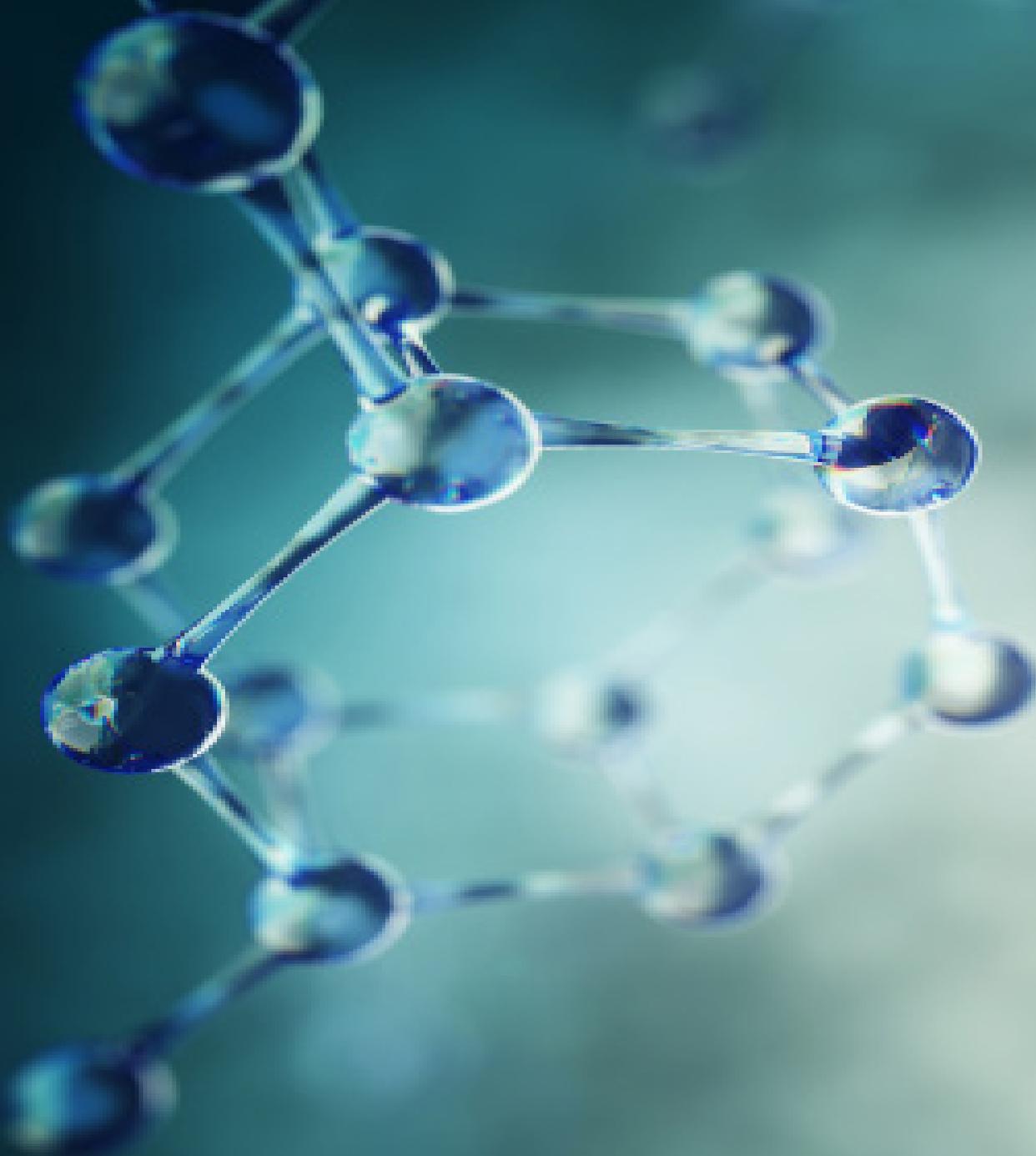




AUJUS

Auburn University Journal of Undergraduate Scholarship

2018 Edition Volume 7



AUJUS is a collaborative effort by students at Auburn University

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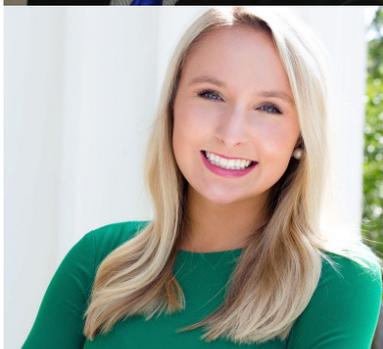
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AUJUS Editorial Team



Associate Editor: Kareem El-Kattan is a senior pursuing a degree in Chemical Engineering and has served as the editor for AUJUS since Spring of 2018. Kareem has been involved in research since the fall of his freshman year. His research, in the Department of Chemical Engineering under Dr. Allan David, focuses on use of iron-oxide nanoparticles as a vessel to test for enzymes present in prostate cancer cells. Kareem is a Student Recruiter, a Cupola Engineering Ambassador, and volunteers at EAMC hospital on the weekends. In his free time he enjoys backpacking, cooking, and cheering on the Auburn Tigers.



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 higher education communications arena.



Associate Editor: Ben Nelson is a senior from Opp, Alabama, and will graduate with a Bachelor of Science in biomedical sciences. While at Auburn, Nelson served the College of Sciences and Mathematics (COSAM) as Peer Advisor, Organismal and Introductory Biology laboratory teaching assistant, and undergraduate research ambassador. He conducted research in the immunology laboratory of associate professor Elizabeth Schwartz, by whom he was directed in his studies of the effects of inflammation on intestinal bacteria. Upon graduation from Auburn, he will further his education at the University of Alabama at Birmingham School of Medicine.



Associate Editor: Phillip Wang is a senior with 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. Virginia A. Davis



"I love that you never know where it [research] will take you," says Dr. Virginia A. Davis, Alumni Professor in Chemical Engineering. Davis began her research journey at a science fair project in the 8th grade. Since then, Davis has mentored more than 50 students with a focus on the fundamental science governing how large materials can be assembled from tiny, nanoscale, building blocks.

"I love that you never know where it [research] will take you"

Davis says she loves research, because it continually stretches her to learn new things and is always rewarding. "Every researcher knows the thrill of finally figuring something out or getting something to work. It is even better when that goes one step further and you see how other people build on the knowledge you helped establish."

Davis says she is supportive of all students who are considering the undergraduate research route and encourages them to get involved. "Go for it! Read as much as you can and try to figure out what excites you, then start talking to potential mentors," Davis says. "Your first research assignment is about learning how to do research, those skills will enable you to explore other topics later."

"Go for it! Read as much as you can and try to figure out what excites you, then start talking to potential mentors."

Davis says her main goal as a mentor is to build her mentees' confidence and research skills. "I want to help facilitate my mentees to figure out what they want to do after they leave Auburn and during their career," Davis says. "I also want to help them have the skills and credentials needed to achieve their goals."

Davis claims the most rewarding aspect of being a mentor is staying connected with her previous mentees. "The biggest reward is hearing from and meeting with previous mentees, and learning about their career pathways and successes," Davis says. "I have mentees who are now working as lawyers, researchers, professors, and leaders in the industry. It is always amazing to meet with a mentee and hear where their journey has taken them."

Mortality and injury rates of wildlife reported by rehabilitators across Alabama

Steven Williamson, Christopher Lepczyk

Abstract

Injured wildlife species are often brought to wildlife rehabilitation centers, where they are assessed, cared for, and eventually released. Injuries are due to many causes, such as anthropogenic sources, predation, disease, and malnourishment. However, little is known about rehabilitation rates (the number of animals brought in or released every year) of these animals. Alabama just recently began requiring rehabilitators to keep records, whereas other states have been doing it for several years. Our objectives were to determine the most common reasons for admitting wildlife species to wildlife rehabilitation facilities in Alabama and to quantify the most commonly brought in species during the first year of statewide records collection. We compiled intake records from 19 rehabilitators during 2014 by contacting district offices of the Alabama Division of Wildlife and Freshwater Fisheries. Records included intake date, species, presenting issue, county of origin, disposition, county of release, release date, and location of release. A total of 834 animals were taken in by rehabilitators in 2014. Nearly 87% of the animals recorded ($n = 726$) were orphaned, injured, or both, with the majority (66%) of the animals being admitted in one wildlife district. The most common animals taken in were squirrels (*Sciurus carolinensis* and *Glaucomys Volans*; $n = 306$) and opossums (*Didelphis virginiana*; $n = 217$). The majority of animals were brought in during August ($n = 158$) and September ($n = 138$), representing approximately 35% of the annual total. Almost 57% of the animals taken in were released after rehabilitation ($n = 474$). The intake records from most wildlife rehabilitators simply listed “orphan” or “injured” as the reason for intake. Identifying the causes for wildlife entering rehabilitation facilities could aid wildlife education programs and better understanding incidences of human-wildlife conflict.

Introduction

Wildlife rehabilitation is the treatment and temporary care of injured, diseased, and displaced indigenous animals, and the subsequent release of healthy animals to appropriate habitats in the wild (Mullineaux, 2014).

However, local governments often outlaw the rehabilitation of non-native species, such as wild pigs (*Sus scrofa*) in Alabama. The benefit to wildlife populations of the practice of rehabilitation has been debated in recent years (Wimberger et al., 2010). Many people believe that wildlife rehabilitation has no effect on most wildlife because relatively a few animals are released into large populations (Molina-Lopez et al., 2013). However, others believe there are several benefits from wildlife rehabilitation and release, including an improvement of an animal's welfare, a reinforcement of the natural population, an identification of causes of the animal's morbidity and mortality, an ability to provide data for passage of new laws, and the generation of positive attitudes towards wildlife by the public (Molina-Lopez et al., 2013). For species that are threatened or endangered, rehabilitation can aid in maintaining the genetic pool (Molina-Lopez et al., 2013). Furthermore, identifying causes of injury and death in wildlife could lead to early identification of disease outbreaks that threaten wildlife, livestock, or people. Finally, laws to limit the impact humans have on wildlife and the ecosystem have been developed by information from wildlife rehabilitation (Molina-Lopez et al., 2013).

Although wildlife rehabilitation can provide valuable information about wildlife, relatively little information exists. The few existing studies to date note low to moderate release rates (<50%; Molina-Lopez et al., 2015; Mullineaux, 2014; Grogan and Kelly, 2013). However, the best indicator of animal survival during rehabilitation is the severity of the illness or injury (Molina-Lopez et al., 2015). Thus, not all animals entering a rehabilitation center necessarily survive. Furthermore, juvenile animals typically have higher release rates, which may show a lack of injury in orphaned animals (Molina-Lopez et al., 2015).

In Alabama, 2014 was the first year that wildlife rehabilitators were required to submit intake records to the state Department of Conservation and Natural Resources. This requirement afforded a unique opportunity to evaluate rehabilitation records from the onset of the program. In assessing the first year of the program in

Alabama, we had two primary objectives: (1) to determine the most common reasons for wildlife species to be admitted to rehabilitation facilities; and (2) to quantify the most common wildlife species admitted to the rehabilitation facilities.

Methods

Within Alabama, five Wildlife District offices manage wildlife rehabilitation licenses for the counties within their district (Figure 1). We contacted the supervising wildlife biologist for each wildlife district and obtained copies of all the intake records submitted within that district. Only four of the five districts had data for 2014; District 5 had no rehabilitators submit for new licenses or license renewal. Records only include mammal and herpetological species because records from the Alabama Wildlife Center and the Southeastern Raptor Rehabilitation Center, which are the only rehabilitation centers in Alabama that admit and rehabilitate avian species, were not available. Once we received the records, we compiled the data following the same record-keeping guidelines as intake records found on the Alabama Department of Conservation and Natural Resources website. These records required rehabilitators to note intake date, species accepted, presenting issue, country of origin, disposition, county of release, date of release, and GPS coordinates or street address of release site. A few records included information from 2013 and 2015, but they were excluded from analysis. We then classified the data into additional categories, including the district where a rehabilitator was located, a unique identification number for each rehabilitator, and the month of intake. We also edited the data to correct misspellings of species names and to group animals that were not identified to species level into broader species classes (e.g., squirrels, reptiles). Our numbers do not include records that were unreadable due to poor scanning or illegible handwriting. We quantified the number of animals admitted per month, number of animals by class admitted per month, number of animals with certain disposition admitted by month, number of animals admitted by district, and number of animals admitted by presenting issue. We analyzed the data by calculating basic summary statistics for the various intake information, as we only had a single year of data and were not testing *a priori* predictions amongst any groups.

Results

In 2014, a total of 834 animals were admitted to rehabilitation facilities across Alabama, of which 24 were missing intake date and thus were not included in the total number of animals taken in by month (Table 1). Of these 834 animals, 37% (n = 306) were squirrels and 26% (n = 217) were opossum (*Didelphis virginiana*; Table 1). The classification of squirrel included both Eastern Gray Squirrel (*Sciurus carolinensis*) and Southern Flying Squirrel (*Glaucomys volans*). There were several other classifications, but they accounted for less than 9% in each case. Of the animals admitted, 87% (n = 726) were labeled with a presenting issue of orphaned (74%) or injured (13%). The other 13% of presenting issues only labeled the animals with an age range, such as adult or juvenile, or as brought in due encounter with domestic animals or some type of disease. Among the four districts that had rehabilitators submit intake records, 66% (n = 549) were from District 4 (Figure 1). The peak of admittance was August (n = 158) and September (n = 138; Figure 2). Of all the possible final dispositions, 57% (n = 474) of the total number of animals admitted were released after spending time at a rehabilitation facility.

Discussion

The greatest number of animals admitted by class were squirrels and opossums. These two classes of animals have high fecundity, with opossums having an average litter size of seven to eight, but the potential for as many as 13. Squirrels average only two to three young, but in good years, females can have two litters. Peak of breeding season in Alabama is in January to February and then again in June to July for both species of squirrels (Burde and Fieldhamer, 2005). With the peak of breeding seasons within those months, young are ready to leave their parental care around May for the first litter of squirrels and then again in August or September for the second litter of squirrels and the litter of opossums. These life-history attributes, along with the proximity of both species to humans, are likely factors for why they comprise such a large number of individuals admitted to wildlife rehabilitation centers. Other animals with high reproductive rates that lived in close proximity to humans were admitted, but they numbered far fewer than opossum and squirrels. These animals that live in close proximity to humans are synanthropes and often gain something beneficial from living in an altered environment such as in urban

and suburban areas (Marchesini, 2016).

The large percentage of animals admitted in District 4 was likely due to a greater number of rehabilitators in this district, compared to the other districts, and the presence of one of the larger mammal rehabilitation centers, the Big Bend Wildlife Rehabilitation Center. Recent legislation allows each district only one rabies-vector rehabilitation center, and Big Bend is the designated center for District 4. However, determining why there were more rehabilitation centers in District 4 was beyond the scope of this study.

Our results differ from previous research results in terms of release rates of wildlife species. Most previous research had release rates < 50%, whereas we found a statewide release rate of 57% (Molina-Lopez et al., 2015). Alabama's release rate could be due to the large number of animals that were labeled and admitted as orphans. People not educated on the life histories of many wildlife species can misidentify an animal's disposition. Some may believe an animal is orphaned or appears too young to be on its own and take it to a wildlife rehabilitator, when such action is not needed. For example, white-tailed deer (*Odocoileus virginianus*) fawns are often found without their mothers, as the fawn will remain in one area, staying hidden while the doe roams to forage. The doe then returns throughout the day to allow the fawn to nurse until it is strong enough to venture out with the doe and escape predators if found (Burde and Feldhamer, 2005). There are a number of reasons for why wildlife species are brought into wildlife rehabilitations centers; however, examining these reasons was beyond the scope of the data available. To alleviate the problems of human-wildlife encounters that result in injury or removing a healthy juvenile from parental care, a campaign to educate the public would help decrease future admittance of injured wildlife or juveniles that are not truly orphaned. Future research can develop yearly trends and identify broad-scale patterns in the state. One aspect that would be beneficial for future research and the state's wildlife professionals is better record keeping. For this study, some records were not considered because of illegible penmanship. Entering records electronically would help alleviate this problem. Furthermore, on-line record keeping would allow for a real-time update that could show which species are being admitted over time and by location.

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Table 1. Total number of animals admitted each month per class to wildlife rehabilitation centers in Alabama in 2014. Class includes both species and groups of species.

Class	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total
Amphibian	0	0	1	0	0	0	0	0	0	0	0	0	1
Armadillo	0	0	0	1	1	0	0	0	0	0	0	0	2
Bat	0	0	1	0	3	3	4	1	1	0	0	1	14
Beaver	0	0	0	1	0	0	0	0	0	0	0	0	1
Bobcat	0	0	0	0	1	0	1	0	0	0	0	0	2
Chipmunk	0	0	1	3	0	0	0	0	1	10	2	2	19
Coyote	0	0	0	0	1	0	0	1	0	0	0	0	2
Deer	0	0	0	0	0	0	17	21	23	9	1	1	72
Fox	0	0	0	3	8	2	0	0	0	0	0	0	13
Possum	0	0	45	54	42	21	11	41	2	1	0	0	217
Rabbit	0	1	3	13	14	8	11	3	4	2	1	0	60
Raccoon	0	0	1	1	9	14	13	6	15	5	6	4	74
Reptile	0	1	0	3	10	1	0	2	1	1	1	0	20
Rodent	0	0	7	0	0	0	1	0	8	1	0	0	17
Skunk	0	0	0	0	0	10	0	1	0	0	0	0	11
Squirrel	2	4	7	3	8	9	26	82	83	31	17	13	285
Total	2	6	66	82	97	68	84	158	138	60	28	21	810



Figure 1. Alabama state map with color-coded wildlife districts.

¹ A total of 834 animals were recorded as being admitted to wildlife rehabilitation centers in Alabama during 2014, but 24 of these animals were recorded without an intake date thus were not included in this table.

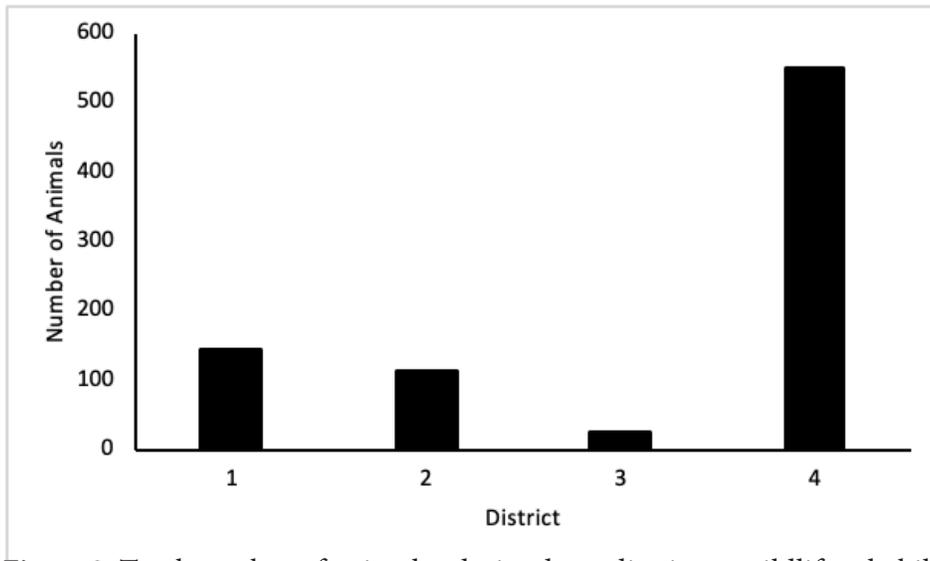


Figure 2. Total number of animals admitted per district to wildlife rehabilitation centers in Alabama 2014.

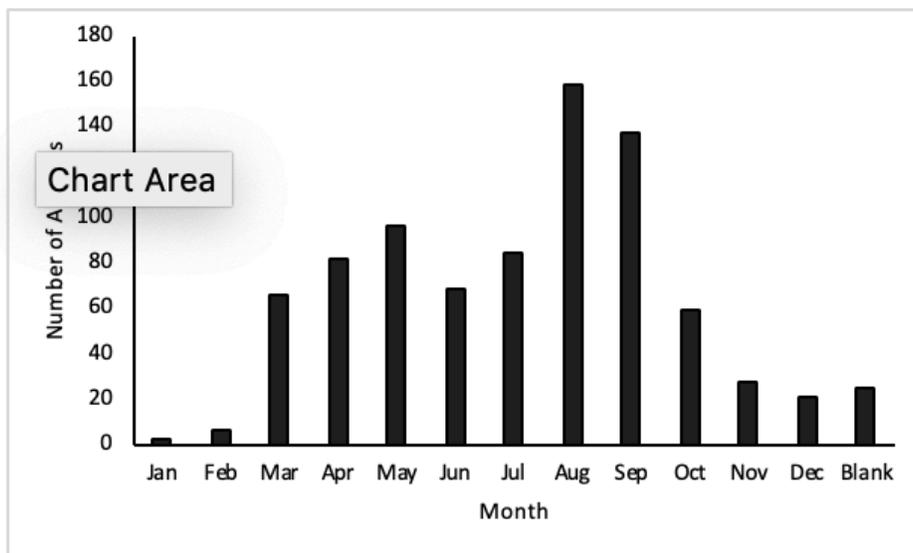


Figure 3. Total number of animals admitted per month to wildlife rehabilitation centers in Alabama in 2014.

Female spirituality in *Marguerite's Hours*

Jordan Wade, Karen Sonik

Abstract

Marguerite's Hours, a Book of Hours originating in France c. 1318-1325, was produced by the Church of St. Omer and contains texts and prayers in Latin and French as well as illuminations by artists of the Franco-Flemish school. It also contains a large amount of marginalia, which depict an unusual number of pregnant women. My research examines the depictions of women in the margins of *Marguerite's Hours* and asks the question: was the frequent portrayal of pregnant women in the margins created with female spirituality in mind? And if so, how would this be identified by the book's medieval female owner? These questions are significant for two reasons: (1) because the purpose of marginalia generally is still unclear, and identifying the function of the marginalia in this book could lead to a greater understanding of marginalia as a whole, and (2) this research provides a deeper understanding of gender roles within medieval Christianity, specifically as pertains to the perception of women. Drawing on the research of medievalists Caroline Walker Bynum and Michael Camille, as well as my own analyses of *Marguerite's Hours*, I argue that, in order to relate to its female patron, the marginal depictions of women in *Marguerite's Hours* deliberately present women literally as (potential or actual) sources of food, mirroring the way Christ offers his body as food to his followers; display the female body in a state of physical change, thus likening the pregnant body to the crucified body of Christ; and downplay the notion of women as opprobrious figures.

Marguerite's Hours, a Book of Hours produced c. 1318-1325 by the Church of St. Omer in France, is remarkable for the numerous images of pregnant women in its margins. Measuring 155 x 105 mm, consisting of 355 vellum pages, and bound in brown calf skin with leather tooling, this book of Latin and French texts and Franco-Flemish illuminations served as a devotional prayer guide for its medieval owner. But the manuscript's repeated portrayals of pregnant women in its margins, interspersed with crowded depictions of fish wearing hats, rear ends poking out of ovens, and decapitated heads atop running legs, are not immediately identifiable as religious, let alone

devotional, even though that is exactly what they are.¹ These women in the margins are identifiable as both Marguerite, the book's patroness, and Saint Margaret, the patron saint of childbirth, and they serve a distinctly religious function by alluding to the societal roles of medieval women (childbirth and nurture), thus applying the manuscript's Christian texts and illuminations to specifically female life experiences. I argue that the marginal depictions of women in *Marguerite's Hours* express medieval female piety by presenting woman as a potential and literal source of food, mirroring the way Christ offers his body as food to his followers; by displaying the female body in a state of physical change, thus likening the changing pregnant body to the crucified body of Christ; and by downplaying the notion of woman as an opprobrious figure.

Marginal images were frequently included in Books of Hours. Like other marginalia, the marginal ladies in *Marguerite's Hours* were based on conventional image types found in illuminations; they could have been created by illuminators, scribes, or both.² And, although scholars have yet to determine which of the multiple theories concerning the function of marginalia are correct, it has been established that their physical location in the margins does not mean they act outside the sacred realm or are separate from the larger functioning and meaning of the main text and illuminations. Since marginalia often includes whimsical, worldly, and sometimes even sexual or grotesque imagery, it may seem odd to us that they adorn the insides of religious texts such as bibles and Books of Hours. But in the Middle Ages, it was not at all uncommon for traditionally secular images and ideas to be used in the discussion or elucidation of religious subjects.³ The image of Saint Margaret on the same page as a bottom sticking out of an oven⁴ would not necessarily have distracted the medieval reader from the prayers, passages, and other religious materials contained in the book. Nor would it sully the image of Saint Margaret herself or in any way eradicate her religious importance. The boundaries between the

¹ Camille 1992: 18

² Camille 1992: 22

³ Camille 1992: 18

⁴ Morgan Library Online Archive: *Marguerite's Hours*, L6

religious and secular worlds at this period were fluid; worldly subjects were used to discuss religion because to the medieval mind, religion was not a separate sphere; religious ideas encompassed and permeated all aspects of life. Marginalia provided scribes and illuminators the opportunity to represent this fluidity visually; marginal images interacted both with and against the rigid structures in manuscript texts. They could variously represent subversions of socio-cultural norms, antitheses to the texts, or gentle parody of the manuscript's contents.⁵

If much of the rest of the marginal imagery in Marguerite's Hours functioned as soft parody of the main text and illuminations, the depictions of Saint Margaret and the patroness, Marguerite, served a distinct purpose. They are physically and visually separated from the other marginalia through their positioning on a raised structure or branch, which grants them a compositional distance from most of the turbulent marginal mayhem — despite their existence in direct relationship to it. Usually depicted on the right side of the page, supported by leaves⁶ or standing alone inside a small rectangular structure topped with small towers and a lavishly detailed arch,⁷ the marginal ladies are given a seat of privilege and clarity amongst fantastically tangled images of animals, detached human body parts, vines, and hybrid creatures. In addition to their physical separation from the figures crowding the margins, the women are granted more negative space and are depicted in slightly larger scale than the other marginal images, as if indicating their importance to the patroness and ensuring that they are not overlooked.⁸ The women are seen in various poses, sometimes standing and sometimes kneeling, but they are almost always facing the illumination or text at the center of the page. They typically have their hands clasped in prayer or hold a book (presumably a Bible) in their outstretched arms, as if ignoring their fantastical surroundings in order to focus solely on the rigid structure provided in the Biblical text and imagery. In some cases, their gaze might also act as the aesthetic factor tying the marginalia and the main texts and illuminations together.

⁵ Camille 1992:21

⁶ Morgan Library Online Archive: Marguerite's Hours, L1

⁷ Morgan Library Online Archive: Marguerite's Hours, L138

⁸ The Morgan Library <http://ica.themorgan.org/manuscript/thumbs/128495>. Occasionally, both St. Margaret and Marguerite are depicted together amongst the throng of whimsical illustrations with one woman on either side of the main text. On most pages, only one woman is shown. Since the women's dress changes style and color throughout the pages of the book, and since both are shown in various stages of pregnancy, scholars have not differentiated one woman from the other. They have only made inferences about the identities being presented.

