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AUJUS Editorial Team



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Associate Editor: Madison Steele is a recent graduate of Auburn University where she earned her Bachelor of Arts degree in Law and Justice with a minor in Political Science. She works as a Communications Assistant for the Office of Undergraduate Research. In her spare time, Madison enjoys reading, attending concerts, watching Auburn sporting events, and traveling. In the fall, Madison will be attending law school at Indiana University McKinney School of Law and plans to focus her career in practicing Civil Rights Law.



Associate Editor: Hazl Torres is a senior pursuing a Bachelor of Chemical Engineering. She was an Office Assistant for the Office of Undergraduate Research. Hazl's other on-campus involvement includes serving as a facilitator for the Alabama Power Academic Excellence Program and being a member of the American Institute of Chemical Engineers. Hazl is also an undergraduate researcher with the Adamczyk Research Group at Auburn University. In her spare time, Hazl enjoys hiking, working out, attending sporting events, and karaoke. Upon graduation, she plans to pursue a career as a chemical engineer.



Editor-in-Chief: Dr. Lorraine W. Wolf is the Auburn University Director of Undergraduate Research and the Lawrence C. Wit Professor in the College of Sciences and Mathematics. She has served as the editor-in-chief of AUJUS since 2013. She has been a faculty member in the Department of Geosciences since 1993, where she teaches courses and mentors undergraduate and graduate students in the field of geophysics. Dr. Wolf's main research interests are in earthquake and geologic hazards.

MENTOR OF THE YEAR: Dr. Angela Calderón



Dr. Angela Calderón first began her journey with research as an undergraduate at the University of Panama, where she discovered a faculty member's interest in studying medicinal plants and how they are used in the traditional medicine of Panama's indigenous people. For her research, Calderón focused on the effects of medicinal plant extracts on fevers and pain. Her research

mentor was a professor in the field of Pharmacognosy, or the study of the anatomy and chemistry of compounds that come from natural sources, such as fungus, bacteria, or plants.

After completing her pharmacy degree, Dr. Calderón worked as a teaching assistant for the School of Pharmacy in Panama for two-years, until she moved to the University of Illinois to complete a master's degree in Pharmacognosy. After completing her master's degree, she relocated back to Panama to work in the University of Panama's School of Pharmacy. Later, Dr. Calderón received a scholarship to pursue a doctoral degree at the University of Lausanne in Switzerland. After completion of her Ph.D., she moved back to Panama to work as a research assistant professor at the University of Panama. Four years later, Dr. Calderón returned to the University of Illinois as a postdoctoral scholar. Concerned about a lack of opportunity for women scientists in Panama, Dr. Calderón accepted an offer of a faculty position at Auburn University in 2008. Here at Auburn, Dr. Calderón focuses on two areas of research: (1) natural products or natural ways to treat infectious diseases, and (2) quality and safety of botanical dietary supplements.

Dr. Calderón's research experience in three different countries showed her the importance of being im-

mersed in other cultures, meeting new people, and learning new languages. It also stressed the influence she could have on students as a research mentor and role model. Dr. Calderón notes the importance of exposing students to research early, because it helps students continue to dream about their future goals. When asked about the most rewarding part of mentorship, Dr. Calderón states,

“Seeing how much my students have grown and developed in their research, seeing the outcomes of their experience, such as publications, brings me a rewarded feeling.”

She explained that seeing students succeed is testimony to the impact she has had on their education and future careers. Dr. Calderón notes that one of her goals is to contribute positively to the experience that females and other minority groups have in the scientific field. “As a female scientist, I know the challenges. I want to contribute by training female students and other minority groups so that they can be advocates for females in science and technology.”

The achievements that have been most meaningful to Dr. Calderón are being awarded with the Provost's Award for Faculty Excellence in Undergraduate Research Mentoring, being recognized for her expertise as an associate editor in her field, and serving as a botanical dietary supplement and herbal medicine expert on U.S.-based committee.



Design and Construction of a Phase Change Flight Experiment for the International Space Station Using Micro-Structured Surfaces

David Teitge, Karthekeyan Sridhar, and Sushil Bhavnani

Abstract

In microgravity environments, the buoyant force that removes a vapor bubble from a surface is greatly reduced. This means that any hot surface where boiling occurs becomes covered by an insulating layer of vapor due to surface tension. To prevent overheating such devices, this project seeks to show that a micro-structured surface is capable of passively transporting the vapor bubbles away from the boiling location. Preliminary parabolic flight experiments performed aboard the NASA Zero-G aircraft by this research team have demonstrated the suitability of the method in a microgravity environment, albeit one limited by g-jitters. To prove this method under a more pristine microgravity environment, an experiment will be performed on the International Space Station. Before this occurs, a ground experiment will be constructed to refine the flight experiment and obtain preliminary data readings. Boiling will be initiated on metal surfaces that are additively manufactured to create a microscopic saw-toothed surface pattern with even smaller nucleation sites on the sloped faces. The surface features will transport nucleated bubbles to a nearby Peltier cooler which will condense the vapor. Demonstrating this method of transport allows for a more powerful, passive cooling technique to be employed in space where gravity is weak and power is limited. Ground based tests have shown promising results where the wall superheat is held relatively constant by the test surfaces. Funding from the National Science Foundation and the NASA Center for the Advancement of Science in Space demonstrates the interest from the scientific community in this capability.

Introduction

When dealing with the high heat fluxes of modern electronics, phase change cooling is one of the more promising solutions to preventing overheating. The performance of phase change systems is dependent on removing the energy-laden vapor bubbles from the hot

surface. Terrestrial applications often use pumps to remove the vapor bubbles, but pumps may not be ideal for low-gravity environments, such as the International Space Station (ISS) or satellites. Due to electrical power restrictions onboard, the increased power draw from a pump may mean other electronic devices must be omitted from the experiment or the experiment must wait until adequate power is available. A passive method of moving the vapor is preferable. Some methods for creating pump-less flow include changing surface wetting characteristics¹ and Marangoni convection.^{2,3} However, the advantage of an asymmetric ratchet surface modification is the potential of vapor mobility while enhancing heat transfer characteristics of the surface. This project seeks to utilize pressure gradients created by surface asymmetry to create this vapor motion as seen in prior experiments.⁴ The surfaces used in these experiments resemble those used by Linke et al. 2006,⁵ who were among the first to capture surface asymmetry-aided motion.

Auburn researchers have been investigating surface asymmetry-aided motion via experiments conducted aboard the NASA Zero-G aircraft, a Boeing 727 flying in parabolic patterns to simulate microgravity. Clear results were captured showing bubbles nucleating and traveling across the micro-structured surface.⁴ The simulated microgravity is not perfect, however, since there are residual g-jitters (random vehicle accelerations) that can affect the vapor motion. Further experiments will be conducted on the ISS, where better isolation from g-jitters can be obtained. The current project seeks to expand upon the previous ground-based experiments and create an experimental setup that mimics the available hardware on the ISS with confined fluid inventory in a borosilicate tube, rather than a large pool of liquid like in the parabolic flight experiments.

Description of Experiment Preparation

The ground-based experiment has gone through several design overhauls in the Auburn-based lab so that surface asymmetry-aided motion can be more accurately isolated and analyzed. The latest experiment was designed to mimic the ampoule (glass tube containing test surface and fluid) that will go to space and be tested in the Pore Formation and Mobility Investigation (PFMI), an oven on the ISS shown in Figure 1. The figure shows some of the main features, including the Sample Ampoule Assembly, where the flight experiment ampoules will be inserted. Also shown are the high-speed cameras located near the base of the assembly, which will be updated for the flight experiment. The PFMI will be taken from storage and placed inside the Microgravity Science Glovebox in the United States lab module of the ISS. The flight experiments will be run on-board before being returned to Earth.

A glass ampoule formed from borosilicate is used as the containment envelope. The glass blowing shop on campus at Auburn is adept at forming unique geometries out of borosilicate. A model of the setup is shown in Figure 2. The port on the far left is manufactured for high vacuum/pressure applications and is used for adding fluid to the glass ampoule. The port on the far right is used for adding the metal surface to the ampoule and as a port for the temperature sensors. A high-vacuum grease is used in conjunction with this port to ensure that the fluid does not leak around the sensor wires. The three ports in the center are used for positioning the metal surface in the middle of the ampoule and pressing it firmly onto the glass. The boiling surface is positioned with the saw teeth sections directly above the heaters. The test surfaces are discussed in detail in a later section.

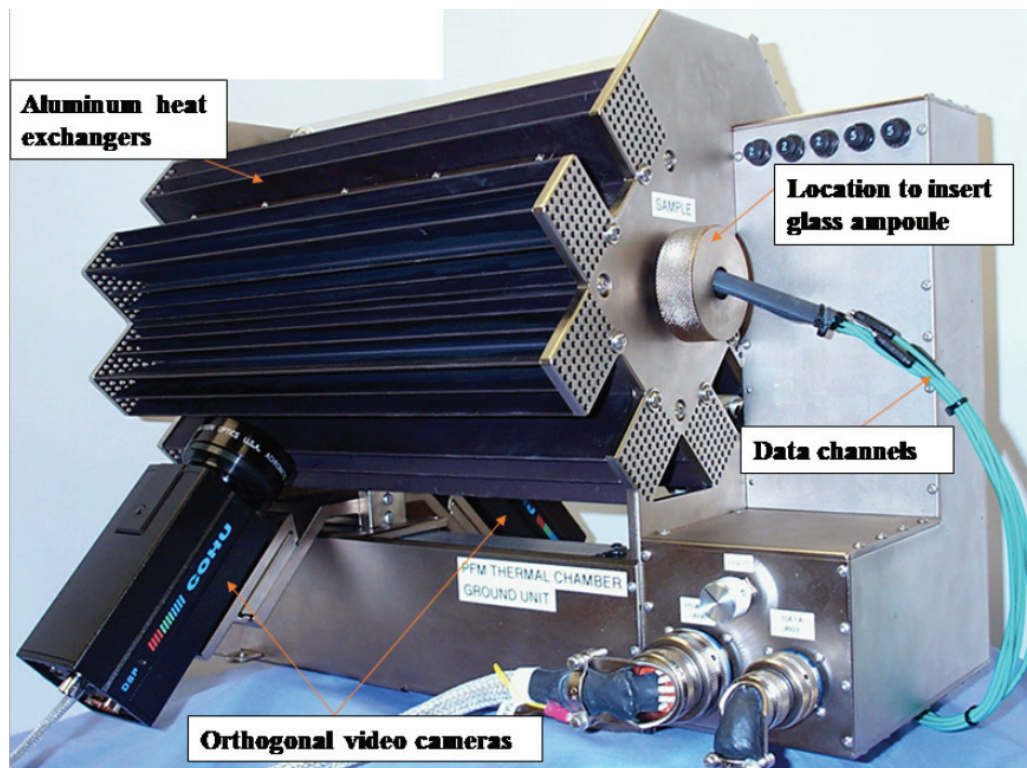


Figure 1. Pore Formation and Mobility Investigation (PFMI) unit on the ISS (adapted from reference 8).

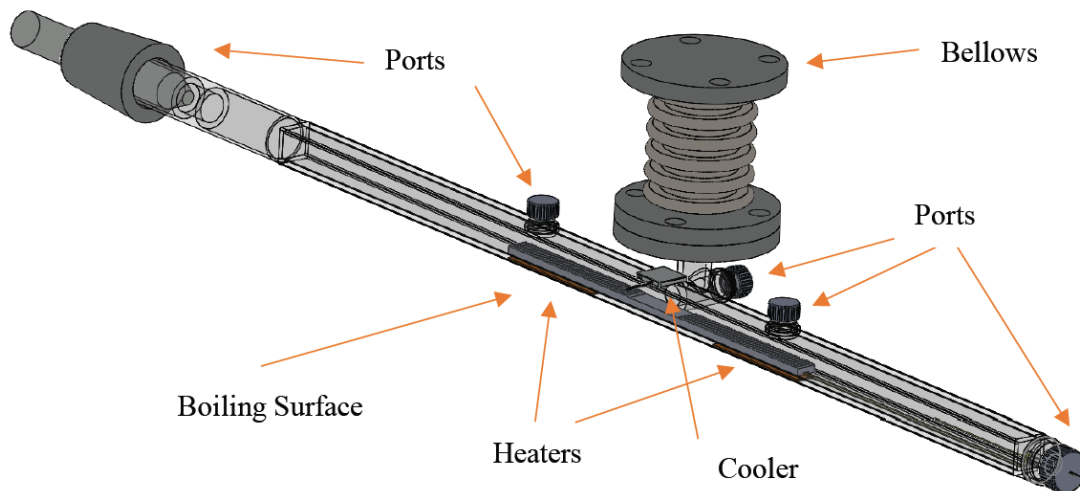


Figure 2. Isometric view of experiment.

The fluid used in these experiments is FC-72, a clear fluid commonly used in electronics cooling. Its popularity in electronics cooling is due to its boiling temperature, 56°C at atmospheric pressure. FC-72 is a highly wetting fluid, meaning that it has incredibly low surface tension and consequently has the tendency to leak from the smallest of openings. Special care must be taken to ensure that the fluid does not leak from the set-up. Neoprene seals and careful application of high-vacuum grease in key areas prevent leaks. An expansion mechanism is used in the form of stainless-steel bellows to allow for changes in fluid volume due to the boiling process. Manufacturers say that steel bellows are not designed for these applications, but the bellows have been used successfully in other experiments.^{6,7} Through testing, the researchers found that the bellows adequately accounted for volume changes in the system.

The heaters used for boiling are simple film resistance heaters composed of thin plastic sleeves with high-resistance wire following a serpentine pattern. Rope heaters are wrapped around the bellows as well. The rope heaters are not to cause boiling but to help keep all the fluid in the system near the same temperature by altering the provided heat flux and avoid excess subcooling. A single Peltier cooler is utilized for the ground experiment as it can produce enough cooling for the process. The use of more Peltier coolers was discussed, especially for the experiment to be run on the ISS, but was forgone from the ground experiment for simplicity's sake. A pressure sensor is screwed into a hole (not pictured in Figure 2) cut into the top

of the bellows to monitor the internal pressure. The temperature sensors used are K-type thermocouples. These are connected to a National Instruments™ data acquisition (DAQ) device and are operated with a LabVIEW™ code.

