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## Editor in Chief

**Lorenzo Cremaschi** is a Professor in the Department of Mechanical Engineering at Auburn University (Auburn, AL). He received his Ph.D. from the University of Maryland (College Park, MD), in 2004. In 2001 he was awarded his M.S. and B.S. diploma degrees from the University of Modena (Italy). Before joining Auburn University, Dr. Cremaschi served as a post-doctoral research associate at Purdue University's Herrick laboratories, followed by assistant and associate professor in the School of Aerospace and Mechanical Engineering at Oklahoma State University (Stillwater, OK).

Dr. Cremaschi's interests focus on buildings' energy efficiency and sustainability, scalable energy systems, water desalination and purification, HVAC and refrigeration systems for the built environment. His current research includes thermodynamics and heat transfer of low Global Warming Potential refrigerants, building air dehumidification processes, frost and defrost, and moisture transport in thermal insulation systems. He is the principal investigator of several research projects and the author of over 95 papers. He revised three book chapters, and gave several presentations at conferences, meetings, and seminars at Universities. Dr. Cremaschi is the inventor of two U.S. patents.

Dr. Cremaschi is an associate editor for the Science and Technology for the Built Environment Journal. He has several honors and awards, including the 2021 Exceptional Service Award from the American Society of Heating, Refrigerating and Air-Conditioning Engineers (ASHRAE), the 2014 Research Excellence Award from the College of Engineering at Oklahoma State University, and the 2013 Innovative Research Grant Award from ASHRAE.

Dr. Cremaschi teaches courses in the area of Thermodynamics, Refrigeration, Renewable Energy Systems, and Cryogenics. These courses are offered at the undergraduate mechanical engineering curriculum's second, third, and fourth years and at the graduate level. Dr. Cremaschi advised 34 graduate students (14 Ph.D. students and 20 M.S. students), over 50 undergraduate students in academic curriculum and career choice. He also mentored 22 undergraduate students during research and scholarly activities and 8 international visiting exchange scholars. Dr. Cremaschi is the faculty advisor of the Auburn University ASHRAE student organization, which was founded at Auburn since his arrival in 2016. He fostered several scholarships and awards for undergraduate students involved with his research.

Dr. Cremaschi is a member of the American Society of Thermal and Fluids Engineers (ASTFE) Executive Committee and the ASHRAE Research and Administration Committee. He was appointed as chair for the International Institute of Refrigeration (IIR) U.S. National Committee, and he is a member of the International Energy Agency (IEA) U.S. Heat Pump National team. He has been serving in several technical and standard committees for the last 15 years. Finally, Dr. Cremaschi was the technical program chair for two ASTFE conferences in 2019 and 2021. He organized numerous technical sessions, panel forums, and seminars within ASHRAE, ASTFE, IIR, and the American Society of Mechanical Engineers (ASME).



#### Production Editor

**Kelleen Wegener** is a third-year graduate student pursuing her Doctorate in Audiology. Kelleen graduated from Ohio University with a Bachelor of Science degree in Communication Sciences and Disorders. She is a Graduate Assistant for the Office of Undergraduate Research. Kelleen's other on-campus involvement includes being a member of the Student Academy of Audiology. In her spare time, Kelleen enjoys working out, spending time with family and friends, and traveling.



#### Associate Editor

**Claudia Ann Rutland** is a PhD student and a graduate research assistant in the Department of Crop, Soil, and Environmental Sciences, studying weed science under Dr. Scott McElroy. She graduated from Auburn University in 2020 with a Bachelor of Science in Microbiology. Her current research focuses on target-site resistance in herbicide resistant weed species, more specifically understanding the mechanisms of resistance in auxin-mimic herbicides. In her free time, she enjoys cooking, reading, painting, and spoiling her dog Obi.



#### Associate Editor

**Pritam Mitra** is a second-year PhD student and graduate research assistant in the Department of Agricultural Economics and Rural Sociology. He obtained his masters degree in Economics. Pritam is from Kolkata, India and his favorite sport is cricket. Other hobbies that Pritam has include playing violin and enjoying music.



#### Associate Editor

**Maliha Yel Mahi** is a second-year graduate student pursuing her doctorate in Mechanical engineering. She has achieved her Master's degree in Mechanical engineering from the University of North Dakota. Her research focuses on Computational Fluid Dynamics (CFD) under the supervision of Dr. Lorenzo Cremaschi. She enjoys reading books, spending time with family and friends, and traveling in her spare time.



#### Associate Editor

**Kanak Parmar** is a PhD student at Auburn University in Aerospace Engineering, specializing in space mission design, trajectory design and analysis, astrodynamics and machine learning. She previously graduated from Auburn University with a bachelor degree in Aerospace Engineering, with a minor in Business, as well as a Master of Science degree in Aerospace Engineering. Her research focuses on the application of machine learning methods to astrodynamical applications to achieve greater on-board autonomy for real-time spacecraft guidance and control, with an emphasis on exploring the novel field of human-AI cooperative learning. In her spare time, she enjoys spending time with family and friends, reading, and playing the viola.





**Associate Editor**

**Ying Yan** is a Ph.D. candidate in the Consumer and Design Sciences. She received her M.I.D. in the Industrial Design program from Auburn University in 2019. She has rich work and research experience in multiple design fields, including industrial design, interior design, consumer behavior, and business. Ying worked at Playcore, a playground company in Chattanooga, as a graduate student and a one-year full-time intern in 2015. Currently, Ying's research focuses on increasing college students' learning motivation. As she develops her research, Ying hopes to promote game-based learning in higher education to bring more fun learning experiences.



**Associate Editor**

**Chenyu** is a fourth-year PhD Candidate the Department of Health Outcomes Research and Policy at the Auburn University Harrison College of Pharmacy. Her research interests include the use of health information technology (HIT) and health behaviors. She is also interested in applying social and behavioral theory to improve patients' health outcomes. Currently, her research focuses on the implementation of physical activity interventions among university students.



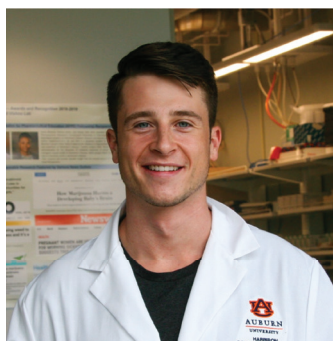
**Associate Editor**

**Sara Schiller** graduated from Auburn in 2016 with a bachelor's degree in Music Education and returned to Auburn for graduate school. Sara is currently pursuing her PhD in Music Education and serves as the Administrative Vice President of the Graduate Student Council. Sara's chosen field of research investigates the positive effect of music education on self-esteem levels in primary aged students.



**Associate Editor**

**Benjamin Jackson** is a fifth year Auburn graduate student in the Department of Chemistry and Biochemistry pursuing a PhD in Chemistry. Benjamin graduate from the University of South Alabama in 2014 with a Bachelor of Chemistry. His graduate work at Auburn University has focused on the use of quantum chemical methods to study systems with diffuse electronic structure with the intent of designing novel catalytic materials for applications in CO<sub>2</sub> conversion.



**Associate Editor**

**Jeremiah Pfitzer** is a graduate student pursuing his Doctorate in Pharmacology in the Harrison School of Pharmacy at Auburn University. He is currently a Graduate Teaching Assistant for the Faculty Affairs and Strategic Initiatives. His current research projects investigate learning and memory alterations from both prenatal cannabinoid exposure and in novel glutamate modulating prodrugs as Alzheimer's disease therapeutics. In his spare time, Jeremiah enjoys cooking, working out, and painting.



**Associate Editor**

**Hannah P. Luz** is a second-year Master's student in Communication at Auburn University's School of Communication and Journalism. Hannah researches in the area of organizational communication, specifically focusing on labor organizing, CCO, and menstrual and reproductive health. She is currently an instructor teaching public speaking, a graduate research assistant, an associate editor for AUJUS, and a senator in the Graduate Student Council. She received her Bachelor's degree in Communication from Auburn University in Spring 2021. After graduating with her Master's degree, Hannah is planning on pursuing a Doctorate in Organizational Communication.



**Associate Editor**

**Ajay Jayswal** is a PhD Student in the Mechanical Engineering Department at Auburn University. He has completed his BS in Mechanical Engineering from Tribhuvan University, Nepal in 2017 and MS (Non-Thesis) in Mechanical Engineering from Auburn University in 2021. His research interests include additive manufacturing of polymers, computer design of textile-based structures, polymer processing and composites, thermo-mechanical characterization, and finite element modeling (FEM). He has published 5 peer reviewed journal articles and 1 conference article.



**Associate Editor**

**Shiva Rahmati** is a PhD candidate in chemical engineering department with expertise in nanomedicine.

# Family Structure and Internalizing Features in Adolescents Who Engage in Illegal Sexual Behavior

Kylie M. Seaton <sup>1</sup> and Kelli R. Thompson <sup>2</sup>

<sup>1</sup> Lab Manager (and 2021 Undergrad. Student Research Fellow), Juvenile Delinquency Lab, Auburn University

<sup>2</sup> Assistant Research Professor, Department of Psychological Sciences, Auburn University

## Abstract

Incarcerated adolescents are more likely to be diagnosed with a psychiatric disorder than their non-offending peers, with depression and anxiety being the two most common. Disrupted caregiving has been a key factor in the high psychiatric disorders observed in many juvenile detention settings. This study aimed to examine family functioning and reports of internalizing features of clinical disorders in a sample of adolescents adjudicated for illegal sexual behavior (AISB). Data were collected as part of a pre-treatment evaluation for youth in the Accountability Based Sex Offense Prevention Program (ABSOPP) located at a residential facility in the Southeast United States (n=966). Results indicated those coming from dual-parent homes with maternal involvement reported significantly higher rates of anxious feelings and lower rates of family discord, as reported by the Millon Adolescent Clinical Inventory (MACI). While cross-sectional in nature and therefore not causal, this research could provide insight into factors contributing to the development of depression and anxiety in incarcerated youths.

**Key Words:** adjudicated youth, sex offenses, family, depression, anxiety

## Introduction

Sex offending continues to be a topic of high demand within the criminal justice system and clinical therapeutic practice. Reports of rape make up over 11% of all known violent crimes, and this may represent only 25% of actual incidences of rape occurring annually, making evident the need for appropriate treatment of sexual offending behavior (FBI 2019; Morgan & Oudekerk, 2019). Adolescents adjudicated for illegal sexual behav-

ior (AISB) form a unique subpopulation of offenders. <sup>1</sup>These adolescents differ from non-sexual offending delinquents in levels of social competence, psychopathology, history of childhood sexual abuse, levels of childhood attachment, and even risk for future offending (Seto & Lalumiere, 2010; Terry, 2013). AISB can have as high as 15% and as low as 4% rearrest rate for later sexual offenses and a significantly higher rearrest rate for later non-sexual offenses when compared to adolescents adjudicated for general delinquent behavior (AGDB) (Caldwell, 2002; Everhart-Newman et al. 2019; Zimring, 2004). Furthermore, re-offending rates are even higher for AISB with more antisocial behavioral patterns (Waite et al., 2005). However, the goal of the juvenile justice system is not to provide retribution for crimes but rather to rehabilitate youth. For AISB, in particular, mental health treatment does work specifically to decrease the number of sexual re-offenses (Caldwell, 2007; Reitzel & Joyce, 2006).

## Delinquency and Mental Illness

When developing effective treatment programs for delinquent youth, there are many different areas to consider, such as underlying or comorbid psychopathology (Chaffin, 2008). Compared to those not adjudicated, youth in correctional facilities have a significantly higher prevalence of psychopathologies, such as depression, anxiety, and ADHD (Fazel et al., 2008). Diving further, AISB often presents features for multiple disorders, specifically depression, anxiety, and personality disorders, at significantly higher rates than any adjudicated youth subpopulation (Seto & Lalumiere, 2010). This is a critical factor to consider when developing treatment programs because untreated mental health problems are a risk factor for later re-offending (Singer et al., 2013). Further investigations, leading to a better understand

<sup>1</sup>Corresponding author: kelli.thompson@auburn.edu



ing of the psychopathological features of AISB, may also provide insight into how to improve the effectiveness of current treatment programs for this population.

It can be challenging to disentangle the etiology of mental illness and delinquency, as the two often develop together. A recent longitudinal study sought to determine which came first in a sample of 806 people from the time they were in fifth grade until the age of 33: the onset of depression, substance use, or delinquent behavior (Kim et al., 2019). Participants were followed every 3 years since 1985. Results indicated that for the sample followed, engaging in a criminal act typically preceded the onset of depression or a substance use disorder (Kim et al., 2019). This is in line with other research indicating depression and anxiety as outcomes of offending behaviors rather than precursors (Jolliffe et al., 2019). While there are many pathways to the development of mental disorders such as depression and anxiety, these studies provide valuable insight into the effects adjudication and incarceration may have on developing youth. Regardless of etiology, understanding the mental health portrait of adolescent offenders, particularly AISB, may contribute to defeating the myth that problematic sexual behavior patterns in adolescents are unable to be “cured” and demonstrate that investing in psychological resources for adjudicated youth can reduce the risk of reoffending (Chaffin, 2008; Terry, 2013).

### **Family Structure, Mental Illness, and Delinquency**

Adjudication in childhood can be an emotionally and mentally taxing experience for a fledgling adolescent. They all at once have to adjust to living in a residential facility, separating from peers, and battling societal judgments because of their sentence. When examining influences on a child’s behavioral and psychological development, it is important to include the role of the family. In addition to the stress of adjudication, youth leave their home environment upon entry and thus possibly leave their parents or guardians for the first time. This forced parental-child separation may cause additional psychological stress. The term “caregiver disruption” is used to describe insufficient attention from caregivers, often resulting in the physical and/or emotional needs of the child not being met (Sitney & Kaufman, 2020; Worley et al., 2011). This term has been used to explain increases in depressive behaviors, anxious symptoms,

delinquent behaviors, and other mental health-related repercussions of parent-child separation in other life situations such as parental divorce or parental relocation (Strohschein, 2005; Apel & Kaukinen, 2008). Upon entering a residential facility, youth are being separated from families in a way not unlike divorce or other forms of family separation (Burkhart & Cook, 2010). In the context of adolescent offenders, it is important to consider the effects disrupted caregiving can have on mental health.

This phenomenon may only partially account for the high prevalence of mental illness in juvenile facilities as adjudicated youth are a very diverse population who come from multiple different home settings outside of the traditional family. For example, many come from foster homes or may even be living somewhat independently with family and friends. The present study utilizes the term family structure to encompass all possible forms of adult guardianship or parental figures which modern families may have. Differences and changes in family structure in an adolescent’s life have been linked to delinquent behaviors. For example, children from separated families have a greater risk of delinquency when compared to children from non-separated homes (Price & Kunz, 2003). However, resiliency must always be considered, and in today’s modern society, families come in many forms (e.g., single-parent households, same-sex parents, adoptive parents, etc.), with resiliency demonstrated in all types. Furthermore, changes in the presence of parental figures have been linked with the mental well-being of children. For example, following a divorce, if the paternal figure relocates, the child is more likely to express symptoms of depression and anxiety and partake in more rebellious behaviors as opposed to maternal relocation (Stevenson et al., 2018). To summarize, research has clearly shown that a child’s family structure, delinquency, and psychopathology are all deeply intertwined.

