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AUJUS is a collaborative effort by students at Auburn University

The AUJUS cover artwork shows the interaction between an antibody and a potassium channel from *Streptomyces lividans*. Antibodies are key proteins in the immune system and understanding the interactions they use to bind to other proteins can enable new methods of designing medicines or diagnosing diseases. The structures are from Protein Data Bank file 2ATK and the image was made with the UCSF Chimera package, which is supported by NIGMS P41-GM103311 [1,2].

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AUJUS Editorial Team



Production Editor: Brooklyn Holt is a senior pursuing a Bachelor of Arts degree in Journalism with a minor in Business. She is a Communications Assistant for the Office of Undergraduate Research. Brooklyn's other on-campus involvement includes serving as an Honors College Ambassador, a College of Liberal Arts Ambassador, a member of Cardinal Key Honor Society, and a Majorette in the Auburn University Marching Band. Brooklyn also works on Auburn's campus as a Precalculus Supplemental Instructor. In her spare time, Brooklyn enjoys taking part in outdoor activities, attending sporting events and visiting the beach or lake. Upon graduation, she plans to pursue a career in the corporate communications arena.



Associate Editor: Ben Nelson is a senior pursuing a Bachelor of Science degree in Biomedical Sciences. In his sophomore year, Ben became involved in immunology research within the Department of Biological Sciences in the lab of Dr. Elizabeth Schwartz. His research focuses on bacteriophage adsorption under inflammatory conditions. Ben is a Project Uplift mentor and serves as a COSAM peer advisor.



Associate Editor: Philip Wang is a senior pursuing a degree in biomedical sciences and has served as the AUJUS editor since spring 2016. Philip has been involved in undergraduate research since fall of his freshman year. His research, under the guidance of Dr. Geoffrey Hill in the Department of Biological Sciences, focuses on animal colorization and its physiological effects. Philip also volunteers at East Alabama Medical Center's Emergency Department, as a coach for Auburn High School's Debate Team, and at the Crisis Center in Birmingham. In his spare time, he enjoys trying new restaurants, backpacking, and writing with fountain pens.



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 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. Wolf's main research interests are in earthquake and geologic hazards.

MENTOR OF THE YEAR: Dr. Sushil Adhikari



"I like exploring things and finding new ways to make things work," says Dr. Sushil Adhikari, Alumni Professor and Director of the Center for Bioenergy and Bioproducts. Adhikari began his research journey while earning his PhD in Biological Engineering at Mississippi State University. Since then, Adhikari has mentored more than 60 students in biosystems research focused on converting trees, grasses and algae into biofu¬els and bioproducts.

"I like exploring things and finding new ways to make things work."

Adhikari says he loves research, because it gives him the opportunity to test new ideas. "There is always something new going on in the laboratory, so life is not boring." According to Adhikari, research is a "new adventure" everyday.

Adhikari encourages all students who are considering undergraduate research to take part in the transforma¬tive experience. "It [undergraduate research] is the best professional experience that you can have," says Adhikari. "Grab it when you can." According to Adhikari, an undergraduate research experience pro¬vides students with professional ethics. Adhikari notes that students learn beyond the equipment in the lab, gaining teamwork skills and the confidence to ask others for guidance.

"[Undergraduate research] is the best professional experience that you can have." Adhikari says his main goal as a mentor is to be flexible and to make a positive impact on his students, even if it is a small one. Through mentoring, Adhikari has discovered that all students learn differently; some are fast learners while others take a little longer to grasp concepts. Adhikari says he tries to find ways to encourage students to continue their research experiences based on the students' strengths and weaknesses.

Having mentored students for 13 years, Adhikari continues to mentor students because he values the trans¬fer of knowledge. "I want to excite them about the research, and [help them] learn certain skill sets that they might not have an opportunity to learn in their curriculum." Adhikari strives to teach students beyond what they learn in the classroom.

"I want to excite [students] about the re¬search, and [help them] learn certain skill sets that they might not have an opportunity to learn in their curriculum."

Adhikari claims that the biggest reward of mentoring undergraduate students is when he receives updates from former students who share with him how valuable their research experience was and how it applies to their current job or graduate studies.

RESEARCH HIGHLIGHTS

Kinematics of Youth Baseball Pitching and Football Passing

Kathryn Clardy, Sarah Gascon, Gabby Gilmer, Jessica Washington, Gretchen Oliver

The overhead throwing motion in youth baseball pitching is associated with overuse injury. In an attempt to prevent this, physicians have recommended that youth athletes play multiple sports, deterring sport specialization at an early age. When youth baseball pitchers are encouraged to participate in additional sports, they tend to take an interest in football, specifically the position of quarterback. Both the baseball pitcher and the football quarterback use an overhead throwing motion. Additionally, when conditioning, it is common for professional baseball pitchers to throw a football to improve arm strength and for football quarterbacks to throw a baseball to improve on arm speed [1,2,3]. Because participating in dual sports is common practice and has training implications, we aimed this study at comparing the kinematics of the baseball pitch and football pass. We hypothesized that significant differences in kinematics would be observed between these different types of throws.

Fifteen male quarterbacks $(13.63 \pm 1.25 \text{ years}; 169.32 \pm 8.01 \text{ cm}; 62.33 \pm 10.17 \text{ kg})$ and eighteen male pitchers $(14.33 \pm 1.58 \text{ years}; 174.94 \pm 7.93 \text{ cm}; 69.05 \pm 13.99 \text{ kg})$ participated. The participants came into the lab for data collection in the spring and summer of 2016. Kinematic data were collected with The MotionMonitor^{**} synchronized with an electromagnetic tracking system at 100 Hz. Quarterbacks threw three passes to a receiver 13.7 m away and pitchers threw three overhand pitches to a catcher 14 m away. The throwing motion of both pitching and passing were divided into four events: foot contact (FC), maximum shoulder external rotation (MER), ball release (BR), and maximum shoulder internal rotation (MIR) (Figure 1).

All data were analyzed using Mann-Whitney U-tests. Baseball pitchers had greater trunk flexion at MIR and external rotation of the shoulder at BR. Baseball pitchers also exhibited greater trunk rotation opposite the throwing arm side at BR. These differences may have been a result of how the pitchers adjust to keep their center of gravity above the stride leg as they throw down from a mound.



Figure 1: Illustration of football and baseball throwing events

Throwing from an inclined surface would result in greater momentum toward the target and be followed with a greater rotation opposite the throwing arm side. Football quarterbacks displayed greater shoulder horizontal adduction at FC, greater shoulder external rotation at BR, and greater

elbow flexion at FC and MER. Each of these differences could be attributed to the increase in size and weight of a football versus a baseball.