A key and consistent aspect of the marginal women in Marguerite's Hours is their portrayal in various states of pregnancy.⁹ The portrayals are not chronological; rather, they might depict a still slender woman on one page and a hugely pregnant one on the next. These dramatic shifts in appearance would likely catch the patroness' eye and bring to mind thoughts of her own potential and past pregnancies, as well as her societal and religious roles and the religious connotations of these pregnancies. In medieval Christianity, men and women were thought to have been given distinctly different roles by God; the man was seen as possessor of God's knowledge and was created to lead, and woman, possessor of his flesh, was made to follow.¹⁰ Because of these distinctions, it was not at all uncommon during the Middle Ages for religious imagery to be geared specifically toward men or women. And because women were possessors of God's flesh, medieval female piety thus utilized sources that were familiar to women or that pertained to what was understood as the woman's sphere — such as food, childbirth, and nurture — in order to enforce the connection between Christ, God's flesh and the provider for all of humanity, and medieval woman, who used her literal flesh to provide for her children.¹¹

The depictions of the pregnant patroness in Marguerite's Hours also cater specifically to female spirituality in their presentation of women as both potential and literal sources of food. The deliberate and heavily emphasized marginal depictions of Saint Margaret and of the pregnant patroness, Marguerite, suggest that the latter will be or already is offering her body as food to another human being. Medieval women often viewed metaphorically the physical changes the pregnant body undergoes; the changing body was compared to the changes Christ's body underwent during crucifixion, and breastfeeding was linked to the concept of Christ's body nourishing his followers.¹² In her book, *Holy Feast and Holy Fast: The Religious Significance of Food to the Medieval Women*, Caroline Walker Bynum emphasizes the role of women as symbols of literal food in women's spirituality, noting that food imagery was much more significant to female religious

⁹ Morgan Library Online Archive: Marguerite's Hours, L200, L126

¹⁰ Bell 1982:743. Medieval women played active roles in society, were oftentimes literate, and sometimes acted as patrons in their own right.

¹¹ Bynum 1987:261

practices than male. For a period after childbirth, women produce breastmilk, a human's first source of nourishment; because of this, women were seen as the ultimate providers. By selflessly providing nourishment from their bodies, women acted in the manner of Christ, who offered his body to his disciples as food, saying "take, eat; this is my body."¹³ While this imagery and thought process would not resonate much with medieval men, it served to link the female owner of the manuscript to the Lord.

The marginal depictions of pregnant women in Marguerite's Hours also speak directly to female piety through the display of the female body in a state of physical change. The marginal presentations of the patroness and Saint Margaret in various states of pregnancy from one page to the next emphasize the Christ-like transformation the female body undergoes and draws associations, again, to breastmilk and other bodily fluids. During the crucifixion, Christ's body is described as undergoing dramatic physical changes.¹⁴ In Isaiah 53:5, Christ is discussed as "*pierced* through for our transgressions, He was crushed for our iniquities; the chastening for our well-being fell upon him, and by His *scourging* we are healed [my italics]."¹⁵ The gruesome wounds inflicted upon Christ were necessary, because his body had to undergo painful changes in order to purge Christians of their sins and thus grant them eternal life. Like any body, the body of Christ, after he had been killed, continued to bleed and secreted waste. This gruesome and human death had to take place in order to save humanity and grant life and was therefore perceived as an essential part of the miracle of Christ's resurrection. Since the female body exudes breastmilk, menstrual blood, and afterbirth fluids, it was "seen as powerful in its holy or miraculous exuding," and was also in this way related with the literal becoming of the crucified body.¹⁶ Through the visual indication of woman as food and the changing body of crucified Christ, the marginalia in Marguerite's Hours serve to relate more personally the manuscript's contents to its patroness and to her fulfillment of both the societal and religious roles of women.

The final indicator that the marginalia in Marguerite's hours were created to promote and perhaps also to

reflect proper female piety is the lack of opprobrious portrayals of women. Because medieval men and women had different religious roles (men were seen as being God's knowledge or spirit and women were his flesh), women were depicted differently in religious imagery depending on whether it was geared toward male or female audiences. In an attempt to discourage sinful actions and retain man's spiritual purity, specifically male piety often used images of women in ways that emphasized feminine weakness, temptation, and corruption, which is why medieval imagery frequently demonized women or depicted them with "moral ambivalence."¹⁷ Derogatory and even disfigured depictions of women were deemed necessary, because the accurately portrayed female face was seen as an object of sin and temptation.¹⁸ Medieval men were also much more apt to view male and female piety dichotomously,¹⁹ and images designed for a male audience might frequently "criticize particular women and...differentiate sharply between...male and female characteristics."²⁰ Marginal images are known to be exaggerated, absurd, and full of commentary. The absence of negative portrayals of women in Marguerite's Hours alone is evidence of the manuscript's specific position in the realm of female piety.

Medieval women, while they sometimes internalized notions of feminine sinfulness and described themselves as "weak" and "sinful," also had positive models and conceptualizations of feminine functioning and behavior. The facial and bodily representations of the women in the marginalia of Marguerite's Hours do not indicate that women are lesser, shameful, or evil. Nor are they grotesque or compared (disadvantageously) to men. They instead, through indications of pregnancy, use the notion of woman as flesh to promote women as positive and productive members of society and link them to Christ. Marguerite and Saint Margaret are portrayed not only without opprobrium but in a positive light, in a manner referencing the actual Marguerite's likely societal roles. Amongst the marginal portrayals of fantastical humans, beasts, and hybrids, the numerous pregnant women in Marguerite's Hours serve as a compositional and ideological link between the seemingly worldly marginalia and the rigidly religious texts and illuminations. And, although the concrete meaning of

¹² Bynum 1987:270

¹³ Matthew 26:26 KJV

¹⁴ Morgan Library Online Archive: Marguerite's Hours, L200, L126

¹⁵ Isaiah 53:5 KJV

¹⁶ Bynum 1987:274

¹⁷ Sauerländer 2006:6

¹⁸ Sauerländer 2006:6

¹⁹ Bynum 1987:264

²⁰ Bynum 1987:263

marginalia as a whole remains unknown, it is clear that these specific marginal depictions of the female body as food and in a state of physical change emphasize the medieval understanding of pregnancy as a time when women were physically in tune with the Lord, thus appealing to and promoting female piety.

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Determining the effects of reproductive performance on bone quality in female house mice

Kayla Frey, Wendy Hood

Abstract

Much of calcium and phosphorus used to support skeletal ossification during prenatal and postnatal mammalian development are derived from the mobilization of maternal bone minerals. As a result, bone loss has been observed in the maternal skeleton during the reproductive cycle. In order to maintain their skeletal integrity, ensure survival, and continue future reproduction, mothers must be able to prevent excessive bone loss. The goal of our research was to evaluate how the number of reproductive bouts and the total number of young produced impacts bone quality in female mice. We evaluated this relationship by quantifying variation in femur morphology and mineral composition of 14 age-matched mice that had the opportunity to breed from reproductive maturity at 2 months until they were 10 months of age. To evaluate femur morphology, we sent one set of femurs to the University of Alabama at Birmingham to quantify the 3D structure of the bones. We quantified the total mineral and calcium content of the bone by inductively coupled plasma spectrophotometry. No significant relationship was established between bone morphology and reproductive performance or between bone mass and mineral content for females that had between zero to seven litters and that vary between weaning 0-44 offspring. These results suggest that female house mice are adept at maintaining bone mineral content and mass throughout their reproductive cycle.

Introduction

To reproduce successfully, female mammals must be able to provide the necessary nutrients for their offspring's physiological development. Developing vertebrae in offspring requires calcium and phosphorus for successful growth to occur, and most mammalian offspring are dependent on their mother for these mineral resources until they reach weaning (Hood & Hobeck, 2015). Evidence from previous studies suggests that increased intestinal calcium absorption during pregnancy provides much of the calcium required by offspring during reproductive development, but that maternal bone loss may still occur in the last months of pregnancy, when rapid mineralization of the

offspring's skeleton occurs (Kalkwarf & Specker, 2002). It is important to evaluate the tradeoffs associated with reproduction and the mineralization of offspring skeleton to determine the extent to which allocation of nutrients to offspring affects mammalian mothers.

Increasing demand for skeletal mobilization leads to increased bone turnover and reduction in bone mineral and strength. Previous studies suggest that bone resorption is more accelerated during the second half of pregnancy, corresponding with an increased fetal demand for mineral during this time (Avendaño-Badillo et al., 2009). This can lead to substantial bone loss during reproduction. In one study, rats lost 28% of their bone mineral content on a 0.4% calcium diet, and up to 53% of bone mineral on a 0.1% calcium diet (Peng et al., 1988). This study concluded that bone loss during reproduction in these rats was not only accentuated by a calcium-deficient diet, but also aggravated by an increase in suckling pups during lactation (Peng et al., 1988).

Bone loss as a cost of reproduction can lead to decreased skeletal integrity in the maternal mammal and negatively affect future reproductive success. It has been shown that reduction in bone mineral density is correlated with reduction in bone strength (Reilly & Burstein, 1975; Marshall et al., 1996). This means that bone loss as a cost of reproduction corresponds with increased risk of fracture. Bone fracture in wild animals can lead to an inability to acquire food and an increased susceptibility to predators, decreasing their chances of survival. In mammalian females, bone fracture leads to a decreased chance of future successful reproductive bouts. Therefore, availability of maternal skeletal mineral can be viewed as a limit for successful reproduction. Additionally, reproductive bone loss can be viewed as a cost of reproduction because there is a trade-off between maternal skeletal integrity and allocation of mineral to offspring.

For this study, we analyzed bone loss during reproduction in house mice (*Mus musculus*). Due to their

proportionally smaller skeletons (Prange et al., 1979), it has been suggested that less maternal skeletal calcium is available for house mice than for larger animals. This likely corresponds to an increased limitation on bone mineral mobilization (Hood, 2012). For this reason, we expect that the mice will experience statistically significant bone loss during their reproductive cycle. In essence, we predict that increased reproductive output in house mice leads to a corresponding decrease in bone quality.

Materials and Methods

We obtained the femurs and vertebrae of 14 age-matched mice postmortem that had the opportunity to breed from reproductive maturity at 2 months until they were 10 months of age. All mice were maintained under semi-natural conditions, and thus their skeletons were assumed to have experienced a level of strain comparable to wild mice.

The right femurs were manually cleaned of residual connective tissue after being soaked in an ultrasonic bath for 30 minutes to loosen the tissue. A Soxhlet apparatus was used to extract neutral lipids with petroleum ether and ethanol until the samples reached a constant mass (12 h). The femurs were then ashed in a muffle furnace (Fisher Scientific Isotemp Muffle Furnace, Dubuque, IA, USA) at 550°C for 24 h. The ash content, or total mineral content, of the samples was determined using the change in mass after ashing. The samples were then digested in nitric acid for one hour and diluted in nanopure water for mineral analysis. All concentrations were determined by mass. The calcium and phosphorus content of the samples were determined via inductively coupled plasma optical emission spectrophotometry (Perkin Elmer Optima 7300DV; Waltham, MA, USA) using the following wavelengths: Ca²⁺ - 317.933 nm and P - 213.617 nm.

The left femurs were sent to the University of Alabama at Birmingham (UAB). There, the 3D structure of the bones was quantified, including cortical thickness, trabeculae thickness, and trabeculae density. UAB used microcomputed tomography to obtain these values. We then used the obtained values in regression analysis to determine if any significant relationships could be established.

Results

There were no significant relationships between bone mass and total number of litters per female aged up to 1 year, average litter size at weaning, or total number of pups throughout life (Table 1). No significant relationships were established between total mineral content and total number of litters per female aged up to 1 year, average litter size at weaning, or total number of pups throughout life (Table 1). No significant relationships could be established between percent calcium and total number of litters per female aged up to 1 year, average litter size at weaning, or total number of pups throughout life (Table 1). Additionally, there were no significant relationships between percent phosphorus and total number of litters per female aged up to 1 year, average litter size at weaning, or total number of pups throughout life (Table 1).

We then tested our variables against Ca/P ratios to see if there was any evidence of a significant relationship. However, we could not show any significant relationships between Ca/P ratio in bone and total number of litters per female aged up to 1 year, average litter size at weaning, or total number of pups throughout life (Table 1). Overall, we found no evidence of significant relationships between bone mass and parity or mineral content of bone for females that had between zero to seven litters, and that vary between weaning zero to 44 offspring.

No significant relationships were established between femur trabecular number and total number of litters per female aged up to 1 year, average litter size at weaning, or total number of pups throughout life (Table 2). There was no significant relationship between femur trabecular spacing and total number of litters per female aged up to 1 year, average litter size at weaning, or total number of pups throughout life (Table 2). There was no evidence of significant relationships between vertebral trabecular number and total number of litters per female aged up to 1 year, average litter size at weaning, or total number of pups throughout life (Table 2). Further, there were no significant relationships established between vertebral trabecular spacing and total number of litters per female aged up to 1 year, average litter size at weaning, or total number of pups throughout life (Table 2). Note that despite the presence of visible variability in the morphological structure of the bones in Figure 1,

Table 1. Comparison of maternal characteristics and femoral bone morphology. Results of linear regression are given.

Maternal Characteristic	Bone Characteristic	F*	df*	P*	Interpretation
Total number of litters	Bone mass	0.26	12	0.6184	Not significant
Average litter size at weaning	Bone mass	0.43	12	0.5266	Not significant
Total number of pups throughout life	Bone mass	0.77	12	0.3994	Not significant
Total number of litters	Total mineral content	0.27	12	0.6139	Not significant
Average litter size at weaning	Total mineral content	0.42	12	0.5369	Not significant
Total number of pups throughout life	Total mineral content	0.74	12	0.4066	Not significant
Total number of litters	Ca %	0.03	12	0.8668	Not significant
Average litter size at weaning	Ca %	1.86	12	0.2000	Not significant
Total number of pups throughout life	Ca %	0.33	12	0.5780	Not significant
Total number of litters	P %	0.26	12	0.6182	Not significant
Average litter size at weaning	P %	0.43	12	0.5269	Not significant
Total number of pups throughout life	P %	0.77	12	0.3994	Not significant
Total number of litters	Ca / P ratio in bone	0.00	12	0.9917	Not significant
Average litter size at weaning	Ca / P ratio in bone	0.10	12	0.7602	Not significant
Total number of pups throughout life	Ca / P ratio in bone	0.02	12	0.8886	Not significant

* F= F value, df= degrees of freedom, P= P value

regression analysis using vertebral trabecular number and spacing, and femur trabecular number and spacing determined that this morphological variability was not statistically significant. Overall, no significant relationships were established between bone morphology or mineral content of bone for females that had between zero to seven litters, and that vary between weaning zero to 44 offspring.

Discussion

We predicted that increased reproductive output would correspond to a decrease in bone mineral. Results of this experiment failed to support this prediction. There was no significant difference between the mineral content and bone morphology of mothers with zero to 44 offspring and zero to 7 litters. These results suggest that female house mice are adept at maintaining bone mineral content and mass throughout their reproductive cycle.

These results contradict our hypothesis that a small mammal with a much smaller skeleton would experience bone loss with an increase in reproductive output if bone mobilization occurred. In a previous study on

female white-footed mice (*Peromyscus leucopus*), there was an observed net bone loss that indicated female white-footed mice trade off skeletal condition for reproduction (Schmidt & Hood, 2014). A separate study on female mice suggested that when faced with increasing litter sizes, the amount of mineral mobilized from the maternal reserves increased correspondingly (Hood, 2012). The results of these studies suggest bone loss on a significant scale during reproduction. However, it is possible that the laboratory conditions of the aforementioned studies affected the experimental results if diminished strain on the skeleton limited the need to maintain bone density.

In a study conducted by Hood & Hobensack (2015), it was shown that when female mice were given the opportunity to climb, and thus experience greater strain on their skeleton, they did not experience a drop in bone mass or mineral content during reproduction. Despite some predicted recovery after weaning, we anticipated a cumulative loss of bone in the mice in our study because they endured more reproductive cycles than those in Hood and Hobensack's study (1 bout per females). Our study did not yield results that support

Table 2. Comparison of maternal characteristics and femoral bone morphology. Results of linear regression are given.

Maternal Characteristic	Bone Characteristic	F*	df*	P*	Interpretation
Total number of litters	Femur trabecular number	0.56	9	0.4770	Not significant
Average litter size at weaning	Femur trabecular number	0.43	9	0.5266	Not significant
Total number of pups throughout life	Femur trabecular number	0.91	9	0.3684	Not significant
Total number of litters	Femur trabecular spacing	0.51	9	0.4947	Not significant
Average litter size at weaning	Femur trabecular spacing	0.89	9	0.3738	Not significant
Total number of pups throughout life	Femur trabecular spacing	0.83	9	0.3899	Not significant
Total number of litters	Vertebral trabecular number	0.95	9	0.3581	Not significant
Average litter size at weaning	Vertebral trabecular number	0.72	9	0.4196	Not significant
Total number of pups throughout life	Vertebral trabecular number	2.03	9	0.1921	Not significant
Total number of litters	Vertebral trabecular spacing	0.98	9	0.3505	Not significant
Average litter size at weaning	Vertebral trabecular spacing	0.95	9	0.3574	Not significant
Total number of pups throughout life	Vertebral trabecular spacing	2.29	9	0.1687	Not significant

* F= F value, df= degrees of freedom, P= P value

this hypothesis. This is likely because under natural conditions, mice experience sufficient strain to stimulate full compensation for the bone loss experienced during the reproduction. Because the mice in this study were kept in semi-natural conditions, the stress and strain experienced by the animals would have been greater than mice kept in a small rodent box. Therefore, the results of this study are predicted to represent the responses of mice in the wild.

Overall, our results suggest that wild-derived house mice in semi-natural conditions experience no loss of female skeleton integrity after reproduction, suggesting the skeleton experiences no cost of reproduction. It is important to note that although no significant relationships were established in this study indicating bone loss throughout the reproductive cycle, visible variability in bone morphology did occur (Figure 1). If bone loss does occur, skeletal maintenance may be compensating for the increase in bone mobilization during reproduction. If these mice can maintain their skeletal integrity during reproduction or quickly recover their bone quality postpartum, they decrease

the risk of bone fracture and susceptibility to predators. This further increases their chances of future reproductive success. By studying wild-derived animals, our results have broader application than experiments conducted under laboratory settings. Future studies should evaluate whether focus on skeletal self-maintenance of the maternal mice had impacts on offspring bone quality. Future studies should also evaluate which variables affect reproductive bone loss and to what extent. These variables may include exercise, dietary mineral intake, and presence of disease. Replications of this experiment should emphasize utilization of a larger sample size.

Acknowledgments

I would like to thank Maria Johnson in the Animal Phenotyping lab at University of Alabama at Birmingham for completing the microcomputed tomography measurements, Dr. Paul Cobine for his assistance with the ICP spectrophotometer, and Auburn University for funding this project through my Undergraduate Research Fellowship.

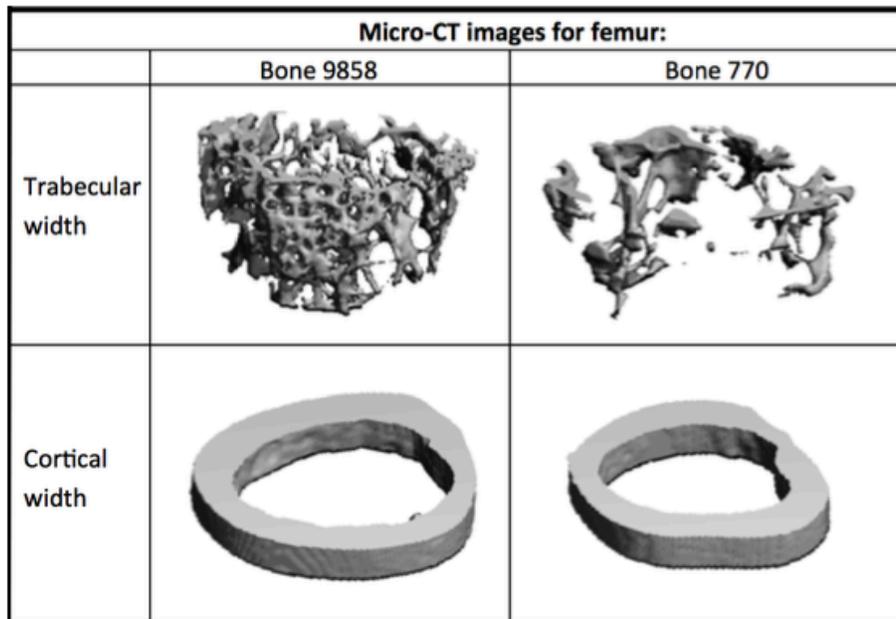


Figure 1. Microcomputed tomography imaging of trabecular and cortical femoral bone.

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The part and the whole: The multiple perspectives of Tara Donovan's *Untitled (Styrofoam Cups)*

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Abstract

From a distance, Tara Donovan's Untitled (Styrofoam Cups) alludes to natural phenomena, yet nearness reveals the Styrofoam material. The artist metamorphizes manufactured cups into a rolling landscape through her own assembly-line process – mimicking the containers' creation in factories. However, in critics' analyses of Donovan's sculptures, they focus on the whole of her creations' formal beauty and advocate that the transformative effect of her assemblages removes the relationship between her mass-produced subunits and their associations with over-consumption. Her methods often produce large amounts of unrecyclable refuse, which further indicate her, like others', complicit participation in consumer culture. Donovan uses a mundane object to create a sculpture fraught with contradictions in order to condemn the environmental damage caused by the very material she relies on. To spread awareness of the ecological criticism contained by Donovan's sculptures, this paper argues that Untitled (Styrofoam Cups) offers a critique of the proliferation of disposable goods through its own dependency on mass-produced products and the process of manufacturing.

As museum visitors walk to the Birmingham Museum of Art's Terrace Café, their attention moves upwards towards a billowing cloud of white forms clinging to the split-level ceiling. Tara Donovan's undulating sculpture *Untitled (Styrofoam Cups)* (2004) hangs permanently from the roof of the café and casts a crisp reflection against the neighboring windows (Figure 1). Upon close inspection, the form reveals its construction of Styrofoam cups. Donovan (b. 1969, New York City) constructed the site-determined installation from 25,000 cups with help from two assistants and thirty volunteers. The work capitalizes on the light-defusing translucency of foam as well as the visual effect of thousands of cups accumulated together. The cellular-like structure and varying opacities of the amassed containers invite comparison to organic shapes ranging from clouds to coral reefs. Donovan's sculptures entice from afar

with their ethereal beauty and then challenge viewers' perceptual limitations by revealing their mundane subunits through proximity; her works play with the multiplicity of perception in recognizing the whole and the part simultaneously and separately.

In critics' analyses of Donovan's sculptures, they focus on her creations' formal beauty and advocate that the transformative effect of her assemblages removes the relationship between her mass-produced subunits and their associations with over-consumption. The artist metamorphizes manufactured cups into a rolling landscape through her own assembly-line process – mimicking the containers' creation in factories. Her methods often produce large amounts of unrecyclable refuse, which further indicate her, like others', complicit participation in consumer culture. Simultaneously, *Untitled (Styrofoam Cups)* formally alludes to natural phenomena from a distance. To spread awareness of the ecological criticism contained by Donovan's sculptures, this paper argues that *Untitled (Styrofoam Cups)* offers a critique of the proliferation of disposable goods through its own dependency on mass-produced products and the process of manufacturing.

The artist relies on the shifts in perception and recognition resulting from viewing her works from different distances and angles. When seen from afar, the individual components of Donovan's sculptures conglomerate into a single, atmospheric mass that disguises the identity of the material.¹ For example, *Haze* (2003) first appears as a soft, undulating wall of white fluff. Intrigued viewers move closer to discern the individual subunits. At close proximity, the whole breaks down into the parts; the identity of the stacked plastic drinking straws reveals itself. The point of recognition of the component depends on scale and volume of the work, but more importantly, the interaction of the viewer to move around and towards the piece. However, critics so far have interpreted Donovan's work only from the distanced view, focusing on the visual transcendence of the work's subunits.

Many art critics focus on the cumulative visual effect of the whole of Donovan's sculptures, merely appreciating their surface beauty. They write that she simply offers a formal appreciation of abundance and unfamiliar forms. In a review of the artist's first solo exhibition in 1998, Ferdinand Protzman set the precedent for a formalist evaluation of Donovan's works, relating her sculptures to landscapes.² Later reviewers mimic Protzman's appreciation of the ethereal beauty of the distanced view of the sculptures and even claim that the accumulations visually obliterate the subunits to eliminate the connotations of the material. In 2006, Helen Winston compares Donovan's forms to natural landscapes, such as overhanging cliffs and snow-covered hills, an effect magnified by distance from the sculptures. While Winston admits that the artist's works carry visual references to these environmental elements, the critic advocates that form and process, rather than metaphorical connotations or allusions to landscapes and household materials, drive the artist's work.³ Protzman and Winston began the focus on the visual elements of the Donovan's sculptures; their writings created a precedent for later writers to deny the symbolism within her sculptures.

In exhibition catalogues on Donovan's works, curators expand the formal analyses first proposed by critics to argue against the presence of ecological connotations in the artist's subunits. Kristina Olson, like Winston, rejects iconographic readings of the artist's materials in favor of a formalist analysis and examination of the process of assembly clearly displayed by the works. For Olson, Donovan's installations capitalize on visual clarity and order; the formal qualities of the whole of her sculptures offer a comforting moment of peaceful visual transcendence rather than a commentary on consumerism originating from the individual material.⁴ Three years later, curator Lillian Davies explored the argument put forth by Olson. Davies advocates that, even though the artist uses the systems and products of mass manufacturing, the beauty of her work glorifies rather than condemns the abundance of disposable goods.⁵ Olson and Davies both recognize the presence of references to the environment in Donovan's work, yet both argue that the whole of her sculptures negate the message carried by the artist's choice of materials. In 2013, curators Oliver Kornhoff and Poul Erik Tøjner even state, "Donovan draws the suggestive power of her art solely from the physical and aesthetic properties of the items she uses" in an

outright rejection of the symbolism of her materials.⁶

Donovan composed *Untitled (Styrofoam Cups)* from one of the most ubiquitous materials of a throw-away society obsessed with to-go drinks; even so, writings on the installation focus on process and form. Critic Christopher Miles, responding to the work's first showing in 2003, remarked on the technical feat of the sculpture's production.⁷ When the Birmingham Museum of Art (BMA) unveiled their commission, curator Susan Sipple Elliott additionally focused on the logistics and numbers of the work's creation.⁸ These readings avoid both the visual relationship of the sculpture to natural forms such as clouds or waves and the innate environmental connotations carried by Styrofoam cups. In favor of attention to the logistics of production, both Miles and Elliott avoid examining the iconography and conceptual connotations of the whole and the part of the BMA installation.