### **Family Structure and AISB**

When compared to a non-delinquent adolescent population, AISB has family structures and experiences that are vastly different. Thus, the family structure may impact the psychopathology of AISB more than the average child or even the average delinquent child. This is not surprising given the higher rates of sexual abuse, often with intra-familial offending patterns,

seen among this population (Grant et al., 2017). Furthermore, compared to a non-delinquent adolescent population, AISB were more likely to have parents who exhibited externalizing behavior problems such as drug or alcohol abuse disorders (Duanne et al., 2003). This is critical because this type of family environment can negatively affect developing youth. AISB tends to have a significantly lower quality of parental relationships and attachment than other adjudicated adolescents including violent offenders (Sitney & Kaufman, 2020; Seto & Lalumiere, 2010; Worley et al., 2011). Furthermore, Sitney and Kaufman (2020) found that AISB tends to have lower quality relationships with their biological father than with their biological mother, among the lowest scores for any group of incarcerated youth. In terms of basic family structure, AISB are more likely to come from single-family households and are more likely to experience parental instability, such as being placed in out-of-home care or even homelessness, than AGDB (Margari et al., 2015; Felizzi, 2015). This is important when examining the etiology of sexual offenses because all these factors have been found to have a negative influence on children and adolescents. This also helps provide evidence of how family structure and quality of attachment and relationships as a result of that structure influence development in this population in a myriad of ways.

The purpose of the current study was to examine the relationship between family structure, mental illness, and delinquency in a sample of AISB. Specifically, the present study investigates whether differences in family structure pre-adjudication are associated with different levels of mental health difficulties post-adjudication. Understanding the relationship between these variables may help clinicians better predict treatment adjustment and effectiveness. Internalizing features of psychopathology, such as depression and anxiety, were chosen as outcome variables given the prevalence among children from separated households (Strohschein, 2005).

## Methods

### Participants

The sample consisted of 966 male AISB at a residential correctional facility in the Southeastern regions of the United States. The average age was 15.82 years, with a 9th grade modal level for the overall sample. More than half the sample identified as Caucasian (56%), with an-

other 41% identifying as African American, and 3% identifying as bi-racial, other, or mixed race. Regarding the family environment, 37% of the sample indicated having a history of Department of Human Resources-involvement, with 52% reporting witnessing domestic violence. Of those who had experienced domestic violence, 20% involved physical violence ( $n=165$ ), and 4% of incidents involved using a weapon ( $n=34$ ). Additionally, 21% reported a history of sexual abuse, 32% a history of physical abuse, and 16% a history of neglect. The majority of committing offenses were hands-on sex offenses such as Sexual Abuse (28.8%), Sodomy (19%), Sexual Assault (3.2%), and 1st Degree Rape/Attempted Rape (10.4%), and 2nd Degree Rape (7.2%). Another 18.1% had non-contact committing offenses such as Sexual Misconduct, Indecent Exposure, and/or some type of Probation Violation. Participants were enrolled in a court-mandated sex offender specific treatment program called the Accountability Based Sex Offense Prevention Program (ABSOPP). The current sample size was derived from archival data collected from youth residing at the facility between 2000 and 2016.

### Procedure

AISB entering the facility were given an expansive pre-treatment assessment which serves as a basis for developing an individualized service plan. AISB are also assessed after completing the treatment program to track treatment progress. However, those data were not a focus of the current analysis. Assessments were conducted by doctoral-level graduate students on practicum rotation at the facility. Data and research conducted with this program have been approved by an official university institutional review board. At the start of each intake, AISB were read aloud the informed consent process and asked if they would like to allow their records to be used for the review.

### Measures

Family structure was assessed through a series of clinical interviews upon entry to the facility. Participants were asked a series of questions regarding guardianship in the home before adjudication and incarceration by a graduate-level clinician who then coded the family environment accordingly after consulting file information for corroborating details. Of the 966 participants surveyed in the current sample, the response to this variable broke down in the following way: biological

mother only ( $n=307$ ), biological father only ( $n=63$ ), both biological parents ( $n=103$ ), biological mother and stepfather ( $n=175$ ), biological father and stepmother ( $n=78$ ), adoptive parents ( $n=44$ ), grandparents ( $n=109$ ), other biological relatives ( $n=49$ ), other non-relatives ( $n=13$ ), or other non-listed ( $n=25$ ). The subtle difference in coding between the last two categories typically dealt with whether or not the custodial caregiver was known to the adolescent. For example, the “other non-relatives” categories were coded as custodial situations involving close family friends or other arrangements made by the courts to reflect some degree of familiarity or prior attachment for the adolescent. The non-listed category typically reflected temporary custodial situations such as foster care or group home living situations.

The Millon Adolescent Clinical Inventory (MACI; Millon, 1993) was used to assess various dimensions of depression and anxiety using scales focusing on internalizing behaviors. The MACI is a self-report, 160-items questionnaire with items featuring a true-false response format useful in analyzing psychopathology in youth (Murrie et al., 2000). It is designed to assess a broad range of psychological problems experienced by adolescents ages 13 to 19. It includes a reliability scale, three validity scales, and three content domain scales (i.e., Personality Patterns, Clinical Syndromes, and Expressed Concerns). Null reports that resulted from spikes in reliability or validity scores were not used in the present study. Those scales pertinent to measuring internalizing features were used in the current analysis. This included two personality pattern scales measuring avoidant personality features of anxiety and dolefulness features associated with depression (Inhibited/Avoidant and Doleful/Depressive); two clinical syndromes scales measuring Anxious Feelings and Depressive Affect; and two expressed concerns scales regarding Self-Devaluation and Sexual Discomfort. The MACI demonstrates moderate to strong internal consistency (.73 - .91) and test-retest reliability (.57 - .92; Davis, Woodward, Goncalves, Meagher, & Millon, 1999; Millon & Davis, 1993), and modest structural validity (see Newman et al., 2015).

## Results

A series of one-way ANOVAs were used to test for group differences in the 10 categories defining family

structure and the six internalizing scales of the MACI. In regards to the MACI personality patterns, there were no significant mean differences between family structure categories for either the Inhibited/Avoidant scale ( $F(9, 965) = 1.54, p=.13$ ) or the Doleful/Depressive scale ( $F(9, 965) = 1.01, p=.43$ ). Regarding the expressed concerns scales, there were no significant mean differences between family structure categories for the Self-Devaluation scale ( $F(9, 965) = 1.11, p=.35$ ). However, there were significant mean differences for the Family Discord scale ( $F(9, 965) = 6.47, p=.00$ ) with both of the other non-relative and other non-listed categories displaying the highest mean scores and dual parent homes with a biological mother present scoring the lowest. Regarding clinical syndrome scales, there were no significant mean differences for the Depressive Affect scale ( $F(9, 965) = 1.16, p=.32$ ). There were, however, significant mean differences on the Anxious Feelings scale ( $F(9, 964) = 2.69, p=.00$ ). Interestingly, those coming from dual parent homes with maternal involvement scored the highest. In contrast, those from the “other” custodial categories scored the lowest on anxiety. See Table 1 for means and standard deviations, and post-hoc comparisons.

## Discussion

The results from this study provided multiple insights regarding the clinical presentation of this population. Scores from various scales of internalizing features of psychopathology were examined based on family structure type. The only scales found to have significant differences between group differences were the Anxious Feelings scale and the Family Discord scale. When analyzed together, these two scales provide very valuable insight into what may be an explanation for this population's overall high psychopathology rates. Those coming from dual-parent homes with maternal involvement had the lowest family discord but the highest reported anxiety levels. Youth in our sample coming from homes involving a stepparent displayed higher anxiety levels when the biological mother remained, and a stepfather was involved. Whereas those from stepparent homes where the biological father remained and stepmother was introduced displayed higher family discord. This could be particularly relevant for a male sample such as this.

When examining household differences, if some house



holds had significantly higher rates of anxiety or depression, one could expect those households to contain a distressing environment to explain the differences. We found those individuals coming from multiparent homes with only one biological parent (i.e., step-parenting) to have some of the highest anxiety scores along with those in multiparent homes involving both biological parents. This was interesting given the assumption that parent separation or absence is a major source of family conflict for children. It may be the case that those individuals from homes where both biological parents are present, but remain in high conflict rather than separating, are feeling the effects of this sustained conflict. For these individuals, the conflict that remains in the marriage is the source of the anxiety rather than the possible separation.

When measuring family discord, results of the current analysis found those with the lowest scores came from homes with maternal involvement. This included single-parent homes headed by the biological mother as well as homes involving the new marriage of the mother. Despite the stress and potential conflict family structure change may bring, the presence of the maternal figure seems to have a stabilizing force in the expressed internalizing features of these young men. Naturally, the highest reported family discord was among those AISB coming from living situations with little to no familial attachment, such as those from the foster care system. Interestingly though, this group also reported the lowest anxious feelings indicating the possible presence of some sort of emotional numbing to deal with the stressors of these kinds of home environments.

The current best explanation for these differences in outcomes may be the concept of “disrupted caregiving.” Recall that disrupted caregiving is used to describe when a child’s needs are not being met by a parent (Worley et al., 2011). For this sample, this analysis comes from when the adolescents have already entered the correctional facility; those who have come from households with parental figures known for closer attachments are now living without their biological parents for the first time (Sitney et al., 2020). Whereas those who come from households without one or both biological parents are not experiencing additional distress because they did not have the same attachment experience, to begin with. Aligning these findings with previous re-

search that has examined the entangled relationship of mental illness and delinquency, results from this study indicate that family structure pre-adjudication may predict post-adjudication mental health struggles of anxiety in AISBs. This falls in line with findings from the previously conducted longitudinal study by Kim et al. that criminal behavior presumes depressive and anxiety disorders and adds in the role of family structure to the understanding of adolescent development (2019).

### Limitations and Future Directions

The largest limitation of this study is that it is an association test, it is not able to provide details for any causal mechanisms. There are, likewise, additional variables that were not included in this study that could help rule out alternative explanations for the results. First, we did not have access to many family history variables. Hence, we were unable to rule out if a family history of similar psychological disorders could have played a role. Additionally, the only family variables included in this study pertained to the family structure, which leaves various familial characteristics, such as actual living arrangements, trauma, conflict level, and divorce history, among others, unspecified in this study. Analyses of family structure differences using other family environment measures could provide a more in-depth insight into the overall familial influences on these adolescents adjudicated for sexual offenses. Finally, the family grouping variable included an “other” category that may be accounted for a decent amount of variability in living environments. Creating additional categories to fit the needs of this sample could provide further information.

One area that ought to be investigated is the addition of variables that could provide or eliminate alternative explanations for these high prevalence rates. Additionally, we did not expect to have family groupings with a stepfather to score so similarly to families with both biological parents, and thus, examining the role of the step-father in the life of these adjudicated youth could be very illuminating. Another possible route could entail investigating the role that family structure may have in the offense itself. Specifically exploring if there is a pattern between family structure, offense types, and internalizing disorders. Lastly, comparing these results to a replication of this study for the general adolescent population could also highlight key population differ-

ences and explanations.

## Conclusions

This study provides a wide range of evidence for how influential the family can be in the lives of those adjudicated for sexual offenses. Due to the results of this study, treatment providers may want to explore possible family causes of distress if an adolescent begins to portray anxious or depressed behaviors. Additionally, this study is evidence to support the notion that because family is so influential on developing adolescents, their families ought to be included in the treatment process, if and when possible. Results also add to the growing understanding of disrupted caregiving, especially among those who have entered a correctional facility away from their families. The practical significance of this study is that it provides insight into this population and a possible explanation for their rates of psychopathology compared to their peers.

## Acknowledgments

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### Author Biography



Kylie Seaton is a recent 2021 Auburn University alumna; she graduated with Summa Cum honors with her Bachelor of Arts in Psychology and Sociology, with a minor in Spanish. Kylie completed this project during her senior year at Auburn, where she was a member of Dr. Thompson's Juvenile Delinquency Lab. Kylie went on to work as the Lab Manager for the Juvenile Delinquency Lab before attending graduate school.



Kelli R. Thompson is the Director of Research for the Juvenile Delinquency Lab in the Psychology Department. Her lab highlights some of the excellent undergraduate research at Auburn University. This spring semester alone, more than a dozen original student research projects were presented and published at regional conferences and in peer-reviewed journals.

# Sample Entropy of Equestrian Locomotion Treated with a Palmar Digital Nerve Block and Lidocaine Solutions

*Jonathan Ting*<sup>1</sup>, *Dan Marghitu*<sup>2</sup>, and *John Schumacher*<sup>3</sup>

<sup>1</sup> Undergraduate Student, Department of Mechanical Engineering, Auburn University

<sup>2</sup> Professor, Department of Mechanical Engineering, Auburn University

<sup>3</sup> Professor, School of Veterinary Medicine, Auburn University

## Introduction

Randomness and non-periodicity are common phenomena in the areas of science [1]. Whether it is in thermodynamics or just flipping a coin, statistical mechanics, or information theory is a method used to quantitatively measure randomness and regularity as information. Information can be measured with a concept known as entropy. The essence of information entropy is that all events/signals have information based on the probability of an event/signal [2]. Describing a signal by its information content was first proposed by Claude Shannon in 1948 which is known as Shannon Entropy [1–3]. Similarly, as measuring the mass of an object with fundamental units like the pound, Shannon Entropy would measure randomness and variability in units of bits [2]. A high probability event would mean that there is less randomness in the event/signal; thus, there would be less information or bits needed to represent that signal [2]. A low probability would mean that the event is random, and thus would need more bits to represent a signal [2]. Though Shannon entropy is easy to apply, the theory does not consider the dynamics of a system [1]. Shannon entropy is best used for analyzing the randomness of binary events like a coin flip and not signals [1]. Introduced by Pincus, approximate entropy, works well for many signals [1,2,4]. Approximate entropy measures the tendency for segments of a signal to repeat themselves throughout the signal within a given tolerance [1,4,5]. However, approximate entropy is a biased measurement that always underestimates the entropy of a system [1]. This is because a portion of the signal is matched with itself during approximate entropy analysis [1]. This match does not represent a real repeat within the structure because that certain template may never appear again in the signal [1]. Developed by

Richman and Moorman (2000), the sample entropy is an improvement of approximate entropy by alleviating self-matching and thus bias [6]. Just like Shannon entropy, a more random signal would yield a higher entropy value. Sample entropy can be applied to any wave system whether it being periodic or non-periodic in nature [6]. As a result, sample entropy is applied extensively to many chaotic mechanical systems like biological systems [1]. For example, Al-Angari et al., used sample entropy to quantify signal of heart rate variability in obstructive sleep apnea syndrome (OSAS) [7]. They found that patients with OSAS have higher sample entropy during times of unobstructed sleep. From previous research, it seems that healthier cardiovascular systems tend to have a higher entropy value. Raffalt et al. used sample entropy to study the dynamics of the hearts of healthy, active subjects and subjects with a sedentary lifestyle [8]. They discovered that individuals who exercise daily, experience an increase in entropy values compared with the subjects who have adopted an inactive lifestyle. However, in gait analysis, an increase in entropy would mean that the gait would be less periodic. Conversely, this would mean the biomechanical system is less healthy. Ahmadi et al. have studied the effect of human aging on walking gait and discovered that the sample entropy values are greater with increasing age or if the subject is distracted while walking [9]. This is because younger individuals can maintain control of their bodily movements better than older individuals or distracted individuals making it easier for them to restore gait stability under unusual biomechanical situations [9]. Therefore, sample entropy values are lower in younger subjects because the gait is less complex due to their better ability to stabilize their biomechanics. This shows that sample entropy is

<sup>1</sup>Corresponding author: jat0076@auburn.edu

a viable technique for determining the health of gait because it is consistent with observation. Horse lameness is a serious economic and prolong the analgesic effect on the horses [11]. Spoormakers et al. assessed side effects, potency, and duration of anesthetic effect between lidocaine and lidocaine with epinephrine for peripheral nerve blocks. They discovered that the analgesic effect from lidocaine with epinephrine lasted for 9 hours whereas the effect of the pure lidocaine solution lasted roughly an hour [15]. Velloso et al. was able to confirm Spoormaker's study by looking into the effect of anesthetics on the gait of lame horses using vector sums of maximum and minimum head height and the sum of hip drop and hip height. From vector sums, Velloso et al. was able to deduce that adding epinephrine to a lidocaine solution does prolong and intensify the analgesic effect [11]. Though Velloso et al. uses vector sums to determine the lameness of each horse, the simplicity of sample entropy makes the theory applicable to many non-linear systems such as biomechanical gait [2]. Because horse movement is unpredictable, sample entropy may also offer a better measurement to this variability. Thus, sample entropy, in addition with inertial sensors, may offer an indisputable and straightforward method in analyzing analgesic effect from localized anesthesia on equine motion. The objective of this paper is to discuss the theory of sample entropy and apply it in analyzing equine locomotion based on the data received from the inertial sensors. The vertical displacement data of the head, measured with the head sensor, was used to determine the sample entropy of the horse's gait. sample entropy was already applied in studying the motion of horses administered detomidine [16]. However, this paper studies the entropy of horses anesthetized with epinephrine, and lidocaine and to determine whether the addition of epinephrine to a lidocaine solution would prolong and intensify the analgesic effect of lidocaine, as stated by Velloso et al., by applying the theory of sample entropy.

