptic dysfunction in the pathology, potentially guiding future treatments.

P109. The human antimicrobial peptide, LL-37, mitigates amyloid beta-induced deleterious effects on apoptosis, motor function, longevity, and gene expression in Alzheimer's disease model Drosophila, Aranza Gomez, Daniela Ochoa, University of California, Santa Cruz, Faculty mentor: Jeremy Lee

Alzheimer's disease (AD) affects an estimated 7 million Americans, a number projected to double by 2060 according to the Centers for Disease Control and Prevention. Aβ42 peptides are thought to contribute to AD pathology due to their aggregation into fibrils and subsequent formation of neurotoxic plaques. In vitro studies have shown that the human antimicrobial peptide LL-37 can reduce Aβ42 fibril formation. This study investigates the potential rescuing effects of LL-37 in vivo using Drosophila melanogaster as a model organism while using genetic, biochemical, and behavioral approaches.Four transgenic Drosophila lines were used, expressing: Aβ42 alone, LL-37 alone, co-expressing Aβ42 and LL-37, and control non-expressing, with neuronal-specific expression. Longevity assays and RNA profiling were conducted to assess phenotypic and molecular effects. Co-expression of LL-37 with Aβ42 resulted in a significant rescue of both eclosion rate and adult survival compared to Aβ42 expression alone. Conversely, LL-37 expression alone had a detrimental effect on survival. RNA sequencing revealed that LL-37 reversed Aβ42-induced gene expression changes, particularly in genes related to proteolysis when flies expressed both proteins. Data from a Rapid Iterative Negative Geotaxis assay suggest that LL-37 may also partially restore locomotive deficits associated with Aβ42 expression.These findings suggest that LL-37 may mitigate AD-related pathology in vivo and highlight its potential as a therapeutic candidate in the treatment and prevention of Alzheimer's disease. The observed rescue in survival supports the idea that LL-37 may interfere with Aβ42 fibril formation and also modulate downstream biological processes affected by amyloid toxicity, such as proteolysis and neurodegeneration.

P110. Neuroimaging reveals differential patterns of human brain activity associated with acts of reflective attention, Emma Withrow, Sonoma State University, Faculty mentor: Evan Lintz

Working memory (WM) is the cognitive system that supports the maintenance and manipulation of mentally represented information. Directing reflective, or internally oriented, attention to a WM representation, also known as refreshing, results in a number of cognitive-behavioral benefits. Findings from functional magnetic resonance imaging (fMRI) studies of reflective attention have shown evidence of increased activity in anterior cingulate cortex that scales with increasing WM load. Due to the low temporal resolution of fMRI, it is unknown whether this increase in activity is

linked to cognitive control signals originating in prefrontal cortex, or if it is related to sensory/perceptual components of the WM representation itself (i.e., orthogonalized representations in visual cortex). Subsequent electroencephalography (EEG) experiments presented participants with a continually changing set of items to maintain in WM, interspersed with cues to refresh or remove certain items from the current set. We uncovered an apparent monotonic relationship between refreshing at various WM loads, and the scalp voltage observed in EEG. Further, neural activity appears to overlap temporally with event-related potential (ERP) components associated with the reinstantiation of neural activity that holds information about the perceptual aspects of WM representations. The current work seeks to conduct exploratory analyses of the existing dataset with the goal of: 1) confirming the previously suggested relationship between neural activity and scalp voltage by controlling for potential confounds related to stimulus presentation, and 2) to apply these same controls to enable a more fair comparison of refreshing and another putative WM process: removal. We hypothesized that not only would the previously hinted-at relationship be supported, but also that direct comparisons of refreshing and removal would demonstrate similar monotonic load-voltage relationships, further supporting the hypothesis that irrelevant-item cues promote refreshing-based strategies rather than an active removal process.

P111. Exploring Cellular Variability in the Leech Nervous System, Christopher Dascalescu, California State University, Sacramento, Faculty mentor: Michael Wright

Our lab is interested in how nervous systems produce behaviors, such as walking, that are similar between individuals but arise from different connectivity patterns. This variability provides flexibility in producing behaviors but also needs to be constrained so that the behavior is functional. We are looking at how variability in neuronal intrinsic properties provides this flexibility. To address this question, we are using the medicinal leech, Hirudo sp., because of their easily identifiable, large neurons that create a stereotyped behavior; swimming. To explore the role of variability in producing behavior, we measured several input-output parameters in four neurons known to play a role in the production of swimming behavior, including a neurosecretory neuron, the Retzius cell, and three mechanosensory neurons that respond to light touch (T cells), pressure (P cells), and nociception (N cells). We introduced two types of input patterns to each type of neurons. The first input pattern was designed to test steady-state encoding properties of these neurons. In these experiments, constructed Frequency Current (FI) Curves to assess how activity patterns change as a function of input current. A second pattern composed of white noise was used to measure dynamic encoding properties of these neurons. Because the input patterns are the same for every neuron, any variability observed must be due to variability in the intrinsic properties of the neurons themselves. With this experimental paradigm, we are trying to discover general principles that underly how nervous systems produce behaviors that arise from variable components.

P112. Resveratrol Metabolites Elicit Neuroprotection via a Cell-Surface Receptor, Genna Kang, University of Southern California, Faculty mentor: Rayudu Gopalakrishna

Resveratrol is a polyphenol commonly found in grapes, berries, and red wine. Both epidemiological and experimental studies suggest that it protects against neurodegenerative diseases such as Alzheimer's disease (AD). After orally administering moderate doses to humans and animals, resveratrol rapidly metabolizes to glucuronide and sulfate conjugates, and only these conjugates are able to be detected in plasma. However, how these conjugates exert neuroprotection is still unknown.Hypothesis: Resveratrol and its metabolites bind with high affinity to the cell-surface 67-kDa laminin receptor (67LR), specifically the "peptide G" region, and rapidly triggers signaling by elevating cyclic AMP (cAMP) to promote neuroprotection.Model system: We used Neuroscreen-1 (NS-1) neuronal cells and induced cell death by serum deprivation. We tested whether resveratrol or its conjugates can protect these cells at subnanomolar or low-nanomolar concentrations that are achievable in the brain.Results: Very low concentrations (0.01–10 nM) of resveratrol and resveratrol glucuronide protected neuronal cells from death. Protection was associated with a rapid, several-fold increase in intracellular cAMP. Both, the neuroprotective effect and the increase in cAMP were blocked by antibodies specific to 67LR. Molecular docking studies indicated high-affinity binding of resveratrol and its conjugates to a palindromic sequence within the peptide G region of 67LR. Binding studies using synthetic peptide G confirmed high-affinity interactions (Kd = 0.2–2 nM). Resveratrol glucuronide bound with a higher affinity than the parent compound, likely due to the presence of a glycosaminoglycan-binding motif in peptide G.Conclusions: Extremely low concentrations of the resveratrol glucuronide conjugate elicits neuroprotection through high-affinity binding to the cell-surface receptor 67LR. Notably, amyloid-β (as a prion complex) binds to the peptide G region of 67LR and promotes neuronal death. Resveratrol metabolites bind to this same site but instead, promote neuroprotection. This provides a potential mechanism by which resveratrol could counteract amyloid-β toxicity and help prevent AD.

P113. Dissecting the distinct but overlapping brain circuits that control attraction and repulsion to smells to mammalian sensed odors, Leslie Suarez, Kiely Hamilton-Payne, California State University Channel Islands, Faculty mentor: Gareth Harris

Dissecting the distinct but overlapping brain circuits that control attraction and repulsion to smells to mammalian sensed odors Leslie Suarez, Kiely Hamilton-Payne, Emily Chang, Amber Seader, Annabelle Tran, Brianna Ramos and Gareth Harris. CSUCI. Biology program. Organisms across the phyla are capable of sensing an array of sensory cues to control or shape complex behavioral responses to survive in a complex environment consisting of an array of attractive and repulsive dangerous cues. Mammalian systems extensively use olfactory and gustatory behavior to fine-tune these sensory-dependent decision- making behaviors. Despite understanding the importance of behavioral responses to cues in the form of odors in shaping decision-making behavior, the

underlying mechanisms that mediate these responses at the level of sensation, processing, integration, and modulation of these sensory dependent responses are not fully understood. To understand these mechanisms, we use the invertebrate worm, C. elegans to characterize attraction to mammalian sensed odorant cues. We show that hermaphrodite worms are attracted to catnip oil cues and identify select sensory mechanisms that mediate this attraction, identifying multiple sensory genes (molecular substrates) that are involved in this chemosensory response to a mammalian sensed cue, that is highly attractive in many cats. We use a population-based chemotaxis behavioral assay to identify attraction to catnip oil and candidate genes involved. We have identified sensory transduction mechanisms, including G-proteins and cyclic nucleotide-gated ion channels, that regulate odor-dependent attraction to mammalian sensed catnip oil cues. We have also identified that nematodes across different species perform differently when exposed to catnip oil. We therefore provide a platform to use C. elegans as a model for studying olfactory-dependent pathways to mammalian cues. Furthermore, we have preliminarily found attraction to catnip alternatives, including honeysuckle and gall fruit, established to be attractive in cats. Future directions include dissecting the molecular mechanism used for attraction to these catnip alternatives and determining the responses of different species. This work overall allows characterization of the neural mechanisms that shape olfactory behavior and decision-making in higher systems.

P114. Glypican Dally Displays Embryonic Lethality, Hope Gillman, San Francisco State University, Faculty mentor: Blake Riggs

Cell division is vital for the development of all multicellular organisms, and is driven by the asymmetric partitioning of cell fate determinants. Proper positioning of these determinants are critical for the formation of complex tissues and organs, including the central nervous system of Drosophila melanogaster. Glypicans are heparan sulfate proteoglycans tethered to the surface of the plasma membrane via a glycosylphosphatidylinositol (GPI) anchor. Through their heparan sulfate chains, they function as co-receptors that bind growth factors and morphogens, helping control how these signals influence cell proliferation and differentiation. Previous researchers in the Riggs lab found that the ER integral membrane protein Jagunal is important and is necessary for asymmetric division of the endoplasmic reticulum during mitosis. It was also found that the glypican, Dally (division a abnormally delayed), was one of the genetic interactors of Jagunal, suppressing the Jagn-RNAi induced rough eye phenotype. This project will be focusing on investigating the lethality of Dally homozygotes in Drosophila melanogaster. This is an excellent model organism to understand how cells are organized and partitioned during cell division as 65% of their genes are homologous to those in humans. We hypothesize that Dally plays a crucial role in the development of the Drosophila central nervous system. In order to assess if the Dally mutation is affecting mortality, we conducted a lethality assay with homozygous Dally deficient fly lines. It was found that the Dally homozygotes are lethal at the embryonic stages. Next steps are to do RNA extraction of Dally mutant allele embryos followed by RT- qPCR analysis in order to

analyze gene expression levels, classifying if the fly line is a null or hypomorphic mutant. Furthermore, our findings will contribute to the understanding of cell division and neurogenesis.

P115. Optogenetic WNT signaling drives AP axis self-organization in a human neural tube model, Justine Leparmentier, University of California, Berkeley, Faculty mentor: David V. Schaffer

Organoids, three-dimensional structures arising in vitro from initially uniform stem cells, recapitulate key aspects of tissue organization and have become essential tools for studying human development, disease, and regenerative medicine. Recent microfluidic approaches have advanced the field by exposing hPSC-derived tissues to opposing morphogen gradients, generating neural tube-like structures with regional identity. However, these systems rely on externally imposed, largely static gradients, offering spatial control but limited capacity to dynamically modulate signaling in time or at the cellular level. The resulting patterning is primarily from imposed gradient rather than an emergent product of cell-autonomous developmental programs. Wnt signaling plays a critical role in establishing anterior-posterior (A-P) identity along the neural tube. Our lab recently demonstrated that optogenetically patterned Wnt activation drives self-organization and A-P polarization in hESC-derived gastruloids [1]. Building on that work, we hypothesized that spatially defined Wnt activation could direct neural tube-like patterning (forebrain, midbrain, hindbrain) through cell-autonomous development triggered by an initial patterned stimulus. We used hESCs engineered with an optogenetic Cry2-based Wnt activation system (OptoWnt) and programmable LAVA (Light-Activated Virtual Actuator) board illumination to apply spatially and temporally defined light patterns in co-cultures of OptoWnt and wild-type cells [2-4]. Salt-and-pepper distribution of Wnt-active and inactive cells exposed to light induced segregation and self-organization in both 2D and 3D contexts. Immunostaining confirmed spatial segregation of forebrain (OTX2) and hindbrain(HOXB4) markers consistent with early A-P neural tube patterning. Bulk RNA sequencing further supported a Wnt dose-dependent specification of regional identity, with high Wnt levels corresponding to hindbrain-like, moderate levels to midbrain-like, and low levels to forebrain-like expression profiles. These results suggest that dynamic optogenetic control, combined with intrinsic cellular heterogeneity, can drive early neural tube-like regionalization and may provide a foundation for extending our lab's gastruloid platform toward modeling aspects of human neural development. References [1] Johnson, H. J., McMullin, D. M., Zimmermann, J., Kim, C., Repina, N., Bhalerao, R., Nowakowski, T., & Schaffer, D. V. (2026). Optogenetic WNT signaling drives germ layer self-organization in a human gastruloid model. bioRxiv. https://www.biorxiv.org/content/10.64898/2026.02.23.707602v1[2] Repina, N. A., Johnson, H. J., Bao, X., Zimmermann, J. A., Joy, D. A., Bi, S. Z., Kane, R. S., & Schaffer, D. V. (2023). Optogenetic control of Wnt signaling models cell-intrinsic embryogenic patterning using 2D human pluripotent stem cell culture. Development, 150(14), dev201386. https://pubmed.ncbi.nlm.nih.gov/37401411/[3] Repina, N. A., Johnson, H. J., McClave, T., Kane, R. S., & Schaffer, D. V. (2020). Protocol to fabricate engineered illumination devices for

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P116. Viscoelastic HA-CS Matrices Alter TrkA Expression That Drives Non-Canonical MAPK/PI3K Signaling to Direct Lineage Commitment, Leah Hecht, University of California, Berkeley, Faculty mentor: David Schaffer

Neural stem cells (NSCs) in the hippocampus enable lifelong neurogenesis, but their regulation – particularly by altered extracellular biophysical cues – is not fully understood. Previous studies have shown that adhesion molecules can bias NSC differentiation in altered mechanical microenvironments. However, other biochemical changes, such as changes in chondroitin sulfate (CS) deposition and their respective sulfation patterns, remain elusive. To study how altered CS levels in the ECM affect NSC lineage commitment, we implant rat NSCs from the hippocampus within an 3D in vitro model to study these biochemical events. More specifically, we employ a hyaluronic-based hydrogel containing varying CS levels with matched stiffnesses to dissect the effect of CS deposition on NSC differentiation. We find that NSCs exhibit a higher prevalence of glial fate when differentiated in the higher CS environments, an effect compounded alongside the previously validated effect of stress relaxation on differentiation. RNAseq analysis indicates that within 24 hours, incorporation of CS increases expression of certain cell membrane proteins and growth factor receptor pathways, while others are pruned; notably, the significant upregulation of Ntrk1 and Itgb7. A KEGG pathway analysis reveals an upregulation of three key pathways, PI3K-Akt, MAPK, and IgSF CAM, indicating that this CS-rich niche transcriptionally regulates the cells, priming them towards gliogenesis. In response to elevated CS levels, cells expressed increased Itgb7 and decreased CD44, along with matrix deposition of Col2a1, indicating changes in adhesion receptor transcription, potentially altering changes in adhesion to the CS microenvironment. Further studies will elucidate the mechanism by which it occurs to understand how CS modulates NSC lineage commitment.

P117. Effects of capsaicin and noxious heat on central sensitization to pain in an insect model, Angus Hamilton, Josiah McGrew, San Francisco State University, Faculty mentor: Megumi Fuse

Chronic pain is an ongoing problem in this country, and with overdose deaths continuing to rise, there is a great need for an alternate solution. The hornworm, Manduca sexta, is becoming a model for studying ongoing sensitivity to pain after mechanical injury. But even though much of the chronic pain experienced in humans arises from thermal injury, this has not yet been studied in our

insect model. Vertebrates and invertebrates both sense thermal stimuli through a variety of TRP receptors. Many chemical ligands, like capsaicin, also work through these channels, simulating thermal nociception in absence of noxious temperatures. M. sexta may prove a valuable model for testing this thermal and chemical sensitization as it possesses TRPA channels to sense heat similarly to Drosophila melanogaster, but from our phylogenetics it seems to lack the TRPV1 channels that capsaicin usually acts through. We tested the response to capsaicin in M. sexta and found that it sensitizes them in a dose-dependent manner, while the TRPV1 channel inhibitor, capsazepine, was largely ineffective at preventing sensitization. Heat also appears to sensitize the insect, verifying the multimodal nature of some M. sexta nociceptors. We further conducted a phylogenetic analysis, where the TRPV1 channels that capsaicin usually acts through cannot be found in M. sexta. We are therefore also testing whether the TRPA1 channel inhibitor HC-030031 can block the effects of capsaicin and laser-based heat, as capsaicin has been shown to activate TRPA1 channels in canine kidney cells in absence of TRPV1 channels.. This data will be discussed in the context of using M. sexta as a model for novel pain management strategies.

P118. Population-Level Associations Between Psychiatric Burden and Brain Aging in a UK Biobank Dataset, Robert Cheng, University of Southern California, Faculty mentor: Andrei Irimia

Psychiatric conditions and socioeconomic factors are increasingly recognized as key determinants of brain health, but their relative contributions to structural brain aging remain unclear. We analyzed UK Biobank participants with T1-weighted MRI and behavioral, socioeconomic, and psychiatric data. Brain age gap (BAG), defined as predicted brain age minus chronological age, was used as a marker of brain aging. Multivariable linear regression models adjusted for demographic, socioeconomic, and lifestyle factors were used to assess associations.Psychiatric burden, particularly depression, was strongly associated with increased BAG, indicating older-appearing brain structure. Socioeconomic deprivation also showed robust positive associations. Lifestyle factors demonstrated comparatively smaller effects, with higher meat consumption associated with increased BAG and higher caffeine intake associated with decreased BAG.These findings suggest that psychiatric and socioeconomic factors are more strongly associated with brain aging than individual lifestyle behaviors, highlighting the importance of mental health and social context in shaping population-level brain health.

P119. APOE4 knock-in mice exhibit blood brain barrier dysfunction without cognitive impairment or astrocyte activation at 14 months, Hita Anand, University of California, Berkeley , Faculty mentor: Daniela Kaufer

Human Apolipoprotein E4 (APOE4) allele is a genetic risk factor for Alzheimer's disease that contributes to blood brain barrier dysfunction (BBBd) and cognitive impairments. In wild-type mice, aging leads to BBBd and memory impairment. However, while APOE4 knock-in mice have elevated BBBd compared to APOE3 controls, there is limited data on whether APOE4 mice have

worse memory function than age-matched APOE3 mice. We show that 14-month-old APOE4 mice develop significant BBBd measured by fibrinogen leakage but no significant cognitive impairment compared to age-matched APOE3 controls. Objectives: Our experiment aims to investigate differences in spatial memory and BBBd in APOE4 mice compared to APOE3 controls and within-genotype correlations of spatial memory and BBBd. Methods: We studied 10 APOE4 and 11 APOE3 male and female mice at 14 months. T-maze, Novel Object Recognition Test, and Barnes Maze were used to quantify cognitive function. IHC staining of fibrinogen and GFAP were used to quantify BBB leakage and astrocyte activation, respectively. Results: APOE4 mice demonstrated no significant impairment in long-term, short-term, and spatial working memory, and showed no significant increase in astrocyte activation relative to APOE3 controls. However, APOE4 mice had significantly elevated BBBd measured by fibrinogen extravasation than APOE3 controls. Conclusion: These results demonstrate that the APOE4 knock-in gene in mice causes increased BBBd measured by fibrinogen extravasation, but no significant change in astrocyte activation and cognitive function. We find a surprising lack of correlation between vascular pathology and memory in APOE4 mice, suggesting the presence of compensatory mechanisms in APOE4 mice that may preserve cognitive function and prevent neuroinflammation despite elevated BBB leakage.

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P120. Assumption-Free Uncertainty Quantification in Epigenetic Clocks, Megan Mitchell, University of California, Los Angeles, Faculty mentor: Matteo Pellegrini

Epigenetic clocks – models that estimate biological age from DNA methylation (DNAm) data – typically provide point predictions without accompanying measures of uncertainty. To address this limitation, our previously published model, BayesAge, used simulated data to generate a range of plausible ages for each individual. However, this approach was computationally intensive and systematically underestimated error. In this study, we evaluated split conformal prediction and its locally weighted variant as computationally efficient and statistically rigorous methods for quantifying the uncertainty of BayesAge's predictions. We found that split conformal inference produced empirically valid confidence intervals, even in finite-sample settings, with minimal residual bias. These findings suggest that conformal inference may be valuable for supporting high-risk decision-making tasks in clinical or forensic contexts, where reliable uncertainty bounds are essential.

