Wyoming INBRE Fall Research Network Retreat



October 15, 2021 (1-5PM)

Join this ZOOM meeting: https://uwyo.zoom.us/j/99843971247

Presentation Schedule

- **1:00-1:15PM**: Welcome remarks. Wyoming INBRE Director/PI: Prof. R. Scott Seville (University of Wyoming)
- **1:15-1:30PM**: Environmental Tolerance Limits of Soil Algae and Fungi. <u>Daniel Casper, Joshua</u> Clayton and Christopher Wenzel (Eastern Wyoming College).
- **1:30-1:45PM**: Evolution of a Dune Specialist Bee Using Genomics. <u>Madi Talley</u>, <u>Quincy</u> Stewart and David Tanner (Western Wyoming Community College).
- **1:45-2:00PM**: Identification of Binding Determinants of PopZ Binding Partners. <u>Thomas Westmoreland</u> and Joshua Holmes (Western Wyoming Community College)
- 2:00-2:15PM: Using Site-Directed Mutagenesis to Investigate the Impact of Defective Citrullination on Microtubule Polymerization and Transport Function. <u>Caitlyn Edwards</u>, Heather Rothfuss, Brian Cherrington, and Florence Teulé-Finley (University of Wyoming at Casper and University of Wyoming-Laramie)
- **2:15-2:30PM**: Title TBD. Mindy McIntosh and Eric Mechalke (University of Wyoming at Casper and Casper College)
- **2:30-2:45PM**: Impact of Wyoming Environment on Oncogenic Protein Production. <u>Jimmy Bautista</u>, <u>Hayley Brown</u>, <u>Tyler Coffman</u>, and Lucy Graham (Central Wyoming College-Riverton)
- **2:45-3:00PM**: Microplastic Science in Wyoming's Remote Dinwoody Cirque. <u>Madison Worthy</u>, Anne Dalton, and Kirsten Kapp (Central Wyoming College-Jackson)
- **3:00-3:15PM**: BIKES: 1000 Miles of Air Quality Research on Two Wheels. <u>Ryan Towne</u>, <u>Alex Minge</u>, <u>Aidan Hereford</u>, and Jacki Klancher (Central Wyoming College)
- **3:15-3:30PM**: You Would Not Believe Your Eyes: Using Predictive Modeling to Estimate Locations of Firefly Populations in the Intermountain West. Ryen Nielsen, Haleigh Vahlbusch, Christopher Hair, and Gavin Martin (Laramie County Community College)
- **3:30-3:45PM**: Three Sisters Gardening, Researching the Life Sustainers. <u>Chelsea-Victoria</u> Turner (Northern Wyoming Community College District/Sheridan College)
- **3:45-4:00PM**: My INBRE Adventure: Tools for Success. Milissa Denevan and Eric Atkinson (Northwest College)
- **4:00-5:00PM**: Wyoming INBRE Program: Core updates and announcements (opportunities for events and funding)

Presentation abstracts

(listing by presentation order)

Environmental Tolerance Limits of Soil Algae and Fungi.

<u>Daniel Casper, Joshua Clayton</u> and Christopher Wenzel Eastern Wyoming College, 3200 West C Street Torrington, WY 82240

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Abstract: A key factor for terrestrial microbial survival is water availability, which is strongly influenced by temperature and light intensity. While under water stress conditions, desiccation may lead to strong cellular dehydration followed by the formation of water-holding structures such as thick extracellular mucilage layers formed from excretion of exopolymeric substances (EPS) leading to a protective layer know as a biofilm. The effects of factors influencing cellular desiccation and subsequent protection on algal and fungal growth have not yet been resolved. The goal of this study is to determine the effects of natural variations (high, moderate and low) of light, temperature and moisture on eukaryotic microbial (fungal and algal) biomass. We hypothesize modified levels of temperature, light intensity, and moisture will result in changes in algal and fungal biomass as determined by specific gene quantities and biomarkers associated with cell membrane structure. Soil biocrust samples were collected from late May to early June, 2021, DNA is being extracted from subsamples by using Nucleospin Soil DNA Extraction Kits (Macherey Nagel, Bethlehem, PA, USA) according to the manufacturer's protocol. DNA concentration is being determined using a NanoDrop One UV-VIS Spectrophotometer (ThermoFisher Scientific, Waltham, MA, USA). Extracted DNA will be examined for presence of target algal genes (i.e., rcbl, tufA, ITS, and 18S) using PCR prior to Real Time (RT-qPCR). Future RT-qPCR will be performed using a ChaiBio Open Real Time PCR System (CHAI, Santa Clara, CA). In addition, a "soil-on-a-chip" micro-fluidic device platform will be used in the study.