## Materials and Methods

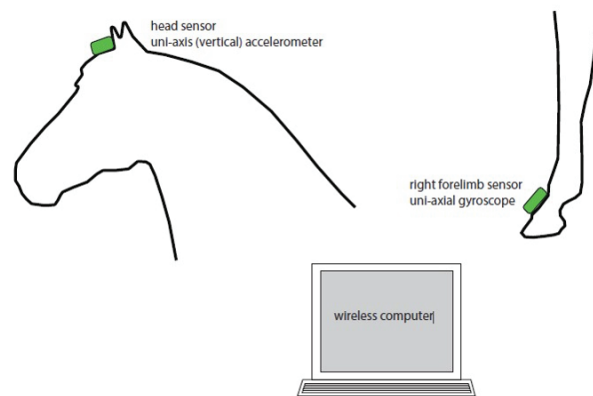
### 2.1. Subjects

All study procedures were reviewed and approved by the Auburn University Animal Care and Use Committee. Six horses with lameness previously localized to the forefoot participated in the study. The mean age of the horses where  $11.5 \pm 4.6$  years old and a mean weight of  $473 \pm 34.5$  pounds. The horses had visually evident

lameness [11].

### 2.2. Sensor application and testing

Two sensors are attached to the horse as shown in Figure 1. Data from the sensors are transferred to a computer via Bluetooth. The first sensor, a single-axis accelerometer, was mounted on the head of the horse; it is used to measure the acceleration of the head during each stride. The second sensor, a single-axis gyroscope, was attached to dorsal surface of the region between the metacarpophalangeal joint and coronary band of the right forelimb to measure the rotation of the foot [11]. Data from the single axis accelerometer was used to determine the entropy of the horse.



**Fig. 1** Sensor application relative to horse.

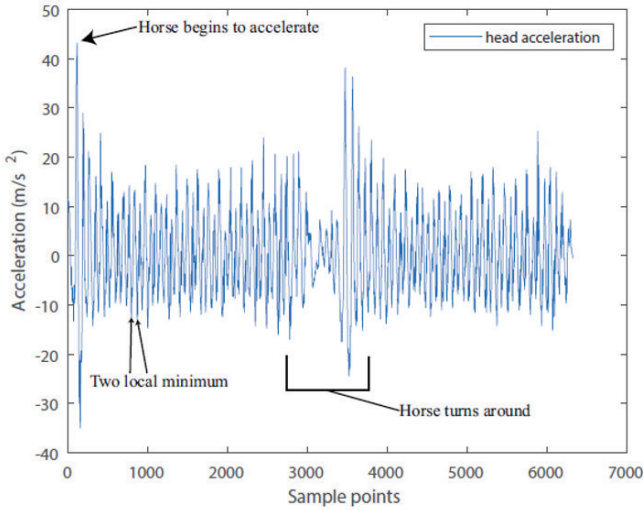
Each horse received a palmar digital nerve block (PDNB) along with three separate lidocaine treatments. The lidocaine treatments involved a 1% lidocaine solution, a 2% lidocaine solution, and a 1% lidocaine solution with epinephrine. Prior to each lidocaine treatment, each horse received a 3-day period between lidocaine treatments to purge residual lidocaine from previous treatments. Before the administration of each lidocaine treatment, the horses trotted 24 strides. Then the horses trotted 5, 10, 15, 20, 25, 30, 45, 60, 75, 90, 105, 110 and 120 minutes after the lidocaine solution was administered to the subjects.

There were two phases of trotting behavior following lidocaine administration. Phase 1 of the treatment was between 0-30 minutes, and phase 2 was between 30-105 minutes after the lidocaine solution was administered. Each horse trotted for at least 12 strides forward, turned around, and trotted 12 strides back. The horses trotted in total of at least 24 strides.



### 2.3. Data post-processing

The head acceleration data was uploaded and processed in MATLAB. First, eighteen strides were separated from the head acceleration data. As shown in Figure 2, two local minimums in the acceleration data are considered as a stride. MATLAB was used to find the location or sample points of each local minimum in the data. That way, eighteen strides can be effectively separated from the data. This process was repeated for the effect of each treatment on the gait of each horse. The purpose of separating the data is to eliminate faulty and needless signals resulting from the horse turning around or the horse accelerating from rest as shown in figure 2.



**Fig. 2** Data separation

The sample entropy algorithm was then applied to the separated data [1,17]. Entropy values obtained from the algorithm were graphed with the administering time as the independent variable. Then the entropy values for all the times after the drugs were administered were averaged between all the horses. A lower entropy value means that the signal is becoming less random and thus the strides are more regular. The average entropy value at 0 minutes was individually subtracted by each of the entropy values, including the initial entropy value, at each time. Subtracting these average entropy values reveals the change in entropy and helps determine the change in regularity of the gait. A negative difference infers that the gait is becoming more regular than at 0 minutes. A Poincare map was applied to all the extracted data to check the regularity of the sample entropy theory.

### 2.4. Sample entropy

The principle of statistical entropy is to quantify information in a signal through probability, as introduced by Shannon [3]. Essentially, sample entropy quantifies a signal's tendency to repeat itself within a given tolerance by measuring its probability for portions of a signal reoccurring in the entire signal. To do this, sample entropy takes portion of a signal and compares its similarity regarding to the rest of the signal. First, when applying sample entropy, all the data points associated with a signal of length  $N$  is separated into vectors of length  $m$ . Take for example, a signal consists of values  $X(1), X(2), X(3), \dots, X(n)$ . If  $m = 2$ , then the signal will be separated into vectors with a length of 2, or in this case,  $[X(1), X(2)], [X(3), X(4)], \dots, [X(n-1), X(n)]$ . The number of separated segments for a signal can be described as:

The separated vectors can then be numbered as  $V_1, V_2, \dots, V(N-m+1)$ . A tolerance  $r$  must be defined before comparing the signals. The absolute difference between each value in one vector,  $V_i$ , with the corresponding values of another vector,  $V_k$ , must be less than  $r$  to satisfy the tolerance and be considered as a match. For two vectors to be considered a match:

$$|V_i(j) - V_k(j)| < r \quad (2)$$

Where  $j$  is any integer ranging from  $1 \leq j \leq m$ . Each vector, excluding itself, is compared with all the vectors that make up the signal. For example,  $V_1$  will be compared with all the vectors, excluding itself, then  $V_2$  is compared with all the vectors, excluding itself, and so on. The number of matches for each segment is denoted as  $nm$  which is divided by the number of separated segments. This results in the probability of the segment from reoccurring in the signal. The probability of each segment is then summed and divided by the number of separated segments to obtain the value denoted as  $C_m$ .  $C_m$  is calculated as:

$$C_m = \frac{\sum_{i=1}^{N-m+1} \frac{nm}{N-m+1}}{N-m+1} \quad (3)$$

The process is repeated; however, the signal is separated into vectors of length  $m + 1$ . The recalculated  $C_m$  value is denoted as  $C_{m+1}$  which is calculated as:

$$C_m = \frac{\sum_{m=1}^{NN-m} \frac{m}{NN-m}}{NN-m} \quad (4)$$

With  $C_m$  and  $C_{m+1}$  values, sample entropy can then be calculated as:

$$S_m = -\log \frac{C_{m+1}}{C_m} \quad (5)$$

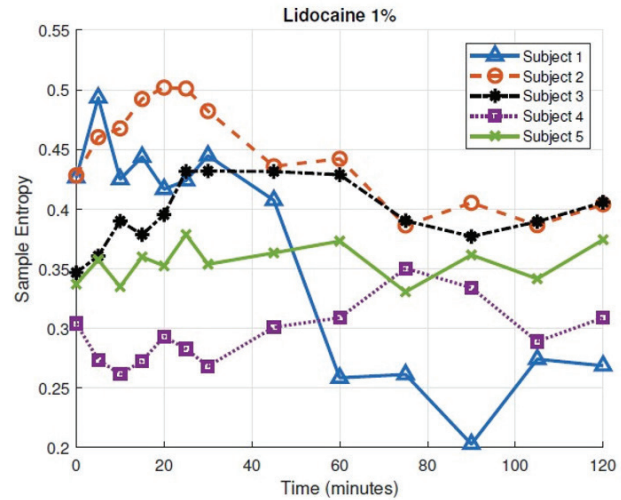
Generally, when a signal is periodic, the probability of a segment repeating itself throughout the signal approaches to 1. This would result in a smaller logarithmic value. Therefore, a more chaotic signal will produce a greater sample entropy value because the probability for signal to repeat itself is lower which, in turn, creates a larger logarithmic value.

A tolerance of 0.15 and a vector length of 2 was established when applying sample entropy. The sample entropy for a sine wave is 0.2244. This is a relatively low sample entropy value because of the sine function's repetitive, non-stochastic nature. For a chaotic signal (Lorentz equation) there is some repetitiveness in the signal, but not to the degree as the sine wave. Therefore, applying sample entropy to this chaotic signal will give a greater sample entropy value of 0.4830. This is to be expected because the chaotic signal is less predictable.

## Results

### 3.1. Treatment 1 (PDNB with 1% lidocaine solution)

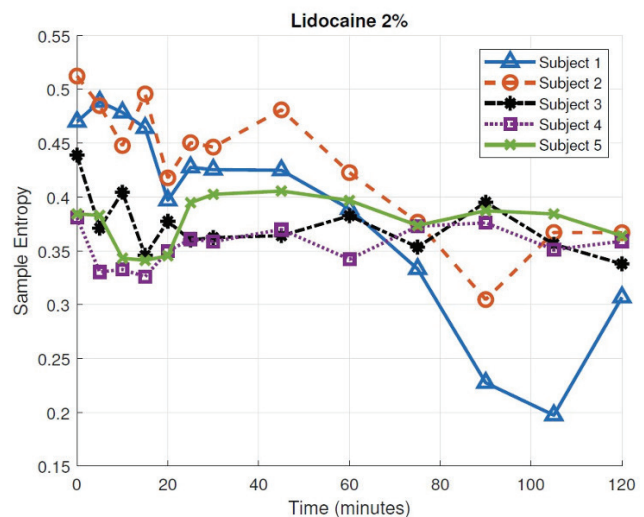
Figure 3 shows the entropy values of all the subjects from 0- 120 minutes receiving the 1% lidocaine treatment (treatment 1). During phase 1 of the treatment, subjects two, three and five experience an increase in entropy throughout phase 1 of the treatment (0-30 minutes). This means that the trotting gait for subjects two three and five are more irregular during phase 1. Thus, the subject is more susceptible to pain, disrupting trotting form. A downward trend in entropy means that the gait for the concerning subject is less chaotic and more regular. This implies that the treatment is improving the gait of the subject. As shown in Figure 3, entropy values for subjects one and four show no meaningful trend throughout the first phase. After phase 1 (30-120 minutes), subjects one, two and three entropy values decrease. Subject 4 has an increase in entropy from 30-75 minutes of treatment then decreases. Subject 5 shows no substantial change in entropy.



**Fig. 3** Entropy values of subjects administered with 1% lidocaine

### 2. Treatment 2 (PDNB with 2% lidocaine solution)

The entropy values for the subjects under a 2% lidocaine treatment are shown in Figure 4. Throughout phase 1, subjects one, two, three and four exhibit a downward trend in entropy, and subject five has a growth in entropy. Subjects three, four and five reveal no trend during phase 2 of treatment. In Figure 4, only subjects one and two show a substantial decrease in entropy throughout phase 2. Subject 1, having the steepest slope, has a sharp rise in entropy at 105 minutes of treatment. A Poincaré map of subject 1's gait at 105 minutes into treatment is shown in Figure 5.



**Fig. 4** Entropy values of subjects administered with 2% lidocaine

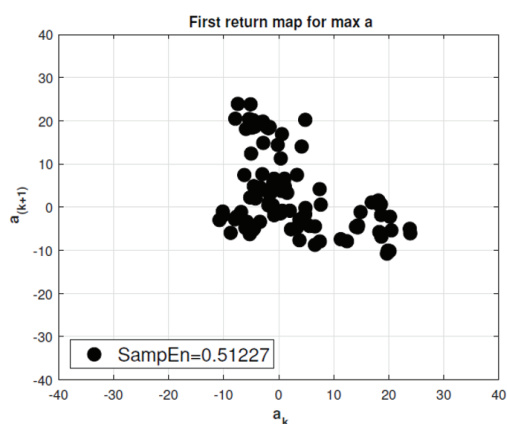
For periodic systems, the map would show one dot.

Conversely, if a signal was quasiperiodic, the Poincare map would show a closed curve distribution of dots. If a signal were to be less random, the Poincare map would show a concentrated cluster of dots. The sample entropy of subject 1 at 105 minutes is the lowest out of all the horses which would mean that the subject has the least random gait. This finding is consistent with the Poincare map shown in Figure 5 where there is condensed grouping of dots. There appears to be fewer dots on the map because of overlapping dots.



**Fig. 5** Poincaré map of subject 1 at 105 minutes into the lidocaine 2% treatment

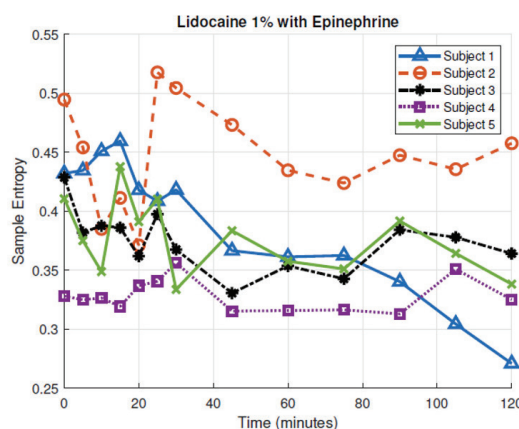
Another Poincare map, shown in Figure 6, was created for subject 2's gait at 0 minutes into the experiment. At this point, subject 2's sample entropy value is high; therefore, it is deduced that the subject has a more random gait. The Poincare map shown in Figure 6 is consistent with the sample entropy theory. Figure 6 shows a random cluster of dots. Unlike the map shown in Figure 5, there appear to have more dots. This is because there are many different peaks in the data due to signal randomness. As a result, more dots are unable to overlap each other, creating a randomly clustered map.



**Fig. 6** Poincare map of subject 2 at 0 minutes into lidocaine 2% treatment.

### 3.3. Treatment 3 (PDNB with lidocaine 1% and epinephrine)

Figure 7 is a graphic showing the entropy values of the subjects experiencing the Lidocaine 1% with Epinephrine treatment. Referencing Figure 7, subjects one and three show a decrease in entropy and subject 4 experiences an increase in entropy during phase 1. Because of the drastic change in entropy in subjects two and five, there is no trend in entropy for these subjects in phase 1. However, subjects two and five have a reduced entropy throughout phase 2 despite subject five having a milder change in entropy. Additionally, subjects one and three have a decrease in entropy, but, like subject 5, subject 3 has a smaller change in entropy. Subject 4 has no substantial change in entropy.