P121. Enhancing RNAi-Based Control of Grapevine Red Blotch Virus Using a Multi-Target dsRNA Spray Formulation, Ahmed Alsulaimawi, Oregon State University, Faculty mentor: Laurent Deluc

Grapevine Red Blotch Virus (GRBV) poses a major threat to viticulture, reducing fruit quality and vine productivity, and vine replacement is currently the only effective management strategy. Spray-Induced Gene Silencing (SIGS), which exploits the plant's natural RNA interference (RNAi) defense through topical applications of double-stranded RNA (dsRNA), offers a non-transgenic and environmentally sustainable alternative to viral control. Previous work demonstrated that dsRNA targeting a single GRBV genomic region (HS9; GRBV RepA gene) significantly reduced viral gene expression in microvines and greenhouse grapevines. Building on these findings, we evaluated whether a multi-target dsRNA solution targeting four viral genes simultaneously could enhance and prolong antiviral efficacy. We compared HS9-derived dsRNA alone against a mixture comprising four GRBV hotspot-derived dsRNAs (HS1, HS4, HS7, and HS9), targeting the movement protein, coat protein, V3 ORF, and replication-associated protein genes, respectively. Both HS-derived dsRNA formulations were complexed with carbon dots (CDs) at a 200:1 CD:dsRNA ratio. GRBV-infected microvine V4 plants were treated with 20 nM, with spray buffer serving as the control. Viral gene expression was quantified by RT-qPCR at 0, 1, 2, and 3 months post-spray (mps). The multi-target formulation significantly reduced RepA expression at 2 and 3 mps relative to controls ($p < 0.05$), with silencing comparable to the single-target treatment

(CD:HS9-dsRNA). These results confirm the reproducibility of CD-mediated SIGS against GRBV and demonstrate that simultaneous targeting of multiple viral genes maintains silencing efficacy, supporting the development of a multi-target dsRNA spray for GRBV control.

P122. Experimentation and Simulation of Near-Infrared Light Penetration in Models of Oral Tumors for Near-Infrared Photoimmunotherapy, Samantha Schilling, Karen Martinez, California State University, Fresno, Faculty mentor: Gemunu Happawana

For over half a century, cancer treatment has primarily consisted of surgery, chemotherapy, and radiotherapy. While semi-effective, they all possess serious side effects, weakening the body's natural immune system and damaging nearby cells. Due to this, a major focus of cancer researchers has been uncovering cures to cancer that cause less harm to the body. One recent discovery is the use of near-infrared photoimmunotherapy (NIR-PIT), a treatment form that uses NIR light and an antibody photoabsorber-conjugate (APC) to induce selective necrotic cell death in targeted cancerous cells. Our research focuses on tracking and testing methods of evenly spreading NIR light throughout oral tumors in order to make NIR-PIT treatment more effective. This research is performed using ballistic gel as a substitute for oral tumors and shining NIR light throughout the tumor with and without varying amounts of silver particles to track how the light spreads. We aim to create a mathematical model of the spread of NIR light throughout an oral tumor in order to help researchers simulate and optimize NIR-PIT treatment for oral tumors.

P123. Comparative Evaluation of Molecular Visualization Platforms for Imaging the SARS-CoV-2 Spike and Surfactant Protein D (SP-D) Binding Complex, Shloka Raghavan, Santa Clara University, Faculty mentor: Angela Haczku

Visualization of protein-protein binding remains a major challenge, particularly for immunologically-relevant interactions. The SARS-CoV-2 Spike protein mediates infection of airway epithelial cells, while surfactant protein D (SP-D) is implicated in innate immune defense through putative neutralization of the Spike. Determining whether this protective function involves direct molecular binding requires reliable computational docking and high-fidelity structural visualization.Objectives: We aimed to compare molecular visualization platforms for their ability to analyze and render the SARS-CoV-2 Spike–SP-D docked complex, specifically evaluating (1) steric clash identification, (2) glycan interaction visualization, (3) structure editing without disrupting the complex, (4) high-quality image export (≥300 dpi), and (5) stability on accessible hardware such as a 2024 MacBook Air.Methods: Quantum algorithmic pruning was applied to reduce the conformational search space prior to docking, followed by Fast Fourier Transform (FFT)-based docking to predict favorable Spike-SP-D binding sites. The resulting complex was imported into three platforms: OneAngstrom's SAMSON, Schrödinger Maestro, and Mol*. The platforms were assessed against the five criteria above.Results: UCSF ChimeraX had to be excluded due to technical compatibility issues. SAMSON produced the highest quality rendered

images and supported detailed visualization of intermolecular contacts; however, it showed instability on laptop hardware, occasionally crashing during rendering tasks. Mol* offered the most stable and accessible experience, enabling rapid loading of docked structures, straightforward chain recoloring, and reliable inspection of contact regions, though with more limited rendering capabilities. Schrodinger Maestro access is still to be obtained to complete this comparison.Conclusion: Visualization platforms differ significantly in their suitability for large, docked protein complexes. SAMSON excelled in image quality, while Mol* provided greater stability and accessibility for structural exploration. Platform selection should be carefully considered and reflect the specific demands and available computational resources for optimal visualization of large protein complexes.

P124. Novel Application of Deep Learning Classification Models in Limbal Stem Cell Deficiency for Staging and Identifying Regions of Subepithelial Scar in Anterior Segment Optical Coherence Tomography, Sarah Wu, University of California, Los Angeles, Faculty mentor: William Speier

Limbal Stem Cell Deficiency (LSCD), an ocular disease characterized by degradation of the corneal epithelium, has no standardized or automated method for disease severity classification or tissue classification. We developed a deep learning-based classification framework that performs automated LSCD analysis directly from anterior segment optical coherence tomography (AS-OCT) images. The framework addresses two complementary tasks: (1) LSCD severity classification (Normal, Stage 1, Stage 2, Stage 3) using a modified InceptionV3 convolutional neural network, and (2) scar versus healthy tissue classification in Stage 3 LSCD using a masked autoencoder (MAE) with a finetuned Vision Transformer (ViT). Our dataset consisted of 134 scans with 3 three scans per eye for the stage classification task, and 113 scans with 3 scans per eye for the scar classification task. The LSCD severity model achieved 86% accuracy across all severity classes on an image level, an F1 score of 0.8589 and AUC of 0.9411 in multiclass classification. The patch-level scar versus healthy tissue model achieved 90.9% accuracy, an F1 score of 0.910 and an AUC of 0.987. These results demonstrate the feasibility of direct image-based classification for LSCD assessment and highlight transformer-based models as a robust, automated approach for characterizing clinically relevant scarring patterns, with the potential to standardize LSCD severity and tissue assessment and reduce clinician workload.

P125. Engineering Coccidioides posadasii with Red Fluorescent Protein via CRISPR-Cas9 for Enhanced In Vivo Imaging and Model Development, Peter Nguyen, University of California, Merced, Faculty mentor: Katrina K. Hoyer

To safely address the therapeutic challenges of Coccidioides spp. (responsible for Valley fever), the aim is to engineer a stable, live-attenuated C. posadasii ΔT mutant expressing red fluorescent protein (RFP). This reliable CRISPR-Cas9 tagging pipeline is crucial for developing advanced clinical models to evaluate host immune responses and screen novel antifungal drugs.An RFP reporter gene will be inserted into the C. posadasii ΔT mutant, targeting a non-essential genome locus using CRISPR-Cas9 transformation. An all-in-one CRISPR vector will be computationally designed encoding the Cas9 protein, a locus-specific single guide RNA (sgRNA) adjacent to an NGG PAM motif, and a targeted repair template. The donor template will carry an RFP reporter gene under fungal promoter control and pyrG knockout for transformation selection. To promote homology-directed repair, the reporter cassette will be flanked by ~1-kb homology arms matching the genomic sequences flanking the cut site. Lytic enzyme treatment will generate protoplasts lacking the fungal cell wall for plasmid delivery via PEG-CaCl2 transformation. After cell wall regeneration, transformants will be isolated via 5-FOA (pyrG) selection and screened through fluorescence microscopy. Finally, structural integration will be validated using junction PCR to confirm correct insertion boundaries, while quantitative PCR (qPCR) will detect single copy integrants.Initial efforts focus on in silico design and procurement of the CRISPR-Cas9/RFP vector. Following PEG-CaCl2 transformation, the goal is to be able to isolate clones with strong, localized nuclear red fluorescence during saprobic and parasitic stages for future biological use.An RFP-expressing avirulent C. posadasii strain provides a vital tool for real-time pathogen visualization and high-resolution host-pathogen interaction studies (e.g. tracking engagement with GFP-expressing host leukocytes). This pipeline validates the capacity to integrate other pre-clinical markers, such as ovalbumin (OVA) to track adaptive immune response during vaccine development and luciferase for in vivo tracking of fungal burden to screen therapeutics.

WCBSURC Poster Session PM - 1:30pm-3:00pm

Poster #127-216

Marine Biology #127-143

P127. Abundance of Microfibers in Sediments from Leo Carrillo State Beach and Catalina Island, California, Abby Lockmann, Kamryn Mortensen, California Lutheran University, Faculty mentor: Andrea Huvard

Microplastic pollution along California is a growing ecological concern for marine and human health. Synthetic microfibers, which are released from textiles during laundering cycles, comprise an overwhelming 70% of microplastic pollutants. With an estimated 100,000 per garment and 9,000,000 fibers shed per laundering cycle, microfibers contribute about two million tonnes of marine debris annually. While few current mitigation strategies exist, including textile engineering approaches, and waste treatment, little is known about the geographical distribution of these pollutants into marine environments, specifically sediments. The purpose of this study is to compare sediment contamination between two geographically different locations; Leo Carrillo State Beach in Malibu, an open-coastal beach, and Avalon Harbor, an enclosed harbor on Catalina Island, 22 miles off shore from Los Angeles, California. Given the pathways of the North Pacific Current, California Current and the proximity to runoff locations, it was predicted that there would be more microfibers in the Malibu location as compared with the Island Harbor. Sediment samples were randomly collected from areas between the subaerial beach and the breaking waves in the intertidal zone. Samples were processed using a 1:2 sediment-to-seawater ratio, then agitated to separate microfibers from sediment. A Büchner funnel filtration system was used to capture fibers on filterpaper, then quantified using light microscopy. Our findings suggest higher microfibers abundance at Leo Carrillo State Beach compared to Avalon Harbor. These findings emphasize the necessity to understand the transport, distribution, and ecological impacts of microplastics and fibers, in order to construct an effective mitigation plan.

P128. A Comparison of Microfiber Quantities in Wild-Caught and Farmed Oysters from the California Coast, Kiara Apeles, California Lutheran University, Faculty mentor: Andrea Huvard

Microfibers are fragmented contaminants from synthetic fabric materials like Polyester and Nylon. They are shed through laundry and wastewater, finding its way to and polluting the oceans. Filter feeders, like oysters, can be used as a biomarker to monitor the environmental conditions of its environment and are important in aquaculture. Farmed oysters are raised in a controlled environment where human activity is limited, unlike the wild oysters. Areas with heavy human activity influences their exposure to microfibers. This study aims to see if there is a difference in microfiber quantities between wild and farmed raised oysters along the coast of California. Wild oysters were collected from Channel Islands pier, and farmed raised oysters were from Chesapeake Bay. Using KOH, organic material was dissolved and filtered to be observed under a microscope to count for microfibers. Data was collected to test the number of microfibers in different sized oysters. There was a significant microfiber difference between the wild and farmed samples ($p < 0.0001$) that was positively correlated with the size of the oyster. A total of 239 microfibers were quantified, 210 sourcing from 20 wild caught Channel Islands oysters, 14 from 7 oysters farmed from Chesapeake Bay, and 15 oysters farmed from Morro Bay. Between the three sample locations, the wild oysters collected from Channel Islands contained a significantly higher amount of microfibers. This supports the idea that because of the controlled conditions, minimal microfiber exposure by human activity decreases the amount of microfibers found inside of oysters and other filter feeders.

P129. Analysis of the Composition and Quantity of Microfibers in Mytilus edulis from Ventura Harbor as Compared with Sediments from the Same Location, Ashley Van, Amber Wey, California Lutheran University, Faculty mentor: Andrea Huvard

As the popularity and practicality of synthetic textiles increase, so does the number of microfibers released into the environment. Microfibers are everywhere in our oceans, not only in open ocean water, but also are found in sediments and many marine animals. To increase our understanding of the microfiber pollution in animals, we aim to investigate microfibers in filter-feeding invertebrates such as mussels. This study's main objectives are to gain evidence of a mussel's viability as a biomarker by comparing it to sediment and to identify the composition of the main pollutants in Ventura Harbor. We wanted to focus on the jetties in Ventura Harbor as they attract a steady stream of surfers, fishermen, and boaters. To separate microfibers, the animals were completely removed from their shells, and the tissue was dissolved in KOH. Then the fluid is filtered to collect microfibers on filter paper. With sediment samples, we used the "wash" technique to suspend microfibers in water before filtration. We found a high correlation between the quantity of microfibers found in mussels ($n=60$) and the sediment in May, June, and July. There was no significant difference in the number of microfibers between the north side and south side of the jetty ($p>0.1$). To determine the microfiber composition, we used FTIR and identified fiberglass

and polystyrene fibers. These findings are essential for monitoring pollution, supporting conservation efforts, and increasing our understanding of the scope of plastic pollution in the ocean.

P130. Exploration of Chiton Populations in Central California, Sofia Ximena Garcia-Isabelli, Santa Clara University, Faculty mentor: Dawn Hart

Chitons, a type of marine mollusc, are vital intertidal grazers. Yet there are gaps within the recent data on their distribution in Central California. To address this gap, this study aimed to investigate species composition, morphology, and habitat association at four sites north and south of Monterey Bay. Using randomized quadrat sampling (0.25 m2) across the microhabitats of the intertidal, identification of differences between the 4 sites was observed. While Nuttallina californica dominated all sites, southern populations exhibited significantly higher abundance and distinct differences in average body length compared to northern groups. Morphological variation was strongly tied to spatial distribution. Chitons were primarily located within rock depressions and surge channels, suggesting microhabitat heterogeneity influences growth and density. Associations with species such as Corallina sp. and Mytilus californianus were consistent across all sites, though southern and northern groups showed unique proximities to varying species. These results indicate microhabitat variation affects morphological distribution within the Monterey Bay.

P131. Characterizing Gene Expression Differences in European Green Crabs (Carcinus maenas) Exposed to Thermal Extremes, Ashray Bangalore, Santa Clara University, Faculty mentor: Yaamini R. Venkataraman

European green crabs (Carcinus maenas) are a highly invasive species on the west coast of North America, spreading from California to southeast Alaska in less than 40 years. Some negative impacts of this species' presence include outcompeting native crab species, destroying eelgrass habitats, and consuming juvenile shellfish which threatens aquaculture. Their success as an invasive species can be attributed to their tolerance to a broad range of temperatures and thermal plasticity. We conducted an experiment in order to understand the impact of temperature on gene expression in C. maenas and identify molecular mechanisms that contribute to their broad thermal tolerance. Green crabs from Washington, USA were exposed to either cold (5ºC), ambient (15ºC), or warm (25ºC) temperatures for six weeks. After the six week exposure, we analyzed gene expression profiles (RNA-Seq) from heart tissues. We expected genes related to metabolic processes would be upregulated in crabs exposed to 25ºC, while similar genes would be downregulated in crabs exposed to 5ºC. Additionally, we expected that heat shock proteins (HSP) and ubiquitin-related genes would be expressed to a greater degree in the 25ºC treatment. This work will allow us to understand what enables this crab species to be a successful invader and predict future spread.

P132. Feeling the Heat: Temperature Effects on the European Green Crab, Sofia Acuña*, Alayna Cipolla*, Jasmin Mazariegos-Rodas, Emberly López-Escobar, Angelisa Abad, Wendy Escobar, Siena Fairbanks, Dominican University of California, Faculty mentor: Diara Spain

Rising atmospheric carbon dioxide associated with human activity is altering ocean conditions, contributing to both ocean acidification and increasing seawater temperatures. These changes may place stress on marine organisms, particularly invertebrates that inhabit dynamic intertidal environments. The invasive European green crab (Carcinus maenas) is known for its broad tolerance to environmental variability, making it a useful model for examining responses to thermal stress. But how resilient is this species when temperatures rise beyond its typical habitat conditions - can it survive a “crab boil”?European green crab specimens were collected from an intertidal zone site in Stinson Beach, CA. They were transported to the aquatic laboratory, then weighed and measured. Individuals were categorized by size and placed into either control or experimental treatments, with each crab housed in a separate container within larger circulating seawater tanks. The control group was maintained at approximately 13 °C, reflecting the temperature conditions at the collection site, while the experimental group was held at 23 °C to simulate elevated ocean temperatures. The experiment duration was nine weeks; specimen body mass recorded at intervals to evaluate potential growth responses to thermal stress.Although European green crabs are generally considered tolerant of temperature fluctuations, our results suggest that sustained exposure to elevated temperatures may negatively affect growth. Within the experimental group, 92% of individuals decreased in body mass, compared to approximately 54% in the control group. These findings suggest that increases in temperature may increase physiological stress, causing a reduction in body mass. As ocean temperatures continue to rise, thermal stress may negatively influence both invasive and native intertidal crab populations.

P133. How low can pH go? Responses of the European green crab, *C. maenas*, Angelisa Abad, Emberly Lopez-Escobar*, Sofia Acuna, Alayna Cipolla, Jasmin Mazariegos-Rodas, Dominican University of California, Faculty mentor: Diara Spain

The invasive European green crab, Carcinus maenas, is widely distributed along the California coast. It is a useful model species for examining how changes in seawater pH may affect the coastal environment of marine invertebrates. Lowered pH levels in seawater can influence calcium carbonate, an essential mineral for the crab exoskeleton structural maintenance. Our research examined the possible impacts of decreased environmental pH on the invasive European green crab. Crabs were collected from Stinson Beach, California, and maintained in Dominican University of California’s aquatic laboratory. The specimens were divided into experimental and control groups which were monitored over a period of nine weeks with recirculating chilled seawater. The experimental group was exposed to reduced pH levels from 6.4 to 6.9. In contrast; the control group was maintained at a pH of 8.1 to 8.2, similar to the collection site. Crabs were weighed at intervals to monitor exoskeleton body mass changes.Crabs in the experimental group

initially showed fluctuations in body mass. While some individuals gained mass and others lost mass, these changes were approximately balanced, resulting in no net difference between the experimental and control groups. These results indicate that the exoskeleton mineral composition may be altering in response to the lowered pH to maintain a relatively stable range for body mass. This invasive species appears well equipped to tolerate environmental changes, there is likely a high degree of physiological tolerance or adaptive capacity under altered pH conditions. Understanding how invasive species respond to changes in pH may help predict how coastal ecosystems could respond as seawater chemistry continues to change.

P134. Global Resort-Based Coral Reef Restoration Initiatives, Gureet Dhillon, Kano Sugawara, Dominican University of California, Faculty mentor: Vania Coelho

Coral reefs are highly diverse ecosystems that offer coastal protection, fisheries and jobs for local communities in tropical areas. Despite their global worth, they are declining greatly in large part because of bleaching events driven by climate change as well as other anthropogenic activities. As a way of responding to this global challenge, stakeholders in the tourism sector have emerged as important contributors to help coral reef health through adopting and integrating restoration projects. This paper focuses on the number of resorts involved in such activities compared to those that are not, in several different parts of the world including the Caribbean, Pacific, and Indian ocean regions. In order to collect the data for our study, we searched on Google for "coral reef restoration resort" in each country where coral reefs are found. Afterwards we looked closely at each resort's website to confirm their participation in those activities. Later we searched on Google Travel for resorts in each country for an estimate of the total number in that area. Our results show that the Caribbean region had the largest percentage of resorts engaging on coral reef restoration (about 2%), followed by Indian (0.4%) and Pacific (0.3%) oceans. In the Caribbean, Saint-Barthélemy had the highest participation (100%), and Mexico the lowest (0.2%). In the Indian Ocean, Seychelles had the highest (9%) and Sri Lanka the lowest (0.1%). Finally in the Pacific region, French Polynesia had the highest (about 36%), whereas the Philippines and Cambodia had the lowest (0.1%). These results contribute to a better understanding of the current extent of tourism-led restoration efforts highlighting the opportunity for further engagement and coordination of these activities globally in concert with the scientific community.

P135. Correlation of Nutrient Concentration and the Size and Complexity of Microbial Cells in the Upwelling System of the California Current Ecosystem, Mackenzie Eckles, California State University Monterey Bay, Faculty mentor: Lydia Baker

The growth and reproduction of primary producers at the base of the marine food web are constrained by the availability of essential nutrients. Variations in nutrient concentrations, driven by physical processes such as upwelling and water mass mixing, can shift the composition and abundance of microbial communities, influencing the structure of the broader ecosystem. Because

nutrient supply also shapes microbial physiology—affecting traits such as cell size and internal complexity—we evaluate whether nutrient concentration correlates with these cellular characteristics across areas of high, intermediate, and low upwelling influence in the California Current ecosystem. Water samples were collected during the summer 2025 MGL2506 research cruise from vertical profiles extending to 500 m depth. Samples were analyzed for nutrient concentrations and for microbial community characteristics using flow cytometry to quantify cell abundance, size, and internal complexity. We found a positive correlation between nutrient levels and both microbial cell size and complexity. In upwelling zones, microbial cells were larger and more complex due to the dominance of nutrient-demanding taxa like diatoms and other eukaryotic phytoplankton. In contrast, nutrient-poor areas were dominated by smaller, less-complex microbes such as Prochlorococcus and heterotrophic bacteria. This study demonstrates how nutrient availability influences overall cell complexity, ecosystem structure, and productivity along the California Current, highlighting the role of physical oceanographic processes in shaping the base of the marine food web.