Results showed kinematic differences that would support the use of different training modalities for the overhand throwing athlete. Therefore, using a football as a weighted ball training technique may increase performance without increasing the occurrence of overuse injury in youth sport. Both baseball pitching and football passing techniques also have biomechanical benefits such as skill acquisition and improved athleticism for young athletes, supporting their long-term athletic development. Participation in both sports could reduce the risk of overuse injuries and burnout because of the differences in the mechanics for the two sports. Future research should investigate sport specialization in larger populations, as well as compare other overhead throwing sports.

Statement of Research Advisor:

Kathryn assisted in data collection, data analysis, and writing of these data.

-Gretchen Oliver, School of Kinesiology

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Fresh Market Tomato Yield and Quality as Affected by Potassium Rates and Sources

Trevor Cofer, Elizabeth Guertal, James Pitts

For many Alabama vegetable growers, fresh market tomatoes represent a significant source of income. Growers often apply high rates of potassium fertilizer at planting because it is thought to be needed to promote growth and uniform ripening. Two tomato growers I spoke with before my project expressed concerns that the area around the stem of the fruit remained yellow throughout the season (called yellow shouldering). Since more potassium is required to color a tomato than to ensure full yield, if there is insufficient potassium at fruit set, the fruit will not color properly, no matter how long it stays on the plant.

The objective of this research was to examine rates and sources of potassium fertilizers and their effects on tomato growth, yield, and fruit quality. The one-year study was conducted during the summer of 2016 at the Auburn University Chilton Research and Extension Center in Clanton, AL. The experiment was arranged in a completely randomized block design, with three replications of each treatment. Potassium was applied at bed formation at rates up to the soil test recommendation of 180 lbs of K₂O/A (0, 60, 90, 120, 150 and 180 lbs K₂O/A) and with four potassium sources (potassium chloride, potassium sulfate, potassium thiosulfate, and potassium magnesium sulfate), all readily height and stem diameter were measured weekly. At one and two months of growth, plant tissue samples were collected and analyzed for percent potassium. At harvest, fruit was counted, weighed, and categorized based on complete or incomplete ripening (via visual inspection). After harvest, soil samples were taken and analyzed for extractable potassium (Mehlich 1 extract).

We found that tomatoes receiving potassium before planting had greater potassium content in leaf tissue than those that were not fertilized with potassium. However, this did not translate to increased fruit yield, nor did it reduce the instances of yellow shouldering of the fruit. Leaf potassium was likely not high enough early in the growing season to promote uniform ripening. Neither the potassium source nor potassium rate significantly affected tomato yield or occurrences of yellow shouldering over the harvest period at the applied rates, although the potassium rate did

significantly affect the plant's height and stem diameter A higher potassium rate promoted taller plants, whereas a lower potassium rate promoted increased stem girth. Soil test extractable potassium showed low to moderate extractable potassium, irrespective of potassium rate or source. Mean and median soil test extractable potassium of all plots were 64 and 60 lbs K₂O/A, respectively. We tested soil from a similar tomato study on the same research station that had no instances of yellow shouldering and found 299 lbs K₂O/A of extractable potassium, an extremely high value. Comparing results from the two studies, we concluded that although the potassium we applied was sufficient to maximize yield (which are how soil test recommendations are given) it was far from sufficient to promote uniform fruit ripening. Such information could help Alabama farmers evaluate their potassium application strategies based on soil testing recommendations.



Figure 1: Tomato affected by yellow shouldering.

Statement of Research Advisor:

Trevor's work with fresh market tomato fertility helps Alabama growers make informed decisions when purchasing their fertilizers. His work showed few differences due to the source of potassium. Thus, growers can purchase the potassium fertilizer that prices out the best for them, and still have good yield potential. —*Beth Guertal, Crop, Soil and Environmental Sciences*

Naturalistic Pen-Based Data Interaction John Cook, Hari Narayanan, Jeff Overbey

The proliferation of affordable multi-touch devices over the last five years has brought the power of data visualization and interaction to the average consumer. However, development and integration of pen-based input via new "smart stylus" tools have not resulted in data visualization applications that respond intelligently to markup and that enhance the naturalistic pen-and-paper interaction these tools are designed to replicate. The aim of this research is to create a system that provides natural pen-based data visualization and manipulation techniques and to evaluate it for feasibility and effectiveness against its touch-based "Windows, Icons, Menus, Pointer" (WIMP) counterparts. I have developed a spreadsheet application for the Apple iPad Pro that recognizes the pen gestures users would naturally use on a paper spreadsheet (circling, crossing out, highlighting, etc.; Figure 1) and maps them to their respective operations.

The application also provides features typically offered by electronic spreadsheets (sorting, computation, etc.).

For this project, I designed a user study in which fourteen undergraduate computing and liberal arts students first completed a number of gestures in isolation, then carried out a sequence of data manipulation tasks using both my application and its leading competitor, Microsoft Excel[™]. The former task provided data regarding the intuitiveness and usability of each naturalistic gesture, while the latter provided the same insights regarding the application and interaction scheme as a whole. An automatic time-stamping function collected timing data for the users' interactions with each application, and an administered questionnaire provided qualitative feedback.

А	в	С	D	E	F	G	н	I	J	к	L
13	14	15	21	17	18	19	20	53	22	23	24
25	68	27	28	29	30	201	32	33	34	35	19
37	38	39	40	0	42	43	44	45	1	28	12
11	42	12	1	X	30	17	13	6	4	15	29
28	0	37	22	45							
+	-	*	1								

Figure 1. Screenshot from the application displaying an example of a 'cross out' gesture to delete (left) and a line gesture to select (right).

The results showed that the pen-based interactions yielded significant time savings over traditional WIMP interactions. On average, completing a set of data interaction tasks was two times faster using pen-based interactions (~5.5 versus 2.7 minutes). Unsurprisingly, previous experience with Excel[™] was a predictor of increased task-completion speed using pen-based techniques (27% faster). Interestingly, liberal arts students completed the tasks 17% faster than computing students. While all of the individual pen gestures were more efficient than their WIMP-based counterparts, some gestures stood out as particularly efficient. Basic arithmetic operations require multiple steps to complete in Excel[™], but only two gestures using the pen-based system. As such, users completed these operations 88% faster using the pen-based techniques. Similarly, deletion and summation require multiple steps to complete using Excel[™], and users completed these tasks 70% and 66% faster (respectively) using the pen-based techniques. The learning curve for the multiple-selection interaction was somewhat high using the WIMP system, and users completed this interaction 62 times faster using the pen-based system when performing the gesture in isolation.