Evolution of a Dune Specialist Bee Using Genomics.

<u>Madi Talley, Quincy Stewart</u> and David Tanner Western Wyoming Community College, 2500 College Drive, Rock Springs, WY 82901

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Abstract: The Killpecker Dune system of southwestern Wyoming is the largest system of transient dunes in the continental United States, and like many other arid regions has been significantly impacted by grazing, mineral extraction, and recreational activities. Faculty at Western Wyoming Community College are cataloging the extant biological diversity of this unique dune system. As a part of these efforts, we are mapping the genomes of dune-specialist pollinators. The first step in this process is to identify a protocol that will maximize per sample yield of long strand DNA. We have compared the extraction efficiency of DNA using a thermomixer or a water bath, and using either a whole insect or an isolated thorax. Our preliminary results suggest that protocols using a shaker are marginally more effective than using a water bath, and that there is no discernable difference between using a whole insect or an isolated thorax. Our next step will be to compare the length of DNA fragments between extraction protocols.

Identification of Binding Determinants of PopZ Binding Partners.

<u>Thomas Westmoreland</u> and Joshua Holmes Western Wyoming Community College, 2500 College Drive, Rock Springs, WY 82901

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Abstract: Polar organizing protein Z (PopZ) has been shown to interact with over 10 different binding partners. This is odd for such a small protein of 177 amino acids. It has been shown to facilitate these interactions being an intrinsically disordered protein. This is a type of protein that natively has no structure and only adopts one in response to environmental cues or protein-protein interactions. Two of the PopZ binding partners (DivL and CckA) have had small truncations to break the interaction of PopZ. This work will expand upon these two truncations. Our first goal is to determine if these truncated regions alone are sufficient for interaction with PopZ. The second goal is to work to identify if there are any sharded traits between these two regions and the other PopZ binding partners. If there is, this would suggest a universal binding determinant. This work could provide novel insight into intrinsic disordered protein-protein interactions.

Using Site-Directed Mutagenesis to Investigate the Impact of Defective Citrullination on Microtubule Polymerization and Transport Function.

<u>Caitlyn Edwards</u>,¹ Heather Rothfuss,² Brian Cherrington,² and Florence Teulé-Finley¹ University of Wyoming at Casper, 125 College Dr., Casper, WY 82601 ²University of Wyoming, Dept. Zoology and Physiology, 1000 E. University Avenue, Laramie, WY 82071

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Abstract: Microtubules are an essential component of the cytoskeleton composed of α - and β -tubulin heterodimers. They are involved in numerous cell processes including cell division, morphogenesis, and intracellular transport with motor proteins. To support this variety of cellular functions, tubulin proteins are prone to posttranslational modifications such as citrullination. Citrullination occurs when calcium-mediated peptidylarginine deaminase (PAD) enzymes change the side chain of arginine, converting it to the non-standard amino acid citrulline. This process has been shown to be crucial in biological processes such as cell differentiation, nerve growth, embryonic development, cell death, and gene regulation. When citrullination patterns are disrupted pathologically, a wide range of diseases can result.

This project investigates the effect of citrullination on microtubule polymerization and kinesin binding affinity. To accomplish this, different mutant human tubulin genes will be engineered through cloning and site-directed mutagenesis, replacing specific arginine targets with lysine residues which cannot be citrullinated. These mutagenized human tubulin genes will be cloned into an insect cell expression vector and transfected into insect cells for production of mutant tubulin proteins to evaluate the impact of citrullination on microtubule polymerization and kinesin binding affinity.

Proteoglycans and the Oyster Mushroom.