P136. Cryopreservative Methods for Storage of Seagrass Wasting Disease pathogen, *Labyrinthula zosterae*, Varsha Vijayakartik, University of California, Davis, Faculty mentor: Jonathan Eisen

Seagrass wasting disease (SWD) results in severe, long-lasting damage to eelgrass ecosystems around the world due to its infectious and persistent pathogenicity. Developing biocontrols for SWD requires an efficient, reliable method to store the causative agent, Labyrinthula zosterae, in the laboratory for experimentation. However, optimal cryopreservation protocols for L. zosterae remain understudied, limiting experimental reproducibility and research progress. The purpose of this study was to develop reliable cryopreservation methods to maximize L. zosterae survival and growth after storage periods of one, three, and six months at -80℃. We evaluated the effects of cryoprotectant (glycerol or DMSO), thawing temperature (30℃ or 50℃), and cryopreservation additives (horse serum and trehalose) on post-thaw survival and growth to identify the most effective storage protocols. The effects of cryopreservation conditions on the average growth of L. zosterae were tested at 3 months, and survival was tested at all time points. There was no statistical difference in the growth of L. zosterae between samples cryopreserved in DMSO and in glycerol. However, DMSO-containing samples exhibited higher survival rates, supporting the use of DMSO in cryopreservation solutions to maximize the survival of L. zosterae at no detectable cost to pathogen growth. Additionally, the inclusion of horse serum resulted in increased pathogen survival at the six-month time point compared to the one-month time point. This result indicates a protective effect of horse serum over extended storage periods with respect to colony growth across all treatment conditions. Finally, the use of trehalose yielded the highest growth of L. zosterae after one and three months, demonstrating that trehalose could be used for short-term cryopreservation of the pathogen. Overall, our work provides multiple cryopreservation treatment

options for short-term and long-term pathogen storage, enabling researchers to maximize the survival and growth of L. zosterae according to specific experimental objectives.

P137. Dental Abrasion Patterns of Male California Sea Lions in Correlation to Sagittal Crest, Liana Lerona, Iman Yousaf, Dominican University of California, Faculty mentor: Doreen Gurrola

The adult male California sea lion (Zalophus californianus) has a prominent sagittal crest that supports strong jaw structure, showing a key difference compared to all California sea lion females. Although feeding habits and dental health have been associated with cranial morphology in marine mammals, it is unknown how sagittal crest growth and tooth abrasion relate to one another in male California sea lions.The purpose of this study is to determine whether sagittal crest height and tooth abrasion severity are correlated in mature males. Analysis was conducted on 247 male skulls from the California Academy of Sciences collection. Digital calipers were used to measure the skulls, including the maximum sagittal height, and a standardized three point scale was used to rate the severity of abrasions on thousands of teeth. In addition to having a higher sagittal crest height than subadults, adult males showed significantly higher levels of moderate and severe tooth abrasion. Sagittal crest height and abrasion severity were positively correlated, with a greater correlation seen in adults, according to scatterplot analysis. Based on results, it is implied that higher tooth wear is caused by increased sagittal crest development, which is probably a reflection of greater bite force and temporalis muscle attachment. The morphological relationships between tooth health and eating performance in male California sea lions are highlighted in this study.

P138. Dental Abrasion Patterns of Subadult and Adult Male California Sea lions, Ameya Gildersleeve, Katalina David, Dominican University of California, Faculty mentor: Doreen Gurrola

Dental abrasion reflects feeding behavior, age-related wear, and overall dental condition in male California Sea lions (Zalophus californians). This study examined tooth abrasion patterns in subadult and adult males using a three-point abrasion scale categorized as mild, moderate, and severe. Male skulls classified as subadult or adult were analyzed, and abrasion patterns were compared between age classes. Subadult males exhibited primarily mild abrasion, with lower levels of moderate and severe wear. In contrast, adults showed substantially greater levels of abrasion, particularly in moderate and severe categories. Overall, adults demonstrated more abrasion than subadults across all categories. These findings suggest that dental abrasion increases with age and cumulative feeding activity. The progression from predominantly mild abrasion in subadults to greater moderate and severe wear in adults highlights the impact of long-term mechanical wear on tooth condition in male California sea lions. These results contribute to understanding age-related dental changes and their ecological significance in marine mammals.

P139. Patterns of Skull and Tooth Echinochrome Staining in Southern Sea Otter (Enhydra lutris nereis), Jenny Hernandez Alvarado, Trinity Lopez Rodriguez, Dominican University of California, Faculty mentor: Doreen Gurrola

This study focused on echinochrome staining in the teeth of southern sea otter skulls (Enhydra lutris nereis) and the correlation between tooth and skull staining. Consumption of purple sea urchins (Strongylocentrotus purpuratus) leads to echinocrome on sea otter skull and teeth, which are a purple-red pigment. Data for this study was collected from the California Academy of Sciences, which has the largest collection of southern sea otter skulls. A total of 372 skulls and over 10,000 teeth were examined. Adults and subadults were compared including males and females. Results showed that most adult skulls (59.8%) had moderate staining with only a small percent (9.2%) showing heavy staining. Subadults showed a higher correlation ($r = 0.598629$) which suggests that staining patterns may be more connected in young otters. Overall, echinochrome staining varies by age and sex and further research is necessary to better understand how sea urchin consumption may affect skull staining and dental wear.

P140. Patterns of invertebrate recruitment associated with kelp forest restoration, Lucas Kadeg, Sonoma State University, Faculty mentor: Brent Hughes

With the decline of the world's major coastal habitats: coral reefs, seagrass beds, and kelp forests, comes the loss of critical ecosystem functions. Kelp forests, such as the bull kelp (Nereocystis luetkeana, hereafter Nereocystis), can act as a foundation species that enhances biodiversity. Since 2014, ~90% of Nereocystis forests have been lost in northern California, and the loss of these kelp forests could have catastrophic impacts on local biodiversity and ecosystem function, such as invertebrate recruitment. Here we describe the recruitment patterns of invertebrates during kelp forest restoration. This was conducted via monitoring recruitment in 2024 and 2025 at two sites (Fort Ross and Timber Cove in Sonoma County, CA) where Nereocystis has been actively restored through reseeding methods. Specimens were obtained using two types of artificial recruitment collectors (ARCs): Plastic brushes and scouring pads, referred to as Tuffys ($N= 9$ of each ARC type at each site). ARCs were selected due to their ability to mimic habitats of newly settled invertebrates. ARCs were swapped out monthly between late spring and early fall. After collecting from the field, ARCs were collected, frozen, and later analyzed at Sonoma State University via dissecting microscope. The invertebrate groups most notably collected in ARCs were clams (especially Hiatella sp.), limpets, mussels, scallops, and other bivalves, snails, urchins, crabs and barnacles; demonstrating high diversity and noteworthy interannual variability in recruitment. Preliminary analysis suggests the potential of greater kelp restoration success yielding differences in recruitment. These functions are important to study as they show how restoring foundation species could restore lost ecosystem functions. These results further reveal the positive relationship of kelp presence to invertebrate recruitment and diversity. This study emphasizes the need to

understand kelp forest functions and the maintenance of biodiversity in ecosystems targeted for restoration.

P141. Waste-to-Resource Valorization of Cladophora Blooms: A Circular Bioeconomy Approach for Soil Amendment and Crop Growth Enhancement, Vanessa Magana, California State University, Stanislaus, Faculty mentor: Alok Arun

Excessive growth of the filamentous green algae Cladophora has become a recurring environmental issue in freshwater systems throughout California's Central Valley. These nuisance algal blooms produce large amounts of biomass that can disrupt aquatic ecosystems and interfere with water management. Although Cladophora blooms are not toxic, the accumulated algae are often treated as waste and removed from waterways. This study explores whether this abundant biomass can be repurposed as a sustainable soil amendment and plant bio-stimulant for crops commonly grown in the region, particularly tomatoes. To test this idea, tomato and lettuce seeds were grown in soils amended with six different concentrations of a Cladophora biomass solution (0.05%, 0.1%, 0.25%, 0.50%, 1.0%, and 10.0%) and compared to untreated control soils. Plant growth was evaluated by measuring germination percentage, root and shoot length, and the seedling vigor index (SVI), which combines germination rate and early seedling growth. Lettuce seedlings grown in amended soils showed improved growth in several treatments compared to the control (SVI = 29.00). The highest vigor was observed at moderate concentrations, especially 0.25% (SVI = 47.33) and 1.0% (SVI = 46.73). Tomato seeds also responded positively to the treatments, with germination increasing from 50% in the control to as high as 85% at the 0.25% concentration. These preliminary results suggest that biomass from Cladophora blooms may improve early plant growth when used as a soil amendment. Our future work will focus on measuring the biomass and other physiological parameters of the treated plants and analyzing the secondary metabolite profile of Cladophora using FTIR to better understand which compounds may be responsible for promoting plant growth

P142. Nest site selection in Northwestern pond turtles (Actinemys marmorata) on a managed livestock rangeland, Mickenna Edy, Sonoma State University, Faculty mentor: Nicholas Geist

The Northwestern pond turtle (Actinemys marmorata) is one of only two of California's native freshwater turtle species, both of which are facing dramatic population declines. Habitat loss, particularly due to agricultural and urban development, as well as the increase in non-native turtles and predator species exacerbates their rapid decline. Despite their threatened status, there is still more to learn regarding their nesting ecology in order to better inform future conservation efforts. We conducted field observations on a population of A. marmorata living in a stock pond on a working cattle ranch in Alameda County, California. Gravid females were tracked both visually and with radio telemetry during nesting season, allowing us to successfully record GPS coordinates as well as additional nest site characteristics for all nesting females. This ongoing,

multi-year study in collaboration with a variety of organizations within the Bay Area will provide valuable information regarding A. marmorata nesting behavior, nest site fidelity, and reproductive success amidst shifting climate patterns leading to better informed management practices.

P143. Development of a 3D-Printed Clinostat for Investigating Microalgal Nutritional Profiles Under Simulated Microgravity, Allison Shewfelt, Imaan Heer, California State University, Stanislaus, Faculty mentor: Alok Arun

Development of a 3D-Printed Clinostat for Investigating Microalgal Nutritional Profiles Under Simulated Microgravity Microgravity can affect microbial growth, metabolism, and nutrient production, which is important to understand for long-term space missions and space-based food production systems. Microalgae are promising organisms for space applications because they can produce oxygen, fix carbon dioxide, and generate nutrients that could support life-support systems. However, conducting experiments in real microgravity environments is very expensive and limited to specialized facilities. To address this challenge, this project focuses on developing a low-cost device that can simulate microgravity conditions in a laboratory.In this study, we developed a 3D-printed clinostat that mimics microgravity by continuously rotating culture samples to randomize the gravitational vector. The design was based on open-source resources but was modified to better fit the needs of cyanobacterial experiments. The device was designed to be compact, affordable, and compatible with standard laboratory culture vessels. Currently, we are testing and integrating sensors into the clinostat to monitor factors such as rotation stability and environmental conditions. These sensors will help ensure that experiments remain consistent and reliable during operation.The next phase of the project will involve growing microalgal cultures using the clinostat under simulated microgravity conditions and comparing them to control cultures grown under normal gravity. Growth performance and nutritional characteristics will then be measured, including biomass production and key nutritional components such as proteins, lipids, and pigments. Overall, this project aims to create an affordable and accessible tool for microgravity research while helping us better understand how microalgae may be used as a sustainable biological resource for future space exploration.

Microbiology #144-155

P144. An Exploration of Mutational Hotspots in Escherichia coli Caused by the Action of a Nucleoid-Associated Protein, HNS, Natasha Sutkin, Alina Garmash, University of California, Los Angeles, Faculty mentor: Jeffrey Miller

Numerous cancers and diseases arise from mutations during DNA replication, yet little is known about the effects of DNA conformation on the formation of mutational hotspots. Mutational hotspots are defined as sites that are much more mutable than others. While hotspot formation can be attributed to favored base pair changes and nearest neighbor sequences, even with identical base pair changes and nearest neighbors, some sites are still more mutable than others. The mutagenic effects of the chemotherapeutic drug Cisplatin (CPT) serve as an example. CPT induces DNA cross-link adducts at neighboring base pair positions, favoring GC → TA and AT → TA transversion mutations. In the rpoB gene of Escherichia coli (E. coli), cisplatin-induced Rifampin resistance (Rif^r) results in 5'-CAG-3' → 5'-CTG-3 mutations at sites 443 and 1538. Despite the identical base pair substitutions and nearest neighbors, we detected 26 occurrences at site 1538 and zero occurrences at site 443, even though both substitutions result in a mutant phenotype. In a second system, we developed mutation-induced inactivation of the thymidine deoxykinase-encoding gene (tdk) through Azidothymidine (AZT) selection. In tdk, CPT induced a dramatic GC → TA hotspot at position 499. Here we show that mutational hotspots are influenced by the conformation of the local DNA, as bacterial DNA is highly folded, supercoiled, and compacted by specific DNA binding proteins, termed Nucleoid-Associated Proteins (NAPs). In investigating the NAPs Fis, HNS, IHF, and HUα, we found that an HNS deficiency results in the disappearance of the dramatic CPT-induced hotspot. However, this hotspot was unaffected by an HUα deficiency. This is the first demonstration that NAPs, and thus DNA compaction, can drive the formation of major mutational hotspots. Future work aims to examine hotspots induced by additional mutagens with an overarching goal of uncovering the fundamental mechanisms behind the pathways that lead to mutation formation.

P145. Analysis of Magnolia Leaf Extract Silver Nanoparticle Aggregation and its Effects on Bacterial Inhibition, Cameron Scott, David Fernando, The Master's University, Faculty mentor: Haley Smith

The antimicrobial effect of silver nanoparticles (AgNPs) has been long established and well characterized. Current literature demonstrates their effectiveness at inhibiting a wide variety of organisms including bacteria, viruses, fungi, protists, and some studies even show effectiveness against a variety of cancer cell lines. Green synthesis of silver nanoparticles represents a viable method for generating AgNPs in a highly cost effective and efficient manner leading to renewed interest in their use as an antibiotic-independent antimicrobial treatments. The use of Magnolia alba leaf extract in green synthesis produces nanoparticles (MAgNPs) that are highly effective

against many bacterial strains, bacteriophages, and fungi. One of the attributes of AgNPs is that the particles tend to aggregate over time, which has been theorized to cause worsening inhibition as aggregation of the particles increases. Characterization of this aggregation and nanoparticle dynamics is critical to understand if the MAgNPs are going to be used for various treatments. This study investigates these dynamics under different biorelevant conditions using UV-Vis-spectroscopy and scanning electron microscopy (SEM) and demonstrates the effect of aggregation on bacterial inhibition. Results show that over the course of 24 hours aggregation of MAgNPs does occur under biorelevant conditions and this is supported by a shift in the peak absorbance of the nanoparticles over time. This was tested across a range of physiologically relevant sodium ion concentrations and results show that the rate of aggregation increases as sodium concentration increases. This demonstrates a further need to characterize MAgNPs under conditions in which they might used if they are to be a viable antimicrobial alternative.

P146. Associations between the human breast microbiome and immune genes in breast cancer, Luke Pontoriero, Joan Argwings-kodhek, Pepperdine University, Faculty mentor: Leah Stiemsma

Breast cancer (BC) affects 1 in 8 women and roughly 50% of breast cancers cannot be explained by environmental or genetic factors. There is growing evidence of the role of the human breast microbiome in breast cancer, but the mechanisms guiding this role are not well understood. The co-evolution of the human microbiome with the immune system represents an interaction between host and microbiome that is understudied with regard to breast cancer pathogenesis. We hypothesized that immune genetic variation is associated with altered abundance patterns of bacterial taxa in BC. We conducted 30X whole genome sequencing on a sample of 45 women who donated 60 tissues, grouped into four distinct tissue types: healthy (H), pre-diagnostic (PD), tumor (T), and adjacent normal (AN), to the Susan G. Komen and Indiana University Simon Comprehensive Cancer Center Tissue Banks. We filtered variants for inclusion in our analysis by quality score, variant impact, relation to immune function, and association with the PD, AN, and T tissues (precancerous and cancerous tissues). We used MaAsLin2 to associate alternate allele frequencies (AAF) and genotypes with amplicon sequence variant (ASV) abundances from breast bacteria previously identified as associated with BC (Hoskinson et al. mSystems, 2022). 13 variants were significantly associated with ASV abundances. The top 4 variants with the highest number of ASV associations with either AAF or genotype were AKR1C3 (genotype: 9 ASVs, AAF: 8 ASVs), ARG1 (genotype: 8 ASVs, AAF: 2 ASVs), CFHR3 (genotype: 13 ASVs, AAF: 4 ASVs) and PRAM1 (genotype: 27 ASVs, AAF: 16 ASVs). Pseudomonas ASVs were the top ASVs associated with these genes (24 total associations). These results suggest interaction of specific breast bacterial taxa with immune variants in BC. We aim to analyze the functional metagenome in association with immune variants to inform how these taxa may influence host immune variation.

P147. Influence of Specific Mutations on Viral Thermostability in Bacteriophage Phi-6 Under Heat Stress, Erika Ebreo, San José State University, Faculty mentor:

As antibiotic resistance continues to limit treatment options, alternative approaches that rely on bacteriophages, viruses that infect and kill bacteria, have gained increasing attention. The ability of bacteriophages to withstand environmental stress is essential for successful infection. Previous work has shown that the bacteriophage phi-6 can improve its survival when evolved with high temperature heat shocks. I engineered mutations found in P5, the lysis protein, to determine how genetic changes alter phi-6 thermal stability. I mutagenized phi-6 and examined how viral thermal tolerance changed across increasing temperatures. Three mutations at amino acid sites 201-207 of P5 increased thermal stability by 1.5-2.5 degrees Celsius. As ongoing work, I am also testing a synonymous mutation and a mutation at the end of the P5 protein (amino acid site 221). I hypothesize that a synonymous mutation does not significantly affect viral thermostability as the amino acid sequence remains unchanged. In contrast, I hypothesize that the amino acid substitution mutation at amino acid site 221 of the P5 protein will significantly increase thermostability through its altering of structure of the P5 protein. Knowing the effects of these mutations will be important for engineering stable bacteriophages that can be used in phage therapy.

P148. Ongoing Biochemical Investigation of Ozonated Black Cumin Seed Oil, Franchesca Maldonado, Mount Saint Mary's University Los Angeles, Faculty mentor: Sylvine Deprèle

Growing interest in the medicinal benefits of ozonated natural oils due to their antibacterial properties has led to increased accessibility and commercialization of these products, particularly as topical treatments. This research continues to investigate the ozonation of black cumin seed oil (BCSO) while focusing on observing the enhancement of its natural antibacterial properties through the reaction of ozone with unsaturated fatty acids present in the oil. Utilizing IR spectroscopy (IR), the point of partial and full ozonation of BCSO was determined to be respectively 40 minutes and 2 hours of ozone exposure, with expected changes in the IR stretches of Csp2-H at 3020 cm-1, Csp3-H at 1375 cm-1, and the C-O at 1200 cm-1. The high stability of the three forms of BCSO was found when tested against UV and heat exposure for a period of 55 and 72 hours, respectively. There were no significant chemical changes observed from IR data collected over the period of exposure for both stressors and only minimal physical changes observed in the heat-exposed oils. To study the antibacterial properties of the various ozonated forms of BCSO, initial trials were designed to resolve the issue of insolubility of BCSO in LB broth. Utilizing Tween-80 surfactant and an ultra sonicator, neat and partially ozonated forms of BCSO successfully formed homogenized emulsions in LB broth that were subsequently utilized in preliminary bacterial inhibition screening completed in broth culture. Results indicated promising signs of inhibition against gram-negative bacteria, Pectobacterium carotovorum. Bacterial inhibition for the neat, partially, and fully ozonated BCSO was further analyzed using a modified disc diffusion method to screen against other gram-negative bacteria, showing significant

inhibition against Acinetobacter baylyi. Future directions include optimizing the emulsification procedure to form homogenized solutions to be used in minimum inhibitory concentration (MIC) assays against various bacteria.