Overall, the feedback from the questionnaire indicated that users found the pen-based interaction techniques more enjoyable, more intuitive (with the exception of a gesture to circle and paste), and easier to use (though users with a large amount of spreadsheet experience were understandably more comfortable using Excel[™]). This positive feedback, combined with the significant increase in user efficiency, indicates that a spreadsheet interface that implements pen-based gestural interaction techniques is quantitatively and qualitatively superior to a traditional WIMP-based interface. While no spreadsheet or other data manipulation applications that provide a pen-based gestural interface exist today, this study proves that such a system is both feasible and preferred by the end users.

Statement of Research Advisor:

John Cook recognized a deficiency in spreadsheet and other data manipulation applications in the marketplace in that while end users increasingly work with such applications on devices like tablets and smartphones with no keyboard or mouse, the interfaces of such applications are still based on keyboard and mouse ineraction. He proposed a research project to develop and test a penbased gestural interface against the traditional interface on a spreadsheet, succeeded in getting the project funded through an Auburn University Undergraduate Research Fellowship, and carried the project to a successful conclusion. His work provides a compelling argument for new interface designs for the "smart" devices of the present and future.

—Hari Narayanan, Computer Science and Software Engineering

Effects of Upper Extremity Pain History on Softball Pitching Mechanics of the Screwball *Gabrielle Gilmer, Jessica Washington, Hillary Plummer, Gretchen Oliver*

Approximately 370 overuse injuries were reported in collegiate and high school fast-pitch softball players from 2004-2009 [1]. However, there are few data on softball pitching injury rates and their etiologies. Investigating pain history in softball pitchers could divulge information about injury-susceptible pitching mechanics. The purpose of this study was to compare pitching mechanics in collegiate softball pitchers with a history of upper extremity pain (UEP) to those with no history of UEP. We hypothesized that statistically significant kinematic differences would be found between pain history groups (stride knee flexion and valgus; pelvis rotation, anterior/ posterior tilt, and lateral flexion; trunk flexion, rotation, and lateral flexion; and elbow flexion). Twenty-nine collegiate softball pitchers volunteered for this study. Participants were divided into those with UEP (n=7; 19.7 \pm 1.3 years; 177.7 \pm 7.3 cm; 77.0 \pm 15.3 kg) and without UEP (n=22; 19.8 \pm 2.0 years; 172.3 \pm 8.4 cm; 80.0 \pm 10.1 kg). Participants were classified as UEP if they sought the medical care of their athletic trainer or team physician within six months prior to the study.

Kinematic data were collected with The MotionMonitorTM synchronized with an electromagnetic tracking system. Eleven electromagnetic sensors were attached to the following locations: (1) the trunk at T1, (2) pelvis at S1, (3-4) bilateral upper arm, (5) flat, broad portion of the acromion of the scapula, (6-7) bilateral forearm, (8-9) bilateral lower leg, and (10-11) bilateral upper leg [2].





Figure 1: Trunk rotation was plotted versus the throwing events (foot contact (FC), top of the backswing (TOB), and ball release (BR)). The healthy group displayed significantly higher trunk rotation at FC and BR when compared to the pain group. ** denotes significance.

Following sensor application, participants were given an unlimited time to perform their warm-up. The screwball was chosen for analysis because it was the most common pitch reported by those with UEP as noted in a health history questionnaire. Participants were instructed to pitch three screwball pitches at maximum effort for strikes over a regulation distance (43ft; 13.11m) to a catcher. Kinematic variables were averaged for the three executed trials at the pitching events of top of backswing (TOB), stride foot contact (FC), and ball release (BR).

Kinematic data were analyzed within IBM SPSS Statistics 23 software using an independent samples t-test with an alpha level of $p \le 0.05$. Pitchers with a history of UEP displayed greater trunk rotation at both FC (95% CIs = -32.70, 4.97, Mean difference=-13.87°; p=0.05), and BR, (95% CIs = -42.57, 3.44, Mean difference= -19.57°; p=0.01) (Fig. 1). The observed trunk rotation indicates that the UEP group's trunk lagged behind over the course of the throw.

The UEP group displayed a trunk position that was more sideways to the target. Specifically, a right-handed pitcher's trunk was more square to the third baseline, with the left shoulder pointing more to the target at FC and BR. This difference in trunk rotation might result in the pitchers having to throw across their body more than necessary, thus putting the shoulder and elbow in a more injury susceptible position. Though it is known that softball pitchers do not rotate square to the target until after ball release [3], the rotational differences in the present study should be further investigated. Positioning the shoulders perpendicular to the target versus shoulders square to the target at BR may contribute to greater upper extremity forces about shoulder and elbow. Future research should analyze larger groups of participants to better determine the relationship between trunk positioning and pitching mechanics.

Statement of Research Advisor:

Gabrielle assisted in data collection, data analysis, and writing of this segment of data regarding pain history and pitching mechanics in collegiate softball pitchers. —*Gretchen Oliver, School of Kinesiology*

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Comparison of a Handheld Glucometer and a Clinical Biochemical Analyzer to Measure Glucose in Porcine Blood Samples

Madison Gohlke, William Nunnelley, Terry Brandebourg

My research question involved developing a sample handling method that allows a human point-of-care glucometer (POCG) to accurately measure circulating blood glucose concentrations in porcine blood. Pigs are an apt biomedical research model for humans, specifically regarding the development of obesity and metabolic disease. As in humans, the accurate measurement of blood glucose concentrations is critical for assessing metabolic status as pigs become progressively obese. Human POCGs are an expedient method for measuring blood glucose in research animals and in clinical practice; the handheld instruments are portable, inexpensive, and yield rapid results. However, glucose values measured by POCGs often differ from values measured by a biochemical analyzer, the accepted clinical laboratory standard. These discrepancies can be large enough to cause potential misdiagnosis, limiting the ability to track the emergence of obesity-induced metabolic disease in our prediabetic animals.

POCGs measure glucose indirectly by utilizing a capillary strip to draw up a small blood sample and then recording the electrical current generated by the chemical conversion of glucose in the sample to ferrocyanide. I attempted to improve the accuracy of the glucometer by manipulating the viscosity and packed cell volume of blood samples, factors that could influence sample behavior on the capillary strip. Glucose was measured with a POCG in whole blood, serum, plasma, and fluorinated plasma (fluorine is a glycolytic inhibitor which

functions to stabilize glucose levels) collected from 152 pigs. These values were then compared to plasma glucose concentrations measured by a chemical bioanalyzer in samples obtained from the same animals.