Donovan's own tight-lipped treatment of her intentions allows for numerous interpretations of her works beyond a focus on the process and visual. Yet, the range of critiques remain limited. Interviews with the artist focus on biographical events in her life as she rose to fame and her methods of making rather than her own intentions for her works.⁹ In 2007, Donovan noted that she leaves her pieces untitled in order to allow audiences to bring their own associations, and thus their own meanings, to the work.¹⁰ However, a comparison of titles found in catalogue listings and exhibition reviews reveals that she has progressively changed her works' titles to mask her artistic intent. In 1997, the artist first displayed her most well-known work, a square yard of compressed toothpicks called *Controlled Caging*.¹¹ In 2005, the title morphed to *Toothpick Cube* and was listed in 2009 as simply *Toothpicks*.¹² Since, the work has most often been identified as *Untitled (Toothpicks)* and *Untitled*.¹³ The original title, *Controlled Caging*, suggests the artist's role in shaping the cube and other iconographical implications, whereas *Toothpicks* and *Untitled (Toothpicks)* places the focus on the materiality of the medium. Her progressive redaction of information in the title prevents wall labels from leading viewers to a specific reading of the work.

Untitled (Styrofoam Cups) visually mirrors a wide array of natural phenomena. Living structures such as

cells, like Donovan's subunits, carry encoded rules for predictable growth that dictate the ways in which they interact, according to Kornhoff and Tøjner.¹⁴ The circular forms of *Untitled (Styrofoam Cups)* reference the replication of cells and clouds.¹⁵ Viewers can detect the entire form's similarity to the landscapes and biological structures, both of which are intimately affected by ecological devastation. The work relies on a product and a process that contribute to an ecologically devastating disposable culture to create an object resembling natural phenomena; as a result, the sculpture subtly presents a critique of contemporary environmental attitudes.

When viewers closely examine the installation, the identity of the Styrofoam cup reveals itself; as a result, viewers can recall the material's ecological impact and discern the work's allusions to environmental concerns. While Jonathan T. D. Neil writes that Donovan's choices of mundane materials lack cultural specificity, like branded Coca-Cola cans,¹⁶ the accessibility of her materials builds on viewers' intimacy with every-day products. Toothpicks, Styrofoam cups, and buttons are materials virtually every viewer knows. Artists originally used Styrofoam for molds and models, but within the last half-century, they have turned to the light and cheap material as a primary medium for sculptures. While the plastic product acts as an energy-saving insulator, more than one-hundred cities have banned its use due to its lack of biodegradability and expense to reclaim.¹⁷ Donovan's use of the synthetic material does not deny but rather points to her, like others', reliance on disposable goods.¹⁸

The very process that formed *Untitled (Styrofoam Cups)* recreates the original production of the cups themselves. Mimicking the assembly-line fabrication of the Styrofoam, the artist and her team re-manufacture the goods into art through the same system of the cups' creation. The allusion to natural phenomena of Donovan's work masks the environmental horror of the throwaway materials she uses. The otherworldly glow of the sculpture mirrors the way in which the sheen and glamour of disposable goods, such as automobiles, cell phones, and computers, disguise the technologies' ecological impact. Donovan's return to the process that created the cups doubles the work's connection to manufacturing and the environmental impact of mass production of disposable goods.

Donovan's methods typically result in many of her materials being discarded as trash, despite her works' critique of the very practice. *Untitled (Toothpicks)* continuously drops toothpicks on to the exhibition space's floor, causing the crisp cube to slump over time. The compression used to form the cube stresses the fibers of the toothpicks; the material can only withstand the process once.¹⁹ When light-sensitive foam of *Untitled (Styrofoam Cups)* grew brittle, the BMA installed a new configuration in 2011. The sculpture aged poorly in the fluctuating temperatures and direct sunlight of the museum café, so assistants recreated the work. The glue attaching the cups' rims becomes yellow and brittle over time. So, seven years after the first installation, assistants replaced the original with a new version in a different configuration of entirely new cups.²⁰ Donovan clearly takes no issue with recreating her works with new materials. She constructs sculptures that will require recreation in future decades, further increasing the work's entwinement with consumer culture.

When diners first approach the café space at the Birmingham Museum of Art, the white mass of *Untitled (Styrofoam Cups)* against the ceiling appears to be a rolling cloud. But, as they near the work to find their table, they discover the identity of the subunits composing the piece. Throughout their meal, they glance upward, amazed at the transformation of the material. As waiters hand to-go beverages to exiting diners, the customers look up from the cup in their hand—destined for a landfill—to the Styrofoam above them. Subtly, *Untitled (Styrofoam Cups)* reminds viewers of the damage that the mundane object in their grip brings to the abstracted landscape they appreciated just minutes ago.

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- ¹Tara Donovan, interview by Kimberly Davenport, "Experts from an email correspondence between Tara Donovan and Kimberly Davenport," *Tara Donovan: Haze* (Houston: Rice University Art Gallery, 2004), 9.
- ²Ferdinand Protzman, "Industrial-Strength Sculpture; At the Hemphill Fine Arts, Tara Donovan's Tar Paper Creation Sticks in the Viewer's Mind," review of *Tara Donovan: Resonances*, Hemphill Fine Arts, Washington, D.C. *Washington Post* (Washington, D.C.), Jan. 22, 1998.



Figure 1: Tara Donovan, *Untitled (Styrofoam Cups)* (2004) at the Birmingham Museum of Art. Museum purchase with funds provided by the Collectors Circle for Contemporary Art, 2005.11, image © Tara Donovan, courtesy of PaceWildenstein, New York

³Helena Winston, "Tara Donovan," review of *Tara Donovan: New York*, New York: PaceWildenstein. *Artus* no. 14 (July 2006), 55.

⁴Kristina Olson, "Tara Donovan," review of *Tara Donovan: New Works*, New York: PaceWildenstein, *Art Papers Magazine* 30, no. 4 (July 2006), 66.

⁵Lillian Davies, "Tara Donovan," in *Vitamin 3-D: New Perspectives in Sculptures and Installation*, ed. Nancy Adajania and Anne Ellegood (London: Phaidon Press Limited, 2009), 98.

⁶Oliver Kornhoff and Poul Eric Tøjner, "Forward," in *Tara Donovan*, ed. Jutta Mattern, Mette Marcus, and Jeanne Rank Schelde (Humblebaek, Denmark: Louisiana Museum of Modern Art, 2013), 7.

⁷Christopher Miles, "Tara Donovan," review of *Tara Donovan: Survey*, Los Angeles, Ace Gallery, *Artforum* 43, no. 9 (May 2005), 253.

⁸Susan Sipple Elliott, "Tara Donovan," *Artus* no. 9 (July 2005), 4.

⁹For interviews with Donovan, see n. 1, 9, and 11. Tara Donovan, interview by Jutta Mattern and Mette Marcus, "Interview," *Tara Donovan* (Humblebaek, Denmark: Louisiana Museum of Modern Art, 2013), 17-20.

¹⁰Tara Donovan, interview by Lawrence Weschler, "Animal, Mineral, Vegetable: The Material Coming to Life," *Tara Donovan* (Hamilton, New Jersey: Isc Press, 2007), 152.

¹¹Michael O'Sullivan, "Parting Ways: An Artist and Her Toothpicks; Tara Donovan's Career and Works are Going Places," *Washington Post* (Washington, D.C.), July 24, 1997; Protzman, "Industrial-Strength Sculpture..." n.p.

¹²Tara Donovan, interview by Collette Chattopadhyay, "Rules for Growth: Tara Donovan," in *Conversations on Sculpture*, ed. Glenn Harper and Twylene Moyer (Hamilton, New Jersey: Isc Press, 2007), 66.

¹³As each installation of *Untitled* requires a different set of toothpicks, it could be argued that each iteration is a separate work meriting distinct titles. However, a full interrogation of the status of these iterations as distinct works is outside the scope of this paper. Davies, "Tara Donovan," 98; Neil, "Tara Donovan," 180.

¹⁴Kornhoff and Tøjner, "Forward," 6.

¹⁵The philosopher Karl Popper examines the visual elements of clouds as mode to understand perception; however, this metaphor is beyond the scope of this paper. For more on this, see Karl Popper, "Of Clouds and Clocks," in *Objective Knowledge: An Evolutionary Approach* (Oxford, Oxford University Press, 1973): 227.

¹⁶For more of Neil's work on Tara Donovan, see Jonathan T. D. Neil, "Legibility of Effort." *Art Review* 18 (January 2008), 74-79; Jonathan T. D. Neil, "The Screen," in *Tara Donovan: Drawings (Pins)*, n.p. New York: Pace Gallery, 2011; and Jonathan T. D. Neil, "Tara Donovan," review of *Tara Donovan*, New York: Pace Gallery. *Art Review* 66, no. 6 (September 2014), 180.

¹⁷The Dow Chemical Company owns the trademark on Styrofoam, a blue, extruded polystyrene foam made for building insulation. Dow clearly delineates that they do not approve of their name "Styrofoam" being applied to coffee cups and packing materials made from polystyrene beads. However, this paper will use "Styrofoam" following common usage of the term as a proprietary eponym used to describe white foam plastic. Judith Tannenbaum, "Styrofoam: From Industrial invention to Artistic Transformation," *Exhibition Notes* no. 29 (Spring 2008), 1-3.

¹⁸Patricia Kelly, "Tara Donovan," review of *Tara Donovan*, Boston, Institute of Contemporary Art, Boston, *CAA Reviews*, March 23, 2009, <http://www.caareviews.org/reviews/1231#.V9nFnZgrLIU>.

¹⁹Protzman, "Industrial-Strength Sculpture..." n.p.

²⁰While the specific details of the contract between Donovan and the Museum remain elusive, it can be assumed that the Museum dismantled and disposed of the Styrofoam cups in order to maintain the integrity of the second installation of *Untitled* (Styrofoam Cups). As noted in an email exchange with Mary Villadsen, dated September 19, 2016.

Quantifying the prevalence of the West Nile Fever, Rift Valley Fever, and Dengue Fever viruses across Mouse Lemurs populations from Madagascar

Emma Hale, Tonia Schwartz, Sarah Zohdy

Wildlife, non-human primates in particular, can serve as reservoirs for arboviruses that can be transferred to humans via mosquitoes (de Thoisy et al., 2001). West Nile Fever Virus, Rift Valley Fever Virus, and Dengue Fever Virus have all been identified in humans in Madagascar (Fontenille et al., 1998; Mathiot et al., 1984; Ratsitorahina et al., 2008). Mouse lemurs (*Microcebus* spp.), primates endemic to Madagascar, may act as potential reservoirs for these viruses, but this has yet to be determined. With this study, we aim to (1) validate the use of blood spots on TropBio™ cards as a method for detecting RNA viruses, (2) determine the prevalence of these viruses in brown mouse lemurs of Madagascar, and (3) test if there is a correlation in the presence of these viruses with habitat degradation.

To validate the protocol, house mouse (*Mus musculus*) blood was spiked with non-infectious viral RNA for Dengue Fever, West Nile Fever, and Rift Valley Fever viruses and inactivated viral particles for West Nile Fever Virus in a dilution series and applied to TropBio™ cards. RNA was isolated from the cards, reverse transcribed to cDNA, and the target viral cDNA was detected by Polymerase Chain Reaction (PCR). Lemur blood samples were collected from intact forest and deforested regions of Madagascar using TropBio™ cards. The samples for each of the viruses were tested using quantitative PCR (qPCR) to determine the prevalence of each of the viruses. The TropBio™ cards were used for both sampling and testing purposes.

Protocols for cDNA reverse transcription and standard PCR using our primers for the viruses have been verified and the PCR product can be visualized on agarose gel. Moving forward, we will refine RNA extraction protocols and qPCR protocols. Once these protocols have been optimized, RNA will be extracted from the Madagascar samples and the presence of the viruses will be quantified with qPCR.

The presence or lack of these viruses in mouse lemur populations of Madagascar can indicate or eliminate a possible source of human outbreak, and a correlation between habitat fragmentation and virus presence can show the effect of habitat degradation on virus presence in endemic species.

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Statement of Research Advisor:

The research being conducted by Emma will be the first study to test for these diseases in lemurs and put their prevalence in the context of deforestation. Further, the viral detection method that Emma is developing will be useful for researchers all over the world to test for these diseases in wildlife.

—*Tonia Schwartz, Biological Sciences*

Longitudinal associations between perceived discrimination and catecholamines

Lydia Homandberg, Thomas Fuller-Rowell, David Curtis

Perceived discrimination (PD) has been associated with lower self-rated health, mortality, and aggregate indexes of physiologic dysregulation (Barnes et al., 2008; Fuller-Rowell, Evans, & Ong, 2012; Schulz et al., 2006). However, few studies have examined associations between PD and catecholamines, or hormones such as epinephrine (adrenaline) and norepinephrine, and even fewer have considered this association using longitudinal data. This is surprising given that over-activation of the sympathetic nervous system has been linked to chronic stress exposure and is thought to be an important mechanism for effects of PD on health (Ahmed, Mohammed, & Williams, 2007; Castro-Diehl et al., 2014).

Our study examined PD as a predictor of changes in urinary epinephrine and norepinephrine concentrations over a three-year period in a sample of college students at a large, predominantly white, midwestern university (N = 149, 45% Black, 55% White; mean age at baseline = 18.8, SD = .96). Two thirds of participants completed a follow-up assessment three years later. Epinephrine and norepinephrine concentrations were obtained from 12-hour overnight urine samples at both time points. PD ($\alpha = .91$) was assessed using 13 items from the Racism and Life Experiences Scales (RaLES; Harrell, 2000). Regression analyses examined the effect of PD on catecholamines at the follow-up time (three years later), adjusting for the initial catecholamine concentrations and other controls. An additional model further adjusted for depression, negative affect, and rejection sensitivity. Full Information Maximum Likelihood estimation was used to deal with missing data. The analytic sample was therefore N=149 across all models.

PD was associated with changes in norepinephrine ($B = .310$; $p = .008$) across the three-year period (Table 1). These effects remained after further adjusting for depression, negative affect, and rejection sensitivity. Analyses for epinephrine yielded the same pattern of findings. These results are consistent with previous

studies showing that PD is associated with physiologic dysregulation and extends that work by establishing a longitudinal association with indicators of sympathetic nervous system activation. This research points to the importance of understanding social elements as risk factors for cardiovascular disease as well as understanding the full spectrum of social influences on health.

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Statement of Research Advisor:

Lydia’s project is the first study to show that perceived discrimination is associated with longitudinal increases in urinary catecholamines, an important marker of sympathetic nervous system activation. I have no doubt that her innovative contributions will be published in a reputable journal and will become the basis for further important inquiry.

—*Thomas E. Fuller-Rowell, Human Development and Family Studies*

Table 1: Models 1-3 results showing race and racial discrimination as predictors of norepinephrine.

	Model 1		Model 2		Model 3	
	<i>B</i>	<i>SE</i>	<i>B</i>	<i>SE</i>	<i>B</i>	<i>SE</i>
Intercept	1.139*	.522	.509	.586	7.468**	2.262
Norepinephrine at T1	.394**	.099	.359**	.093	.407**	.098
Age	-.329**	.116	-.340**	.114	-.360**	.108
Sex (Female)	.094	.118	.036	.107	.018	.112
Race (Black)	.029	.093	-.140	.102	-.017	.166
Heavy Drinker	-.020	.095	-.046	.089	-.058	.094
Cigarette Smoker	.178	.137	.151	.115	.145	.110
BMI	-.081	.082	-.057	.077	-.086	.086
Income: Needs	.016	.115	.020	.108	-.016	.105
Perceived Discrimination			.310**	.117	.416**	.127
Depression					-.142	.098
Negative Affect					.077	.117
Rejection Sensitivity					-.221	.192

Note. All continuously coded predictor variables were standardized to have a mean of zero and *SD* of 1 in all models.

Carotenoids protect against oxidative inactivation of an iron-regulatory protein in the marine copepod *Tigriopus californicus*

Phillip Wang, Ryan Weaver, Geoffrey Hill

Carotenoids are red, orange, and yellow pigments that are the source of coloration in many animals. Carotenoids are thought to have many physiological roles such as immune function and vitamin A synthesis. Carotenoids have been convincingly demonstrated to act as antioxidants *in vitro*, but evidence for their *in vivo* antioxidant activity is contentious. In this study, we used the marine copepod *Tigriopus californicus* to test the antioxidant capacity of carotenoids.

In the wild, *T. californicus* obtains carotenoids by consuming algae. However, *T. californicus* can be cultured on yeast, which lack carotenoids. Copepods raised on yeast have small amounts of carotenoids. However, by raising these carotenoid-free yeast copepods in a carotenoid solution, carotenoids can be redeposited into their integument.

We used tert-butyl hydroperoxide (tBHP) as our oxidative stressor. We exposed 320 yeast-fed and carotenoid-restored copepods to either 0 mM or 1 mM tBHP for one hour and then tested for oxidative damage by measuring total aconitase activity (Fig. 1). Aconitase is a protein that contains an iron-sulfur cluster that is sensitive to oxidative damage. In the presence of reactive oxygen species, ferrous (Fe^{3+}) iron will be reduced to ferric iron (Fe^{2+}), causing aconitase inactivation.

Under normal conditions yeast-fed copepods and carotenoid-restored copepods had equal aconitase activity (Yeast: 78.5 ± 10.9 , Carot: 80.9 ± 5.1 , $n = 3$, $t = -0.293$, $p = 0.78$). Exposure to tBHP decreased aconitase activity in carotenoid-restored copepods by 6% (Carot-tBHP: 75.83 ± 10.26 , $n = 3$, $t = -0.624$, $p = 0.55$), whereas yeast-fed copepods exposed to tBHP had a 34% decrease in aconitase activity relative to unexposed yeast-fed copepods (Yeast-tBHP: 51.5 ± 11.7 , Yeast: 78.5 ± 10.9 , $n = 3$, $t = -3.35$, $p = 0.01$).

We tested for the functional role of carotenoids as antioxidants by exposing carotenoid-deficient and carotenoid-supplemented copepods to an oxidative challenge and found that carotenoids act to protect against inactivation of a critical metabolic enzyme. Our results corroborate *in vitro* studies on the antioxidant activity of carotenoids and provide support for a biologically relevant role of carotenoid accumulation as a protectant against oxidative stress in the wild. Future research will incorporate the same carotenoid-rich/carotenoid-free copepod model to investigate other physiological attributes typically associated with increased carotenoid intake.

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 recently submitted this research to a peer-reviewed journal as a second author. –Geoffrey Hill, *Biological Sciences*

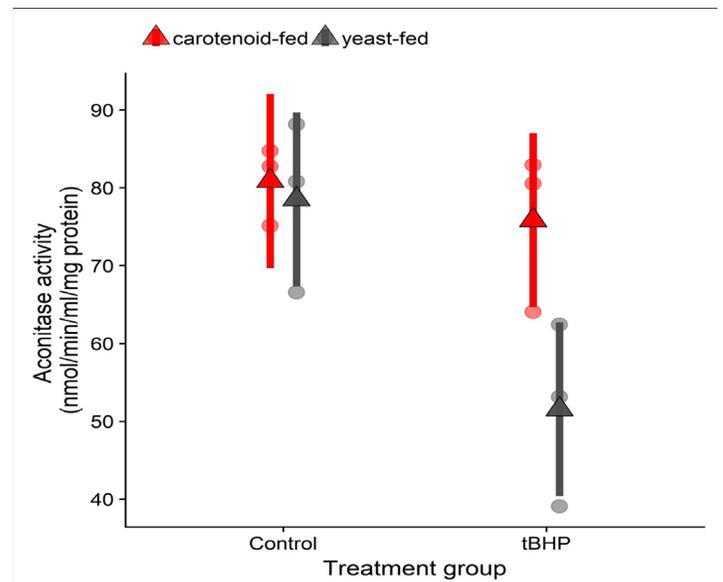


Figure 1: Aconitase activity of carotenoid supplemented (red triangles) and carotenoid deficient (gray triangles) copepods exposed to tBHP versus control.

Design and development of a low temperature tube calorimeter for ammonia refrigerant

Ford Gibbes, Lorenzo Cremaschi

In 1987, with the ratification of the Montreal Protocol, the United Nations agreed that environmentally harmful refrigerants would need to be phased out. This decision has created a need for refrigerants with low global warming (GWP) and low ozone depletion potential (ODP). Ammonia refrigerant is promising in this regard because it possesses both of these characteristics and is also a naturally occurring compound. Unfortunately, ammonia is toxic and corrodes copper; thus it is not commonly used in HVAC systems. Recent government policies encourage the use of low GWP refrigerants and the interest in ammonia has grown since. However, there is a lack of information regarding the frictional pressure drop during two-phase flow phase change processes when ammonia is used inside the tubes of a heat exchanger.

In this project, a tube calorimeter is used to measure the differential pressure drop across a 180° U-bend. The calorimeter is constructed from 3/8", 3/4", and 1" stainless steel tubing with pressure transducer taps at specific distances. The bend radius to tube diameter ratio ranges from 1.2 to 2.5. Flow visualization data are recorded across the 180° glass U-bend using a high-speed camera and a pressure chamber. The data from this study are used to improve pressure drop models of ammonia.

Increased energy efficiency and more predictable heat transfer can be expected for heat exchangers that are designed using the new model. Because of this, ammonia has the potential to become an attractive refrigerant for wide usage in HVAC and refrigeration applications. Currently, the testing facility is nearing the end of construction and testing will be feasible late 2018.

Statement of Research Advisor:

Ford assisted in the design and construction of two heat transfer fluid loops and two refrigeration loops. Additionally, he was responsible for all design and construction of the test apparatus. The new test facility developed under this project will allow our research group to gather new data for pressure drops in U-bends of heat exchangers for ammonia refrigerant-based refrigeration systems. The facility isolates and quantifies the energy losses and inefficiencies associated with U-bends in heat exchangers.

—*Lorenzo Cremaschi, Mechanical Engineering*

Preclinical strategies evaluating the treatment of triple negative breast cancer

Elena Skarupa, Shanese “Lani” Jasper, Robert “Rusty” Arnold

Breast cancer is the second leading cause of cancer death among women. Early detection and diagnosis, along with targeted therapies, have drastically improved survival rates. However, this improvement does not extend to patients with metastatic, triple-negative breast cancer (TNBC). TNBC is an aggressive cancer that is estrogen receptor-negative, progesterone receptor-negative, and HER-2 negative. Therefore, TNBC cannot be targeted via hormone therapies or novel molecularly targeted therapies, such as anti-HER2 therapies. Toxic chemotherapeutics, such as Doxorubicin, remain the primary treatment option. However, Doxorubicin exhibits cumulative dose-limiting cardiotoxicity that limits its clinical utility. In order to create and optimize therapeutic options for the treatment of TNBC, a better understanding of these tumors and their drug interactions is necessary. Often, two-dimensional (2D) *in vitro* models are employed to assess these interactions, but these models do not effectively recapitulate the complexity and multicellular nature of *in vivo* tumors. Three-dimensional (3D) *in vitro* models present more physiologically relevant models for assessing the interplay between drug and tumor by attempting to mimic the complexity of the tumor microenvironment.

For this study, the potency of Doxorubicin was determined using conventional cell toxicity assays. Both the SRB assay, which measures the total protein content of cells, and MTT assay, which evaluates the mitochondrial enzymatic activity of cells, were performed. The assays were conducted using classic 96-well flat-bottom plates with drug exposures ranging from 100 to 0.0001 uM Doxorubicin in the human TNBC cell-line, MDA-MB-231-LUC-GFP. They were evaluated following 24, 48, and 72 hours of drug exposure. The potency (IC₅₀ value) was calculated after 72 hours of drug exposure. Cytotoxicity correlated positively with Doxorubicin concentration and length of Doxorubicin exposure.

The development and evaluation of the 3D model have been conducted in parallel with the 2D experiments, which serve as a reference platform for evaluating the 3D model using tumor spheroids. Future studies will focus on determining potency of Doxorubicin after the 72-hour time point using a single cell (tumor only) and multicellular (tumor and cells associated with tumor stroma, such as macrophages or fibroblasts) 3D models, as well. Future studies will also focus on evaluating differences in gene and protein expression between the 2D and 3D cell models with particular interest in HIF-1, P21, and other markers associated with tumor growth and metastases. HIF-1 is the Hypoxia-Inducible Factor, which is a transcription factor that responds to decreased available oxygen in the cellular environment. P21 is a cyclin-dependent kinase inhibitor, which inhibits the normal cell cycle. The goal of this work is to determine if 3D models can be used to give a more accurate depiction of the *in vivo* tumor response in an effort to develop better treatment options for TNBC.

Statement of Research Advisor:

Over the last year Elena has worked to establish and test a 2D- and 3D-tumor spheroid platform that can be used to examine the effect of individual cell types on tumor growth and responsiveness to chemotherapy. This platform will permit examination of different drug treatment schedules and performance of various nanomedicines with the goal of improving the treatment of aggressive metastatic cancers.

—Robert “Rusty” Arnold, Harrison School of Pharmacy

Expression of the INK4AB/ARF tumor suppressor transcription factor MSK1 in canine breast cancer: Quantification through qPCR and correlation with established phenotypes

Jonathan Dismukes, Patricia DeInnocentes, R. Curtis Bird

MSK1 is a regulator of a protein that encodes for INK4A/p16, a vital tumor suppressor gene that is found defective in both human and canine cancers. While the cause of many of these defects are known and lie within mutations of the genetic coding for INK4A/16, the other causes are most likely due to up-stream mutations, possibly with MSK1. Canine mammary tumors cells are used in this study, since dogs are excellent models for human cancer and exhibit comparable molecular targets. Prior research exhibited heightened expression of MSK1 within tumor cell tissue after traditional polymerase chain reaction (PCR) amplification and gel electrophoresis.

To quantify the levels of this expression, quantitative-PCR (qPCR) was utilized using a SYBR Green fluorophore, which preferentially binds to double-stranded DNA during amplification. Therefore, the levels of fluorescence correlate directly with the amount of product as it increases exponentially. Following analysis, MSK1 was confirmed to have more starting product within tumor cell lines than the normal epithelial cells, albeit varying degrees among the six lines used.

Future work will compare the data to the known phenotypes of the respective cells. Additionally, further tests exploring downstream protein kinases of INK4A/p16 and CDKs 1 and 2 with flow cytometry and fluorescence microscopy will further verify expression and location of the transcription factor during the rapid growth of tumor cells. With these tools, description of MSK1 profiles among the different canine mammary cell lines and levels of expression will lead to evidence about defects within the mechanism of the INK4A/p16 tumor suppressor and changes in cellular location. Ultimately, this knowledge of the faulty cellular mechanism within canine cancer will allow for potential gene therapy treatments to correct the known defects in both dogs and humans.

Statement of Research Advisor:

Jonathan helped develop a research strategy to first determine if the canine breast cancer cells expressed this newly described transcription factor thought to regulate cell proliferation and then designed a qPCR assay to assess the levels of expression. Though it took most of the summer to first develop and then optimize the assay, he was able to detect this important regulator of gene expression and to quantify its expression levels in cancer cells he grew in culture. This technology will allow the further investigation of this important transcription factor and its contribution to cancer cell proliferation.