P149. The Antimicrobial Potential of Ozonated Pumpkin Seed Oil, Natalie Garcia Quezada, Mount Saint Mary's University Los Angeles, Faculty mentor: Sylvine Deprèle

The continuous treatment of bacterial infections with drugs has contributed significantly to microbial resistance to antibiotics. This concern is ongoing as bacteria possess innate defense mechanisms and can reduce toxic oxygen species using specific enzymes; ozone has been used in dermatological experiments to either inhibit or kill microbial colonies. The purpose of this research is to enhance the holistic effects of pumpkin seed oil (PSO) through ozone infusion and develop a topical treatment for various skin ailments that can be infiltrated by opportunistic pathogens. Pumpkin seed oil is an ideal candidate for ozonation as it is known for its antibacterial and anti-inflammatory properties as well as its richness in vitamins A and E. Throughout ozonation, the chemical composition of PSO is tracked using Infrared Spectroscopy (IR), specifically the functional groups at 3020 cm-1, 1375 cm-1, and 1200 cm-1 which indicate change in unsaturation within the oil. PSO reaches partial ozonation (pPSO3) at 30 minutes and full ozonation (PSO3) at 65 minutes. To determine the stability of each oil form, stress tests were performed with heat (250 watts) for 72 hours and UV (245 nm) for 56 hours, during which IR was used to evaluate chemical changes. The high stability demonstrated by each oil provided a segue for biological testing, which began with the formation of microemulsions containing PSO or pPSO3, Tween 80 surfactant, and Luria-Bertani broth. Once developed, turbidity of the broth was assessed to determine the inhibitory effects of PSO and pPSO3 emulsions against Pectobacterium carotovora, during which pPSO3 displayed greater inhibition when compared to PSO. To confirm antimicrobial properties, disc diffusion was used to qualitatively evaluate the inhibition zones formed by PSO3 against Pectobacterium carotovora and Staphylococcus aureus. Future experiments include the optimization and formulation of PSO3 microemulsions as well as further testing against ESKAPE relative pathogens.

P150. Transcription-coupled repair factor Mfd modulates biofilm production in Bacillus subtilis, Carmina Chavez, University of Nevada, Las Vegas, Faculty mentor: Eduardo Robleto

Bacillus subtilis is a soil-dwelling bacterium that forms complex, three-dimensional biofilms. These biofilms confer an evolutionary advantage, enabling microbial communities to optimize resource use and withstand environmental stressors. B. subtilis biofilms are particularly important due to their symbiotic relationship with plants and potential roles in bioremediation of industrial contamination. Within a biofilm, B. subtilis cells can differentiate into matrix producers, motile cells, competent cells, or spore-forming cells. Because cellular differentiation depends on transcriptional regulation, we investigated the role of Mfd, a conserved transcription-coupled DNA

repair factor, in biofilm development and population heterogeneity. Recent findings from our lab suggest that Mfd contributes to the regulation of biofilm-associated genes. We hypothesize that Mfd promotes colony complexity and phenotypic variation by regulating biofilm gene expression under stress. To explore this, we compared the wild-type strain NCIB 3610 to an Mfd-deficient mutant by analyzing colony morphology and conducting a 20-day serial passage to assess the long-term evolution of the population. Prior studies indicate that B. subtilis populations can evolve distinct colony morphotypes, which may reflect changes in biofilm gene expression and function. Preliminary data from the Δmfd mutant revealed altered colony architecture and reduced structural complexity, suggesting a role for Mfd in biofilm formation. These results support a role for Mfd in regulating biofilm associated gene expression and promoting phenotypic heterogeneity within B. subtilis. Understanding the genetic mechanisms that influence biofilm structure and deepen our insight into bacterial physiology has potential applications in agriculture and bioremediation.

P151. Uncovering Efflux Regulators that Control the Expression of EmrAB-TolC, Takoda Lakpour, California Lutheran University, Faculty mentor: Dana Harmon

The EmrAB-TolC efflux pump found in Escherichia coli is involved in the removal of toxic molecules and cellular metabolites. Transcription of emrA and emrB is controlled by EmrR, and is cotranscribed with emrA and emrB in the emrRAB operon. It is not known if transcriptional regulators involved in regulating the expression of other efflux pumps also control the expression of emrRAB. We investigated four regulators – SoxR, SoxS, Rob, and AcrS – for their role in controlling emrRAB expression. These regulators also control the expression of acrAB, the main efflux pump in E. coli. The goals of this study were to characterize the emrRAB regulatory network and determine if there is overlap with acrAB. Using an emrRAB promoter gfp fusion construct (emrRABp-gfp), we have measured the expression of emrRAB in 4 different mutant backgrounds. To this end, utilizing strains from the KEIO collection, a library of E. coli strains containing deletion mutants of these regulators, replaced by a kanamycin resistance cassette. After removing the kanR cassette, we have transformed our markerless mutants with a plasmid containing the emrRAB-gfp fusion. In addition to the soxRS, rob, and acrS, we have also investigated additional strains deleted for other genes known to regulate acrAB or emrRAB expression. Overall, the data did not reveal statistically significant differences in expression in our mutant backgrounds. However, while the anticipated effects were not observed, the outcomes reflect a need for methodological refinement in future experiments.

P152. UV mutants of Ustilago maydis: Role of Nitrogen Source in Filamentation, Hadley Jones, University of Louisville, Faculty mentor: Michael Perlin

Ustilago maydis (UM), a smut fungus, presents a unique threat to maize crops. Wildtype UM exhibits dimorphic growth behavior, i.e., yeast-like budding when in a haploid sporidial state, and upon mating, it switches to a filamentous dikaryotic hyphal form. Filamentation is thought to be

key to the infectious patterns and tumor formation associated with UM. Interestingly, haploid cells have been found to exhibit filamentation in nitrogen-poor environments. The ump2 gene of UM is required for such filamentation in response to low ammonium, since its disruption, in haploid strains (e.g., FB1) or solopathogenic haploid strains (SG200), eliminates this filamentous response. Second site mutants that rescued the filamentation response of ump2 disruption strains were identified in cells exposed to UV light. It is also well understood that UM has multiple pathways of source-dependent nitrogen utilization. To determine whether the filamentation response is limited to ammonium availability, here, we investigate the 3 respective UV mutated SG200 and FB1 strains and their filamentation patterns when exposed to high and low concentrations of different nitrogen sources, including: ammonium sulfate, ammonium nitrate, potassium nitrate, sodium nitrite, gamma-aminobutyric acid, beta-alanine, as well as an amino acid profile both coupled and uncoupled with ammonium sulfate. Subsequent comparison to wildtype and non-UV exposed cells in the same nitrogen sources aims to elucidate both the relationship between the mutated genes that rescued the ump2 deletion phenotype, and, more generally, nitrogen utilization pathways in UM.

P153. Implementation and Optimization of CRISPR-Cas9 gene editing in Methylobacterium extorquens AM1, Vihari Kotipalli, Ruth Casab, San Jose State University, Faculty mentor: Elizabeth Skovran

Methylobacterium extorquens AM1 is a GC-rich methylotrophic bacterium that can recover rare earth elements (REEs) from electronic waste. Our goal is to genetically engineer M. extorquens for improved REE recovery as a biorecycling platform. We chose to implement CRISPR-Cas9 gene editing to rapidly generate desired mutations as alternative methods take longer to create genetic changes. We synthesized codon-optimized Cas9 variants and constructed tunable Cas9 and guide RNA expression systems. Cas9 expression levels can be induced by adding cumate to the growth medium, while guide RNA levels are upregulated using methanol and downregulated using lanthanum. In this study, we developed an optimal gene editing protocol by altering Cas9 and guide RNA ratios. To evaluate CRISPR-Cas9 editing efficiency, we designed a phenotypic screen that utilizes the organism's methanol oxidation pathway. While both edited and unedited strains grow on non-selective media, those with the desired mutation survive under selective pressure. By first upregulating guide RNA expression using methanol vapors, then downregulating expression using lanthanum, we achieved a 60% editing efficiency. Work in progress suggests efficiency can be further improved by using guide RNA sequences that bind closer to the site of the desired mutation. It is possible that adding methanol vapors acted as a selective pressure that enriched for edited cells, thereby inflating the efficiency of our system. Future studies will examine editing efficiency when using different lanthanum concentrations and screen for mutations that are not affected by the selective pressure from methanol vapors. We will also identify whether the use of a nickase-Cas9 variant, which makes single-stranded instead of double-stranded DNA breaks, impacts gene editing efficiency. Successful implementation of CRISPR-Cas9 gene editing will

facilitate our ability to engineer mutations for M. extorquens AM1 to improve our REE biorecycling platform.

P154. Investigating How Genetic Variation in Malaria Vaccine Candidate PfRh5 Alters Immune Evasion, Elon Atlaw, Yale University, Faculty mentor: Amy Bei

Malaria is one of the leading causes of death globally and continues to pose a major threat as eradication efforts have stalled in recent years. Vaccine development offers another control strategy, with current sporozoite-stage vaccines showing promise, but their efficacy is challenged by the parasite's complex life cycle and genetic diversity, highlighting the need for next-generation vaccines targeting other stages of the life cycle. The RH5 protein in P. falciparum (PfRh5) is a promising target for a blood-stage vaccine due to its conservation, its essential role in the PCRCR complex that binds human erythrocyte receptor, Basigin (BSG), and its susceptibility to vaccine-induced human monoclonal antibodies. A recent phase 2b trial showed that the RH5.1 vaccine had the highest efficacy of any blood-stage vaccine tested to date, but the impact of genetic variations in RH5 on immune evasion and vaccine efficacy is currently unclear. We propose to study the effect of single-nucleotide polymorphisms (SNPs) in PfRh5—D243N, D249G, and F505Y—found in endemic regions of Senegal, on immune response, invasion, and antibody binding. To this end, I first aim to assess the functional roles of PfRh5 variants on immune neutralization using CRISPR-Cas9 gene editing of P. falciparum parasites. My second aim is to identify genetic polymorphisms in PfRh5 in culture adapted parasite lines from Thiès, Senegal. After identifying SNPs, I will investigate the neutralization ability of antibodies isolated from vaccines against Thiès isolates through growth inhibition activity assays. Our objective is to investigate the role of selected PfRh5 genetic variants, in immune evasion both through a cellular and biochemical angle. Preliminary sequencing of P. falciparum isolates from Thiès identified four PfRh5 SNPs, including variants previously reported in highly endemic regions, supporting the presence of genetically diverse RH5 alleles in this population. In parallel, dual-vector CRISPR-Cas9 constructs targeting PfRh5 have been successfully generated and transfected into parasites, establishing the experimental platform needed to evaluate how these variants influence antibody neutralization and erythrocyte invasion. Overall, this project investigates genetic variation in PfRh5 to better understand its role in immune evasion and to inform the design of next-generation blood-stage malaria vaccines

P155. The Effect of Peripartum Antibiotic Treatment on Maternal Vaccination Conferred Protection against Neonatal Pertussis, Mythili Thota, University of Georgia, Faculty mentor: Eric Harvill

Bordetella pertussis (Bp), the main bacterial agent of whooping cough, is a highly contagious respiratory pathogen that poses considerable risk to neonatal health prior to routine immunizations. Maternal pertussis immunization during the third term of pregnancy allows Bp-specific antibodies

to be transferred transplacentally and through breastfeeding, which serves as a crucial mechanism of neonatal immunity against Bp. However, the maternal use of antibiotics during pregnancy has been shown to impact the level of antibodies transferred from mother to infant. While we know that maternal antibody transfer provides significant protection to newborns against severe disease, the impact of maternal antibiotic treatment before birth on the transfer of protection remains unclear. Here, we will show that peripartum maternal antibiotic treatment disrupts maternal vaccination conferred protection to neonates against Bp by decreasing maternal antibody presence. To assess, dams will be left untreated or be given a dose of cefazolin, gentamicin, or penicillin, intranasally, 3 days pre-parturition. Resulting pups will be challenged intranasally with 1×10^4 CFU/15 μL of Bp at 5 days old and then assessed 3 days post bacteria challenge, to evaluate transferred protection. Protection is measured through quantifying the bacterial burden in the lungs of the pups, and measuring Bp-specific antibodies in the serum, milk spots, and lungs of the pups. This study will assess the efficacy of maternal vaccination conferred protection when clinically relevant peripartum antibiotic regimens are administered. Understanding how maternal interventions influence the amount of maternal antibody transferred is essential for optimizing strategies to protect newborns against Bp.

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P156. Determining the role of SOX9 in neural crest development, Veda Jadaprolu, University of California, Davis, Faculty mentor: Crystal Rogers

Neural crest (NC) cells are transient multipotent stem cells that form in the developing neural tube. They undergo an epithelial-to-mesenchymal transition (EMT) which allows them to migrate through the embryo and differentiate into more than 30 different tissue derivatives. NC cells form the peripheral nervous system and craniofacial bone and cartilage. The NC gene regulatory network (GRN), which is a hierarchical map of the proteins essential for NC cell development, migration and differentiation, but the NC GRN remains incomplete. To expand our understanding of the molecular cues driving NC development, we characterized the expression and function of SOX9, a marker of definitive NC cells. SOX9 is a transcription factor necessary for the formation of craniofacial bone and cartilage. Using the chicken embryo (Gallus gallus) as our model, we have uncovered the relationship between SOX9 and other NC markers using knockdown experiments followed by in situ hybridization chain reactions (HCR) and immunohistochemistry (IHC) to study the effects of SOX9 knockdown on SNAI2 and PAX7 gene and protein expression across different developmental stages. We identified that SOX9 is necessary for SNAI2, but not PAX7 expression and the progression of NC development. Our work will help uncover the role of SOX9 in NC EMT, providing insight into conserved molecular mechanisms that underlie both embryonic development and pathological EMT processes such as cancer metastasis.

P157. Effect of Serotonin Receptor Modulation by Psilocin on Cardiac Neural Crest Derivatives and Embryonic Fasciculation, Mandoline Nguyen, Sarmad A. Alani, Loyola Marymount University, Faculty mentor: Maxellende Ezin

Psilocybin has been recently designated as breakthrough therapy by the U.S. Food and Drug Administration (FDA) for treatment-resistant depression. Psilocybin, the psychedelic agent of Psilocybe mushrooms, is metabolized into the active congener, psilocin, in the liver. However, the impact of psilocin on embryonic and fetal development is unknown. Psilocin broadly interacts with the serotonergic system as an inverse agonist of serotonin receptor 2B (5-HT2B) and as an agonist of 5-HT1A, 2A, 2C. In addition to regulating physiological functions in the adult, serotonin regulates embryonic development. Serotonin regulates migration and differentiation of embryonic neural crest cells (NCCs). These cells give rise to critical structures in embryos, such as the cardiac valves. Therefore, we aim to study the effect of disrupting serotonergic receptors on the developing embryo using exposure to psilocin. Methods: Chicken embryos were treated with psilocin doses of 10 uM, 20 uM, or 100 uM in ovo at Day 1 (equivalent to 20 days of human fetal development) before the heart has formed. These embryos were then collected at Day 10 (58 days of human fetal

development), when the heart has formed. During collection, Day 10 embryos were filmed in ovo to assess embryonic movement known as fasciculation. Once collected, these embryos were cryosectioned to visualize the structures of the heart. Morphometric analysis of fasciculation and cardiac valves was performed using FIJI. Preliminary Results: Embryos treated with psilocin exhibit increased rates of muscular fasciculation, with limbs moving longer distances relative to controls. Additionally, psilocin-treated embryos present with external phenotypes that indicate heart failure, such as hemorrhage and edema. Lastly, treated embryos formed abnormal cardiac valves, such as tissue breaks and gaps in the tips of tricuspid valve leaflets. Conclusion: Because NCCs also contribute to the craniofacial bones, we also aim to study the unknown effect of psilocin on craniofacial bone development by staining the bones and cartilage of the head.

P158. Assessing the Role of COUP and ETS in PCV Identity, Audrey Combs, Devashree Hemant Agarwal, San Jose State University, Faculty mentor: Theresa Dinh

Cancerous tumors hijack the body's immune response and often grow near capillaries to exploit nutrient supply, yet immune cell trafficking into tumors remains limited. A key barrier lies in the identity of endothelial cells (ECs) in post-capillary venules (PCVs), which normally control immune cell entry via adhesion molecules such as ICAM-1 and EphB4. However, there is a knowledge gap concerning the transcriptional programs that specify PCV identity and regulate addressin expression. Prior work shows that COUP-TFII (NR2F2), a venous master regulator, promotes high endothelial cell (HEC) identity in lymphoid organs, but transcriptional control of non-lymphoid PCVs is unclear. ETS family transcription factors govern EC identity and angiogenesis, making them strong candidates to form COUP-TFII-ETS heterodimers that regulate PCV-specific genes. Using the UCSC Genome Browser, we identified evolutionarily conserved COUP-TFII: ETS binding sites in the promoter regions of PCV- and addressin-related genes. By electrophoretic mobility shift assay (EMSA), we demonstrate that COUP-TFII and ETS proteins bind the ICAM-1 and EphB4 promoters. Thus, the Dinh lab hypothesizes that COUP-TFII and ETS form heterodimers that drive PCV-selective gene expression and promote vascular segmentation. This work will clarify how endothelial cells are specialized in vivo, with direct relevance to immune-related diseases such as cancer, autoimmune disorders, and endometriosis.

P159. Fission vs Regeneration: Distinct Differentiation Dynamics in an Acoel, Keana Ravello, University of San Francisco, Faculty mentor: James Sikes

Asexual reproduction by fission requires tightly coordinated cell differentiation and axis repatterning to generate clonal progeny from a single organism. Although regeneration and fission have similar outcomes, it is unclear whether tissue specification and differentiation of functional cell types follow the same spatiotemporal pattern. The acoel flatworm Convolutriloba longifissura robustly regenerates post-injury and utilizes a distinctive double fission process to asexually reproduce. Since regeneration and fission require respecification of both head-tail and left-right

body axes, C. longifissura is a powerful model for comparatively studying these postembryonic developmental processes. Here, we characterize the spatial and temporal dynamics of differentiation during both fission and regeneration in C. longifissura. Using fluorescently conjugated lectins to label specific carbohydrate motifs, we track the emergence and patterning of previously undescribed epidermal secretory and adhesive cell populations. Cell type differentiation during fission occurs in a spatially restricted manner that progresses with the advancing fission plane, whereas during regeneration the same cell types differentiate more slowly and appear synchronously. These findings provide new insight into how coordinated cell-type differentiation occurs during alternative post embryonic developmental trajectories in acoels.

P160. Investigating the Role of Integrin α6 in Small Intestinal Crypt Morphogenesis, Nicole Ahsan, Yale University, Faculty mentor: Kaelyn Sumigray

The epithelium of the mammalian small intestine undergoes rapid turnover, regulated by intestinal stem cells (ISCs). ISCs are housed at the bottom of crypts, cup-like invaginations from the luminal surface of the small intestine. Crypts begin forming in mice at postnatal day 0 (P0) and become fully compartmentalized from villi by P10 through the invagination of intervillous regions into the stroma. We previously found that fibroblasts are essential for proper crypt invagination during postnatal development. Fibroblasts are the main producers and regulators of the extracellular matrix (ECM), which provides structural support and external stimuli that regulate stem cell behavior and fate. There is minimal literature covering the ECM's role in supporting crypt morphogenesis. Integrin a6, a laminin receptor, is a key regulator of these interactions as it anchors epithelial cells to the basement membrane and transduces signals to regulate cell proliferation, migration, and differentiation. We hypothesize that fibroblast-dependent ECM remodeling regulates crypt morphogenesis, in part through epithelial-ECM interactions mediated by integrin a6. Through immunofluorescence staining of vibratome-sectioned tissues from wild-type mice and postnatally fibroblast-ablated mice at P2, P5 and P10, we found that fibroblast ablation resulted in reduced integrin a6 intensity at P2 and P10. Understanding how fibroblasts regulate integrin and ECM interactions in forming intestinal architecture may inform therapeutic strategies for diseases involving ECM dysregulation, including cancer. Future studies will investigate how cell differentiation and migration are affected by fibroblast ablation.

P161. Tissue-specific labeling of Septin12 with split-fluorescent proteins, Emily Le, University of California, Merced, Faculty mentor: Stephanie Woo

Septin12 is an understudied protein in zebrafish endodermal development, and we aim to visualize its role using a split-fluorescent protein tagging system. Septin12 is part of a family of cytoskeletal proteins called Septin. This protein is relatively highly expressed from 5-9 hours post-fertilization in sox32-positive cells (sox32 is a marker for the zebrafish endoderm). During this time, endoderm cells are highly migratory. Septin12 levels then drop after 9hpf, a time when endoderm cells

migrate less and take on more epithelial characteristics. We believe that Septin12 may have a role in endodermal cell migration, and visualizing how it is dynamically localized in the cell is important in probing this role. We will do this using a split-fluorescent protein approach. This newer method enables the tissue-specific visualization of broadly expressed proteins at their endogenous levels. Research on this new method would allow for newer and more efficient methods of protein labeling, and this could benefit developmental biology and biomedical research. My lab focuses on the endoderm layer of the zebrafish, which eventually forms the gut, liver, pancreas, etc. However, the endodermal cells are smaller in number, and they are underneath other layers, which makes it more difficult to visualize proteins in this endoderm layer. To address this, we investigate the split-fluorescent protein tagging system, and my research project this semester will focus on a gene knock-in and visualizing this using a split-fluorescent protein system. It will also provide more insight into live cell imaging.