LabVIEW™ 2016 is used to collect temperature data for the process and export to an Excel™ file. A picture of the LabVIEW™ wiring diagram is shown in Figure 3. The sampling rate and number of thermocouples are set with the elements around the “DAQ Assistant” block. The temperatures from each thermocouple are averaged over a set time and passed to the “Set Dynamic Data Attributes” blocks, where the data are corrected to a National Institute of Standards and Technology (NIST) traceable calibration standard. The information is then exported to an Excel™ file and saved.

Fabrication of Experimental Surface

It is generally understood that bubbles are generated in the cavities of a heated surface, however small these cavities might be. If one wishes to study bubble nucleation in detail, preferential locations for nucleation must be created and all other nucleation sites must be eradicated (or at least greatly reduced). For these reasons, the metallic surfaces used in these experiments are sanded and electro-polished (discussed later). These surfaces, shown in Figure 4 and Figure 5, are additively manufactured (a.k.a. 3D-printed) out of metal. To minimize lead time, the ground experiment surfaces are printed in stainless steel at the National Center for Additive Manufacturing Excellence at Auburn. For

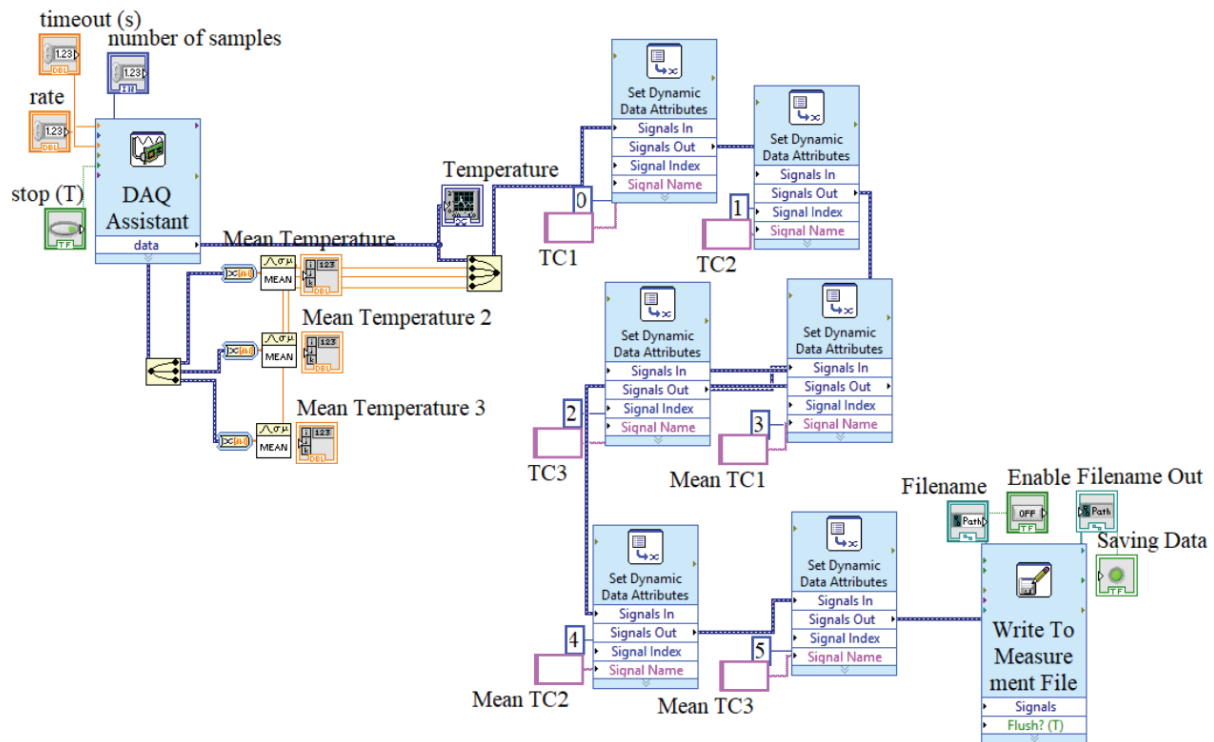


Figure 3. LabVIEW™ code.

the ISS experiment, the test surfaces are printed out of a titanium alloy, which has a coefficient of thermal expansion much closer to that of the glass being used.

The numerous engineered nucleation sites cause vapor bubbles to be formed in concert across the surface. The combined motion of these bubbles creates bulk fluid motion across the surface that aids in heat transfer.

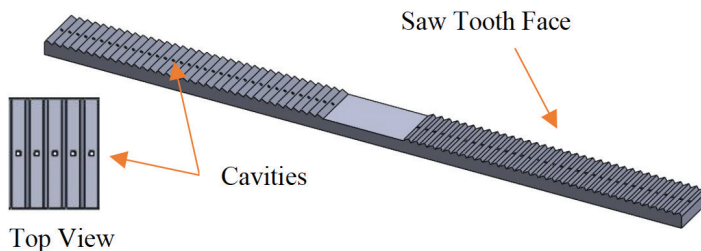


Figure 4. Test surfaces.

There are seven different surfaces printed. A control surface is printed without the saw teeth to see bubble movement in the absence of net pressure gradients. Surfaces are also be printed with cavities on every tooth face or on every other tooth face. The angles for the saw teeth can be either 30°-60° or 15°-75° (where the 15°-75° teeth are shorter since the tooth pitch is always 1 mm). The surfaces also can be either 120 mm long or 180 mm long. The difference in length between these surfaces aids in determining if any edge effects are present. The surfaces with teeth are summarized in Table 1 and can be visualized with the help of Figure 6.



Figure 5. Titanium test surfaces after 3D-printing.

Once the surfaces have been printed and cut off the base plate, the remaining support structure is milled off to ensure the base is perfectly flat. Because the test surfaces are made by selectively melting metal powder, the surfaces are often slightly coarse. To reduce the roughness of the sides, low-grit sandpaper is used to knock down the large bumps. To help smooth out

all faces of the metal pieces, a process called electro-polishing is utilized. The process is similar to electro-plating, but the cathode and anode are switched so that instead of adding a thin layer of metal, a thin layer is removed. A mixture of 75% phosphoric acid (by volume) is used with 25% sulfuric acid for added potency. This process targets bumps, edges, and other areas with low radii of curvature to lower surface roughness and help open the through-cavities along the center of the surfaces.

With the surfaces prepared, a groove must be cut along the center of the base to allow for the thermocouple wires to measure the surface temperature without obstructing the fluid and vapor above the surface. This groove is cut with a cutting wheel while the test surface is locked into a vise.

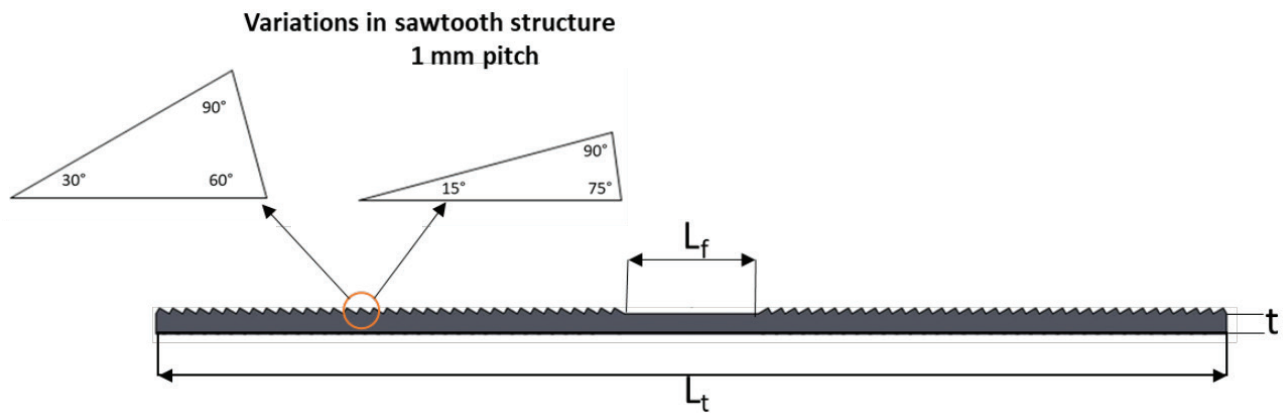


Figure 6. Test surface variations described in Table 1.

Table 1. Test surface variables.

Sawtooth Structure	Cavity Density	Base Thickness (t)	Length of Flat Portion (L_f)	Total Length of Test Surface(L_t)
60°, 30°, 90°	All	2 mm	20 mm	120 mm
60°, 30°, 90°	Alternate	2 mm	20 mm	120 mm
75°, 15°, 90°	All	2 mm	20 mm	120 mm
75°, 15°, 90°	Alternate	2 mm	20 mm	120 mm
60°, 30°, 90°	All	2 mm	20 mm	180 mm
60°, 30°, 90°	Alternate	2 mm	20 mm	180 mm

Experimental Procedure

The thermocouple wires must first be affixed to the inside of the groove with Artic Silver™ adhesive, a type of thermal paste. Thermal paste is also applied to the base of the test surface. The test surface is then slid into place and positioned with the help of the ports in the middle of the ampoule. Care must be taken to ensure the surface is pressed firmly and flatly against the base of the ampoule. With the test surface in place, the thermocouple wires can be brought out through the loading port before sealing the port with high-vacuum grease. The heaters can then be taped to the underside of the glass ampoule directly underneath the saw tooth faces. With the heaters in position, a vacuum is pulled, the fluid is added, and degassing can begin. The degassing process involves heating the fluid so that any dissolved gases come out of solution and can be extracted from the system. A Graham condenser is used to allow the gas to escape while condensing the vapor so that it drips back into the system. The Peltier cooler is added, and the ampoule is leveled. The high-speed camera must be focused on the surface near the nucleation site and enough lighting is provided with an LED lamp. Power is provided to the heaters and cooler and, when boiling occurs, the data collection software is used to record temperature data while video is recorded. The data are reduced to determine the lateral velocity of the bubbles as they leave the cavities (done by using the saw pitch as a scale since each is one millimeter apart). Assuming that the density of vapor is uniform throughout the bubble, the center of each departing bubble can be estimated. As the vapor bubble departs from the cavity, the center can be estimated at a location ~2-3 mm from the surface to obtain lateral velocity. The process is repeated for each of the test surfaces.

Results and Discussion

Data from a preliminary test of the ground experiment are shown in Figure 7. The heat flux in W/cm^2 that is added by the film heaters is presented as a function of superheat, the difference between the surface temperature and the fluid boiling temperature. Although these data are from a preliminary test, they do exhibit signs of the boiling phenomenon expected. At around $1.25 \text{ W}/\text{cm}^2$, the superheat stays constant due to the transition from natural convection to nucleate boiling. As the heat flux increases in the nucleate boiling regime, the rise in superheat is negligible when compared to the

natural convection regime. It can also be thought of as the slope in the natural convection regime being much less than the slope in the nucleate boiling regime. This difference is because nucleate boiling is a more efficient mode of heat transfer than natural convection. The results are in line with conventional pool boiling knowledge, where nucleate boiling can dissipate more heat while maintaining a low rise in surface temperature. The engineered cavities located on the shallow slopes act as nucleation sites for vapor bubbles to grow and depart from the surface.

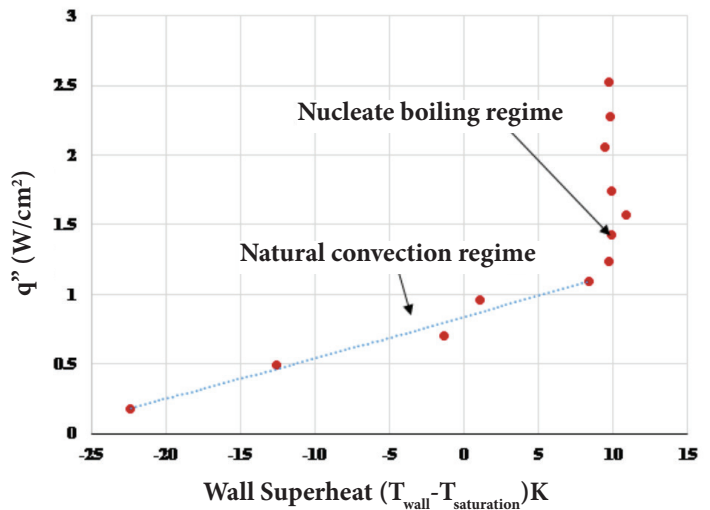


Figure 7. Heat flux provided vs wall superheat (defined as the difference in wall temperature and saturation temperature of the liquid).

The images in Figure 8 show stills from the high-speed video capture for a 15° - 75° sawtooth surface with cavities on every other face. As expected, the bubble breaks away from the surface because of buoyancy, but does so perpendicular to the surface profile, thereby providing a lateral component of velocity.

Conclusions and Future Work

The data collected so far exhibit the expected wall superheat trends and the jump from natural convection to nucleate boiling. Preferential nucleation and motion off the surface can be observed in the high-speed video. The lateral component imparted to the departing bubbles is indicative of a successful proof-of-concept test.

The ground-based experiment is currently ongoing. The ampoule for the ISS is being designed now with the help of the implementation partner, Techshot Inc. The projected launch window for the ISS experiment is early 2020, when the payload will be delivered by the SpaceX Dragon spacecraft. Due to increased interest from the ISS Labs, the original plans for the experiment will be expanded upon to include additional geometries and equipment.

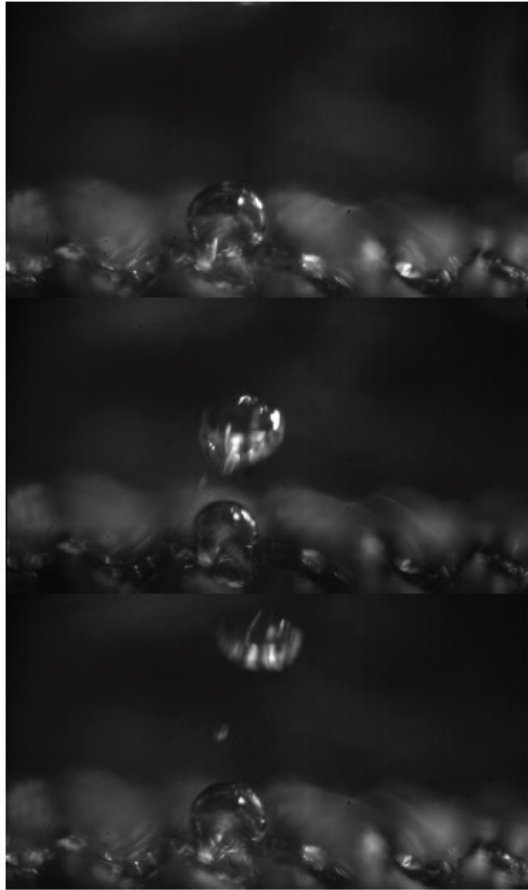


Figure 8. High speed video stills where lateral bubble motion is visible (shot with Phantom v310 outfitted with an Infinity microscope lense (1280x800, 1000 fps)).