—*R. Curtis Bird, Pathobiology*

Correlation of pelvis kinematics with ground reaction forces, muscle activation, and hormones during a drop vertical jump

Gabrielle Gilmer, Michael Roberts, Gretchen Oliver

Female athletes are 2 to 9 times more likely to tear their anterior cruciate ligament (ACL) than males [1]. Clinical tests and prevention programs focusing on the lumbopelvic-hip complex (LPHC) are used to identify injury susceptibility [2]. Despite implementation, females continue to have higher ACL injury rates. Recently, relaxin, a peptide hormone similar to insulin, was identified as interfering with the structural integrity of ACLs in females but not males [2]. We aimed to answer the question: how are pelvis kinematics during the drop vertical jump (DVJ) related to ground reaction forces (GRF), muscle activation, and serum relaxin concentrations (SRC)?

Twenty-two female athletes (21.7 ± 3.7 yrs.; 64.8 ± 8.2 kg; 1.8 ± 0.3 m) participated. Participants performed a DVJ while equipped with various sensors. A DVJ involves the participant standing on a box 0.505 m high, dropping off the box to the ground, immediately jumping as high as they can in the vertical direction, then landing. Kinematic data were collected at 100 Hz using an electromagnetic tracking system. GRF data were collected at 1000 Hz using a Bertec force plate. Electromyographic data were collected at 1000 Hz using a Delsys system. Blood samples were collected when SRC peaked and processed using a Quantikine Human Relaxin-2 Immunoassay.

Pearson product-moment correlations revealed significant correlations ($p < 0.05$) between the following: anterior/posterior GRF and pelvis lateral flexion, rotation, and adduction; vertical GRF and pelvis adduction; quadricep to hamstring ratios and pelvis flexion, adduction, lateral flexion, and rotation; gluteus medius activation and pelvis rotation, adduction, and lateral flexion; and SRC and pelvis adduction.

These correlations reveal the interconnection between ACL injury risk factors during a DVJ. Larger anterior/posterior GRF and smaller vertical GRF indicate the center of mass is not appropriately distributed over the stance legs [3]. Larger anterior/posterior GRF was associated with the athlete's pelvis tilting and rotating towards the dominate side and collapsing in towards their midline. Pelvic collapse towards the midline was also associated with decreased vertical GRF. These positions put the pelvis in a more injury susceptible position and decrease the energy transfer along the kinetic chain, which could result in distal segments being placed in injury susceptible positions (Figure 1).

The relationships between muscle activation and pelvis kinematics agree with previous literature that document low activation of the gluteal and hamstring musculature coupled with high activation of the quadriceps is associated with low energy transfer from the LPHC [4]. Athletes who had higher quadricep to hamstring ratios placed their pelvis in a position that was anteriorly tilted, collapsed towards the midline, tilted and rotated to the dominate side position. In contrast, athletes who displayed higher gluteus medius activation kept their pelvis in a more neutral location.

The relationship between pelvis adduction and SRC suggests higher SRC is associated with an adducted hip. Hip adduction during the DVJ is an injury susceptible position, due to decreased LPHC stability. LPHC instability and increased SRC should be of great concern for sports medicine personnel. This finding reiterates the need for coupling biomechanical evaluation with monitoring hormonal concentrations.

Ultimately, these results suggest biomechanical, neuromuscular, and hormonal risk factors for ACL injury are not independent and should be evaluated together. Further studies are needed to determine if any causal relationships exist between these variables.

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Statement of Research Advisor:

Gabrielle’s work is an inspiration for future undergraduate research fellows. Gabrielle was able to utilize her research experiences to develop, design, and carry out a solid study that will further the world of sports medicine and biomechanics.

—Gretchen Oliver, *Kinesiology*

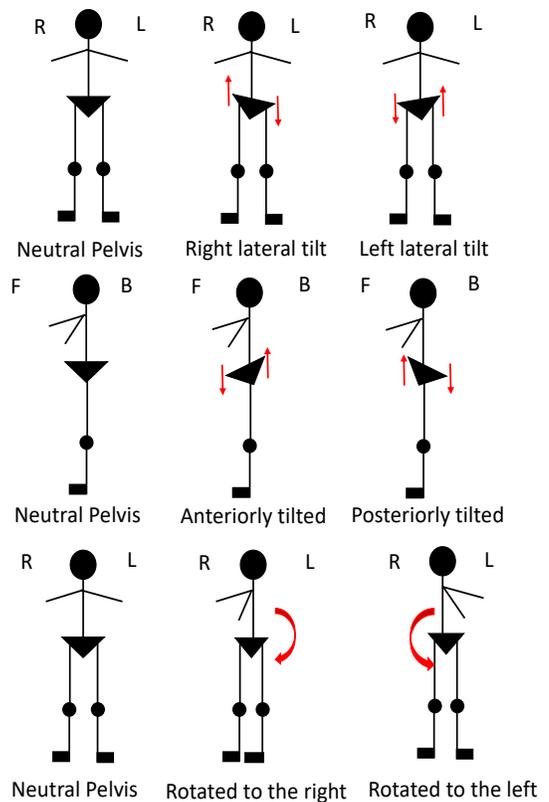


Figure 1: The three-dimensional positions of the pelvis are shown.

Mechanics of professional polo players exhibiting pain vs. players without pain

Abigail Brittain, Gretchen Oliver

As one of the oldest sports still played today, it is somewhat surprising that very little data have been collected on the sport of equestrian polo. Of the available data, the focus has been either on the kinematics and kinetics of the offside forehand swing^{1,2} or injury statistics^{3,4} of polo athletes. No study has yet to link the two areas of focus: mechanics and injury. The purpose of this study to identify significant differences in swing mechanics between female polo athletes with and without pain.

Ten female professional polo athletes (33.0 ± 10.4 yrs.; 1.69 ± 0.06 m; 66.9 ± 9.3 kg) participated. After signing an informed consent and completing a health history questionnaire, participants were attached to an electromagnetic tracking system (trakSTAR™, Ascension Technologies, Inc., Burlington, VT, USA) synced with The MotionMonitor® (Innovative Sports Training, Chicago, IL., USA). Each participant then warmed-up and executed three match effort offside forehand shots from a wooden horse. Warm-up time as well as equipment (mallet and helmet) were not standardized in effort to minimize testing environment adjustments. Testing procedures were approved by the university's Institutional Review Board prior to all testing.

Five of the ten participants indicated they were currently experiencing pain and five indicated having no pain, allowing for participants to be divided into two equal groups, those with pain and those without pain. Kinematic data were analyzed at three swing events: take away (TA), top of backswing (TOB), and ball contact (BC) (Figure 1). An independent samples t-test was conducted to determine significant correlations between trunk and swing-side shoulder kinematics and reports of current pain. Significant differences were calculated using IBM SPSS Statistics 21 software (IBM Corp., Armonk, NY) for normally distributed data with an alpha level set apriori at $\alpha = 0.05$. Significant differences were found between pain and shoulder elevation at TA ($t(7.54) = 2.999, p = 0.18$); shoul-

der horizontal abduction at TOB ($t(7.061) = -2.868, p = 0.24$); and shoulder elevation at TOB ($t(7.322) = 3.030, p = 0.18$).

Based on this study, polo players exhibiting pain display different kinematics of the offside forehand swing, compared to those with no pain. The findings of this study suggest that athletes who display higher shoulder elevation at TA and TOB, as well as greater shoulder horizontal abduction at TOB, may be more likely to also experience pain. This means that bringing the mallet excessively high and farther from the midline of the body may be related to incidence of pain susceptibility in polo athletes. The study supports previous work that reported higher forces at the elbow when shoulder horizontal abduction is greater¹. Based on these results, more research regarding pain location, kinematics, and kinetics is warranted.

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2. Oliver GD, Gilmer G, Barfield J, Brittain A. Swing mechanics of the offside forehand in professional female polo athletes. *Journal of Orthopedic Research and Therapy*. 2018.
3. Costa-Paz M, Aponte-Tinao L, Muscolo DL. Injuries to polo riders: a prospective evaluation. *Br J Sports Med*. 1999.
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Statement of Research Advisor:

Abigail's work documents some of the first data published regarding polo swing mechanics. Abigail was able to utilize her research experiences carry out a solid study regarding polo swing mechanics that will not only further the sport of polo but also world of sports medicine and biomechanics.

—Gretchen Oliver, Kinesiology

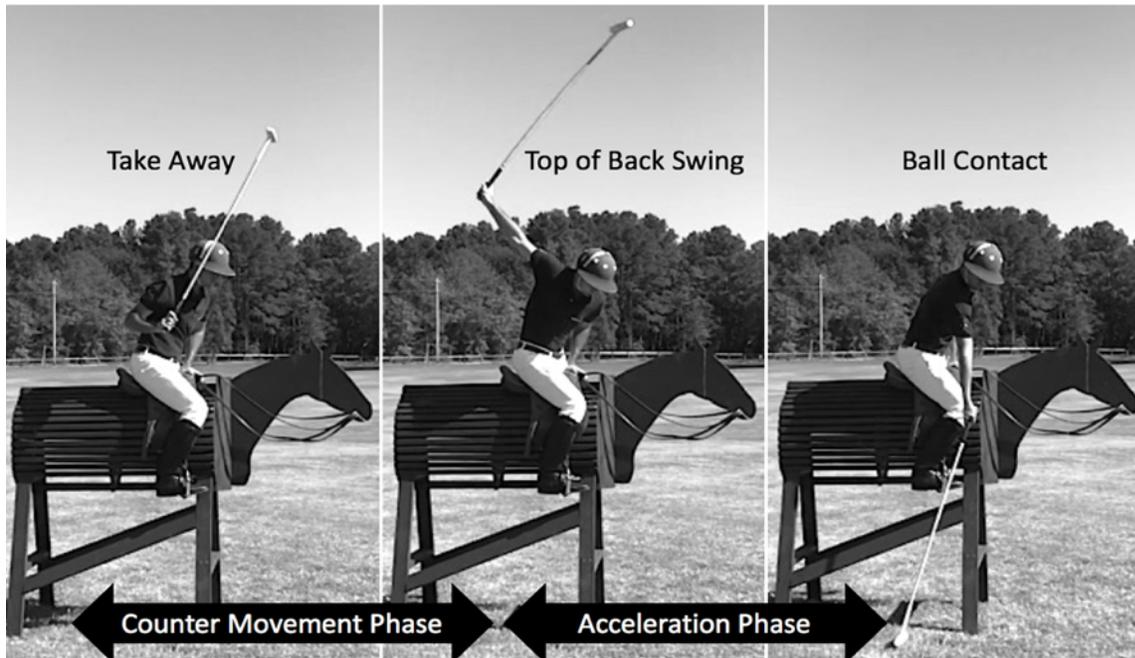


Figure 1: Events of offside forehand polo swing.

Synthesis and properties of PTEMA

Jihyuk Kim, Michael Minkler Jr., Bryan Beckingham

The growing world population is leading to an increasing demand for energy. This demand places responsibilities on us to seek either alternative renewable energy sources or more efficient means of energy usage. The search for more energy-efficient technology has led to research on devices such as thermoelectric generators to recover low-grade waste heat. Specifically, conductive polymers, like polythiophenes, can greatly enhance the feasibility of these devices. Polyalkylthiophenes are widely studied due to their favorable solid-state and optoelectronic properties, well-controlled synthesis, and tunable microstructure. The optoelectronic and solid-state properties can be easily altered by changing the polythiophene microstructure. This allows us to synthesize a wide variety of polythiophenes presenting an opportunity to enhance the performance of solar panels, thermal electric generators, organic photovoltaics, and sensors due to their tunable microstructure.

To better understand how varying microstructure affects the optoelectronic and physical properties of polythiophenes, we synthesized a novel 2-(2-(thiophen-3-yloxy)-ethyl) malonate monomer in order to enable the synthesis of a polythiophene with unique properties. Figure 1 shows the synthesis of this monomer, which involves several steps. One of the main difficulties in synthesizing the monomer was the synthesis of 3-(2-bromo)-ethyloxythiophene from 3-methoxythiophene. These two intermediate monomers have very similar structures, making their separation difficult. Upon analysis of 3-(2-bromo) ethyloxythiophene via ¹H NMR spectroscopy, 3-methoxythiophene peaks were noticed, and it was determined that the 3-methoxythiophene should be removed via vacuum distillation or through further reaction. The 3-(2-bromo) ethyloxythiophene was successfully separated via vacuum distillation and the successive steps of the monomer synthesis were performed without interruption.

In future work, this monomer will be polymerized using Grignard Metathesis polymerization to yield poly(2-(2-(thiophen-3-yloxy)-ethyl) malonate). Grignard Metathesis polymerization along with the base work up will transform the malonate group to malonic acid. The final product will be poly(2-(2-(thiophen-3-yloxy)-ethyl) malonic acid (PTEMA). Since this polymer has not yet been successfully synthesized, intense characterization including UV-Vis spectroscopy, ¹H NMR spectroscopy, FTIR spectroscopy, gel-permeation chromatography, differential scanning calorimetry, and x-ray diffraction will be performed following synthesis. This polymer is expected to possess halochromic behavior, as demonstrated by a chemically similar polythiophene. The functionalization of the polythiophene with malonic acid will allow the polymer to change color with a change in pH, and potentially alter other optoelectronic properties. This derivative of polythiophene presents a possible route for the fabrication of a polymeric pH sensor that indicates a change in pH through optical and electronic means.

Statement of Research Advisor:

Jihyuk successfully navigated a difficult series of reactions and found a successful means for purifying his target molecule, 2-(2-(thiophen-3-yloxy)-ethyl) malonate. His work lays the foundation for further synthetic efforts for the synthesis of a multifunctional polymer that would combine the semiconducting properties of polythiophenes with the pH-responsiveness of malonic acid.

—Bryan S. Beckingham, *Chemical Engineering*

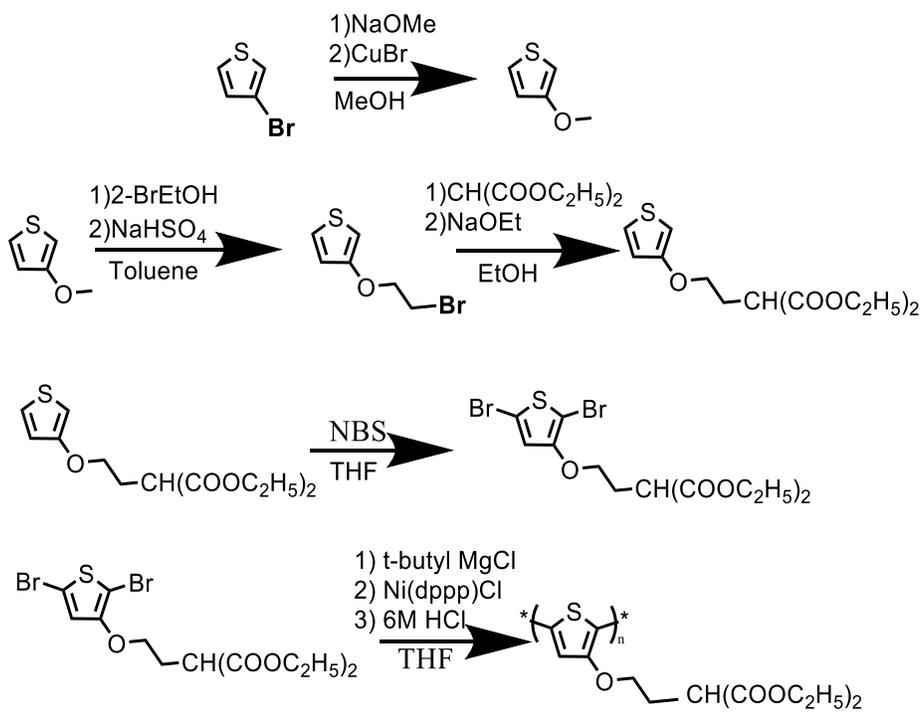


Figure 1: Schematic diagram of PTEMA synthesis.

Assessing interobserver replicability in the scoring of enthesal markings through 3D technology

Kyle McLaughlin, Kristrina Shuler

This study is aimed at 3D technologies and their potential to advance research methods in the physical anthropology field. The focus of the research was on enthesal markings, or where muscles and tendons attach to bones. These bony sites' physical appearances will change over time, depending on use or lack of their use during a lifespan. These physical changes can give us insight into the activities in which the individual took part. For example, Shuler et al. (2012) assessed entheses from a site in Moundville, Alabama, in comparison to small farmsteads and villages in other parts of Alabama and Mississippi and found that enthesal development in these groups is consistent with predictions that some early agriculturalists (1000-1500 CE) experienced greater activity-related stress than hunter-gatherers (500-900 CE) regionally. Others such as Noldner (2013) have examined the variation of enthesal markings for individuals by burial locations within sites, such as a colonial Spanish mission Tipu, Belize. The researchers reported that individuals with robust and rugged entheses who were buried far away from the church at this site were more likely to have been low-status laborers.

In research, entheses are given a score based on a standard of physical descriptions. However, there is often a lack of agreement between researchers and their scores because scoring depends on how a researcher interprets the entheses' appearance. The goal of this study was to determine whether scanning and printing an entheses would lead to more agreement among researchers' scores. Forty-two entheses (subscapularis, pectoralis major, extensor carpi radialis longus, quadriceps femoris, and soleus) from the Newton Plantation osteological series were scored from seven bones used to create three identical scoring sets: dry bones, corresponding 3D-scans, and 3D-replicas. NextEngine© scans (29K resolution) were printed via Makerbot Replicator©. Entheses were then scored independently by two observers of varying experience levels using a set of standards. Results indicated

scoring agreement about 50% of the time for each representation of the entheses. Low replicability may stem from subjectivity in scoring, although experience did not seem to impact agreement. Small samples scored within a brief (one-hour) period allowed observers see the same entheses back to back across the bone, scan, and print. This methodology may have influenced the observers' scores; however, more intraobserver study is needed to determine if this is the case. This preliminary study does not suggest a major difference when scoring entheses from real bone versus 3D technology, or between observers, though the lack of agreement between observers may suggest the need to refine the scoring method.

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Statement of Research Advisor:

Skeletal muscle attachment sites are commonly evaluated by biological anthropologists to gain insight into patterns of labor and lifetime activities in past communities. Kyle's research compared interobserver differences in two of the most commonly used methods of assessment in the field with novel three-dimensional (3D) scanning and printing methods.

—*Kristrina Shuler, Anthropology*

Does community conservation improve human and wildlife health in eastern Madagascar?

Jordan Broadhead, Sarah Zohdy

The purpose of this study was to compare the health and well-being of people and wildlife in community protected and non-protected forests in eastern Madagascar. Forest loss on the island of Madagascar contributes to both the endangerment of some of the world's most threatened species and the spread of poverty-linked infectious diseases in human populations. The practice of slash-and-burn agriculture has resulted in the loss of 40% of the island's forest since 1950. Community-driven conservation efforts may offer the potential to improve human health and well-being while simultaneously protecting endemic wildlife.

In this study, we captured small mammals using Sherman traps and assessed the species diversity, invasive rodent distribution, mass, and ectoparasite counts for small mammals in both the protected forest and in a disturbed habitat. To assess human health, Association Mitsinjo, the local community conservation center, administered surveys to the local villages. One village was surrounded by protected forest and two were surrounded by deforestation. Surveys assessed the economic status, education, and personal health of the inhabitants.

We found that small mammal diversity in the protected forest was greater than in the disturbed forest. Invasive rodents were more prevalent in the disturbed forest. Mass and ectoparasite counts were not significantly different between the two sites for any species captured ($p=0.38$ and $p=0.31$). The two villages surrounded by disturbance had 103 responses from 50 households with a mean age of 33. The single smaller village surrounded by protected forest had 15 responses from 5 households with a mean age of 29. Comparing the health of villages over 6 months prior to the survey, people living in the protected forest village had more fever incidents but fewer fleas and no ticks, lice or diarrhea. People living in disturbed villages had fewer fever incidents but more lice, fleas, ticks, and diarrhea. We surveyed 16 guides and 46 agricultural workers. Comparing the health of people in these

occupations over 6 months prior to the survey, we found people who work as ecotourism guides had more ticks and diarrhea, fewer fever episodes, and no lice or fleas. People who work in agriculture had many more fever occurrences, more lice, and more fleas but fewer ticks and cases of diarrhea. People living in the protected forest village reported less income than those living in the disturbed village. Guides reported greater income than agricultural workers.

This research suggests that community conservation can benefit human health and well-being while simultaneously protecting native forests and endemic species diversity. Community conservation has the potential to be an effective public health strategy.

Statement of Research Advisor:

Jordan Broadhead has a deep interest in disease ecology and how human and animal health are linked. Jordan led this particular project investigating whether wildlife conservation can play a role in improving human health and well-being. Jordan made two trips to the island of Madagascar for this research, and in collaboration with Association Mitsinjo, the University of Antananarivo, and students from the Auburn College of Veterinary Medicine and Emory University, Jordan participated in all parts of the project from wildlife field sampling to helping with survey design and analysis and presentation of results at national conferences.
—Sarah Zohdy, *Forestry and Wildlife Sciences*

Mental health integration through accountable care organizations

Cassidy Roby, Rene McEldowney

Mental health treatment continues to be the “elephant in the room.” Psychiatric services need to be more adequately integrated into primary and hospital care, yet are often superseded by physical healthcare issues due to the negative stigma of mental ailments. The high prevalence of mental health disorders and lack of access to facilities are indicators of need for greater integration of mental health services. Accountable Care Organizations (ACOs) aim to increase coordination between physicians in order to improve quality and increase savings, and thus provide a proper framework for assessing current mental health integration efforts.

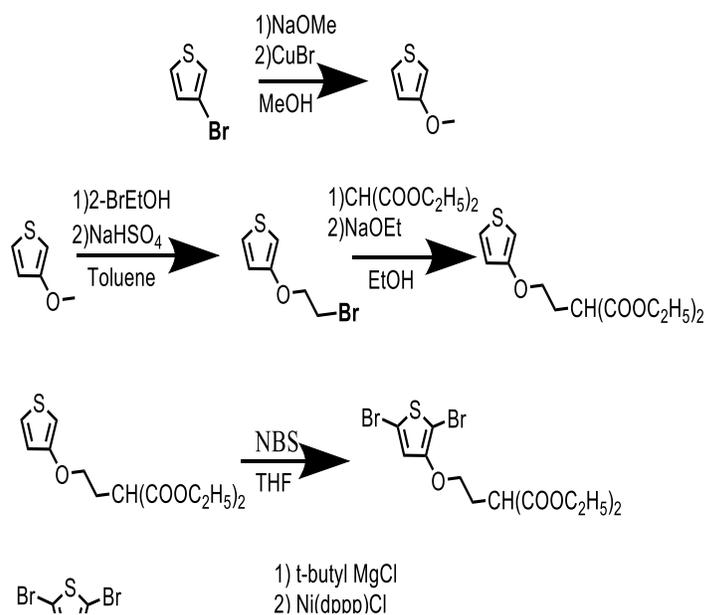
Our research examined the current integration of mental health services in hospital-led ACOs by looking at adolescent, consultation, education, emergency, geriatric, outpatient, partial hospitalization, and residential treatment services offered using the data from the American Hospital Association (AHA) 2014 database. In our analysis we defined “mental health integration” as the availability of psychiatric services offered by that hospital entity. We acknowledge the limitations of this definition. Our assumption is that if psychiatric services are more readily available to patients, they will be more inclined to use them. Our analysis showed all psychiatric services, with the exception of residential treatment, are more adequately integrated into hospital-led ACOs than non-ACOs. Residential psychiatric treatment did not appear to have any statistically significant differences between ACOs and non-ACOs. Recent increased prevention efforts may account for the lack of services in both ACOs and non-ACOs. We speculate that the lack of residential treatment in hospitals is a positive sign of preventative medicine being able to treat psychiatric patients without needing inpatient rehabilitation services. We make this speculation with the understanding that inpatient rehabilitation is often more time consuming, expensive and usually intended for very extreme psychiatric cases. Further research is needed to confirm the reasons behind this lack of residential treatment services.

These results show that ACOs provide greater quality of services to mental health patients than non-ACOs, and imply that ACOs are achieving their goal of providing coordinated care. Further research is needed to find if mental health integration has helped ACOs achieve greater cost savings. This study serves as a reference point to track the progress of mental health services in a rapidly changing healthcare system.

Statement of Research Advisor:

The research Cassidy conducted demonstrates the ability for adolescent mental health services to be fully integrated into hospital-owned Accountable Care Organizations at the primary provider level. Utilizing the American Hospital Association comprehensive database, Cassidy’s analysis indicates that hospital-owned Accountable Care Organizations are an effective way to assimilate adolescent mental health services into the scope of primary medical care. Cassidy is a Health Services Administration Spruiell Scholar and an extremely accomplished individual and student.

—Rene McEldowney, Health Administration



The fine structure of the tentacular apparatus of *Mnemiopsis leidyi*

Dorothy Mitchell, Anthony Moss, Gen Dong

Ctenophores are gelatinous zooplankton found throughout Earth's oceans, from the depths of oceanic trenches to the surface of shallow estuarine systems. Their unique morphology and physiology have long intrigued researchers working on various topics in development, neurophysiology, and evolution. Ctenophores have become popular research organisms because they were recently found to be the last common ancestor to all animals. I have investigated the cellular architecture of the ctenophore tentacular bulb and associated tentacles. The ctenophore *Mnemiopsis leidyi* was chosen because of its year-round availability and its resilient physiology. *Mnemiopsis* bears tentacles that arise from two centrally positioned tentacular bulbs. They have fine tentillae that extend perpendicular to the tentacles, which contain 'sticky' colloblast cells that aid in prey capture.

My goal was to visualize tentillar organization at a cellular level, using light microscopy and transmission electron microscopy (TEM). Preparations for microscopy were chemically fixed, infiltrated and embedded in Spurr's epoxy resin. I generated 0.25-micron-thick cross-sections of the tentacle bulb using an ultramicrotome, then stained and viewed at up to 100x under a microscope equipped with a digital color camera. Since slide preparation causes dehydration and collapse of the watery tissue, I additionally cryosectioned frozen ctenophore bulbs to determine if dehydration changed the structural profile. I also performed analysis of 60-nm-thick 'thin sections' using TEM. I sectioned resin-embedded samples with glass knives, followed by staining with lead citrate and uranyl acetate. Thin sections were mounted on copper grids, dried and examined by TEM on the Zeiss EM10 microscope in Auburn University Research Instrumentation Facility (AURIF) at up to 30,000X magnification. We were able to form a high-resolution 'mega-image' (totaling 1.2 terabytes) of a tentacular bulb, revealing previously unseen and unknown cellular structures and cell-cell interactions.

We discovered the following: (1) the oral-most end of the bulb and canal was not attached to the body; (2) what we interpret to be a layer of food-absorptive cells that corroborates a recent report (Giribet, 2016); (3) a structural feature we interpret to be bulb neuropile surrounding a region rich in what we interpret to be neural stem cells; (4) a region of tentillar longitudinal fission and growth (TLFG) that 'waves' laterally to either side of the bulb to 'feed' tentillar structure within the tentacular groove of the food groove; and (5) several unknown cell types, including apparently apoptotic cells, at different stages of differentiation.