P162. Variation in Expression of Ecdysone-Related Genes Throughout Development of *Gryllus lineaticeps*, Avery Garcia, University of California, Berkeley, Faculty mentor: Caroline Williams

Determined jointly by environmental and genetic factors, wing polymorphisms are a widespread physiological phenomenon in insects, with different wing morphs often specializing for different life-history strategies. In the California field cricket, Gryllus lineaticeps, wing dimorphism reflects a trade-off between dispersal and reproduction. Juveniles enter adulthood either as sexually immature, flight-capable long-wing morphs or as sexually mature, flightless, short-wing morphs. While preliminary studies have suggested that morph determination may be under neuroendocrine control, little is known about the specific genetic regulation of these hormones guiding juvenile development. The hormone ecdysone is a strong candidate for influencing morph determination given its implication in tissue-remodeling events and regulation of ecdysis (molting). In order to clarify the potential correlation between the expression of ecdysone-related genes and wing morph determination, I characterized gene expression timelines for eight ecdysone biosynthesis and receptor genes in head and wing bud samples taken from both long-wing- and short-wing-destined morphs of Gryllus lineaticeps across the last juvenile stage, when wing morph is determined. Gene expression analysis demonstrated that four ecdysone biosynthesis and signaling genes (shade, phantom, spook, ECR) are upregulated in both long-wing- and short-wing-destined morphs, typically peaking either on the fifth or seventh day of the final juvenile stage. This indicates that ecdysone contributes greatly to molting progression in both morphs, supporting the juvenile-to-adult transition. Additionally, comparison between long-wing and short-wing morphs exhibited a significantly larger magnitude of upregulation of most ecdysone production and signaling genes in short-wings. This result suggests that ecdysone plays an important role in wing morph determination in Gryllus lineaticeps, with elevated ecdysone biosynthesis potentially biasing development in favor of a short-wing morph. Understanding genetic and hormonal

influences like ecdysone and their effect on wing morph determination holds broader implications for understanding life-history determination at the molecular level.

P163. Spatial and Temporal Expression of the 2B Receptor During Early Embryogenesis, Saumaun Zadeh, Loyola Marymount University, Faculty mentor: Maxellende Ezin

Spatial and Temporal Expression of the 2B Receptor During Early Embryogenesis The serotonergic system acting through receptor 2B plays an important role in adult vertebrates, regulating processes such as sleep cycles in the central nervous system and smooth muscle tone in the cardiovascular system, with potential involvement in hypertension. In the vertebrate embryo, serotonin has been found at developmental stages of blastulation, gastrulation, neurulation and early organogenesis. Yet, mainly because the location of many serotonergic receptors are unknown at these stages, the function of serotonin in early vertebrate embryos is largely unclear. To begin to answer these questions, we are determining the developmental spatiotemporal expression of serotonin receptor 2B, using the chick embryo as a model organism. Chick embryos were collected from Hamburger Hamilton stages 4 (gastrulation) to 12 (early neurulation). The embryos were fixed in 4% paraformaldehyde, and processed for indirect immunohistochemistry with an anti-5HT2-B antibody. Whole mount imaging of stained embryos provided evidence that 2B receptor expression is found in the neural tube, neural crest, and somites. Interestingly the neural tube showed left-right asymmetrical expression. Future directions consist of completing staining, imaging, and sectioning our selected stages.

P164. Embryonic Transcriptome of Botrylloides violaceus from Long-Read Direct RNA Sequencing, Alexa Gutu, Jack Poole, California Polytechnic State University, San Luis Obispo, Faculty mentor: Elena Keeling

New developments in next-generation sequencing (NGS) have enabled the direct sequencing of long-read RNA transcripts. This study applies Oxford Nanopore Technology's (ONT) direct RNA sequencing (dRNA-seq) to generate a draft transcriptome for Botrylloides violaceus embryos. Colonial ascidians such as B. violaceus are chordates, the group of invertebrates most closely related to vertebrates. Transcriptomics has the potential to reveal genetic factors associated with their capabilities to perform both sexual and asexual reproduction and undergo whole-body regeneration (WBR). This study aims to assemble and annotate an embryonic transcriptome for comparison to a previously created adult transcriptome, as cellular processes that create body structures during embryogenesis must be mimicked during regeneration. To create the embryonic transcriptome, RNA was isolated from B. violaceus embryos and directly sequenced using the ONT MinION flow cell, producing roughly 2.5 million total reads. Read quality was measured with FastQC, confirming consistently high read quality across the dataset (median Q score 22.3), and reads were assembled using RNABloom2. Assembly quality was measured with QUAST, and the resulting transcriptome comprised approximately 30,000 sequences representing 10.1% of the

reference genome. Functional annotation was then performed using a BLAST2GO pipeline against an updated reference genome, resulting in approximately 42% of mapped reads annotated. Gene Ontology (GO) term analysis characterized biological functions and cellular pathways represented in the transcriptome. BUSCO analysis confirmed completeness of the transcriptome, with 66.2% complete transcripts for conserved Metazoan genes. This study demonstrates that direct RNA sequencing using Oxford Nanopore Technology is a feasible approach for generating high quality transcriptomic data in non-model organisms similar to B. violaceus.

P165. Adeno-associated virus-based gene therapy for diabetic corneal wound healing and stem cells, Ebony Lubin, California State University, Northridge, Faculty mentor: Gilberto Flores

Diabetes mellitus is an epigenetic disease that causes several ocular complications which can lead to blindness. In the cornea, it causes diabetic keratopathy, which is presented with delayed wound healing and limbal epithelial stem cells (LESC) dysfunction. Previously, we found that non-canonical WNT5A is hypermethylated in diabetic corneas, which results in suppressed Wnt-5a expression. Addition of recombinant Wnt-5a significantly improved diabetic wound healing and LESC phenotype. In this study, we utilized recombinant adeno associated virus (rAAV) as a gene delivery platform to introduce WNT5A into diabetic limbal epithelial cells (LEC) and organ-cultured corneas. Human diabetic cultured LEC and organ-cultured corneas were treated with rAAV6-eGFP as a control and rAAV6-WNT5A. The cells were successfully transduced with the virus as observed by eGFP positive staining with minimal cell death, observed by annexin V (apoptosis) and propidium iodide (necrosis) staining. We found that delivering WNT5A via rAAV6 in both diabetic LEC and organ-cultured corneas stimulated wound healing and putative LESC marker expression, keratins 15 and 17 by western blot or immunostaining. Thus, rAAV6 offers a safe new gene therapy approach to treat diabetic keratopathy.

P166. Use of Diabetic Limbal-Derived Induced Pluripotent Stem Cells to Derive Normalized Limbal Epithelial Progenitor Cells, Samantha Yao, California State University, Northridge, Faculty mentor: Gilberto Flores

Diabetes mellitus is the most widespread blinding disease in adults. It causes corneal complications, such as keratopathy and impaired wound healing due to limbal epithelial stem cell (LESC) deficiency (LSCD). Treatments include keratolimbal grafts and transplantation of cultured limbal epithelial cells (LEC) but are limited without a standardized source of LESC. Derivation of normalized differentiated corneal cells from diabetic induced pluripotent stem cells (iPSC)-derived LEC may be a therapeutic strategy for autologous transplantation. We used a preexisting protocol to differentiate iPSCs into LESC-like cells. Immunostaining was performed on days 2, 14, 21 for pluripotent (Oct4), LESC (ΔNp63, K17), and corneal (K12) marker expression. Our results support the generation of normalized LESCs from impaired diabetic LECs by iPSC redifferentiation. With

further investigation, this novel approach may serve as a potential transplantation modality for LSCD and diabetic keratopathy.

P167. Biomimetic Corneal Model: Stromal ECM Induction for Studying Human Corneal Regeneration, Raquel Blanco, California State University, Northridge, Faculty mentor: Gilberto Flores

Existing corneal models fail to recapitulate native ECM architecture and epithelialstratification, limiting their translational relevance. Here, we present a biomimetic humancorneal model that promotes ECM production by primary human limbal stromal cells(LSCs) using vitamin C stimulation. Enhanced ECM deposition generates a relevant stromal microenvironment that supports the adhesion, proliferation, and organization of primary human limbal epithelial cells (LECs). This co-culture system recreates key features of native corneal stromal–epithelial organization, overcoming limitations of conventional flat substrates. The resulting ECM-rich construct provides a physiologically relevant platform for investigating corneal biology, epithelial–stromal crosstalk, disease mechanisms, and regenerative therapies.

P168. Examining Senescence in Aged Mouse Livers treated with UK5099, Karina Huitzil, California State University, Northridge, Faculty mentor: Gilberto Flores

As organisms age, fundamental changes occur, particularly in tissue regeneration. Aging is associated with the reduction of stem cell activation and proliferation. A major contributor to this reduction is the accumulation of senescent cells, which disrupts tissue repair and regeneration. Evidence has previously shown that UK-5099, a pharmaceutical drug that changes cellular metabolism, enhances hair follicle stem cells (HFSCs). Treatment with UK-5099 has shown to have a positive impact on the hair cycle by promoting HFSC activation. We hypothesize that UK-5099 treated mice would have lower senescence levels in aged liver tissues. To examine senescence across aged livers, we looked at β-Galactosidase staining in young, aged, and aged UK-5099 livers. We found that UK-5099 treatment reduced the high senescence observed in aged liver tissues. By investigating how UK-5099 influences senescence in aged liver tissues, this work aims to clarify its potential effects in the context of aging.

P169. Piezo1 Function in Intestinal Smooth Muscle Cells, Ashley Menodza-Urbina, California State University, Northridge, Faculty mentor: Gilberto Flores

Piezo1 is a mechanosensitive ion channel that helps maintain intestinal homeostasis. When Piezo1 is deleted in smooth muscle cells, it leads to significant structural and cellular changes. To see how these changes develop over time, we analyzed distal intestine mouse tissues from 3 to 21 days post Tamoxifen induced Piezo1 knockout. Using whole-mount confocal immunofluorescence microscopy and qPCR, we focused on smooth muscle cell density, apoptosis, proliferation, and

mitochondrial dynamics. Our results show that deleting Piezo1 causes a loss of smooth muscle cells, with the greatest reduction seen after day 7, in the circular muscle layer. This loss of smooth muscle is connected with increased apoptosis but no increase in proliferation, showing that Piezo1 is needed in smooth muscle cell survival and intestinal homeostasis. These results suggest that Piezo1 plays a key role in maintaining cellular homeostasis in the gut.

P170. Investigating the role of thyroid hormone on cardiomyocyte size and cell division using digital holographic imaging, Matt Chu, San Jose State University, Faculty mentor: Alexander Payumo

After heart injury, pre-existing adult mammalian cardiomyocytes have limited capacity to divide into new muscle cells to regenerate the tissue. Neonatal mammalian cardiomyocytes on the other hand, retain proliferative capacity for a short window after birth and can regenerate the heart. During this postnatal transition, cardiomyocytes undergo cell cycle arrest where they become hypertrophic and multinucleated, which is partly driven by increasing thyroid hormone levels after birth. How thyroid hormone signaling inhibits cardiomyocyte proliferation is not fully understood. In this project, we isolate postnatal day 1 (P1) to P3 cardiomyocytes from neonatal rats and culture them under serum-free chemically-defined conditions to model the suppressive effects of the thyroid hormone, triiodothyronine (T3), on cardiomyocyte proliferation stimulated through inhibition of glycogen synthase kinase 3 (GSK3). We observe that treatment with 3 nM T3 decreases cardiomyocyte-specific incorporation of 5-ethynyl-2'-deoxyuridine (EdU), an S-phase marker, by 2-fold (59.66% to 29.82%). Using three-dimensional digital phase holographic imaging to capture time-lapse videos with minimal phototoxicity, we track the dynamics of single cardiomyocytes and capture changes in cell optical volume and cell division frequency. We observed a 2.5-fold decrease (26.9% to 11.48%) in complete cell division events. Despite decreasing the frequency of cell division events, T3 did not alter cardiomyocyte optical volume increase within 24 hours post-GSK3 inhibitor treatment, suggesting a possible inhibition at the level of the G1-S transition. Future experiments will aim to determine whether thyroid hormone enables cardiomyocytes to bypass their size-increase threshold for triggering cell division, thereby promoting hypertrophic growth. By better understanding the role thyroid hormone plays in changing cardiomyocyte morphology and proliferative capacity it can aid in understanding how to reverse this in adults after heart injuries.

P171. PTPN14 Regulates Hippo–TAZ Signaling to Maintain Endothelial Homeostasis and Prevent Vascular Malformations, Chenyang Xu, University of California, Davis, Faculty mentor: Daniah Beleford

Vascular malformations arise from abnormal development of arteries, veins, capillaries, and lymphatic vessels and can lead to significant morbidity and mortality. The Hippo signaling pathway plays a central role in regulating organ development and tissue homeostasis, in part by

controlling angiogenesis. The Hippo pathway regulates cell proliferation, differentiation, and apoptosis, primarily through contact inhibition in epithelial and endothelial cells. YAP and TAZ are transcriptional co-activators of the Hippo signaling pathway, with distinct tissue-specific expression patterns; TAZ is highly expressed in vascular endothelial cells. Hippo signaling pathways are regulated by genetic modifiers. One genetic modifier is Protein Tyrosine Phosphatase Non-Receptor Type 14 (PTPN14). PTPN14 interacts with YAP in epithelial cells and has been implicated in multiple diseases, including cancer. However, its role in endothelial Hippo signaling remains undefined. Here, we demonstrate that PTPN14 is essential for maintaining Hippo pathway activity in endothelial cells by sequestering TAZ in the cytoplasm and promoting its degradation under normal physiological conditions. Loss of PTPN14 results in increased nuclear localization of TAZ, uncontrolled cell proliferation, and thereby promotes vascular malformations. To link human genetic variation to molecular mechanisms, we generated a comprehensive PTPN14 variant map and employed AlphaFold-based structural modeling to predict how disease-associated variants may disrupt PTPN14-mediated protein interactions that normally restrain TAZ activity. Together, these findings identify PTPN14 as a critical regulator of endothelial Hippo-TAZ signaling, clarifying its role in normal vascular development and providing mechanistic insights into the human vascular malformations.

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P172. Live-cell imaging studies of CSPα during phagocytosis by retinal pigment epithelium cells, Sarabjit Kaur Dhillon, Bryn Mawr College, Faculty mentor: Adam Williamson

Kufs Disease, also known as Ceroid Lipofuscinosis, Neuronal 4 (CLN4), is an adult lysosomal storage disorder and neurodegenerative disease that causes progressively devastating symptoms such as muscle coordination loss and seizures. CLN4 is inherited through autosomal dominant mutations in the DNAJC5 gene, which encodes Cysteine String Protein alpha (CSPα). CSPα is a co-chaperone in cells that supports the fusion between vesicles and the cellular plasma membrane for exocytosis. For example, in the neuron, CSPα interactions within the SNARE pathway lead to vesicle release into the synapse. It is also thought that CSPα coordinates exocytosis of misfolded proteins as a part of the "misfolded-associated protein secretion" process to lessen stress on the cell to break these down. Collectively, these studies of CSPα connect it to having important roles in membrane biology. Given the extensive and dynamic membrane remodeling events coordinated during phagocytosis, we wondered whether CSPα plays a functional role in this universal immune process. We chose the retinal pigment epithelium (RPE) as a model system due to the high endogenous expression of CSPα in these cells. We reconstituted phagocytosis in live RPE cells and monitored GFP-CSPα dynamics during phagocytosis of synthetic cell corpses. We found that CSPα+ puncta fuse with nascent phagosomes on the timescale of minutes, solidifying CSPα as a novel early phagosomal protein. The next steps of this work include screening whether CSPα interacts with cellular structures involved in phagolysosome formation and generating mutants to learn which domains of CSPα drive phagosomal localization. New information about the functional role of CSPα in phagocytosis may allow for steps towards analyzing the molecular dysregulation in Kufs Disease.

P173. Uncovering the Phagocytic Mechanisms behind Batten Disease: Imaging CLN Protein Localization in a Human RPE Cell Model, Samantha Wagner-Robertson, Bryn Mawr College, Faculty mentor: Adam Williamson

Batten disease is the most common neurodegenerative disorder in children. Mutations in one of thirteen neuronal ceroid lipofuscinosis (CLN) proteins cause dysfunctional phagocytosis and, consequently, disease onset. Variability in disease presentation and progression among patients reflects the heterogeneity found across these thirteen proteins. Despite this diversity, a common early indicator of Batten disease is vision degradation. In this project, I aim to uncover the cellular biology of Batten disease CLN proteins during phagocytosis using a human retinal pigment epithelium (RPE) cell model. Using live-cell confocal microscopy, I examine the clearance of

human embryonic kidney (HEK) cell corpses by wild-type and mutated RPE cell lines expressing fluorescently labeled CLN proteins. This approach allows for the analysis of protein localization towards and surrounding corpse debris. My findings demonstrate rapid localization of multiple CLN proteins to corpse debris seconds after engulfment. Interestingly, this behavior is observed across CLN proteins with differing functions and locations. Notably, CLN3, a lysosomal transporter; CLN7, a chloride channel; and CLN12, a polyamine transporter, all localize to cell debris at comparable intensities. CLN3, the most commonly mutated Batten Disease protein, is among the strongest localizers. These findings suggest an active role for CLN proteins in RPE phagocytosis and debris degradation, with current studies examining each protein's specific contribution to phagocytic debris clearance in the retina. Future studies will look to identify associated organelles involved with specific CLN protein activity through the use of Rab GTPases, as well as further structural analyses of each protein to illuminate broader links between CLN dysfunction and disease progression.

P174. Reassessing the antiviral mechanism of APOBEC3G in HIV restriction, Josel Perez, College of the Holy Cross, Faculty mentor: Ann Sheehy

APOBEC3G (A3G) is a human cellular restriction factor with activity that inhibits HIV replication. The protein is primarily expressed in immune cells and interferes with viral reverse transcription via its well-characterized cytidine deaminase activity and potentially alternative non-catalytic mechanisms. Our lab has generated a library of A3G variants and is characterizing these variants for enzymatic and anti-viral functions. While catalytic function was assessed with an antibiotic-resistance assay (bacterial cells), anti-viral activity was quantified with an infectivity assay in human cells. Using these categorizations, 6 distinct A3G variants have been identified exhibiting decreased catalytic activity but also strong anti-viral activity. This is significant because the primary known mechanism A3G uses to suppress HIV infection is catalysis, these variants could be used to probe alternative antiviral function of A3G. We are also in the process of sequencing these HIV genomes post-infection to determine whether the loss of mutational capacity seen in our antibiotic assay reflects true protein activity in an infection setting. If these 6 variants exhibit a potent antiviral activity in the absence of enzymatic function, it could help determine the regions of A3G responsible for its enzyme-independent antiviral activity.

P175. Recurrent Endotoxin Exposure Drives a Heritable Maladaptive State in Macrophages, Dhruv Suresh, University of California, Los Angeles, Faculty mentor: Anthony Covarrubias

Resident macrophages are long-lived innate immune cells that are continuously exposed to pathogens, yet the impact of recurrent endotoxin exposure on human health span has not been studied. Recent evidence suggests acute exposures to pathogens have a capacity to remodel a macrophage's epigenome to suppress excess and unnecessary inflammation, therefore we hypothesize the long-term impact of a remodeled epigenome establishes a distinct heritable

program that drives immunocompromised states in macrophages throughout aging. This study examines whether macrophages can survive repeated endotoxin stimulation in vitro and determines whether this causes age-related immune decline in vivo. Additionally, we aim to delineate the molecular mechanisms of endotoxin induced epigenetic remodeling throughout the life-span of a macrophage. To test this, primary macrophages derived from the bone marrow were stimulated repeatedly with endotoxin, then assessed for acute cell death, cellular senescence, and the capacity to restore metabolic functions. We found that repeated exposure to endotoxin did not induce macrophage senescence nor cell death; DNA damage resolved and macrophages regained proliferative capacity. However, progeny retained a persistent maladaptive memory characterized by diminished effector function. Interestingly, repeated inflammatory exposures establish a heritable maladaptive state distinct from senescence. Future work will test whether epigenetic perturbations can restore chromatin accessibility and evaluate infection outcomes using reporter mice, labeled bacteria, and graft infection models to further define the physiological impact of maladaptive macrophage memory. Additionally, epigenetic remodeling will be performed using ATAC-seq to test chromatin plasticity. In vivo relevance will be assessed using M1-reporter mice subjected to repeated inflammatory exposures and bacterial infection models. This work will provide an understanding on how repeated inflammatory challenges impair bacterial clearance in aging and identify key mechanisms to reverse the effects of an aged immune system.

P176. Comparative analysis of motile cilia arrays on tetrahymena bacteria using immunofluoresce, Diego Valderrama, Santa Clara University, Faculty mentor: Brain Bayless

Motile cilia are hair-like extracellular appendages that move in coordinated motions to transport fluid and cells in the body. Abnormalities in motile cilia function can result in serious human health issues, such as respiratory diseases, hydrocephalus, fertility issues, and cilia-related congenital heart diseases. Despite their importance in human health, an understanding of how ciliary arrays coordinate to move extracellular fluid is poorly understood. Due to human motile cilia being a technically challenging culture, Tetrahymena thermophila has emerged as a model organism for ciliary array research. Tetrahymena are aquatic protists whose cilia are structurally and molecularly conserved with human cilia, which makes them a valuable model organism. By exploring the parameters of ciliary array organization with respect to extracellular fluid flow, we hope to gain an understanding of how motile cilia coordinate within an array. To gather preliminary information on these ciliary arrays, immunofluorescence was performed on multiple strains of Tetrahymena to image their cilia and basal bodies. From these images, the length and arrangement of these structures are measured and then compared between the various strains of Tetrahymena. Our experiment focused on the starved states of Tetrahymena americanis, borealis, ellioti, and thermophila. The data gathered from immunofluorescence is ultimately assessed alongside swim speed data for these strains, to identify optimal cilia array structure characteristics for fluid movement.