As expected, glucose concentrations in whole blood as measured by POCG were highly variable and poorly correlated with plasma glucose concentration measured by the biochemical analyzer $(r_{2}=.34)$. However, glucose concentrations as measured by POCG in serum (r2=.57), plasma (r2=.64), and fluorinated plasma (r2=.89) were more strongly correlated with plasma glucose concentrations measured by the biochemical analyzer. Bland-Altman analysis revealed that mean differences in glucose concentrations determined by biochemical analyzer and by POCG in whole blood, serum, plasma, and fluorinated plasma were 43.5, 33.5, 12.4, and 6.9 mg/dl, respectively, mirroring the correlation analysis. These results suggest that measuring glucose with a POCG in the fluorinated plasma fraction of the blood yields a glucose value almost identical to that of a biochemical analyzer.

Finally, to validate this method under physiologically meaningful conditions, glucose values were measured in pigs that were fasted overnight to induce low blood glucose levels and again when pigs were fed to induce high blood glucose levels. Glucose values obtained by the POCG closely matched values obtained by a biochemical analyzer; differences in results fell well within a diagnostic window, and the instrument reliably identified fed and fasted animals. This indicates that a POCG may be used to accurately detect physiologically meaningful differences in porcine glucose values from fluorinated plasma samples.

This sample handling protocol improves and validates a rapid and reliable method for assessing metabolic status in pre-diabetic pigs as they become increasingly obese, allowing continued study on this research model.

Statement of Research Advisor:

Maddy discovered that significant discrepancies existed between blood glucose values obtained from pig blood depending upon the method she used to measure circulating concentrations. To address this issue, she conducted experiments to determine the effects of various sample handling methods on porcine blood glucose values obtained by a handheld point-of-care glucometer (POCG). She ultimately determined that the use of a human POCG to measure glucose concentration in fluorinated porcine plasma yields results that are most similar to those produced by a biochemical analyzer, a gold standard method in clinical settings. This supports her original hypothesis that utilizing the liquid, cell-free fraction of blood when determining porcine glucose concentrations with a POGC can increase accuracy and reliability of the glucose measurements to a degree that is comparable to methodologies utilized in clinical diagnostic laboratories. In doing so, Maddy has made an impressive and substantial contribution to the methodology the lab uses to measure a key metabolic marker.

— Terry Brandebourg, Animal Sciences

Controlling the Ion Distribution in Artificial Muscles for More Dynamic Actuation Response *Ethan Hofer, Patrick Bass, Jiachen Liu, Yancen Cai, Zhongyang Cheng*

Electroactive polymers (EAPs), also known as artificial muscles, are an exciting new branch of smart materials. Ionic EAPs, a subset of electroactive polymers, contain an electrically insulative polymer matrix as well as ions that can freely move through the polymer. When subjected to a voltage, ions move to different sides of the film depending on their charge, and the size difference of these ions expands one

side of the polymer, resulting in an overall bending or curling actuation. These EAPs, with their unique function and low operating voltage, are being studied for potential applications ranging from airplane wings to artificial organs. The goal of this project was to improve EAP actuation and control the ion concentration throughout the polymer for a more dynamic response.



Figure 1. Ionic EAP bending mechanism when a surface charge is applied

We created EAPs using a solution casting process of polyvinylidene fluoride (PVDF) copolymer doped with cobalt perchlorate. Based on thermal analysis and the optic properties of the polymer films, we found that two different phases often form during the casting process. Tests showed the second phase is detrimental to the actuation response, so we investigated modifications to the casting process to create films with a uniform structure. Methods such as increasing and decreasing the air flow during the casting process as well as dampening the mechanical vibration of the furnace in which the polymers were cast were examined. Additionally, post-casting surface treatments were done to determine their effects upon the performance of the EAPs.

While the second phase within the thin film was never completely eliminated, notable progress was achieved in decreasing its presence. The surface treatments did not aid in the EAP actuation as they would dissolve the cobalt perchlorate in the polymer, but the dampening of the mechanical vibrations improved the film quality considerably. The evaporation rate during the solution casting is one of the key factors to controlling the microstructure of the films. Because the solution is cast on glass slides, the evaporation rate is difficult to control, as the solvent will evaporate at different rates at different locations on the slides. Because of the two phases, a region's observed actuation did not accurately represent the concentration of ions in the region. This made it challenging to create an EAP with a designed actuation shape based on the local concentration of ions and the electromagnetic field. Consequently, further research will focus on forming a uniform PVDF thin film. Once this is accomplished, the effect of electromagnetic fields on the ion concentration throughout the film will be studied to create ionic EAPs with a dynamic bending actuation.

Statement of Research Advisor:

The EAPs studied by Mr. Hofer exhibit a new type of electromechanical response (i.e., actuation). There are many factors/issues to be solved/addressed to create engineering devices, or actuators, using the EAPs. In this project, Mr. Hofer solved one of the factors by designing experiments and identifying possible issues for future study.

-Zhongyang Cheng, Materials Engineering

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Identification of Phase I & Phase II Metabolites of Maca Constituents

Da Sol Jung, Thankhoe Rants'o, Angela Calderón

Maca (*Lepidium meyenii* Walpers) is a plant cultivated in central Peru, and its root is a common active ingredient in botanical dietary supplements for its effects on a variety of health issues such as sexual dysfunction, anemia, and cancer. The plant contains compounds that have cytotoxic and antioxidant activities, but information on the metabolism of the bioactive compounds is limited. The goal of this study was to identify Phase I and Phase II metabolites of the bioactive constituents of maca extracts, such as induction and inhibition of CYP3A4 and their interactions with anticancer drugs, for further study.

To predict the interactions of the maca compounds and their metabolites with anticancer drugs, Phase I and Phase II reactions of maca constituents were conducted, and the resulting metabolites were monitored and assessed for their increase in polarity and decrease in toxicity as well as possible inhibition of CYP3A4. Before performing the metabolic experiments, the permeability of the components was determined by incubating three different extracts in dichloromethane, methanol, and acidic methanol with buffer in a parallel artificial membrane permeability assay (PAMPA), which mimics the role of intestinal membrane. Then, the compounds were categorized into two groups: those that passively diffused across the membrane and those that did not. Both groups were incubated with human liver microsomes, phosphoric buffer at pH of 7.4, and nicotinamide adenine dinucleotide phosphate (NADPH) and/or uridine 5'-diphosphoglucuronic acid (UDPGA) to initiate Phase I and Phase II reactions and to obtain corresponding metabolites. Furthermore, for the Phase II experiment, alamethicin was added to the sample to activate UDPGA sequestered in the hepatocytes, and saccharolactone was added to inhibit glucuronidase, which reverses the Phase II reaction. The obtained metabolites of maca were analyzed with rapid resolution liquid chromatography, and their identity was confirmed via various layers by comparing obtained liquid chromatography (LC/MS) and mass

spectrometry (MS/MS) data to previously published literature or database and elucidating the structure of a compound with ACD/Spectra software.