We view our findings as groundwork to investigate the function and identity of neuronal stem cells. These cells are of much interest because *Mnemiopsis* demonstrates the ability to rapidly regenerate and heal its wounds. Little is known about the function of the cells involved in tissue repair and how these cells interact with surrounding effectors. EdU (5-ethynyl-2'-deoxyuridin) analysis (Salic and Mitchison, 2008) by others showed a source of cell proliferation in the aboral portion of the ctenophore *Pleurobrachia pileus* (Alie et al., 2010). However, *Pleurobrachia* is structurally distinct from *Mnemiopsis*. Future studies will precisely determine the nature of the cells uncovered and relate them to their differential gene expression during cellular development.

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Statement of Research Advisor:

Dorothy's work on the ultrastructural organization of the *Mnemiopsis* tentacular bulb and tentacular/tentilar structure has produced major strides in our understanding of this sensory integration center. This is a very difficult and time consuming, even tedious effort, and yet Dorothy's enthusiasm and boundless energy, and attention to detail, together with the careful overview of her graduate student supervisor in addition to myself, has broken open the cellular organization of this mysterious system. This work is of considerable importance because ctenophores are thought to be among the earliest metazoan (i.e., multicellular animals) to evolve, and are thought to therefore have one of the most ancient nervous systems. That the ctenophores are so ancient poses an interesting possibility: namely that they could be a potential precursor to all current nervous systems in more complex, 'higher' animals, including ourselves.

—Anthony Moss, *Biological Sciences*

Identifying active tectonics in the New Madrid Seismic Zone using LiDAR and geologic data

Caleb Eldridge, Lorraine Wolf

The New Madrid Seismic Zone (NMSZ) is located in the central U.S. and includes parts of west Tennessee, eastern Arkansas, and eastern Missouri. This earthquake zone is considered the most seismically active area east of the Rocky Mountains, though most of the earthquakes are small and not always felt by humans (magnitudes < 3). In 1811-1812, however, three major earthquake sequences occurred with estimated magnitudes up to M 7.8. Earthquakes of that magnitude today would cause massive damage to cities and towns along the Mississippi River valley, including Memphis and St. Louis. Studies suggest an earthquake recurrence interval of approximately 500 years during the last 1200-year period.

The purpose of this project was to analyze recently available Light Detection and Ranging (LiDAR) data for portions of the NMSZ to identify surface deformation that is possibly related to active faults in the region. Most faults in the central U.S. are hidden by a thick covering of alluvial sediment; thus, researchers have to rely on subtle changes in surface topography and remote sensing of subsurface structures for evidence of deeply buried active faults.

For this project, we compiled a comprehensive database of geologic and geophysical information for the study area using Geographical Information Systems. This database consists of Digital Elevation Models (DEMs), mapped locations of known or suspected basement faults, geologic maps and cross-sections, and information on earthquake locations and magnitude. The new LiDAR data was added to the database and provided the highest resolution topography available for the area. By combining features identified in the LiDAR data with earthquake locations and magnetic field anomalies, potentially active faults can be identified. Aeromagnetic data were downloaded and processed using wavelength-separation methods. A literature search was conducted to collect articles describing magnetic anomalies and subsurface geology that could be related to regional seismic activity and surface features observed in the LiDAR models.

The LiDAR data collected show prominent earthquake-induced soil liquefaction deposits aligned in a northeast-southwest orientation that match the orientation of faults in the area. The LiDAR data also show linear ridges that align sub-parallel to the liquefaction deposits and that may suggest deformation from buried faults. Analysis of the magnetic data shows anomalies that share the northeast orientation and that could be related to seismic activity in the area. They also show the presence of igneous plutons that may concentrate tectonic stress and may explain some of the observed seismicity. These hypotheses may be further explored in future work.

Statement of Research Advisor:

Caleb's work on compiling data sets related to seismicity in the NMSZ represents a solid contribution to earthquake research in intraplate areas. Unlike that seen at tectonic plate margins, evidence of active faulting and deformation in the mid-continent is subtle and often not visible. Through his analysis of newly acquired, high-resolution topography, Caleb has identified surface features that suggest the presence of active, but hidden faults.

—*Lorraine Wolf, Geosciences*

The effect of bioretention media treatment of stormwater on *Daphnia* populations

Rachel Kuntz, Thorsten Knappenberger, Eve Brantley

Untreated stormwater entering streams can result in contamination of drinking water supplies and shell-fishing waters, prohibition of recreational use (e.g., fishing, swimming, and boating), injury to aquatic biodiversity, danger to public health, and increased flooding. Total suspended solids, nutrients (including phosphorus and nitrogen), pathogens, and petroleum-based contaminants are common stormwater pollutants of concern. Bioretention cells are landscape features that may serve as stormwater-control measures by reducing the impact of non-point source pollutants through filtration, absorption, and flow velocity reduction. The purpose of this experiment was to examine the lethal effect of local stormwater runoff and to evaluate the influence of bioretention media on stormwater toxicity.

Adult *Daphnia pulex* (water flea) were exposed to stormwater runoff collected in Auburn, Alabama, to observe the effects of runoff toxicity. Comparisons in survivorship were made between *D. pulex* exposed to untreated stormwater runoff and runoff treated with bioretention media, respectively. We hypothesized the bioretention media would decrease the toxicity and increase survival across *D. pulex* populations.

Approximately 85% of *D. pulex* exposed to the stormwater runoff treated with the bioretention cell survived, compared to 54% exposed to untreated runoff. This difference in survivorship is statistically significant ($p < 0.05$) and indicates bioretention media may minimize stormwater toxicity that, in turn, benefits aquatic communities in urban areas.

Activity levels of the *D. pulex* were observed and recorded after 30 minutes, 12 hours, 24 hours, and 48 hours of exposure to the treated and untreated stormwater. Sixty-one percent exposed to untreated runoff died within the first 30 minutes of exposure; 0% of the *D. pulex* exposed to treated runoff died during the same time frame. 81% of the fatalities experienced

from exposure to treated runoff did not occur until after 12 hours of exposure. Considering most storms in Alabama typically do not last longer than one hour, the likelihood of aquatic organisms being exposed to the same concentration of toxicity for over 12 hours is very low. This suggests bioretention media can be an effective solution to reduce toxicity of stormwater runoff and protect surrounding aquatic organisms from harmful polluted stormwater runoff. Future studies may evaluate different bioretention media, runoff from other impervious surfaces types (new asphalt, concrete, roof top), and sublethal impacts such as reproduction or movement.

Statement of Research Advisor:

Rachel's research is an important contribution to our understanding of how stormwater affects aquatic organisms, and it demonstrates the effectiveness of bioretention media on reducing stormwater toxicity and mitigating stormwater impacts on *Daphnia pulex*.

—Thorsten Knappenberger, *Crop, Soil and Environmental Science*

Electron-hole hopping as catalytic self-preservation: how catalase-peroxidase from *Mycobacterium tuberculosis* avoids the perils of peroxide decomposition

Patrick Sahrman, Kirklin McWhorter, Jessica Krewall, Douglas C. Goodwin

Catalase-peroxidase (KatG), an enzyme produced by bacteria and fungi, is especially prominent among some of the world's most prolific pathogens (e.g., *Mycobacterium tuberculosis*). These organisms use KatG to defend against H_2O_2 produced by host immune responses. Contrary to all other members of its enzyme superfamily, KatG is bifunctional, capable of catalase and peroxidase activities. Further, against long-standing predictions that its two activities would be mutually antagonistic, we have shown that peroxidatic electron donors (PxEDs) stimulate KatG's catalase activity.

A narrow active site access channel prevents PxEDs from directly reducing KatG's heme cofactor, suggesting that the protein itself must facilitate free radical transfer from within the active site and channel this absence of an electron to the periphery of the enzyme via oxidizable amino acids (i.e., electron-hole hopping) to prevent irreversible inactivation. This links the active site to the solvent-accessible surface of KatG. In this manner, peroxidase activity (i.e., the use of PxEDs) serves to uphold KatG's robust catalase activity. We have shown that one prominent electron hole-hopping pathway begins with oxidation of an active site tryptophan (W321). A methionine (M377), an amino acid which contains an oxidizable sulfur, 3.9 Å away from W321 is a prime candidate for the next step in this mechanism (Figure 1).

To investigate this possibility, we used site-directed mutagenesis to generate M377I KatG, replacing methionine with the non-oxidizable amino acid isoleucine. M377I KatG variant displays similar catalase activity to that of wild type, indicating M377 is not directly involved in catalase turnover. Subsequent additions of substrate after an initial reaction with H_2O_2 in the presence of ABTS, a peroxidatic electron donor, show a decreased initial rate, suggesting M377 is pertinent in the mechanisms of catalase recovery.

M377I is incapable of aiding in through-protein radical transfer from W321, leading to irreversible inactivation of enzyme due to advanced oxidation of the protein even in the presence of ABTS. As with both wild-type and W321F KatG, a variant of KatG with the previously mentioned tryptophan replaced with a non-oxidizable phenylalanine, inclusion of a PxED sustains the catalase activity of the M377I variant to the complete consumption of H_2O_2 . We propose that possible pathways for through-protein radical transfer expand as the distance from the active-site heme cofactor increases, thereby producing a corresponding decrease in contribution from any particular oxidizable amino acid in catalase activity preservation. Future research will focus on generation of more variants of KatG to identify amino acids involved in the hole-hopping mechanism, as well as a definitive look at the iron oxidation states of the heme cofactor using Mössbauer spectroscopy.

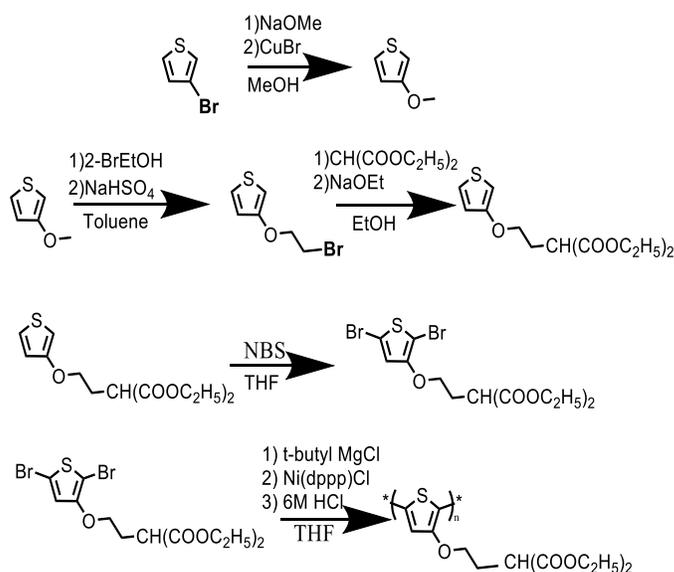


Figure 1. Possible hole-hopping routes from the KatG active site. Blue amino acids indicate possible starting points for hole-hopping routes, while red amino acids indicate possible continuations of each route.

Statement of Research Advisor:

Patrick Sahrman's research has helped to elucidate how redox enzymes protect themselves from inactivation by the highly reactive intermediates they must generate as part of the chemical reactions they catalyze. Redox enzymes are essential in every realm of biology, and the particular enzyme Patrick worked on is central to how pathogens like *Mycobacterium tuberculosis* defend themselves from host defenses

—*Douglas Goodwin, Biochemistry*

A shape-memory alloy-actuated hand exoskeleton

Sean Herrera, Michael Zabala

Many musculoskeletal disabilities restrict motor hand movement to a degree that limits day-to-day function. The purpose of my research was to create a biomechanical glove that aides in finger movement for those with such disabilities. This was achieved by heating and cooling of shape-memory alloy (SMA) wire laced into a finger of a glove. SMA wires have a unique ability in which they contract in length under a heated condition, and return to their original length under a cooled condition, producing a “shape memory” effect. The exoskeleton fingers can thus curl from wire contraction with resistive heating of the wires.

Two applications were tested with motion capture: (1) a 3D-printed finger and (2) a custom-designed biomechanical glove. Data were also collected from a human finger to serve as an anatomical standard. For each finger, the angle that the mid phalange makes with the proximal phalange was calculated as a function of time. Motion capture data were processed so that the flexion of the 3D-printed finger and the biomechanical glove could be compared to that of the human finger.

The time to reach full flexion for the human finger, the 3D-printed finger, and the glove-assisted finger was approximately 1.2 s, 2.1 s, and 1.85 s, respectively; the change in the mid phalange joint angle over this time was 33.2 degrees, 64.7 degrees, and 29.8 degrees, respectively (Figure 1). Data from the 3D-printed finger and the human finger both show increasing values over time, indicating that the SMA wire in the 3D-printed finger was successful in actuating finger movement in the correct direction. The shape of the human finger flexion curve was different from that of the 3D-printed finger (linear vs exponential). This might be a result of greater joint friction in the printed finger than in the human finger, thus creating irregularity in the rate at which the 3D-printed finger actuates. The glove’s flexion curve and the human finger’s flexion curve were very similar in shape and range. The glove-assisted finger flexed at a similar rate as did the human finger. The similarity of the human finger flexion and the glove-assisted flexion demonstrates the glove’s capability of actuating an impaired user’s finger to produce natural, human-like movement.

This study demonstrates the performance of a custom-designed biomechanical glove that assists finger flexion through the controlled heating and cooling of SMA laced into the ring finger. Kinematic data of the 3D printed finger validate use of SMA wires for such finger actuation. The data from the glove-assisted finger and the human finger demonstrate that the glove can produce human-like finger flexion on an immobile hand. Future studies involve increasing actuation speed, load-carrying capabilities, and implementing a feedback control system for position and speed control of the fingers of the glove.

Statement of Research Advisor:

Sean has designed, built, and tested a powered hand exoskeleton that is actuated with shape-memory alloy wire laced within a glove. His design has the potential to aid in hand movement rehabilitation and to provide strength augmentation to the user.

—Michael Zabala, Mechanical Engineering

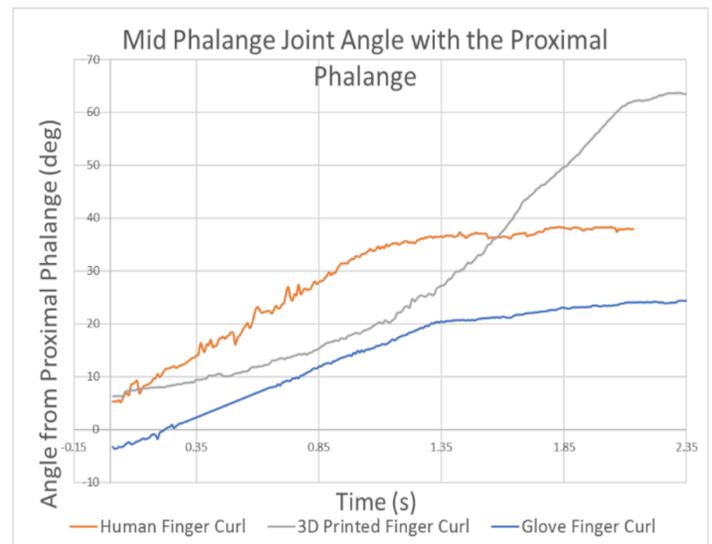


Figure 1. The joint angle that the mid phalange makes with the proximal phalange as a function of time during the flexion process of a human finger, 3D-printed finger, and glove-assisted finger.

Anaerobic digestion of food waste and poultry litter for biogas production

Matthew Preisser, Brendan Higgins

Millions of pounds of food waste are generated in the United States every year, and Auburn is no exception. This food waste burdens the environment by contributing to pollution associated with landfills and is an inefficient use of natural and economic resources. Alabama alone produces over 1.5 million pounds of poultry litter per year. Poultry litter contains harmful bacteria (e.g., salmonella) and high concentrations of nutrients that can contribute to water pollution. Currently, most of this litter is applied on the surface, but concerns about nutrient build-up in soils may soon limit this practice. A possible solution to divert poultry litter and other forms of food waste from the landfill is through the anaerobic digestion process. Anaerobic digestion has the potential to divert food waste and convert it into methane-rich biogas that can be used as a heat and electricity source.

The objective of this research project was to determine if locally sourced poultry litter and food waste are viable feedstocks for anaerobic digestion at Auburn University. It was hypothesized that both locally sourced poultry litter and food waste from on campus dining venues (the Village Dining) could produce biogas with high concentrations of methane, which can be used to generate heat. To test this, 160 mL batch reactors were loaded with varying amounts of inoculum containing methane-producing bacteria, water, and waste. They were then left in an incubator at 35°C for close to a month (Figure 1). Pressure readings were taken every two days to measure the gas buildup inside the reactors. In reactors loaded with a 1:1 ratio of waste to inoculum by volatile solids, a bio-methane potential (BMP) of 80.8 mL-CH₄/gram of food waste and 183.5 mL-CH₄/gram of poultry litter was achieved.

Through this research project, it was determined that a more efficient way to measure biogas production, other than measuring pressure changes, was needed in the laboratory. This is because the build-up of biogas (the metabolic end-points) in the headspace can inhibit further digestion. A more suitable approach is to

allow biogas to leave the reactor while measuring its flow rate. A low-flow gas measurement device that monitors water displacement over time was consequently constructed. This device uses a time of flight (ToF) infrared ranging sensor and Arduino, a commercially available microprocessor, to monitor six different bioreactors at a single time. A Matlab™ code was also developed to be able to convert water displacement into a volumetric flow rate. Future students in the lab will be able to calibrate this device and use it to monitor larger scale reactors.

Moving forward, there is a desire to use the anaerobic digestion process in conjunction with food waste generated by TigerDining. A closed-loop food cycle that converts uneaten and wasted food into heat for greenhouses on campus has the potential to reduce Auburn University's natural gas needs.

Statement of Research Advisor:

Matt has carried out a project on anaerobic digestion of poultry litter and food waste in satisfaction of the requirements of his Undergraduate Research Fellowship. This research holds the potential to advance the conversion of waste materials into renewable energy and organic fertilizer. Closer to home, Matt's project can have a transformative impact on food waste management practices at TigerDining on the AU campus. His results and development of a gas monitoring system have laid the groundwork for testing of fed-batch reactors operated over a long-time horizon.

—Brendan Higgins, Biosystems Engineering

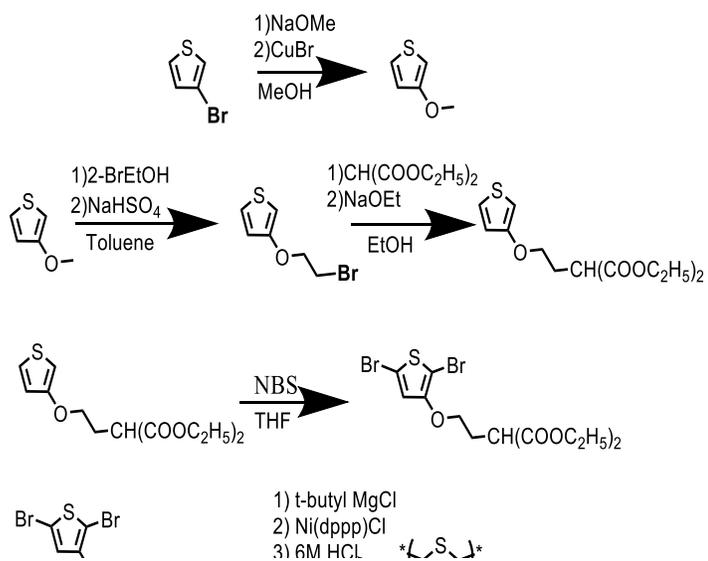


Figure 1: 160 mL bioreactor containing inoculum, waste, and water (left) placed in an incubator at 35°C (right).

The biofluid mechanics of expiratory central airway collapse in patients with chronic obstructive pulmonary disease

*Andrew Lenzie, Vrishank Raghav, *Surya Bhatt, M.D.*

Over 11 million people in the U.S. suffer from chronic obstructive pulmonary disease (COPD), the third leading cause of death. COPD is a progressive disease that irritates, inflames, and weakens the lungs. An excess of mucus is produced that obstructs the lungs and makes breathing difficult. Smoking is the primary cause of COPD; with no cure, much of the medicine is preventative [1]. Collapse of the central airway of greater than 50% has been associated with cigarette smoking and COPD. This condition has been named expiratory central airway collapse (ECAC), because the collapse occurs upon exhalation. This has been shown to result in an overall reduction of respiratory health of the COPD patient [2]. It is hypothesized that this collapse results in additional resistance to air flow and hence amenable to interventional therapy. The aim of this project was to develop a bench-top lung simulator to mimic the collapse of the central airway and measure the resistance to fluid flow.

The lung simulator was designed as a flow loop with water as the medium. Using water instead of air allows for the use of pumps, and the water can be measured more easily. A bladder pump allows for compressed air to pressurize the chamber within which it is held, thus allowing the bladder full of water to compress and push water through the loop. Therefore, the bladder pump allows for controlled beating similar to both cardiac and pulmonary rhythms. Water is held approximately three feet above the pump and flexible tubing models the central airway, thus providing a pressure head on the system. The reservoir allows for water to be pumped continuously in the piping of the system. The flexible tubing is necessary to model the collapse of the central airway. A box was designed to contain the tubing and to be air tight. Air is compressed into the box with a modified sphygmomanometer. The tubing within the box is collapsed by positive pressure. The bladder pump, reservoir, and chest cavity box are connected in series to complete the flow loop. The bladder pump is controlled by a

computer system and controller to provide successive compressions. This drives the water through the flexible tubing in the chest cavity box and up into the reservoir. Pressure and flow sensors have been installed and are being calibrated.

The flow loop design has been finalized, but sensor calibration is required to begin experimentation. Further investigation with the measurement of the changes to pressure and flow rate across the collapsed tubing (airway) compared to no collapse is necessary. This study will help physicians determine corrective measures by evaluating the effectiveness of interventional therapies, such as stenting of the trachea to stop the airway from collapsing. Future endeavors include patient specific 3D-printed airways from CT scans.

Statement of Research Advisor:

Andrew's research focused on understanding the bio-mechanics of the recently discovered problem of expiratory central airway collapse among smokers who are suffering from chronic obstructive pulmonary disorder. He worked towards developing a bench-top physiological analog to study the mechanics of central airway collapse.

—*Vrishank Raghav, Aerospace Engineering*

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**S.B. is a pulmonologist at the University of Alabama at Birmingham.*

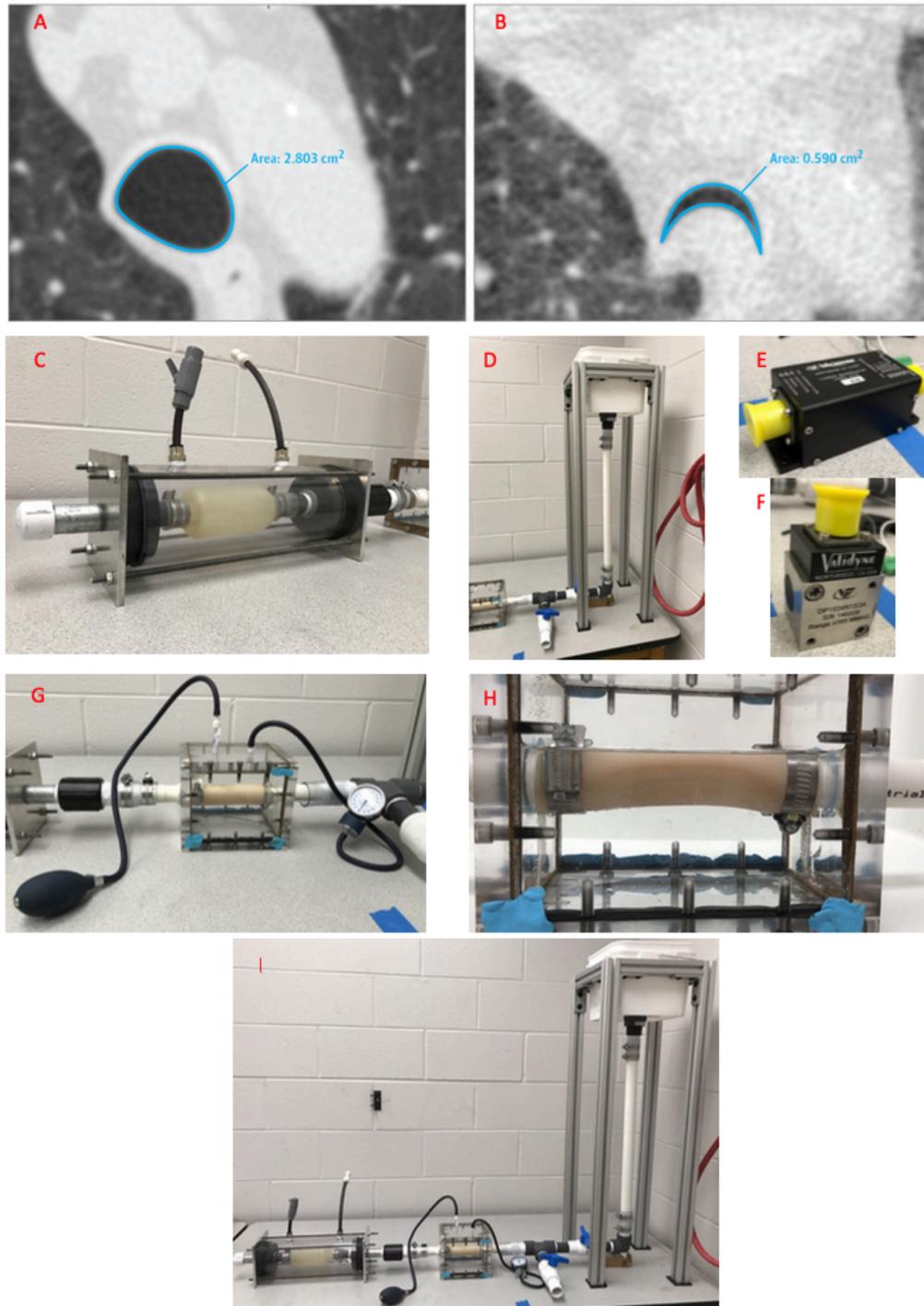


Figure 1. (A and B) CT images of ECAC patient (see reference [2]), (C) bladder pump, (D) reservoir, (E and F) pressure sensor system, (G) trachea box with modified sphygmomanometer, (H) collapsed tubing, (I) respiratory simulation flow loop.

Estimating fosa (*Cryptoprocta ferox*) population parameters in the rainforests of Madagascar

Cullen Anderson, Zach Farris, Sarah Zohdy

Understanding population parameters and dynamics of carnivores is vital to protecting these threatened taxa and the ecosystems they inhabit¹. Across the globe, carnivores are especially vulnerable to various anthropogenic pressures (e.g., deforestation, habitat fragmentation, introduced exotic carnivores) due to their relatively small populations and large home ranges¹. Madagascar's carnivores (Family Eupleridae) are no exception as they are among the least-studied and most-threatened carnivores in the world². Research is sparse on these mammals, and to date, no studies have been conducted on populations occupying one of Madagascar's most important protected regions: the Andasibe-Mantadia landscape.