P177. Tetrahymena Swim Speed, Cameron Zirkel, Santa Clara University, Faculty mentor: Brain Bayless

Motile cilia are extracellular appendages that beat in a whip-like motion to direct fluid flow in the body. Defects in motile cilia can lead to serious human health conditions, such as Primary Ciliary Dyskinesia (PCD), a genetic disorder that often leads to chronic respiratory infections, bronchiectasis, infertility, ear infections, hearing loss, and neonatal distress. Aquatic protists Tetrahymena, with their diverse species and clear ciliary responses, offer a valuable model for exploring the mechanisms underlying ciliary movement, which may have implications for understanding and treating PCD. Various Tetrahymena strains have been utilized for these studies, however, there does not seem to be a standard correlation between basal body organization and swim speed. In this study, we examined the basal body organization and swim speed of Tetrahymena americanis, borealis, corlissi, pyriformis, shanghaiensis, thermophila, and vorax. Our results suggest that larger cells swim faster than smaller cells, as close spacing along ciliary rows is also correlated with quicker swim speed. Overall, we are beginning to determine the parameters of ciliary array organization that affect the output of directed fluid mechanics. In my undergraduate research on Tetrahymena ciliary function, I have explored how environmental conditions impact motile cilia. Under fed and starved conditions, one sees how the availability of nutrition greatly influences swim speeds and how Tetrahymena species adapt to these environmental conditions. My research adds to the general knowledge of ciliary function, with possible implications to further understanding the rare genetic disorder of PCD. Through my involvement in research, I have learned the importance of cellular biology, acquiring essential analytical and problem solving skills. Though I am new to the world of research, I hope that my work makes a worthwhile contribution to the field of science.

P178. Berberine Reduces the Viability and Proliferation of A-673 Cancer Cells in vitro, Nicolas Caputo, California Lutheran University, Faculty mentor: Chad Barber

Berberine Reduces the Viability and Proliferation of A-673 Cancer Cells in vitro Abstract There is an increasing need for complementary treatments to cancer as patients face adverse side effects of chemotherapeutics. Attention to herbal medicines historically used in Traditional Chinese Medicine has increased as a possible source of bioactive compounds that can support conventional therapies. Berberine, a natural isoquinoline alkaloid herbal compound, has shown promising anti-cancer effects in other studies. In this study, we tested various concentrations of berberine chloride (10, 20, 40, 80uM) against A-673 cells in vitro, at 24, 48, and 72hr marks, using flow-cytometry, microscopy, and hemocytometry. We found that berberine decreased cell proliferation and viability, inducing a dose and time-dependent cell death in the A-673 cells. Although cell death increased, berberine was not found to affect apoptosis in cells after 72 hours as determined by FITC/PI staining and flow-cytometry. Berberine in this experiment exhibits anti-cancer effects on A-673 cells by decreasing proliferation and viability, showing its potential as

a complementary therapy. Future directions include additional flow-cytometry trials to further characterize the mode of cell death, testing against additional cell lines, as well as primary tissue such as canine tumors.

P179. Confinement Modulates Mechanotransduction in Prostate Cancer Bone Metastasis Organoids, Iris Sloan, Brianna Gaughan, University of California, Los Angeles, Faculty mentor: Daniel Kamei

Prostate cancer (PCa) is the second most prevalent cancer in men worldwide, which commonly metastasizes to bone. Metastasized PCa cells move into the pores of trabecular bone, becoming physically confined. This activates integrins and downstream mechanotransduction pathways such as YAP and the Unfolded Protein Response (UPR), established drivers of PCa progression. To model this signaling, we fabricated PDMS microwells of varying diameters (200μm-500μm), matching the various sizes of bone pores. PC3 cells were successfully inserted into the wells and cultured for 10 days. We investigated XBP1s and c-Myc, key transcription factors that participate in the UPR. Furthermore, we probed the activity of YAP and Ki67, markers of PCa proliferation and progression. Immunofluorescence imaging was used to visualize the levels of each transcription factor. Confinement altered growth dynamics and proliferation. XBP1s, Ki67, c-Myc, and YAP levels increased in confined cells relative to unconfined controls. Ultimately, this microwell model simulates the mechanically complex bone marrow niche, enabling us to study metastasized PCa under physiologically meaningful conditions.

P180. Comparative Analysis of Motile Cilia Arrays on Tetrahymena Bacteria Using Immunofluorescence Spectroscopy, Alena Gagnon, Diego Valderrama, Santa Clara University, Faculty mentor: Brian Bayless

Motile cilia are hair-like extracellular appendages that move in coordinated motions to transport fluid and cells in the body. Abnormalities in motile cilia function can result in serious human health issues, such as respiratory diseases, hydrocephalus, fertility issues, and cilia-related congenital heart diseases. Despite their importance in human health, an understanding of how ciliary arrays coordinate to move extracellular fluid is poorly understood. Due to human motile cilia being a technically challenging culture, Tetrahymena thermophila has emerged as a model organism for ciliary array research. Tetrahymena are aquatic protists whose cilia are structurally and molecularly conserved with human cilia, which makes them a valuable model organism. By exploring the parameters of ciliary array organization with respect to extracellular fluid flow, we hope to gain an understanding of how motile cilia coordinate within an array. To gather preliminary information on these ciliary arrays, immunofluorescence was performed on multiple strains of Tetrahymena to image their cilia and basal bodies. From these images, the length and arrangement of these structures are measured and then compared between the various strains of Tetrahymena. Our experiment focused on the starved states of Tetrahymena americanis, borealis,

ellioti, and thermophila. The data gathered from immunofluorescence is ultimately assessed alongside swim speed data for these strains, to identify optimal cilia array structure characteristics for fluid movement.

P181. Anti-cancer Effects of Myricetin, a Plant-based Compound, Nevign Angelique Besas, California Lutheran University, Faculty mentor: Chad Barber

Moringa oleifera, colloquially known as horseshoe radish or malunggay (Tagalog), is a plant used extensively as cultural herbal medicine throughout African and Asian countries, such as Nigeria, India, and the Philippines. Moringa contains a variety of bioactive compounds throughout its entirety as a plant, leading to its interest in biomedical research applications. Myricetin is a flavonoid that is abundantly found in Moringa that exhibits strong anti-oxidant properties. Previous studies have shown that Myricetin demonstrates anti-diabetes, anti-inflammatory, and anti-cancer effects in vitro. Our research aims to further investigate the anti-cancer properties of Myricetin in vitro by assessing its effects on cell viability, migration, apoptosis, and inflammatory cytokine secretion in various concentrations of 0uM, 25uM, 50uM, and 100uM. Our study found that Myricetin significantly reduces cell migration, decreases cell viability, and promotes apoptosis in cancer cells in vitro in a dose-dependent manner. Additionally, we found that Myricetin inhibits the secretion of IL-6 in cancer cells, an inflammatory cytokine linked to tumor growth and metastasis. Further studies will be conducted to examine the effects of Myricetin on protein and gene expression, in long-term treatments, and potency when used with common cancer drugs in treated cancer cells. Our findings emphasize Myricetin as a potential chemosensitizer for current targeted therapies to increase the effectiveness of these pre-existing therapies against more aggressive forms of cancer. As cancer continues to affect the global population, there is an increasing need for new and more effective treatments, especially as more patients develop resistance to existing therapies.

P182. Developing Medical Countermeasures: Investigating the immune responses to a live attenuated vaccine for Francisella tularensis, Aidan Jones, United States Air Force Academy, Faculty mentor: Derek Barthels

Francisella tularensis (Ft), a facultative intracellular bacterium causing tularemia, poses a minimal community risk in the United States, but is a major bioterrorism threat due to its weaponization potential. A 1970 World Health Organization study estimated that aerosolizing 50 kg of Ft over a city of 5 million people could kill 19,000 and incapacitate 250,000. A 1997 study also projected costs of $5.5 billion ($11.1 Billion in 2025) per 100,000 people infected. Ft is a Tier 1 Select Agent according to the Center for Disease Control and Prevention, and while it is treatable with antibiotics, the potential for antibiotic resistance and distribution challenges may limit their effectiveness. Various groups in the United States have tried to develop an Ft vaccine to remedy this deficiency, but none have yet achieved FDA approval. We are proposing Ft SCHU S4 ΔclpB

(ATI-1701) as a live attenuated vaccine for tularemia. Animal studies to date have shown ATI-1701 to be safe and effective, with as few as six CFU ATI-1701 protecting up to 100% of mice from subsequent lethal challenge. While most vaccines' effects are measured by humoral responses, Ft's intracellular nature implicates that measurements of immunocytes' responses may be useful for further development. This study explores immune responses of THP-1 macrophages and peripheral blood mononuclear cells (PBMCs) to ATI-1701. Specifically, these experiments investigate which immunity-related molecular pathways ATI-1701 activates in PBMCs. These results offer insights to help find potential mechanisms of immunity, which are used to help bridge the gap between preclinical and clinical studies of ATI-1701 as a vaccine candidate.Abstract Disclaimers: The views expressed in this presentation are those of the author and do not necessarily reflect the official policy or position of the U.S. Air Force Academy, the Air Force, the Department of Defense, or the U.S. Government.

P183. Enhanced Regeneration Rate in Planaria Suggests Effectiveness of Red-Light Therapy, Angelica Velasco, Alison Grammond, The Master's University, Faculty mentor: Haley Smith

Red-light therapy (RLT) is an emerging method for cellular regeneration and repair in the beauty and medical industry. With sources advertising effects such as improving inflammation, acne, and even helping slow the progression of dementia, RLT holds potential for improving health care as an accessible and non-invasive medical tool. As an emerging tool, there is much to be investigated regarding its efficacy and the cellular mechanisms that underlie its effects. Due to the remarkable regenerative capacity of planaria, they serve as a good model for exploring the effectiveness of RLT. To explore this, we assessed the regenerative capacity of bisected planaria in the presence of red light compared to full-spectrum white light. We utilized reformation of anterior eye spots from posterior planarian fragments as a marker of regeneration. After observing the planaria over the course of one week following bisection, it was determined that planaria exposed to red light regenerated their eye spots notably faster than those exposed to full-spectrum white light. A single dose of exposure to red light for ten minutes resulted in an average eye spot regeneration time of 11 hours quicker than controls. These findings suggest that red light has a positive impact on the rate of planaria regeneration and provide support for a possible role of RLT in general tissue regrowth and repair.

P184. Higher frequency of homologous chromosome pairing in human adult aortic endothelial cells, Belia Zetter Angulo, Isabella Bibayoff, Sonoma State University, Faculty mentor: Lisa Hua

During mitosis, pairing of homologous chromosomes can be detrimental and has been correlated with gene misregulation, chromosomal aberrations, and various pathological diseases. We previously demonstrated that homologous chromosomes are spatially segregated, or antipaired, in neonatal human endothelial cells at metaphase/anaphase, which may help prevent abnormal

recombination. However, it is unclear if this antipairing persists in adult endothelial cells. To test whether the antipairing, or one homolog per nuclear hemisphere motif, is conserved in adult endothelial cells, we examined human aortic endothelial cells at metaphase. Using ImmunoFISH and high-resolution confocal microscopy to visualize the chromosomes and centrosomes, we found that small homologous chromosomes 13, 15, 17, 19, 21, 22, and the sex chromosomes, XY, exhibit a loss of spatial segregation in human adult aortic endothelial cells. In contrast, fewer adult endothelial cells showed a loss of segregation for the larger chromosomes 1, 4, and XX, suggesting a gradual decline in the fidelity of spatial segregation of homologous chromosomes. Notably, we observed a higher frequency of abnormal pairing in both small and large chromosomes in adult aortic endothelial cells as compared to neonatal umbilical vein endothelial cells. These findings suggest that mechanisms governing chromosome antipairing may decline with aortic endothelial cell age, leading to increased susceptibility to abnormal pairing and cardiovascular disease.

P185. Verification of endothelial cell identity with a CD31/PECAM-1 marker for human adult endothelial cells, Sarah Sanchez Hurtado, Sonoma State University, Faculty mentor: Lisa L. Hua

It was discovered that homologous chromosomes spatially segregate across the axis defined by the two centromeres in a dividing human cell, a process called antipairing. Antipairing has been reported for neonatal human umbilical vein endothelial cells (HUVECs). A potential mechanism for preventing somatic homologous pairing and recombination. Mitotic recombination can lead to genetic instability, including the loss of heterozygosity which could be detrimental to the cell. Yet, it is unknown whether this antipairing organization observed in neonatal endothelial cells is maintained throughout adulthood. Preliminary findings from our lab indicate a loss of antipairing in human adult aortic endothelial cells (HAECs) derived from six patients. This suggests that the antipairing organization may be disrupted in adult endothelial cells. However, it remains uncertain whether the endothelial cell phenotype is preserved in these adult cells. To test whether the adult HAECs preserve their endothelial cell identity, we completed immunofluorescence staining for an endothelial cell junction marker, CD31/PECAM. CD31/PECAM is a cell-to-cell anchoring junction marker that maintains endothelial cell junctional integrity that localizes to the cell's outer membrane. Neonatal HUVECs are used as our positive control, while retinal pigment epithelial-1 (RPE-1) cells, an epithelial cell line, serve as our negative control. Our data shows CD31/PECAM localization to the outer membrane of HUVECs and HAECs, with no such localization observed on the outer membrane of the RPE-1 cells. Taken together, this data suggests the adult HAECs have preserved their endothelial identity, whereas the absence of marker staining would have implied the loss of identity. Findings of our study have provided new insight into chromosome organization and its loss of fidelity in aged endothelial cells, suggesting a link to vascular pathology.

P186. Using a mouse cell model to understand chromosome organization patterns, Shelby Giannamore, Sonoma State University, Faculty mentor: Lisa Hua

A non-sex cell contains two copies of each chromosome, one from each parent. These homologous chromosomes in human cells separate to opposite sides of the cell. This organization may function to prevent abnormal exchange of genetic material, which has been linked to genomic instability. To test whether other mammalian models also maintain this spatial segregation of homologous chromosomes, we selected mouse fibroblast cells, as mice share approximately 90% genetic similarity to humans. The spatial segregation of chromosomes 17, X and Y in mouse cells were previously established, suggesting a shared chromosome organization pattern among mice and humans. Our preliminary data of mouse homologous chromosomes show a lack of spatial segregation of chromosomes 1, 2, 17, 19, and X. These results suggest that spatial segregation may be lost. These findings will determine whether spatial segregation is conserved in mice, providing new information on mouse model usage in human biology.

P187. Subphenotype Characterization within Unsupervised Clustering, Akshat Ubale, University of California, Los Angeles, Faculty mentor: Teresa Kortz

Pediatric sepsis is a leading cause of pediatric death globally, with a disproportionate burden in sub-Saharan Africa. A lack of tools for early risk stratification prevents targeted care. Using data to subphenotype can help solve this issue.Objectives:Using unsupervised clustering to identify subphenotypes. Then, using the subphenotypes to compare clinical characteristics, biomarker profiles, treatment, and mortality across subjects.Methods:A prospective cohort study was conducted at the Muhimbili National Hospital in Tanzania with 325 children with severe sepsis aged >28 days to 14 years. Participants were followed until discharge or death. Clinical data were collected in Tanzania, while metagenomic sequencing, RNA sequencing, and biomarker assays were completed at UCSF. Leiden clustering was then applied to the data. We evaluated cluster differences with median values or percentages combined with p-values, with a significance threshold of $p < 0.05$.Results:Cluster 1 (n=109), cluster 2 (n=100), and cluster 3 (n=116) had significant clinical differences in median age in months ($p=0.019$), median white blood cell count ($p<0.001$), and median glucose in mmol/L ($p=0.029$). The biomarkers median procalcitonin in ng/mL, median ferritin in ng/mL, and median C-reactive protein in mg/L, all had significant differences across the clusters with $p<0.001$. Among the clusters, 1 had the highest median level of each biomarker. While the mortality rate without steroid treatment was significant with $p<0.001$, the mortality rate with steroid treatment was not, with $p=0.995$. Conclusion: The three subphenotypes identified by unsupervised clustering display substantial disparity in multiple clinical characteristics, inflammation as indicated by biomarkers, and mortality without steroids. These findings emphasize the potential of clustering-based stratification to create more targeted decisions to ameliorate outcomes in sub-Saharan Africa.

P188. Investigating Pharmacological Inhibition of Protein Tyrosine Phosphatases as a Novel Therapeutic Intervention for Diabetic Kidney Disease, Ayaka Sonehara, University of California, Davis, Faculty mentor: Fawaz Haj

Diabetic kidney disease (DKD) is a serious consequence of diabetes characterized by progressive podocyte dysfunction and loss, leading to breakdown of the glomerular filtration barrier (GFB). Podocytes are specialized epithelial cells that form the final boundary of the GFB, and animal studies show that loss of 20% of podocytes is sufficient to induce DKD. Moreover, podocyte-specific insulin receptor knockout in animal models recapitulates DKD features. Insulin signaling is modulated, in part, by protein tyrosine phosphatases (PTPs), which dephosphorylate key components. We investigated the effects of pharmacological inhibition of PTP1B, T-cell PTP (TCPTP), and Src homology PTP2 (SHP2) in E11 podocytes. Podocyte motility was assessed using a scratch migration assay under normal- and high-glucose conditions over 48 hours with AC484 (dual PTP1B/TCPTP inhibitor) or SHP099 (SHP2 inhibitor). Insulin signaling was assessed by immunoblot of insulin receptor, ERK, and AKT phosphorylation, following insulin stimulation at 0, 5, and 20 minutes with or without AC484 or SHP099.PTP inhibition reduced podocyte motility and altered insulin signaling. Complementary in vivo studies are ongoing to evaluate the effects of these inhibitors in a diabetic mouse model of DKD. These studies provide insight into PTP-mediated mechanisms that may preserve podocyte integrity and limit DKD progression.

P189. CRISPRa-Mediated Activation Reveals Proliferation Dynamics in Estrogen Receptor Positive Breast Cancer Cells, Jessalyn Chen, Chloe Nacorra-Scruggs, Pepperdine University, Faculty mentor: J. Antonio Gomez

The development of new cancer therapies is greatly hindered by the scarcity of actionable genetic targets. Most cancer therapies on the market are designed around inhibiting oncogenes or exploiting cancer dependencies, but while these targets have played an essential role in cancer treatment, their effectiveness is reduced by mutational heterogeneity. To overcome this limitation in therapeutic development, more attention has turned to the role of epigenetics in cancer cell replication. For instance, it is increasingly recognized that some tumor cells have evolved mechanisms to silence genes detrimental to cell growth rather than inactivating them via mutational changes. Our hypothesis is that cancer cells may epigenetically silence genes with anticancer properties. Thus, our study aims to determine if reactivating epigenetically silenced genes alters the rate of cancer cell proliferation. Specifically, we have used a dCas9-CRISPRa system to selectively activate 7 genes with proposed involvement in proliferation of MCF7 breast cancer cells. Our results indicate that epigenetically activated genes can alter cell replication rates early after gene activation or induce changes in cell cycle exit at later stages of activation. These

results provide strong proof of concept that epigenetically silenced genes can expand the repertoire of actionable targets in cancer treatment.

P190. Proliferation of Estrogen Receptor Positive Breast Cancer Cells following Transcriptional Activation using CRISPRa Epigenetic Editing, Chloe Nacorra-Scruggs, Pepperdine University, Faculty mentor: J. Antonio Gomez

The development of new cancer therapies is greatly hindered by the scarcity of actionable genetic targets. Most cancer therapies on the market are designed around inhibiting oncogenes or exploiting cancer dependencies, but while these targets have played an essential role in cancer treatment, their effectiveness is reduced by mutational heterogeneity. To overcome this limitation in therapeutic development, more attention has turned to the role of epigenetics in cancer cell replication. For instance, it is increasingly recognized that some tumor cells have evolved mechanisms to silence genes detrimental to cell growth rather than inactivating them via mutational changes. Our hypothesis is that cancer cells may epigenetically silence genes with anticancer properties. Thus, our study aims to determine if reactivating epigenetically silenced genes alters the rate of cancer cell proliferation. Specifically, we have used a dCas9-CRISPRa system to selectively activate 7 genes with proposed involvement in proliferation of MCF7 breast cancer cells. Our results indicate that epigenetically activated genes can alter cell replication rates early after gene activation or induce changes in cell cycle exit at later stages of activation. These results provide strong proof of concept that epigenetically silenced genes can expand the repertoire of actionable targets in cancer treatment.

P191. Targeting the C9orf72–SMCR8–WDR41 Complex: A Screening Platform for Small-Molecule Modulators in ALS, Shriprithi Vel Murugan, University of California, Berkeley, Faculty mentor: James Hurley

Mutations in C9orf72 are the leading genetic cause of amyotrophic lateral sclerosis (ALS) and frontotemporal dementia (FTD). C9orf72 forms a complex with SMCR8 and WDR41 (the CSW complex), acting as a GTPase-activating protein (GAP) for ARF1 to regulate vesicular trafficking. Despite its pathological importance, small-molecule modulators of CSW have not yet been identified. Here, we present a robust drug discovery pipeline for the CSW complex. By optimizing a biochemical GAP assay, we screened a ~5,000-compound library at two distinct time points: 30 minutes to identify activators (~2% hit rate) and 90 minutes to identify inhibitors (~2.5% hit rate). Hits were validated through orthogonal methods, including dose-response assays, thermal shift assays, and in silico predictions using Boltz2. Finally, we utilized cryo-EM to resolve an initial model of the C9orf72–SMCR8 (CS) subcomplex, significantly improving micrograph contrast through the addition of Late Embryogenesis Abundant (LEA) proteins. This work establishes a framework for targeting the CSW complex as a therapeutic strategy for neurodegeneration.