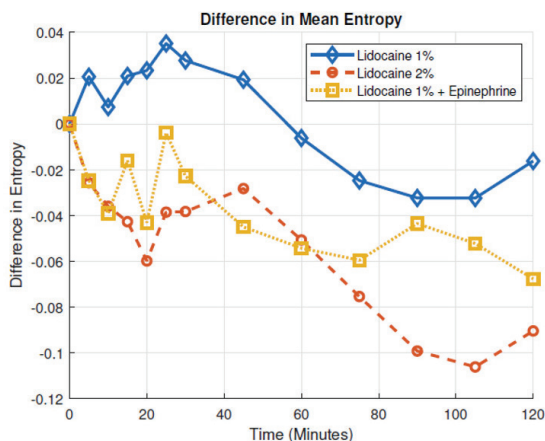


**Fig. 7** Entropy values of subjects administered with lidocaine 1% with Epinephrine

### 3.4. Difference in mean entropy

To clearly see if entropy has changed for the subjects after each treatment, the entropy values were averaged for each time for all three treatments. All the average entropy values were then subtracted with the initial average entropy. Figure 8 shows the change in average entropy for all subjects and their respective treatments. As suspected, there is a delayed reaction during phase 1 of the lidocaine 1% and epinephrine treatment; however, after phase 1, there is a steady decline in entropy for the lidocaine 2% and lidocaine 1% with epinephrine treatments. For the Lidocaine 1% treatment, the mean entropy values increase with respect to the average initial entropy during phase 1. Though the en

trophy exhibits a negative change during phase 2 for the lidocaine 1% treatment, it is less drastic compared to the other two treatments. The lidocaine 2% treatment has a negative change in entropy in all phases of treatment. Negative change in mean entropy correlates to a more regular gait. Schumacher notes that the Lidocaine 1% and Lidocaine 2% effect diminishes near the end of the treatment. However, Figure 8 challenges this by showing the decreasing difference in entropy at minutes. This means that the gait for all the subjects is more regular despite the lessening effect of the drugs.



**Fig. 8** Difference in mean entropy of all subjects.

## Conclusions

The purpose of this study was to determine whether lidocaine in addition with epinephrine and a palmar digital nerve block could make the gait more regular for a longer time. By applying sample entropy, it is confirmed that lidocaine and epinephrine with a palmar digital nerve block suppresses erratic head movement for greater duration of time.

## Statment of Research Advisor

Jonathan Ting has done great work in implementing a more streamlined and less invasive method in diagnosing equine lameness. Based on analysis, we have determined that sample entropy values decrease more significantly in the lidocaine 2% and lidocaine 1% with epinephrine analgesic solutions.

-Dan Marghitu, Samuel Ginn College of Engineering

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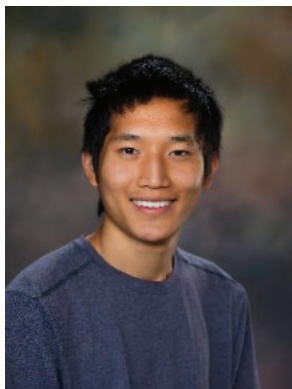


Dr. Dan Marghitu is a Professor in the Samuel Ginn School of Engineering at Auburn University. He is a mechanical engineering professor and is known for teaching classes like vibrations and kinematics and dynamics of robotics.



Dr. John Schumacher is a Professor Emeritus for the School of Veterinary Medicine at Auburn University. His research interests involve the study of equine motion, particularly developing novel and refined diagnostic methods for localizing the source of pain in equine lameness.

## Author Biography



Jonathan Ting is a senior- year student pursuing a B.S. degree in Mechanical Engineering at Auburn University. He has played a key research role in the study *Sample Entropy of Equestrian Locomotion Treated with a Palmar Digital Nerve Block and Lidocaine Solutions*.

# Extraction of DNA from Honeybee, *Apis mellifera*, Beeswax for Genetic Analysis of the Microbiome

Kylie S. Weis<sup>1</sup>, Neha Potnis<sup>2</sup>, and Geoffrey Williams<sup>2</sup>

<sup>1</sup> Undergraduate Student, College of Agriculture, Auburn University

<sup>2</sup> Professor, Department of Plant Pathology and Entomology, Auburn University

## Abstract

Beeswax has the potential to harbor a diverse microbiome that may be an indicator of honeybee colony health. In order to characterize the microbiome with a culture-independent method, DNA must first be extracted from the beeswax, amplified by PCR, and sequenced. Currently, there is no established protocol for DNA extraction from beeswax. The properties of beeswax can make DNA extraction difficult and inhibit PCR. To find an effective protocol of DNA extraction for sequencing, various beeswax sampling tools were used in combination with different DNA extraction methods in this study. Samples were then assessed for their ability to amplify by PCR. Multiple methods yielded amplification by PCR with varying degrees of success.

## Introduction

The honeybee, *Apis mellifera*, is an important financial asset for beekeepers in addition to being essential for the agricultural industry via pollination (Calderone, 2012). However, the Bee Informed Partnership (BIP) National Colony Loss and Management Survey reported that US beekeepers experienced an estimated loss of 32.2% of their colonies during the winter of 2020-2021 (Steinhauer et al., 2021). The current consensus for the cause of colony loss is a conglomerate of stress factors such as parasitic mites, insecticides, food shortages, weather, and pathogens (Neumann & Carreck, 2010).

While studies have been conducted on pathogens in the hive environment, these studies are often of small scope, only utilize culturing methods, and focus on one pathogen rather than the entire microbiome. Technologies exist to sequence DNA and identify microorganisms found within the microbiome; however, these methods have not been readily used for investigating the microbiome of beeswax. This is likely due to difficulties in extracting DNA from wax.

DNA extraction methods use the chemical properties of DNA and contaminants to yield purified DNA for downstream use in PCR or sequencing. Various methods and commercial kits are available for DNA extraction. Kits, such as Qiagen's DNeasy PowerSoil Pro Kit and DNeasy PowerFood Microbial Kit, are designed to extract DNA from certain sample types and to remove specific contaminants from those samples that would inhibit PCR. Numerous substances, including proteins, polysaccharides, phenols, and lipids, can inhibit PCR and prevent amplification of the 16S region used for DNA sequencing (Richards, 1999). While the composition can vary, beeswax contains hundreds of compounds, including a large amount of lipids and proteins (Hepburn, 1986). Due to the hydrophobic nature of lipids, problems can occur in DNA extractions that rely on aqueous solutions. The properties of lipids complicate not only the separation of microorganism cells from the beeswax but also the removal of PCR inhibiting contaminants from the sample. Preliminary attempts at DNA extraction from beeswax have resulted in excess contamination or the degradation and loss of DNA from multiple purification steps.

The goal of this study is to assess multiple sampling tool types and DNA extraction methods to optimize a protocol for DNA extraction from beeswax. This protocol can then be used to analyze the hive microbiome and gain better insight into honeybee health and unexplained colony loss.

## Methods Sampling

To determine the best area for microbiome sampling within the interior hive environment, samples were taken with a sterile cotton swab from the hive entrance, the top of a frame, the interior wall of a brood

<sup>1</sup>Corresponding author: ksw0029@auburn.edu

box, the backs of bees, beeswax covering the brood, and inside an open cell. The swabs were sonicated in 0.01% MgSO<sub>4</sub> buffer and spiral plated on nutrient-rich agar. The plates were incubated for 3 days at 37° C, and bacterial colonies were counted. Samples taken from beeswax covering brood produced the highest number of colonies (Fig. 1), likely due to the high amount of contact with bees and high nutrient content from the beeswax composition. Beeswax covering brood was, therefore, determined to be the best area to focus on for this study.

Samples were taken from the beeswax covering brood of five unrelated honeybee colonies. Three types of sampling tools, a dry sterile cotton swab, a cotton swab saturated with 0.01% MgSO<sub>4</sub> buffer, and autoclaved flat wooden picks, were selected for use. Both dry and buffered cotton swabs were twirled with moderate pressure over the beeswax covering brood in an 8x8 cm area for 30 seconds. The wooden picks were inserted into the beeswax covering brood to remove the capping containing approximately 100mg to 250mg of beeswax. Dry swabs and the wooden pick samples were stored in microcentrifuge tubes and temporarily kept on ice until placed in a -80° C freezer. Buffered swabs were kept on ice and underwent DNA extraction within two hours after sampling.

### DNA Extraction

Four different DNA extraction methods were used in this study. The Qiagen DNeasy PowerSoil Pro Kit was selected since it is designed for small quantities of DNA, uses a bead beating tube for spore extraction and a spin filter column, and removes most PCR inhibitors. The Qiagen DNeasy PowerFood Microbial Kit was selected as it is designed for small DNA quantities and uses a bead beating tube and a spin filter column. It is designed to remove PCR inhibitors specific to foods such as lipids. The Bio-Rad InstaGene Matrix, which utilizes chelating particles to remove PCR inhibitors, was selected for both cost and ease of use. Crude preparation was also selected for cost and ease of use. The Qiagen PowerSoil Pro Kit, Qiagen PowerFood Microbial Kit, and Bio-Rad InstaGene Matrix were used according to manufacturer directions for each sample tool type for all 5 honeybee hives sampled. An exception is that 400ul instead of 200ul of InstaGene Matrix solution was used per sample to ensure coverage of the swabs and to

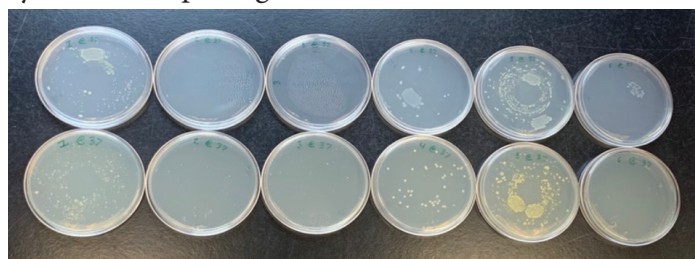
increase inhibitor removal capacity. Furthermore, the wooden pick or cotton swab was used in place of soil or food for the respective kit. The crude preparation of DNA extractions involved adding 500ul of nuclease free water to the samples, vortexing, and boiling at 101° C for 10 minutes. Samples were vortexed again and centrifuged at 12,000g for 2 minutes. The supernatant was used as a PCR template. All samples, except crude preparation samples, were analyzed by NanoDrop for DNA concentration and purity.

### PCR

The 16S rRNA gene PCR amplification was performed using universal primers 27F (5'-AGAGTTTGATYMTGGCTCAG-3') and 1492R (5'-GGTTACCTTGTTACGACTT-3'). The amplifications were carried out under the following conditions: initial denaturation at 95°C for 10min, followed by 35 cycles of denaturation at 95°C for 15 seconds, primer annealing at 50°C for 60 seconds, and extension at 72°C for 120 seconds, with a final elongation at 72°C for 10 min. For each PCR reaction, the following was used: 5ul of template, 1.5ul MgCl<sub>2</sub>, 5.0ul buffer, 0.5ul dNTPs, 2.0ul 27F primer, 2.0ul 1492R primer, 0.20ul Taq polymerase, and 8.5ul nuclease free water.

### Results

Culturing of the samples from different areas within the hive yielded varying results (Fig. 1). Samples from beeswax covering brood produced the largest number of bacterial colonies with at least three distinct morphologies seen. There were no bacterial colonies obtained from the wooded surfaces in the interior of the hive, although divots were observed in the media. Diverse bacterial colonies were obtained from the hive entrance; however, more growth was seen for these samples at 27°C than 37°C, indicating, along with the morphology of some of the colonies, that these bacteria were likely environmental species and not honeybee symbionts or pathogens.



**Fig. 1.** Cultures from various areas of the honeybee



hive. The top row is minimal media, and the bottom row is nutrient-rich agar. From left to right, the samples were taken from the hive entrance, top of the frame, wall of the brood box, backs of bees, beeswax covering capped brood, and inside an open cell.

Gel electrophoresis was used to confirm that after extraction, samples could be amplified by 16S PCR (Fig. 2). All samples taken with any of the three sampling tools used with the Qiagen PowerSoil Pro Kit were able to be amplified. Samples used with Qiagen PowerFood Microbial Kit were able to be amplified when samples were taken with a wooden pick or a dry cotton swab. The buffer saturated swabs had mixed success, with only 3 out of 5 samples amplifying. DNA extraction with InstaGene Matrix produced minimal amplification with the dry cotton swab and no amplification with wood or buffered swabs. Crude preparation did not produce any amplification. The band seen for dry swabs under C2 in Fig. 2 was due to a gel loading error.

Bands showing that the 16S rRNA region of bacteria was able to be amplified appeared for the negative controls for all sample collection tool types. The negative controls, in this case, consisted of an unused swab or wooden pick. Sanger sequencing of the negative controls showed that the contamination came from shared lab equipment.

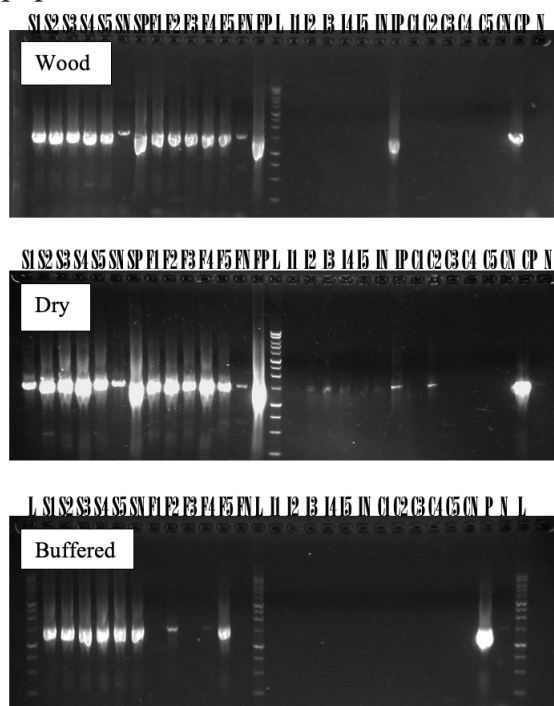


Fig. 2. Gel electrophoresis results of 16S PCR, where S

refers to samples extracted with Qiagen PowerSoil Pro Kit, F refers to Qiagen PowerFood Kit, I refers to Bio-Rad InstaGene Matrix, C refers to crude preparation, P refers to the positive control, N refers to a negative control, and L refers to a 1kb ladder. A white band at around 1,500bp indicates successful amplification of the 16S rRNA gene region. Note that the brightness of bands does not indicate amplified DNA quantity. The gel for dry swabs was overexposed to better visualize the InstaGene bands.

Before PCR, samples were analyzed for DNA concentration and purity with NanoDrop. Table 1 lists the average concentration of DNA in ng/ul for sample tool type and DNA extraction method. On average, the samples collected with wooden picks contained more DNA than the other sampling tools. The dry swabs and buffered swabs had comparable amounts of DNA. It should be noted that the MgSO<sub>4</sub> buffer contained bacterial contamination that may have inflated the amount of bacteria, and therefore DNA, that can be collected by a buffer-saturated swab. Samples processed with the Qiagen PowerFood Microbial Kit had a DNA concentration double that of the Qiagen PowerSoil Pro Kit. Although the InstaGene method produced a high concentration of DNA, this method did not yield consistent PCR results.

**Table 1.** Average DNA in ng/ul extracted from the Qiagen PowerSoil Pro Kit, the Qiagen PowerFood Kit, and InstaGene Matrix for the various collection tool types.

	Soil Kit	Food Kit	InstaGene
Dry swab	3.62	6.92	115.26
Buffered swab	3.28	7.00	100.64
Wood	5.80	18.96	608.94

The presence of contamination was determined by NanoDrop spectroscopy through 260/280 and 260/230 absorbance ratios. The average readings for samples that could be amplified by PCR were 17.1 ng/ul with a 260/280 ratio of 1.88 and a 260/230 ratio of 0.15. The average readings for samples that were not amplified were 218 ng/ul with a 260/280 ratio of 1.15 and a 260/230 ratio of 0.41. A T-test determined the readings for concentration, 260/280 ratio, and 260/230 ratio to be significantly different between the samples that were able to be amplified by PCR and the samples that were



not able to be amplified ( $p < 0.005$ ).