The permeability test illustrated that among twenty-seven known chemical constituents of maca, six chemical constituents showed high permeability-these compounds have passively diffused through the membrane from donor site to acceptor site-while other twenty-one constituents displayed low permeability. The second part of the experiment suggested the presence of twenty-three metabolites of maca chemical constituents, and further analysis with MS/ MS, database, literature, and chemical software identified and confirmed eight metabolites produced via Phase I and Phase II reactions of maca extract. Among these eight metabolites, five metabolites were produced through hydrolysis and oxidation while three metabolites were produced through glucuronidation. While NMR analysis will provide further structural confirmation, a method to discern the compounds should be organized prior to the analysis, since botanical extract contains a wide range of compounds that are not necessarily bioactive or of interest. After finalizing the confirmation of produced metabolites, the samples will be tested for its possible inhibiting effect against CYP3A4 and will be incubated with and without anticancer drugs to observe the inhibition and interaction of the metabolites. The study provides a confirmed chemical profile of maca that may offer insight into active compounds that are known to be effective against cancer.

Statement of Research Advisor:

Da has developed an approach to assess the formation of Phase I and Phase II metabolites of intestinally permeable maca constituents and to elucidate the corresponding structure with mass spectrometry. The maca metabolites identified in this study have not been reported in the primary literature. The established approach will help to in the prediction of any potential maca-anticancer drug interactions.

—Angela I. Calderón, Pharmacy

Comparing the Floodplain Hydrology of a Recently Restored Stream Bank to a Natural, Undisturbed Floodplain

Benjamin King, Thorsten Knappenberger

The scientific community has been turning their focus to the watershed level when tackling sediment and pollution related issues. This paradigm shift has increased the number and size of the stream restoration projects that have been accomplished. However, there is a lack of data and inadequate attention given to the effectiveness of these restoration efforts.

In December 2015, an eroded reach of Parkerson Mill Creek was restored. The stream bank degradation only took place on one side of the floodplain while the other side remained functional. Hence, the stream restoration was directed at only the degraded side. This reach now offers the unique opportunity to study the hydrology of a recently restored floodplain compared to a natural reference floodplain within the same reach.

To assess the hydrology of the restored and natural sides of the floodplain, we developed a groundwater-sampling plan in which four groundwater wells, evenly spaced at intervals of 20 feet, were established along a perpendicular transect within each floodplain. Each groundwater well was equipped with a groundwater-level sensor. The goal was to understand how rainfall, discharge volumes, and groundwater levels are interconnected and how groundwater levels differ between a recently restored floodplain (left floodplain) and a natural, reference floodplain (right floodplain).

Results showed that groundwater moved more swiftly through the disturbed (restored) side of the reach due to a lack of established horizons within the soil profile. This result was predicted since this soil had been completely disrupted and had not been through the same weathering processes as the established, non-disturbed bank of the stream. However, we noted that the restored floodplain has established vegetation and has been stable despite multiple storm events that have filled the stream to overflowing. Future restoration efforts should be pursued well after the initial work has been completed to understand better the healing process that a floodplain undergoes after being completely disturbed. Although traditionally limited by budget, these follow-up studies are needed to gain a full understanding of what is truly accomplished by a restoration effort.

Statement of Research Advisor:

Ben has worked on this project very independently and he had to deal with situational factors that he could not influence but that affected his work. Yet he invested time and effort to mitigate unforeseeable issues, for example, how to dig wells to appropriate depths. I think he has had a great experience of planning, conducting, analyzing, reporting, and presenting field research. Through Ben's research, we have now a better understanding of the hydrology of restored stream reaches vs. reference conditions.

—Thorsten Knappenberger, Crop, Soils and Environmental Sciences

Functional Studies of Genes Transcriptionally Regulated by Calcium in Xylella fastidiosa *Courtney Kloske, Sy Traore, Prem Kandel, Leonardo De La Fuente*

Xylella fastidiosa (X. f.) is a bacterium that causes multiple lethal diseases in many economic crops including grapes, citrus, peach, plum, almond, coffee, pecan, and more recently olive. It was first described as Pierce's disease in the grapevines of California vineyards. *X. f.* lives only in xylem vessels, which carry water and nutrients from the roots to the rest of the plant. Proliferation inside the xylem leads to formation of a biofilm that obstructs the plant's vascular system, causing nutrient deprivation in the plant.

Previous research by our group showed that calcium increases the growth of the biofilm, and therefore, the virulence of the bacterium. Whole transcriptome analysis identified 17 *X*. *f*. genes that were consistently upregulated by calcium at different times, including 12 without an assigned function. One gene (PD0926) was selected and encodes for a hypothetical protein. The objective of this study was to determine the role of PD0926 in the virulence of *X*. *f*. To test its virulence, homologous recombination was used to cleanly delete the PD0926 and replace them with the chloramphenicol antibiotic resistance gene. Sequence verification was performed to verify successful knockout of the genes of interest in the *X*. *f*. genome. We successfully knocked out gene PD0926 in the WM11 and Temecula strains of *X*. *f*. These mutated strains will serve as a resource for performing further *in vivo* and *in vitro* testing. Microfluidic chambers will be used to test the biofilm formation of the mutated strain, and twitching motility will be observed. An *in planta* test will be performed to show the progression of the disease within a tobacco plant. These tests will help us understand the virulence of the mutated strain. With this knowledge, a more holistic view of the bacterium will help with potential eradication of this bacterium and its related diseases.

Statement of Research Advisor:

Courtney's work was important to create mutants in *X*. *f*. in order to understand the biology of this pathogen. We decided to venture into unknown territory with the target genes chosen. Despite the fact that we could not finish what we originally planned, Courtney's research was an example of the challenges of doing research and how to deal with non-anticipated hurdles in the process.

-Leonardo De La Fuente, Plant Pathology

The Time Course of DPP-IV, CD26+ T-Cells and IL-6 Following a DOMS Protocol in College-Aged Participants

Elise Mann, Leslie Neidert, Anna LaMantia, Heidi Kluess

Delayed onset muscle soreness (DOMS) can occur following exercise and results in damaged muscle fibers and the release of many chemicals such as potassium, histamines, and cytokines. A particular cytokine, interleukin 6 (IL-6), is known to increase in response to muscle injury and participates in the muscle recovery process. There is evidence that a serine protease, known as dipeptidyl peptidase IV (DPP-IV), cleaves IL-6, making it inactive. DPP-IV can be soluble in the blood and interstitial fluid, or bound to the T-cell membrane, making T-cells CD26+. The focus of our study was to observe the time course of soluble DPP-IV, CD26+ T-cells and IL-6 during recovery from DOMS.

