STRATEGIC INITIATIVE
Undergraduate Research Funding for STEM Majors at the University of Idaho
FINAL PROJECT REPORT

Submitted to:
Higher Education Research Council
Idaho State Board of Education
P.O. Box 83720
Boise, Idaho 83720-0037

Submitted by:
University of Idaho Office of Undergraduate Research
875 Perimeter Drive
Moscow, ID 83844
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Table of Contents

Appendix 1: Copies of ICUR Posters for SBoE supported SURF Students

Appendix 2: Copies of Posters for OUR supported Summer Students

Executive Summary

Undergraduate research is recognized as a high-impact educational practice that increases the rates of student retention and engagement. At the University of Idaho, it is practiced throughout all units on campus, and it is centrally placed in the institution's strategic plan. The Office of Undergraduate Research (OUR) is taking the lead in enabling research opportunities for undergraduates at UI as it manages various competitive student grant programs that directly support student research.

During AY 2022-23, generous funding from the State Board of Education (SBoE) permitted UI to continue its Summer Undergraduate Research Fellowship (SURF) Program. This intensive 10-week summer research experience actively engages undergraduates in faculty-mentored, independent research. Over the course of the program, students are mentored toward increased independence on their projects. Each student is provided with a $5,000 stipend in the form of a fellowship which allows them to devote full time effort to their projects. Each student is also provided with $1,000 to help offset materials and supplies and other project-related expenses. Selection of student participants is a competitive process in which students submit research proposals to the Office of Undergraduate Research and the Director utilizes faculty affiliates in each college to review the proposals and provide selections. The Director then makes sure that projects all well-represented across colleges. It is important to note that not all projects this cycle required a supply budget. This allowed for us to fund additional students with SBoE funds. For FY 2022-23, the SBoE funding supported **12 SURF** awards. All these students supported by SBoE funds attended and presented the results of their projects at the 2023 Virtual Idaho Conference on Undergraduate Research in July of 2023. These students will also be required to present their results at the UI Undergraduate Research Symposium on April 22, 2024.

Additionally, due to partnerships with programs such as Idaho INBRE as well as various departments on campus, the OUR was able to support **an additional 4 Summer Student projects** that, while outside of STEM disciplined, provided excellent research experiences to these undergraduates. These students were also required to present at the virtual ICUR 2023 and summaries of their projects and a few of their posters are also provided in this report.

End of project feedback from students and their mentors was overwhelmingly positive. Significantly, none of the undergraduate research projects described here would have been possible without the support provided by the State Board of Education. We sincerely thank the Higher Education Research Council and the Idaho State Board of Education for making these experiences possible for our students.

Sincerely,

Kristopher V. Waynant
Director, Office of Undergraduate Research
Associate Professor, Chemistry
kwaynant@uidaho.edu

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – SURF 2023

Fellowship Recipient: **Dillon West**
Faculty Mentor: *Nathan Schiele, Biological Engineering*
Project Title: *Investigating the Effects of TGF-βR2 Inhibition in Mesenchymal Stem Cells*

Abstract: Tendons transfer forces from muscle to bones and are vital in joint movement and injury prevention. However, they have poor self-regeneration capabilities and current rehabilitations are unable to restore damaged tendons to their prior strength. Tissue engineering using mesenchymal stem cells (MSCs) is widely being investigated as a potential means of functional tendon replacement or repair. A current obstacle in this field is the lack of thorough understanding of how to direct MSCs differentiation into specialized tendon cells and control functional tendon formation. The cytokine TGF-β is known to play a key role in this, though its specific mechanisms and signal propagation require further investigation. This research will examine the effects of inhibiting TGF-β type-II receptor to create a better understanding of its signaling pathways within the cell and its role in the production of functional tendon markers. The outcomes of this research will enhance the field of tendon tissue engineering by exposing potential pathways responsible for functional tendon marker creation, allowing for more informed manipulation of MSCs in future studies and regenerative therapies.

Objectives: Obj. 1: Inhibit TGF-βR2 via the chemical Morin and identify impacts on LOX and downstream signaling. **Obj. 2**: Inhibit TGF-βR2 via RNA interference and identify impacts on LOX and downstream signaling.

Results: Morin Inhibition: Initial experiments utilizing morin as a chemical inhibitor required a few iterations as high concentrations of DMSO resulted in cytotoxic effects on the cell cultures (Fig.1A). The first solution of morin in DMSO at 50 μM prepared resulted in a concentration of about 10% DMSO in cell media, which had cytotoxic effects on the cells. The second attempt of preparing a suitable solution resulted in a 2% concentration of DMSO in media, however cytotoxicity was still present. The third attempt allowed a 0.5% concentration and had negligible effects on the cells. However, treatment and morin control groups showed little to no difference in cell morphology, which indicated ineffective inhibition. Western blot analysis further proved morin as an ineffective choice for chemical inhibition of TGF-βR2.

RNA Interference Inhibition: Fluorescent control indicated only the 10 nM concentrations of the constructs had successful transfection into the cells, which led to the 0.1 and 1 nM concentration groups being disregarded. The siRNA construct S1 showed the most effective inhibition based on cell morphology, and its LOX protein band was lighter compared to the negative control construct when evaluated with a western blot, which indicates that TGF-βR2 does play a role in the production of LOX.

Discussion & Future Direction: Morin appears to be an ineffectual choice for inhibition of TGF-βR2 based on the experiments conducted here and the limited literature supporting it. Once the DMSO cytotoxicity was no longer an obstruction, qualitative analysis indicated no substantial difference between morin-treated cell groups and its controls. RNA interference, on the other hand, was able to effectively inhibit TGF-βR2, which negatively impacted LOX levels. This indicates that TGF-βR2 may be crucial in the production of LOX. However, this experiment so far is only n=1, and therefore needs to have further technical replicates ran to validate the initial findings. In the future, the protocol for siRNA inhibition will be ironed out, and the experiments will be repeated to ensure consistency of outcomes.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks. Spent $538 on supplies.

Acknowledgement: This student's poster is included in Appendix 1

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – SURF 2023

Fellowship Recipient: **Lacey Hunt**
Faculty Mentor: *John Shovic, Electrical Engineering*
Project Title: *Precision Agriculture Adoption and Integration Case Study*

Abstract: Remote sensing systems for precision agriculture allow for technology and automation to improve the profitability and sustainability of modern farms. The adoption of these systems in the United States has been slow, due to the high cost and difficulty using them. The University of Idaho, in collaboration with Laurel Grove Wine Farm in Winchester, Virginia, has been in the process of developing a remote sensing system designed to be low-cost and flexible in its agricultural applications. This new system lacks deterministic testing results for (1) reliability, (2) overall cost, and (3) usability by farmers. This research project aims to test the system's performance in a new application domain: a heritage apple orchard at Sandpoint Organic Agriculture Center. This proposal outlines the tasks related to testing the performance, cost, and user-interaction of the remote sensing system to determine its viability in furthering the usage of precision agriculture in the United States.

Objective: Assembly and Installation of SCARECRO
The researcher installed a fully functioning instance of the SCARECRO system into SOAC consisting of two WeatherRack stations, two Datagators, two Gateways (one for use in the orchard, the other stored locally for testing purposes), and one Middle Agent. A new database instance was also established, as well as physical building of hardware supports in the orchard. An explanation of SCARECRO's operational architecture is necessary for later exception and fault troubleshooting explanations. The WeatherRacks collect environmental data such as temperature, humidity, rainfall, wind speed and direction, and sunlight. These sensor readings are sent to the Gateway via a wired connection, Bluetooth, or 433MHz radio waves depending on the type of sensor. The Datagators collect readings from sensors that are located farther from the Gateway (in this case, soil moisture sensors) and transmit this data to the Gateway via MQTT through a local Wi-Fi Hotspot. The Gateway is responsible for eliminating duplicate messages, keeping track of sensor connections, and sending data to the Middle Agent. The Gateway sends data to the Middle Agent, a cloud computer which handles all data transmission between the Gateway and the database, via MQTT through a Wi-Fi connection to AirVandal Gold. The database stores all collected data for the SOAC installation, which can be retrieved via database queries for display on a dashboard website or used in AI research applications. All components are solar-powered, so South-facing solar supports were also built into each component stand.

Conclusions: This case study demonstrated successful implementation of the SCARECRO system into an entirely new application domain. This system, even after unprecedented obstacles, proved this specific data collection method to be more cost-effective and robust than its commercially available competitors. Currently, despite this system being not quite ready for widespread deployment, is on the right track to be a viable option for a wide variety of agricultural settings looking to incorporate important precision agriculture methods into their work. To further develop the SCARECRO system towards widespread availability, more work must be completed. Some future projects that would aid in this process include other test sites, incorporation of the SOAC horticulturist sensor suggestions, research into the cause of the periodic disconnection from the network, and further troubleshooting of Datagators on site. A new test site (piloted with the locally assembled test Gateway) would allow further conclusions and improvements to be made regarding the implementation of this system in a different agricultural application.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks. A series of electrical equipment and data sensors were also purchased for this project totalling $854.

Acknowledgement: Poster can be found in Appendix 1

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – SURF 2023

Fellowship Recipient: **Daniel Blanchette**
Faculty Mentor: *Diana Mitchell, Biological Sciences*
Project Title: *Segmentation analyses to identify and quantify microglia in 3D image stacks*

Abstract: Using time-lapse confocal microscopy to record and observe microglia behavior in living zebrafish embryos, the Mitchell lab investigates the molecular basis of dynamic migration and phagocytic behavior of microglia in the central nervous system. Microglial cells regulate brain development, maintain neuronal networks, and repair neural injuries. Timelapse imaging provides crucial insight into the behavior of these cells. Other methods, such as manual cell counting, are subject to human bias and are tedious, repetitive, and time-consuming. The Long computer science lab utilizes Python and open-source modules to develop an automated pipeline as a programmatic solution. These methods aim to expedite and optimize the lab's manual processes. This research project seeks to segment microglia when applied to 3D time series image stacks. Computer segmentation of microglia imaging relies on pixel values to generate bitmasks. Microglia segmentation is challenging because the cell's irregular shape and size can be obscured, resulting in omitted pixel intensity values. Two programs were developed to test the viability of Otsu, multi-Otsu, and Yen automatic thresholding[2]. Both methods separate the raw image into two layers: the foreground and background. Numerical data is generated via histograms to predict optimal threshold values for each algorithm. Prototype implementations of these programs demonstrate viability for microglia counting. However, misclassification of microglia occurs when the predicted pixel threshold is outside the histogram's range. Further refinement of these programs is crucial as they will be foundational for future object classification methods and microglia tracking, optimizing the lab's data processing capabilities and time.

Objectives and Results: The prototype program takes an entire directory tree (folders and subfolders) from the researcher's computer after they adjust the path for their image data. Users also designate an output folder path(to the desktop or otherwise) for storing the results. The thresholding methods of the program are applied to each image in the video frame subfolders. For debugging and lab conformation, a .csv file is generated with data for each frame, z stack position, cell counts, what branched value was used for the threshold, and the threshold value. To help with misclassification, other algorithms can be used to separate the objects. Canny edge detection may be a viable way to distinguish cells close to one another. The overall goal is to ensure separation in all frames of the z-stack to render microglia in 3D space accurately. However, misclassification is not a deal breaker for the lab as their metrics include margins of error for the traditional method of manual counting. While the program being off by one on the cell count may not impact data collection. However, cases where the program counts are greater than or equal to ten cells, may risk skewing the results to the detriment of the lab. One performed better than the other for these programs(Otsu and Yen). Key findings suggest that while both suffered from object misclassification, Otsu and Multi-Otsu were better at setting thresholds to include pixel values closely related. On the other hand, the Multi-Otsu and Yen program performed better when there were high differences between pixel intensity values. Based on the preliminary data, Multi-Otsu and Otsu for the branching logic was the better choice. As segmentation is the foundation for all tracking methods to follow, it is paramount to continue to refine the current program and iterate upon it until a viable cell count is produced. The next step is to classify these 3D objects' shapes for the microglia and upload them to a database. This step will be daunting due to the permutations of known microglia shapes and others that require further observational research via recordings. Once these pieces are in place, tracking methods utilize machine learning and Artificial Intelligence(A.I.) to predict better what composes microglia from the rest of an image.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks.

Acknowledgement: This student's poster can be found in Appendix 1

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – SURF 2023

Fellowship Recipient: **Destinee Ditton**
Faculty Mentor: *Nathan Schiele, Biological Engineering*
Project Title: *Visualization of Tendon Organization of Rat Tails and Hindlimbs in 3D Using Micro-CT*

Abstract: Tendons are connective, collagenous tissues that transmit mechanical forces to allow for body movement, but tendons injure easily and have poor healing capabilities. New treatment methods are needed, requiring a better understanding of multiscale structure-function relationships through comprehensive imaging. Micro-computed tomography (CT) is an imaging modality that produces three-dimensional (3D), high-resolution images, but is limited to high density objects as the x-rays render low-density soft tissues transparent. To address this issue, we explored a contrast-enhanced technique using a serial dehydration in ethanol and employing a chemical stain, specifically phosphotungstic acid (PTA), to image tendon using 3D micro-CT imaging. Utilizing this new contrast enhancing method, 3D images from tissues initially were analyzed and processed to identify and trace tendon fascicles and analyze the anatomical organization of immature (postnatal day 21) rat tails and hind limbs. From this analysis, we found that tail tendon fascicles appeared to cross between fascicle bundles, whereas it was previously assumed that the fascicle bundles maintain specific tracks within the rat tail tendons. Based on these initial findings, the objective of this study was to visualize the tendons in the tails and hind limbs of both younger and older, sexually mature rats to further validate our imaging technique and evaluate fascicle organization.

Results: The dehydrated and PTA-stained P21 and P50 tails and hindlimbs have clearly visible tendons and other soft tissues, providing additional evidence that this staining and imaging technique is applicable to younger and older (sexually mature P50) rats. Even with their much larger size and increased tissue density with age, PTA staining appears to be effective and penetrated the tissues and enhanced soft tissue contrast. Tendon fascicle organization was examined in both P21 and P50 tails. When viewing the P21 tails and moving from a more proximal to distal location, individual tendon sub-fascicles appear to move from one fascicle bundle to a neighboring fascicle bundle. However, the P50 tails appear to have tendon fascicles that divide into smaller subfascicles, moving from a more proximal to distal location. Tendon insertions in P50 tissue appear to follow a specific pattern of outer fascicle bundles moving to insert at the proximal side of the tail vertebra. Overall, validation of this imaging technique in both young and mature rats has allowed for tracking changes in tendon organization and structure with development. Coupling this contrast-enhancing technique with micro-CT scanning enables clear visualization of tendon and its organization and insertion sites, which advances future studies to determine tendon structure and organization in situ. Ultimately, better understanding of tendon structure-function relationships, including how they transmit mechanical forces, will improve treatment methods for restoring tendon function after an injury.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks.

Acknowledgement: This student's poster can be found in Appendix 1

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – SURF 2023

Fellowship Recipient: **William Auten**
Faculty Mentor: *Kristopher Waynant, Chemistry*
Project Title: *Design, synthesis, and redesign of therapeutic peptides using SPPS, native chemical ligation, and AutoDock Crankpep*

Abstract: The use of molecular modeling is quickly becoming an essential tool for the experimentalist. In this project, the molecular docking software AutoDock CrankPep (ADCP) will be utilized to analyze a variety of parameters on peptides to determine which parameters are the largest contributors in producing active compounds when synthesized experimentally and give higher accuracy to the computational results. Our hypothesis is that ADCP parameters (whose exactness we are trying to figure out) can be manipulated through inspiration from wet lab data to give scores more like real world values. This will then lead to faster, more efficient methods for the discovery of peptide-based drug/biologically active compounds.