Mindy McIntosh¹ and Eric Mechalke² University of Wyoming at Casper¹ and Casper College², 125 College Dr., Casper, WY 82601

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Abstract: The fungal cell wall is made up almost exclusively of bioactive molecules that are not found in the human body. It is these molecules that trigger our immune defenses. The polysaccharides that produce the medicinal properties found in mushrooms compose 90% of the cell wall. The cell wall is a tough, fibrous macromolecular complex network of a chitin fibrillar skeleton that is alkali-insoluble and an amorphous solid that is alkali-soluble. Extracting polysaccharides requires breaking the outer layer of the wall first and moving inward using pH and temperature control in order to cause a weak-to-strong extraction environment. Materials needed for this vary, depending on their solubility and structure. It should be noted there are many extraction "recipes" that use many different chemicals and methods. This provides a gateway to creating new methods for extracting the polysaccharide.

Producing healthy mushrooms that contain high levels of polysaccharides depends on the growing environment. Mold and bacteria are a constant threat against mushrooms and special precautions need to be taken to avoid pathogens when growing them. It is imperative to keep a clean and sterile growing environment if one is to grow healthy, happy mushrooms.

Impact of Wyoming Environment on Oncogenic Protein Production.

<u>Jimmy Bautista</u>, <u>Hayley Brown</u>, <u>Tyler Coffman</u>, and Lucy Graham Central Wyoming College, 2660 Peck Avenue, Riverton, WY 82501

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Abstract: The environment can have a profound impact on physiological processes, including the initiating of oncogenic processes. Asbestos, UV radiation, and cigarette smoke are some examples of environmental triggers that have been well studied as carcinogens. The Wyoming environment is of particular interest, as there are some characteristics that have not been well-studied in regards to the development of cancer. Forest fire smoke exposure is especially noteworthy, as it has been named a potential carcinogen, but studies have not elucidated this fully. Both the composition of forest fire smoke and the effect on firefighter health have been explored, but incidental exposure of the public and the subsequent health impacts have not been well researched. As forest fires become more prevalent in the American West, the potential inflammatory and carcinogenic effects of the smoke on the affected public should be more robustly investigated. To address this, we aim to explore the impact of forest fire smoke on lung cells by measuring oncogenic protein expression after exposure to smoke.

Microplastic Science in Wyoming's Remote Dinwoody Cirque.

Madison Worthy, Anne Dalton, and Kirsten Kapp Central Wyoming College, 240 S. Glenwood St. #124, P.O. Box 4795, Jackson, WY 83001

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Abstract: Microplastic pollution in alpine ecosystems is an emerging scientific field with potentially high adverse impacts on human and environmental health. In July 2021, a team of student researchers from Central Wyoming College collected surface and core snow samples to study the presence of microplastics particles (under 5mm in length) in remote alpine areas of the Wind River Range in Wyoming. Three sites were selected across the Dinwoody Cirque, consisting of two permanent snowfields with heavy recreational use, and one snow field in a less-traveled area. In total, 23 samples were collected including 18 surface samples and five vertical column samples. Microplastic research is still developing, with no standardized sampling protocols. Here we present our methodology for collecting snow samples in remote wilderness areas, and highlight the unique challenges of sampling in dynamic, alpine environments. While the samples have not yet been analyzed, results will be incorporated into a larger regional study on microplastic pollution that will continue this winter. Recent studies indicate the presence of microplastics in similar alpine ecosystems including the French Pyrénées and the Italian Alps. The Dinwoody Cirque is located within the remote Fitzpatrick Wilderness area and is home to Gannett Peak, the highest mountain in Wyoming. Each year, a large number of recreationists traverse this area of the Wind River Range. The presence, or absence, of microplastics in samples from this area will inform how widespread microplastics are in the Greater Yellowstone Ecosystem and whether human activity is a leading contributor to microplastic pollution.

BIKES: 1000 Miles of Air Quality Research on Two Wheels.

Ryan Towne, Alex Minge, Aidan Hereford, and Jacki Klancher Central Wyoming College, 120 Enterprise Blvd Lander, WY 82520