Our study used six healthy college-aged participants who underwent a weightlifting protocol designed to induce DOMS in the bicep brachii. We first determined the participant's one-repetition maximum bicep curl. The participant then completed 15 sets of 15 repetitions at 65% of his/her one-repetition maximum. Measurements were taken before, immediately after, and 3, 24, and 48 hours following the DOMS protocol. Measurements included blood samples from the DOMS-treated limb to measure local plasma DPP-IV activity, CD26+ T-cells, and IL-6. Plasma DPP-IV activity was measured on the untreated arm via a finger capillary draw to determine if the effect was localized or systemic. Soreness measures were also taken to ensure that the participants experienced DOMS in the treated bicep. These included a soreness ranking using a visual analog scale and a pressure-sensing algometer at three different points along the treated bicep.

Increased pressure sensitivity indicated by decreased algometer pressure occurred at 24 hours (see Figure 1A; p < 0.05) and maximal soreness indicated by an increase in Visual Analog Scale measure was experienced at 24 and 48 hours (p < 0.05), suggesting that participants developed DOMS in the treated bicep. Plasma DPP-IV in the DOMS-treated arm significantly increased 9.9 \pm 5.7% at 24 hours and 9.3 \pm 9.6% at 48 hours (see Figure 1B) with no change in the non-treated arm. Plasma IL-6 significantly increased 166.1 \pm 124.3% immediately post protocol and 164.9 \pm 91.26% at three hours (see Figure 1C) before returning to baseline by 24 hours. There was no significant change measured in CD26+ activated T cells over time.

The results suggest that IL-6 peaks early after muscle injury, but plasma DPP-IV increases later in the DOMS process. This may imply that the increase in DPP-IV is for modulating the time course of IL-6. Overall, the increase in DPP-IV is localized to the site of injured muscle fibers because an increase in plasma DPP-IV was not observed in the control arm. We also did not see an increase in activation of CD26+ T-cells. This may be due to the small muscle mass utilized in this study or possibly the severity of the DOMS induced was not great enough to cause measurable activated T-cells. The relationship between DPP-IV and IL-6 is important for understanding the inflammatory response and recovery process after exercise-induced muscle damage. In the future, this lab plans to analyze the time course of blood flow in response to DOMS.



Figure 1: Mean and standard deviation for each time point for the algometer (A), plasma DPP-IV (B) and plasma IL-6 (C). * p<0.05 different from pre.

Statement of Research Advisor:

This project was a continuation of our lab's interest in the function and location of DPP-IV release. Elise was integrally involved in planning and executing this study. She obtained training to take blood samples and managed the entire project including recruitment, data collection and analysis. Leslie Neidert and Anna LaMantia assisted Elise in this project. —*Heidi Kluess, Kinesiology*

High Prevalence of *Anaplasma Platys* Infection in Alabama White-Tailed Deer

Seth Rankins, Sarah Zohdy, Stephen Ditchkoff, Bernhard Kaltenboeck, Dongya Gao, Shelby Zikeli

The objective of our study was to investigate the influence of morphology, age, and ectoparasite loads on the susceptibility of white-tailed deer (*Odocoileus virginianus*) to *Anaplasma spp. Anaplasma spp.* are gram negative bacteria in the order Rickettsiales and are the causative agent of anaplasmosis. These bacteria are spread by ticks and lice and infect erythrocytes. There are multiple species of *Anaplasma* that infect a wide range of mammals, including companion animals, domestic livestock, wild ruminants, small mammals, and even humans. As white-tailed deer serve as a reservoir host for *Anaplasma spp.* across the South, we were interested in identifying which deer are likely to serve as carriers, allowing us to manage deer populations in a manner that would help protect ourselves and our animals from anaplasmosis.

This study was conducted at the Auburn University Deer Research Facility in Camp Hill, Alabama. The vegetation within the facility, which is roughly 40% open area maintained for hay production and 60% forest, is representative of the landscape encountered throughout the South. Male deer in the facility are chemically immobilized each year to maintain a marked population and to collect data for numerous ongoing projects. For our study, we took blood samples from 29 deer that were captured in the fall of 2016. Body and antler measurements, as well as full body (excluding the legs) ectoparasite counts were taken for the same individuals. The blood samples were screened for *Anaplasma spp.* with a 16S rRNA gene FRET-qPCR.



Figure 1: *Anaplasma platys* infection level in white-tailed deer, determined by the number of copies of the target gene in 0.1 mL of genomic DNA extract, was positively correlated with tick loads (P=0.038).

Twenty-four of the 29 deer screened were positive for Anaplasma platys (no other species of Anaplasma were detected). Body weight of male deer fluctuates drastically throughout the year; therefore, we used a principle components analysis to create a continuous variable for body size based upon chest girth, total body length, and hind foot. We then ran a regression analysis to examine the effect of deer body size on infection level of A. platys, and we found that there was no effect (P = 0.644). Additionally, a regression indicated that antler size did not influence the infection level of deer (P = 0.840), and a Fisher exact test indicated that age did not influence infection level (P > 0.05). However, we did find that infection levels increased with increasing tick loads (P = 0.038) (Figure 1). Our results suggest that morphology and age do not influence which deer serve as reservoir hosts for anaplasmosis, but the presence of a vector is vital for the

life cycle of this bacteria. It is surprising that *A. platys* is the species found in the deer, since it has never been documented in white-tailed deer and canines serve as the primary reservoir host. Yet considering the prevalence of free roaming dogs and the ubiquitous nature of deer and ticks, it is not unreasonable to think that *A. platys* is widespread in deer across the South. It is imperative to conduct further research to better understand how *A. platys* exploits deer, allowing us to prevent livestock and human exposure.

Statement of Research Advisor:

This research has considerably expanded our knowledge concerning the role of white-tailed deer as a reservoir for *Anaplasma spp.*, and the finding that *Anaplasma platys* was the primary strain detected was extremely novel. —*Stephen Ditchkoff, Forestry and Wildlife Sciences*

Finding Formulas for the Number of Pieces Resulting from k (n-1)-Dimensional Linear Cuts on a Unit Ball in Euclidean n-Space

Elizabeth Scholss, Michel Smith

Our research question examined the maximum number of pieces into which a convex region can be cut using k number of (n-1)-dimensional linear cuts. This problem is related to many other mathematical fields, like topology and geometry, and it also has connections to the medical field. A CT scan uses combinations of many X-ray images at different angles to produce cross-sectional images, which is exactly what this research problem examined in the different convex regions. Specifically, we studied the regions by analysing our drawings and formulating proofs.