Objectives: Protein-peptide interactions are among the most important interactions in living organisms. Peptides are essentially keys, or activators, that unlock specified functions of proteins. Many of these peptides are hormones. Proteins take on a variety of functions in cells, from structure to transport, from activating cell growth to causing cell death. For this project we will focus on SUMO1 (small ubiquitin-like modifier 1). SUMO1 is a post-translational modifier and is interesting due to its potential in fighting cancerous cell growth. SUMO1 is a well-studied protein and has known peptidic ligands. These peptides are of interest due to their potential therapeutic use. The benefit of a natural peptide as a drug is that peptide production could be induced through genetic therapies. Peptides are notoriously poor drugs when administered orally as most of our digestive system works to destroy peptides before they can enter the lumen. By identifying new natural peptides that bind to SUMO1 and inhibit cell growth, the project can then become a feedback loop that builds from the modeling and learns from the binding experiments to build a better peptide drug. Modeling is a very useful tool that can be used to reduce the amount of work for experimenters. AutoDock CrankPep has been praised for its efficiency and its ability to dock peptides with more residues than other docking methods. ADCP is a *de novo* method. This means that the conformation of the peptide is determined while docking (2). ADCP manages to be less computationally taxing and runs simulations faster than other *de novo* methods.

Conclusions: While this project is still ongoing, there are several advancements that were made over the summer, including gaining access and familiarity with the operation and maintenance of an Isothermal titration calorimeter (ITC), as well as an automated solid-phase peptide synthesizer. Several peptides were synthesized using the automated synthesizer, they were purified with high performance liquid chromatograph (HPLC) and characterized using electrospray ionization mass spectrometry (ESI-MS). Additionally, to create long peptides for this project I continued to advance my skills in a peptide to peptide linking strategy called native chemical ligation (NCL). In NCL, no protecting group is needed and simply requires a cysteine residue somewhere in the sequence. Progress was made and the synthesis of target molecules (connected peptides) was confirmed using mass spectrometry.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks.

Acknowledgement: This student's poster can be found in Appendix 1

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – SURF 2023

Fellowship Recipient: **Peter Wieber**
Faculty Mentor: *Nathan Schiele, Biological Engineering*
Project Title: *Development of a Bioreactor to Mechanically Stimulate and Evaluate Cell Self-assembled Neotendons*

Abstract: Tendon injuries are common but current repair methods are inadequate. Even with surgical repair, scar tissue and incomplete tendon healing result in prolonged weakness which presents a risk for re-rupture. Thus, new regenerative treatment methods are required. To develop new treatments, mesenchymal stem cells (MSCs) have been explored in tissue engineering strategies to form tendon replacements. Much of the current research is focused on recapitulating adult tendon structure and function using biomaterial scaffolds, but tissue engineering approaches aimed at modeling embryonic tissue development may lead to a new understanding of tissue formation. Our lab has developed a tissue engineering method to guide MSCs to form neotendons through directed cell self-assembly, which appear to mimic the early highly cellular stages of embryonic tendon formation. However, maturation of these neotendons may be limited without the dynamic mechanical stimulation associated with *in vivo* muscle contraction. Applying mechanical stimulation is a unique challenge due to the delicate nature and small size of the neotendons. Thus, our objective was to develop a bioreactor system to mechanically stimulate the neotendons and assess how that stimulation alters cell morphology.

Results: From the cytotoxicity assay, day 1 of treatment with the VeroClearTM(VC) resin extract media saw slightly more rounded cells in the media. However, by day 3 the differences between treatment and control diminished. Cell density appeared to decrease by a small margin when treated with the resin extract, but the resin would be classified as only slight cytotoxicity as described in ISO 10993-5:2009. Therefore, we deemed the VeroClearTM(VC) as an acceptable material and used it to print the main body of the bioreactor chamber (Fig 1C). The bioreactor design meets the design criteria, connects to our prior system [1] and can apply dynamic mechanical loading to neotendons. Additionally, despite their small size (~6 mm long) and fragile nature, the neotendons withstood the process of mounting, gripping, and applying 1% tensile strain. Staining demonstrated that 24 hours after a loading cycle, cells in the neotendon maintained a cohesive structure. No differences were detected between loaded and static controls, and future work will explore higher loading magnitudes and longer timepoints. There are a few suggestions for future work as well including increasing the time of pre-culture or treating the neotendons with a tenogenic factor such as TGF-β2 before applying loading. Lastly, some warping of the bioreactor occurred with multiple ethanol sterilization cycles suggesting that an alternative method (e.g., autoclaving) should be explored to increase the lifespan of the 3D resin-printed parts. In conclusion, we developed and tested a bioreactor system that can successfully apply mechanical stimuli to a cell-self assembled neotendon. Using this system, we can better understand how mechanical tensile loading impacts the early stages of tendon tissue formation *in vitro*.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks.

Acknowledgement: This student's poster can be found in Appendix 1

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research
Grant – SURF 2023

Fellowship Recipient: **Jennie Tafoya**
Faculty Mentor: *Robert Rinker, Electrical Engineering*
Project Title: *Tower of Lights Wireless Edition*

Abstract: Small, compact, and powerful, hardware Arduinos are microcontrollers that enable a user to more easily interact with the software utilized in many everyday applications. The unique functionality that the Arduino possesses is the ability to run a multitude of actions and processes synchronously, opening opportunity to perform a variety of processes otherwise difficult to handle on one controller. With the way that the microcontroller's interface is built, it allows those working on the Arduino to work closely with embedded systems. In my project, I plan to further study and exercise the processes that the Arduino can handle, expanding the capability to mass wireless functionality for the purpose of ease of reliable entertainment display in our Vandal Homecoming's Tower of Lights light show. Between the integrated development environment and multi-functional controller, the Arduino enables fluid study between hardware and software components. This summer, I plan to work closely with Dr. Rinker to improve my knowledge and solidify the bridge between the hardware and software world to create a product that envelopes user friendliness with wireless functionality.

Objectives: To expand TowerLights show from Fall 2022 with a completely wireless setup for Fall2023. The Tower of Lights operates by using mechanisms called "light bars". These are LEDs attached by aluminum wire to a microcontroller, all taped onto a wooden beam with a notch running down the middle for wire to run through. There are forty beams made for all forty rooms of the south-facing windows of the Theophilus Tower, between the 2_{nd} and 11_{th} floors. The wireless setup for the TowerLights show involves using Zigbee's Xbee Pro wireless transmitter, modeling the set up utilized for the Vandal Marching Band's LED glasses performances. Rather than using the connections made in the network closet on each floor from a plug-in to the wall, the idea for wireless is to provide portability. By attaching its power source to the beam, the LED circuit can be used in virtually any location.

Conclusions: With my project, I am happy to exclaim that I created a working prototype for the wireless edition of the Tower of Lights. To do this, I had to re-design the circuit involved between the power source, the microcontroller, and the LEDs. With the wired circuit set up, the power was drawn from a bigger power source. With the wireless setup, the power supply was limited to a nine-volt battery, which hindered the emission of light from the LEDs. The green and blue component of the LEDs required a minimum of 3.2 volts to light up each LED. With three LEDs in series, 9 volts simply wasn't enough. To counteract this problem, I studied and analyzed the distribution of power throughout the circuit set-up and concluded that a parallel circuit would deliver enough power without having to make any major changes to the circuit board that the microcontroller sits on. However, with three LEDs, the parallel circuit was lop-sided and may have involved extra resistance to distribute the luminous intensity evenly. By adding another LED, the parallel circuit balanced out. Once the circuit was designed, I soldered it together, taped, and tested it extensively.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks. Supplies were purchased at $955.

Acknowledgement: This student's poster can be found in Appendix 1

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – SURF 2023

Fellowship Recipient: **Juwon Elelu**
Faculty Mentor: *Herbert Hess, Electrical Engineering*
Project Title: *Reaped electromagnetic energy*

Abstract: Radio Frequency (RF) can be said to be a collective term for electromagnetic energy. This form of energy is derived from electromagnetic radiation which travels through a space between transmission antenna and receiving antenna. The receiving antenna only captured a small amount of RF energy. The purpose of this research is to design and build a system that can harvest the unused RF energy and convert it to useful electricity. We are surrounded by lots of devices that use RF energy in our day-to-day activities. Such devices include mobile phones, remote controls, microwaves, Wi-Fi, radio transmission towers etc. Most of these devices make use of batteries as a source of energy or as backup energy. These batteries are changed from time to time. The used batteries are disposed of in such a way that they cause environmental pollution. So, the need for alternative sources of energy for powering these devices cannot be overemphasized.

Objectives: Reaped electromagnetic harvester was designed to collecting unused RF radiation from its surroundings and convert AC to DC power. The project consisted of the following segments:
• <u>Transmission antenna</u>: The antenna responsible for collecting the radio frequencies. A patch antenna was decided for this project because they are small and flat allowing them to fit easily with circuit boards. The antenna collects energy at radio frequencies of 2.4GHz and uses the microstrip feedline as a feeding method. FR4 epoxy was used as the dielectric substrate for the patch antenna.
• <u>AC to DC converter</u>: Schottky diodes are responsible for collecting the AC current from the reaped RF energy at the antenna and converting into the DC current responsible for powering the device.
• <u>Microcontroller:</u> The microcontroller is responsible for collecting the DC current produced by the converter and performing the given task. The task given was to sense the amount of power passing through into a capacitor and once it had reached a certain amount it would stop the flow of current with the aid of a transistor, until power was needed to be stored again termed: switch mode control.

Project Accomplishments: Reaped electromagnetic harvester was designed to achieve the goal of collecting unused RF radiation from its surroundings and converting it to DC power. I designed and simulated my antenna with the aid of HFSS software to determine which material would be most suitable to make the antenna. From there I proceeded to search for a Schottky diode suitable to convert AC to DC, then I proceeded to make my PCB to allow me to test the parts for DC using an oscilloscope. Results showed that we were getting DC as an output which was a success, but this also led to the discovery that the diode could filter output, thereby acting as a capacitor. It was unexpected but it played in our favor and saved us resources. I used an oscilloscope to determine whether we were successfully getting DC from our circuitry and the outcome was a success. As for the microcontroller I was able to program it to control external hardware which sets the basis for future work. There is room for improvement for both the device and me. I hope to finish the original program after better understanding the MSP430, from there I can combine all units of the device to make it whole and functional. From there I hope to include a sensor into the device to create room for more experiments. And as time goes by future researchers can try new things as well.

Summary of Budget Expenditures: $6,000
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks. Unfortunately, as Juwon is not an American citizen, his $5,000 stipend was taxed accordingly (30%) and he only received $3,500.
- The student purchased consumables such as PCB boards, transistors, diodes, etc.
- The student printed a poster for $100.

Acknowledgement: This student's poster can be found in Appendix 1

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – SURF 2023

Fellowship Recipient: **Katherine Ryssel**
Faculty Mentor: *James Van Leuven, Animal Veterinary and Food Sciences*
Project Title: *Usage of P. Larvae tailocins to treat American Foulbrood disease in honeybees*

Abstract: Tailocins are antibacterial proteins produced by some bacteria to kill closely related bacterial strains. In nature, this benefits the microbe by offering a way for them to control the bacterial composition of their environments. However, in a lab setting, researchers can use these tailocins to treat against unwanted bacteria. The purpose of this research project is to test whether a tailocin treatment is a viable option in protecting honeybees against the deadly American Foulbrood and European Foulbrood diseases, which are caused by the bacterial species *Paenibacillus larvae* and *Melissococcus plutonius*, respectively. To test this, the proposed research will involve three major parts over the course of the 10-week period. Collection and purification of tailocins from honeybee pathogens will occur over the first two weeks, in vitro analysis of tailocins against strains of P. larvae and M. plutonius will be completed by the fifth week, and in vivo treatment of infected bees by tailocins will be explored for the final 5 weeks. This work addresses the urgent need to develop novel antibacterials and reduce our dependence on traditional antibiotics.

Project Accomplishments: Of the seven *P. Larvae* strains which were tested, five showed tailocin-associated activity. Of those five, two had consistent and replicable results. Possible tailocin activity is considered when a sample creates a zone of inhibition on a bacterial lawn. This can be easily distinguished from the plaques created by a prophage. When diluted, a prophage sample would create multiple smaller plaques on the lawn. A bacteriocin (such as a tailocin) would not create individual plaques, and instead, diluted samples show a faded zone of inhibition resulting from a less efficient killing of bacteria in the area. This kind of activity was seen from strain 368W against 5 of the 7 strains. One of the strains it did not show activity against was itself, which was expected as producing strains are resistant to their products. Strain 25747 showed inhibitory activity against two strains, including one of the strains that 368W was ineffective against. 25747 also showed prophage activity against strain 2605. Additionally, other strains showed activity under certain conditions. Strains Y-3650 and 16425 both showed very light activity against 368W when lawns had very low bacterial concentrations. Strain 3670 showed activity against 4 or 5 other strains in two tests, but such activity has not yet been consistently replicable. Three samples were taken in for TEM imaging. Only two of these samples were clear enough to take pictures. Sample 368W had structures which somewhat resembled F-type tailocins but could be a virus. No tailocin-like structures were found in sample 3670, although some interesting structures were seen. Although samples 368W and 3670 were clear enough to take images, they were still very thick. In the future, a better method of isolation must be found prior to imaging. It may be important to find an imaging facility which has the capability to run a glow discharge on the copper grids. Glow discharge may prevent the clumping of certain charged molecules and therefore allow clearer images to be taken.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks. Katherine purchased $1000 in supplies.

Acknowledgement: This student's poster can be found in Appendix 1

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – SURF 2023

Fellowship Recipient: <u>Alyssa Livingston</u>
Faculty Mentor: *Eva Strand / Courtney Conway, Natural Resources*
Project Title: *Northern Idaho Ground Squirrel Hypothesis Testing*

Abstract: Using ARCGIS and aerial imaging, I will compare changes in abundance of Northern Idaho Ground Squirrel populations to changes in forest canopy coverage over the past 50 years. I will also overlay these data to spatial footprints of past forest fires within areas that Northern Idaho Ground Squirrels inhabit. These comparisons will allow me to test the prevailing hypothesis for the population declines of this rare species: by testing the prediction that forest fire suppression has allowed tree growth and encroachment into the squirrel's habitat. This hypothesis predicts that the areas with the most pronounced population declines of Northern Idaho Ground Squirrels will have the most pronounced forest encroachment among colonies of Northern Idaho Ground Squirrels.

Project Accomplishments: Two main analyses were run on ArcGIS Pro: zonal statistics of change in tree cover over the past 40 years within the historical range of the Northern Idaho Ground Squirrel and within the squirrels' colony areas only. Results of the first test show that there has been an increase in tree cover in the historical range over the past 40 years. The effect size shows a 50% increase in tree cover through the entirety of the historical range. The tree cover in the historical range changes from 24% to 32% from 1986 to 2022 respectively, meaning that tree cover has increased in the historical range by roughly 8%. Results of the second test show that there has also been an increase in tree cover in the squirrels' colonies over the past 40 years. The effect size again shows a 50% increase in tree cover within extinct and extant colonies. The inhabited range of extant and extinct colonies shows tree cover changing from 7% to 9% from 1986 to 2022 respectively, meaning tree cover percentage has increased roughly 2% in the inhabited range in this time. The fire suppression hypothesis is supported based on two predictions. Forest canopy cover has significantly increased over the last 40 years in both the historical range of the Northern Idaho Ground Squirrel and in the explicit colonies within their range. Canopy cover within the historical range (15% cover) was higher than canopy cover within the actual colonies (5% cover). Experts on this rare species believe that these squirrels need areas with <15% canopy cover. The hypothesis is supported based on our results.