A sample taken with a dry cotton swab extracted with the Qiagen PowerSoil kit was able to be sequenced using Illumina Miseq with an acceptable ( $>10,000$ ) number of reads. Samples used with other extraction methods have not yet been submitted for sequencing.

## Discussion

The resulting bacterial colony growth from the different hive areas leads to further questions that can be investigated in future studies. Investigating why there are no culturable bacterial colonies from the interior wooden surfaces of the hive could lead to new knowledge regarding the i-microbial properties of the hive environment. Further studies concerning the bacterial composition of the microbiome found at the hive entrance can provide insight into the vector ecology of honeybees as carriers of environmental bacteria such as those that may be plant pathogenic.

Although there appears to be bacterial contamination in all DNA extraction samples, this serves to demonstrate which DNA extraction methods can be successful when bacteria are known to be present. Among the protocols tested, using a wooden pick or dry cotton swab and either the Qiagen PowerSoil Pro Kit or Qiagen PowerFood Microbial Kit was the best option for ensuring amplification of the 16S rRNA gene region, which can then be used for sequencing. Since bacteria are not highly abundant inside the hive, using a wooden pick that can collect a larger sample and the Qiagen PowerFood Microbial Kit, which yielded the highest DNA concentrations of the methods that worked, may be the best option for assessing the microbiome. If cost is a factor, the Qiagen PowerFood Microbial Kit is the more economical option compared to the Qiagen PowerSoil Pro Kit. It should be noted that the bead beating tubes in the Qiagen PowerFood Microbial Kit is too small for the swabs or wooden picks. The beads may be transferred to the microcentrifuge tube used for sample collection. The inconsistent results of the Qiagen PowerFood Microbial Kit, when used with the buffered swabs, show that there may be an incompatibility with the kit and  $MgSO_4$  and, therefore, should not be used together.

Wooden picks may sample more invariably than cot-

ton swabs. Unless the wooden picks are weighed before, and after sampling to record the mass of beeswax taken, the samples may vary significantly. Meanwhile, swabbing an 8x8 cm area for 30 seconds generally saturates a dry swab with wax and normalizes the samples.

Results indicate that higher concentrations of DNA after extraction do not indicate a greater likelihood of being amplified by PCR. Instead, lower concentrations of extracted DNA are associated with amplification ability and may indicate a higher degree of purification. However, 260/280 ratios seem to be most indicative of the ability to amplify. A reading of 1.8 for a 260/280 ratio is considered pure for DNA (Thermo Scientific, 2012). The samples that were able to be amplified had an average 260/280 ratio of 1.88, while the samples that were unable to amplify had a ratio that was significantly lower. A ratio of 2.0 or more for 260/230 ratios indicates pure DNA, with lower values indicating contamination. The 260/230 ratios seen in this study were all significantly lower than 2.0. However, this did not affect the ability to amplify; in fact, the group that was able to be amplified had a lower 260/230 ratio. Low 260/230 ratios were found to have no significant effect on the ability of RNA samples to successfully undergo qRT-PCR (Cincinnati et al., 2008). In this case, low 260/230 ratios can be disregarded as an indicator of potential PCR inhibition.

While previously, there has not been an established protocol for DNA extraction from honeybee beeswax, this study shows that DNA extraction from beeswax, 16S rRNA gene amplification, and sequencing of the hive microbiome is possible. This will allow for future studies of the hive microbiome that may show the microbiome to be an indicator of honeybee colony health.

## Acknowledgments

Gratitude is extended to the Auburn University College of Agriculture and the Office of Undergraduate Research for partially funding this project. The guidance, shared expertise, lab resources, and financial support of Dr. Neha Potnis was highly appreciated. Credit for the 16S PCR protocol goes to Rishi Bhandari. The assistance of the AU-Bees lab members Dan Aurell, Selina Bruckner, and Clinton Wall was essential to sampling and maintaining the honeybee colonies.

-Geoffrey Williams, *Entomology and Plant Pathology*

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researching the genetic components of plant-microbe interactions.



Neha Potnis, Ph.D. is a Professor at Auburn University, where she began working in 2016. She earned her Ph.D. in Plant Pathology at the University of Florida and is an NSF CAREER award recipient. Her work includes investigating host specificity and virulence in *Xanthomonas* species affecting tomato and pepper.



Geoffrey Williams, Ph.D. is a Professor at Auburn University, where he began working in 2016. He earned his Ph.D. in Biology at Dalhousie University, Canada. He is the President of the Bee Informed Partnership and on the executive board of COLOSS. His work includes studying the effects of parasites and pesticides on honeybee health.

## Author Biography



Kylie S. Weis is a third-year student pursuing a B.S. degree in Applied Biotechnology at Auburn University. She has led key research roles in experimental design, sampling, lab work, and analysis for this project. She has a background in plant pathology and plans to continue her education by

# Comparing Small and Medium-Sized Outbursts Of Comet 29p

Johannes J. Allen<sup>1</sup>, Youssef Moulane<sup>2</sup>, Dennis Bodewits<sup>3</sup>

<sup>1</sup>Undergraduate Student, Department of Aerospace Engineering and Physics, Auburn University

<sup>2</sup>Postdoctoral Researcher, Department of Physics, Auburn University

<sup>3</sup>Associate Professor, Department of Physics, Auburn University

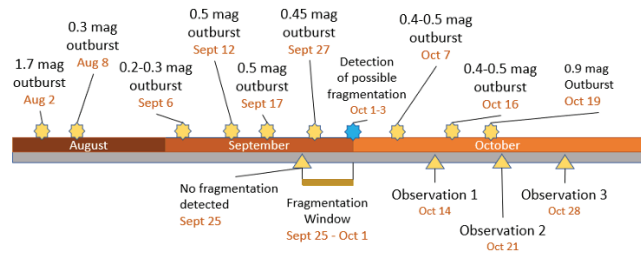
Comet 29P/Schwassmann-Wachmann<sup>1</sup> (hereafter 29P) is a highly active member of the centaur population, notorious for its unusual outburst behavior. Hovering at large heliocentric distances, 29P is too cold for the sublimation of water ice to drive its outbursts. Despite the low temperatures, frequent outbursts of differing sizes have been observed for 29P. This unusually high frequency and magnitude of outbursts at such cold temperatures have left astronomers puzzled. With data collected from the Hubble Space Telescope (HST) during a medium-sized and a small-sized outburst of 29P, the differences between outbursts of two different magnitudes were explored. Understanding the differences between small and medium-sized outbursts of 29P can provide insight into how the characteristics of its outbursts scale with size.

Despite having an orbit and size consistent with an Asteroid, 29P has a composition and outburst behavior similar to that of a Comet, thus classifying it as a Centaur. Centaurs are small icy bodies with orbits between Jupiter and Neptune, which are thought to dynamically link the Jupiter-Family Comets (JFCs) to the Trans-Neptunian Objects (TNOs) of the outer solar system. Based on their composition and outburst behavior, many Centaurs, like 29P, are often referred to as both Comets and Centaurs. Although the outburst mechanism of most comets is the sublimation of water ice, 29P is too cold for its water ice to sublimate enough to drive an outburst. For this reason, the mechanism driving 29P's outbursts is currently unknown. However, what is known, is that 29P does exhibit frequent outbursts of various magnitudes.

**Table 1** Filters used for observations.

Filter	Range (nm)	$\lambda_{eff}$ (nm)	Color
F689M	6450 - 7325	6873	Red
F487N	4828 - 4918	4874	Cyan
F845M	7895 - 9018	8431	Red-IR
F350LP	3197 - 10710	5558	Visible

The Target of Opportunity (ToO) observations collected with the HST were acquired following reports of a possible fragmentation event of 29P before Oct. 1st, 2019 [1]. The observing cadence captured 29P within two days of a 0.9 magnitude, and thus medium-sized, outburst. This set of observations provided data both preceding and following this 0.9 magnitude outburst, which would allow us to probe the development of the ejecta. Between the purported fragmentation event and the HST observations, 29P underwent two small outbursts of magnitude 0.4-0.5 on Oct. 7th [2] and Oct. 16th of 2019 [3], followed by the medium-sized 0.9 magnitude outburst around Oct. 19th, 2019 [4]. A timeline of the observation window, purported fragmentation window, and reported outbursts from August to October of 2019 is given in Fig. 1.



**Fig. 1** Timeline of reported outbursts of 29P around the time of observation.

Three sets of observations were conducted using the

<sup>1</sup>Corresponding Author: jza0052@auburn.edu

HST in October of 2019. The first set is a collection of eight images on Oct. 14th, 2 in the F689M filter, 2 in the F845M filter, and 4 in the F487N filter. The second set is also eight images collected on Oct. 21st, using the same three filters and their respective number of images. The third set is a collection of 12 images on Oct. 28th, 4 in the F689M filter and 8 in the F350LP filter. Table 1 shows the wavelengths and corresponding colors of these 4 filters.

One of the most important pieces of information for characterizing and comparing comet outbursts is the flux. The flux is a measure of how much light from an object reaches the detector. A python script was written to calculate the flux of the comet within a 0.8-arcsecond aperture. The flux values were then separated into data subsets based on their respective filter, such that a plot of flux vs. time could be made for each filter (Fig. 2). These plots show a significant increase in the flux between Oct. 14th and Oct. 21st, which is consistent with the reported 0.9 magnitude outburst observed on Oct. 19th [1]. The significant increase in flux was observed in all three filters used across those two observation dates. There was a 26% increase in the Red-IR band, a 31% increase in the red band, and a 67% increase in the Cyan band. In addition to the 0.9 magnitude outburst occurring on Oct. 19th, a much smaller 0.2 magnitude outburst is thought to have occurred on Oct. 28th. As shown in the F350LP and F689M plots, a noticeable jump in the flux occurs between 12:40 and 13:40 on Oct. 28th. In the full visible spectrum, the flux increased by 18% within one hour before continuing to fall back down. This jump in the flux is also seen as an 11% increase in the red band filter.

## Statement of Research Advisor

Johannes worked on the analysis of Hubble Space Telescope (HST) observations of an object far from the Sun that is continuously out-bursting. In his project, he developed a detailed timeline of various outburst events surrounding the HST observations, which provides important context for the interpretation of our data. He prepared the HST data by carefully removing cosmic rays contaminations, and by further processing the telescope images he isolated and analyzed the properties of the material ejected by different kinds of outburst.

- Dennis Bodewits, College of Science and Mathematics

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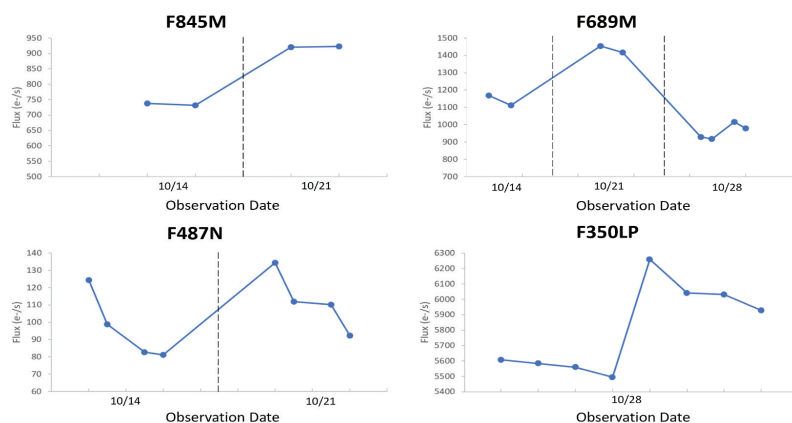


Fig. 2 Flux vs. Observation date for all 4 filters.



## Authors Biography



Johannes J. Allen is a senior completing concurrent B.S. degrees in Aerospace Engineering and Physics at Auburn University. He has been conducting research in the Cometary Science research group at Auburn for the last two years. In addition to his research work, he is a Teaching Assistant for Physics and the president and co-founder of the Astronomical Society at Auburn Univ.



Youssef Moulane, Ph.D., is a postdoctoral researcher in the Physics department at Auburn University. He got his B.S. and M.S. in High Energy Physics & Astrophysics at Cadi Ayyad University in Morocco and his Ph.D. in Astronomy and Astrophysics at the University of Liege in Belgium. While getting his Ph.D., he also did research at Le Havre University in France and the European Southern Observatory in Chile.



Dennis Bodewits, Ph.D., is an Associate Professor in the Auburn University Physics Department. After getting his Ph.D. in Laboratory Astrophysics at the University of Groningen in the Netherlands, he worked as a Research Scientist at NASA Goddard Space Flight Center and the University of Maryland. He also has an asteroid named after him, “10033 Bodewits”!

# Nanocellulose-based Antimicrobial Systems for Mitigating *E. coli* outbreaks in water bodies

Brianne Dickson<sup>1</sup>, Gabriel Au<sup>2</sup>, Diego Gomez-Maldonado<sup>3</sup>, Brendan Higgins<sup>4</sup>, and Maria Soledad Peresin<sup>5</sup>

<sup>1</sup>Undergraduate Student, College of Science and Mathematics, Auburn University

<sup>2</sup> Undergraduate Student, Department of Biophysics, John Hopkins University

<sup>3</sup>Post-Doctoral Research Professor, College of Forestry and Wildlife Sciences, Auburn University

<sup>4</sup>Assistant Professor, Biosystems Engineering Department, Auburn University

<sup>5</sup>Assistant Professor, College of Forestry and Wildlife Sciences, Auburn University

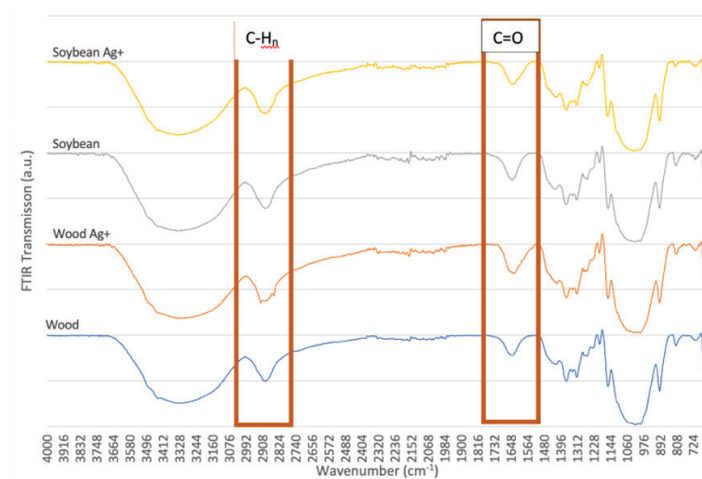
In Alabama, there has been a significant *E. coli* pollution problem in water sources due to an overabundant feral hog population. A study tested the impacts these feral hogs have on the water quality and found that in treatment watersheds, the values of *E. coli* were up to 40 times greater than the reference watershed due to fecal material (Bolds et al., 2021). This is an obvious threat to entire populations that have their water sources contaminated by hogs.

One solution to this problem may be the use of silver nanoparticles, which are well known for their antimicrobial properties. Silver ions liberated by them can enter the cells through pores in the membrane, damaging proteins and DNA, disrupting the functioning of the cell, and resulting in cell death (Liao et al., 2019). For silver nanoparticles to function, they first need to be anchored to other structures to control the release and long-term use of these silver ions. Nanocellulose-based hydrogels are an attractive alternative as they can be used as reactants for synthesis while serving as an anchoring structure.

Cellulose nanofibrils are attractive due to their morphology, high surface area, low density, and sustainable sourcing. Cellulose can form hydrogel structures which can be used as scaffolds for synthesizing silver nanoparticles. To form them, premade nanocellulose spheres are immersed into Tollens' reagent where the hydroxyl and carboxyl groups in cellulose fibers interact with the silver ammonium oxidizing to aldehyde groups. Silver interacts with these fibers, building silver seeds and once heated, crystallizes to form silver nanoparticles (Wu et al., 2014).