Acknowledgments

Support provided by the Alabama Space Grant Consortium is gratefully acknowledged.

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Development of Algal-Bacterial Wastewater Treatment Systems That Are Effective in the Presence of Antimicrobial Processing Aids Used in the Poultry Processing Industry

Justin Box and Brendan Higgins

Poultry processing plants utilize a range of antimicrobial processing aids to sanitize plant equipment and bird carcasses to meet stringent food safety standards. Recently, the U.S. Poultry and Egg Association recognized that these processing aids could present problems for their existing wastewater treatment approaches. While effective at killing pathogens, antimicrobials have the unfortunate side effect of harming the “good” bacteria, particularly nitrifiers, that are critical to wastewater treatment. Past research on anaerobic digestate has shown that adding algae to wastewater treatment processes can help bacteria more effectively carry out nitrification, even in a toxic environment. It is unknown if algae can help nitrifying bacteria overcome the adverse effects of antimicrobial aids in the treatment of poultry processing wastewater. The research objective of this study was to compare the effectiveness of an algal-bacterial process to a bacteria-only wastewater treatment process in the presence of an antimicrobial aid commonly used in the poultry industry: peracetic acid (PAA).

The first step in this project was to determine the concentration of PAA at which the growth of *Chlorella sorokiniana* algae is inhibited. The result from this dose-response study showed that *C. sorokiniana* has a similar sensitivity to PAA as nitrifying bacteria, with an EC50 of about 7.5 mg/L PAA.

To understand the effect of PAA on an algal-bacterial wastewater treatment process, we performed a batch microcosm study by adding algae to a bacterial consortium treating poultry processing wastewater in the presence and absence of 7 mg/L of PAA. PAA was spiked into the culture daily over 120 hours. Subsequently, ion chromatography was used to measure the change in water quality by analyzing ammonium, nitrite, and nitrate concentrations. Additionally, quantitative polymerase chain reaction (qPCR) was

performed on the final biomass samples to evaluate the effect of PAA by quantifying the genes for ammonia (AmoA) and nitrite (nxrb1-F) oxidizing bacteria. Figure 1 includes the results for ion chromatography and qPCR.

The ion chromatography results show that 7 mg/L of PAA suppresses nitrification; however, the results show the suppression of nitrification is reduced when algae are present in the system, therefore allowing for nitrification to occur. This is seen by the conversion of ammonium to nitrite/nitrate by the bacteria present in the algal-bacteria system. The ammonium levels dropped significantly in the algae cultures due to the improved algal uptake and bacterial utilization of ammonium. The qPCR analysis indicated that PAA suppressed the ammonia and nitrite oxidizers; however, algae clearly supported these oxidizers in both the presence and absence of PAA. These results indicate the possibility of using an algal-bacterial process to effectively treat poultry processing wastewater, even in the presence of PAA.

Statement of Research Advisor

The results from this project constituted a significant advance in our understanding of how algae can increase nitrification capacity of bacteria when treating poultry processing wastewater. To our knowledge, this is the first report showing that algae can support ammonia-oxidizing and nitrite-oxidizing bacteria in the presence of PAA.

– Brendan Higgins, Biosystems Engineering

Events Boundary Effects on Speech in Physical and Virtual Reality Environments

Brianna P. Burks, Dallin J. Bailey, Cheryl Seals, Majdi Lusta, Tianshi Xie, Nikolay Sargsyan, Jueting Liu, and Soundarya Korlapati

Speech-language pathologists (SLPs) provide rehabilitative services for persons with communication disorders such as aphasia and apraxia of speech. Both of these disorders disrupt an individual's ability to communicate. While SLPs often see strides of progress during session work, the progress may not carry over to other environments, such as home. The carryover from therapy to real life is referred to as generalizations and is often difficult for people with aphasia. One possible reason for this may be that patients are changing learning contexts, which has an impact on memory and speech, especially for older adults. Event cognition states that the mind automatically separates events, often by physical boundaries. We wanted to see if event cognition effects on memory and speech production, which have been found in physical reality, could also be found in a virtual reality environment. To test this, we created a virtual replica of our physical laboratory to allow experiment building and testing of memory and speech production in both the physical and virtual environments. In addition, a table and moveable picture cards matching the physical picture cards used in the laboratory setting allowed for basic experiments concerning memory and speech to be carried out in both the physical and virtual realities.

We are currently collecting data related to event cognition and the event boundary effect using the virtual environment. Experimental studies include replicating the speech production findings of Meagher and Fowler (2014). Meagher and Fowler found that the initial length of time to produce the word was longer and became shorter as the participant remained in that environment. However, when they switched to a new environment or room, the time to produce the words increased and had a duration that resembled the initial production in the first environment. Results of this study may have implications for better understanding event cognition effects on speech production, which, in

turn, may impact generalization in speech therapy for individuals with aphasia or apraxia of speech.

Statement of Research Advisor

Brianna led the development of a project combining speech-language pathology, psychology, and computer science. The virtual reality platform developed has potential to be adapted for many further research investigations.

-Dallin J. Bailey, *Speech Language, and Hearing Sciences*

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Frequency Characterization of Thigh Soft Tissue Artifact During a Relaxed and Activated State

Scot Carpenter, Michael Zabala, Jacob Larson, and Anthony Marino

Marker-based motion capture systems are commonly used for biomechanical analyses to measure human movement by tracking the positions of reflective markers placed on the surface of the skin. A 3D model of the human subject can be created using the position data from the markers. This provides an approximate location of the position and orientation of the underlying bones during a particular movement. A well-known limitation of this method of data collection, however, is the relative movement of skin and other soft tissue over the bone known as soft tissue artifact (STA). In order to accommodate for this discrepancy, a low-pass filter at 6 Hz has been shown to be somewhat effective at removing STA and has been used as the typical low-pass filter. However, the frequency at which data should be filtered has been shown to depend on the movement type as well as various physical characteristics of human subjects.

For this study, the objective was to analyze how the STA frequencies of the thigh differ based on an activation

state of the thigh musculature. Four subjects participated in the study, and reflective markers were placed on each subject using a 79 marker set based on the point cluster technique. The test consisted of subjects standing with their right foot on a platform, which was raised to a height such that the knee was at 90 degrees of flexion. The subjects were instructed to relax the thigh and perturb the skin by twisting the thigh with one hand about the femoral longitudinal axis to its maximum possible angle and quickly releasing, allowing the soft tissue to oscillate freely. The subjects were then asked to repeat the procedure, while activating the musculature of the thigh. Three trials were taken for both conditions on the right leg. The marker trajectories were processed in MATLAB[®] by creating an anatomical reference frame on the thigh and tracking movement of the centroid of the 9-marker thigh cluster relative to the origin of the segment. The angle of twist about the femur was calculated for each trial and converted to the frequency domain with a Fast Fourier Transform (FFT) to analyze the frequency components of the signal.

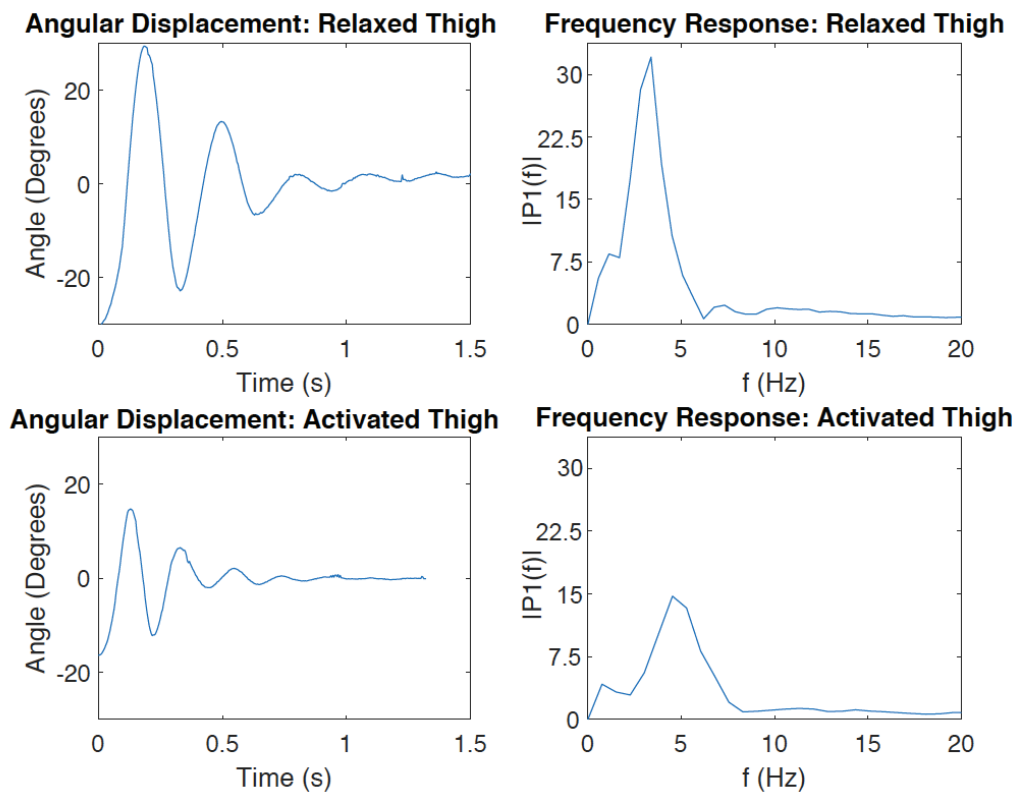


Figure 1. Time history and frequency response plot for a single subject with $|P1(f)|$ = peak amplitude (deg).

The results indicated that the angular frequency of the thigh increased with muscle activation, where a relaxed and activated thigh had an average frequency of 4.04 and 5.60 Hz, respectively. More importantly, the average frequency is less than 6 Hz regardless of activation. This means that some STA might not be removed from motion capture data when the typical 6 Hz cut-off frequency is used. The results of this study have indicated the possibility of allowing STA into motion capture results while using a 6 Hz lowpass filter. Future work in determining the relationship between STA and physical traits of the subject, such as age, weight, sex, and percent body fat, is needed to further determine STA frequencies for various subject characteristics.

Statement of Research Advisor

Scot has done tremendous work that has determined thigh muscle oscillation frequency in a contracted and relaxed state for this group of subjects. These data are new to the field and have practical utility in improving biomechanical analyses dependent on surface-based motion capture markers.

– *Michael Zabala, Mechanical Engineering*

Investigating the Role of the Lateral Entorhinal Cortex in Alzheimer's Disease

Marissa Dogan, Sharay E. Setti, Jeremiah Pfitzer, and Miranda N. Reed

The purpose of this study was to discover potential mechanisms that mediate the spread of Alzheimer's Disease (AD), a fatal neurodegenerative disease that affects 5 million U.S. citizens every year (Alzheimer's Disease facts and figures, Alzheimer's Association, 2019). AD disrupts and destroys normal neuronal function via the formation of beta-amyloid plaques (between neurons) and neurofibrillary tangles made of hyperphosphorylated tau protein (within neurons). One of the first brain regions to exhibit this pathology is the lateral entorhinal cortex (LEC); this study is aimed at understanding the role of this brain region in the progression of the disease.

We began by delivering mutant tau protein to the LEC in previously healthy mice via intracranial injection. After allowing sufficient expression time of four weeks, we assessed for learning and memory impairments in these mice using two memory tasks: object recognition and trace fear conditioning. At the conclusion of behavioral testing, we then assessed for the presence of tau pathology using immunohistochemical staining of brain slices. We found the presence of early tau pathology mice injected with mutant tau (AAV-TauP301L; data not shown). In addition, these mice had cognitive deficits (Figure 1), indicating that our experiment sufficiently modeled early stage disease alterations.

In AD, tau pathology beginning in the LEC eventually spreads to the hippocampus along a circuit called the perforant pathway. Evidence from the literature suggests that misfolded tau may be released into the space between neurons, or the synapse, due to increases in neuronal activity (Pooler et al., 2013). Once there, the misfolded tau can be taken up by nearby neurons, eventually compromising the healthy tau in these neurons. To determine if this was the case, we again delivered mutant tau protein to the LEC. In addition, we delivered a viral vector containing light-sensitive opsin receptor to neurons in the LEC. Neurons expressing this light-sensitive opsin receptor can then be activated by exposure to a certain wavelength of light. We implanted

a fiber optic cannula through which we exposed these neurons to light, resulting in increased neuronal activity. For our comparison, we increased neuronal activity (stimulated) in one hemisphere of the brain and not in the other hemisphere (non-stimulated). We determined that tau pathology spread further in the stimulated hemisphere, versus the non-stimulated hemisphere (Figure 2). These results indicate that increased neuronal activity resulted in increased spread of tau along connected neural networks. Currently, we are continuing work to determine if this increased spread of tau pathology also results in worsened pathology and memory deficits.

The LEC has been identified as the site where some of the first alterations appear in AD, but the role of the LEC in memory is poorly understood. Understanding the role of the LEC in memory would allow for creation of diagnostic tasks sensitive to these early alterations.

Statement of Research Advisor

Marissa's work strongly indicates that tau pathology in the LEC can induce detectable memory deficits and that increasing neuronal activity in the LEC can cause tau pathology to spread to connected neural networks. Future work will identify whether decreasing neuronal activity can prevent the spread of tau.