P192. Pattern Perturbation: Phenotypic Outcomes of Drug Treatment During Homeostasis and Fission, Eve Awad, University of San Francisco, Faculty mentor: James Sikes

Modification and re-specification of the adult body plan are uncommon in animals. However, organisms that reproduce asexually can modify body plans within the adult soma that would otherwise remain static. Fission is an asexual reproductive strategy that requires organisms to re-specify their body axes, yet the spatiotemporal dynamics of gene expression during fission and how these processes regulate fission rate remain poorly understood. The acoel Convolutriloba longifissura employs a unique double-fission strategy, making it a powerful model for investigating the molecular and morphological mechanisms underlying large-scale patterning changes associated with fission. In this study, we pharmacologically disrupted key developmental signaling pathways to examine their roles in both fission rate and homeostasis. Perturbation of Wnt, Hedgehog, and BMP signaling resulted in morphological defects during homeostasis and inhibited fission, providing insight into the molecular mechanisms that may initiate fission and regulate patterning processes. Given the deeply conserved roles of these pathways in embryonic development, our findings suggest that fission co-opts embryonic patterning mechanisms to initiate fission and repattern the adult soma.

P193. Defining the mechanism of action of Aurora B during cytokinesis, Eduardo Sanchez, University of California, San Diego, Faculty mentor: Karen Oegema

Cytokinesis is the process by which a mother cell divides to form two daughter cells. Cytokinesis is accomplished by assembly of a contractile ring on the cell cortex that encircles the cell equator and constricts to change cell shape and partition its contents into two daughter cells. Failure to complete cytokinesis can lead to cell death or contribute to cancer development; approximately 40% of cancers are thought to arise from a cell lineages that experienced failed cytokinesis resulting in whole genome doubling. Cytokinesis is regulated by the small GTPase RhoA; when RhoA is active on the cortex, it promotes constriction. During cytokinesis RhoA activation is controlled by the centralspindlin complex, composed of 2 molecules each of CYK-4 and ZEN-4. Centralspindlin is recruited to a microtubule-based central spindle that forms between the separating chromosomes and then diffuses to the overlying cortex to promote RhoA activation and contractile ring assembly. Centralspindlin is thought to be held inactive by the 14-3-3 protein PAR-5, and the mitotic kinase Aurora B, which localizes to the central spindle, may promote cytokinesis by relieving PAR-5-mediated inhibition of centralspindlin. Here, we test this model in the C. elegans embryo. Using a molecular replacement system for the ZEN-4 component of centralspindlin, we asked whether mutations that prevent PAR-5 from binding to ZEN-4 could rescue the cytokinesis defects observed in Aurora B depleted C. elegans embryos. Time lapse spinning disk confocal microscopy revealed that mutations preventing association of ZEN-4 with PAR-5 significantly rescue cytokinesis in Aurora B depleted embryos. Because mutations in

candidate Aurora B phosphorylation sites in centralspindlin do not compromise cytokinesis, we hypothesize that that Aurora B phosphorylates PAR-5 directly, causing PAR-5 to detach from the centralspindlin complex which can then promote cytokinesis.

P194. α-Synuclein Expression Blocks Lipophagy and Decreases Chronological Lifespan, Brandon Fong, University of California, Davis, Faculty mentor: Ken Kaplan

α-Synuclein (α-syn) has been identified as a definitive biomarker for Parkinson's Disease (PD), yet its role in the initiation, progression and ultimately the pathogenesis of PD is unclear. Applying the ideas around physiological allostasis to the loss of autophagy pathways, we propose that chronic cell stress over long periods of time could explain the journey of cells from normal to dysfunctional and ultimately disease states. Emerging evidence shows α-syn accumulates at the nuclear endoplasmic reticulum (nER), a critical subcellular region that mediates lipid transfer activities to other organelles as well as autophagy-mediated trafficking of ER and lipid droplets (LDs) to the vacuole/lysosome. Indeed, LD autophagy, or lipophagy, is critical for maintaining normal lipid and energy balances in cells and evidence suggests that it is defective in PD patients' neurons. We hypothesize that induced α-synuclein expression increases the cell stress load via organelle stress at the nER and disrupts lipophagy. Using a yeast model system to study this pathway, we have monitored lipophagy under glucose restriction (GR) in the presence or absence of human α-syn expression. Remarkably, we have shown that α-syn expression blocks lipophagy under GR. We are currently investigating the mechanism by which α-syn blocks lipophagy, but preliminary results suggest it may do so by disrupting the organization of the proteins that mediate both lipophagy and ER-phagy through contact sites at the nER. This novel finding raises the possibility that progressive loss of neuronal function is a consequence of disrupted lipophagy and altered energy metabolism. Additionally, acute GR is associated with activation of lipophagy and chronological lifespan (CLS) extension. We have found that α-syn expression reduces CLS extension, raising this as a possible mechanism associated with neuronal dysfunction in PD patients.

P195. Practical Visualization of Dividing Cells in Response to Injury and Infections, Diana Torres Ortiz, University of California, Merced, Faculty mentor: Nestor J. Oviedo

In most multicellular organisms, mitotic activity rises after injury or infection. This cellular response is unavoidable, but questions still exist about the spatiotemporal dynamics of cell proliferation. To gain insights about cellular dynamics in response to injury and infection, we use whole-mount immunostaining to detect proteins of interest. These can later be quantified to establish the expression in a specific cell, tissue, or organism. Our lab uses Schmidtea mediterranea, a planarian flatworm, as a model organism to study stem cell regulation, cancer, and host-pathogen interactions. Our interest in planarians stems from their remarkable regenerative capabilities, driven by their stem cells (neoblasts). Planarians are capable of overcoming infections

and repairing tissue after injury. One of the main reasons for overcoming injury and infections is the increase in actively dividing cells. We performed immunohistochemistry to detect phosphorylated histone-3, a marker of actively dividing cells, which can be observed and quantified by fluorescent microscopy. In planarians, we found that 6 hours after injury or infection, there is a noticeable increase in mitotic cells throughout the organism. Remarkably, we observed a differential rise in mitoses at 48 hours, whereas in response to infections, it occurs within 24 hours. Future work will address the molecular mechanisms underlying differential cellular dynamics during cell division to design strategies for future clinical interventions.

P196. Pigment Cells Mediate Pathogen Clearance in Planarians, Ashley Liao, University of California, Merced, Faculty mentor: Néstor J. Oviedo

Candida albicans is one of the most prevalent human fungal pathogens, leading to infections including oral thrush and yeast infections. However, current fungal infection models have major limitations. They do not allow us to visualize epithelial infection progression in a whole organism paired with a non-invasive infection system. The planarian flatworm Schmidtea mediterranea serves as a powerful in vivo infection model for studying host-pathogen interactions due to non-invasive infection methods and effective innate immune responses. These allow planarians to quickly eliminate and recover from C. albicans infections in 10-12 days although the mechanisms are largely unknown. Melanocytes (pigment cells) are known to have immune functions including pathogen recognition, phagocytosis, and antigen processing and presentation. During infection, non-uniform pigment was observed on the planarian epithelial surface. Here, we investigated the role of pigment cells in planarian antifungal immune response. To remove pigment cells, we continuously exposed planarians to white light. We reduced ≥80% of pigment cells from the planarian's body area in 7-15 days using this strategy. To confirm pigment loss, planarians were imaged and pigment changes were quantitatively analyzed using PlanaraChrome, a program used to quantify relative pigment levels. Depigmented planarians were infected with C. albicans for 3 days, following an updated planarian infection protocol via soaking. Depigmented planarians were prone to fungal infections and succumbed within 72 hours. This indicates that pigment cells play a critical role in host recovery and survival. Planarians have highly conserved genes, making them a suitable model for studying antifungal innate immune mechanisms. Determining the mechanisms involved in pathogen clearance will shed light on infection response in other organisms.

P197. Targeting Methionine Vulnerability in Hepatocellular Carcinoma Using an Genetically Engineered Salmonella Biologic, SGN1, April Zhao, Institution, Faculty mentor: Ping Mao

Liver cancer is one of the most prevalent malignancies worldwide and ranks third in cancer-related mortality. Hepatocellular carcinoma (HCC), accounting for approximately 75–85% of primary liver cancer, is the most common and clinically aggressive subtype, with limited effective treatment options. Methionine dependency is a well-recognized metabolic vulnerability in many

malignant cancers. However, systemic dietary methionine restriction or administration of free L-methioninase is not clinically feasible due to the essential role of methionine in normal physiology and the systemic toxicity associated with global methionine reduction. To exploit this metabolic dependency, we deployed SGN1, a genetically engineered Salmonella Typhimurium strain designed to overexpress L-methioninase and specifically reduce methionine levels within the tumor microenvironment. Currently, SGN1 already gained approval from the FDA, China NMPA, and China Taiwan TFDA agencies for clinical trials. Under methionine-restricted (MR) culture conditions, multiple HCC cell lines exhibited significant dose-dependent suppression of proliferation, reduced clonogenic capacity, G0/G1 cell-cycle arrest, increased apoptosis, and impaired migration and invasion of the HCC cells. Consistently, SGN1 treatment in vitro significantly inhibited HCC cell proliferation in a dose-dependent manner, supporting its ability to functionally mimic methionine depletion. In a subcutaneous HCCLM3 (a human HCC cell line)-derived xenograft model, a single intratumoral administration of SGN1 significantly reduced intratumoral methionine levels and suppressed tumor growth in a dose-dependent manner; higher bacterial doses resulted in progressively greater reductions in tumor volume and tumor weight without affecting body weight. In an immunocompetent Hepa1-6 syngeneic model, intravenous SGN1 administration similarly inhibited tumor growth in a dose-dependent manner with no significant impact on body weight. These results demonstrate that SGN1 exerts robust antitumor activity in both immunodeficient and immunocompetent models, with favorable systemic safety profiles. Collectively, these findings demonstrate that methionine depletion suppresses malignant phenotypes in HCC and support SGN1-mediated enzymatic methionine degradation as a promising therapeutic strategy.

P198. Determining the Subcellular Localization of TLR10 Using Flow Cytometry, Jia Li Chen, Chiomimi Pearl Ahans , University of San Francisco, Faculty mentor: Sangman Kim

Toll-like receptors (TLRs) are innate immune pattern recognition receptors (PRRs) that detect pathogen-associated molecular patterns and regulate downstream immune responses. TLR10 remains the only TLR whose ligand specificity, cellular localization, and immunological function are not fully defined. Emerging studies suggest that TLR10 may play a role in anti-inflammatory responses and can dimerize with TLR2 to generate immune signaling. Previous studies in our laboratory demonstrated that stimulation with Mycobacterium tuberculosis promotes the trafficking of TLR10 and TLR2 to the cell surface. Their subsequent dimerization was detected using Förster Resonance Energy Transfer (FRET). However, the subcellular localization and trafficking of TLR10 remain poorly understood. Here we show that HEK293T cells transfected with TLR2, TLR10, or co-transfected with TLR2+10 exhibit strong intracellular fluorescence intensity for TLR10 and TLR2+10. These cells were analyzed by flow cytometry under permeabilized and non-permeabilized staining conditions. We observed significantly higher TLR10 signal under the intracellular compared to extracellular staining conditions in both TLR10 alone and TLR2 + TLR10 transfected cells. These findings suggest that TLR10 is predominantly localized intracellularly in transfected HEK293T cells. In the future, stimulation with probiotic bacteria combined with flow cytometry analysis may further clarify the pathways regulating TLR10 localization and its potential role in immune signaling.

~~**P199. Evaluation of Gene Characteristics and Effects on Multi-Peptide Cleavage Efficiency**, Saharsh Madullapalli, Sophia Zhang, Aspiring Scholars Directed Research Program, Faculty mentor: Zane Chen~~

~~[Withdrawn]~~

~~2A sequences are 18-22 long oligopeptides referring to a specific region of the viral genome, and have been previously investigated in terms of use in gene co-expression, particularly in polycistronic vectors. These vectors allow for the expression of multiple genes, which are introduced into an organism through plasmids for various applications, such as cellular reprogramming, protein tagging, and gene therapy. However, a critical aspect of our project is understanding how gene transcript length and structure can influence the efficiency of expression. We aim to test and evaluate how changes in transcript length and structure may affect overall expression efficiency, ensuring optimal performance for downstream applications. Our project utilizes western blot and mass spectrometry to track the effects of gene characteristics between genes of varying lengths (small, medium, large) that affect efficiency of 2A sequence cleavage. This evaluation can be a potential stepping stone for future uses in therapeutics.~~

Molecular Biology #200-216

P200. Characterizing Growth and Induction Dynamics in Yeast Surface Display, Christell Martinez, California State University, Channel Islands, Faculty mentor: Gamze Camdere Tapia

Yeast surface display is widely employed in protein engineering for the presentation and selection of proteins on the surface of Saccharomyces cerevisiae. To support robust implementation of this technique, we characterized key experimental parameters in two commonly used yeast strains. Growth curves were measured under standard culture conditions, and doubling times were calculated to determine growth dynamics relevant to transformation timing. These analyses provided strain-specific insights into culture expansion rates. In parallel, we performed an induction time course for protein display, quantifying expression across multiple post-induction time points. This dataset revealed the kinetics of surface protein presentation and identified time windows most appropriate for downstream screening applications. Together, these characterizations establish a practical framework for anticipating culture behavior and induction response in yeast display experiments. The results provide foundational data that enable more consistent planning and execution of display-based protein engineering workflows.

P201. Mapping the UVRAG Promoter Region to Better Understand Transcriptional Regulation, Bijan Gonzalez, California State University, Northridge, Faculty mentor: Cindy Malone

Ultraviolet radiation resistance-associated gene (UVRAG) is a multifunction protein with roles in tumor suppression, autophagy, and DNA damage repair. Transcription is the process of turning DNA into RNA and is regulated by the promoter region located upstream of the gene. The promoter recruits RNA polymerase and contains transcription factor binding sites (TFBS), transcription factor proteins that bind to regulate gene expression. The aim of this project is to identify which regions of UVRAG promoter contribute to the highest levels of transcriptional activity. Bioinformatics analysis was used to identify the TFBS within the UVRAG promoter by matching nucleotide sequences from various programs to confirm the transcription factor in the promoter. The UVRAG promoter was ligated into pGL3-Basic Luciferase Reporter Vector to create a recombinant plasmid (RP) -1367 bp. Four sequential deletion constructs (DC) -1143 bp, -932 bp, -721 bp, and -530 bp of the RP were generated using site-directed mutagenesis (SDM), shortening the promoter region by 200 base pairs. All constructs were transiently transfected into human embryonic kidney 293T cells to quantify luciferase expression using a dual luciferase assay. The -1367 bp RP exhibited a 25.6-fold increase when compared to the promoterless pGL3-Basic indicating an active promoter. The -530bp RP showed a 26.6-fold increase when compared to the promoterless pGL3-Basic suggesting that TFBS driving transcriptional activation will be found within this region. It is hypothesized that SP1 or USF1 could be the main driving element due to their known activation function. Two additional sequential DC will be engineered

using SDM and transfected to further identify the TFBS responsible for maximal transcriptional activity. Identifying these regulatory elements will improve our understanding of how the UVRAG promoter controls gene expression and contributes to cellular processes such as tumor suppression, autophagy, and DNA damage repair.

P202. Analysis of RAD51AP1 Promoter Using Luciferase Expression, Fernando Martinez, California State University, Northridge, Faculty mentor: Cindy Malone

RAD51-associated protein 1 (RAD51AP1) is a DNA repair gene associated with homologous recombination, chromatin binding, and telomere maintenance. To further understand the regulation of the RAD51AP1 gene, its transcriptional activity must be analyzed by identifying positive regulatory elements within the promoter sequence and the core promoter, which yields the highest amount of gene expression. The RAD51AP1 promoter must be directionally cloned into pGL3-Basic, a vector containing a multiple cloning site (MCS) with restriction enzyme sites and the reporter gene luciferase. The RAD51AP1 promoter was amplified through PCR, followed by a restriction digest on both the amplified promoter and the MCS of pGL3-Basic to create complementary sticky ends which were ligated together through complementary base pairing to create a recombinant plasmid. Transient transfections into human embryonic kidney cells (HEK293T) with the recombinant plasmid produced a 20-fold increase in luciferase expression compared to the pGL3-Basic control, confirming an active RAD51AP1 promoter. Bioinformatics analysis was used to map the RAD51AP1 promoter for positive regulatory elements such as transcription factor binding sites, which are areas where regulatory proteins bind to initiate transcription, and the CpG island, which is an area rich in cytosine and guanine nucleotides. Using site-directed mutagenesis (SDM), new restriction enzymes will be introduced into the RAD51AP1 promoter by designing primers with specific nucleotide mutations to create deletion constructs that will identify which regulatory elements have a positive effect on transcriptional activity. All constructs will then be transiently transfected into HEK293T cells and transcriptional activity will be quantified using a dual luciferase assay. Identifying the core promoter and positive regulatory elements of the RAD51AP1 promoter will contribute valuable insight on gene regulation of a DNA repair gene to prevent the dysregulation of the RAD51AP1 gene.

P203. Analyzing Transcription Factors in the Promoter Region of USP47, Isabella Ballesteros, California State University, Northridge, Faculty mentor: Cindy Malone

The central dogma of biology is DNA being transcribed into RNA and translated into protein. A promoter is a region of DNA upstream of a gene that contains transcription factor binding sites (TFBS), which allow proteins to bind to specific DNA sequences to initiate transcription, thus dictating when a gene is turned on or off. Ubiquitin Specific Peptidase 47 (USP47) is associated with tumor development, metastasis, cell growth, and genomic stability. This research aims to analyze transcription factors in USP47 by identifying the TFBS that have the most positive regulatory effect on gene expression. pGL3-basic contains luciferase, the light-producing enzyme, and a multiple cloning site (MCS), a small segment of DNA within a vector holding recognition sites for different restriction enzymes. Using directional cloning, primers are designed with MCS enzyme recognition sites to flank the promoter sequence on both 5' ends, and through polymerase chain reactions, the promoter is amplified. A restriction digest then attaches the promoter onto pGL3-basic, where restriction enzymes have opened the MCS. The ligation of the promoter into the vector creates a recombinant plasmid. Deletion constructs are created through site-directed mutagenesis by designing primers that introduce restriction enzyme recognition sites to remove a region of the promoter from the 5' end, thereby identifying the TFBS with the greatest regulatory effect. Cells will be transfected with the recombinant plasmid containing the deletion constructs to measure transcriptional activity, where higher luciferase production indicates TFBS with greater regulatory effect on gene expression. The ability to characterize the transcription factors of this gene can aid the overall understanding of its mechanistic role in various pathologies for future studies.

P204. Identifying positive regulatory elements associated with the FOXR1 promoter, Lindsay Reyes, California State University, Northridge, Faculty mentor: Cindy Malone

In gene expression, DNA is transcribed into RNA to produce proteins. Located upstream of a gene, the promoter region contains the transcription start site and initiates transcription by providing binding sites for positive regulatory elements, including RNA polymerase and transcription factor binding sites, proteins that enhance transcription. The three different types of transcription initiation are focused, dispersed, or mixed and depend on the type of transcription start site pattern. The forkhead box R1 (FOXR1) gene encodes for a transcription factor involved in neurological cell development, leading to brain diseases like Neuroblastoma. The objective is to identify positive regulatory elements of the FOXR1 promoter. The pGL3-Basic vector was used to directionally clone the FOXR1 promoter as it contains a multiple cloning site (MCS) upstream of its reporter gene luciferase, which shows a detectable output in the form of bioluminescence. Primers were designed for the FOXR1 promoter with flanking sequences corresponding to recognition sites on the pGL3-Basic MCS. Using PCR, the FOXR1 promoter was amplified before being digested with the same restriction enzymes as the pGL3-Basic MCS. The resulting sticky

ends were ligated to form the pGL3-Basic FOXR1 promoter recombinant plasmid. Sequential 5' deletions on the FOXR1 promoter will be designed using site directed mutagenesis by introducing restriction enzyme sites to create a series of deletion constructs. All of the constructs will be transfected into mammalian cells to allow for measurable expression of the luciferase gene which reflects transcriptional effectiveness. Obtaining a better understanding of the FOXR1 promoter's transcriptional ability can give insight into the genetic processes behind these health conditions.