The objectives of this project were to generate silver nanoparticles using nanocellulose spheres from different raw materials as scaffolds and test their efficiency at inhibiting *E. coli* growth. Two common raw materials, wood and soybean hulls, were chosen to determine which has the best-reducing capacity.

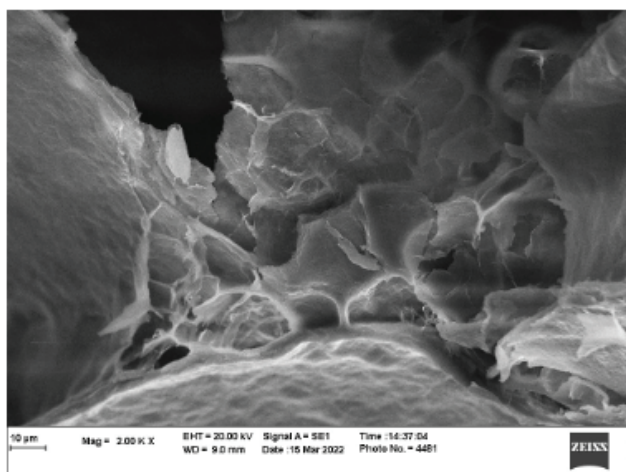
Characterization using Fourier-transform infrared spectroscopy (Figure 1) showed a change in the carbon hydrogen-bond due to aldehyde formation. There is also a decrease in carboxyl groups as they are used during the reaction.



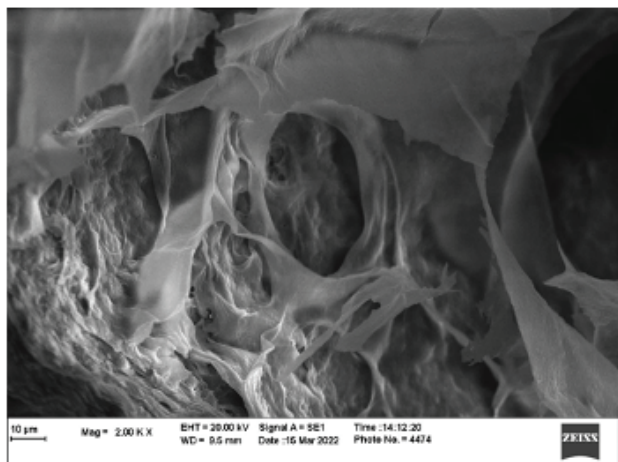
**Figure 1.** Fourier Transform Infrared Spectroscopy of Four Materials

Scanning electron microscopy (Figure 2-5) showed an increase in rugosity in the presence of silver nanoparticles, inferring that the higher presence in soybean-spheres might be caused by the additional components such as pectin.

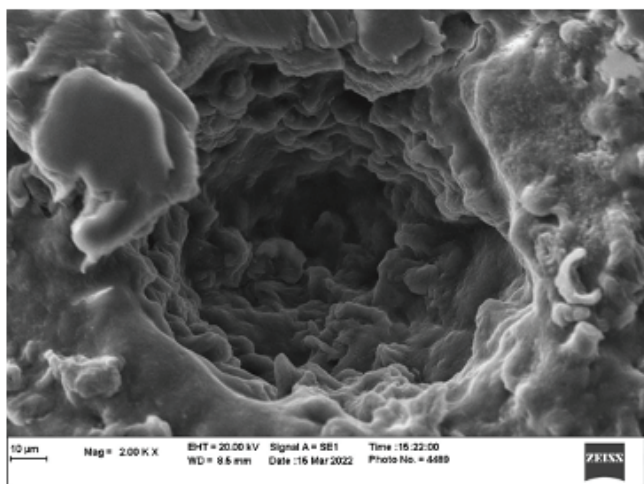
<sup>1</sup>Corresponding Author: brd0011@auburn.edu



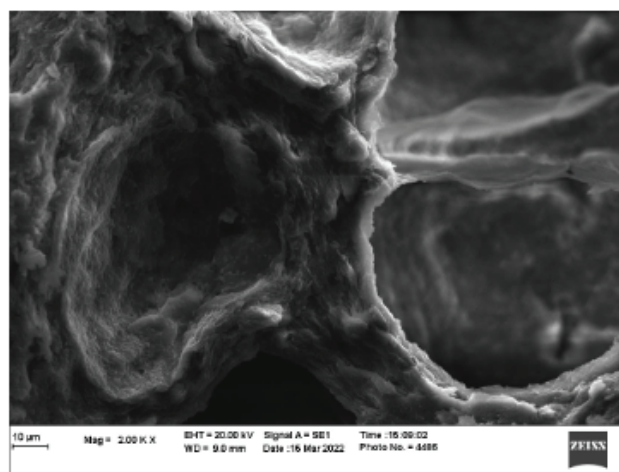
**Figure 2.** Scanning Electron Microscopy Wood with Silver



**Figure 3.** Scanning Electron Microscopy Wood without Silver.

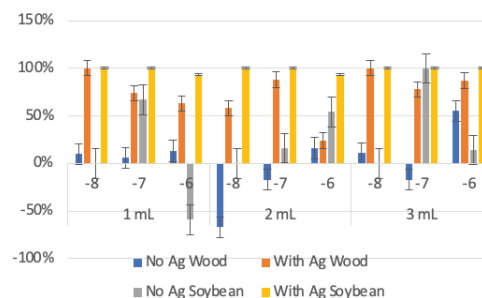


**Figure 4.** Scanning Electron Microscopy Soybean with Silver.



**Figure 5.** Scanning Electron Microscopy Soybean without Silver.

A bacterial suspension was performed through serial dilutions to obtain bacterial concentrations similar to those found in watersheds. These bacterial concentrations include 10<sup>-8</sup>, 10<sup>-7</sup>, and 10<sup>-6</sup> colony forming units. Ten milliliters of each concentration were obtained, and three milliliters of each concentration were run through tubes at volumes of 48.4-, 10.89-, and 16.64-millimeters of spheres. The last milliliter of the solution was collected and plated along with a control plate. Plates were placed in an incubator at 37 °C overnight, and the resulting colonies were counted and recorded. Figure 6 indicate the antimicrobial efficacy in wood and soybean spheres with and without silver nanoparticles. The 1mL, 2mL and 3mL on Figure 6 indicate the depths of spheres in the tube and the -8, -7, and -6 indicate the bacterial concentrations. Overall, we found that there is an increase in antimicrobial efficacy with silver nanoparticles. Silver nanoparticles in the soybean spheres seem to have greater efficacy than wood.



**Figure 6.** Comparison of Antimicrobial Efficacy in Wood and Soybean Spheres with and without Silver



Nanoparticles.

### Statement of Research Advisor

During this project, Brienne has developed a method to produce nanocellulose-based hydrogel beads and decorated them with silver nanoparticles for reducing *E. coli* content in water. This work advances our understanding of this system so we can move forward with field trials that has great potential for alleviate environmental concerns related to the bacterial contamination in water bodies.

-Maria Soledad Peresin, Forestry Wildlife and Sciences

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### Authors Biography



Brienne Dickson is a Senior pursuing a B.S. degree in Microbiology at Auburn University with plans to pursue PA school upon graduation. She has been working as an undergraduate research fellow in the Sustainable Bio-based Materials Laboratory.



Gabriel is a sophomore currently pursuing a degree in biophysics at Johns Hopkins University. At the Sustainable Bio-based Materials Laboratory, he helped develop antimicrobial and antigen assays as an NSF CAREER intern.



Diego Gomez-Maldonado is a Post-Doctoral Research Fellow in the Sustainable Bio-based Materials Laboratory at Auburn University. He is very active in helping undergraduate students expand their knowledge with research.



Brendan Higgins is an assistant professor in Biosystems Engineering at Auburn University. He studies biological processes that incorporate algae and bacteria for waste remediation, water quality, biofuels, and high-value nutraceuticals.



Maria Soledad Peresin is an assistant professor of forest biomaterials in the Auburn University College of Forestry and Wildlife Sciences. She is the principal investigator for the Sustainable Bio-based Materials Laboratory.



# Determination of the Effectiveness of a Novel Liposome Formulation of Proteasome Inhibitors

Laura E. Downey<sup>1</sup>, Andrey V. Maksimenko<sup>2</sup>, Robert D. Arnold<sup>3</sup>, Alexei F. Kisselev<sup>4</sup>

<sup>1</sup> Undergraduate Student, Department of Science and Mathematics, Auburn University

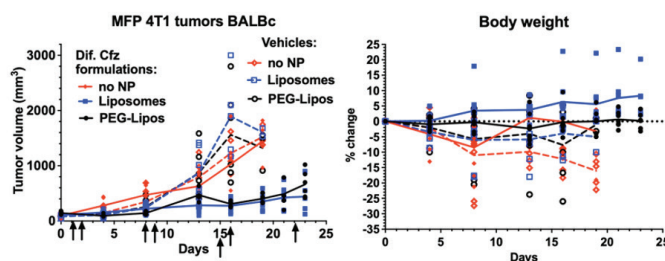
<sup>2</sup> Visiting Scientist, Harrison College of Pharmacy, Auburn University

<sup>3</sup> Professor, Harrison College of Pharmacy, Auburn University

<sup>4</sup> Associate Professor, Harrison College of Pharmacy, Auburn University

Proteasomes are multi-subunit protein recycling machines of cells turning damaged and no longer needed proteins into building blocks to make new proteins[1]. Rapid growth and elevated rates of protein synthesis in cancer cells increase the production of abnormal proteins increasing the load of the proteasome. Proteasomes are also involved in the specific destruction of regulatory proteins such as transcription factors. These proteins are essential for cancer cell proliferation and survival. Taken together, these factors create a therapeutic window for proteasome inhibitors in various cancers. Inhibiting the proteasomes of the cancer cells by blocking the active site(s) of the proteasome causes selective apoptosis of malignant cells. FDA-approved proteasome inhibitors, bortezomib, carfilzomib, and ixazomib, are used for the treatment of multiple myeloma, a bone marrow cancer. However, the usage of proteasome inhibitors in solid tumors is limited by their poor tumor penetration and toxicity to normal tissues. By packaging FDA-approved proteasome inhibitors into liposome nanoparticles, selective delivery may protect drugs from metabolic inactivation, improve efficacy, tumor penetration, target engagement, and reduce toxicity to normal tissues[2]. To test this, tumor growth and the target engagement by the drug were evaluated in a murine model of triple-negative breast cancer. These cancers lack targeted therapies and are highly dependent on proteasome activity [3]. However, clinically achievable concentrations of FDA-approved proteasome inhibitors are not high enough to inhibit the growth of these tumors [4].

We created two types of nanoparticles, <sup>1</sup>conventional nanoparticles with a non-modified lipid envelope and pegylated-long-circulating nanoparticles, which carried polyethylene glycol (PEG) on their surface. PEG is a biocompatible and biodegradable polymer used to extend the half-life of nanoparticles in vivo because it prevents liposomes from being recognized and cleared by the innate immune system. We have compared the efficacy of conventional liposomes and PEG-liposomes of Cfz in a murine model of triple negative breast cancer. Tumors were created by injecting 105 4T1 cells into a mammary fat pad. This murine breast cancer cell line closely resembles human triple negative breast cancer in its molecular properties. When tumors became palpable, mice were treated with free, liposome, or PEG-liposome encapsulated Cfz twice weekly on consecutive days to match the clinical schedule of Cfz in myeloma patients. Tumor volume was measured with a caliper over the course of experiment. Mice were sacrificed when they lost more than 15% body weight as per approved IACUC protocol.



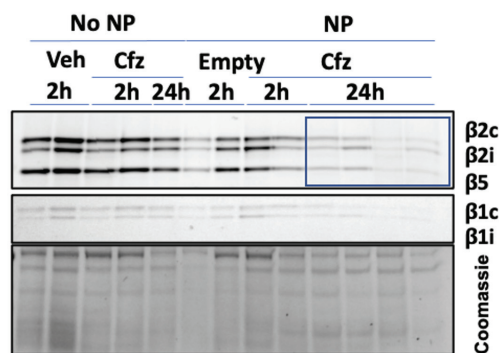
**Fig. 1** Liposomal formulation dramatically improves efficacy of Cfz in a murine model of triple negative breast cancer. Mice bearing 4T1 tumors in a mammary fat pad were treated with different formulation of Cfz (3 mg/kg) or vehicle on days indicated by arrow. MFP, mam-

A remote loading technique [5] was used to encapsulate Carfilzomib (Cfz) into pre-formed liposome nanopar-

<sup>1</sup>Corresponding author: led0043@auburn.edu

mary fat pad.

As can be seen on Fig. 1, traditional formulation of Cfz did not block growth of these tumors although Cfz was used at doses close to the maximal tolerated dose in mice [6]. On the other hand, tumor growth in mice treated with the same dose of Cfz in the liposomal formulations was dramatically delayed. Thus, liposomal formulation dramatically improved the efficacy of Cfz in this model of triple negative breast cancer. We also showed that the liposome formulations had less weight loss, compared to free drug. This suggests that these formulations were less toxic to healthy tissue compared to free Cfz. In a separate experiment (Fig. 2), we have measured intratumoral inhibition of proteasome by Carfilzomib. Tumor tissue was harvested 2 and 24 hours after a single dose of Cfz and lysed. The samples were incubated with activity based fluorescent probes that affinity label the  $\beta 1$ ,  $\beta 2$ ,  $\beta 5$  active sites of Cfz targets of the 20S proteasome [7]. Cfz is an irreversible inhibitor, and its binding to proteasome active sites prevents their labeling with the probes. After labeling, extracts were fractionated in polyacrylamide gel in the presence of lithium dodecyl sulfate to separate subunits (LDS-PAGE), followed by visualization of subunits by fluorescent imaging (Fig. 2). Darker bands indicate activity while the absence of the bands indicates proteasome inhibition. The  $\beta 5$  site is the prime target of Cfz, but our laboratory has previously determined that, in order for Cfz to kill triple-negative breast cells, it must co-inhibit either  $\beta 1$  or  $\beta 2$  sites [4]. As can be seen on Fig. 2, in the non-liposomal formulation of Cfz and empty nanoparticles caused little to no inhibition of the proteasome. Contrary to it, strong to near-complete inhibition of all active sites was observed in tumors that were exposed to inhibitors for 24 hours. Thus, the liposomal formulations of carfilzomib dramatically improved intertumoral target engagement.



**Fig. 2** Liposomal formulation of Cfz improve intratumoral inhibition of proteasomes. LDS-PAGE of tumor extracts after affinity labeling of proteasome subunits, which were detected by fluorescent imaging. Subsequent Coomassie staining was used as a loading control. Tumors were harvested at times indicated after treatment with a single 3mg/kg does of Cfz.

## Statement of Research Advisor

Laura Downey played a very important role in this project conducting all in vitro analysis of animal tissues. We will use her results in the NIH grant application, and eventually in a publication. This project was carried out under very difficult circumstances after Dr. Andrey Maksimenko, who created nanoparticles and conducted animal experiments, died unexpectedly. We were unable to hire a replacement essentially depriving us of opportunity to conduct additional experiments that would have generated additional samples for Laura to analyze. Laura presented these findings at the Student Research Symposium, where she won the Second prize in COSAM poster competition, and at the recent Inaugural Harrison College of Pharmacy Research showcase, where she got an award for the second-best poster in Cancer Research.

- Alexei Kisselev, Harrison College of Pharmacy

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## Authors Biography



Laura E. Downey is a junior-year student pursuing a B.S. degree in Biomedical Sciences with a Pre-medical concentration at Auburn University. She is also currently double minoring in Public Health and Dance. She played key research roles in quantifying tissue samples and testing multiple cell lines with various proteasome inhibitors. She received second place for University-Wide Undergraduate Student in Science, Technology, Engineering and Mathematics in the Auburn Research Symposium as well as Outstanding Presentation Cancer Focus at the HCOP Research showcase this April.