We conducted the first survey and estimation of carnivore populations in this area using photographic sampling across three protected forests (Analamazaotra, Mitsinjo, VOI). We focused on fosa (*Cryptoprocta ferox*), Madagascar's largest and most widespread carnivore and an umbrella species for this threatened region, meaning protection of this top carnivore can result in the protection of a wealth of biodiversity occupying the same habitat^{3,4}. Using the program Presence™, we estimated naïve occupancy (occupancy not accounting for probability of detection) and detection. We investigated variables influencing these parameters, including human detection and human-altered environments (e.g., villages, roads, forest edge).

With 24 camera stations, 19 sampling days, and 362 trap nights, we found fosa trap success was 1.29, with 5 captures and a naïve occupancy estimate of 0.22. These figures do not refer to numbers of fosa; rather, they are indicative of fosa detectability and prevalence within the study area. Due to a limited study period and decreased winter activity, our models were unable to estimate true occupancy (e.g., accounting for imperfect detection), which is likely much higher than naïve occupancy indicates. However, our results still indicate fosa are negatively affected by humans and human-altered environments (Figure 1). Distance to forest edge best explains fosa detection

($\Delta AIC=0.00$, AIC Weight=0.552), but fosa detection displayed a negative relationship with distance to road, distance to anthropogenic area, and human detection as well. These factors also likely threaten other endemic wildlife, such as endangered lemurs, in this region.

We are currently analyzing data from a more expansive survey conducted across these sites during warmer months (October-November 2017) to compare estimates across seasons. With the longer survey during Madagascar's summer, we anticipate fosa trap success and detection will increase, and our models will be able to estimate occupancy. Further research is needed to provide such estimates for other elusive native carnivores in the region.

Fosa are one of the least-studied carnivore species in the world, and their vulnerable status and umbrella species role makes studying and managing them particularly important^{2,4}. This study's findings have direct implications for management relating to this tourist-driven conservation area. Having more robust, reliable estimates of fosa populations in this region will allow Madagascar National Parks and local non-governmental organizations (e.g., Association Mitsinjo) to make more informed conservation decisions for threatened carnivores. Additionally, this information will provide insight on relationships with local people, their behaviors, and their domestic animals, all of which pose serious risk to Madagascar's imperiled wildlife.

Statement of Research Advisor

For his research project, Cullen conducted the first robust survey and estimation of the native and exotic carnivore populations across the Andasibe-Mantadia region of Madagascar, thus providing the government of Madagascar (Madagascar National Parks) and local NGO's (Association Mitsinjo) with critical population estimates and a thorough evaluation of the impacts of anthropogenic pressures on native, threatened wildlife.

He is currently working to disseminate these important findings via publications and professional presentations at national and international meetings.

—Zach Farris and Sarah Zohdy, School of Forestry and Wildlife Science

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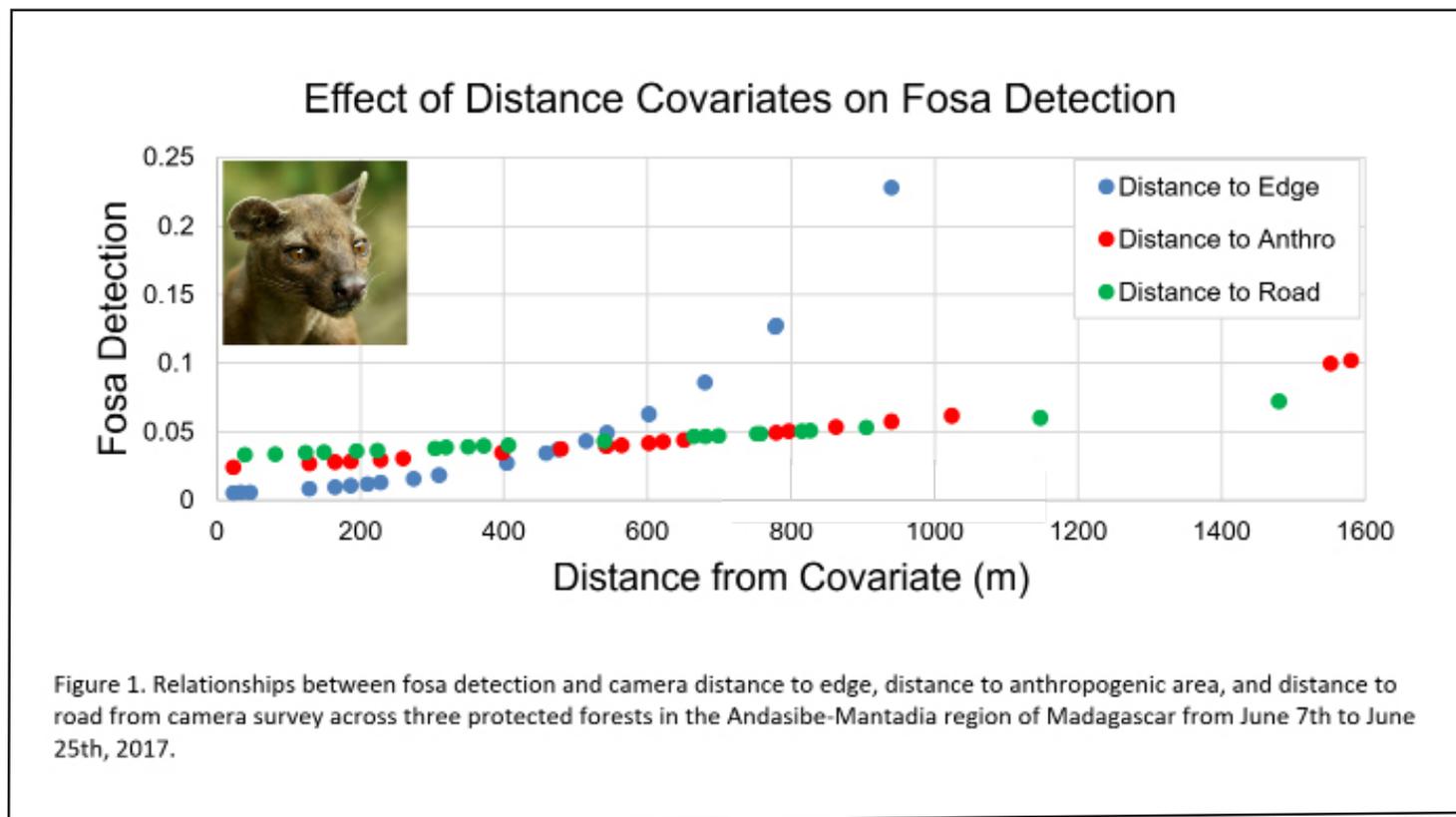
¹Weber, W., & Rabinowitz, A. (1996). A Global Perspective on Large Carnivore Conservation. *Conservation Biology*, 10(4), 1046-1054.

²Brooke, Z. M., Bielby, J., Nambiar, K., & Carbone, C. (2014). Correlates of Research Effort in Carnivores: Body Size, Range Size and Diet Matter. *PLoS ONE*, 9(4).

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Manufacturing of nanoparticle reinforced materials through electrospinning

Kiana LaBombard, Sabit Adanur

Electrospinning is a process of manufacturing nano-fibrous webs using an electric force to transport threads of a polymer solution. Polyvinyl Alcohol (PVA) was chosen as the base polymer for all testing purposes. PVA is known for having high tensile strength, but the tensile strength is decreased by high humidity. To regulate humidity, all tests were performed inside a sealed box with a dehumidifier.

Two different materials were chosen as additives to the PVA: carbon nanotubes (CNTs) and nanoclay. CNTs have high modulus, elasticity, and strength. CNTs are also effective electrical and thermal conductors. Nanoclay is not easily dispersed throughout polymer mixtures, but once it is evenly dispersed, it has been shown to improve mechanical strength, barrier properties, and flame-resistance.

Three separate experiments were conducted for this study:

1. PVA with no additives (control)
2. PVA with carbon nanotubes
3. PVA with nanoclay

For each experiment, 15 percent by weight (wt%) of PVA was mixed with distilled water and was heated for four hours at 100 °C. For PVA with CNTs, less than 1 wt% CNTs was added to the PVA/water mixture. For PVA with nanoclay, 5 wt% nanoclay was added to the PVA/water mixture. Once the mixture was finished heating, it was cooled and electrospun.

Scanning Electron Microscopy (SEM) was employed to visualize the individual fibers of the electrospun webs. PVA with no additives has fibers that are very sporadic, going in different directions and having varied diameters and connection points. Unlike PVA with no additives, PVA with CNT fibers have a more uniform structure, holding the shape and diameter of each fiber. PVA with nanoclay has the most sporadic fibers, with stacking and changing directions and diameters of the fibers.

Three individual tests/measurements were performed on the samples of the electrospun webs: tensile strength, thickness, and air permeability. From the tests/measurements, it was observed that PVA with no additives has the highest tensile strength and elastic modulus, PVA with CNTs has the best air-permeability, and PVA with nanoclay has the greatest thickness and average force.

Electrospun webs are becoming more popular because of their ability to be used in filtration, textile manufacturing, medical technologies, composite materials, and chemical catalysts. Depending on the application of the electrospun web, PVA with CNTs or nanoclay exhibit improved performance. Future work is needed to explore alternative polymers and additive configurations that were not discussed here.

Statement of Research Advisor:

Kiana has resurrected the research in electrospinning after a disruption due to the move of the Polymer and Fiber department. She obtained good results and presented them in the Fiber Society international conference in Athens, GA in Fall 2017.

—*Sabit Adanur, Polymer and Fiber Engineering*

New structural and electronic insights on supported $(VO_x)_m/(M_1O_y)_n/(M_2O_z)_{bulk}$ catalysts for the oxidative dehydrogenation (ODH) of light hydrocarbons

Kaitlyn Lawrence, Natalie Stephens, Jorge Moncada, Raj Thakur, Carlos A. Carrero

A heightened demand for commodity chemicals such as propylene and butylene paired with recent advances in hydraulic fracturing has motivated industry to turn to a domestic production feedstock: shale gas. Traditionally, C₃-C₄ olefins were produced as byproducts using steam cracking of naphtha, an expensive, energy-intensive route. Therefore, as demand for olefins continues to soar, researchers have investigated other pathways of on-purpose production of C₃-C₄ olefins.

One pathway that has been extensively studied in recent years is catalytic oxidative dehydrogenation, or ODH. The reaction is proven to run with relatively high yields of desired product even with relatively low energy inputs. Group V metals perform well in ODH due to their unique chemical nature, and supported vanadium oxide catalysts have proven a viable option of the Group V metals due to their low cost and high performance. [1] It has been found that synergistic effects in supported multi-component metal oxides enhance olefin productivity in ODH of light hydrocarbons. [2,3] The goal of this project was to synthesize a library of ternary supported metal oxide catalysts, all of which utilize vanadium as a main component $(VO_x)_m-(M_2O_y)_n/(M_3O_z)_{bulk}$, while varying the M₂ and M₃ metals (Si, Ti, Zr, Al, Ce, Nb). Supported catalysts utilize an inert support material to which the catalytic active sites are anchored, whereas bulk catalysts are composed of a uniform catalyst through the solid particle. Support is often used to lower the cost of catalysts while maintaining a high number of active sites.

In this project, the ratio of V:M₂ metals was tuned with the goal of achieving monolayer coverage, a term used to describe the two-dimensional dispersion of MO_x active sites on the outer layer of the support. These samples were characterized by Brunauer-Emmett-Teller surface area tests (BET), inductively coupled plasma atomic emission spectroscopy (ICP-AES), Raman spectroscopy (Figure 1), and H₂-TPR (Figure 2).

The catalysts were then tested catalytically in a custom reactor set up. We performed the ODH of isobutane to produce isobutylene (Figure 3).

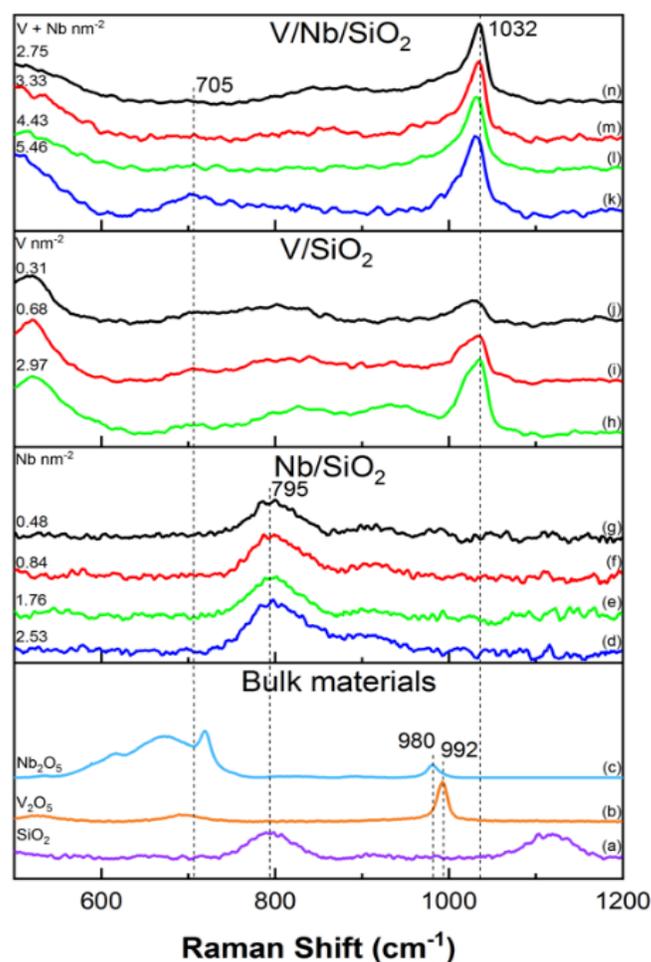


Figure 1: *In situ* Raman spectra for bulk, supported binary and ternary metal oxides. Metal coverage obtained from BET and ICP is labeled along its correspondent Raman spectra.

The final goal was to establish structure-reactivity relationships for the design of more selective, stable and active catalysts for future ODH experiments. The catalytic activity of supported binary $(M_2O_y)_n/(M_1O_x)_m$ and ternary $(M_3O_z)_{bulk}-(M_2O_y)_n/(M_1O_x)_m$ metal oxide catalysts essentially depends on the presence of two-and

three-dimensional M_2O_y and M_3O_z species, the $M_3:M_2$ ratio (coverage of each component), and their spatial relation. We found that the extent of synergistic effects strongly depends on these same factors. Our study shows that the extent of synergistic effects is greater on reduced metal oxides. This is because we cannot reduce all V_{5+} and Nb_{5+} sites to V_{3+} and Nb_{3+} under our reaction conditions and therefore, metal cations (V_{5+} , V_{4+} , V_{3+} , Nb_{5+} , Nb_{4+} , Nb_{3+}) at different amounts and oxidation states are present on the catalyst's surface. The formation of these M_{n+} species depends on both the reaction conditions (e.g., temperature, residence time) and the reducing agent (e.g., hydrogen, hydrocarbon).

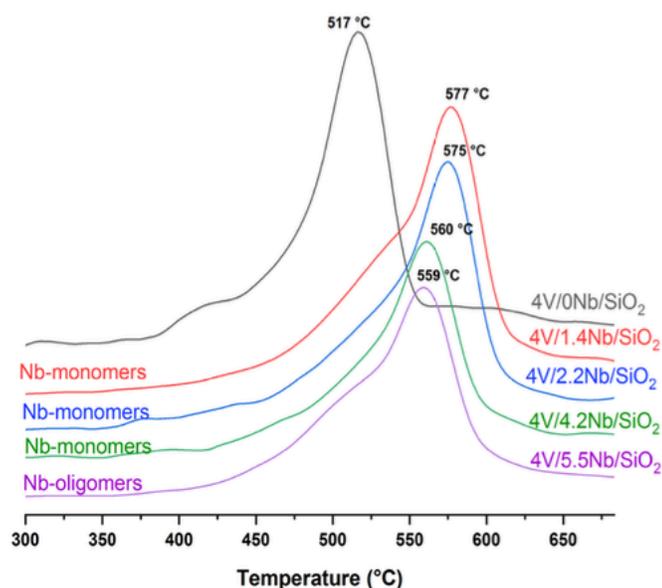


Figure 2: H_2 TPR profiles for VNb/SiO₂ catalysts. Explicit indication for samples containing Nb monomers and oligomers are shown on the left side of each curve.

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Statement of Research Advisor:

Kaitlyn has been learning and developing new strategies to prepare supported metal oxide catalysts for the upgrading of natural gas. During the last year, Kaitlyn has determined the most important variables to control and tune the desired molecular species needed to perform specific catalytic reactions. Currently, we are both compiling these important results in a peer-reviewed journal (ChemCatChem) and using them to continue our research in selective oxidation catalysis for light hydrocarbons.

—Carlos A. Carrero, *Chemical Engineering*

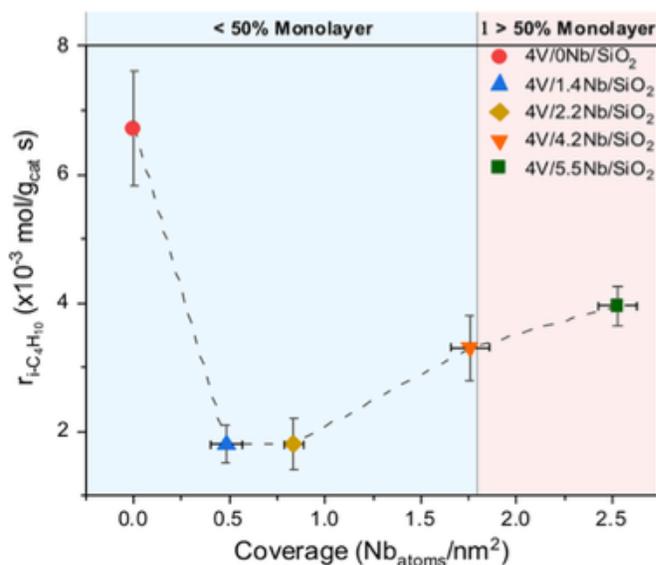


Figure 3: Isobutane consumption rate as a function of Nb coverage at 420 °C. Catalyst mass: 30-40 mg. Total flow: 60 sccm.

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The engineer in me

Njeri Bennett, Christine Schnittka

This project was aimed at investigating the impact of science, technology, engineering, and math (STEM) curricula centered on spatial thinking skills, science content knowledge, and attitudes towards science and engineering in low socioeconomic (SES) areas. This research topic is important because students of color from lower SES areas may be less likely to get exposed to STEM before they step foot into a college classroom. In this project, 47 students of color, ranging from kindergarten to twelfth grade, were exposed to an interactive and fun way to experience science and engineering activities. My goal was to help them translate their experience as a student of color to subjects that they find challenging, like math and science. As a major in Industrial and Systems Engineering with a minor in Africana Studies, I wanted to link the unique identity of Black inventors to science to show the direct impact that racial representation has in STEM fields.

In the first task, each student was asked to “Draw a Scientist/Engineer” and given a form to fill out with supporting information about their character. This form asked for the character’s personal information, work setting, job description, and a description of what the engineer/scientist is doing in the drawing. Each student also received a brown engineering journal that they used each week to draw project designs, parts, materials, spatial thinking tools, and people. For the first few weeks, I introduced spatial thinking activities that challenged each student’s orthographic skills.

The next phase of the project focused on introducing scientists and engineers from marginalized groups like women or African Americans. In this exercise, I read a description of a scientist or engineer and each student would draw the person that I was describing. Afterwards, I would do an activity related to the scientist or engineer that we focused on for that day. For the last two weeks of the project, I exposed the students to more spatial thinking games and activities.

Although the initial reason for this project was to expose students of color to curriculum centered on spatial thinking skills and science content knowledge, I wish to examine the participants’ attitudes towards

math, science and engineering in future work. In addition, I feel that my identity as a Black, female engineering student at Auburn University served as a catalyst to my research project. From this experience, I plan to create a STEM curriculum that will expose students from underrepresented groups to the intersecting identities of Black and Brown people in the engineering community. My hope is that this curriculum will increase the interest in engineering as well as love for math and science during primary and secondary education.

Statement of Research Advisor:

My research is on engineering design-based curriculum development for youth, with a particular emphasis on developing spatial thinking skills. Njeri was key to my work during the fall of 2017, as she participated in curriculum development, curriculum implementation, and data collection and analysis. Her assistance was invaluable.

— *Christine Schnittka, Curriculum and Teaching*

Characterization of mitochondrial variation between oocytes harvested from lean and obese pigs

Madison Gohlke, Paul Dyce, Terry Brandebourg

The overarching goal of our work is to study the influence of mitochondrial-nuclear crosstalk on development of obesity and metabolic disease, and specifically, whether the mitochondrial genome plays a significant role in obese phenotypes and whether this role can be manipulated to control the development of metabolic disease. However, underlying this hypothesis is the assumption that there is indeed mitochondrial variation between lean and obese individuals and that the resultant metabolic phenotype is not merely driven by environmental effects such as an unhealthy diet. My research question involved examining differences in mitochondria between oocytes collected from genetically lean and obese pigs to justify the pursuit of mitochondrial manipulation as a means to circumvent metabolic disease.

Experimental models were a Yorkshire pig (a lean, selectively bred, meat-breed pig) and a Mangalitsa pig (an obese, genetically unimproved, lard-type pig). Oocytes were used as the tissue of choice because current efforts in mitochondrial manipulation indicate intervention in the embryonic stage; additionally, paternal mitochondria are excluded from the fertilized embryo. Ovaries were collected from both animals at the time of commercial harvest. Oocytes were manually aspirated and placed in media. Cumulus-oocyte complexes (an oocyte and the surrounding cumulus cells that provide it with support and signaling) were morphologically graded to predict maturation and fertilization potential. Oocytes that have more than five complete layers of cumulus cells (grade A) are expected to be more successful at maturation and fertilization. Oocytes were then stained with brilliant cresyl blue (BCB) to predict general developmental competence. Oocytes that are BCB positive have higher developmental competence. MitoView™ Green, a fluorescent mitochondrial stain, was then added to visualize the mitochondria themselves. Images of each oocyte were captured and assessed using ImageJ®, an image-analyzing software. Finally, oocytes were denuded of cumulus cells and stained with Nile Red, a fluorescent stain that binds cellular lipids, to visualize lipid content of the cells. Images were again captured and analyzed.

Oocyte grading showed that a larger percentage of Mangalitsa than Yorkshire oocytes were grade A, meaning that Mangalitsa oocytes had higher maturation and fertilization potential. BCB staining showed that a larger percentage of Mangalitsa than Yorkshire oocytes were BCB positive, suggesting that Mangalitsa oocytes were more developmentally competent. Mitochondrial staining showed that Yorkshire oocytes had stronger fluorescent signals (implying higher numbers of mitochondria) than Mangalitsa oocytes. Interestingly, BCB positive oocytes also had higher numbers of mitochondria than BCB negative oocytes regardless of breed, indicating that developmentally competent oocytes have more mitochondria, whether genetically lean or obese. Finally, Nile Red staining showed that Mangalitsa oocytes had higher overall lipid content as well as a higher ratio of polar to neutral lipids within individual oocytes. Neutral lipids are associated with fat storage within a cell, whereas polar lipids are associated with structural components.

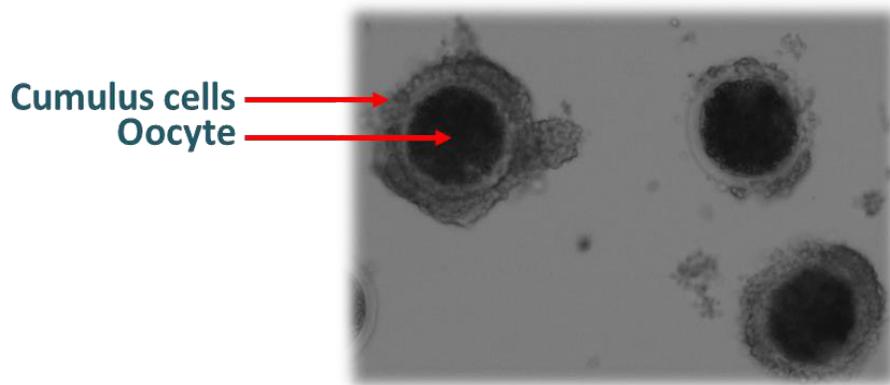
Taken together, these data are consistent with the hypothesis that there is mitochondrial variation between lean and obese pig oocytes and that mitochondrial manipulation may indeed prove a worthwhile strategy to combat metabolic disease.

Statement of Research Advisor:

Maddy conducted an important pilot study to verify that differences in oocyte mitochondria exist between lean and obese pigs. Our overall aim is to examine the role of mitochondrial in obesity-induced metabolic disease. To accomplish this, we are pursuing a mitochondrial-nuclear DNA exchange strategy whereby mitochondria (and thus the inherent mitochondrial DNA) will be switched between embryos of lean and obese pigs. Maddy's results suggest that differences do exist between the mitochondria of Yorkshire (lean) and Mangalitsa (obese) pigs. Maddy's data are consistent with her hypothesis that disparities in mitochondria account in part for the divergent metabolic phenotypes exhibited across breeds. In doing so, Maddy has made an impressive and substantial contribution to the lab as Maddy's work provides critical support for further

pursuing mitochondrial nuclear exchange between Mangalitsa and Yorkshire pigs.

—Terry Brandebourg, Animal Sciences



Oocyte Grade	Layers of Cumulus Cells
Grade A	≥5 layers
Grade B	2-4 layers
Grade C	≤1 layer

Figure 1. Photomicrograph of cumulus-oocyte complexes (an oocyte and the surrounding cumulus cells that provide it with support and signaling) and table detailing grading criteria. Cumulus-oocyte complexes were morphologically graded to predict maturation and fertilization potential. Oocytes that have more than five complete layers of cumulus cells (grade A) are expected to be more successful at maturation and fertilization.

The potential role of inflammation in oocyte health and developmental potential

Sara A. Gorman, Casey C. Read, Kaitlyn M. Phillips, Terry D. Brandebourg, and Paul W. Dyce

This research investigated the relationship between inflammation and the developmental potential of bovine oocytes. Cumulus granulosa cells play a critical role in supporting the developing oocyte during its development. Cumulus granulosa cells have previously been shown to have a pro-inflammatory immune response signaling through Toll-like receptor-4 (TLR-4). In order to investigate potential inflammatory effects on oocyte development, we utilized a single cumulus-oocyte-complex (COC) *in vitro* maturation, fertilization and culture system. This system allowed us to collect cumulus granulosa cell samples and retrospectively separate them into low developmental potential (LDP) or high developmental potential (HDP) groups. Samples were labeled as LDP if they stalled at the 2-cell stage and HDP if they reached the blastocyst stage, following fertilization and development. We then compared the expression of proinflammatory cytokines, including tumor necrosis factor alpha (TNF α), interleukin 6 (IL-6), C-X-C chemokine 5 (CXC-5), and monocyte chemoattractant protein 1 (MCP1). Also, in order to investigate the ability of granulosa cells to respond to pathogens, we isolated granulosa cells and treated them with various concentrations of lipopolysaccharide (LPS). LPS is known to trigger an immune response in responsive cells through upregulating the expression of pro-inflammatory cytokines. We found cumulus cells in the HDP group had lower expression levels of IL-6 (0.52 ± 0.45 vs. 6.68 ± 3.80) and TNF α (0.84 ± 0.18 vs. 2.58 ± 0.37) when compared to the LDP group. Furthermore, LPS treatment of granulosa cells tended to increase the expression of IL-6, TNF α , and MCP1.