– *Miranda N. Reed, Drug Discovery and Development*

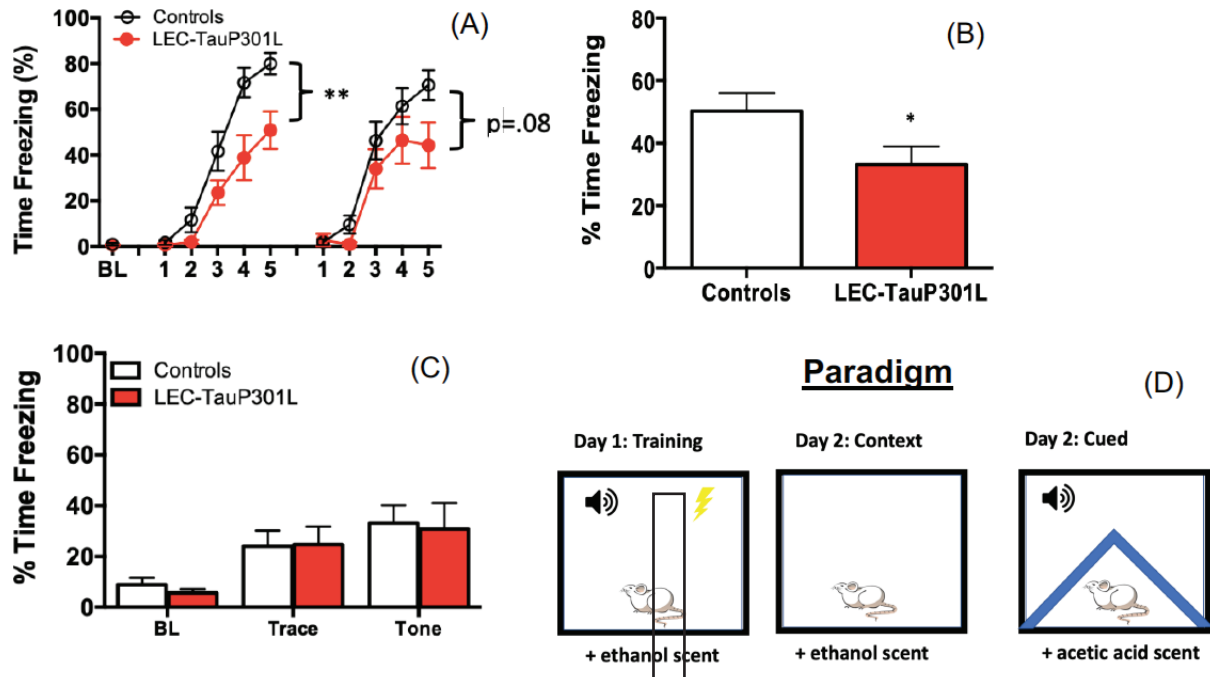


Figure 1. (A) AAV-TauP301L mice do not differ from controls in baseline freezing. AAV-TauP301L mice freeze significantly less than controls during trace and tone acquisition. (B) AAV-TauP301L mice freeze significantly less than controls during contextual testing. (C) AAV-TauP301L mice do not differ from controls in amygdala-based tone retention testing. (D) Representative paradigm. Data represented as means \pm SEM.

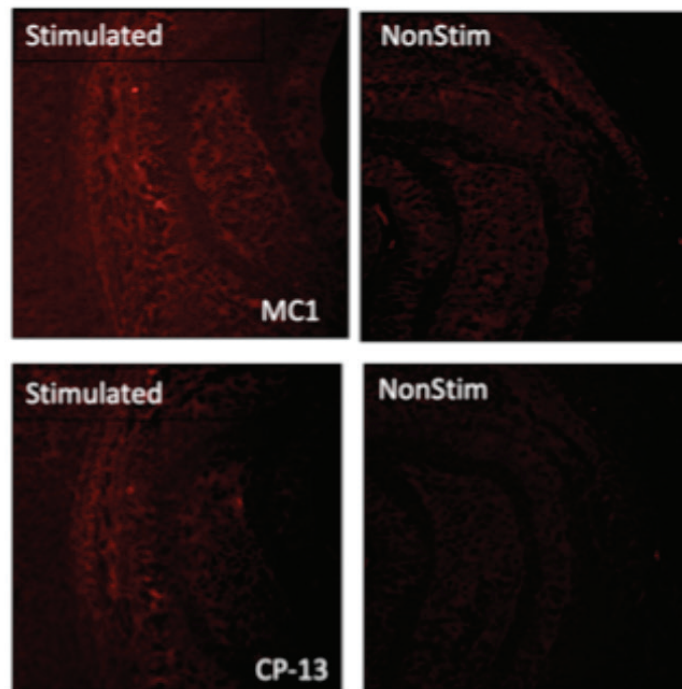


Figure 2. Presence of MC-1 and CP-13 in the hippocampus of stimulated brain regions but not non-stimulated brain regions.

Effects of Physical Activity and Sport Participation on Mental and Physical Health in Rural, Low-Income High School Students

Sarah Beth Dolinger, Robyn Feiss, and Melissa Pangelinan

Research shows there are many physical, cognitive, and social factors that influence health and well-being of an individual, particularly during adolescence. To this end, sport participation has been shown to have a positive influence on physical health during adolescence¹ and allows adolescents to increase physical activity levels.² Furthermore, studies show increased physical activity levels and sport participation are associated with decreased mental health burdens.^{3,4} The purpose of my research was to study the relationship between sport participation and physical activity levels and mental health in rural, low-income, high school students. This sample group was chosen specifically because adolescents from low-income backgrounds have elevated risk for mental and physical health disparities.⁵ Little research, however, has been conducted in this population.

Wellness fairs were hosted at four Title I high schools. A total of 253 10th and 11th grade students participated (114 males, 139 females). This study was approved by the IRB (Protocol 18-109 MR 1803); parental consent was obtained prior to each fair. Participants completed the PROMIS[®] Pediatric Anxiety,⁶ Pediatric Depressive Symptoms,⁶ Psychological Stress Experiences,^{7, 8} Physical Stress Experiences,⁷ and Physical Activity⁹ questionnaires. Additionally, students completed the FITNESSGRAM[®] physical fitness testing, which consisted of anthropometrics (height, weight, body mass index [BMI]), body composition (% fat mass, % lean mass), resting heart rate (RHR), blood pressure, muscular strength and endurance (push-ups, curl-ups), and aerobic capacity (Progressive Aerobic Cardiovascular Endurance Run [PACER]). Body composition was measured using a TANITA[®] total body composition analyzer (SC-331S Total Body Composition Analyzer, TANITA[®]) and blood pressure and heart rate were measured via an Omron[®] automatic blood pressure monitor (5 Series Upper Arm Blood Pressure Monitor BP742N, Omron Healthcare).

Stepwise linear regression was used to examine the influence of sex and race on sport participation and physical activity. Additional stepwise linear regressions were used to examine the influence of sex, race, physical activity, and number of sports on mental and physical health measures.

Contrary to our hypotheses, females reporting greater physical activity levels reported higher anxiety symptoms ($p = .02$). The same was true for one-sport ($p < .01$) and multisport ($p < .01$) females showing greater depressive symptoms. Though Caucasian/white adolescents showed lower depressive ($p < .05$) and psychological stress ($p < .01$) symptoms with greater number of sports, non-white adolescents showed no relationship. These results suggest other factors are contributing to higher mental health burdens in this population. Moreover, 36.4% of students reported moderate or severe anxiety symptoms, 36.0% reported moderate or severe depressive symptoms, and 28.1% reported high or very high psychological stress symptoms. Sport participation was related to improved physical fitness measures (i.e., systolic blood pressure [$p = .04$], RHR [$p < .001$], push-ups [$p < .01$], curl-ups [$p = .02$], and PACER test [$p < .01$]) suggesting sport participation may be a way for rural, low-income adolescents to improve physical health outcomes. Yet according to BMI, 15.8% of participants were overweight and 23.3% were obese. Results suggest the need for further examination of these relationships in the present population.

Statement of Research Advisor

Sarah Beth Dolinger is a current undergraduate research fellow (2018-2020). Over the course of her second year fellowship (2019-2020), Sarah Beth was a major contributor to the management of the Wellness Fair program, including disseminating information to the schools, distributing and collecting consent forms, as well as managing data collection and data archiving.

She contributed to the data analysis for the present study and the interpretation of the study results for publication. Sarah Beth has presented these results at the Student Research Symposium at Auburn University, as well as at two national conferences (National Conference on Undergraduate Research and the annual meeting of the North American Society for Psychology of Sport and Physical Activity).

– *Melissa Pangelinan, Kinesiology*

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Click Chemistry Towards Allyl-Polymer MRI Contrast Agents: Solubility, Kinetics, And Magnetic Responses

Mingrui (Lily) Gong, Bryan Beckingham, and Michael Minkler

The general principle of Magnetic Resonance Imaging (MRI) is that it exerts a strong external magnetic field to force protons in the body to align with the magnetic field. The MRI sensor then captures the energy released by the proton and records the T1 relaxation time, or the time that the proton takes to recover from the “excited” state to the equilibrium state. Water protons in different tissue structures provide different T1 values, which allow MRI to generate the scan or images. However, sometimes more detail or contrast between different tissues is required to evaluate the area of interest. In this situation, a MRI contrast agent is introduced to enhance the contrast between nearby water protons, typically by shortening the T1 relaxation time. Most of the current MRI contrast agents are gadolinium-based. Unfortunately, studies have shown that gadolinium can deposit in the body and potentially lead to severe side-effects, especially for patients with renal problems. To reduce this potential risk, a low-toxicity and biocompatible MRI contrast agent is sought to investigate as a possible replacement for gadolinium-based contrast agents. Here, we investigate poly (allyl glycidyl ether) (PAGE)-based materials due to its biocompatible polyethylene glycol (PEG) backbone and pendant allyl, which is amenable for click chemistry (thiol-ene cou-

pling) as shown in Figure 1.

This pendant allyl group can incorporate magneto-responsive groups that can impact the T1 relaxation time. The experimental approach is to functionalize PAGE with various content of a magneto-responsive groups and examine its impact on the T1 relaxation time. As an initial test of this chemistry, a small molecule was synthesized by the reaction of histamine and γ -thiobutylolactone. This molecule contains a thiol group for reacting the PAGE-pendant allyl groups and a histamine residue for later quaternization. Once isolated, this small molecule was used to functionalize PAGE through click chemistry and quaternize it with iron(III) chloride (FeCl_3) to introduce the magneto-responsive character. ^1H Nuclear Magnetic Resonance (NMR) was used to stimulate the general condition in MRI and to determine its impact on T1 relaxation time. This synthesis process was successfully completed to obtain a 20 mol% functionalized PAGE that was used to investigate the T1 relaxation time in dimethyl sulfoxide (DMSO). The ratio between functionalized PAGE and DMSO was varied from 5 mg/mL to 50.1 mg/mL. Overall, T1 relaxation times decrease as the content of functionalized PAGE increases in the solution.

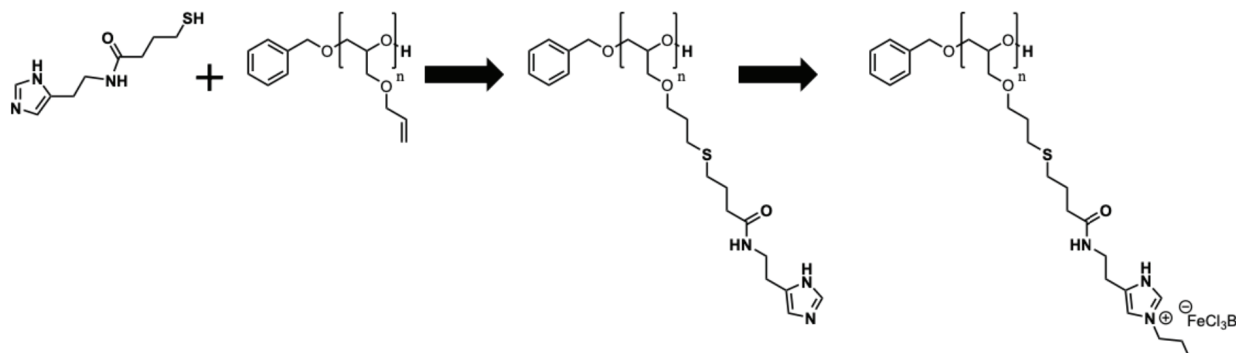


Figure 1. Reaction scheme for the synthesis of magneto-responsive PAGE.

There are still many questions that remain unanswered. For instance, we wish to understand how this T1 relaxation time compares with common Gadolinium-based contrast agents, investigate how functionalized PAGE behaves when placed in water, and possibly test it with actual cells or tissues. This experiment successfully investigated an alternative materials approach to low-toxicity and biocompatible MRI contrast agents.

Statement of Research Advisor

Lily has been very productive in learning new chemistry and techniques for synthesizing new materials for next-generation bio-compatible MRI contrast agents. Her work has laid the foundation for future work in this area and I am greatly looking forward to her future research contributions.

-Bryan Beckingham, Chemical Engineering

Investigating the Influence of Preparatory Arm Swing on Ankle Kinematics and Force Production During the Horizontal Jump

Daisy Hansana and Wendi Weimar

Preparatory movements prepare the body to perform ballistic movements. During the horizontal jump, the preparatory movement includes a vigorous arm swing and a countermovement marked by hip and knee flexion. While several studies investigated the mechanical role of the arm swing in enhancing the performance of the horizontal jump, the reason for this enhancement remains unclear. Previous work has indicated that the arm swing raises the center of mass and increases the force into the floor but has not found if alterations in the preparatory arm swing can alter ankle kinematics and/or the forces exerted on the floor by the body. This project, by adding an extra arm swing to the preparatory movement, seeks to determine if this extra motion prepares the body to experience a greater gain from the stretch shortening cycle during the counter movement of the horizontal jump. The stretch shortening cycle is a term used to describe a phenomenon demonstrated by counter movements, in which the body stretches the muscles about to be used in the performance of a ballistic task. This stretch stores elastic potential energy that is released as kinetic energy during the propulsion phase of a ballistic movement, like the horizontal jump. Our hypothesis is that the extra arm swing will remove some of the “slack” in the muscle prior to the stretch and thereby allow for the storage of more elastic potential energy during the stretch.

To test this hypothesis, we measured changes in ankle motion and the forces exerted into the floor during the horizontal jump, during a normal jump performance, and during a performance in which an extra arm swing is added to the preparatory movement. Our pilot study included four participants between 19 and 30 years of age who voluntarily consented to perform the two-day trial that lasted 1.5 hours. Each participant was given an unlimited time to warm up, after which reflective markers were attached to the participant in order to track the movement of the body segments. Following the warm-up, the participants performed six jumps of maximal effort under two different conditions:

1. Condition 1: Three jump trials that began with an extra arm swing preparatory arm movement (bilateral swing) prior to initiation of the jump.

2. Condition 2: Three jump trials utilizing their “normal” or typical start form.

Foot placement along with stance remained the same throughout all six trials, whereas the order of jump types was randomized via coin flip. Data from each jump were collected using a 10-camera Vicon[®] optical motion capture system at 240Hz, and an AMTI[®] force plate (1000Hz).