Andrey V. Maksimenko received his M.S. and Ph.D. in Chemistry from M.V. Lomonosov Moscow State University in Russia. After graduation, he moved to France, where he has held a variety of positions in industry and academia, and where he developed a strong interest in drug delivery. Dr. Maksimenko has been a visiting scientist at the Harrison College of Pharmacy from January 2018 till his untimely passing in March 2021.



Robert D. Arnold, Ph.D., is a Professor in the Department of Drug Discovery and Development. He completed a B.S. in Biochemistry and a Ph.D. in Pharmaceutical Sciences. After completing postdoctoral training at the University at Buffalo and Roswell Park Comprehensive Cancer Center, he was at the University of Georgia. He joined Auburn University and the Harrison College of Pharmacy in 2012. His research focuses on development of composite lipid-based nanoparticles and use of alternate dosing schedules to improve the treatment of primary cancer and metastatic disease.



Alexei F. Kisselev has been an Associate Professor of Drug Discovery and Development at the Harrison College of Pharmacy since 2017. He received his M.S. and Ph.D. in Chemistry from M.V. Lomonosov Moscow State University in Russia. Dr. Kisselev has conducted post-doctoral research



# Threat Reactivity as a Potential Risk Factor for Posttraumatic Stress Symptoms and Substance Use

*Dorothy E. Dreelin<sup>1</sup> and Brian J. Albanese<sup>2</sup>*

<sup>1</sup> Undergraduate Student, Department of Psychological Sciences, Auburn University

<sup>2</sup> Assistant Professor, College of Liberal Arts, Auburn University

Exposure to a potentially traumatic event (PTE) is incredibly common among U.S. adults, with approximately 90% reporting traumatic event exposure in their lifetime, and many reporting exposure to multiple PTEs (Kilpatrick et al., 2013). Trauma exposure can lead to an array of negative physical and mental health symptoms associated with stress-related conditions such as post-traumatic stress disorder. Our “Traumatic Stress Risk Factors and Daily Symptoms” study aims to examine underlying risk factors for posttraumatic stress symptoms (PTSS), specifically threat reactivity and negative reinforcement sensitivity. These risk factors will be assessed in relation to PTSS severity and maintenance over time among a trauma-exposed sample. Normal functioning of our body’s threat response system is imperative for ongoing survival, and PTSS has been shown to alter the way our body reacts to external threatening stimuli (Lobo et al., 2011). A blunted neural response to threatening stimuli has been shown in chronic and severe PTSS, while a hypersensitive neural response has been shown in people with moderate PTSS (D’Andrea et al., 2013). Further research on these risk factors will provide a better framework to understand the conditions that result in altered threat responding in PTSS and how this connects to other forms of psychopathology such as substance use disorders (McCauley et al., 2012) and suicidality (Albanese et al., 2021).

Eligibility requirements for participants (n=100, aged 18-65) include a history of trauma exposure and some PTSS, including at least avoidance or traumatic intrusions. We will first administer a clinical interview to screen participants for eligibility and symptom severity. Following screening, we will schedule participants for an in-person laboratory appointment in which in

which they will complete a non-invasive electroencephalogram (EEG) procedure while engaging in a series of computerized tasks designed to examine threat reactivity and sensitivity to negative reinforcement. To measure threat reactivity, participants will complete a picture-viewing task where they are presented with pleasant, neutral, and threatening images. Either a circle or a square will then flash onto the screen, and the participant must quickly and accurately press a button indicating which shape appeared. This task reflects threat reactivity and the ability to maintain attention on a goal-directed task in the context of a threat compared to the pleasant and neutral conditions. I hypothesize that participants who struggle to maintain attention during this task will experience more severe PTSS and report greater substance use. If participants have trouble maintaining attention, then they will have significantly greater brain activity, approximately 400-1000 milliseconds after viewing the stimulus. Participants will complete self-report measures of drug and alcohol use as well as alcohol use motives to assess engagement in substance use and potentially problematic substance use motives (e.g., drinking to cope and conform).

We will then invite participants to complete a two-week ecological momentary assessment (EMA) period where they will complete a brief survey four times a day for two weeks. The EMA period provides an opportunity to evaluate daily fluctuations in anxiety, mood, sleep patterns, and PTSS in the participant’s typical environment in real time (Shiffman, Stone, & Hufford, 2008). It also provides a chance to test how neurobehavioral mechanisms assessed in the lab predict changes in PTSS and substance

<sup>1</sup> Corresponding Author: ded0020@auburn.edu



use patterns in a real-world setting. The EMA period can provide rationale as to how and when PTSS and substance use co-occur (Lane, Waters, & Black, 2019). We will invite all participants to complete a follow up appointment three months later, which will include another clinical interview to observe the maintenance of their PTSS.

The present work may contribute to the development of targeted interventions to normalize threat responses among those with PTSS. It will also allow us to gain more knowledge about the comorbidity of PTSS and substance abuse. We look forward to collecting data on this study over the next few years and will be actively recruiting participants in the near future.

### Statement of Research Advisor

The Research and Interventions for Stress-related Conditions (RISC) lab in the Department of Psychological Sciences aim to better understand neurobehavioral mechanisms underlying vulnerability for posttraumatic stress disorder (PTSD), suicide, and other anxiety and stress-related disorders. Dorothy's research expands these areas in important ways by investigating one possible vulnerability factor for PTSD symptomology. Dorothy's impressive contributions highlight the outstanding undergraduate scholarship conducted in the RISC lab.

- Brian J. Albanese, College of Liberal Arts

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### Authors Biography



Dorothy E. Dreelin is a senior pursuing a B.A. degree in Psychology at Auburn University. She has been heavily involved in research at Auburn University since she began managing the Research and Interventions for Stress-related Conditions (RISC) lab in May of 2021. This upcoming fall, she

will be applying to Clinical Psychology Ph.D. programs with the intention of starting her Ph.D. in August of 2023. For her Ph.D. and future research, she intends to study risk and protective factors that overlap with both substance abuse and posttraumatic stress disorder.



Brian J. Albanese is an Assistant Professor in the Department of Psychological Science and Director of the Research and Interventions for Stress-related Conditions (RISC) lab at Auburn University. He received his B.S. in Psychology from the University of Connecticut and Ph.D. in Clinical Psychology from Florida State University in 2020 after completing a clinical internship at the Charleston Consortium.

# Alcohol Related Alterations in TNF - $\alpha$ In Binge and Moderate Drinkers

Josh Enger <sup>1</sup>, Benjamin Campbell <sup>2</sup>, Darren Beck <sup>3</sup>, Clayton Ridner <sup>2</sup>, Lily Crone <sup>1</sup>, Austin McClanahan <sup>3</sup>, Juliet Wilson <sup>3</sup>, Summer West <sup>3</sup>, and Sara K Blaine <sup>2</sup>

<sup>1</sup> College of Science and Mathematics

<sup>2</sup> College of Liberal Arts

<sup>3</sup> Edward Via College of Osteopathic Medicine

Alcohol is one of the most popular recreational drugs with roughly 70% of Americans reporting alcohol use within the last year (NIH, 2020). It is known that alcohol indirectly influences the immune system via its effects on the hypothalamic-pituitary-adrenal (HPA) axis, resulting in a dampening of the immune response (Eskandi & Sterberg 2002). Additionally, alcohol has direct effects on inflammatory and immune processes (Molina, et. Al., 2010), i.e., alcohol directly decreases white blood cell function (Gacouin, et. al, 2012) and quantity (Szabo & Mandrekar, 1999).

For example, alcohol also decreases expression of TNF- $\alpha$  through activation of its molecular inhibitor. TNF- $\alpha$  is a pro-inflammatory cytokine prone to decreased expression following the consumption of alcohol due to the interaction of alcohol with the NF-B inhibitor. TNF- $\alpha$  is a pro-inflammatory cytokine produced by macrophages/monocytes during infection which participates in a wide array of signaling pathways within cells. TNF- $\alpha$  is also an important protein in resistance to cancer and infection (Idriss & Naismith, 2000). Alcohol interacts with the NF-B inhibitor, decreasing phosphorylation and allowing it to bind to NF-B- thereby inhibiting the production and expression of TNF- $\alpha$ . This binding inhibits proper production of TNF- $\alpha$  in chronic drinkers. Importantly, these immunosuppressant qualities of alcohol consumption have been linked to greater susceptibility to respiratory infections (Happel and Nelson, 2005, Szabo and Mandrekar 2009).

Because of the tendency of a binge drinker to consume more alcohol on a more regular basis when compared to a moderate drinker, we hypothesized that the expression of TNF $\alpha$  following consumption of alcohol will be significantly less than moderate drinkers after alcohol

cue exposure and alcohol consumption. Understanding the potential negative ramifications of alcohol consumption on the immune system is especially relevant as the COVID19 pandemic continues on college campuses.

**Table 1.** Demographics. This table breaks down the demographics of selected individuals. (Total N=64 and the \* symbol notates a significant difference between BH/MD groups at p=0.05.)

SUBJECT VARIABLE	MODERATE SOCIAL DRINKERS (N=31)	BINGE/HEAVY SOCIAL DRINKERS (N=33)
<b>GENDER</b>		
FEMALE	14 (45%)	16 (48%)
MALE	17 (55%)	17 (52%)
<b>RACE</b>		
AFRICAN AMERICAN*	5 (16%)	1 (3%)
CAUCASIAN	23 (74%)	32 (97%)
ASIAN AMERICAN*	3 (10%)	0 (0%)
HISPANIC	6 (19%)	6 (18%)
<b>AGE*</b>	28 (7)	23.73 (4.5)
<b>YEARS OF EDUCATION*</b>	16.84 (2.5)	15.76 (1.6)
<b>DRINKING VARIABLES</b>		
NUMBER OF AUD FIRST DEGREE RELATIVES	0.26 (0.58)	0.24 (0.58)
YEARS OF REGULAR DRINKING	7.53 (6.89)	4.97 (5.16)
DRINKING DAYS IN PAST MONTH*	9.89 (7.35)	14.61 (6.37)
TOTAL AMOUNT CONSUMED IN PAST MONTH*	23.46 (19.18)	78.27 (63.70)
CAHALAN QFVI USUAL NUMBER OF DRINKS*	2.45 (0.93)	5.11 (2.84)
CAHALAN QFVI MAX NUMBER OF DRINKERS*	4.97 (1.83)	9.21 (2.75)

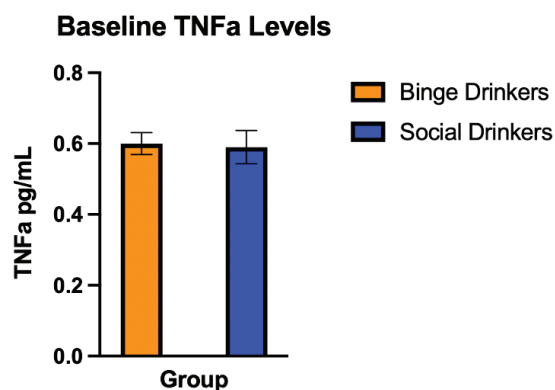
Participants were recruited based on their responses to a survey screening for drug use and abuse, alcohol use and abuse, history of mental health disorders, as well as age, race, and weight. Using parameters set by the NIH, the participants were classified into either the binge or moderate group. The classification was not mentioned to participants, and both groups experienced equal treatment. The group classified as “binge” was composed of participants who consumed 15+ alcoholic beverages a week for men, and 12+ alcoholic beverages a week for women. In these use patterns, we also looked to identify “binge” occurrences where the participant consumed 5-7+ drinks in one instance roughly once a month. The group classified as “moderate” consumed less than 15 drinks a week for men, and less than 12 drinks a week for women.

<sup>1</sup>Corresponding author: sara.blaine@auburn.edu

Participants came into the AUBIE lab on 2 non-consecutive days and administered a urine drug screen and a breathalyzer breath alcohol content test. Next, baseline blood samples were collected by a registered nurse prior to the alcohol taste test and placed in an ice bath for preservation prior to centrifugation. Next, during The Alcohol Taste Test, participants are presented with three mugs of beer. Participants are instructed to drink enough to determine if they are same beer or different beers. Following the ATT, the participant was monitored for 45 minutes and a final blood sample was collected.

The blood samples were then spun in the centrifuge at 3000 RPMs for 12 minutes at 4°C to separate the blood components. The plasma from the samples was then pipetted into participant specific 5mL test tubes and brought to Dr. Beck's lab at VCOM Auburn to be analyzed via an ELISA assay to determine and quantify the presence of TNF- $\alpha$  within participant samples. ELISA assays are an efficient method in quantifying blood proteins. A blood sample is tagged using antibodies that select for a specific blood protein, TNF- $\alpha$ , and are run through software that can detect the quantity of TNF- $\alpha$  by identifying the number of fluorescent antibodies.

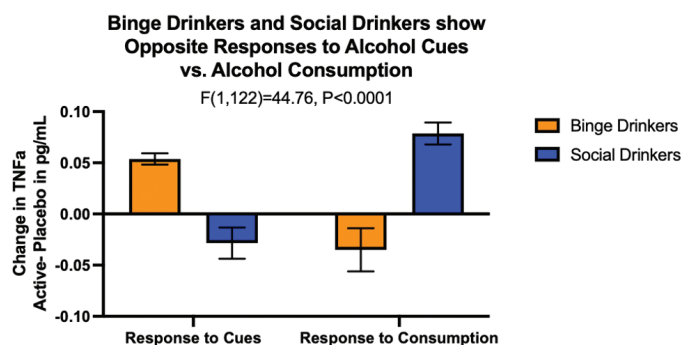
There were no significant difference of baseline levels of TNF- $\alpha$  between the two drinking groups, as shown in Figure 1. This demonstrates that there two groups were young and healthy, i.e., without a health condition affecting baseline immune activity.



**Fig 1.** No difference in baseline levels of TNF- $\alpha$  were found between groups of drinkers, ( $t(62)=0.1797$ ,  $p=0.85$ ).

A statistically significant interaction effect on TNF- $\alpha$

levels was seen among group membership, alcohol cues, and alcohol consumption (Figure 2). As the moderate drinking group was exposed to cues, their levels of TNF- $\alpha$  decreased. After the consumption of alcohol, TNF- $\alpha$  levels in MD drinkers increased, as expected in healthy individuals. The binge drinking group experienced an opposite reaction with TNF- $\alpha$  release during exposure to cues and a lack of release following consumption. With decreased levels of TNF- $\alpha$ , the ability of fight off inflammation is compromised. These findings are represented graphically in Figure 2.



**Fig 2.** Comparison of levels of TNF- $\alpha$  between groups.

Additionally, the response elicited by the binge drinking group is indicative of a priming effect, whereby initial exposure to reminders of drinking led to more drinking. The moderate drinking group does not elicit a priming response and experiences a release of TNF- $\alpha$  following the alcohol stimulus. The moderate group releases TNF- $\alpha$  following the consumption of alcohol. This response, by the moderate drinking group, is a healthy, unadjusted response to alcohol. The binge group, however, experiences the priming effect by releasing TNF- $\alpha$  initially when exposed to the alcohol cues, but before the consumption of alcohol.

Through the analysis of blood TNF- $\alpha$  via ELISA assays, it was found that TNF- $\alpha$  levels following exposure to alcohol cues differed significantly between groups. The analysis indicated that TNF- $\alpha$  levels in binge drinkers increased when exposed to alcohol visual cues but decreased following consumption. On the other hand, it was found that TNF- $\alpha$  levels in moderate drinkers decreased when exposed to alcohol cues but increased following consumption.



## Statement of Research Advisor

As part of a larger project funded by NIH, TNF- $\alpha$  response to alcohol cues and consumption were measured in binge drinkers and social drinkers. During the COVID19 pandemic, Josh noted the importance of a healthy immune response in fighting the virus. He therefore proposed to examine the differences in TNF- $\alpha$  response between the groups and found preliminary evidence that binge drinkers may have a compromised immune response to alcohol.