This research is currently ongoing to confirm the observed response. These results support the hypothesis that granulosa cells can respond to LPS, via the upregulation of pro-inflammatory cytokines, causing a potentially negative effect on bovine oocyte developmental competence. A limited number of samples were run concerning the LPS treated cells, thus making the results suggestive, not conclusive. Further research into the specific mechanisms continue.

Statement of Research Advisor:

Inflammation has long been known to negatively affect fertility outcomes, though the mechanism remains unexplained. Sara's work helped solidify a potential relationship between oocyte developmental competence and cumulus granulosa inflammatory cytokine expression.

—Paul Dyce, Animal Sciences

Evaluation of açai and maca extracts for CYP3A4 enzyme induction

Elizabeth Lopez, Yilue Zhang, Da Jung, Kodye Abbott, Satyanarayana R. Pondugula, Angela I. Calderon

The most abundant liver cytochrome P450 isoform is CYP3A4, an enzyme responsible for the metabolism of at least 60% of approved drugs. Notably, most anticancer agents are metabolized by CYP3A4. Increased consumption of botanical dietary supplements (BDS) has been observed to increase CYP induction and metabolism, and therefore decrease efficacy of drugs like anticancer agents. The purpose of this investigation was to establish the degree of CYP3A4 induction caused by two extracts commonly used in BDS by cancer patients and their potential for botanical-drug interactions.

Maca root (*Lepidium meyenii* Walp) has historically grown in the Andes Mountains and has been consumed as part of the local diet. Recent research suggests that maca root has anti-inflammatory and cancer cell growth inhibitory properties. Açai (*Euterpe oleracea* Mart) is a berry-producing palm grown across South America. Medicinal use of açai has gained popularity in North America due to its antioxidant activity. Current research has suggested that phenolic acid and other antioxidants in açai have antiproliferative activity on cancer cells. Both maca and açai were investigated for CYP3A4 induction potential and effects on the efficacy and toxicity of anticancer agents in the liver.

Methanol and acidic methanol extracts of maca and açai were prepared and subject to parallel artificial membrane permeability assay (PAMPA). PAMPA is an *in vitro* model for simulating the intestinal absorption of bioavailable compounds via passive diffusion. The donor site of its 96-well plate simulates intestinal content pre-absorption and non-passively diffused compounds post-absorption, while the acceptor site simulates the compounds that passively diffuse the intestinal membrane. PAMPA was used to separate the passively diffused CYP-inducing constituents of açai and maca in the acceptor site from non-passively diffused CYP3A4-inducing constituents in the donor site. These extracts were then subjected to investigation on human hepatocyte cells to determine the extent of CYP3A4 mRNA expression.

Initially, CYP-induction from açai and maca were added to the PAMPA donor site at concentrations of 1.5 µg/µL and 0.75 µg/µL in buffer and incubated at 37°C for five hours. Then, samples from the PAMPA donor and acceptor sites were applied to hepatocyte cell lines and incubated for 22 hours in the initial induction assay. Hepatocytes treated with both donor and acceptor site extract samples underwent RNA extraction and qPCR analysis to determine mRNA expression of CYP3A4. Only açai extracts significantly induced CYP3A4 compared to the DMSO control. Cytotoxicity was noticed in açai methanol and acidic methanol at 1.5 µg/µL treatments after 20 hours' incubation.

When both extracts were subjected to PAMPA, passively diffused compounds in açai methanol extract at 1.5 µg/µL showed significant CYP3A4 induction in hepatocytes after PAMPA assay, while maca extracts exhibited inhibitory activities. Finally, induction potential for non-passively diffused and passively diffused compounds in both açai extracts were compared. Higher hepatic CYP3A4 induction was found in liver cells treated with non-passively diffused constituents in the donor site. This finding suggests that other mechanisms such as ion channels and active transporters are involved in CYP3A4-induced intestinal transportation. In short, these results suggest that açai has the potential to produce botanical-anticancer drugs interactions by the mechanism of CYP3A4 induction.

For future investigation, elucidation of the chemical profile of the compounds that pass the PAMPA membrane and induce CYP3A4 is being considered. Specifically, identifying these compounds will require analysis via liquid chromatography-mass spectroscopy and Mass Profiler software. These data will be compared to data regarding CYP3A4 inhibition in human liver microsomes due to maca and açai extracts. The experiment may be repeated using manufactured maca and açai supplements to investigate the efficacy of these findings in mass produced botanical products available to the public consumer as compared to laboratory-prepared extracts.

Statement of Research Advisor:

Betty was trained on the CYP3A4 inhibition and induction assays to carry out the project and learned how to review and analyze papers from the literature about CYP3A4 inhibition and induction and about biological activity and chemistry of maca and açai. Her study was focused on the assessment of the interference of liver metabolic enzymes by two top botanicals in the U.S. market of dietary supplements and their potential to produce interactions with drugs.

—*Angela I. Calderón, Harrison School of Pharmacy*

Development of an *in vitro* model to evaluate novel genes regulating inflammation and fibrosis

Peyton Kuhlers, Michael Greene

Non-alcoholic steatohepatitis (NASH) is a condition in which fat accumulates in the liver and is accompanied by inflammation. This condition can eventually lead to liver fibrosis, cirrhosis, and hepatocellular carcinoma. The goal of this project was to use the CRISPR-Cas9 system to develop an *in vitro* model capable of identifying novel genes that regulate the development of inflammation and fibrosis in the context of NASH. The mechanism of the disease is not entirely understood, and the CRISPR-Cas9 system has not been used in this context.

To validate our *in vitro* NASH model, AML-12 mouse liver cells were treated with either 0.4 mM palmitic acid (PA), 25 ng/ml TNF- α , or hypoxia for 24 hours to induce Collagen I expression, which was assessed by western blot. Collagen I is a marker for fibrosis and NASH. Next, the AML-12 cells were infected with a plasmid containing the gene for the Cas9 enzyme. This plasmid also included resistance to the antibiotic blasticidin, which allowed for selection of stable AML-12 cells expressing Cas9. Multiple single cell colonies were cultured and Cas9 expression was determined by western blot (Figure 1A). The Cas9 expressing AML-12 cells were then treated with the same experimental NASH conditions and Collagen I expression was analyzed by western blot.

As shown in Figure 1, both regular AML-12 and Cas9 expressing AML-12 cells exhibited significant elevations in Collagen I expression, normalized to the expression of GAPDH, when treated with palmitic acid (2.4 ± 0.2 -fold and 3.3 ± 1.5 -fold, respectively), but not when treated with TNF α (0.6 ± 0.2 -fold and 0.9 ± 0.2 -fold, respectively). We observed that there was not a significant difference in Collagen I expression between the two cell types (Figure 1). These results verify that AML-12 cells and our Cas9 expressing AML-12 cells can be used as a model for NASH and fibrosis.

The next step for our project is to utilize the CRISPR-Cas9 system to mutate the gene *TGFBR2*, which

has a known role in the development of NASH. We hypothesize that by mutating this gene we will see a reduction in Collagen I expression. The final step will be to use the CRISPR-Cas9 system to mutate genes that were found to be differentially expressed in the livers of mice with NASH. The reduction in Collagen I expression found from knocking down *TGFBR2* will be used as a benchmark to assess the role that these novel genes may have in the development of NASH.

Statement of Research Advisor:

Peyton has performed the key experiments necessary to validate our new *in vitro* model of NASH. Our Cas9 expressing cells will be used to screen for novel regulators of NASH. Our novel model may transform the manner in which new targets for NASH are screened —*Michael Greene, Nutrition, Dietetics, & Hospitality Management*

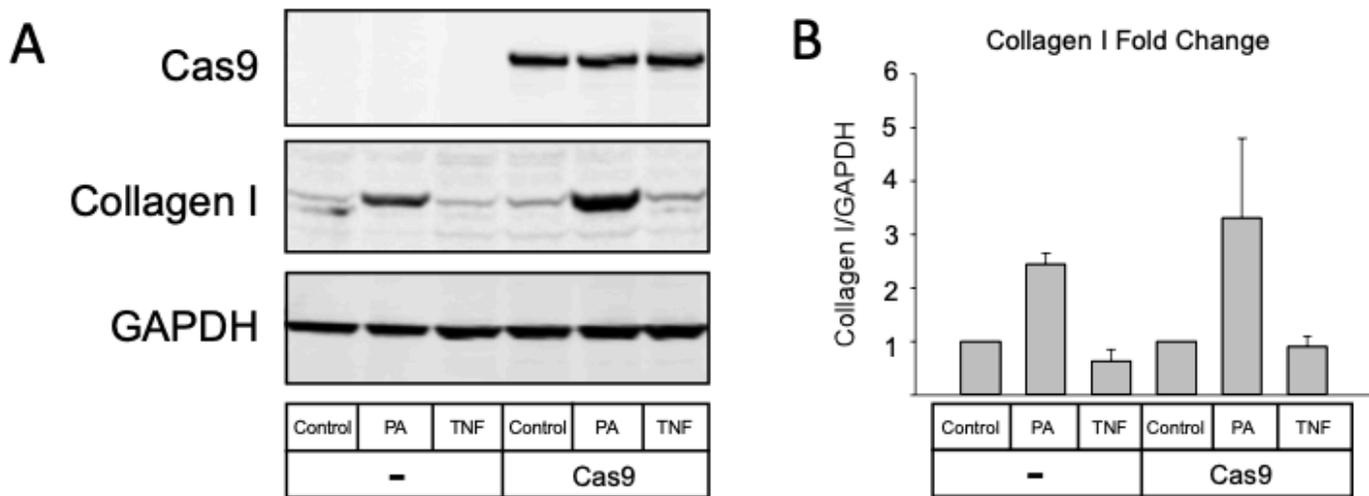


Figure 1. Induction of Collagen I expression in AML-12 cells. **A.** Representative images of Cas9, Collagen I and GAPDH Western blots. **B.** Quantitation (mean \pm standard error) of Collagen I expression from 3 independent treatments.

Quantifying the mammal diversity of the Louise Kreher Forest Ecology Preserve

Autumn Patterson, Cullen Anderson, Christopher A. Lepczyk

Trail cameras are a cost-effective tool that minimizes the impact of human disturbance while collecting large amounts of data (Figure 1). This lack of disturbance increases the possibility of detecting animal species and observing their behavior, which has made trail cameras a popular tool for assessing biodiversity throughout the world. Given the lack of knowledge about mammals in urban areas throughout the southeast United States, our goal was to inventory mammal diversity in six urban green spaces of Auburn, Alabama.

In this study, we set out 21 camera traps in five of the six urban green spaces in Auburn, Alabama, between fall 2017 and spring 2018. Currently, data from one of the five locations, the Louise Kreher Forest Ecology Preserve, has been fully analyzed. Cameras were placed approximately 0.3 m above ground at trail intersections in nine locations of the preserve, with each camera being about 0.16 km apart from one another. Each camera was set to take pictures at three-photo intervals, every four minutes, and cameras were checked every four weeks. When analyzing the images, we identified each animal to its species, and then counted the number of each species present in all images. To date, 17,839 images have been collected over 819 trap nights, with 954 sightings of mammals.

Image analysis resulted in the identification of 13 terrestrial mammals in the Louise Kreher Forest Ecology Preserve using Alabama Wildlife, Volume One for reference (Mirarchi 2004). The identified species can be found below (Table 1).

The number of mammals observed is nearly 20% of the known mammal diversity of the state (68 species). Notably, one camera malfunctioned, resulting in a loss of two weeks of data.

Our next step is to collect and analyze images from five other greenspaces in Auburn, Alabama. In addition, the next phase of research will use bait traps to increase the likelihood of observing a variety of small mammals and will result in a broader representation

of the mammals in Auburn. Upon completion, the data collected will be available for residents and management organizations and will offer a detailed understanding of how Auburn's mammalian community vary across the city based on environmental attributes.

Statement of Research Advisor:

Autumn carried out an important assessment of mammal diversity in Auburn that contributes knowledge about where animals live and how they behave in urban areas. This initial research will provide a baseline on which to build a broader understanding of wildlife in and around Auburn and other cities of the southeast United States.

—Christopher A. Lepczyk, School of Forestry and Wildlife Sciences

References:

Mirarchi, Ralph E., ed. (2004). *Alabama Wildlife, Volume One. A Checklist of Vertebrates and Selected Invertebrates: Aquatic Mollusks, Fishes, Amphibians, Reptiles, Birds and Mammals*. The University of Alabama Press, Tuscaloosa, AL., 209 pp.

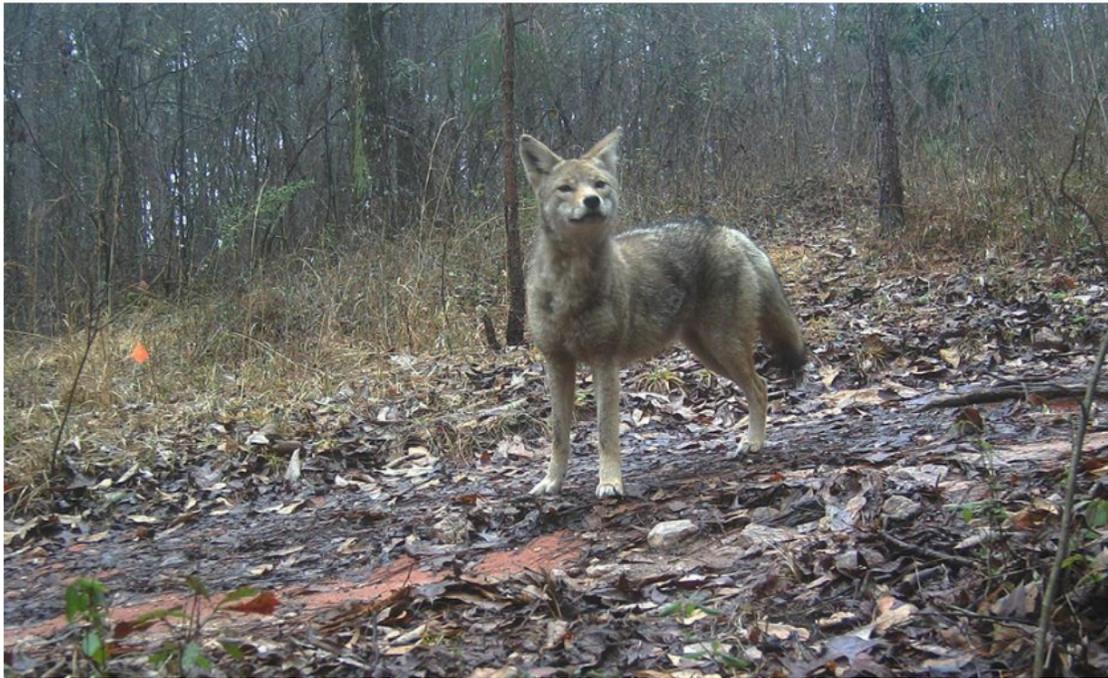


Figure 1. Image of coyote (*Canis latrans*) captured by a game camera.

Table 1. Alphabetized list of terrestrial mammals identified in the Louise Kreher Forest Ecology Preserve.

Scientific Name	Common Name
<i>Canis latrans</i>	Coyote
<i>Canis lupus familiaris</i>	Domestic Dog
<i>Dasyopus novemcinctus</i>	Nine-banded armadillo
<i>Didelphis virginiana</i>	Virginia opossum
<i>Felis catus</i>	Domestic cat
<i>Lynx rufus</i>	Bobcat
<i>Odocoileus virginianus</i>	White-tailed deer
<i>Procyon lotor</i>	Raccoon
<i>Sciurus carolinensis</i>	Eastern gray squirrel
<i>Sylvilagus floridanus</i>	Eastern cottontail
<i>Tamias striatus</i>	Eastern chipmunk
<i>Urocyon cinereoargenteus</i>	Gray fox
<i>Vulpes vulpes</i>	Red fox

Effect of rumen-protected methionine on beef cow-calf pairs performance during pre-and post-calving

Betsy Brown, Soren Rodning, Gastón Alfaro, Sonia Moisés

Methionine is one of the first limiting amino acids in cattle [1]. A limiting amino acid is the one that is found in shortest supply in relation to the nutritional requirements needed by the animal. By being one of the first limiting amino acids, methionine is supplemented because of its relatively short supply in the diet of the animal [1]. In all ruminant species, nutrients provided by the diet are utilized by the microorganisms present in the rumen. The product of the fermentation of those nutrients by the microorganisms reaches the small intestine, where it can be absorbed and used by the ruminant. By using a rumen-protected methionine (RPM), the cow has more available methionine reaching the small intestine, which will enhance the animal's performance for milk production and fetal development. Supplementing RPM in dairy cows has yielded an increase in animal performance [2], milk-fat percentage [3] and immune-metabolic status during the peri-partal period [4]. By implementing the same technique in beef cattle, we hypothesize that RPM supplemented beef calves borne from cows supplemented with RPM during the last 90 days of gestation will have increased muscle growth and daily body weight gain. If this hypothesis is supported, cattle farmers may be able to increase their calves' ability to gain weight without using artificial growth factors, such as hormones, while still increasing profits. Furthermore, improving animal performance will lead to more animals to be used for beef production and thus benefit society.

The specific purpose of the research project is to evaluate the effect of RPM during late gestation in the performance of cows in winter and fall calving seasons and determine how this maternal supplementation of RPM affects the offspring's growth before weaning (through the cow's milk) and after weaning by RPM supplementation to the offspring for 100 days. For this study, 22 cows were used for both the winter and fall calving season. From these two groups, 11 cows received RPM for 90 days before calving and the other 11 cows were used as a control.

During the testing and observation of the calves and cows at birth, the RPM calves weighed heavier by a difference of ~11 lbs. more than the control calves ($P = 0.02$) (Table 1). Furthermore, the calves given the RPM supplementation had an increased average daily gain (ADG) over the control group between birth until weaning ($P = 0.03$) (Table 1). Although calf body weight and average daily gain did not have a significant treatment \times time interaction, they did demonstrate a time effect (Figure 1). The RPM group only had a significant increase in the ADG over the control between birth and weaning and between 25 and 50 days after weaning. However, the RPM group was steady compared to the extreme fluctuations of the control group. The heifers, first time mothers, used in the project were in two groups with an exact average body weight, 997 lbs, to be used as a baseline (Table 1 Cow BWO). This baseline allowed us to see if the mothers were able to maintain a healthy weight after giving birth and lactating. At the end of the calving season, there were 47 lb. difference between the RPM heifers and the control group, favoring the RPM (Table 1).

The RPM calves had a significant increase in ADG and body weight while still being with their dams or immediately after weaning than when being directly supplemented with RPM in their diet (Table 1). These results suggest that the calves responded more effectively through milk rather than direct RPM supplementation, supporting our hypothesis that RPM cows have greater milk production compared to control cows. Since the RPM supplementation while in the womb increased the weight of the calf at birth, producing calving difficulties (Table 1), we suggest that RPM should not be administered to beef heifers. In conclusion, RPM supplementation improves calves' performance before weaning, probably due to a greater milk yield in RPM supplemented cows.

Table 1. Cow-calf pairs performance data.

Cow#	Treatment	Pelvic_Area	Days_on_RPM	Calf_sex	Calf#	CowBW0	CowBW1	BW0	BW1	ADG(0-1)	BW25	ADG(1-25)	BW50	ADG(25-50)	BW80	ADG(50-80)	Notes
1531	CON	320	106	M	8076	1025	952	65	191	1.48	239	1.85	276	1.61	336	2.31	
6078	CON	320	125	M	8032	985	856	68	218	1.45	274	2.15	303	1.26	326	0.88	
6098	CON		171	M	8111	1085											Cow died after giving birth
6113	CON		158	M	8105	1100											Sick calf-Died
6122	CON	288	123	M	8026	875	904	60	176.5	0.94	226	1.90	255	1.26	303	1.85	
6131	CON	302	131	M	8052	1050	930	70	154.5	0.86	202	1.83	223	0.91	266	1.65	
6142	CON	280	155	F	8101	995	984										Female
6159	CON	276	131	M	8053	940	859	70	181	1.18	220	1.50	247	1.17	293	1.77	
6164	CON	300	126	M	8036	1000	886	75	181	1.06	229	1.85	246	0.74	301	2.12	
6166	CON	278	151	M	8098	900	910	90	152.5	1.42	196	1.67	209	0.57	258	1.88	
15108	CON	323	136	M	8074	1010	948	65	157.5	1.02	202	1.71	238	1.57	281	1.65	
1525	RPM		114	M	8007	1055											No C-section - Stillborn
1596	RPM	320	117	M	8011	985	984	80	227	1.34	273	1.77	282	0.39	357	2.88	
6034	RPM	310	153	M	8100	1185	1220	75	195.5	2.20	216	0.79	262	2.00	269	0.27	
6075	RPM	310	119	M	8016	1060	984	90	249	1.62	316	2.58	331	0.65	386	2.12	
6082	RPM	283	127	M	8041	980	890	85	204	1.34	262	2.23	306	1.91	359	2.04	
6089	RPM	280	169	M	8109	910	886	80	138.5	2.08	171	1.25	200	1.26	234	1.31	
6105	RPM	320	156	M	8104	980	946	80	159	1.74	180	0.81	220	1.74	267	1.81	
6106	RPM	300	120	M	8017	950	862	85	250	1.66	299	1.88	350	2.22	397	1.81	
6110	RPM		141	M	8086	920											C-section - Stillborn
6126	RPM	285	131	M	8051	960	920	65	168.5	1.03	206	1.44	264	2.52	319	2.12	
6153	RPM	255	127	M	8042	980	956	89	189.5	1.18	241	1.98	274	1.43	290	0.62	C-section - Alive
AVG	CON	298	138		997	914	70	177	1.18	224	1.81	250	1.14	296	1.76		
	RPM	296	134		997	961	81	198	1.58	240	1.64	277	1.57	320	1.66		
P value		0.79	0.66		1.00	0.24	0.02	0.19	0.03	0.41	0.46	0.19	0.14	0.30	0.75		

Abbreviations: Cow# =Cow ID number; Treatments: control (CON) and rumen-protected methionine (RPM); Calf# =calf ID number; Calf sex: male (M), female (F); CowBW0=cow body weight in pounds at first day of study; CowBW1=cow body weight in pounds at right after calving season finished; BW0=calf body weight in pounds at birth; BW1=calf body weight in pounds at weaning; ADG(0-1)=average daily gain between birth and weaning; BW-25=calf body weight at 25 days after weaning; ADG(1-25) average daily gain between weaning and 25 days; BW(50)=calf body weight at 50 days after weaning; ADG(25-50)=average daily gain between 25 and 50 days; BW80=calf body weight at 80 days after weaning; ADG(50-80)=average daily gain between 50 and 80 days. Notes: reason for removing cow from study. Statistical significant differences where declared at P < 0.05.

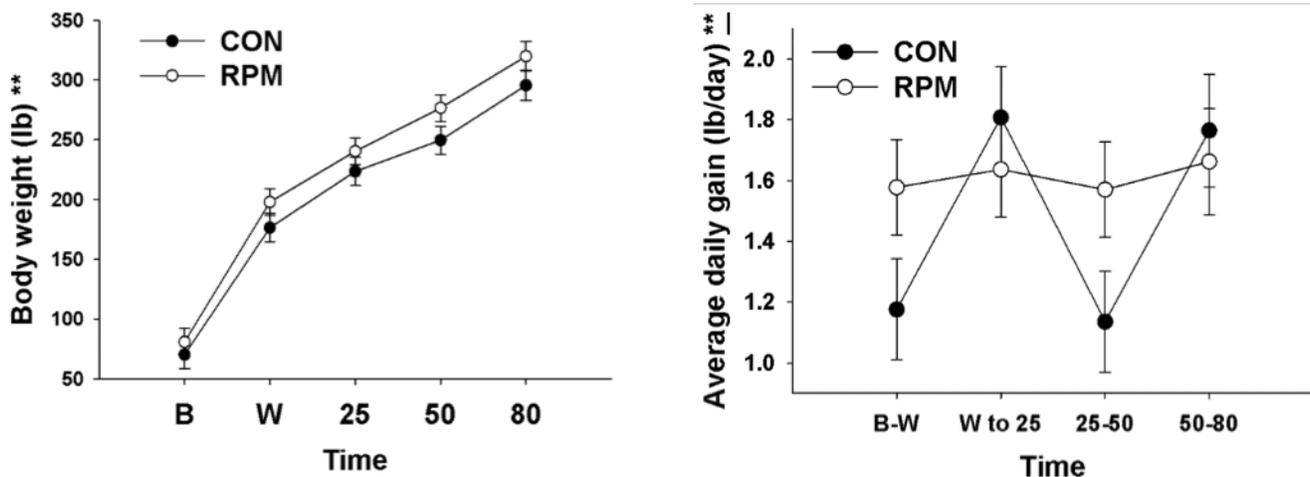


Figure 1. Body weight and average daily gain of beef offspring that received (RPM) and control calves (CON). B = Birthday, W= weaning, 25 = 25 days after weaning, 50 = 50 days after weaning, 80 = 80 days after weaning. B-W = Birthday to weaning, W to 25 = from weaning to 25 days after weaning, 25-50 = from 25 to 50 days after weaning, 50-80 = from 50 to 80 days after weaning. Statistical significant differences were declared at P < 0.05 and tendencies at P > 0.05 and < 0.1.

Statement of Research Advisor:

Betsy's work documents some of the first data published regarding the use of rumen-protected methionine during late gestation and in postnatal life in beef cattle. Her work is part of an ongoing study. During her summer internship, Betsy learned a muscle biopsy technique on Longissimus dorsi muscle of beef calves, how to extract blood samples from the jugulars vein and how to weight animals without the use of a squeeze chute. Through Betsy's research, she gained experience handling beef cattle that will be advantageous in her future career as a veterinarian.

—Sonia Moisés, PhD, Department of Animal Sciences

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2. Zhou, Z., et al., Better postpartal performance in dairy cows supplemented with rumen-protected methionine compared with choline during the peripartal period. *J Dairy Sci*, 2016. 99(11): p. 8716-8732.
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Assessment of the neurotoxic effects of prenatal nicotine and alcohol exposure on hippocampus and cortex of rodents

Ayaka Fujihashi, Murali Dhanasekaran

The focus of this study was to observe the effects of prenatal exposure to nicotine and alcohol. Women who report drinking during pregnancy have a higher incidence of also smoking during pregnancy; however, very few clinical data exist on the cumulative effects of both nicotine and alcohol during pregnancy. Thus, this study investigated the neurological and behavioral changes in rats exposed to nicotine and alcohol during gestation with specific emphasis on the hippocampus and cortex of the offspring.