Results indicated that the extended arm swing condition yielded the following outcomes:

- (a) more ankle dorsiflexion during the counter movement.
- (b) more ankle plantar flexion at take-off
- (c) greater peak vertical force
- (d) greater peak posterior force.

An increase in each of these variables implies increased performance on the horizontal jump. The increased ankle dorsiflexion during the counter movement indicated that a greater stretch was realized at the knee and the hip. The increased plantar flexion at take-off indicates that the body pushed through a greater range of motion at take-off. The increases in vertical and horizontal forces indicates that the body exerted greater forces into the ground, which should be realized in greater jump performance.

At this time, it is difficult to discern whether the inclusion of an additional preparatory arm swing produced the kinematic changes at the ankle, or whether the changes in ankle motion were a byproduct of the changes brought about at the hips and knees. Regardless, the increase in force production indicates that the additional preparatory arm swing is beneficial to hori-

zontal jump performance.

Statement of Research Advisor

Daisy investigated the interconnectedness of the body by evaluating the influence of a pre-preparatory movement on the kinematics and kinetics of the ankle during the horizontal jump.

-Wendi Weimar, Kinesiology

Impact of the U.S.-China Trade War on the Alabama Soybean Industry

Seth Ingram and Ruiqing Miao

Soybeans are one of the primary crops produced within Alabama's thriving agricultural industry. The soybean sector has a total annual impact of more than \$1 billion on Alabama's economy.¹ The purpose of this study is to evaluate the impact of the U.S.-China trade war on the Alabama soybean industry. Many studies have been completed on the broader impact of the trade war on the U.S. soybean industry as a whole. However, this study is unique in its focus specifically on the implications for the soybean industry in Alabama. It is critical for Alabama soybean producers and industry leaders to understand the impact of this trade dispute on the sector in order to be prepared to make informed management decisions when facing similar situations in the future.

The collection and analysis of quantitative data pertinent to the Alabama soybean industry from the beginning of the trade war through the end of 2020 served as the focal point of this study. Acreage and price data provided by the United States Department of Agriculture (USDA) highlighted the trade war's impact on the planting decisions and profitability of Alabama producers. In addition, soybean export figures provided by the Alabama Department of Commerce presented the sector's struggle to replace the Chinese market for Alabama soybeans at the height of the trade war, as well as the resurgence of exports to China following the Phase One trade agreement.

A detailed analysis of these data confirmed that the U.S.-China trade war has significantly impacted the Alabama soybean industry since its beginning in 2018. The data reveal that the elimination of the Chinese export market for Alabama soybeans from late 2018 until January 2020 led to sharp decline in prices received by producers in the state. During this time, many Alabama producers opted to plant other field crops with more favorable immediate market outlooks, such as corn, in place of soybeans. Additionally, government support in the form of Market Facilitation Program payments played a critical role in mitigating the negative implications of the trade dispute for Alabama soybean producers.

Following the announcement of the Phase One trade agreement in January 2020, China reopened to Alabama soybean exports and reclaimed its position as the largest market in the soybean sector for Alabama. Accordingly, soybean acreage and prices in the state have rebounded. Overall, my study shows the U.S.-China trade war severely stressed the Alabama soybean industry, but the industry has proven to be resilient and looks to be in route to a sound recovery.

Statment of Research Advisor

This analysis contributes to the field by examining the impact of the U.S.-China trade war on the Alabama soybean industry. Existing studies have mainly focused on U.S. soybean industry as a whole or some key producing states of soybeans in the nation. The present study focuses on Alabama and shows how soybean acreage and prices in the state changed during the trade war.

-Ruiqing Miao, Agricultural Economics and Rural Sociology

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Corrosion of Additively Manufactured 316L Stainless Steel Bio-Implants in Simulated Human Body Fluids

Catherine McConatha and Xiaoyuan Lou

Additive manufacturing (AM) technology is becoming increasingly popular for building complex parts in various industries, including medicine. A surgical procedure for cranial reconstruction with a custom designed metal implant has been performed using AM technology¹. Implants such as this are lifelong replacements for body parts, and AM provides an efficient way to create implants and solve other complex medical problems. Medical devices are traditionally made from wrought materials, but because of the increasing popularity and availability of AM technology, medical professionals are transitioning to AM methods. AM is a popular method in bio-implants because it allows for customization of surgical implants. The material commonly used is 316L stainless steel, a bio-compatible metal. However, the corrosivity of human body fluids can cause breakdown of metal and adverse side effects for the patient.

The objectives of this study are to (1) optimize the laser AM process to fabricate high quality AM 316L SS parts, (2) characterize microstructure of AM 316L SS, (3) understand localized corrosion of AM parts versus wrought parts in simulated body fluids, and (4) understand localized corrosion in as-built AM parts versus heat-treated AM parts in simulated body fluids.

Additive samples were created using a Concept Laser Mlab Direct Metal Laser Melting (DMLM) machine using 316L SS powder. Each sample was etched to reveal microstructures and cellular structures, which were analyzed using a scanning electron microscope (SEM). SEM characterizations were performed on untested AM as-built in both vertical and horizontal orientations, heat-treated AM 316L, and wrought 316L in order to identify important microstructures that may be related to corrosion resistance. The samples were tested using a GAMRY Reference 600+™ Potentiostat. Two cyclic potentiodynamic polarization (CPP) tests were performed for each sample. Figure 1 includes one CPP

graph for each material type. The data were analyzed to evaluate the susceptibility of the material to localized corrosion, or pitting. Hank's Balanced Salt Solution (HBSS) with 5.96 g/L of HEPES Buffer Solution with a pH of 7.4 +/-0.2 was used as the electrolyte.

Prior to printing the AM samples, a study was conducted to determine the most efficient printing power, speed, and hatch spacing. A printing power of 90 W, a speed of 600 mm/s, and a hatch spacing of 80 µm were selected. Hatch spacing is the distance between two adjacent scanning paths of the laser used to melt the metal powder. This parameter was found to greatly affect the size, type, and quality of defects present in the specimens.

The microstructural components of each material were drastically different, a result that coincides with the data found from CPP testing. Based on the data, it is clear that both as-built and heat-treated AM 316L SS demonstrated high resistance to localized corrosion in HBSS. In addition, additive 316L SS has superior resistance to localized corrosion when compared to wrought 316L. Lastly, unique microstructures in as-built and heat-treated AM 316L SS parts contribute to their superior corrosion resistance.

Statement of Research Advisor

Catherine contributes to the understanding of the degradation process of additively manufactured stainless steels in the human body environment, which is a critical measure for medical implant devices. She also characterized the defect structures of AM parts as a function of laser processing parameters, which contributed to a collaboration with another university. She did an excellent job on her research.

-Xiaoyuan Lou, Mechanical Engineering

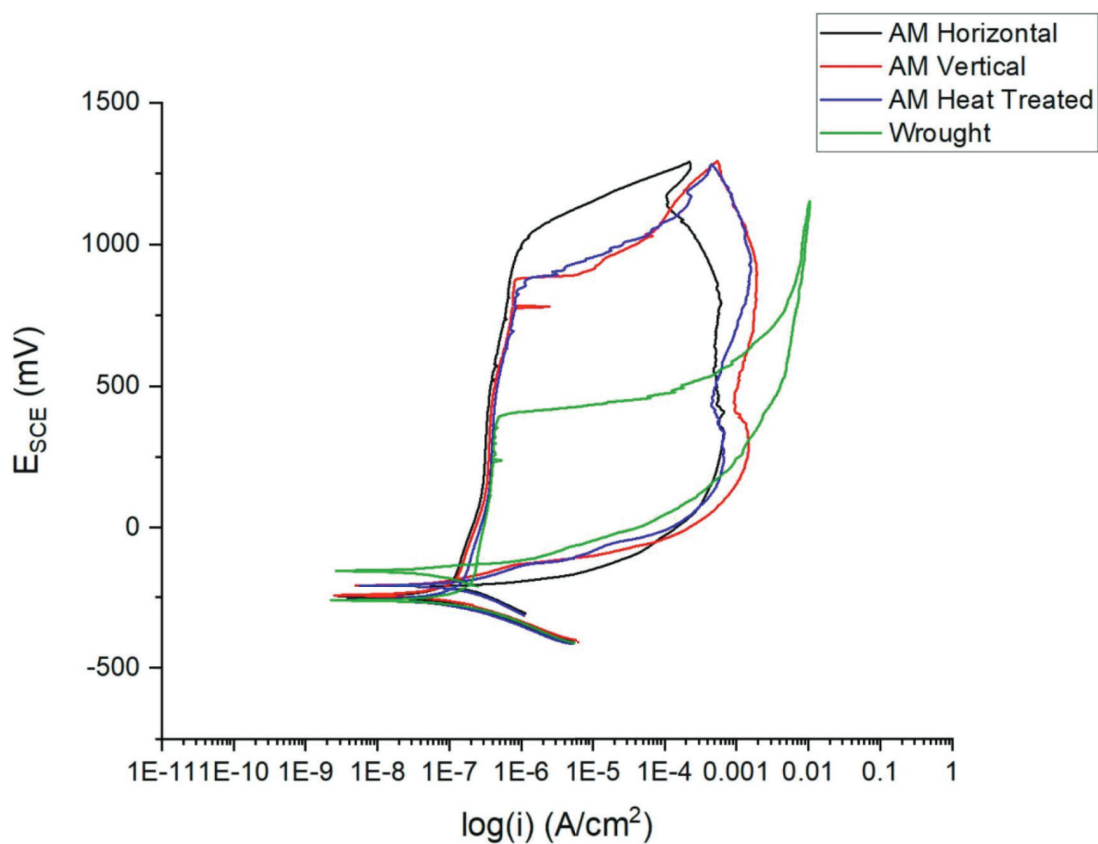


Figure 1. Cyclic potentiodynamic polarization graph for each tested material type.

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Anthropometric Factors in Softball Hitting Performance

Johann Phan, Kenzie Friesen, and Gretchen Oliver

Baseball is considered one of America's pastimes, and as such, is one of the most popular sports today in America among youth. Despite the popularity of the sport, there are limited data available when examining hitting mechanics, especially in conjunction with anthropometric measurements of the hitters. Hitting is a major component to team success in baseball; therefore, research dedicated to understanding performance parameters that improve hitting performance is valuable to the development of the sport. Research has found arm length is an important anthropometric factor in elite players of other sports, particularly those that require extensive use of the upper extremity.¹⁻⁵ Given that the upper extremity is an important link within the kinetic chain in hitting, the objective of this study was to find the effect of arm length and other physical traits on baseball hitting performance.

Nine youth baseball players (11.2 ± 1.3 years, 149.8 ± 9.6 cm, 51.1 ± 14.1 kg) volunteered to participate in the study. The Institutional Review Board of Auburn University approved all testing protocols and informed written consent was obtained from each participant prior to participation. Each participant was then measured for bilateral hip and shoulder isometric strength and range of motion using a dynamometer and inclinometer, respectively. Hip measurements were conducted with the participants sitting on an athletic training table, with their hips and knees both flexed to 90 degrees. Shoulder measurements were completed with the participants lying supine on the table, with their shoulder abducted 90 degrees and their elbow flexed 90 degrees. Participants were then hooked up to an electromagnetic motion capture system synced with motion analysis software, which was used to record weight, height and arm length for all participants. Yakkertek®, a ball-tracking device, was used to measure batted ball distance, launch angle and exit speed for all trials (Figure 1). Each participant warmed up for as long as needed, then completed ten swings from a front tossed pitch. Of the ten swings, the four trials with the closest launch angle to the average launch angle for the ten trials were used for data analysis.



Figure 1. Yakkertek device used to measure batted ball characteristics.

Pearson's correlations ($p < 0.05$) revealed a significant relationship between player mass and batted ball distance (Figure 2), as well as player height and exit speed (Figure 3). Therefore, these findings would suggest overall height and mass have a greater impact on hitting performance than arm length, strength and range of motion measures.

These results suggest that neither arm length nor isometric strength has a significant effect on batted ball performance in youth. These findings could indicate the overall importance of total body kinetic chain sequencing versus absolute brute strength in hitting performance. However, it should be noted that these findings may also be due to the inconsistency of isometric strength and lean muscle mass in youth athletes. Future research should be conducted to examine the relationship between anthropometric factors and hitting performance among a larger age

range of athletes accounting for those with bigger body characteristics and more hitting expertise.

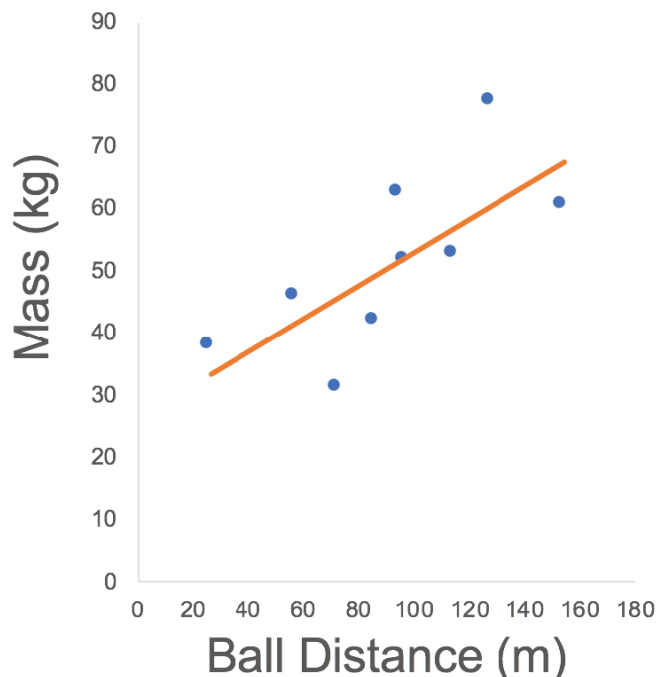


Figure 2. Relationship between mass and ball distance.