Future Research: I will be continuing this research as my thesis project for my degree. In the future I will be performing the same zonal analysis test on the colony range but split into two groups: extant and extinct. We would like to see if there is a correlation in extinct population areas and higher tree cover percentages. We would then like to do the same thing but use extant (stable or increasing) vs extinct + extant (decreasing). The population data that we have on this species is limited and spotty due to the nature of the squirrel and its hibernation patterns allowing only a small window for field data collection. We would still like to run a linear regression on the data we do have with our tree cover data to see if there is a correlation. We also hope to work with Idaho Fish and Game to get population data that is more conducive to our research to directly compare our data. Through my future research I plan to use more aerial imaging to determine more historic tree cover changes for extinct populations to determine whether there is a correlation. Most populations that are labeled as extinct were noted to be as such in the early 1980's, so I will be looking at aerial images from extinct colony areas and transporting them into ArcGIS Pro to get the zonal statistics and tree cover data for them for times further back than the Rangeland Analysis Platform can go (1986). We would finally like to get canopy cover data in corridors between colonies to see if there is ability for dispersal anymore or not.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks.
- $150 was spent on travel to Field Sites, $75 was spent on creating a poster.

Acknowledgement: This student's poster can be found in Appendix 1

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – SURF 2023

Fellowship Recipient: **Mason Kilker**
Faculty Mentor: *Sebastian Stoian, Chemistry*
Project Title: *Synthesis and Structural Characterization of Zinc Complexes with N, S donor ligands.*

Abstract: Zinc complexes, which are diamagnetic with no unpaired electrons, are sought after for their unique chemical and biological applications as enzyme analogues or bioinspired catalysts, being relevant to several different areas in pathophysiology and bioinorganic chemistry. These potential applications require an understanding of the structural factors that lead to the desired properties of these compounds. The main goal of this proposal is to prepare a novel series of zinc complexes and to elucidate their coordination modes and structures both in solution, by NMR, and in solid state by *x*-ray crystallography. These complexes will incorporate a tetradentate ligand with N,S donor atoms, either S,S′-**b**is(2- **p**yridylmethyl)-1,2-**thio**ethane (**bpte**) or S,S′-**b**is(2-**p**yridylmethyl)-1,3-**thio**propane (**bptp**), and two additional anionic co-ligands NCX-, where X= O; S; Se; and BH3. Because zinc complexes with **bpte** and **bptp** are virtually unexplored, this work will provide access to a large selection of zinc compounds with several diverse structures and will contribute to a greater understanding of steric factors at play in zinc complexation.

Project Accomplishments: We have synthesized and structurally characterized three new complexes, Zn(bpte)(NCS3)2, [Zn(bpte)(EtOH)$_2$](ClO4)$_2$, and Cu(bpte)$_2$. The solid-state structures of these compounds were elucidated using X-ray crystallography. In addition, a previously synthesized complex, Zn(bpte)(NCBH$_3$)$_2$, was examined using Nuclear Magnetic Resonance (NMR) spectroscopy. Finally, we have optimized the synthetic protocol used to obtain Zn(bpte)(NCBH$_3$)$_2$. The bpte and bptp ligands were successfully synthesized using procedures adapted from published methods. Their structures and sample purities were assessed using nuclear magnetic resonance (NMR) spectroscopy. However, we have only explored the coordination chemistry of bpte. [Zn(bpte)(NCS)$_2$]. A new complex, [Zn(bpte)(NCS)$_2$], was synthesized. The structure of this compound was elucidated through X-ray crystallography. The complex was obtained as clear block crystals and exhibits a slightly distorted octahedral geometry. N-Zn-S angles are approximately 76 degrees (76.4°, 76.7°), and axial N atoms exhibit an angle of 167.7°. The experimental structure is shown in Figure 1 together with the model included in the proposal.

Over the summer of 2023, I was fortunate enough to take part in one workshop and one conference. I attended the Whitworth X-Ray Crystallography Workshop at their Spokane campus, where some basics of X-ray crystallography were covered. Additionally, I was able to present at ICUR in conjunction with my fellow undergraduate researchers.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks.
- Spent $703 on solvents, reagents, and characterization.
- Workshop Registration was $225.

Acknowledgement: This student's poster can be found in Appendix 1

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – SURF 2023

Fellowship Recipient: **Zaiden Espe**
Faculty Mentor: *Frank Gao, Mathematics and Statistical Science*
Project Title: *Optimizing the Nelder-Mead method via Genetic Programming*

Abstract: This project aims to discover related methods and optimizations to the Nelder-Mead method. The Nelder-Mead method is an algorithm used to solve the unconstrained minimization problem, or function optimization problem that transforms a geometric object throughout the function space. To explore the limits of the Nelder-Mead method, genetic programming will be used. Genetic programming is the most robust approach because it will allow me to search a large space of modifications to the Nelder-Mead algorithm. The research will focus on adjusting the scalar parameters and reflections of the Nelder-Mead transformations. The result will be an algorithm based on the Nelder-Mead method. The expected outcome will exceed the standard Nelder-Mead method and variations.

Objectives: To test the frequency of selection of various reflection vectors in a copy of the comparison Nelder-Mead algorithm. The reflection vectors were called the "rank", "inverse gradient", "slope", "evaluation", "mirror", and "top rank" reflections. Each of these vectors would be calculated at each iteration. Then the resulting vector was used as the centroid is used in the standard Nelder-Mead method to calculate the reflection. The rank vector was calculated using a weighted average of the n-best points in the n-dimensional simplex, with better ranked points having a linearly higher weight than worse points dependent on their rank. The inverse gradient vector was calculated by multiplying the inverse of the matrix generated by subtracting the worst vector from each vector in the simplex with the vector generated by subtracting the evaluation of the worst vector from the evaluation of the other vectors. This final vector gives an estimate of the gradient at the worst point that becomes more accurate as the simplex shrinks. The slope reflection is calculated by finding the slope from the worst point to each other point in the simplex, and then multiplying the new slope vector by the matrix formed by the difference of each vector and the worst vector. The evaluation reflection was calculated by using a weighted average of the n-best points in the simplex. The weights were determined by taking the square root of the difference between the function evaluation of the point and the function evaluation of the worst point. The mirror reflection was calculated by orthogonally projecting the worst vector through the space determined by the n-best vectors. The top rank reflection was formed by taking the rank average, but only with the top $n/2$-best vectors. Each reflection was tested as the replacement for the centroid vector in the standard Nelder-Mead method. The reflections were also tested in various combinations with each other, and all together, each being evaluated by the algorithm and then the best performing reflection chosen as the reflection. The algorithm was tested with n+k points, k being two or larger. The standard Nelder-Mead algorithm uses n+1 points. For values of k, the algorithm would reflect k-points, using the n-best points to determine the reflection. The value of two for k was tested most extensively. The tests compared how which reflections were chosen for each reflected vector by rank and how the reflections were manipulated via expansion, contraction, or standard reflection.

Results: The most successful algorithm used the standard reflection and mirror reflection with a simplex containing n+2 points. The worst point was always reflected using a mirror reflection except every ten iterations the standard reflection was used, and the second worst point was always reflected using a standard reflection.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks.

Acknowledgement: Poster can be found in Appendix 1

15

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – Summer OUR Students' 2023

Fellowship Recipient: **Kendall Mitton**
Faculty Mentor: *Bert Baumgaertner*
Project Title: *Anxiety and the Effectiveness of Fact-Checking*

Abstract: As levels of misinformation rise in the United States, so does the threat to efficient and effective policymaking. Most Americans are misinformed about a variety of topics and actively think and behave against the recommendations and empirical findings of experts. That said, the lack of fact-checking misinformation and failure to accept corrective information has fostered a variety of different problems within our borders (e.g., Qanon/PizzaGate). Anxiety may be a factor in this phenomenon, but it has been largely overlooked in previous research. Building off existing scholarship surrounding these variables, I will conduct an online survey experiment to test the relationship between anxiety and the effectiveness of fact-checking misinformation. The results of this survey will help further our broader understanding of the effect anxiety has on our behavior, as well as when fact-checking misinformation is effective. If society can identify when and how to effectively provide fact-checks, we may be able to minimize the threat misinformation poses to our Democracy.

Objectives: Many scholars have already conducted experiments on the manipulation of information exposure. Most notable for my research is the work of Albertson & Gadarian (2015) and Nyhan & Reifler (2018). Albertson & Gadarian (2015) identify several effects anxiety can have on the information we seek out and the information we are provided with. Furthermore, Nyhan & Reifler (2018) identify ways in which fact-checking effectively works to correct misinformation and misperceptions. While the findings of both these studies provide important knowledge on information exposure, there is a gap in this research that should be further explored. Moreover, the gap I want to bridge is between the types of information/opinions individuals maintain and how or when individuals assimilate new information. Strictly speaking, what effect does anxiety have on the effectiveness of fact-checking misinformation? It is important to fill this gap in our knowledge so that social and political experts will be in a better position to address issues related to misinformation.

Results: To properly identify the relationship between anxiety and misinformation fact-checks on Covid-19 vaccine opinions, I conducted an interaction data analysis. Using an interaction analysis, I was able to look at the interaction of my treatment and control groups for both independent variables on my dependent variable. Put simply, I was able to examine the four following groups: Control = {anxiety control/fact-check control}, Anxiety Dummy = {anxiety treatment/fact-check control}, Fact-Check Dummy = {anxiety control/fact-check treatment}, and the Interaction = {anxiety treatment/fact-check treatment}. Given my research was not focused on the interaction of two control groups, the following results will summarize the interaction of an Anxiety Dummy, a Fact-Check Dummy, and an Interaction. Regarding the Anxiety Dummy variable (i.e., the variable examining respondents in the anxiety treatment/fact-check control group), I found that anxiety has a significant but negative effect on a respondent's individual perceptions about the Covid-19 vaccine. As one can see in the first row of the table, a 1-point increase in the respondent's level of anxiety leads to a .35-point decrease in that individual's perception towards the Covid-19 vaccine. In short, as a respondent's anxiety level increases, the more negative their thoughts about the vaccine will be. Moving forward, I did not find any significant effect of the Fact-Check Dummy variable (i.e., the variable examining respondents in the anxiety control/fact-check treatment group) on individual perceptions about the Covid-19 vaccine. Similarly, I did not find any significant effect of the Interaction variable (i.e., the variable examining respondents in the anxiety treatment/fact-check treatment group) on individual perceptions about the Covid-19 vaccine.

Summary of Budget Expenditures: The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks.
Acknowledgement: This student's poster can be found in Appendix 2.

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – Summer OUR Students' 2023

Fellowship Recipient: **Alexandro Aguilar**
Faculty Mentor: *Roger McVey*
Project Title:

Abstract: The purpose of this project is to research, perform, and analyze the piano music of Manuel Maria Ponce and study the influence of Mexican folk music on it, while exploring my own Mexican heritage. My goal is to highlight Ponce as a composer of an underrepresented background by performing his music at two different concerts, one of which I will oversee planning and organizing. The music of Mexican composers such as Ponce, deserves to be in the standard repertoire of classical piano as his artistic merit could be compared to that of other European composers such as Frederic Chopin or Maurice Ravel. It is important to highlight music from people of color and help expand the scope of music as a studied form of art if the study of classical piano were to stay relevant in modern day. I will do this by conducting research study on Ponce's life, influences, and music. I will be showcasing the compositions of Manuel Ponce while bringing awareness to the music of composers from non-euro-centric backgrounds.

Project Accomplishments: From reading on Manuel Ponce's life, analyzing his works for solo piano, and researching the music of Mexico, I found that the traditional folk genres of Mexico had a heavy influence on the compositional style of Manuel Ponce. The Jarabe, Cuban Contradanzas, Sons and Huapangos, and Corridos, all have heavy correlation from their musical motifs to those found in the compositions of Manuel Ponce. Ponce drew inspiration from the rhythmic motifs of these folk genres. There is a musical idea called a "Sesquialtera" that is found in both the music and dance of Mexico. The Sesquialtera is the flexibility between ternary and binary based rhythmic ideas. Manuel Ponce uses the Sesquialtera in an obvious manner in his works Scherzino Mexican and the B sections of the Cuatro Danzas Mexicanas, giving his works a dance topic derived from the rhythmic motifs. Ponce draws melodic inspiration from the traditional corridos that came from the Mexican Revolution which occurred during his early years as a composer. Ponce draws inspiration from the parallel third voicings of corrido melodies and the motific descending diatonic thirds that resolve phrases. He uses this in his Scherzino Mexicano and Romanza De Amor.

I plan to present my work done this Summer in the form of a lecture recital at the Lionel Hampton School of Music in September during Latinx Heritage Month. I will present the music SURF Grant Proposal of Mexico and the life and compositions of Manuel Ponce in the form of a lecture followed by a performance of the works I worked on by Ponce. I also plan on auditioning/applying to present the same work at the Music Teachers National Association National Conference in March of next year. I plan on continuing to research these topics while also learning and performing the works of Manuel Ponce for other events such as my junior and senior performance recitals.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks.

Acknowledgement:
- This student's poster can be found in Appendix 2
- This student's poster won a 3-minute presentation competition held at UI on the eve of ICUR 2023.

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – Summer OUR Students' 2023

Fellowship Recipient: **<u>Amara Bailey</u>**
Faculty Mentor: *Dr. Omi Hodwitz*
Project Title: *Missing and Murdered Indigenous Women, Girls, and Two-Spirit (MMIWG2) database*

Abstract: The United States and Canada have histories littered with abuse against Indigenous populations. The systemic racism which previously justified genocide and displacement of these communities is upheld today and felt throughout Indigenous communities. The devaluing of Indigenous lives is felt most notably by the epidemic of Missing and Murdered Indigenous Women, Girls, and Two-Spirit individuals (MMIWG2). Underlying this issue is an information deficit surrounding the thousands of cases that exist across Indigenous communities. The MMIWG2 database aims to resolve this problem by being an exhaustive culmination of already existing data from community-led efforts. The MMIWG2 database research involves the collection of MMIWG2 cases, and subsequent authentication of everyone's case.

Project Accomplishments: This summer, approximately 500 cases of MMIWG2 in the United States were coded. Of those 500, only 407 were confirmed and authenticated. The data collected thus is representative of approximately 10% of the collected potential cases. The use of this data to understand the MMIWG2 epidemic would be irresponsible and paint an incomplete picture of the phenomenon. However, the data currently available is valuable in understanding who MMIWG2 are, the demographics of their offenders, as well as a basic understanding of the effectiveness of law enforcement in closing MMIWG2 cases. Of the total confirmed cases, 392 have known ages. Of these, 94 MMIWG2 were under 18 at the time of their victimization. Overwhelmingly, MMIWG2 identify as female. 405 confirmed cases have known genders, of which, 269 are female, 134 are male, 1 is non-binary, and 1 is two-spirit. Only 87 cases have identified at least one offender. Of these, 75 offenders are male and 3 were younger than 18 when they offended. The response by law enforcement in each case is incredibly variable. The role of tribal police is determined by the occurrence of an incident on tribal land; of the confirmed cases, 72, less than 20%, of incidents occurred on reservation. In terms of case resolution generally, there were only 364 cases with known outcomes. 54.7% of these are resolved, meaning an individual or their remains have been located. The remaining 45.3% of cases are still missing a body. Only 21.2% of total confirmed cases had both a known offender and a recovered body.