We were able to find some of the formulas for our problem for k-dimensional convex figures. By using the summation formulas: $\sum(_i=1^{n}(n)) i^{k}$, we have found explicit formulas up to i=6 for g_i (n), where g_i (n) is the number of regions into which each figure is cut and i is the dimension number. We have also found a recursive formula for i-dimensions: We have also worked on 2-dimensional convex shapes with convex regions removed from it. This problem can be translated into more dimensions with more regions removed. We have found a recursive formula for this region if one straight cut can pass through all the holes, assuming the holes are in a straight line:

$$f_i(n) = (n-1) + f_{i-1}(n),$$

where $f_i(n)$ is the number of regions into which the shape is cut, *i* is the number of holes inside of the shape, and *n* is the number of straight cuts. We have also found that if these holes are not in a straight line, then the number of pieces into which the region can be cut is the same as the largest number of holes that are in a straight line.

$$g_i(n) = g_i(n-1) + g_{i-1}(n-1).$$

This research has connections to the medical field and could help doctors better understand the mathematical principles behind the machines they use. Further research could investigate the formula for finding the number of holes into which higher dimensional regions can be cut. We can also continue research directed at finding a general, non-recursive formula for the number of pieces into which each region can be cut.

Statement of Research Advisor:

For the first result of our research, we used an inductive technique to obtain the formulas. Although the

solution was known to me, a different technique was used by Elizabeth. The problem was then generalized to a convex body with a convex hole or a set of holes of different configurations. Elizabeth used different techniques to obtain some new partial solutions to this generalization. Additional properties of the counting functions were derived. For example, the leading coefficient was calculated exactly without induction; this was also new to both researchers.

—Michel Smith, Mathematics

Multi-Bend Antenna Optimization by Genetic Algorithms

James Smith, Michael Baginski

The goal of our research was to design antennas that broadcast electromagnetic energy in very specific, predefined regions. Signal strength topology may appear to be nearly pixilated and is typically impossible to realize using commonly available antennas. Highly directional antennas are very beneficial in tracking radars where maximum "power on target" is crucial for telecommunication systems trying to maximize coverage. Telecommunication systems and specifically cellular towers trying to focus the signal power in populated areas and eliminate coverage in unpopulated areas (city reservoirs, landfills, etc.) will greatly benefit from these antenna designs.

The optimization technique used in this research is called a genetic algorithm (GA). This technique mimics the biological processes of evolution, molding and shaping a "population" of wires over several generations to perform optimally in a specific task. Objects are optimized on their "fitness," or how well the object performs compares to the ideal object. Two key advantages of a genetic algorithm are that it can search an infinitely large number of possible solutions and it can avoid local minima and maxima by introducing mutations.

The predecessor for this work was the "crooked wire antenna," which can be imagined as a straight piece of

wire arbitrarily bent many times to achieve a specific antenna power pattern [1]. The location and degree of the bends were determined by a GA that selects the antenna designs that best creates the desired pattern. Our research extends the "crooked wire antenna" design to achieve even better results using a type of "branching antenna," where the antenna wire configuration resembles the branch structure of a tree. A specially designed GA determines the length and placement of each of the wire branches. The key difference in this work from the "crooked wire antenna" is that each wire is allowed to branch out in arbitrary directions, similar to a tree.

To test our hypothesis, we ran trials of our developed genetic algorithm comparing branching versus non-branching antenna results for several test cases. Table 1 shows averaged fitness results of such a test case, using 10 trials per combination of branches per node and depth of antenna. The total number of wires for each antenna is a result of its branches per node and depth, as shown in Figure 1. This is presented in the results to show that branching wire antennas are not out-performing non-branching wire antennas by consisting of more total wires. Our most important finding was that the averaged fitness, a single value that represents how close the antenna is to the "ideal antenna"

(with the more positive value being the better antenna), is much higher for antennas allowed to branch. Figure 2 shows the radiation patterns for the antenna giving the highest fitness in this experiment.

Future research will apply this technique to a practical problem, likely including further restraints. We plan to show that the multi-bend antenna can be used for cost-saving in industry.

Statement of Research Advisor:

This research is significant because, unlike its predecessor, all the conceptual designs are realizable using antenna branching that reduces the load bearing of the individual antenna member. These findings will be transformative to any field needing unique antenna radiation.

-Michael Baginski, Electrical and Computer Engineering



Table 1: GA Experiment Results



Figure 1: Antenna structure



Figure 2: Ideal and actual radiation patterns for branching antenna with 2 branches per node and a depth of 5 (31 total wires)

Teaching Spatial Visualization in Chemistry *Natalie Stephens, Christine Schnittka*

Spatial visualization refers to one's ability to mentally visualize, rotate, and transform objects. It is an important skill that helps with problem solving, with understanding and applying math and science concepts, and with engineering design. While the necessity for spatial skills is known, it is still not clear how to best infuse spatial thinking into primary education.

Applying spatial skills to the study of chemical molecular geometries makes it possible to identify and visualize molecular shapes to determine their chemical properties. The goal of this research was to determine if spatial recognition software on a smartphone could help high school chemistry students develop stronger spatial skills when applied to chemical molecular geometry. The software used was a free phone app, Aurasma. This app recognizes a designated picture, called a trigger image, and displays a 3D augmented reality model created by the designer. These 3D images can be designed and accessed by any user.

For this research, 3D images of 14 chemical molecular geometries were designed. Students in four chemistry classes at Southern High School (a pseudonym) were given access to traditional ball and stick models to learn the structures of the 14 molecules. The students had to identify the molecular geometries of these various chemical compounds. Then, two classes, one regular and one advanced, were given access to trigger images and the Aurasma app to visualize the 3D structures. The students using the app checked their guess of the molecular geometry and used the app to review the material.

All the students were given pre- and post- tests of their ability to identify the molecular geometries of chemical compounds. These evaluations were quantitatively analysed to understand if the students with the added technology had an increase in their 3D chemical geometry skills over the group that just used traditional ball-and-stick models. Surveys were administered to determine whether participants used the app to review the material at home, and if they felt that the app helped them learn the material better than traditional physical models.

The advanced class that used the app had a statistically significant increase in their test scores (p=0.01) and they reported in their surveys that they believed the app helped them advance their understanding of molecular geometry. The regular class had an increase in their test scores as well (p=0.12) but it was not as significant as the advanced chemistry class. Even though the advanced chemistry class had a greater increase in their test scores than the regular class, they did not report using the app more often. The advanced students' willingness to learn and their higher level of fundamental understanding of molecular geometry allowed them to significantly improve their scores through the help of the Aurasma app.