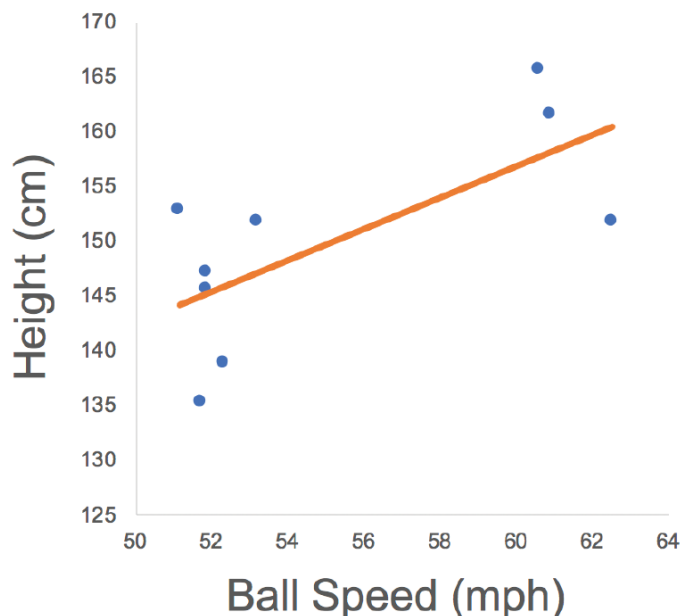


Figure 3. Relationship between height and exit speed.

Statement of Research Advisor

Johann's research has established normative data on a small subset of athletes that will allow for more in-depth exploration into total body mechanics and anthropometrics in hitting athletes of different skill levels.

– Gretchen Oliver, Kinesiology

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Mitochondrial Network Dynamics: Benefits of a High Metabolic Demand (Lactation) Condition in Rats

Ryleigh Randall, Hailey A. Parry, Hayden W. Hyatt, Wendy R. Hood, and Andreas N. Kavazis

Pregnancy and lactation are physically and metabolically challenging events in a female's lifetime. In this regard, the female body undergoes significant metabolic changes over the course of pregnancy that support the growing fetus and prepare for lactation. During pregnancy, the female body facilitates energy storage that can lead to increased risk of metabolic diseases such as type 2 diabetes. Lactation initiates a beneficial metabolic shift that reverses the gestation-induced energy storage to that of milk production.

Breastfeeding reduces postpartum adiposity by facilitating lipid transport to mammary glands for milk production. Importantly, the liver has a high metabolic rate and plays a key role in various pathways including milk production. The metabolic alterations that occur during pregnancy and lactation require cellular remodeling to facilitate these changes. Therefore, the purpose of this study was to investigate the effects of lactation on mitochondrial fission and fusion and autophagy in the liver.

Female rats (ten weeks old) were assigned to: (1) nonreproductive, (2) reproductive without lactation (did not suckle their pups), and (3) reproductive with lactation (suckled their pups) groups. Half of the animals from each group were sacrificed at peak lactation (14 days postpartum). The other half were sacrificed twelve weeks post weaning (weaning was done at 21 days postpartum). This resulted in six groups with eight animals per group. Several markers of mitochondrial fusion and fission and autophagy were measured in the liver via Western blots.

Autophagy is a process in which cells recycle old or damaged components and can play a role in liver remodeling during reproduction. In this regard, during peak lactation, the lactating animals had larger liver mass compared to those that did not lactate. However,

the liver mass in the lactating animals at the post weaning time were smaller compared to the animals that did not lactate. Our results suggest that an increase in autophagy may, in part, explain this size difference, as autophagy and cell growth are known to be conversely regulated.

Mitochondria are the powerhouse of the cell; they transform chemical energy from the food we eat into adenosine triphosphate (ATP), the energy currency of the cell. Therefore, it is not surprising that mitochondria are constantly undergoing remodeling based on the energy needs of the body. Two of the main process of mitochondria remodeling are fusion (the process of two smaller organelles combining to form one larger organelle) and fission (the process of cleaving one larger organelle into two smaller ones). Our results indicate that the liver mitochondrial network is regulated via the fusion pathways, but further research is warranted.

In conclusion, lactation has rapid and persistent beneficial effects and provide additional support of the Stuebe and Rich-Edwards "reset hypothesis" which posits that lactation helps to reset maternal metabolism postpartum.

Statement of Research Advisor

Ryleigh is a hard worker, an extremely fast learner, and a dependable student researcher who has helped to collect these data to better understand female reproductive health.

– *Andreas Kavazis, Kinesiology*

Effect of the Wooden Breast Myopathy on Broiler Chicken Collagen and Myogenic Regulatory Factor Protein Expression

Lauren Shortnacy, Joshua J. Flees, Charles W. Starkey, and Jessica D. Starkey

The global demand for chicken meat continues to rise because it is a nutritious and affordable source of protein. In response to the increased demand for chicken meat, the poultry industry has selected broiler chickens for rapid growth rate and high breast meat yield. Wooden Breast is a condition recently observed in rapidly growing, high-yielding broiler chickens reared for meat. The *pectoralis major*, or breast muscle, of chickens affected with Wooden Breast is extremely rigid or hard to the touch and can result in a negative eating experience for consumers. Currently, the broiler industry is losing millions of dollars each year due to various issues related to this meat quality defect. However, the reason why the Wooden Breast defect occurs is still unclear. Muscle growth is regulated by the expression and function of myogenic regulatory transcription factors such as myoblast determination protein 1, or MyoD. Insights regarding the amount and type of collagen infiltrating the affected muscle tissues as well as the muscle stem cell transcription factor protein expression profiles at various stages of production are needed. Knowledge regarding differences in affected and unaffected birds at the cellular and molecular level will aid in the effort to understand the problem in the muscles of the rapidly growing broilers affected with the Wooden Breast phenotype.

The goal of this research was to test the hypothesis that broiler chickens affected with the Wooden Breast myopathy will have elevated concentrations of myogenic regulatory factors and collagen in their breast muscle. Muscle samples were obtained from broilers affected with Wooden Breast ($n = 8$) and unaffected/normal broilers ($n = 8$) and reared to either 25 or 43 days of age. The Wooden Breast phenotype can be visualized at 25 days of age. The common processing age for U.S. broilers is 43 days. In accordance with these milestones, total protein was extracted in this study from the samples ($n = 32$ total samples) and electrophoresed on polyacrylamide gels and transferred to PVDF

membranes. Target protein concentrations were assessed using an established multiplexed, fluorescence-based Western blot detection method. The background corrected volume of each target protein signal (Figures 1C and 2C) was normalized to total protein (Figures 1D and 2D) in the respective lane or channel for DNA fragments to travel when electric current is applied.

On day 25, there was no change in the protein expression of collagen ($P = 0.7072$; Figure 1A) and MyoD ($P = 0.9636$; Figure 1B) between birds unaffected and affected by Wooden Breast. On day 43, collagen protein expression was significantly increased in birds affected by the Wooden Breast muscle defect compared to birds unaffected by the myopathy ($P = 0.0051$; Figure 2A). There was no change in the expression of MyoD on day 43 ($P = 0.7027$; Figure 2B). Results from this experiment show that collagen expression increases on day 43 when broilers are severely affected by the Wooden Breast defect compared to broilers that are unaffected, but expression of MyoD was similar.

Ultimately, the results of this study provide the basis for further exploration of the Wooden Breast meat quality defect and possible selection targets for producers to work with to eliminate this undesirable condition in commercial broiler chicken production.

Statement of Research Advisor

Lauren's work related to determining the proteomic profiles of broilers unaffected and affected with the Wooden Breast myopathy is novel and will serve as the basis for further experiments in our laboratory to be conducted by future students. The results of Lauren's project will be included in a peer-reviewed journal article of much larger scope that will ultimately serve as a contribution to the scientific literature concerning the Wooden Breast broiler meat quality defect.

– Jessica Starkey, Poultry Science

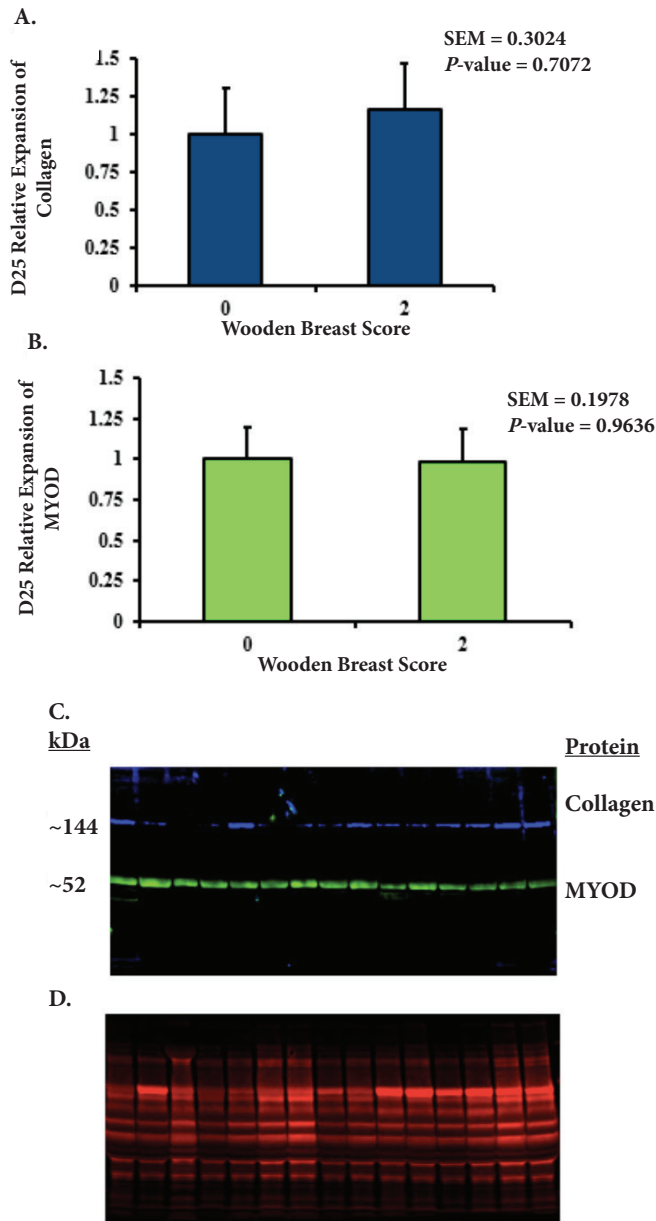


Figure 1. Relative protein expression of collagen and MyoD in pectoralis major muscle samples from normal (score 0) and Wooden Breast-affected (score 2) broiler chickens at 25 d posthatch ($n = 8$ per score). The expression of collagen (Panels A and C (blue bands)) and MyoD (Panels B and C (green bands)) proteins were quantified relative to total protein (Panel D). Means were separated at $P < 0.05$.

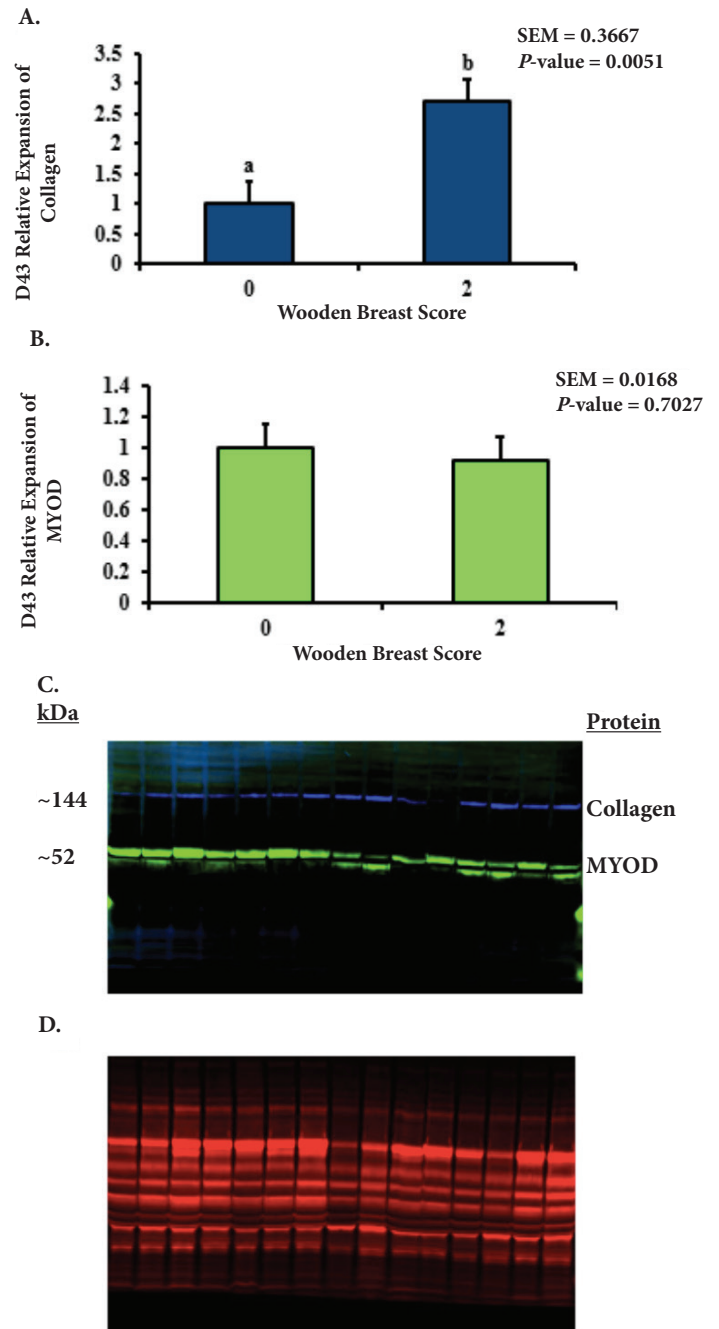


Figure 2. Relative protein expression of collagen and MyoD in pectoralis major muscle samples from normal (score 0) and Wooden Breast-affected (score 2) broiler chickens at 43 d posthatch ($n = 8$ per score). The expression of collagen (Panels A and C (blue bands)) and MyoD (Panels B and C (green bands)) proteins were quantified relative to total protein (Panel D). Means were separated at $P < 0.05$ and bars with different letter superscripts differ.

Identification of Key Genes Influencing Aggressiveness and Metronomic Treatment for Prostate and Breast Cancer

Elena Skarupa, Taraswi M. Ghosh, and Robert D. Arnold

Prostate and breast cancers are among the leading causes of cancer deaths in the United States. Treatment options for prostate and breast cancer are surgical resection, sterilizing radiation, hormone ablation therapy, and/or chemotherapy. Chemotherapeutic drugs alone or in combination remain the primary treatment options for aggressive and metastatic cancers; however, dose-limiting toxicity and drug resistance limits their clinical utility. Therefore, novel treatment strategies that improve survival or delay disease progression are needed. Our overall goal is to tailor therapies by using tumor specific molecular signatures for individual patients with aggressive metastatic cancers.