Our study used fifteen pregnant Sprague-Dawley rats, separated into three groups of five: (1) a control group, (2) a group administered a 10% v/v alcohol in tap water solution (roughly equivalent to 1-2 drinks a day for humans), and (3) a group administered 10% v/v alcohol in tap water solution as well as 6mg/kg/day of nicotine via a subcutaneous semi-osmotic pump (roughly equates to moderate to heavy smoking in humans). Six weeks after birth, the hippocampus and cortex of the offspring of the respective groups were collected and tested for each group's long-term potentiation, as well as levels of markers of oxidative stress in the cortex. Long-term potentiation, an indicator of how effectively neurons are communicating in the hippocampus, allows for comparison of relative synaptic plasticity of the three experimental groups. Both alcohol- and alcohol with nicotine-exposed animals showed significant deficit in long-term potentiation (LTP) as compared to the control. However, there was no significant difference between the alcohol group and the alcohol with nicotine group. Both alcohol and alcohol with nicotine groups exhibited significant increases in markers of oxidative stress and mitochondrial dysfunction; however, the alcohol with nicotine group exhibited less neurotoxicity than the alcohol alone group. This, along with LTP results, indicate that both alcohol and nicotine have detrimental effects, but no conclusion can be made about the combinatory effects of prenatal alcohol and nicotine exposure. Future studies will focus further on elucidating the mechanisms of neurotoxicity in the developing rodent fetuses.

The adverse effects of prenatal nicotine and alcohol exposure on human reproductive outcomes are a major scientific and public health concern. Thus, this work on prenatal alcohol and nicotine exposure may lead to new scientific perceptions and novel and suitable therapeutic actions in the future.

Statement of Research Advisor:

Ayaka was involved with animal and chemical procurement, designing the experiments, analyzing the data, writing the results, presenting in conference/meetings, and she will be involved with publishing in a peer-reviewed journal.

— *Murali Dhanasekaran, Harrison School of Pharmacy*

Elucidating the role of periplasmic nitrate reductase (Nap) in *Pseudomonas aeruginosa* physiology

Amanda Myles , Sang-Jin Suh

Pseudomonas aeruginosa is an important bacterial pathogen that predominantly infects immunosuppressed patients. This pathogen is recalcitrant to antibacterial therapy because of its natural resistance and its propensity to form thick biofilms that inhibit penetration of antibiotics and antibodies. Therefore, understanding its basic mechanisms of survival in the host may suggest better therapeutic approaches by targeting its environmental adaptability.

P. aeruginosa is the infectious agent for almost 90% of mortality in people with cystic fibrosis (CF). In the CF lungs, *P. aeruginosa* is believed to live in anaerobic/hypoxic environment. Under those conditions, *P. aeruginosa* relies on nitrate respiration for energy generation. Periplasmic nitrate reductase (Nap) is one of the three nitrate reductases in *P. aeruginosa*. The physiological role of Nap in *P. aeruginosa* stress resistance and pathogenesis has not yet been elucidated. I hypothesize that Nap may catalyze redox balancing in *P. aeruginosa*, functioning in the aerobic dissipation of excess reducing power, perhaps preparing the cell for the transition to long-term anaerobiosis.

As a first step in elucidating the Nap function in *P. aeruginosa*, a *napD::lacZ* reporter gene fusion was used to characterize the conditions that may affect expression of the genes that encode for the Nap function. Previous data in the lab had suggested that *nap* operon may be under the regulation of the general stress response regulator RpoS in *P. aeruginosa*. The previous data was validated and *nap* expression was demonstrated to be regulated by RpoS. In addition, the potential effect of nitrate in the growth medium during aerobic growth was assayed. Although the nitrate data were inconclusive under aerobic growth conditions, the RpoS regulation of *napD::lacZ* was apparent in all of the nitrate concentrations tested. Future plans are to determine the potential effect of nitrate on *nap* expression under anaerobic growth conditions when nitrate could be utilized to generate energy for the cell.

Whether the *nap* expression is affected by the redox state of the bacterium was also addressed. I hypothesized that when *P. aeruginosa* is grown on more reduced carbon sources, it would accumulate more reduced molecules (e.g., reduced nicotinamide adenine dinucleotide) than when grown on more oxidized carbon sources and, thus, express *nap* operon higher for redox balancing. The *P. aeruginosa* wildtype PAO1 *napD::lacZ* fusion strain was grown on four carbon sources of the tricarboxylic acid cycle (citrate, succinate, fumarate, and malate) and the *napD::lacZ* expression was measured. It was determined that the *napD::lacZ* expression correlated with the reduced state of the carbon source. The *nap* operon expression was highest when grown on citrate and lowest on malate. These data suggest that Nap may be involved in redox balancing in *P. aeruginosa* if the initial hypothesis is correct. In near future, whether Nap plays a role in anaerobic growth of *P. aeruginosa* will be elucidated to determine whether Nap could be a potential target for therapeutic approach to combat this important bacterial pathogen.

Statement of Research Advisor:

Amanda's preliminary data suggest a potentially important role of Nap in the initial adaptation of *P. aeruginosa* to hypoxic/anaerobic environment of the host. Understanding how *P. aeruginosa* adapts to the host environment to persist and thrive will be important in developing approaches to combat this pathogen.
— Sang-Jin Suh, Department of Biological Sciences

In Vivo tracking of *Staphylococcus aureus*

Madison Schoeberl, Peter Panizzi

My project was to track *Staphylococcus aureus* (*S. aureus*) infections in a living animal system over time by use of optical and photo-acoustic imaging methods. We chose to use a bioluminescent strain of *S. aureus* that constitutively expresses light so we could identify bacteria as the infection spread. Our goal was to see if we could monitor development of kidney abscesses in murine glomerulus. As there was little previously done in the way of characterizing this particular light-producing strain, we confirmed that our target strain *S. aureus* Xen36 expresses an important virulence factor called staphylocoagulase (SC). SC is a potent activator of prothrombin, so we were able to assess presence of SC by using a chromogenic substrate assay specific for thrombin activity. We infer that the high level of staphylocoagulase should translate to greater infectivity and virulence potential of a given strain *in vivo*.

To determine whether the bioluminescence can be tracked in a murine model after injection, we intravenously injected the pathogen at a sublethal dose. The distribution and kinetics of the infection were analysed using small animal bioluminescence imaging mice. It took multiple trials to determine an optimal pathogen load. The colony-forming units injected were adjusted to allow for kidney infections to develop. To complement our studies, we plan to add photo-acoustic imaging to better locate infections in anatomical structures. Collectively, the study closes current gaps that exist in the understanding of the best ways to track *S. aureus* in a host. This understanding will lead to the eventual development of new adjunctive therapies designed to be used with antibiotics in the future.

Statement of Research Advisor:

It is our hope that one day we can design tools to specifically identify infections so that patients, in the end, get the right antibiotic therapy. Madison's project is at the forefront of modern microbiology as molecular imaging is underutilized and, when used, the results are often misunderstood or too vague to be meaningful. We hope to design better ways of imaging infections while supporting our finding with basic microbiology techniques and histological analysis.

— *Peter Panizzi, Harrison School of Pharmacy*

Afrofuturist collages, time-lags, and post-soul identities

Jordan Wade, Kathryn Floyd

In 2012, Pierre-Christophe Gam created an Afrofuturist website for the French-African pop singer, Taali M (www.taali-m.com), whose “Afro-pop” music mixes sounds inspired by ska, 80s pop, ragga, and traditional African chant. Like Taali M’s collage of musical inspirations, Gam’s website for the singer stitches together a diverse range of images, including a 1970 photograph of a Kuba king, a 1950s photograph of a Fulani woman, and digitally rendered images of 16th century Yoruba Queen Mother Heads.¹ By collaging these images from Africa’s past into a sleek, digitally rendered setting, Gam grants the website a decidedly “Afrofuturist” aesthetic. My research addresses Gam’s use of an Afrofuturist framework, specifically focusing on how he collages technology with imagery appropriated from Africa’s past as a means to represent Taali M’s individual identity, address and reframe perceptions about race and cultural purity, and provide a visual representation of the concept of black post-soul identity.

Afrofuturism was coined in 1993 but has been present in popular culture since the mid-20th century—from 1960s jazz musician, Sun Ra, to the 2018 *Black Panther* movie—and has since been defined as a “critical perspective” by which black subjects are projected into the future, thus transforming spaces of racial misrepresentation or oppression into spaces for conversation. Since Gam’s Taali M website and most other Afrofuturisms draw on imagery, sounds, or ideas from Africa’s past, and since people do not presently hold the ability to literally warp time, Afrofuturists create a “time-lag” in an artificial future where black people have the technological power to pluck objects, people, and ideas from the past and into a contemporary, technological setting, thus disrupting linear constructions of time and subverting notions of Africa as archaic and technologically unadvanced.² The website’s collaged aesthetic also serves to deconstruct present cultural classifications thrust upon African

communities by European colonizers in the 19th-20th centuries. Because Gam collaged such a diverse range of African imagery into the website, it is difficult for viewers to culturally categorize it. This blurs the notion of “cultural purity,” which, in the West, is often associated with primitivity.

In addition to using Afrofuturist collage to reclaim and recontextualize African technology use and cultural classifications, by collaging notions of the individual (representations of Taali M specifically) and the group (imagery that might represent Africans and diasporic Africans as a whole), Gam creates a visual reflection of the concept of dual consciousness, or post-soul, which, in contemporary scholarship, refers to the multiplicities contained in individual black identities, similar to the amalgamation of time periods and cultures used to represent Taali M.³ This research is significant, because Afrofuturism itself is a fairly new area of study and because Afrofuturists often use a combination of futuristic imagery and allusions to Africa’s past, but the significance of this sort of collage aesthetic and its relationship to post-soul identity has yet to be clearly and critically addressed.

Statement of Research Advisor:

Jordan’s research into the Afrofuturist design of the website for African pop artist Taali M is an example of the innovative art historical frameworks and methods emerging today in response to global contemporary art and visual culture. Her case study tackles complicated questions around race, history, identity, and media, as it brings together critical theory with popular culture. Her analysis sheds light on the way global artists construct flexible and empowering meanings that operate across multiple platforms, not only reflecting their agency, but also creating it.

—Kathryn Floyd, *Art and Art History*

¹ *Making Africa*, <http://makingafrica.net/2015/04/featured-work-pierre-christophe-gam-taali-m/>.

² Bhabha, 1992: 57.

³ DuBois, 1897: np. See also Gilroy, 1993: 13.

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¹Making Africa. “Featured Work: Pierre-Christophe Gam: Taali M.” Accessed September 9, 2017.<http://makingafrica.net/2015/04/featured-workpierre-christophe-gam-taali-m/>.

²Bhabha, Homi. “Postcolonial Authority and Post-modern Guilt.” In Cultural Studies, ed. Lawrence Grossberg, Cary Nelson, and Paula Tredichler. New York, NY: Rutledge Press, 1992.

³Du Bois, W.E.B. “The Conservation of Races.” In The Souls of Black Folk, ed. David W. Blight and Robert Gooding-Williams, 228-238. Boston, MA: Bedford Book, 1897.

⁴Gilroy, Paul. The Black Atlantic: Modernity and Double Consciousness. Cambridge, MA: Harvard University Press, 1993.

Detecting uranium and toxic heavy metals using molecular sensors

Kevin Wyss, Emily Hardy, Anne Gorden

Although nuclear power is extremely efficient, generates no harmful greenhouse gasses, and uses no petroleum resources or fossil fuels, the increased use of nuclear power for civilian energy has been met with skepticism. This is largely due to fears of radioactivity and contamination from nuclear waste or disasters, such as a power plant accident. Radioactivity cannot be easily detected seen, tasted, smelled, or felt. Most modern detection methods require the use of technical equipment or training and may not be feasible or time effective to use in disaster situations.

Our research tries to develop new strategies to remedy the possibility of nuclear contamination. We have developed molecules that detect and bind with uranium or other radioactive and toxic heavy metals such as cadmium and the lanthanides. Our goal is to synthesize a small amount of powder that can simply be stirred into a sample of drinking water and change colors within seconds in the presence of contamination. Some of the challenges associated with this project include the fact that many dissolved metals have similar chemical properties, whether or not they are toxic and radioactive. For example, making the molecule sensitive to the presence of uranium and not iron is central in creating a dependable sensor. Another important quality of an effective sensor is the ability for the sensor to indicate extremely low concentrations of hazardous materials, as extremely small amounts can still be harmful to humans. By tweaking, synthesizing, testing, and analyzing the sensor molecule, it has been greatly improved. The sensor molecule binds with the positively charged metal ions through oxygen and nitrogen atoms, a process known as coordination. Different metal ions result in slightly different coordination environments, and if the sensor is effective, this can manifest as a visible color change as shown in Figure 1.

In this study, multiple sensors were designed and synthesized and synthetic procedures were streamlined and refined¹. The synthetic procedure for one such molecular sensor is shown in Figure 2.

Once the sensors were able to be made at high purities and on large scales, we worked to explore and characterize how the sensor behaved in solutions with different metal contaminations². Through single crystal X-Ray diffraction, we were able to visualize how the sensor molecules actually bind with the uranium when the molecular sensor powder is added to the contaminated samples. The crystal structure of a sensor molecule coordinated to a uranium containing uranyl ion is shown in Figure 3.

Statement of Research Advisor:

Although it seems like a small amount of steps, Kevin worked diligently with his graduate mentor to perfect the synthetic steps and carefully isolate and purify his organic compound to use in the metal complexation and characterization studies. He has optimized the procedure and has continued to characterize the interactions of this species with other metals.

—Anne Gorden, *Chemistry and Biochemistry*

References:

- ¹ Hardy, E. E.; Eddy, M.A.; Wyss, K.M.; Gorden, Anne E.V. "An example of unusual pyridine donor Schiff base uranyl (UO₂²⁺) complexes," *Chemical Communications*, 2017, 53, 5718-5720. (DOI: 10.1039/c7cc02747h)
- ² Wyss, K.M; Hardy, E. E.; Gorden, Anne E.V. "An Example of Enhanced Emission of a Pyridine Containing Schiff Base Zinc(II) Complex," submitted

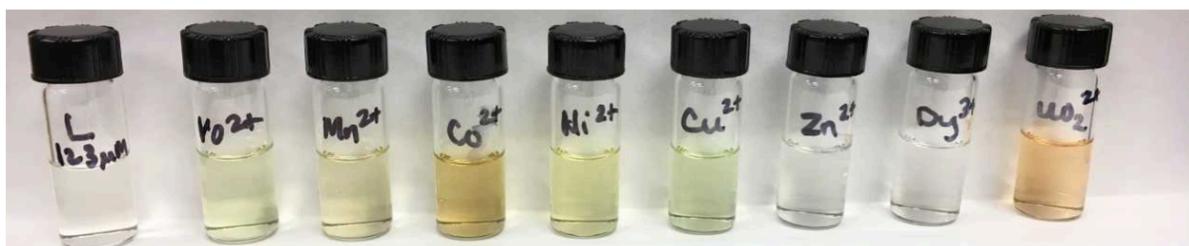


Figure 1: Differing colorimetric signals produced by sensor molecule in varying metal solutions. Uranium contamination is on the far right.

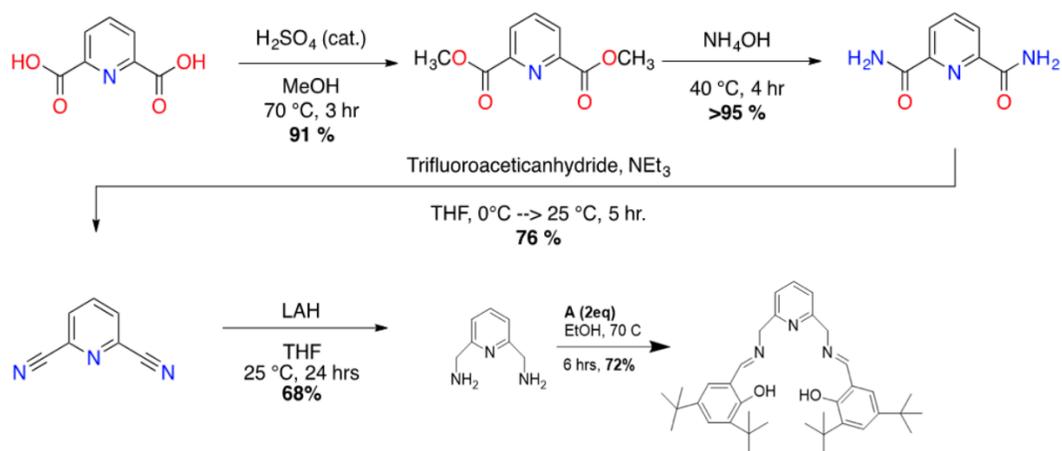


Figure 2: A scheme showing the synthetic steps required to create one of the molecular sensors used in this study.

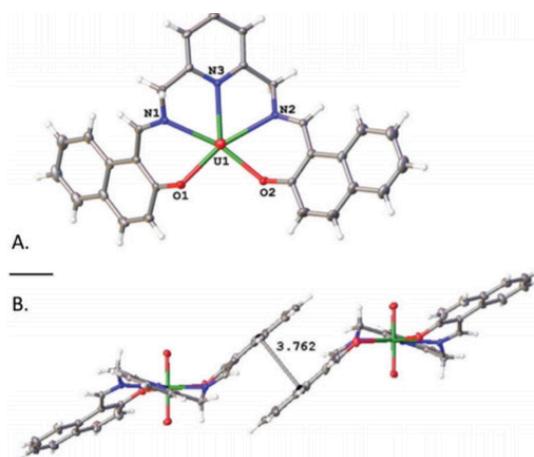


Figure 3: The crystal structure of one of the molecular sensors used in this study, coordinated to uranyl.

Physiological responses of the invasive cane toad (*Rhinella marina*) from Florida: How stress affects immune response and locomotor performance

Emma Yonan, Steven Gardner, Mary Mendonça

The purpose of this study was to determine the energy and physiological trade-offs from *Rhinella marina*, or cane toads, on its invasion front and in sites that were previously populated. An invasion front is the edge of a moving or expanding population. *Rhinella marina* are native to areas in South America, but invasive populations have been found in Florida. *Rhinella marina* were originally introduced for crop protection against sugarcane pests, but failed. They were also accidentally released by the Miami Airport (Lever, 2001).

In this project, the toads were given a set length of track at different temperatures in one experiment and then given an endurance challenge. We hypothesized that the northern population (invasion front) would be less willing to move because of the energy trade-off, and instead, secrete toxins to defend themselves from predators.

Twelve toads collected for this experiment, 6 from Miami and 6 from New Port Richey (the invasion front) in Florida. Toads were challenged to progress up and down a 200-cm track in a temperature-controlled chamber. Mass was recorded before every run, while snout-vent length (SVL) and leg length were recorded for each toad prior to the locomotion challenge. The different populations were run on alternating days at 8:00 pm under only red lights. The toads were acclimated for 24 hours in the chamber before each run and tested at 25°C then 19°C. The first part of the experiment was a burst speed test. Twelve toads were placed onto a track at either 25°C or 19°C and “encouraged” to hop by gentle tapping with a pencil/pen. Each individual toad was encouraged to hop until it had gone a total distance of 4 m. During this period, a stopwatch was used to measure the duration of time it took each toad to travel 4 m, and the number of hops and the number of taps required to encourage the toad to travel the full distance were recorded. For the endurance test, which was the second part of the experiment, the same twelve toads from earlier experiment

were placed back onto the track at 25°C, followed by the same challenge at 19°C, and encouraged to hop until stopping. During this trial, the time it took the toad to reach the total distance before stopping, the number of taps it took before each toad stopped, the number of hops taken by each toad before stopping, and the total distance traveled in the track were recorded.

Although the total distance traveled by the toads was not significantly affected by location, the unwillingness to move, characterized by a lower number of taps, a lower number of hops, and less time before the toad stopped moving, was significantly higher in New Port Richey toads compared with Miami toads (Figure 1). Also, larger toads were more willing to move for both populations. This result could reflect behavioral differences in the toads from the two locations, as Australian cane toads near the invasion front have been characterized by unwillingness to move when placed in a track and by a higher likelihood of secreting poison when tapped, compared with toads from the more established populations.

Further work will expand the size of each trial population and use lactate fall information to better establish fatigue and endurance. Additional work will also involve running toads brought newly into captivity and comparing them to toads that have been held in captivity longer.

Statement of Research Advisor:

Emma Yonan was an enthusiastic undergraduate who put in a great many hours of careful testing of the endurance of invasive cane toads from core introduced populations (Miami) versus those from populations on the invasion front in northern Florida. She became familiar with aspects of invasion biology, physiology, and statistical analysis to produce a study that is definitely publishable.

—*Mary Mendonça, Biological Sciences*

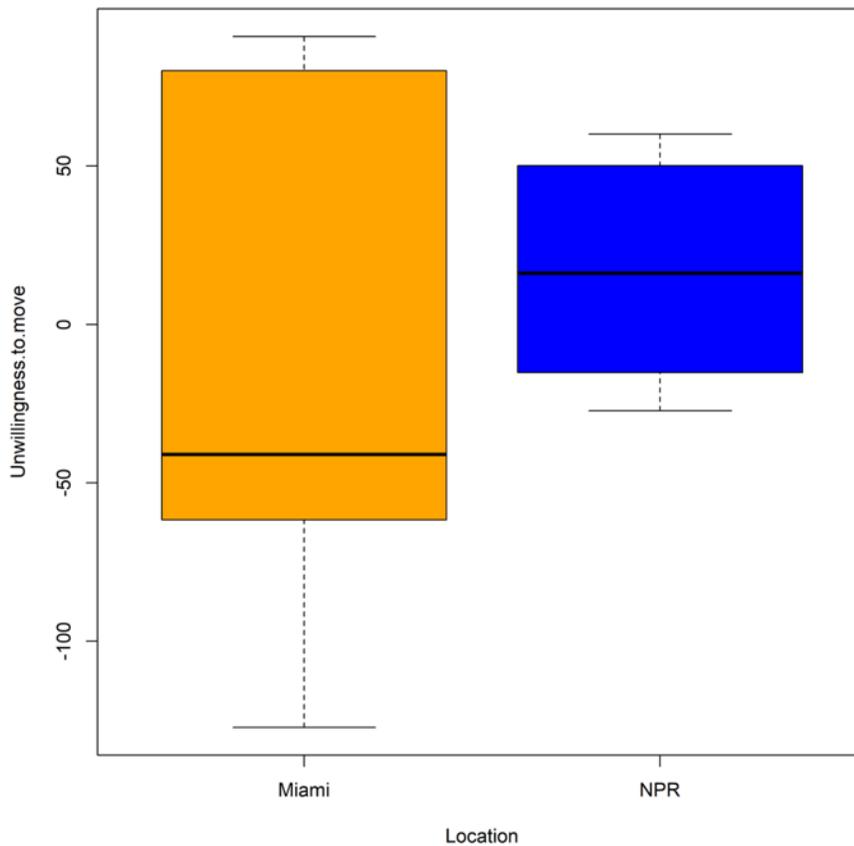
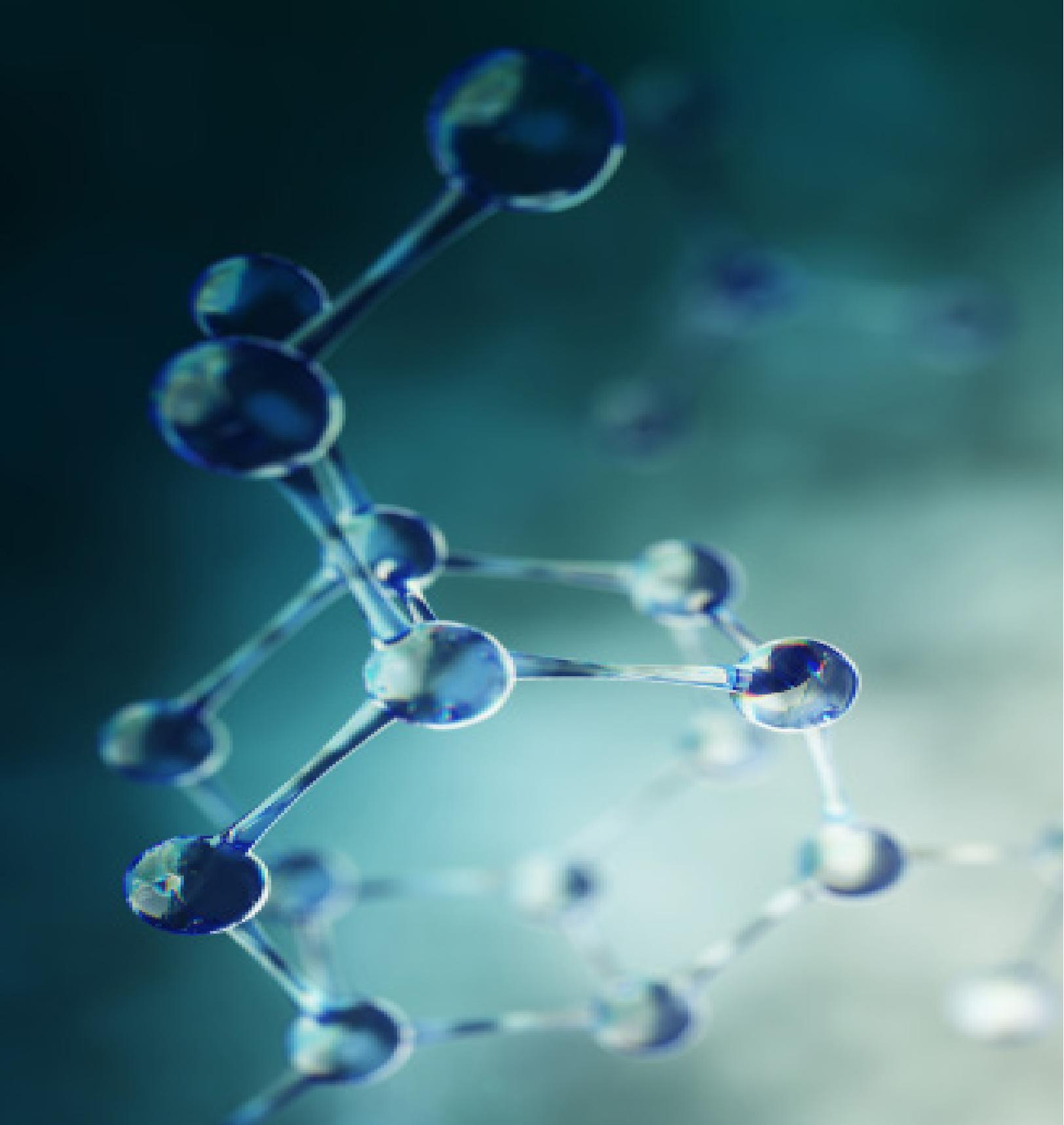


Figure 1. Unwillingness to move for Miami and New Port Richey populations of *Rhinella marina*.

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Lever, Christopher (2001). *The Cane Toad. The history and ecology of a successful colonist.* Westbury Publishing. ISBN 1-84103-006-6.



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