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Abstract: The negative impacts of anthropogenic pollution and wildfires on both human and environmental health have become increasingly evident. During the 2020-2021 fire years, particulate matter and other air quality parameters were identified as exacerbating factors for respiratory conditions such as asthma, COPD, and allergies; linked to increased rates of morbidity from COVID-19; and correlated with increased atmospheric transport of black carbon particulate matter. In June 2020, the CWC BIKES research team deployed portable air quality meters and launched a six week expedition to assess air quality along a 1000 mile stretch of mixed remote Wilderness, rural, and urban environments from Eureka, MT to Atlantic City WY. A portable air quality meter was employed for approximately 8 hours per day to measure particulate matter from 1.0um to 10.0um, Volatile Organic Compounds (VOC's), and Nitrogen Dioxide (NO₂). This research addressed two key research questions: 1. Are low cost portable air quality sensors appropriate for scientific research and 2. What trends in air quality are revealed by the data? It was hypothesized that there would be measurable quantitative differences between air quality in rural and remote areas vs urban areas and that overall air quality would be negatively impacted in areas with forest fire activity. Preliminary assessment of the value of the air quality technology used for this research is positive, but with concerns surrounding both accuracy and precision. Early data analysis suggests that traffic, wildfires and proximity to urban areas can be correlated with poor air quality even in remote environments.

You Would Not Believe Your Eyes: Using Predictive Modeling to Estimate Locations of Firefly Populations in the Intermountain West.

Ryen Nielsen, Haleigh Vahlbusch, Christopher Hair, and Gavin Martin

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Abstract: Lampyridae populations and distribution have been extensively documented in the Eastern United States, but less is known about their western counterparts. Currently, our knowledge of Lampyridae distributions in the region relies on the contributions of citizenscientists who report their sightings to organizations like the Western Firefly Project, NMHU which tracks Lampyridae populations across Utah. While the Western Firefly Project, NMHU is primarily Utah specific, their data can predict populations in surrounding states through the creation of a model that utilizes derived geographic distribution and environmental conditions. We created such a model and used it to determine areas that had probable Lampyridae locations with a specific focus on Wyoming.

Three Sisters Gardening, Researching the Life Sustainers.

Chelsea-Victoria Turner and Ami Erickson

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Abstract: The three sisters gardening is a commonly utilized traditional Native American practice. Not only does planting in this format allow each crop to benefit one another, but the food produced provides a balanced and nutritional diet. Four versions of the three sisters gardening were observed along with one control. The four versions were as follows: a traditional version using circular mounds and the center of planting, a variation of the traditional circular planting, a circle of life formation, and a series of alternating rows. The control crops were planted with spacings between each crop variety. Production value was analyzed based on crop production per plant. Each plant was given a rank following the final harvest count as well as a rank based on chlorophyll fluorescence. An overall rank was calculated based on all the ranks a plot had previously received and the best producers were determined from their ending rank. The control had the lowest rank followed by the alternating lines, which ended up being viewed as another version of a control. The three plots that did the best were the versions that incorporated the three sisters so that they were growing with one another and thriving together.

My INBRE Adventure: Tools for Success.

Milissa Denevan and Eric Atkinson Northwest College, 231 West 6th Street, Powell, WY 82435

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Abstract: INBRE has been a way to learn applicable skills and knowledge that are essentially tools to help me further my career. I am a non-traditional student at Northwest College majoring in Natural Resource Biology.

During my first semester, spring 2021, I joined two student groups who were researching DNA extraction from small mammal feces searching for potential zoonotic diseases. This was my first experience with this type of lab work. I learned how to extract DNA in the lab as well as how to read metabarcodes using the QIIME2 pipeline. The lab work was a result of teamwork, the beginning of a network of peers and scientist that I would become very familiar with. Summer 2021 I interned with Professor Atkinson banding songbirds. We took morphometrics on all the birds we caught in the mist nets, as well as a saliva sample to test for West Nile Virus (WNV), a blood sample to test for antibodies and a blood smear for a malaria test. Surprisingly, we found that the summer 2021 WNV prevalence was low as shown by ELISA(RAMP) tests. INBRE has provided me with the tools I need to move forward in my career. I have many ideas for further research, such as looking for prevalence of wild type endophytes in tall fescue or exploring the prion disease CWD. After graduating, I would like to work for Wyoming Game and Fish or a similar agency. I have also considered continuing my education, as I would like to eventually get a bachelor's degree in education. Then I can pass the tools I have learned from INBRE to the students I will teach.

Wyoming INBRE Program: Core updates and announcements (opportunities for events and funding) by the Wyoming INBRE administrative team:

- Dr. Scott Seville, PI/Director (sseville@uwyo.edu)
- Dr. Florence Teulé-Finley, Program Coordinator (fteule@uwyo.edu)
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