Using human prostate cancer cell lines, PC-3 (metastatic castration resistance; mCRPC) and LNCaP (androgen-sensitive), were used to determine the antitumor activity of conventional (CONV) and metronomic (METRO) dosing of the anticancer agent topotecan (TOPO) by protein staining (SRB) and metabolic activity (MTT). Concentration-dependent decrease in SRB and MTT were observed following increased drug concentrations. Changing the media/drug daily to mimic METRO increased potency (IC_{50}) ~8-fold ($p < 0.05$) compared to CONV dosing after 72hr exposure.^{1,2} These data demonstrated that METRO dosing resulted in similar anticancer activity as CONV dosing, but at lower drug concentrations. Cancer pathway gene-based expression studies following METRO and CONV dosing *vs.* control (no-drug treatment) were performed. We identified several genes as signatures for aggressiveness in prostate cancer (PLAU, TGFB1, SERPINE1, MET, TIMP1, CXCL8, MCAM, ITGA3) and altered the following genes METRO therapy (CDKN1A, ERBB2, SERPINB5, ITGA3, PLAU, GZMA and ITGA4).

Although differences in gene expression were observed, their translation into proteins needed to be verified. The specific goal for this project was to validate the expression of a number of candidate genes *via* protein

immunoblotting (Western analysis). Protein was isolated from a diverse panel of prostate cancer cell lines (PC-3M, PC-3, DU-145, LNCaP, and 22RV-1: arranged most to least aggressive (Figure 1A). Immunoblotting was achieved using a 4-14% gradient gel and Actin-Beta as a housekeeping (control) protein. My research focused on validation of top significant genes with immunoblotting. Here we show the effect of treatment schedule (CONV- *vs* METRO-TOPO) on SERPINE1, a gene associated with aggressiveness in prostate cancer. SERPINE1 is a serine proteinase inhibitor gene that is important in making plasminogen activator inhibitor 1 (PAL-1), which promotes tumor progression. Research has shown that SERPINE1 controls plasmin and initiates migration and remodeling of body tissues.¹

In summary, METRO administration of TOPO increased its antitumor activity in comparison to CONV high-dose chemotherapy. Unique gene signatures were identified in aggressive cancer and following METRO dosing. Immunoblot analysis verified that SERPINE1 was downregulated significantly ($p < 0.05$) following METRO dosing (Figure 1B). The finding supports the hypothesis that METRO dosing schedules could be utilized to personalize patient therapy, based on an individual's molecular signature. Future studies will continue validation of treatment-mediated effects on the genomics and transcriptomics associated with various molecular signatures using molecular (CRISPR-Cas9 gene editing) and conventional pharmacological techniques in both aggressive prostate and breast cancers. The effect of tumor stroma (e.g., macrophages or fibroblasts) in 2D and 3D-tumor spheroids on treatment efficacy will be determined.

Statement of Research Advisor

The development of advanced “omic” tools have ushered in a new era of research that will provide greater mechanistic insights into disease progression and treatment response. Elena has worked to validate

molecular signatures that we have identified for aggressive prostate cancer and therapeutic response to metronomic therapy. Her research will support development and optimization of novel dosing strategies that may be tailored for a patient's specific cancer.

– Robert “Rusty” Arnold, Drug Discovery & Development

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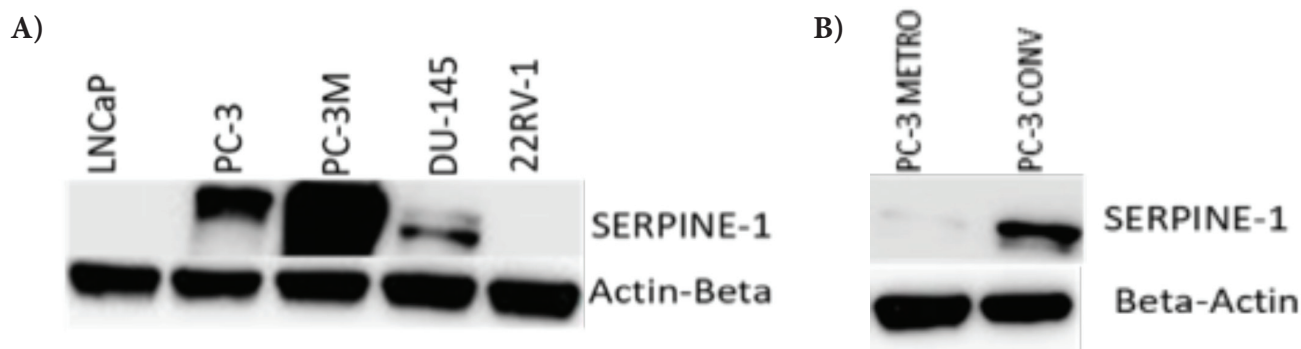


Figure 1. *In vitro* validation- Immunoblotting. (A) **SERPINE-1** was identified as the top marker for aggressiveness in mCRPC (unpublished data) and its expression was increased according to aggressiveness. The most aggressive cell lines (PC-3M and PC-3) showed greatest amount of **SERPINE-1** protein. (B) METRO-TOPO treatment downregulated SERPINE-1 compared to CONV.

Late-Stage Peptide Diversification via Twisted Amide Bond Formation

Xavier Streety, Mahesh Sriram, Victor Adebomi, and Monika Raj

Peptides occupy a biomolecular niche between that of small molecules and proteins. Given that they can function not only as substrates in biochemical pathways, but as signaling molecules, peptide therapy presents a unique opportunity for medicine to closely mimic natural pathways.¹ As new challenges, such as mutant viral and bacterial strains with resistance arise, the need for modification of these peptide therapeutics has increased. In recent years, the use of acyclic twisted amides as reactants for cross-coupling reactions has been investigated. These twisted amides feature a distorted amide bond that has reduced double bond character and thus allows for the bond to be broken and replaced with various new bonds.²

We sought to find a method to selectively introduce these twisted amides into peptides and test their reactivity toward cross-coupling and nucleophilic acyl substitution reactions to introduce new functionalities. Amide bonds form the backbone of peptides and thus are a desirable synthetic handle for modification. Therefore, methodology to selectively modify peptides using these amide bonds leads to more possible peptide derivatives.

We started this process by investigating oxazolidinone and thiazolidinone, derivatives of serine and cysteine, respectively, using density functional theory (DFT) calculations to determine whether these structures exhibited a twisted amide bond. DFT is a computational

quantum mechanical modeling method used to investigate the electronic structure of large molecules. From these initial calculations, we determined four potential moieties for twisted amides applicable to peptides: the aforementioned oxazolidinone and thiazolidinone structures in addition to their thiocarbonyl analogs.

Following the calculations, we moved to the synthesis of these moieties on small molecules before moving to the synthesis of these moieties on tripeptides. Following each step of synthesis, each product was purified using silica gel chromatography and confirmed by mass spectrometry and nuclear magnetic resonance (NMR). For work on small molecules, the methyl ester derivative of either serine or cysteine was used as a starting point. These small molecules with added carbonyl or thiocarbonyl groups were then reacted with benzoyl chloride to yield the desired potential twisted amide functionality. These potential twisted amides were tested as reactants in cross-coupling and nucleophilic acyl substitution reactions.

Work on small molecules gave positive results as these molecules featuring twisted amide moieties were successfully used in transamidation, esterification, Suzuki-Miyaura coupling, and Friedel-Crafts acylation. This result supports the formation of twisted amides as a method to introduce modifications to peptides.

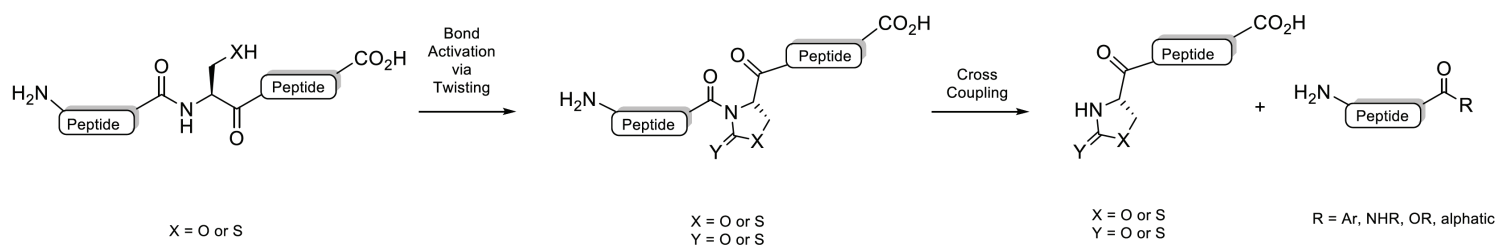


Figure 1. General scheme for peptide diversification using twisted amides.

The introduction of twisted amides to tripeptides is ongoing. Once these twisted peptides are formed, they will be tested as reactants in the previously mentioned coupling reactions to form modified peptide derivatives. Positive results in these tests support using the introduction of twisted amides to peptides as a method for late-stage diversification.

Statement of Research Advisor

Xavier has developed a new pathway for the synthesis of peptides in solution on a large scale. He also developed a new method for activating the unactivated, highly stable amide bonds for various cross-coupling reactions by twisting them.

– *Monika Raj, Chemistry and Biochemistry*

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Determining KSHV Copy Number Using Quantitative PCR (qPCR) with ERV-3 Sequence Embedded in Genomic DNA as a Reference

Jake Tatum, Huachen Ganand, and Joanna Sztuba-Solinska

Kaposi sarcoma-associated herpesvirus (KSHV) is an oncogenic herpesvirus and causative agent of Kaposi's sarcoma, primary effusion lymphoma, and Multicentric Castleman disease (Figure 1A)[1]. KSHV displays two phases of infection cycle. During latency, the viral DNA persist as an episome attached to the host chromatin and displays limited expression of viral genes. During the lytic phase, the full repertoire of genes is expressed, and viral progeny are produced. Latent viral replication of herpesviruses differs from lytic viral replication in at least three distinct ways: (1) latent replication initiates at ori-P and proceeds bidirectionally, while lytic replication originates from a distinct origin, referred to as ori-Lyt, and proceeds through a rolling circle mechanism; (2) latent viral replication depends on host cellular DNA polymerase and accessory factors, while viral lytic replication utilizes its own DNA polymerase and other factors; and (3) latent DNA replication occurs in synchrony with host DNA replication, maintaining a stable number of viral episomal DNA in each cell, while lytic replication amplifies DNA 100 to 1,000 fold. The goal of my project is to set up a real-time quantitative PCR (qPCR) assay aimed at measuring the KSHV episomal DNA load in naturally infected and recombinant cell lines.

The model system is KSHV positive body cavity-based lymphoma cells (BCBL-1), which allows the controlled chemical manipulation of viral infectivity cycle [2]. The cells are maintained in RPMI-1640 medium (ThermoFisher, 11875085) supplemented with 10% fetal bovine serum (FBS), 1% Penicillin/Streptomycin, 1% L-Glutamine at 37 °C in 5% CO₂. The treatment with histone deacetylase inhibitor, sodium butyrate, stimulates the viral latent-to-lytic switch by uncoiling of DNA leading to transcriptional activation of viral lytic genes. Carefully timed collection of cells post sodium butyrate induction, allows examining the KSHV episomal DNA copy number at immediate early (8 hours post-induction, (hpi)), early (24 hpi), and late

(48-72 hpi) stages of lytic, as well as before the treatment, during latency (0 hpi).

Two reference plasmids encoding K6 and ERV-3 genes were used to create standard curves that allow quantifying the viral episome and genomic DNA copy number, respectively. The K6 gene codes for viral macrophage inflammatory protein 1 and is present in each viral episome at a single copy. The ERV-3 is endogenous retrovirus that integrated into the human genome 40-100 million years ago and is also at a genomic copy number of one per human cell [3]. Both plasmids were first cloned into *Escherichia coli* DH5 alpha bacterial cells, purified with the Qiagen plasmid Midi kit, quantified, and used to create serial dilutions that are directed to qPCR analysis. Specific primers were used to amplify both products (Table 1).

Two standard curves of the cycle threshold values (Ct) plotted against the logarithm of the copy number were used to deduce the total episomal and genomic DNA copy number in each experiment (Figure 2). Having been established, this assay will be used in future work to (1) estimate the viral DNA copy number at different time-points of KSHV infectivity cycle in BCBL-1 as well as other patient-derived primary effusion lymphoma cell lines, and (2) address how specific mutations introduced within KSHV locus encoding important long noncoding RNA, i.e., polyadenylated nuclear (PAN) RNA, affect viral replication. The work is significant in that reproducible, sensitive, and specific quantitative techniques are needed to assess various hypotheses regarding the viral load and its correlation with different experimental conditions and clinical samples.

Statement of Research Advisor

Jake Tatum has established two standard curves for K6 and ERV-3 genes that are essential for estimating the KSHV episomal DNA copy number in BCBL-1 and patient-derived primary effusion lymphoma cell lines.

This assay will be also applied for studying specific mutations within KSHV locus encoding polyadenylated nuclear (PAN) RNA, a key regulator of KSHV infectivity cycle. His work largely contributes to the understanding of KSHV molecular biology and pathogenicity.

-Joanna Sztuba-Solinska, Biological Sciences

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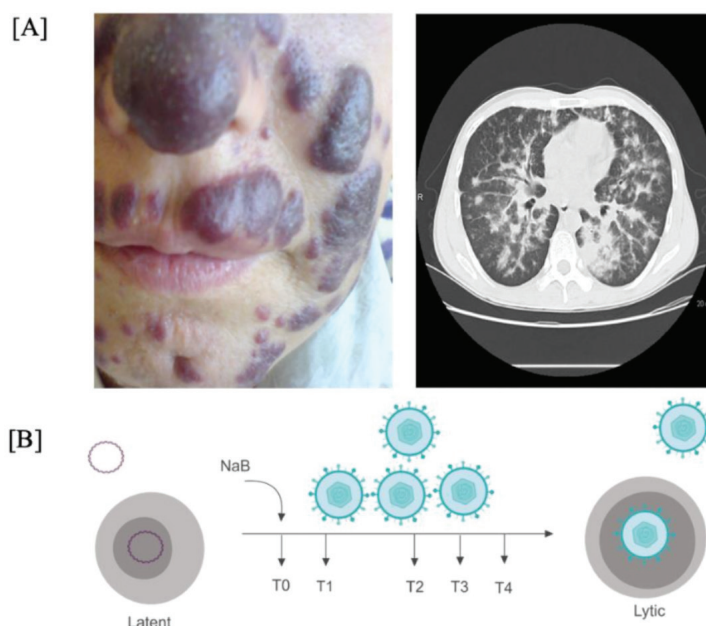


Figure 1. [A] Symptoms of Kaposi's sarcoma related to KSHV infection. [B] Experimental set-up used for manipulating KSHV latent (T0) and immediate early (T1), early (T2) and late lytic (T3-T4) lytic stages of KSHV replication.