The data reveal a disturbing trend; many MMIWG2 cases are and remain unsolved. The significant failure to close these cases may be indicative of the ineffectiveness of law enforcement or poor reporting systems for MMIWG2. In many of the unconfirmed cases, no record of the individual existed. In almost all the confirmed cases, Facebook groups and other community-based reporting systems were heavily relied upon, as many individuals were not reflected in state-wide or federal databases. The continuation of investigation into this phenomenon is vital to understanding and eventually preventing the epidemic of MMIWG2. As the number of cases confirmed and coded increases, the data becomes more valuable and versatile. The data collected this summer only verify the intense need for continued research into an issue with almost no study or literature.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks.

Acknowledgement: This student's poster can be found in Appendix 2

Final Project Report: Office of Undergraduate Research (OUR) Undergraduate Research Grant – Summer OUR Students' 2023

Fellowship Recipient: **Hunter Rouse**
Faculty Mentor: *Joshua Bailey*
Project Title: *Does Functional Power Threshold test differ between bikes*

Abstract: Advancements in technology have continued to push the limits on how cyclists optimize training programs based on intensity. Power measures provide instantaneous feedback reflective of each pedal stroke versus heart rate measures which possess a delayed response. A Functional Threshold Power (FTP) test to establish the average power output during a maximal effort ride is an effective tool to create intensity zones for training.1 FTP tests are traditionally performed outdoors. However, with poor weather or limited routes, they can be performed indoors on stationary trainers. The Wahoo Kickr stationary cycle was designed to resemble a true road cycling experience while staying indoors. Prior to utilizing the Kickr as a tool for completing an FTP test, potential differences between the cyclist road bike and the Kickr need to be established. Therefore, the purpose of the study is to investigate if an individual's performance during an FTP test is affected by the type of stationary bike. Cyclists will perform a 20-minute maximal effort FTP test on their personal bike and one on the Kickr stationary bike. It is hypothesized cyclist will have higher average power output when performing the FTP test on their own bike.

Summary of Budget Expenditures:
- The student was awarded a $5,000 stipend to conduct this research for 40h/ week over 10 weeks.

Acknowledgement:
- This student was selected to give a 45-minute Paired Research Presentation with his mentor at the ICUR 2023 and did not present a poster.

Appendix 1

Determining the role of the TGFβ Type II Receptor on lysyl oxidase production

Dillon B. West, Colin R. Marchus, and Nathan R. Schiele
Department of Chemical & Biological Engineering, University of Idaho, Moscow, ID

University of Idaho — IdahoINBRE

Abstract

Tendons transfer forces from muscle to bone and are vital to movement. Unfortunately, tendons are injured frequently and have poor self-healing capability. Current therapies are unable to restore damaged tendons to their prior strength. Tissue engineering using mesenchymal stem cells (MSCs) is being investigated in strategies to develop functional tendon replacements or repairs. A current challenge is the limited information on the regulators of MSC differentiation into specialized tendon cells (tenogenesis) and control of functional tendon formation. Prior work showed that the transforming growth factor beta (TGFβ) family of cytokines are critical in regulating tenogenesis and the production of lysyl oxidase (LOX), an enzyme that mediates collagen crosslinking and functional tendon formation. TGFβ may activate the TGFβ Type II Receptor (TGFβRII), a transmembrane protein that recruits and phosphorylates TGFβRI when bound by a TGFβ ligand However, how TGFβRII signaling controls the TGFβ response in MSCs to regulate LOX production is unclear. The objective of this study was to determine how inhibition of TGFβRII impacts LOX levels in MSCs. We are exploring inhibiting TGFβRII using both a chemical agent as well as siRNA and evaluating the impacts on MSCs and LOX levels. The outcomes of these studies will show how TGFβ signaling through TGFβRII in MSCs regulates LOX levels, which can be used to direct functional tendon tissue formation and future regenerative therapies.

Background

- Tendons are connective tissues that absorb and transmit forces to allow body movement that injure easily and heal poorly
- The cytokine TGFβ is believed to be responsible for the expression of functional tendon markers such as lysyl oxidase (LOX)
- LOX is an enzyme that mediates the crosslinking of collagen in tendons
- There is a limited understanding of how the signaling pathways between TGFβ Type I and Type II Receptors may differ
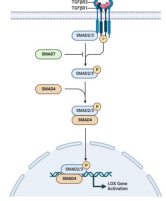
Figure 1: The TGF-β signaling pathway starts with the dimerization of TGFβRI & RII upon ligand binding. SMAD 2/3 are then recruited & phosphorylated before recruiting SMAD 4, translocating into the nucleus, & enhancing LOX expression (BioRender)

Objective: *Identify how TGFβ Type II Receptor impacts lysyl oxidase production*

Experimental Design

- The role of TGFβRII can be investigated via inhibition methods
- Morin was found in literature to be a potential chemical inhibitor of TGFβRII
  - However, there was little support and TGFβRII may have the ability to auto-phosphorylate
- RNA interference utilizing siRNA (short interference RNA) was selected as a secondary method to disrupt TGFβRII levels
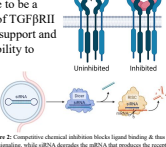
Figure 2: Competitive chemical inhibition blocks ligand binding & thus cell signaling, while siRNA degrades the mRNA that produce the receptor (BioRender)

Experiment 1: TGFβ Type II Receptor Chemical Inhibition

Methods & Materials

- Murine MSCs were plated in 10% FBS
- After 24 hours, media changed to 1% FBS
- After 48 hours, treatments include:
  - Morin & TGFβ2, morin vehicle control (DMSO, VC) and TGFβ2, morin and TGFβ2 VC (H₂O), & morin VC and TGFβ2 VC.
  - Morin was at 0.5, 5, & 50 μM and TGFβ2 at 50 ng/mL.
- Cells were imaged, collected at 1, 3, 7 days

Morin may not impact LOX

LOX | β-Actin | 0.5 μM | 5 μM | 50 μM
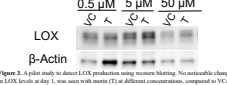
Figure 3. A pilot study to detect LOX production using western blotting. No noticeable change in LOX levels at day 1, was seen with morin (T) at different concentrations, compared to VCs.

Trial 1 — 50 μM
Trial 2 — 50 μM
Trial 3 — 50 μM

(Day 1, Day 3, Day 7; DMSO +TGFβ2, Morin +H₂O, Morin +TGFβ2, DMSO +H₂O)
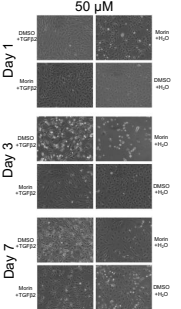
Figure 4: High concentrations of DMSO resulted in abnormal cell morphology and cell death. Day 3 plates had low cells densities and resulted in cell death when treated with TGFβ2.

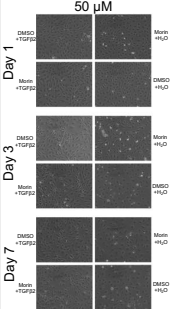**Figure 5:** DMSO concentrations were lowered for Trial 2. However, the concentrations at 50 μM exceeded 0.5% DMSO in media, still resulting in an abnormal environment and reduced cell growth even with TGFβ2 treatment.
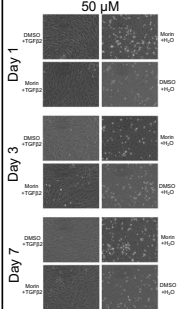
Figure 6: DMSO concentrations were brought to below 0.5% DMSO in media for Trial 3. Treated MSCs appear healthy and TGFβ2 treatment induced fibroblastic cell morphology.

Experiment 2: TGFβ Type II Receptor siRNA Inhibition

Methods & Materials

- Murine MSCs were plated in 10% FBS
- After 24 hours, the cells are washed, and media changed to 1% FBS
- After 48 hours, a solution of lipofectamine and siRNA was prepared and pipetted into plated cells for transfection
  - 0.1, 1, and 10 nM were tested for each construct and control
- 48 hours after transfection, the cells were treated with 50 ng/mL of TGFβ2
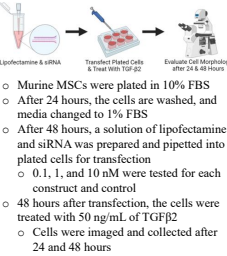 - Cells were imaged and collected after 24 and 48 hours

Fluorescent Control

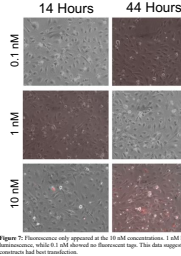14 Hours | 44 Hours (0.1 nM, 1 nM, 10 nM)

Figure 7: Fluorescence only appeared at the 10 nM concentrations. 1 nM had sparse luminescence, while 0.1 nM showed no fluorescent tags. This data suggested 10 nM constructs had best transfection.

TGFβ2 Treatment

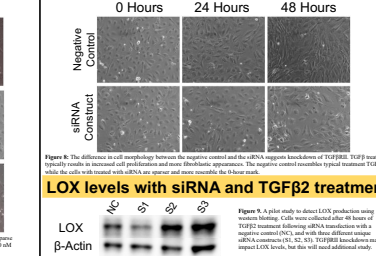0 Hours | 24 Hours | 48 Hours (Negative Control, siRNA Construct)

Figure 8: The difference in cell morphology between the negative control and the siRNA suggests knockdown of TGFβRII. TGFβ treatment typically results in increased cell proliferation and more fibroblastic appearances. The negative control resembles typical treatment TGFβ, while the cells with treated with siRNA are sparser and more resemble the 0-hour mark.

LOX levels with siRNA and TGFβ2 treatment
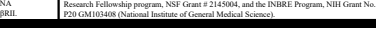
LOX | β-Actin | NC | S1 | S2

Figure 9. A pilot study to detect LOX production using western blotting. Cells were collected after 48 hours of TGFβ2 treatment following siRNA transfection with a negative control (NC), and with three different unique siRNA constructs (S1, S2, S3). TGFβRII knockdowns may impact LOX levels, but this will need additional study.

Discussion & Future Directions

The observable cell morphology differences between morin and its vehicle control are negligible. The cell morphology differences between the siRNA construct and its negative control suggest a more thorough inhibition of TGFβRII. In the future, additional timepoints and siRNA knockdowns will be analyzed to quantify the levels of SMAD activation and LOX protein levels to better understand the role of TGFβRII.

Acknowledgements

Precision Agriculture Adoption and Integration Case Study

Lacey Hunt, Dr. John Shovic, Mary Everett, Garrett Wells, Walter Neils
DEPARTMENT OF COMPUTER SCIENCE, UNIVERSITY OF IDAHO

SCARECRO — University of Idaho

Introduction

- Precision agriculture describes a variety of emerging technological systems and tools for use in crop and livestock farming
  - Improve profitability for farmers [1]
  - Potentially reduce negative climatic impacts of farming [2]
- Remote sensing is a tool that has not been widely adopted, despite availability of these systems [3]
  - This is due to high cost and lack of usability
- Sensor Collection And Remote Environment Care Reasoning Operation (SCARECRO) system
  - Modular, extensible, open-source system that uses low-cost hardware to resolve cost and usability issues
  - Consists of a gateway, middle agent, weredog, and sensors
  - Send data collected by sensors to database to be stored, displayed, and used in AI models
  - Already implemented in Laurel Grove Wine Farm in Winchester, VA
- This project aims to test this system's performance in a new application domain: a heritage apple orchard at Sandpoint Organic Agriculture Center (SOAC)
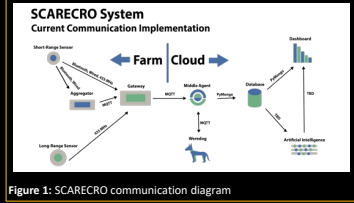 - Observe performance, compare cost, and evaluate usability

Figure 1: SCARECRO communication diagram

Methods

- Performance
  - Perform routine system maintenance and determine uptime
  - Write automated tools in Python to find system faults
- Cost
  - Calculate labor and component implementation cost
  - Compare to other similar remote sensing systems
- Usability
  - Integrate dashboard to display relevant data
  - Regularly survey farmer regarding system functionality

Figure 2: Left to right: Datagator, Gateway, WeatherRack Station

Progress

- Assembly and installation of all system components is complete (Figure 2)
  - Gateway to eliminate duplicate messages, keep track of sensor connections, and send data to the database
  - Aggregator (Datagator) to collect and transmit readings from sensors located further away from the gateway
  - WeatherRack (both WR2 and WR3 models used) to collect weather data
  - Middle agent to handle data transmission to gateway and/or database on cloud computer
- Integration of a dashboard to view collected data from database (Figure 3)
- System usage and usability surveys
  - Conducted virtually over a 10-week period (will start late summer)
  - Used to discover the usability of the system and dashboard as well as ask for suggestions to improve the system in the future
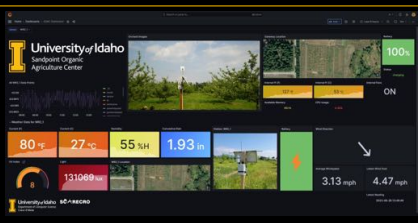- Began cost analysis of current structure and comparison to similar systems

Figure 3: SOAC Dashboard

Results

- Performance
  - System assembly and implementation was successful
  - Consistent data coming in
  - Occasional continuous network connectivity issues
- Preliminary research showed financial advantage to using SCARECRO as compared to other similar systems
- Initial discussions with horticulturalists at SOAC proved this system's usefulness and room for input

Future Work

- Conduct and analyze continued farmer surveys
- Implement SOAC horticulturalist suggestions
  - Light canopy sensors to quantify benefits of pruning
  - Degree day modeling for pest control
- Installation of additional local test unit (Harbor Center)
  - Help test reliability in a new setting
- Troubleshoot connection issues in gateway

References

[1] M. Boehlje. (2021, February 22) The Value of Data/Information and the Payoff of Precision Farming. [Online]. Available: https://ag.purdue.edu/commercialag/home/resource/2021/02/the-value-of-data-information-and-the-payoff-of-precision-farming/

[2] P. A. C. T. Force. (2021, November 10) Task Force for Reviewing the Connectivity and Technology Needs of Precision Agriculture in the United States." Available: https://www.fcc.gov/sites/default/files/precision-ag-report-11102021.pdf

[3] Lowenberg-DeBoer, J. and Erickson, B. (2019, July 1). Setting the Record Straight on Precision Agriculture Adoption. Available: https://acsess.onlinelibrary.wiley.com/doi/full/10.2134/agronj2018.12.0779

Acknowledgements

Segmentation analyses to identify and quantify microglia in 3D image stacks

¹UNIVERSITY OF IDAHO, ²LEWIS-CLARK STATE COLLEGE
³DEPARTMENT OF COMPUTER SCIENCE, ⁴DEPARTMENT OF BIOLOGICAL SCIENCES
Dan Blanchette[1], Dr. Samuel Seth Long[2,3], Dr. Diana Mitchell[1, 4]

ABSTRACT

Using time-lapse confocal microscopy to record and observe microglia behavior in living zebrafish embryos, the Mitchell lab investigates the molecular basis of dynamic migration and phagocytic behavior of microglia in the central nervous system. Microglial cells regulate brain development, maintain neuronal networks, and repair neural injuries. Timelapse imaging provides crucial insight into the behavior of these cells. Other methods, such as manual cell counting, are subject to human bias and are tedious, repetitive, and time-consuming. The Long computer science lab utilizes Python and open-source modules to develop an automated pipeline as a programmatic solution. These methods aim to expedite and optimize the lab's manual processes. This research project seeks to segment microglia when applied to 3D time series image stacks. Computer segmentation of microglia imaging relies on pixel values to generate bitmasks. Microglia segmentation is challenging because the cell's irregular shape and size can be obscured, resulting in omitted pixel intensity values. Two programs were developed to test the viability of Otsu, multi-Otsu, and Yen automatic thresholding[2]. Both methods separate the raw image into two layers: the foreground and background. Numerical data is generated via histograms to predict optimal threshold values for each algorithm. Prototype implementations of these programs demonstrate viability for microglia counting. However, misclassification of microglia occurs when the predicted pixel threshold is outside the histogram's range. Further refinement of these programs is crucial as they will be foundational for future object classification methods and microglia tracking, optimizing the lab's data processing capabilities and time.