Further research could examine if there is transfer of general spatial thinking skills from the advancement of the students' chemical molecular geometry understanding. While this study gained some understanding about the benefits of utilizing the Aurasma app for molecular geometry, there are still many research opportunities to learn about spatial thinking and chemistry.

Statement of Research Advisor:

Natalie's work is innovative, and has already attracted the attention of other teachers who want to help us investigate its effectiveness in different classrooms. Natalie designed and implemented the 3D images used in the study. Her work is the first of its kind, and we anticipate publishing in both practitioner and research journals, so that science teachers and education researchers can learn about the work and build upon it.

-Christine Schnittka, Education

Airfoil Analysis Using Smoothed Particle Hydrodynamics

Daniel Stubbs, Stephen Nichols

Our project sought to develop a method for enabling smoothed particle hydrodynamics (SPH) to simulate fluid flow over a body using an inflow-outflow boundary condition that allows fluid to enter and to exit the computational domain.

With this boundary condition, inflow-outflow-type simulations can benefit from the advantages SPH has over traditional computational fluid dynamics (CFD). For example, SPH is a particle-based method which, unlike traditional CFD, requires no grid mesh to be defined. Not requiring a mesh allows SPH to provide time savings and to handle simulations such as flow over complex shapes, mixing problems, or multiphase flow with relative ease. By giving SPH the ability to model the flow in and out of a domain accurately, SPH would naturally extend to a wider range of applications and would aid engineers as an efficient tool providing valuable insight into the intricate details of aerodynamic flows. Such insight would assist engineers in developing more efficient and capable designs.

This project began with an existing SPH simulation code already capable of simulating flow within a closed domain. The code was then extended by developing methods allowing for fluid particles to flow through the domain, leaving through an outlet and re-entering through an inlet. As part of this inflow-outflow condition, methods were developed for resetting the particle properties upon exiting and re-entering the domain as well as for providing buffer regions at the outlet and inlet to ensure that the simulation would remain stable. Once these capabilities had been added to the simulation code, test cases were run on fundamental fluid dynamics problems such as flow over cylinders and airfoils. The results of these simulations were compared against widely available and accepted verification data.

Results from this project show that the inflow-outflow condition is working well, but that there are still some underlying issues within the core equations used in the simulation, particularly regarding boundary conditions and data extraction from solid boundaries. Figure 1 shows the aerodynamic flow around a two-dimensional airfoil shape, which exemplifies the current capabilities of the simulation. The next steps



Figure 1: SPH simulation of a NACA0012 airfoil in an aerodynamic flow.

for this project are to investigate the equations being used, to determine methods to improve the model's accuracy, and to remove some of the shortcomings of the simulation in its current state. Future projects will seek to make the code capable of 3-D combustion reaction simulations as well as giving the code the ability to run on modern high-performance computing technologies, such as multi-node supercomputers and GPU clusters.

Having developed a working method for allowing fluid to flow in and out of the computational domain in an SPH simulation, this project has laid the ground work and provided valuable lessons for future efforts with SPH. With this foundation in place, SPH is on course to be fully capable of simulating complex fluid flow over any geometry, simple or complex, allowing insight into the fluid dynamics around shapes that challenge existing technology.

Statement of Research Advisor:

Daniel's research project focused on extending SPH from its typical regime of incompressible fluids in closed domains to compressible flows with open boundaries. During his research, Daniel designed a stable, buffered inflow-outflow boundary condition, developed a methodology to improve the placement and use of solid boundary particles, and studied multiple combinations of the Lagrangian forms of the compressible conservation equations.

-Stephen Nichols, Aerospace Engineering

Role of Carotenoids as Antioxidants in the Marine Copepod, *Tigriopus californicus*

Philip Wang, Ryan Weaver, Geoffrey Hill

The food, pharmaceutical, and cosmetic industries are directing increasing attention to compounds with antioxidant properties. This interest arises from an antioxidant's capability to quench reactive oxygen species (ROS) such as H₂O₂ and O₂⁻, which play a key role in aging and pathological conditions [1]. ROS attack bio-membranes and cause damage to lipids, proteins, and DNA [2]. Antioxidants prevent oxidative damage by reducing ROS and preventing the radical chain reactions that would otherwise lead to damage of cellular components. Animals internally synthesize many antioxidants, such as superoxide dismutase and glutathione, but can also obtain antioxidants from their diet.

Carotenoids are organic pigments synthesized by plants that are the source of red, orange, and yellow colors in many plants and animals. Carotenoids have antioxidant properties and can quench ROS by dispersing the radical's energy into a solvent, as shown in many in *vitro* studies [3]. Increased dietary intake of carotenoids has been correlated with a variety of physiological benefits, such as increased immune function and increased pathogen resistance across multiple species [4]. Many scientists propose that the physiological benefits of increased carotenoid consumption arise from their antioxidant properties. However, evidence for carotenoids' in *vivo* antioxidant activity is contentious.

We used the marine copepod, *Tigriopus californicus*, to test the antioxidant properties of carotenoids in animal systems. In the wild, *T. californicus* obtains carotenoids by consuming algae. In the lab, *T. californicus* can be raised on a yeast diet to completely remove carotenoids from its system. Carotenoids can be reintroduced into the copepods by supplementing their yeast diet with powdered carotenoid. We fed 600 yeast-raised copepods either yeast or yeast with carotenoid supplement for 48 hours. We then placed half of them in a 100 μ M tert-butyl hydroperoxide (tBHP) solution to stimulate the production of ROS. After 24 hours of exposure, we counted copepods to assess mortality (Fig. 1).

The mortality data are promising and show that carotenoid-fed copepods had a lower mortality than carotenoid-deficient copepods in both the control and pro-oxidant exposure groups. However, there was no different



Figure 1: Mortality of carotenoid-deficient and carotenoid-supplemented copepods for both the pro-oxidant exposure and control treatments shown with 95% upper and lower confidence levels.

in mortality between the control groups and the pro-oxidant groups, a result that suggests our concentration of pro-oxidant had no effect on the copepods' mortality. We will repeat the experiment with a higher concentration of tBHP for a shorter amount of time to hopefully induce more oxidative damage in the exposure groups. We will also measure the amount of malondialdehyde using high-performance lipid chromatography to quantify the level of lipid peroxidation in each group.

Statement of Research Advisor:

Philip worked as an independent investigator on the study of the role of carotenoids in protection from oxidative damage in copepods. He designed the study, confronted the numerous problems that arose in the execution of the research, and is now taking a leading role in the analysis and interpretation of data. —Geoffrey Hill, Biological Sciences

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