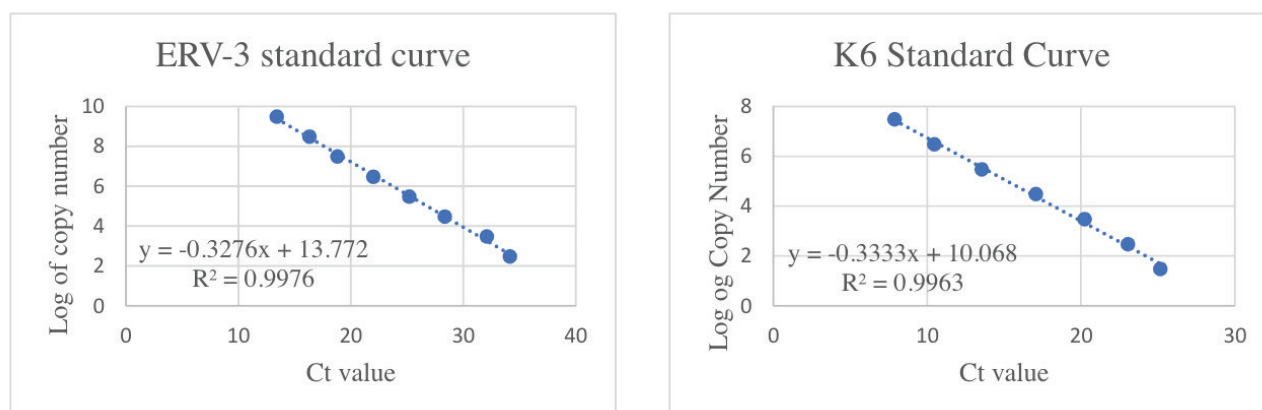


Figure 2: Two standard curves established with control plasmids ERV-3 and K6.

Table 1. Oligonucleotides used in the following study.

Name	5' -3' sequence	Purpose
K6_Forward	CGC CTA ATA GCT GCT GCT ACG G	Amplification of K6 specific viral locus
K6_Reverse	TGC ATC AGC TGC CTA ACC CAG	
ERV3_Forward	CAT GGG AAG CAA GGG AAC TAA TG	Amplification of ERV-3 genomic product
ERV3_Reverse	CCC AGC GAG CAA TAC AGA ATT T	

ROTC Cadets' Help-Seeking Attitudes: Examining the Role of Leadership Style

Kristyn Waldron, Haley Sherman, and Mallory Lucier-Greer

In 2016, approximately 20% of U.S. active-duty military members met the criteria for a mental health disorder (Deployment Health Clinical Center, 2017). However, less than half of military members with a mental health disorder seek support services (Cuyler & Guerrero, 2019). Some correlates of help-seeking have been identified, such as fear of being viewed as weak and fear of experiencing rejection within one's military unit. This study investigated how a person's approach to leadership is associated with dimensions of help-seeking among a sample of emerging military leaders who are college/university students enrolled in Reserve Officers' Training Corps (ROTC) programs (i.e., ROTC cadets). Specifically, this study sought to answer two research questions:

Research question 1: *What is the factor structure of the Attitudes Toward Seeking Professional Psychological Help (Fisher & Farina, 1995) measure among ROTC cadets?*

Research question 2: *How is leadership style associated with help-seeking?*

Data were collected from 69 ROTC cadets via an online survey; 42% were women, 37% were men, and 21% did not disclose their gender. The average age was 20.68 years old. An exploratory factor analysis (EFA) was conducted to examine the factor structure of the *Attitudes Toward Seeking Professional Psychological Help* measure. The EFA revealed two factors: *openness to seeking treatment and value in seeking treatment*, both of which align with the original construct of the scale (Fisher & Farina, 1995; Table 1). Mean scores suggest that this sample of ROTC cadets is relatively *open to seeking treatment* ($M = 2.62$, $SD = 0.89$, range 0-3) and they see *value in seeking treatment* ($M = 2.78$, $SD = 0.78$, range 0-3).

The relationship between leadership style and help-seeking was examined through linear regression analyses, accounting for age. Leadership style was assessed with two measures: the Northouse's Leadership Scale (2001), which consisted of the *task-oriented leadership* and *relationship-oriented leadership* subscales, and the Socially

Responsible Leadership Scale, which included the *socio-cultural conversations with peers* and *commitment to responsibilities* subscales (SRLS; Dugan & Komives, 2010). Higher scores on relationship-oriented leadership and the ability to have socio-cultural conversations with peers were significantly related to more openness to seeking treatment. In other words, leaders who are invested in their relationships with their team and are able to have meaningful, sometimes difficult, conversations were more likely to be open to seeking mental health services. None of the leadership measures were related to value in seeking treatment.

Implications of this work are meaningful across several disciplines. First, multiple dimensions of help-seeking have been identified in other work and confirmed in this sample of ROTC cadets. It appears that help-seeking is both a mindset (e.g., do you see value in seeking services?) and a potential for behavior (e.g., would you be willing to see a professional?). Along with other variables identified as salient predictors of help-seeking in prior research, some dimensions of leadership may also be of importance. Military leadership trainings may benefit from promoting relationship-oriented leadership and emphasizing the importance of having socio-cultural conversations with peers. These aspects of leadership may promote positive mental health within leaders and, in turn, the units they lead.

Statement of Research Advisor

Kristyn's research is an important step in addressing mental health in the military. By examining a sample of ROTC cadets, she is getting a pulse of what military leadership will look like in the next 5-10 years. These results are encouraging as this sample of emerging military leaders appear to value and be open to seeking mental health services, which is vital for their own success as a service member and for the unit's they lead.

-Mallory Lucier-Greer, Human Development and Family Science

Table 1. Results of exploratory factor analysis (EFA) of the 10 items from the help-seeking scale among a sample of ROTC cadets. **Note.** The blue and orange highlight indicates that the item loaded onto either factor 1 or 2.

Please indicate the degree to which you agree or disagree of each statement.	Factor 1: <i>Openness to seeking treatment</i>	Factor 2: <i>Value in seeking treatment</i>
Item 1. If I thought I was having a mental breakdown, my first thought would be to get professional attention.	.664	
Item 2. Talking about problems with a counselor/therapist seems to me as a poor way to get rid of emotional problems.		.569
Item 3. If I were experiencing a serious emotional crisis, I would be sure that counseling/therapy would be useful.	.870	
Item 4. I admire people who are willing to cope with their problems and fears without seeking professional help.		.575
Item 5. I would want to get professional help if I were worried or upset for a long period of time.	.864	
Item 6. I might want to have counseling/therapy in the future.	.716	
Item 7. A person with an emotional problem is not likely to solve it alone; he or she is more likely to solve it with professional help.	.694	
Item 8. Given the amount of time and money involved in counseling/therapy, I am not sure that it would benefit someone like me.		.686
Item 9. People should solve their own problems, therefore, getting counseling/therapy would be their last resort.		.830
Item 10. Personal and emotional troubles, like most things in life, tend to work out by themselves.		.785

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Biocidal Properties of Europa Lander Solid Rocket Motor Adhesive, Loctite® EA9394, Against *Hypsibius dujardini*

Natalie R. Williams, Mark R. Liles, and Morgan R. Sisk

An ice-covered moon of Jupiter, Europa is a prime candidate as a potentially viable environment in NASA's search for extraterrestrial life. Since Europa is a celestial body unlike any NASA has explored before, new issues regarding forward contamination of Europa and sterility of the spacecraft arise. These novel problems that often accompany exploration of new frontiers must be solved in accordance with NASA planetary protection standards, and launch of the Europa Lander is ultimately contingent upon the fulfillment of these criteria.¹

The Europa Lander Mission launch criteria requires spacecraft decontamination of terrestrial microorganisms to limit the probability of contaminating Europa's surface. These standards include reducing microorganisms present on the lander by at least 10^{-6} and a final European contamination probability of $10^{-4}\%$.² For the mission to be a success, the microbiome of certain spacecraft problem areas must be quantified, and the inherent antimicrobial or biocidal properties of native rocket motor materials must be characterized. We discovered that a frequently used rocket motor adhesive, Loctite® EA9394, can reduce the number of viable *Bacillus atrophaeus* vegetative cells by 10 million-fold, and spores by greater than 100-fold, after short-term exposure. Though spore-forming bacteria have traditionally been the primary targets of planetary protection efforts, eukaryotic *Hypsibius dujardini* (i.e., tardigrades, or "water bears") is the best animal model for planetary protection given their known resistance to radiation, desiccation, and space environments.² This study evaluated susceptibility of *H. dujardini* to exposure to Loctite® EA9394.

Viability assays were performed with populations of active *H. dujardini* in water and several concentrations of water-soluble extracts of Loctite® EA9394 using 70% ethanol as a positive control and spring water as a negative control. Final concentrations of extracts in

solution ranged from 1.6 mg/mL to 0.025 mg/mL. After 24 hours, each replicate was observed under a microscope at 100X magnification. Immobile tardigrades were classified nonviable, while spontaneously mobile tardigrades were classified viable.

We found that increasing adhesive concentration correlated with an increase in tardigrade death. There was a statistically significant difference of percent viability from 1.6 to 0.8 mg/mL of extract when compared to negative control, corresponding to 100% and 94.9% viability reduction, respectively. In summary, active *H. dujardini* populations suffered an average of 100% viability loss when exposed to 1.6 mg/mL of water-soluble extracts for 24 hours. The minimum inhibitory concentration at which 50% viability was observed (MIC_{50}) was approximately 0.2 mg/mL.

The results of this study suggest the risk posed by radiation and desiccation-resistant *H. dujardini* could be mitigated through the biocidal activity of Loctite® EA9394. These reductions can be incorporated into the launch risk assessment for the Europa Lander Mission. As this is an ongoing project, next steps include inducing anhydrobiosis in *H. dujardini* and testing these dormant, extremotolerant populations against Loctite® EA9394 to calculate percent viability reductions as done before, as well as identifying the sporicidal and/or tardigradical compounds in the adhesive using mass spectrometry.

Statement of Research Advisor

Natalie Williams has made very significant contributions to the NASA Europa Lander mission in identifying an adhesive that can be used to reduce microbial contamination of the solid rocket motor insulation. She has developed many skills in quantifying adhesive effects on viable numbers of bacterial spores and tardigrades, and these data will be very useful for NASA's planetary protection mission.

– Mark Liles, Biological Sciences

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Motor Learning with EEG Controlled Virtual Muscle

Duncan Wisniewski and Mark Adams

This research addresses whether fast skill motor learning, a subset of motor skill learning that acts to rapidly decrease response time as an activity is repeated, occurs when using an electroencephalogram (EEG) virtual muscle control system. Indirectly, this addresses the question of the motor homunculus' role in motor learning. Previous studies on the topic of motor learning implicate the motor homunculus as playing a necessary role, since its activation is seen in fast skill motor learning tasks. However, it has been unclear whether the motor homunculus activated because the use of muscles was necessary in the task or because the area is needed for motor learning. This study seeks to clarify the reason for activation of the motor homunculus and does so by using a virtual muscle, not the motor homunculus, to participate in a fast skill motor learning task. This issue is important because some brain-computer interface-controlled prosthetics use EEG as a control system and may be unable to properly utilize motor learning if it requires the muscle being used to be a part of the motor homunculus.

To investigate this topic, I constructed a custom EEG virtual muscle by using the Fast Fourier Transform to find the power in the 7-14 Hz range and using it to control a program that simulates a single player game of pong. Using alpha wave power to control the height of a paddle on one side of the screen, the participant attempts to reflect the ball back towards the other side of the screen. As participants played, their response time to each new ball was recorded. The participants' response times indicated no negative trend that would signify the presence of motor learning. Motor learning would be signified by a decrease in response time as the participants played the game longer; however, the average response times remained constant throughout the time spent playing.

The lack of motor learning while using a virtual muscle system means that those using prosthetic limbs may not be able to form muscle memory for some tasks; however, the motor homunculus is adaptive and can adjust to the loss of a limb. Given this area's

tendency to adapt, it may be able to adjust to adding new areas as well. A further study using functional magnetic resonance imaging (fMRI) could test whether the motor homunculus is being used during the fast skill motor learning pong task, and whether the motor homunculus is necessary to the development of muscle memory. The findings of this study indicate that it may take significantly longer than usual to develop muscle memory when using an EEG-controlled prosthetic, since the motor homunculus may need time to adapt to the prosthetic.

Statement of Research Advisor

Duncan's research explored whether motor skills could be learned through a virtual process, which is significant to people who have lost limbs and use prosthetics. He successfully implemented an EEG-based virtual muscle control system to monitor the development of motor skills through mental repetition. Although the data collected indicated that actual movement may be necessary to develop motor skills, the research opened the door for additional studies to determine whether the motor homunculus is being used during the fast skill motor learning pong task, and whether the motor homunculus is necessary to the development of muscle memory.

-Mark Adams, Electrical Engineering

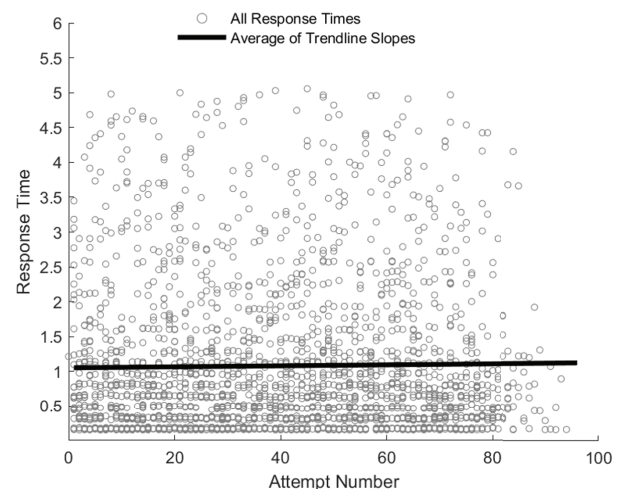


Figure 1. Response Times Relative to Pong Attempt Number Scatterplot with Trendline.

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