INTRODUCTION

- Microglial cells regulate brain development, maintain neuronal networks, and repair neural injuries.
- Bit masking is the method of isolating specific color value combinations (objects of interest) from the rest of a digital image.
- Previous versions of this program were applied to 2D images with a set threshold value. The aim of this iteration is to adapt dynamic thresholding methods to 3D image stacks.

METHODS
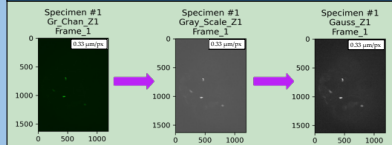
Figure 1: Copying of raw image with applied gray scale then Gaussian blur[1].
(NOTE: all images were recorded at 20x magnification).
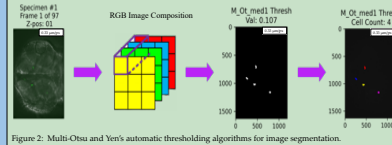

Figure 2: Multi-Otsu and Yen's automatic thresholding algorithms for image segmentation.
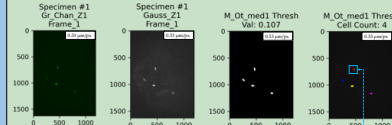
RESULTS

Figure 3: Processed image with Multi-Otsu and Otsu algorithm.

Frame ID	Z-pos	Cell Count	Thresh Method	Threshold Value
1	1	4	M_Ot_med1 Thresh	0.107
1	2	6	Fixed Thresh	0.11
1	3	6	M_Ot_med1 Thresh	0.116
1	4	9	M_Ot_med1 Thresh	0.107
1	5	8	Fixed Thresh	0.11
1	6	7	Fixed Thresh	0.11
1	7	7	Fixed Thresh	0.11
1	8	14	Fixed Thresh	0.11
1	9	14	Fixed Thresh	0.11
1	10	15	Fixed Thresh	0.11
1	11	15	Fixed Thresh	0.11
1	12	13	Adj Max Thresh	0.11
1	13	6	Adj Max Thresh	0.092
1	14	8	Adj Max Thresh	0.11
1	15	7	Adj Max Thresh	0.11
1	16	7	Adj Max Thresh	0.11
1	17	15	most Max Thresh	0.094
1	18	24	most Max Thresh	0.09
1	19	7	most Max Thresh	0.109

Table 1: Multi-Otsu and Otsu results.
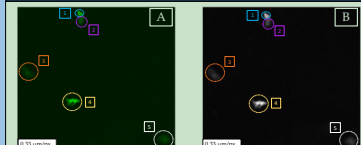

Figure 4: Mis-classified object

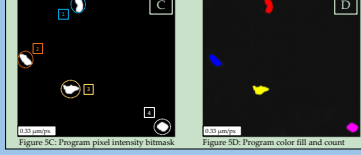## RESULTS (CONT.)

Figure 5A: Manual count of microglial cells
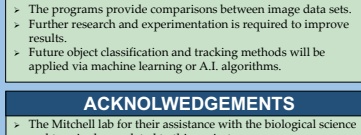
Figure 5B: Verification count with denoising.
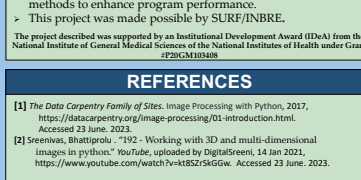
Figure 5C: Program pixel intensity bitmask

Figure 5D: Program color fill and count

CONCLUSIONS / FUTURE WORK

- The programs provide comparisons between image data sets.
- Further research and experimentation is required to improve results.
- Future object classification and tracking methods will be applied via machine learning or A.I. algorithms.

ACKNOWLEDGEMENTS

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[2] Sreenivas, Bhattiprolu. "192 - Working with 3D and multi-dimensional images in python." YouTube, uploaded by DigitalSreeni, 14 Jan 2021, https://www.youtube.com/watch?v=kt8SZrSkGGw. Accessed 23 June. 2023.

Visualization of tendon organization and fascicle crossover in 3D using micro-CT

Destinee M. Ditton[1], Colin R. Marchus[1], Michele R. Brumley[2], and Nathan R. Schiele[1]
¹Department of Chemical & Biological Engineering, University of Idaho, Moscow, ID, ²Department of Psychology, Idaho State University, Pocatello, ID

Abstract

Micro-computed tomography (CT) is an x-ray-based imaging modality that produces three-dimensional (3D), high-resolution images of whole-mount tissues, but is limited to dense tissues, such as bone. The x-rays readily pass through tendons, rendering them transparent. Contrast-enhancing chemical stains have been explored, but their use to improve contrast in tendon for micro-CT imaging had not been systematically evaluated. To address this, our previous investigation showed that phosphotungstic acid (PTA) staining increased x-ray absorption of tendon to enhance tissue contrast and obtain 3D micro-CT images of rat tendons within the tail and hindlimb. Further, we demonstrated that tissue hydration state significantly affected soft tissue contrast. Utilizing this new contrast enhancing method, 3D images from tissues were analyzed and processed to identify and trace tendon fascicles and analyze the anatomical organization of the tail and hind limb. From this analysis, we found that tail tendon fascicles appeared to cross between fascicle bundles, whereas it was assumed that the fascicle bundles maintain specific tracks within the tendon. Based on these initial findings, we extended our study and investigated the tails and hind limbs of younger and older, sexually mature rats to evaluate tendon fascicle crossover as a function of age. Ultimately, contrast-enhanced 3D micro-CT imaging will lead to better understanding of tendon structure, and relationships between the bone and soft tissues.
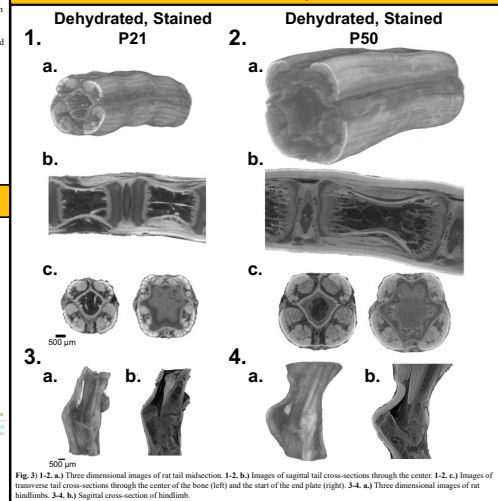
Background

- Tendons are connective tissues that absorb and transmit forces to allow body movement that injure easily and heal poorly
- Need to understand structure-function relationships with comprehensive imaging
- Soft tissues are difficult to visualize *in situ*
  - Using a micro-CT scanner allows 3D visualization of tissue, but requires high density materials
  - Phosphotungstic acid (PTA) stains collagenous tissues for observation with micro-CT
- Identified most effective contrast-enhancing technique for micro-CT imaging of tendon

Fig. 1) a.) Transverse cross-section of rat tail through the center of the bone and b.) through the intervertebral disc showing anatomical structures.

Objective: Validate a contrast-enhancing imaging technique for a range of ages and evaluate structural changes in the tendon with development
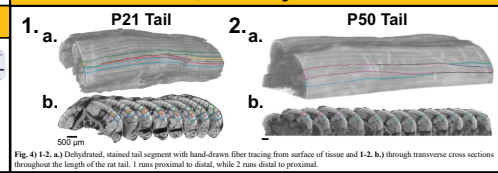
Methods and Materials

Fig. 2) Diagram of method for collection and preparation of tissues, P21 (n=3), and kangaroo rats (n=2), for imaging in Skyscan 1275 micro-CT scanner.

- P21 rats stained for 10 days and imaged at 70 kV and 59-63 μA
- P50 rats stained for 11 days and imaged at 70 kV and 40-61 μA
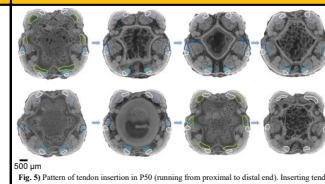- Kangaroo rats stained for 1 ½ months and imaged at 60-70 kV and 57-86 μA

Imaging technique allows for comparison of skeletal development

Fig 3) 1-2. a.) Three dimensional images of rat tail midsection. 1-2. b.) Images of sagittal tail cross-sections through the center. 1-2. c.) Images of transverse tail cross-sections through the center of the bone (left) and the start of the end plate (right). 3-4. a.) Three dimensional images of rat hindlimbs. 3-4. b.) Sagittal cross-section of hindlimb.

Tendon fascicle crossover seen in younger and older, sexually mature rats

Fig. 4) 1-2. a.) Dehydrated, stained tail segment with hand-drawn fiber tracing from surface of tissue and 1-2. b.) through transverse cross sections throughout the length of the rat tail. 1 runs proximal to distal, while 2 runs distal to proximal.

Tendon insertion pattern visible in tails

Fig. 5) Pattern of tendon insertion in P50 (running from proximal to distal end). Inserting tendons are outlined in blue, insertion sites in green, and the next set of inserting tendons in white.
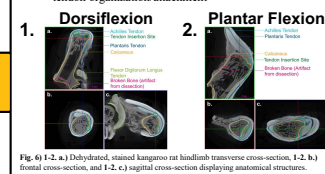
Discussion & Conclusion

- Bone calcification and tissue growth seen in age comparison
- Tendon crossover seen in both P21 and P50, however, yet to see tendon fascicles joining different bundles in P50
- Within tail, tendons insert into bone in a specific pattern
- New procedural findings, including stain stability and skin removal impact on stain penetrance

Further Applications

- Implications of method development includes:
  - Fascicle/fiber tracing across length of sample
  - Examination of tendon development across age groups
  - Analysis of tendons in different species/genotypes
- Examination of other animals/rat species anatomy
  - Analysis of kangaroo rat musculoskeletal system and tendon organization/attachment

Fig. 6) 1-2. a.) Dehydrated, stained kangaroo rat hindlimb transverse cross-section, 1-2. b.) frontal cross-section, and 1-2. c.) sagittal cross-section displaying anatomical structures.

Acknowledgements

Design, synthesis, and redesign of therapeutic peptides using SPPS, native chemical ligation, and AutoDock Crankpep

William Auten[1,2], Matt Womelduff[2], Kristopher Waynant[1], F. Marty Ytreberg[2], Darren A. Thompson[1,3]

[1]Dept. of Chemistry, University of Idaho, [2]Dept. Of Physics, University of Idaho, [3]Peptidaho Research Consortium
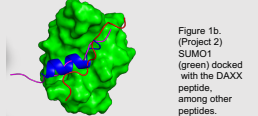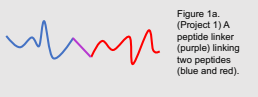
Abstract

The use of molecular modeling is quickly becoming an essential tool for the experimentalist. In this project, the molecular docking software AutoDock CrankPep (ADCP) was utilized to analyze a variety of parameters on peptide sequences to determine which are the largest contributors in producing active compounds when synthesized experimentally and which provide the most accuracy to the computational results. Our hypothesis is that ADCP parameters (whose exactness we are trying to figure out,) can be manipulated through inspiration from wet lab data to give scores more similar to real world values. This will then lead to faster, more efficient methods for the discovery of peptide-based drugs / biologically active compounds.

Introduction

Two Projects (Figure 1).

- **Project 1**: Linking peptides utilizing native chemical ligation and a novel peptide linker.
  - Resulting dipeptide has two C-termini.

- **Project 2**: Devise peptide modeling method for ADCP.
  - Current ADCP runs produce data that strays from *in situ* data.

Figure 1a. (Project 1) A peptide linker (purple) linking two peptides (blue and red).

Figure 1b. (Project 2) SUMO1 (green) docked with the DAXX peptide, among other peptides.

Acknowledgments

Project 1 Methods – Linker Synthesis and Native Chemical Ligation

Linker Synthesis

8 mmol Phenylenediamine + 4 mmol Succinic Anhydride → (4 hours, 4 mL DMF) → Linker Intermediate + 3 mmol NHS → (1 equiv DIC to NHS, 30 min) → Activated Linker

1. Linker-YGGFL Synthesis

o-aminoanilide linker + YGGFL-Resin → ~30 min → Linker-YGGFL

2. Diazotization

Linker-YGGFL → NaNO₂ → Acyl-benzotriazole

3. Thioesterification

Acyl-benzotriazole → MPOH → Thioester

4. Native Chemical Ligation[5]

Biotin + Thioester → Novel Related Analogue

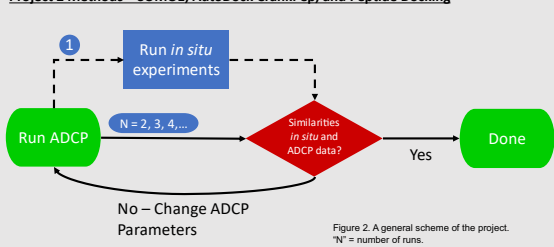## Project 2 Methods – SUMO1, AutoDock CrankPep, and Peptide Docking

Figure 2. A general scheme of the project. "N" = number of runs.
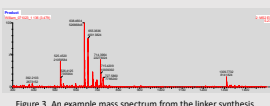
Project 1 Results

Figure 3. An example mass spectrum from the linker synthesis.

Project 2 Results

Figure 4. Comparison of several ADCP runs. The header of each column describes tested parameters of the run, these being the number of replicates and the number of Monte Carlo steps. The green highlights in the "Sequence" column represents residue mutations of the wildtype (DAXX), represented in the first row. The yellow and red highlights accentuate the highest and lowest affinity of each set of parameters/column respectively.

Discussion

Current Work:

- Currently generating *in silico* affinities using ADCP
- Currently collecting chromatograms and mass spectra for the peptide linker project

Future Work:

- Will use SPPS to synthesize peptides in ADCP work, then ITC to generate experimental binding affinities
- Finish collecting characterization data
- Finish collecting docking affinities *in situ* and *in silico*
- Compare *in silico* and *in situ* docking affinities and run different parameters to possibly determine which parameters allow for more accurate binding predictions

Work Cited

(1) *Targeting SUMO Signaling to Wrestle Cancer.* Jessie S. Kroonen 1 and Alfred C.O. Vertegaal.