P205. Role of TREM2^{+} Macrophage-Derived Exosomes in Chronic Liver Diseases, Siwen Cui, University of California, San Diego, Faculty mentor: Debanjan Dhar

Chronic liver diseases, including metabolic dysfunction–associated steatohepatitis (MASH) and alcohol-associated liver disease (ALD), are major causes of liver-related illness worldwide. These conditions are characterized by excessive lipid accumulation in the liver, hepatocyte injury, inflammation, and progressive fibrosis that can ultimately lead to cirrhosis or hepatocellular carcinoma. Macrophages, a key population of immune cells in the liver, play important roles in regulating inflammation, tissue repair, and communication between different liver cell types during disease progression. Recent studies have identified Triggering Receptor Expressed on Myeloid Cells-2 (TREM2) as an important regulator of macrophage function and liver homeostasis. Loss of Trem2 has been shown to worsen MASH; however, its role in ALD remains poorly understood. Here, we show that the absence of Trem2 exacerbates hepatic inflammation, increases lipid accumulation in hepatocytes, and enhances activation of hepatic stellate cells (HSCs), the primary fibrogenic cells responsible for scar formation in the liver during ALD progression. These findings suggest that TREM2 plays a critical role in macrophage-mediated communication with hepatocytes and HSCs. We further investigate the mechanisms underlying this intercellular communication and demonstrate that macrophage-derived exosomes, small extracellular vesicles that transport proteins, lipids, and RNA between cells, mediate TREM2-dependent signaling during chronic liver disease. These exosomes may serve as important carriers of molecular signals that influence hepatocyte metabolism and HSC activation. By facilitating communication between immune and parenchymal liver cells, TREM2-dependent exosomal signaling may help maintain liver homeostasis during metabolic stress and chronic injury. Understanding this mechanism could reveal previously unrecognized pathways involved in liver disease progression and identify potential therapeutic targets for treating MASH, ALD, and other chronic liver diseases.

P206. Probing autophagy activity in Ras and Myc tumors, Yadanar Khin, Danino Corsis, San Jose State University, Faculty mentor: Bree Grillo-Hill

Cancer is a disease where cellular growth is increased, causing physiological dysfunction. Two of the most mutated genes in human cancers are Ras and Myc. Ras mutations are present in 20% of human cancers, and Myc mutations are present in 70% of human cancers. Cancer cells are under constant stress from altered metabolism and unregulated growth. Autophagy is a catabolic lysosome-dependent process vital for cell health and recycling of cellular building blocks. In autophagy, proteases in lysosomes break down protein aggregates, dysfunctional organelles, and cell membranes into their basic building blocks so that they can be recycled into new tumor cells. Autophagy is dysregulated in cancers, however the mechanism is not understood. Our goal is to compare autophagic activity between Myc-induced tumors, Ras-induced tumors, and healthy tissues. We will use the autophagy marker LysoTracker to label tumors in Drosophila wing imaginal discs and image them using confocal microscopy. We will perform quantitative analysis of these images to determine how levels of LysoTracker are different across our tissue samples. Additionally, we will look for nonautonomous effects on neighboring healthy cells. We hypothesize that Ras tumors will show increased abundance of Lysotracker, suggesting increased autophagy, whereas Myc tumors will show lower levels of LysoTracker. Data from these experiments will determine how tumors with different genetic drivers respond to cellular stresses in different ways, and may suggest distinct therapeutic approaches that are based on the genetic drivers of tumors.

P207. Red but not dead: Investigating Membrane Repair in Yeast During Desiccation, Riley Thorne, California State University, Channel Islands , Faculty mentor: Hugo Tapia

Organisms that withstand extreme water loss must preserve membrane integrity to remain viable. In yeast, trehalose and stress response pathways are thought to protect against desiccation damage, but their role in membrane repair remains unclear. This study examined recovery after rehydration in three Saccharomyces cerevisiae strains: wild type BY4742, a tps1Δ single deletion mutant lacking trehalose synthesis, and a tps1Δ glc3Δ hog1Δ triple deletion mutant impaired in both trehalose accumulation and stress signaling. Cultures were desiccated and stored for 2, 15, 30, or 90 days at 25 °C under 4% relative humidity, then rehydrated to assess recovery. Membrane repair was evaluated using propidium iodide staining to distinguish intact from damaged cells. Initial findings suggest that wild type cells retain greater viability compared to deletion mutants, and that viability decreases with longer storage times, with the triple mutant exhibiting the most severe loss. Future work will expand this analysis to additional relative humidity conditions to determine how environmental factors influence membrane repair. Defining the molecular basis of desiccation tolerance may guide new approaches for stabilizing biologics and preserving cellular materials.

P208. Investigating Genetic Risk Factors for Kidney Disease Using Large-Scale Genomic Datasets, Fiona Chan, University of California, Berkeley, Faculty mentor: Vivek Bhalla

Investigating Genetic Risk Factors for Kidney Disease Using Large-Scale Genomic DatasetsFiona Chan, Molecular and Cell Biology at UC BerkeleyFaculty Mentors: Dr. Vivek Bhalla and Dr. Vivek Charu, Division of Nephrology, Department of Medicine, Stanford University School of Medicine and Department of Pathology, Stanford University School of Medicine Kidney disease affects more than 1 in 7 adults, with many conditions driven by genetic factors. By identifying genetic variants contributing to kidney disease, we can expand early detection and understanding in disease progression. The study's aim is to identify and assess emerging renal genetic variants from a case control study using large genomic datasets and analysis. We draw on two large health research programs, NIH All of Us and UK Biobank, and a genetic testing database from Natera Renasight kidney gene panel. The Natera database has individuals who were referred for genetic testing after being clinically diagnosed with kidney disease, creating a unique population to investigate variants in. From there, we identified candidate genetic variants for deeper investigation. Our evaluation included variant analysis, literature review, and investigation of knockout model findings. Variant quality is also assessed using Sherlock and Integrative Genomics Viewer (IGV). We narrowed down a set of 131 genes with European ancestry to 7 genes for variant analysis, which are ITSN2, CABIN1, GCDH, NOTCH3, MIB1, ERBB4, and MANBA. With these candidate genes, we are investigating loss of function effects of variants and further assessing genetic variant quality. By utilizing large datasets to investigate candidate genes, we can uncover novel genes and facilitate genetic counseling and detection.

P209. Chemical activation of in vitro cardiomyocyte migration: a potential strategy to mammalian heart repair, Xiaopei Chen, University of California, Berkeley, Faculty mentor: Guo Huang

Cardiovascular disease is the leading cause of death in the United States and heart failure has been declared a global pandemic. In myocardial infarction, blocked blood vessels cause extensive cardiomyocyte death due to the human heart's limited regenerative capacity. Injured mammal myocardium is replaced with non-contractile fibrotic scar tissue, leads to deteriorating heart function and chronic heart failure. Model organism such as zebrafish can regenerate hearts throughout lifetime, while the regenerative window for murine animals closes within one week post birth. Previous research on mammalian regeneration focused primarily on cardiomyocyte proliferation, whereas zebrafish studies indicate that successful regeneration also requires mediated and directed cardiomyocyte migration. The role of migration in mammalian cardiomyocyte regeneration remains contentious. Here, we investigate the intrinsic migratory capacity of the neonatal rat cardiomyocytes. Using scratch assays, we find evidence of postnatal day 1 (P1) rat cardiomyocytes active migration in vitro, whereas postnatal day 7 and beyond (P7-P10) rat

cardiomyocytes do not exhibit migratory activity. We inquire if it is possible to reactivate the migratory activity in P7-P10 rat cardiomyocytes by chemical treatment. We found several putative compounds that can reactivate migratory activity of P7-P10 from screening 1,840 FDA-approved small molecule drugs. These findings reveal the developmental dynamics and underlying mechanism of mammalian cardiomyocyte migration, which may provide insight into future therapeutic strategies for enhancing cardiac regeneration and treating heart failure.

P210. Dried and Tried: Yeast Spores, Trehalose, and the Battle Against Desiccation, Ryan Moses, Karen Galvan, California State University, Channel Islands, Faculty mentor: Hugo Tapia

Dried and Tried: Yeast Spores, Trehalose, and the Battle Against DesiccationRyan Moses, Karen Galvan, Joshua Sarmina, Sheila Ferer, Alina Vale, Gaby Amador and Hugo Tapia Water loss typically causes cell death, yet some organisms—including the baker's yeast Saccharomyces cerevisiae - can survive extreme desiccation. A key factor for yeast survival is trehalose, a protective disaccharide thought to help cells enter a glass-like state, a process called vitrification. This theory suggests trehalose stabilizes cellular components and reduces damage during drying and rehydration. In yeast, diploid cells can also undergo meiosis to form spores with tough walls that further enhance desiccation tolerance. Our work tests how trehalose synthesis and sporulation influence survival. We compared Wild-type, heterozygous (TPS1/tps1Δ), and homozygous (tps1Δ/tps1Δ) trehalose mutants. Preliminary results show that wild-type and homozygous deletion strains sporulate, but the heterozygous deletion fails to do so, indicating an unexpected link between trehalose gene dosage and sporulation. Growth rates remain similar under normal conditions, and ongoing tests will determine how trehalose levels and spore formation shape recovery after long-term drying. We will also assess how these strains respond to other environmental stresses—including heat, salt, and freezing—alongside desiccation. Understanding these mechanisms may reveal how eukaryotic cells maintain viability through severe water loss

P211. Investigating the Role of ATR in Chromatin Compaction and Genome Stability, Carla Becerra Sabrera, Yale University, Faculty mentor: Lilian Kabeche

Chromosomal instability (CIN) is a hallmark of many cancers and often arises from defects in mitotic chromosome segregation. ATR (ataxia telangiectasia and Rad3-related) kinase is a central regulator of genome integrity, best known for its role in the DNA damage response. Recent studies suggest that ATR also contributes to proper chromosome segregation during mitosis. However, the mechanisms through which ATR regulates mitotic chromosome architecture remain unclear.The objective of this study was to investigate whether ATR regulates chromatin compaction through modulation of Topoisomerase IIα (TOP2A), an enzyme essential for chromosome organization and decatenation during mitosis.To test this hypothesis, U2OS cells were treated with ATR inhibitors and/or TOP2A inhibitors to examine potential functional interactions between these pathways.

Chromosome spread assays were performed to quantify changes in mitotic chromosome compaction, and western blot analysis was used to evaluate Aurora B activity, a downstream substrate of ATR involved in mitotic regulation.Our analysis revealed that inhibition of ATR, TOP2A, or both resulted in decreased chromosome compaction compared to control conditions. These findings suggest that ATR activity contributes to proper chromatin compaction during mitosis and may act through modulation of TOP2A-related pathways.Together, these results support a previously underappreciated role for ATR in maintaining mitotic chromosome structure and genome stability. Understanding how ATR regulates chromosome compaction may provide insights into mechanisms underlying chromosomal instability in cancer, with potential implications for developing targeted treatments.

P212. Engineering E. coli to Express DP1 for Protection Against Paraquat-Induced Oxidative Stress, Luke Maki, United States Air Force Academy, Faculty mentor: Matthew Lerdahl

New research has prompted a paradigm shift in radiobiology, recognizing that lethal damage correlates primarily with oxidative modification of critical cellular proteins, rather than solely with DNA double-strand breaks. Extreme radioresistance, exemplified by Deinococcus radiodurans, is most likely achieved by accumulating low-molecular-weight -antioxidant complexes which selectively protect the cellular proteome, thereby preserving the function of essential DNA repair machinery. Drawing inspiration from this natural defense, the synthetic decapeptide DEHGTAVMLK (DP1) was designed based on amino acid residues abundant in D. radiodurans extracts. When complexed with Mn2+ and orthophosphate (MDP), this antioxidant system demonstrates extraordinary efficacy, preserving enzyme activity against oxidative damage caused by massive doses of gamma-radiation (up to >60 kGy) and conferring 100% survival against otherwise lethal doses of IR in mic. MDP has also been successfully utilized to preserve antigenic epitopes, enabling the development of radiation-inactivated vaccines. Despite this proven therapeutic potential, external synthesis of or reliance on ultrafiltrates is not a sustainable countermeasure for human application. This project seeks bioengineer Escherichia coli to express DP1 both intracellularly and, critically, extracellularly. Extracellular secretion is being investigated using a synthetic csg operon to harness the native Type VIII secretion pathway of E. coli. Success will be determined by confirming overexpression and demonstrating enhanced resistance in the recombinant E. coli strains when challenged with superoxide-inducing agents like paraquat. Preliminary results indicate successful overexpression of DP1 in E. coli and subsequent enhanced survivability when BL21 (DE3) is supplemented with exogenous MDP. This study sets the critical foundation required to confer effective radiation protection in humans for both civilian and military applications.

P213. Determining whether nociceptive pain-like thresholds vary with weight in larval Manduca sexta, Sathya Correa, San Francisco State University, Faculty mentor: Megumi Fuse

Over 20% of the population suffers from chronic pain, but there are few effective models to study its causes. Manduca sexta is a promising model for nociception (a pain-like response) with a highly reproducible defensive strike as a proxy for pain. The defensive strike is easier to elicit after injury (sensitization), and this response is long-lasting, suggesting that M. Sexta may be used as a model for chronic pain. Previously, studies on sensitization due to noxious stimuli in M. Sexta larvae were normalized for weight, but no research has tackled the correlation between weight and threshold of naive or sensitized animals. We assessed the baseline threshold of naive animals as well as those sensitized to a strong pinch (noxious stimulus) for animals of varying weight, all within the 5th larval stage. In order to sensitize the insect, we pinched the cuticle, and determined their strike threshold, using von Fray filaments following the lab up and down protocol. We describe differences over a 24 hour window, when larvae almost double their weight, and discuss the limitations to our bioassay in the context of weight and developmental changes.

P214. Re-Engineering Bexarotene: EC50 Screening Reveals Novel and Potent RXR Agonists with Therapeutic Promise, Hani Khan, Arizona State University, Faculty mentor: Peter Jurutka

Bexarotene (Bex) is an FDA-approved rexinoid used in the treatment of cutaneous T-cell lymphoma (CTCL) and is also being investigated for potential use in Alzheimer's disease. As a rexinoid, Bex binds to the retinoid-X-receptor (RXR) and modulates transcription of genes involved in cellular differentiation, apoptosis, and cell proliferation. However, to produce the observed clinical anti-cancer effects, Bex is utilized at higher concentrations when administered to patients. Consequently, this drug can cause adverse dose-dependent side effects such as hyperthyroidism, hyperlipidemia and cutaneous toxicity. These side effects are linked to the high drug concentrations required to achieve antitumor efficacy, thus underscoring the need for more potent rexinoid therapeutics with comparable effectiveness. Therefore, we developed several novel analogs with structural similarities to the parent Bex compound and employed an effective half-maximal concentration (EC50) assay to determine analog activity profiles relative to Bex. The EC50 assay was carried out using a dual-luciferase mammalian-2-hybrid assay (M2H) in human embryonic cells (HEK-293), and activity was measured through a quantitation of light emission. To calculate EC50 values, a preliminary 14-point concentration window was utilized with analog concentrations ranging from 1×10^{-9} M to 3×10^{-6} M. Following initial screening, the concentration window was refined as necessary to generate symmetrical EC50 binding curves. EC50 values were then calculated using a nonlinear regression and four parameter logistic curve fitting to determine the concentration required to achieve half-maximal activation for each compound. With refined concentration windows and nonlinear regression analysis, our results revealed that several analogs demonstrated a lower EC50 (higher potency) than Bex. These

included A101, A105, and A113 with EC50 values of 0.3 nM, 8 nM and 5 nM, respectively, versus Bex (10 nM). Overall, these findings support several analogs as potential next-generation CTCL therapeutic candidates, with improved RXR agonist potency relative to Bex.

P215. Characterizing the Phenotypic and Transcriptional Effects of Pitx2 in Threespine Stickleback Fish Teeth, Rebecca Atlas, University of San Francisco, Faculty mentor: Sophie Archambeault

Pitx2 is a gene required for the proper development of multiple tissues in vertebrates, from humans to fish. For example, humans with Pitx2 mutations often develop Axenfeld-Rieger Syndrome, characterized by tooth, eye, and craniofacial abnormalities. In mice, Pitx2 is involved in the proliferation and differentiation of cells that give rise to growth factor- and enamel-secreting cells, without which the developing tooth buds arrest. These findings are consistent with primary and adult tooth abnormalities in humans, although mice do not replace their teeth. To better understand the role of Pitx2 in the development and replacement of teeth, we are using the threespine stickleback fish (Gasterosteus aculeatus), which replace their teeth throughout life, and have fewer primary teeth when Pitx2 is mutated. I am characterizing the phenotypic and transcriptional effects of Pitx2 mutations in threespine stickleback fish teeth. First, we are characterizing the spatiotemporal expression of Pitx2 in paraffin sections of wildtype stickleback teeth using Hybridization Chain Reaction (HCR). I expect Pitx2 expression to mimic patterns seen in other vertebrates, consistent with a conserved function across vertebrates. We found that, similar to those vertebrates, Pitx2 is restricted to the basal lamina and tooth germ epithelium during odontogenesis, with high levels of expression in the inner and outer dental epithelium. Next, we explored the effects of the Pitx2 mutation on the maturation of ameloblasts, the cells that secrete enameloid. We stained for an ameloblast marker, Mmp20b, using HCR in homozygous wildtype, heterozygous, and homozygous mutant fish. We found Mmp20b staining in later stages of wildtype tooth development in the expected time and place of enamaloid secretion, confirming our approach. Preliminary results suggest a decrease in staining in homozygous mutant fish. This work improves our understanding of Pitx2 in stickleback tooth development by determining whether expression and function are conserved from mice to stickleback.

P216. Structural Breakdown of Vascular Scaling in Tumor Angiogenesis, Charissa Mak, University of California, Los Angeles, Faculty mentor: Van Savage

Tumor vasculature exhibits pronounced structural heterogeneity, including irregular branching, tortuosity, and vessel dilation, which undermines efficient perfusion and contributes to hypoxia and therapeutic resistance. In contrast, classical allometric theories such as the West–Brown–Enquist (WBE) framework predict regular scaling relationships between vesselradii and lengths in healthy vascular networks. The extent to which tumor vasculature conforms to or deviates from these theoretical predictions remains insufficiently quantified at the network level.In this study, we leverage Angicart++, a computational framework for extracting vascular graphs from imaging data, to quantify vascular structure in lung, liver, kidney, and skin tumor datasets. Specifically, we compute parent-to-daughter radius (β) and length (γ) scaling ratios, as well as asymmetry metrics, across reconstructed vascular trees from multiple patients. These measurements are analyzed to assess variability across tumor regions and to compare empirical distributions with theoretical predictions from energy-minimization and space-filling vascular models.Preliminary analyses indicate that tumor vessels exhibit systematic deviations from classical scaling expectations, with increased variability in β and γ ratios and higher asymmetry in branching patterns relative to idealized networks. We also identify correlations between local network irregularities and regional differences in vascular density and branching hierarchy. These results provide a quantitative framework for characterizing tumor vascular architecture and highlight the utility of Angicart++ for translating imaging data into biologically interpretable metrics.This pipeline enables scalable analyses across tumor types and imaging modalities, supporting future studies of how vascular geometry influences perfusion efficiency and metabolic heterogeneity. By linking empirical network metrics with theoretical models, this work advances a mechanistic understanding of tumor vascular dysfunction. Importantly, the extracted scaling and asymmetry metrics provide structured, physically interpretable features for theory-informed machine learning models, enabling data-driven prediction of vascular function while preserving constraints from vascular optimization theory.

P217. Beyond Mere Muscle: Handgrip Strength as a Marker for Biological Aging, Petri Alva, Sonoma State University, Faculty mentor: Bulent Sokmen

Purpose: Muscular strength and cardiorespiratory fitness are key predictors of health, functional independence, and longevity. While lower-body strength and maximal oxygen uptake are well established biomarkers, the role of handgrip strength as an indicator of biological aging remains less defined. Emerging evidence suggests that handgrip strength may reflect not only muscular function but also cognitive and neurological health. The purpose of this study is to evaluate whether handgrip strength is associated with key indicators of physical and cognitive function and to determine its utility as a practical biomarker of biological aging. We hypothesize that handgrip

strength will be positively associated with these variables and may serve as a practical marker of overall physiological function.
Methodology: This study aims to examine the relationship between handgrip strength and measures of muscular strength, muscular endurance, cardiovascular fitness, and cognitive function in adults aged 40 to 100 years. Participants completed a single testing session including assessments of handgrip strength, cognitive function using the Montreal Cognitive Assessment, cardiovascular fitness via the 6-minute walk test, lower-body endurance using the 30-second sit-to-stand test, pulmonary function, and isokinetic muscular strength.
Results and Discussion: This study seeks to further evaluate handgrip strength as an accessible biomarker of biological aging and functional capacity.

P218. Beyond Mere Muscle: Handgrip Strength as a Marker for Biological Aging, Nate Cohen, Zach Jones, Sonoma State University, Faculty mentor: Bulent Sokmen

Purpose: Neuromuscular electrical stimulation (NMES) and resistance exercise modalities such as concentric and eccentric contractions have been commonly used to enhance muscle performance and neuromuscular function. However, their combined effects on muscle contractile history (PAP), peak force, and rate of force development remained unclear. The purpose of this study was to examine how NMES, as well as concentric and eccentric contractions of agonist and antagonist muscle groups, influenced PAP, muscular strength, power, and electromyographic activity.
Methodology: Healthy resistance-trained adults aged 18 to 44 years participated in a repeated-measures design involving six laboratory visits over 4 to 6 weeks. Following baseline assessments, participants completed experimental sessions that included NMES application and targeted concentric and eccentric contraction protocols. Peak force and rate of force development were assessed during isometric knee extension using an isokinetic dynamometer, while muscle activation was measured via surface electromyography.
Results and Discussion: This study aimed to provide insight into how different neuromuscular interventions influenced muscle performance and activation, contributing to improved training and rehabilitation strategies.

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