(2) Cistrone, P. A.; Bird, M. J.; Flood, D. T.; Silvestri, A. P.; Hintzen, J. C. J.; Thompson, D. A.; Dawson, P. E. Native Chemical Ligation of Peptides and Proteins. *Curr. Protoc. Chem. Biol.* **2019**, *11* (1), e61. https://doi.org/10.1002/cpch.61.

Development of a bioreactor to mechanically stimulate cell self-assembled neotendons

Peter J. Wieber, Colin R. Marchus, and Nathan R. Schiele

Department of Chemical & Biological Engineering, University of Idaho, Moscow, ID

Abstract

Tendon injuries are common but current repair methods are inadequate. Even with surgical repair, scar tissue and incomplete tendon healing result in prolonged weakness which presents a risk for re-rupture. Thus, new regenerative treatment methods are required. To develop new treatments, mesenchymal stem cells (MSCs) have been explored in tissue engineering strategies to form tendon replacements. Our lab has developed a tissue engineering method to guide MSCs to form neotendons through directed cell self-assembly, which appear to mimic the early highly cellular stages of embryonic tendon formation. However, neotendon maturation in culture may be limited without the dynamic mechanical stimulation associated with *in vivo* muscle contraction. Applying mechanical stimulation is a unique challenge due to the neotendons' delicate nature and small size. To address this issue, a new bioreactor system was developed using Solidworks modeling software. The bioreactor was designed to be watertight and incorporate a novel loading mechanism that secures and applies tensile loads to the neotendon, while remaining compatible with an existing motor and controller. Before resin 3D printing the bioreactor, resins were tested for cytotoxicity, and minimal changes in cell viability were observed. Using this new bioreactor design, neotendons were dynamically stretched (1% strain at 0.1 Hz for 4 hrs) and the cell morphology was qualitatively evaluated, compared to static controls. Outcomes of this study will advance tendon tissue engineering strategies by improving our understanding of how mechanical loading impacts tendon formation.

Background

- Tendons are essential for the transfer of forces from muscle to bone
- Current treatments are ineffective often due to scar formation
- Much of the current stem cell research works to mimic adult tissue structures
- Mimicking embryonic tissue may be more clinically relevant when it comes to tissue regeneration and repair
- Mechanical stimulation has been shown to induce tenogenesis in MSCs
- Scaffold-free 3D cell culture has proved difficult to mechanically stimulate

Objective:

To develop a bioreactor system to mechanically stimulate 3D scaffold-free cell self-assembled neotendons and to assess how said stimulation alters cell morphology

Methods and Materials

Cytotoxicity Assay
- Resin samples were designed using Solidworks 3D modeling software and were 3D-printed on a Stratasys J850 printer
- Potential volatile organic compounds and cytotoxic residues were extracted by incubating resin samples in cell culture media for 1, 2, and 3 days
- Cells were grown with resin extract media in a 12-well plate for 3 days
- Cytotoxicity was qualitatively assessed following ISO 10993-5:2009

Bioreactor Design Methods
- Components were designed using SolidWorks 3D modeling software
- Bioreactor was 3D-printed with VeroClear™ (VC) Resin on a Stratasys J850
- Accessory components were 3D-printed with PLA plastic on an Ender 3 printer

Mechanical Stimulation Methods
1. Neotendon were grown at a seeding density of 3 million cells per construct
2. After 48 hours of preculture, neotendons were loaded into bioreactor chamber
3. Neotendons were cyclically loaded with 1% strain at 0.1 Hz for 4 hours
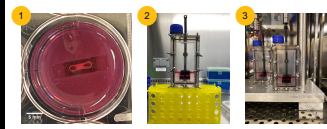
Design Criteria and Implementation

Soft requirements
- Accessories fit in a 50 mL centrifuge tube for easy storage and sterilization (1-5, 7, 9)
- Maintain alignment for future mechanical property test applications (1-3, 5-7)
- Minimize media usage (8, 9)
- Minimize cell trauma (1, 3, 5-7)
- Simplify loading procedure (1-9)
- Reduce undesired mobility (1-9)
- Reduce alterations to the existing neotendon architecture (3, 6, 7)
- Improve reproducibility (1-9)
- Easy to adapt/modular design (1-5, 7-9)

Hard requirements
- Non-cytotoxic (1-9)
- Watertight (6)
- Compatible with existing motor and controller (1-9)
- Able to be disinfected (1-9)
- Secure neotendon without slipping or breaking (3, 4, 6-9)
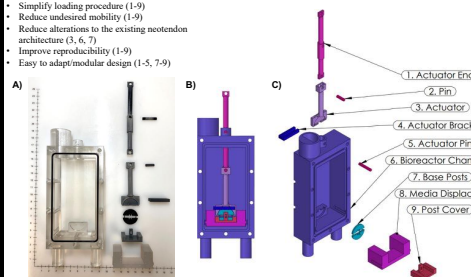- Allow for gas diffusion (6)

1. Actuator End
2. Pin
3. Actuator
4. Actuator Bracket
5. Actuator Pin
6. Bioreactor Chamber
7. Base Posts
8. Media Displacer
9. Post Cover

Figure 1: Real images and digital render of the bioreactor assembly. If a part meets a criteria, the part number is listed under said criteria. **A)** Exploded view (Ruler increments = 1 cm). **B)** Front view render. **C)** Exploded view render with labels.

Minimal Cytotoxicity from VeroClear™ Resin

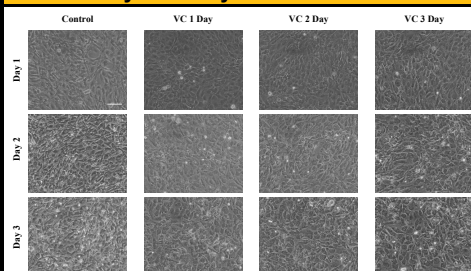Control | VC 1 Day | VC 2 Day | VC 3 Day

Day 1 / Day 2 / Day 3

Figure 2: Phase contrast images of cells following treatment with resin extracts. Following the qualitative grading scale from ISO 10993-5:2009, only slight reactivity was observed from culturing with VeroClear™ (VC) Resin extract. This is indicated by low levels of cell death and only slight growth inhibition. Column labels indicates the length of time that the resin was incubated in the media (1, 2, and 3 days respectively) while row labels indicate the length of the treatment with the extract. Scale bar = 200 µm.
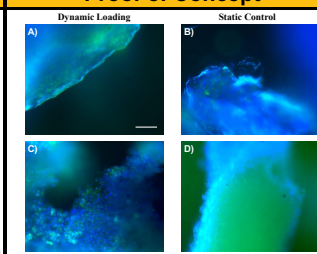
Proof of Concept

Dynamic Loading | **Static Control**

A) B) C) D)

Figure 3: Representative fluorescent images of neotendons. **A, C)** Neotendon post mechanical stimulation. **B, D)** Static control neotendon. While cell nuclei are present, and actin is being produced, it is difficult to comment on cell morphology. Blue = DAPI stained cell nuclei. Green = Phalloidin stained actin cytoskeleton. Scale bar = 200 µm

Discussion

- VC Resin was non-cytotoxic and did not leak
- Components successfully integrate with the motor and controller
- Ethanol sterilization led to bioreactor deterioration
- Bioreactor is capable of providing mechanical stimulation to the neotendons
- Loading and collecting process avoids breaking the neotendon
- **Design is feasible for future mechanical studies**

Conclusion and Outlook

- Resolve neotendon disintegration
  - Longer preculture
  - Treat with TGF-β2
- Address durability issues
  - Sterilize by autoclave, UV
  - Explore alternate materials
- Improve imaging process to reduce blurring and improve qualitative analysis
  - Remove neotendon from uneven agarose
  - Spinning disk confocal microscope to address the 3D nature of the neotendons
- Western blot for quantification of protein

Figure 4: Characteristic problems seen over the course of the experiments. **A)** Breakdown of the neotendon after fixation in 10% formalin. **B)** Flaking on the periphery of the bioreactor.

Acknowledgements

WIRELESS TOWER OF LIGHTS

An undergraduate research presentation for the Idaho Conference of Undergraduate Research, conducted at **University of Idaho**

THE SITUATION

Every year, the University of Idaho hosts a lights show in the windows of the eleven-floor Theophilus Tower after the Homecoming serpentine march. Forty rooms are equipped with LED lights situated on wooden beams. All of these beams are connected into the wall through wire, which routes down with more wire to a networking closet in the basement, where more wire is connected to a large microcontroller, or the "brains" of the operation. *This is too much wire.*

The microcontroller communicates with all of the rooms to light up the LEDs, changing according to code written in C++, which is connected to a laptop via a lengthy wire out to the field where the music and lights display is broadcasted.
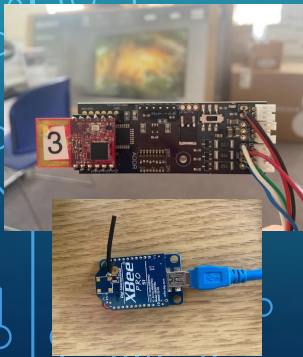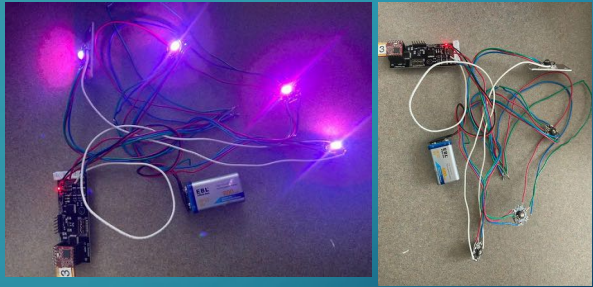
THE PROBLEM

This is a lot of wire to deal with. Not only does it make it difficult to set-up, all connections need to be ensured, and there is a small loss of energy through the wires to the top-level floors. To make the set-up for the Tower of Lights show easier, I collaborated with Dr. Rinker to create a wireless set-up for the Tower of Lights. To do this, we needed to think through two different problems: power utilization and wireless communication. Now that the power wasn't being drawn from a wired set-up, we needed to think how the LEDs could supply their own energy. For power, we re-designed our LED circuit to accommodate a common nine-volt battery. For communication, we utilized a wireless transmitter device that connects into the laptop and forty wireless transceivers attached to a small microcontroller, one for each room.

THE RESULTS

By switching to wireless technology using transceivers, we were able to address the communication challenges and overcome the issue of efficiency through wireless technology. We re-formatted the communication by placing hand-held microcontrollers on each beam and adding a transceiver to each. The mother of all transceivers that communicates to all is a transmitter that is connected via USB into a laptop using the Linux operating system. By re-designing the LED circuits, we were able to provide power to each LED beam and also add an extra LED to each beam. Setting two sets of two LEDs in parallel, or side-by-side, rather than in series, or a line-up, made the power running through the circuit more efficient without noticeably reducing luminous intensity.

Reaped Electromagnetic Energy

DEPARTMENT OF ELECTRICAL AND COMPUTER ENGINEERING, UNIVERSITY OF IDAHO

Juwon Elelu

Abstract

Radio Frequency (RF) can be said to be a collective term for electromagnetic energy. This form of energy is derived from electromagnetic radiation which travels through space between a transmission antenna and receiving antenna. The receiving antenna only captured a small amount of RF energy.

The purpose of this research is to design and build a system that can harvest ubiquitous unused RF energy and convert it to useful electricity. The device was composed of several units. First was the antenna responsible for collecting the Electromagnetic energy from the environment as AC current. In order for the device to power a load the current has to be in DC, and so an AC to DC converter or rectifier was used. This formed the second unit of the device. The last unit of the device, the microcontroller served as the load that was programmed to store a certain amount of energy in a capacitor by switch mode control. These units were designed individually, assembled as a circuit, then tested to get results.

Introduction

We are surrounded by lots of devices that use RF energy in our day-to-day activities. Such devices include mobile phones, remote controls, microwaves, Wi-Fi, radio transmission towers etc. Most of these devices make use of batteries as a source of energy or as backup energy. These batteries are changed from time to time. The used batteries are disposed of in such a way that they cause environmental pollution. So, the need for alternative sources of energy for powering these devices cannot be overemphasized.
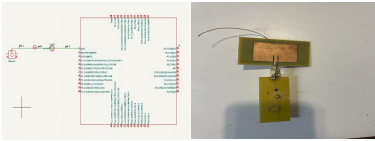
Figure 1: Battery pollution https://gsiwaste.com/battery-recycling-is-important-for-environmental-health/

Figure 2: Electromagnetic energy harvesting system

Results

- Both Antenna and circuit board were designed and produced in hopes of collecting frequencies of 2.4GHz which is used in Wi-Fi.

Figures 3 & 4: Circuit board schematic and its physical form

- An oscilloscope was used to check the antenna and rectifier for signs of DC current being produced. Results show that they succeeded in performing their respective tasks. But in addition to this it was discovered that the diode also filters the output which was an unexpected behavior but useful. Useful in the sense that it could also act as a capacitor, allowing us to no longer need an actual capacitor and thereby saving resources
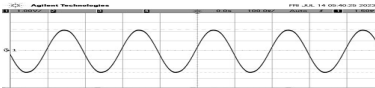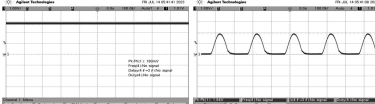
Figure 5: Result from reading of Antenna

Figure 6: Result of input current before measuring rectifier

Figures 7 & 8: Result of output when measuring rectifier but with presence of filtration [6]; and result of output without presence of filtration

- The last unit, the microcontroller we picked, was an MSP430FR2476. According to the datasheet, this microcontroller was designed for a nominal power of 600uW which was acceptable for our purpose. It also had potential for switch mode control. Research was done to understand the microcontroller and how to control the pins, so a simple LED blink program was made as a start, serving as proof our original program is achievable.
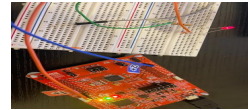
Figure 9: External LED blink program using MSP430FR2476 microcontroller

Conclusions and Future Directions

In summary of what has been accomplished so far. I designed and simulated my antenna with the aid of HFSS software to determine which material would be most suitable to make the antenna. From there I proceeded to search for a Schottky diode suitable to convert AC to DC, then I proceeded to making my pcb to allow me to test the parts for DC using an oscilloscope. Results showed that we were getting DC as an output which was a success, but this also led to the discovery that the diode could filter output, thereby acting as a capacitor. It was unexpected but it played in our favor and saved us resources. I used an oscilloscope to determine whether we were successfully getting DC from our circuitry and the outcome was a success. As for the microcontroller I was able to program it to control an external hardware which sets the basis for future work.

There are rooms for improvement for both the device and myself. I hope to finish the original program after better understanding the MSP430, from there I can combine all units of the device to make it whole and functional. From there I hope to include a sensor into the device to create room for more experiments. And as time goes future researchers can try new things as well.

References

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- Hart, D. W. (1997). Introduction to Power Electronics. Prentice-Hall, Inc.
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Learn more about me and my research project at the following website:

Tailocin treatment of American Foulbrood Disease in honeybees

Department of Biological Sciences | University of Idaho

Katherine Ryssel, James Van Leuven, Tracey Peters

ABSTRACT

Tailocins are antibacterial proteins produced by some bacteria in order to kill closely related bacterial strains. In nature, this benefits the microbe by offering a way for them to control the bacterial composition of their environments. However, in a lab setting, researchers can use these tailocins to treat against unwanted bacteria. The purpose of this research project is to test whether a tailocin treatment is a viable option in protecting honeybees against the deadly American Foulbrood disease, which is caused by the bacterial species *Paenibacillus larvae*. To test this, the proposed research will involve three major parts: Collection and purification of tailocins from honeybee pathogens, in vitro analysis of tailocins against strains of *P. larvae*, and *in vivo* treatment of infected bees by tailocins. This work addresses the urgent need to develop novel antibacterials and reduce our dependence on traditional antibiotics. Currently, research remains in the *in vitro* stage of testing.

METHODS

- 7 strains of *P. Larvae* were labeled A-E.
- Samples were grown into day cultures.
- Day cultures were subjected to stressful conditions to induce bacteriocin production.
- Samples were filtered to isolate the any tailocin-like particles
- Samples were spotted onto lawns of the original bacterial strains.
- Lawns were examined for inhibitory activity.

RESULTS

Five of 7 strains showed activity, but only 2 gave replicable results which resembled the inhibition of growth associated with tailocins. Strain B showed activity against four of the seven strains. One of the strains it did not show activity against was itself, which was expected as producing strains are resistant to their products. Strain D showed inhibitory activity against two strains, including one of the strains that B was ineffective against. D also showed prophage activity against strain C. Additionally, strains A and E both showed very light activity against B when lawns had very low bacterial concentrations. Strain G showed activity against 4 other strains in one test, but the results were never able to be replicated.
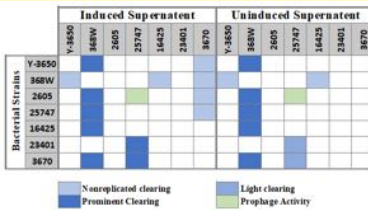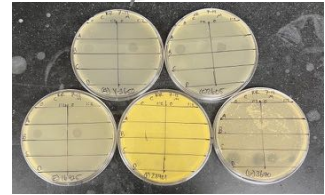

Figure 1. This matrix shows the results of spot testing supernatants against bacterial lawns. The left half of the figure represents induced samples, while the right half represents uninduced controls.

FUTURE WORK

- Activity from samples must be confirmed at tailocin activity by TEM imaging.
- DNA sequences will be obtained and used to locate gene clusters responsible for tailocin production.
- Gene clusters will be used to clone tailocin proteins and create samples of a higher concentration.
- *In vivo* experiments: Infection of honeybees with *P. Larvae* and treatment with active tailocins.

The hope is that these tests will prove useful in the development of a possible commercial treatment of *P. Larvae* infections in hives.

No Fire on the Mountain: Has fire suppression caused population declines of an imperiled ground squirrel?

A Test of the Fire Suppression Hypothesis

Ecology and Conservation Biology
University of Idaho livi8992@vandals.uidaho.edu

Alyssa Livingston
Courtney J. Conway
Eva Strand

Abstract

The Northern Idaho Ground Squirrel (*Urocitellus brunneus*) is listed as threatened under the Endangered Species Act. The prevailing hypothesis for the species decline is encroachment of trees into its habitat due to fire suppression. However, this hypothesis has not been rigorously tested. Using geographic information systems and aerial imaging, I compared changes in abundance of the squirrels' populations to changes in forest canopy coverage over the past 40 years. These comparisons allowed me to test the prevailing hypothesis for the population declines of this rare species by testing the prediction that forest fire suppression has allowed tree growth and encroachment into the squirrel's habitat. This hypothesis predicts that the areas with the most pronounced population declines of Northern Idaho Ground Squirrels will have the most pronounced forest encroachment in the areas used by these squirrels. Our results indicate a 50% increase in tree cover over the colonies and the historical range for the last 40 years.
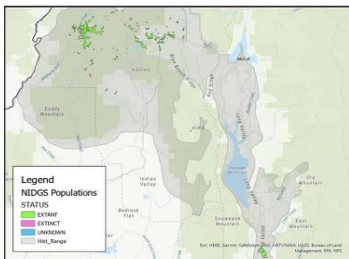
Project Description

The Northern Idaho Ground Squirrel (NIDGS) is a threatened species endemic to central Idaho. With already small numbers dropping, the species is threatened by extinction. The main hypothesis for population decline is habitat loss due to fire suppression, causing more trees to grow and encroach into natural clearings, which are the squirrel's habitat. This leads to a loss of suitable habitat for the squirrel and subsequent population decline. This hypothesis, however, has never been directly tested. The main objective for this project is to test the prevailing hypothesis for the squirrel's population declines.

Methods

- Programs utilized for this project: ArcGIS, Rangeland Analysis Platform, Google Earth Engine, and aerial photographs from the USDA over the past 40 years.
- Obtained ground squirrel count data from state and federal agencies to document variation among colonies in population declines.
- Documented change in tree cover over the past 40 years within Northern Idaho Ground Squirrel 1) historical range and 2) mapped colonies.
- Synthesized all these data to determine if the encroachment of trees is a likely cause of the squirrel's decline in abundance.
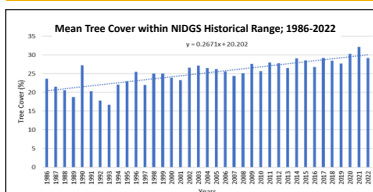

Figure 1: Map of Northern Idaho Ground Squirrel (NIDGS) colonies illustrating their status – extant, extinct, or unknown.

Figure 2: Change in the mean tree cover for the historical range of Northern Idaho Ground Squirrels (NIDGS) from 1986-2022.

Results

Two main analyses were run on ArcGIS Pro: zonal statistics of change in tree cover over the past 40 years within the historical range of the Northern Idaho Ground Squirrel and within the squirrels' colony areas only. Results of the first test show that there has been an increase in tree cover in the historical range over the past 40 years. The effect size shows a 50% increase in tree cover through the entirety of the historical range. Results of the second test show that there has also been an increase in tree cover in the squirrels' colonies over the past 40 years. The effect size again shows a 50% increase in tree cover within extinct and extant colonies.
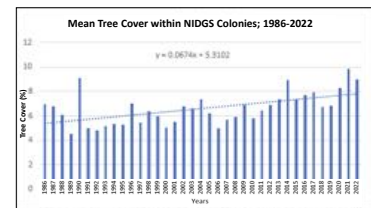

Figure 3: Change in the mean tree cover within colony footprints of Northern Idaho Ground Squirrels (NIDGS) from 1986-2022.

Conclusions

The fire suppression hypothesis – that squirrel population declines are caused by tree encroachment due to fire suppression - was supported based on 2 predictions. Forest canopy cover has significantly increased over the last 40 years in both the historical range of the Northern Idaho Ground Squirrel and also in the explicit colonies within their range. Canopy cover within the historical range (15% cover) was higher than canopy cover within the actual colonies (5% cover). Experts on this rare species believe that these squirrels need areas with <15% canopy cover.

Synthesis and Structural Characterization of Zinc Complexes with N,S Donor Ligands

DEPARTMENT OF CHEMISTRY, UNIVERSITY OF IDAHO

Mason Kilker, Alina Andrasi, and Sebastian Stoian

University of Idaho

University of Idaho — Office of Undergraduate Research

Abstract

Zinc (Zn) complexes are diamagnetic species with no unpaired electrons sought after for their unique chemical and biological applications as enzyme analogues or bioinspired catalysts; as such, Zn complexes are relevant to several different areas in pathophysiology and bio-inorganic chemistry.

The inorganic field of metal complexation has a diverse and important role in biological functions. For example, the binding of molecular oxygen (O_2) to hemoglobin involves forming coordinate bonds at alveoli of the lungs and breaking them in the tissues, while O_2 remains bound to the heme group during transport[1]. The exact signaling mechanism has not been reported in literature[1].

Alkaline phosphatases are hepatic enzymes distributed widely in mammalian cells[2]. They are Zn-based complexes that are thought to have a role in cell signaling and catalyzing protein folding, though their chemistry and functions are not well understood. High mammalian alkaline phosphatase levels have been associated with liver and heart disease, with any physiologic role yet to be underlined[2].

Given the Zn-regulated biological functions, it is important, of course, to acquire a fundamental understanding of model Zn-complexes and their properties in order to apply them further. The main goal of this short summer project was to prepare several novel series of Zn complexes and to elucidate their coordination modes, structures, and properties in solution and in solid state.

Introduction

The main goal of the project was to synthesize several Zn(II) complexes with a coordination number of 6. The complexes were to incorporate the tetradentate ligand S,S'-**b**is(2-**p**yridylmethyl)-1,2-**t**hio**e**thane (**bpte**) with two anionic co-ligands NCX, where X=O; S; and BH_3.

Zn(II) used for these reactions was sourced from zinc perchlorate hexahydrate salt. Anionic co-ligands were obtained from their sodium or potassium salts, and **bpte** was synthesized using published methods [3].

X-ray crystal diffraction at 100K was used to determine the composition of the complex in solid state, and ^{1}H-NMR was used to determine the composition in liquid state (DMSO-D6 solvent). NMR was also used to verify the purity of **bpte**.

Several crystallization techniques were employed, including vapor diffusion and slow evaporation.
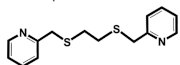
Figure 1: Structure of the bpte ligand

Results

Several novel Zn complexes were synthesized and characterized using x-ray crystallography and NMR. All compounds are hexa-coordinated and exhibit an octahedral geometry. The compounds were synthesized by allowing Zn perchlorate hexahydrate to react with bpte for 20 min., followed by the addition of a potassium or sodium salt of the anionic co-ligand.

[Zn(bpte)(NCBH$_3$)$_2$]

This octahedral zinc complex was isolated as clear block crystals and exhibits a slightly distorted octahedral geometry around the metal center with an N-Zn-S angle of ~80°.

X-ray Crystallography

Single crystals were sent for x-ray diffraction. The result, as compared with the anticipated drawn structure, is shown below.
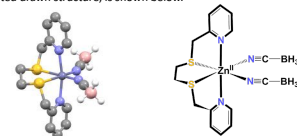
Figure 2: X-ray crystallography structure determination (left) and drawn structure (right) of [Zn(bpte)(NCBH$_3$)$_2$]. Hydrogen atoms other than those on the co-ligand were omitted for simplicity.

Nuclear Magnetic Resonance (NMR)

The NMR spectrum (below) was recorded for a sample obtained by dissolving several single crystals in DMSO-D6. The NMR indicates that the complex structure observed in solid state is maintained in solution.
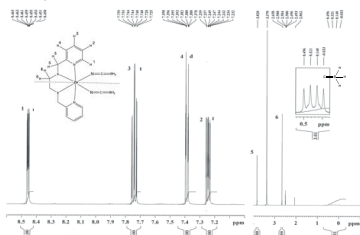
Figure 3: Nuclear magnetic resonance spectrum of [Zn(bpte)(NCBH$_3$)$_2$].

[Zn(bpte)(NCS)$_2$]

This octahedral Zn complex also exhibits a slightly distorted octahedral geometry around the metal center. Single crystals were sent for x-ray diffraction. The result, as compared with anticipated drawn structure, is shown below.
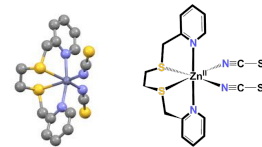
Figure 4: Comparison of the X-ray single crystal (left) and ChemDraw® (right) structures of Zn(bpte)(NCBH$_3$)$_2$.

Conclusions and Outlook

1. A series of zinc complexes with a N,S donor ligand have been synthesized.
2. X-ray diffraction has been used to elucidate the solid-state structure.
3. Nuclear magnetic resonance has shown the structure to be rigid and maintained in solution.
4. Future work will explore other Zn complexes by attempting to make a dimer.
5. We will also explore other metals such as copper (II) and cobalt (II) for future structural analysis.
6. Further, we plan to study the interaction of these diamagnetic Zn complexes with biomolecules such as deoxyribose nucleic acid (DNA) or ribose nucleic acid (RNA).

References

[1] Carbon Monoxide Poisoning: Pathogenesis, Management, and Future Directions of Therapy: Rose, J.J. et. al.: Am J Respir Crit Care Med, pg 596, 2017; Vol. 195.

[2] Alkaline phosphatase: beyond the liver: Fernandez N. et, al: Veterinary Clinical Pathology; 3rd edition (pg 223-227); 2007; Vol. 36.

[3] 1,2-Bis[(pyridin-2-ylmethyl)sulfanyl]-ethane and its dimorphic hydro-chloride salt: Lennartson A. et. al.: Acta Crystallographica Section C, 9th edition (pg m287-o372); 2011; Vol. 9

Selective Reflections for the Nelder-Mead Algorithm

Zaiden Espe   Department of Mathematics, University of Idaho

University of Idaho

Abstract

The Nelder-Mead Algorithm is an established algorithm to search for a minimum to a function. Since its first description, the algorithm has had various improvements made to it. With this project, I searched for improvements to the algorithm through changes to its reflective direction and geometric structure. I found in testing that directions that maintain the shape of the algorithm's search structure perform better than those that do not. The algorithm's speed can also be increased in higher test dimensions by increasing the number of points in the simplex by one. With these alterations, the Nelder-Mead algorithm can find solutions to problems it previously could not and do so with fewer function evaluations.

Introduction

The Nelder-Mead method optimizes a function of n variables by creating a simplex with n+1 points in an n-dimensional space. It then uses a combination of five steps to transform the simplex by moving the point with the worst evaluation to a hopefully better location. The direction in which it moves this point is what this project focused on studying. The standard algorithm moves the point in the direction of the center of the rest of the points, called the centroid. The centroid is calculated by taking the mean of the n-best points. Once the last ranked point has been moved, the algorithm starts again with the new set of points and iterates until a stopping condition is met. In a single iteration, the basic algorithm does not directly use a lot of information that is stored within the simplex.

Methods

In a single iteration, the basic algorithm does not directly use all the information that is stored within the simplex. The algorithm overlooks the fullest extent of rank and value of the n-best points during an iteration. To use this information, I calculated new direction vectors to use as a basis for the reflection. To gather more information, I added an extra point to the simplex as a second point of reflection and used another mirror reflection to allow more movement of the simplex.
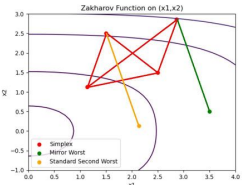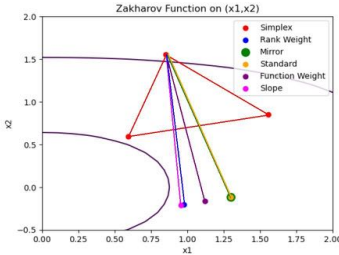
Figure 1. An example of the reflection and mirror steps on an n+2 point simplex evaluated on the Zhakarov function.

Figure 2. An example of each of the five tested reflections given a simplex evaluated on the Zhakarov function.

Results

This project found that the additional point to the simplex improved the performance of the base algorithm without any changes to the reflective direction as the dimension of the function increased. The mirror transformation used with the extra point also helped the algorithm perform faster on multiple test functions and find more accurate minimums. However, more aggressive reflections that did not uphold the shape of the simplex as closely as the standard reflection or mirror reflection would often cause the algorithm to fail to find an acceptable minimum or perform within an acceptable speed.

		Standard NM		Combination NM	
Function	Dim	f-min	f-eval	f-min	f-eval
Quadratic Sum	20	2.314075e-16	3406.3	8.677412e-17	2713.0
Quadratic Sum	50	5.711390e-16	10720.7	1.845766e-17	8503.3
Quadratic Sum	100	8.047840e-16	25268.9	2.882725e-17	11907.0
Zakharov	20	2.706404e-16	10146.6	8.644623e-16	3183.2
Zakharov	50	4.287115e+01	50000.1	2.887030e-15	7065.0
Zakharov	100	3.349520e+03	50000.3	4.790109e-15	13240.1
Dixon-Price	20	6.666667e-01	4050.6	6.666667e-01	29507.9
Dixon-Price	50	6.666667e-01	16461.5	6.667530e-01	50000.5
Dixon-Price	100	6.683384e-01	50000.1	6.937013e-01	50000.0
Rosenbrock	20	3.986624e-01	23634.1	9.347150e+00	50000.1
Rosenbrock	50	4.261088e+01	50000.1	4.657021e+01	50000.0
Rosenbrock	100	9.510262e+01	50000.0	9.703387e+01	50000.7
RH_Ellipsoid	20	1.804961e-15	3547.4	5.376130e-16	2779.9
RH_Ellipsoid	50	5.784937e-16	11366.7	5.304931e-16	6778.2
RH_Ellipsoid	100	1.759490e-14	30437.1	1.978980e-15	11952.2

Figure 3. Data collected comparing the standard Nelder-Mead algorithm to the one found in the project. Note that "Dim" stands for dimension of the test function. "RH_Ellipsoid" stands for the Rotated Hyper-Ellipsoid function.

Conclusions

These results open avenues to understanding the functioning of the Nelder-Mead algorithm and improving its design. Future research can be done to explore the effect of using even more points in the simplex, new transformations that continue to maintain the shape of the simplex and fine-tune the selection of multiple types of reflections in a single algorithm run.

References

Nelder, J. A., & Mead, R. (1965). A simplex method for function minimization. *The Computer Journal*, 7(4), 308–313. https://doi.org/10.1093/comjnl/7.4.308

Appendix 2

The Folk Influences on the Piano Works of Manuel M. Ponce

Alexandro Aguilar (agui6621@vandals.uidaho.edu) and Dr. Roger McVey (rmcvey@uidaho.edu)

University of Idaho
Lionel Hampton School of Music

Motivation and Project Background

The compositions of musicians from underrepresented backgrounds have gone largely overlooked in the core literature for piano and in academic fields such as musicology and theoretical analysis. Inadvertently, the field of music has missed out on rich cultural inspiration by doing so. It is important to highlight music from people of color and help expand the scope of music as a studied form of art if the study of classical piano were to stay relevant in modern day.

For this project, I prepared four pieces for solo piano composed by Mexican composer, Manuel Maria Ponce, for performance. Research was done on the history of native folk music in Mexico and how it influenced Ponce's works for the piano.

The four works of focus throughout the project are Scherzino Mexicano, Romanza De Amor, Quatro Danzas Mexicanas, and Intermezzo No. 1. Latin folk genres of music and dance such as **Huapangos, Sons, Cuban Contradanzas, Jarabes**, and **Corridos** were discovered to have heavy influence in the rhythmic, melodic, sonic, and harmonic motifs found in Ponce's compositions for the piano.

Manuel M. Ponce (1882-1948)

- Born on December 8, 1882, in Fresnillo, Zacatecas
- Began studying the piano at age 6
- Made his first composition at age 9
- Attended the National Conservatory of Music in Mexico City in 1901 for one year
- Traveled to Europe in 1904 to study composition and piano in Italy and Germany
- Returned to Mexico City in 1908 and taught composition and music history at the National Conservatory
- In 1923, Ponce met Spanish guitarist, Andres Segovia, a lifelong friend who helped spread Ponce's influence as a composer around the world.
- Moved to Paris in 1925 for 7 years to work on his compositional technique.
- Moved back to Mexico in 1933 and spends the last 15 years of his life composing and teaching at the National Conservatory.
- Manuel Maria Ponce passed away in Mexico City on April 24, 1948

The Music of Mexico

The Mexican composer Carlos Chavez gives a definition to the term "Mexican Music" as "the native music of the ancient Mexicans, the music of Spanish or other origin implanted in Mexico, and the production in Mexico of a mixture of these elements."

Sons/Huapangos

- Mexican folk music and dance genre identifiable by the **sesquialtera** rhythmic motif and zapateado (tap dance) dancing
- Simultaneous 2 against 3 rhythmic motifs and alternating ternary and binary rhythms
- The **Huapango** is a regional sub-genre of the **Son**, known as the "Son Huasteco"
- The "Son Jaliscience" originating in the region of Jalisco evolved into what we know as modern **Mariachi** music.
  - Call and response melodic motifs, originating from work songs sung by field workers, influenced the Son Jaliscience, and later, mariachi music.

Ex. 1 & 2: Common rhythmic motifs found in the Huapango, both examples of a sesquialtera or hemiola

Corridos

- Mexican ballad genre identifiable by its narrative story telling
- Falsetto style vocals
- Melodies typically resolving in thirds
- Circle of fifths-based harmony
- Melodies voiced in thirds
- Chromaticism treated as ornamentation to melody,

The Piano Works of Manuel M. Ponce

Scherzino Mexicano (1909)

- "Dominant-Tonic" or circle of fifths based harmony, familiar to late 1800s to early 1900s Mexican **salon** music
- A melody voiced in intervals of thirds, akin to Mexico's early **Corridos**
- Familiar than Ponce frequently used in his works for solo piano
- **Huapango/Son** influenced alternating ternary and binary rhythms (sesquialtera)
- Mariachi-influenced call and response phrase structure

Ex. 3: Scherzino Mexicano - Measures 1-5

The Piano Works of Manuel M. Ponce (cont.)

Quatro Danzas Mexicanas (1941)

- Syncopated rhythms familiar to **corridos** in the A section
- Ambiguous chromatic harmony
- Two-part AAB form influenced by Cuban **Contradanzas**
  - A as a faster introductory theme
  - B as a slower and more contrastive theme
- Four and eight measure phrases
- Alternating groupings of duple and triple **Habanera**-based rhythms, familiar to the **sesquialtera**.

Ex. 4: Quatro Danzas Mexicanas - Danza I - Measures 28-31

Conclusions and Future Directions

A lecture recital will be held at the Lionel Hampton School of Music where I will present my research on the traditional folk music of Mexico, a biographical overview of Manuel Maria Ponce, and I will perform the four pieces of music that were of focus throughout this project, Intermezzo No. 1, Quatro Danzas Mexicanas, Scherzino Mexicano, and Romanza De Amor.
I will also plan on applying to present my research at poster sessions hosted at music pedagogy conferences around the country such as the MTNA National Conference.

Scan this QR Code for a list of musical references pertaining to this project:

References

1. Guerra, Dahlia Ann. "Manuel M. Ponce: A Study of His Solo Piano Works and His Relationship to Mexican Musical Nationalism". ProQuest Dissertations Publishing, 1997.
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Anxiety and the Effectiveness of Misinformation Fact-Checks

DEPARTMENT OF POLITICS AND PHILOSOPHY, UNIVERSITY OF IDAHO

Kendall Mitton

Abstract

As levels of misinformation rise in the United States, so does the threat to efficient and effective policy-making. Most Americans are subject to misinformation, causing them to actively think/behave against the recommendations and empirical findings of experts. That said, in the absence of misinformation fact-checks and the presence of individual defiance toward corrective information, a variety of problems have been fostered within our borders (e.g., events like *Pizzagate and* conspiracies about Covid-19). Anxiety may be a factor contributing to this phenomenon, but it has been largely overlooked in previous research. Building off existing scholarship surrounding these variables, I conducted an online survey experiment to test the relationship between anxiety and fact-checks on individual perceptions. After analyzing my results, I found increased levels of anxiety and the presence of fact-checks do not have a significant effect on an individual's perception about the Covid-19 vaccine. However, anxiety itself has a negative and significant impact on an individual's perception about the effectiveness of the Covid-19 vaccine.

Overview

Background:
- What factors contribute to an individual's belief in misperceptions?
- Previous research shows...
  - Misinformation in the U.S. has created adverse complications for a range of different salient issues in politics, science, and medicine (Flynn et al., 2017).
  - In general, human emotions can have a significant impact on our actions and beliefs (Albertson & Gadarian, 2015).
- *Hypothesis: I hypothesize an increase in the level of anxiety individuals have will lead to a decrease in the effectiveness of a misinformation fact-check.*

Research Design:
- Nationally representative online survey experiment
- Original Survey Data: June 2023
- 607 respondents
- 2x2 between-subjects experiment
  - Independent Variables:
    - Anxiety Treatment (i.e., Artificial Intelligence article)
    - Anxiety Control (i.e., Virgin-Birth in Crocodile article)
    - Misinformation Treatment (i.e., Facebook post about the Covid-19 vaccine being harmful)
    - Misinformation Control (i.e., fact-check from PolitiFact about the Covid-19 vaccine being helpful)
  - Dependent Variables:
    - 5 opinion questions measured with a Likert scale

Results

- Interaction Analysis (Figure 1)
  - Anxiety Dummy: respondents induced with anxiety
  - Fact-Check Dummy: respondents presented with fact-check
  - Interaction: respondents from both groups above
- Figure 1 shows...
  - There is a negative but significant correlation between anxiety and individual perceptions. Substantively, a 1-point increase in a respondent's level of anxiety leads to a .35-point decrease in that respondent's average perception about the effectiveness of the Covid-19 vaccine.
  - Neither the presence of a fact-check nor the combination of both anxiety and a fact-check have a significant effect on an individual's perceptions about the Covid-19 vaccine.
  - A respondent only having a high school diploma, or being a republican, has a negative and significant impact on individual perceptions about the Covid-19 vaccine.
  - A respondent being white has a positive and significant impact on individual perceptions about the Covid-19 vaccine.

Negative Impact →
Positive Effect →
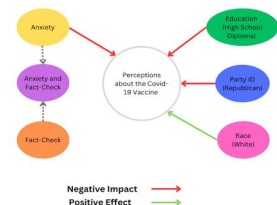
Conclusions and Future Directions

- Does the presence of anxiety and misinformation fact-checks influence an individual's perceptions?
  - My research suggests no (kind of).
- This could likely be due to the topic (i.e., Covid-19 vaccines) I chose.
- In future research, one should explore the effects of other emotions that may lead an individual to hold misperceptions. Furthermore, one could examine other topics where common misperceptions are held.
  - For example, what effect does trait aggression have on an individual's misperceptions. Or what factors cause people to hold misperceptions about election fraud in the US?

Figure 1

	Dependent variable: Individual Perceptions
Anxiety Dummy	-0.35***
	(0.11)
Fact-Check Dummy	-0.09
	(0.11)
Interaction	0.08
	(0.15)
Age	0.004
	(0.002)
Gender (Male)	0.09
	(0.08)
Education (High School Diploma)	-0.40***
	(0.10)
Party ID (Republican)	-1.08***
	(0.10)
Race (White)	0.64***
	(0.11)
Average Income ($80,000 - $84,999)	0.09
	(0.29)
Region (West)	-0.06
	(0.10)
Constant	3.04***
	(0.16)
Observations	607
R^2	0.25
Adjusted R^2	0.24
Residual Std. Error	0.93 (df = 596)
F Statistic	19.77*** (df = 10; 596)
Note:	*p **p ***p <0.01

References

Flynn, D. J., Nyhan, B., & Reifler, J. (2017). The nature and origins of misperceptions: Understanding false and unsupported beliefs about politics. *Political Psychology, 38*, 127-150.

Albertson, B., & Gadarian, S. K. (2015). *Anxious politics: Democratic citizenship in a threatening world*. Cambridge University Press.

Missing and Murdered Indigenous Women, Girls, and Two-Spirits in the United States

Amara Bailey (bail8404@vandals.uidaho.edu)
Dr. Omi Hodwitz (omi@uidaho.edu)
Department of Culture, Society, and Justice

University of Idaho
Department of Culture, Society and Justice

Research Goal

The United States has a history littered with abuse against Indigenous populations. Today, there is an epidemic of Missing and Murdered Indigenous Women, Girls, and Two-Spirit individuals (MMIWG2). Despite Indigenous Women, Girls, and Two-Spirit individuals being at greater risk of victimization than other populations, their disappearances and murders are under-documented (Hodwitz & King, 2021). The MMIWG2 database aims to resolve the information deficit surrounding this phenomenon by being an exhaustive culmination of existing data from community-led efforts. Through the collection and subsequent authentication of each individual's case, the MMIWG2 database provides much-needed context to this epidemic for governing institutions in the United States to give this issue the time, attention, and resources it deserves. In addition to identifying the trends of this phenomenon, the MMIWG2 database provides an opportunity to analyze the response and effectiveness of law enforcement.
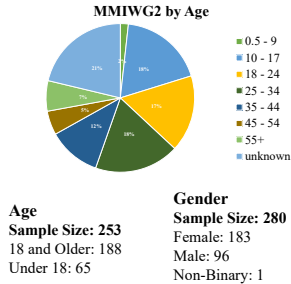
Research Questions

What is the demographic breakdown of MMIWG2s?
What is the demographic breakdown of their offenders?
Given the role of tribal police in law enforcement, how many incidents occur on reservation and require their involvement?
How many MMIWG2 cases are resolved?

Research Methods

Utilizing publicly available resources such as reliable news publications, existing public databases, government documents, and other community-led efforts, we are able to verify the authenticity of each MMIWG2 case. There are over 1,000 cases of MMIWG2s in the United States between 1980 and 2021 in the current data set. Roughly 30% have been identified as confirmed cases so far.
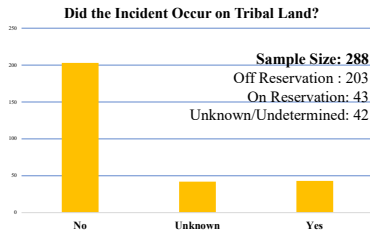
Victim Demographics

MMIWG2 by Age

- 0.5 - 9
- 10 - 17
- 18 - 24
- 25 - 34
- 35 - 44
- 45 - 54
- 55+
- unknown

Age
Sample Size: **253**
18 and Older: 188
Under 18: 65

Gender
Sample Size: **280**
Female: 183
Male: 96
Non-Binary: 1

Offender Demographics

Out of **56** cases where there is at least one known offender

Age
18 and Older: 53
Under 18: 3

Gender
Male: 52
Female: 4

Incidents on Reservation

Did the Incident Occur on Tribal Land?

Sample Size: **288**
Off Reservation : 203
On Reservation: 43
Unknown/Undetermined: 42

Case Resolution

Of the total confirmed cases, **244** have known outcomes. **56.1%** of these are resolved, meaning an individual or their remains have been located. The remaining **43.9%** of cases are still missing a body.

Case Status of Resolved Cases

Sample Size: **137**
Murdered: 101
Missing: 12
Unidentified Body: 24
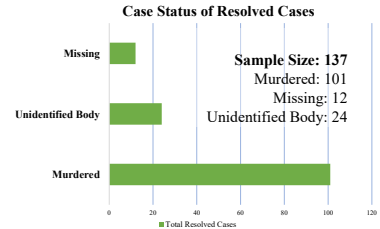
The data reveal a disturbing trend; the majority of MMIWG2 cases remain unsolved. Of the total confirmed cases, only **18%** have both a known offender and a recovered body. Low resolution rates may indicate the ineffectiveness of law enforcement or a lack of accessible reporting systems for MMIWG2s.

References

King, S. and Hodwitz, O. (2020). What's the plan? Broadening the MMIWG2 conversation in North America, Comparative American Studies An International Journal, 17:3-4, Retrieved March 15, 2022, from https://www.tandfonline.com/doi/full/10.1080/14775700.2021.1877082

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