-Sara K. Blaine, PhD, College of Arts and Sciences

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## Authors Biography



Joshua Enger is a senior year of Biomedical Science at Auburn University. He recognized the importance of immune function during the COVID19 pandemic and developed this project for his Undergraduate Research Fellowship. He plans on continuing his research in medical school.



Benjamin Campbell is a post-graduate research assistant. He has compiled statistics for all facets of the research study and contributed his technical knowledge of computer programming and lab management.



Dr. Darren Beck is a member of the faculty of VCOM studying Cell biology and physiology. ELISA assays were run in his lab and under his supervision.



Summer West is a rising third year medical student at Edward Via College of Osteopathic Medicine. She performed ELISA assays.



Clayton Ridner, is a graduate student in the Cognitive and Behavioral Neurosciences Program at Auburn in Fall 2020. He performed psychological interviews of participants and supervised experimental procedures.



Austin McClanahan is a rising third year medical student at Edward Via College of Osteopathic Medicine. He performed ELISA assays.



Lily Crone, is a junior-year student pursuing a bachelor's of science in Neuroscience from Auburn University. Lily worked as a research assistant, handling participant blood samples and compiling data.



Dr. Sara Blaine is an Assistant Professor in the Department of Psychological Sciences as an Assistant Professor in 2019. TNF- $\alpha$  levels in response to alcohol cues and consumption were already being measured via her NIH funded Study, R00 AA25401.



Juliet Wilson is a rising third year medical student at Edward Via College of Osteopathic Medicine. She performed ELISA assays.

# Methods for Investigating the Role of Siderophores in Clinical and Agricultural Isolates of *Fusarium*

Harrison P. Estes<sup>1</sup> and Jeffrey J. Coleman<sup>2</sup>

<sup>1</sup> Undergraduate Student, Department of Entomology and Plant Pathology, Auburn University

<sup>2</sup> Associate Professor, Department of Entomology and Plant Pathology, Auburn University

The fungal genus *Fusarium* is comprised of many ubiquitous species that present challenges as both clinical and environmental pathogens (Coleman, 2015). Despite the availability of many modern techniques for genetic modification, it is challenging and sometimes impossible to alter this genus. Specifically, deletion of multiple genes requires an extensive, tedious process with often unsatisfactory results.

The goal of this project was the design and generation of a recyclable dominant selectable marker cassette that, upon insertion into a gene causing a mutation, would result in the disruption of transcription and termination of any product being created.

Like many other fungal genera, *Fusarium* produces secondary metabolites known as siderophores to chelate iron from its surroundings (Greenshields et al., 2007). Upon creation of the recyclable dominant selectable marker construct, the plasmid would be tested in one of the three primary siderophore-producing genes in a member of the *Fusarium solani* species complex (FSSC), which can infect both agricultural and clinical hosts. Following insertion into a single gene generating the desired mutation, this construct could then be modified to lose the dominant selectable marker but maintain a small insertion of DNA keeping the siderophore encoding gene nonfunctional.

A similar strategy would be used to disrupt and mutagenize the other siderophore-producing genes. Once produced, the triple mutant of all the siderophore producing genes would then be cultured to examine its ability to survive compared to the wild type. We hypothesize that upon the production of this mutant, it

will show that iron plays a more significant role in the<sup>1</sup> FSSC than previously thought. These mutants will fail to survive and will not cause as severe symptoms as the wild type in their hosts.

So far, the construct is still being designed due to challenges experienced in its assembly. Multiple molecular techniques and strategies have been taken to approach the difficulties, but further work is required to ensure the production of the proper vector. Once the plasmid vector can be utilized, though explicitly designed for the inhibition of the production of siderophores, it is not limited in its application solely for this purpose. It will be able to disrupt any gene or group of genes, allowing for a more efficient investigation of the many unknown biochemical pathways in *Fusarium*.

The importance of this research project lies in the commonality of *Fusarium*, the lack of a method to modify the fungus efficiently, and the few options available to treat its infections. This project will provide information about the siderophores of *Fusarium*, which can be utilized to treat and prevent diseases in clinical and agricultural environments. Additionally, the created vector will be utilized to investigate other metabolic pathways and the development of other novel treatments.

## Statement of Research Advisor

Iron serves as an important cofactor for many enzymes and is a limiting nutrient during fungal virulence within a host. Many fungi have evolved several mechanisms to overcome this limitation, including synthesizing many iron-binding compounds such as siderophores. As generation of multiple mutations within a single fungal strain is difficult, Harrison's project aimed to develop a

<sup>1</sup>Corresponding author: hpe0004@auburn.edu

method to overcome this challenge. His research has demonstrated that construction of this novel vector is not straight forward and will require a time consuming and tedious process of multiple cloning steps.

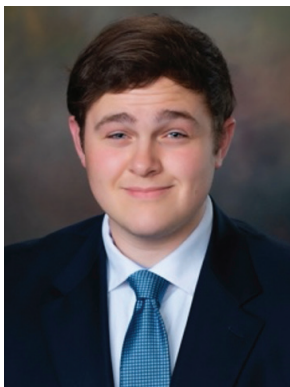
- Jeff Coleman, *Entomology and Plant Pathology*

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## Authors Biography



Harrison P. Estes is a junior-year student and Undergraduate Research Fellow pursuing a B.S. degree in Applied Biotechnology at Auburn University. He has played key research roles in determining methodology for the assembly of the vector. From Pike Road, Alabama, Harrison has spent the previous two years in Coleman Laboratories contributing to three projects in molecular mycology, fungal ionomics, and virulence studies of clinical and agricultural isolates of *Fusarium*. Once in graduate school, he plans to pursue a Ph.D. and continue his research career focusing on agricultural biotechnology, medical mycology, and fungal genomics.



Jeffrey J. Coleman is an Associate Professor in the Department of Entomology and Plant Pathology at Auburn University. After obtaining his Ph.D. in Plant Pathology at the University of Arizona, Dr. Coleman conducted postdoctoral research in medical mycology at Harvard Medical School. Since joining the faculty at Auburn University in 2014, he has developed a laboratory focused on investigating fungal pathogenesis using advanced molecular techniques.



# Testing Interoceptive Dysfunction as a Mediator between Sleep Disturbances and Suicidal Ideation among Military Service Members

*Walton G. Ferguson<sup>1</sup>, William Grunewald<sup>2</sup>, and April R. Smith<sup>3</sup>*

<sup>1</sup> Undergraduate Student, Department of Psychological Sciences, Auburn University

<sup>2</sup> Graduate Student, Department of Psychological Sciences, Auburn University

<sup>3</sup> Associate Professor, Department of Psychological Sciences, Auburn University

Suicide among military service members (MSM) is a major public health problem. Veterans are at increased risk for suicide compared to non-veterans (Kaplan et al., 2012; McCarthy et al., 2009). Additionally, the suicide rate for active-duty service members has become comparable to that of the general population following sharp increases in recent years (Pruitt et al., 2019). Public health entities such as the Veterans Health Administration have taken steps to reduce the burden of suicide among MSM, but the need to identify factors that confer risks for suicidality among MSM is still significant. Thus, the current study attempted to identify two potential risk factors/mechanisms for suicidality among MSM: insomnia severity and interoceptive dysfunction. Interoception is defined as the central nervous system's ability to monitor and notice signals coming from the body. Likewise, dysfunctional interoception can occur whenever external factors hinder one's ability to accurately assess such signals. Interoceptive dysfunction influences various psychopathologies, and insomnia is a known risk factor for suicidality among MSM (Forrest et al., 2015; Troxel et al., 2015). These constructs may work conjointly to predict suicidality among MSM. Specifically, interoceptive dysfunction may underlie the relationship between insomnia severity and suicidal ideation. Individuals with sleep difficulties may have difficulty perceiving their internal sensations as a result of poor sleep quality. This interoceptive dysfunction may culminate in suicidal thoughts, given the well-established link between decreased interoception and suicidality. Therefore, we investigated longitudinal relationships between interoceptive dysfunction, insomnia severity, and suicidality among a sample of MSM. We hypothesized interoceptive dysfunction to act as a mediator between insomnia severity and suicidal ideation.

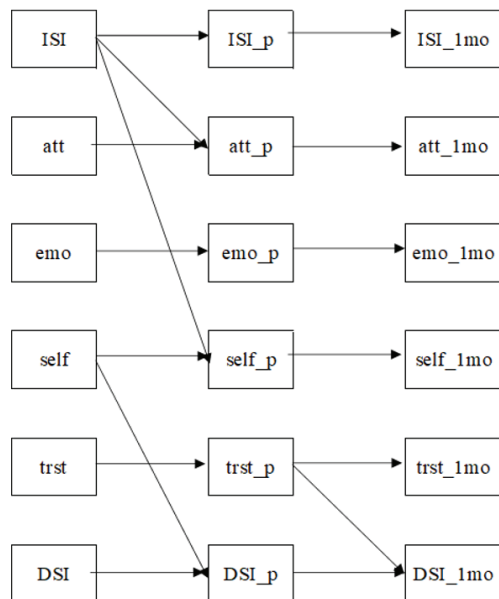
For this study, archival data were collected from 195<sup>1</sup> MSM enrolled in a randomized control trial meant to reduce suicidality among MSM. Participants completed self-report surveys at three timepoints separated by one month each that measured suicidal ideation, insomnia severity and interoceptive awareness (4 subscales included). Analyses focused on a 3-timepoint longitudinal autoregressive cross lagged mediation model using MPlus. Longitudinal paths from insomnia severity to interoceptive dysfunction, interoceptive dysfunction to suicidal ideation, and insomnia severity to suicidal ideation were estimated, each separated by one timepoint. In addition, normal tests of mediation of insomnia severity to suicidal ideation by interoceptive dysfunction were conducted.

Results of this model are presented in Figure 2. Insomnia severity significantly predicted interoceptive dysfunction over time, while interoceptive dysfunction predicted increased suicidal ideation over time. Interoceptive dysfunction did not mediate relationships between insomnia severity and suicidal ideation. Significant relationships between insomnia severity and interoceptive dysfunction suggest that insomnia severity may be a risk factor for interoceptive dysfunction in that poor sleep quality may lead to reduced ability to recognize bodily sensations. Furthermore, our results replicate previous research (Forrest et al., 2015) by highlighting that disconnection from one's bodily sensations increases suicide risk.

Given our results, we recommend assessing for both interoceptive dysfunction and insomnia severity as potential risk factors for suicidal ideation among MSM. Furthermore, results suggest that interoceptive dys

<sup>1</sup>Corresponding author: wgf0007@auburn.edu

function may be a promising intervention target to reduce suicidality among MSM. Clinicians may consider improving interoceptive abilities by utilizing mindfulness, exposure, and cognitive-behavioral techniques.



**Fig. 1.** Significant effects observed from the model. 'p' = post, '1mo' = 1 month timepoint. 'ISI' = Insomnia Severity Index Scores, 'att' = MAIA "attention" subscale, 'emo' = MAIA "emotional regulation" subscale, 'self' = MAIA "self-regulation" subscale, 'trst' = MAIA "trusting" subscale, 'DSI' = Depressive Symptom Index suicidality subscale scores.

### Statement of Research Advisor

Walton Ferguson led the development of this project testing the relations between sleep disturbances, interoception, and suicidality among military service members (MSM). Under the supervision of graduate student, Will Grunewald, Walton learned how to conduct and interpret the longitudinal path analyses for this study. These results can inform clinical assessments and interventions for MSM.

- April R. Smith, College of Liberal Arts

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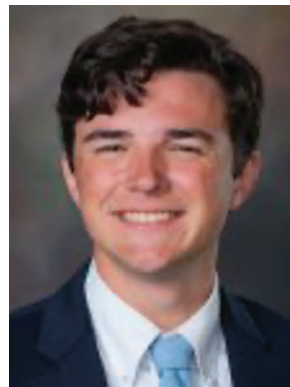
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### Authors Biography



Walton Ferguson is studying Psychology and Statistics at Auburn University. He intends to pursue graduate studies in Psychology. His research interests include learning, neuroplasticity, and interoception and currently has one publication with the REDS lab.



Will Grunewald is a third-year clinical Psychology PhD student at Auburn University. His research interests include identifying risk factors for disordered eating, body image disturbance, and suicidality, with a particular emphasis on these risk factors among vulnerable populations. He also studies the overlap between conformity to masculine norms and Muscle Dysmorphia.



Dr. Smith is currently an Assistant Professor of Psychology at Auburn University and the director of the Research on Eating Disorders and Suicidality (REDS) Laboratory. She received her B.A. in Psychology and Plan II from the University of Texas at Austin in 2004. She received her Ph.D. from Florida State University's Clinical Psychology Program in 2012, and completed her clinical residency at the University of California, San Diego.



# Cognitive Distortions and Empathy in Adolescents Adjudicated for Sexual Offenses

Emma Fisher <sup>1</sup> and Kelli R. Thompson <sup>2</sup>

<sup>1</sup> Undergraduate Student, Department of Psychological Sciences, Auburn University

<sup>2</sup> Assistant Research Professor, Department of Psychological Sciences, Auburn University

This study examined the potential relationship between cognitive distortions and empathy in a sample of adolescents adjudicated for illegal sexual behavior (AISB). According to the U.S. Department of Justice, approximately 32% of arrests made for sexual offenses involve suspects identified as minors (Eastman, 2004). Improving efforts to understand the etiology of adolescent sexual offending benefits all other assessment, treatment, and policy efforts involving these youth and their families (Seto & Lalumiere, 2010). Two common treatment goals when working with this group are increasing levels of empathy and cognitive restructuring (Eastman, 2004). Cognitive distortions are ways by which people rationalize and excuse their illegal or otherwise socially inappropriate behavior through maladaptive thought patterns, beliefs, and ideas (McCrady et al., 2008). These distortions may block empathic responses by misinterpreting the sexual encounter without those moral feelings typically associated with abuse, such as remorse.

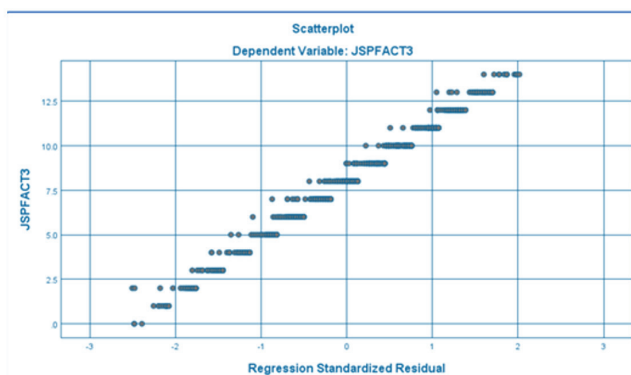
Data was collected from a pre-treatment interview of 732 male adolescents ages 12-17 in a court-mandated residential treatment facility. Cognitive distortions were measured using the Adolescent Cognition Scale, a 32-item forced choice (T/F) questionnaire describing a range of sexual attitudes, values, or behaviors (Hunter et al., 1991). A sample item may read, "A very young child can make a decision about having sex with me." An endorsement of "true" would indicate a cognitive error about socially appropriate sexual behavior. The total number of cognitive errors was entered as the predictive variable. Empathy was assessed using the Juvenile Sex Offender Assessment Protocol-II (JSOAP-II; Prentky & Righthand, 2003). The JSOAP-II is a 28-item clinician administered actuarial risk assessment designed for and widely used with AISB (Petersen, 2010). The JSOAP-II provides four scales of risk assessment

and an overall risk score. The intervention scale (Factor<sup>1</sup> 3), which looks at accepting responsibility, internal motivation for change, empathy, remorse, guilt, and cognitive distortions, was entered as the outcome variable in the current analyses.

Linear regression was performed to assess the relationship between cognitive distortions and a lack of empathy. The overall sample mean on the Adolescent Cognition Scale was 4.36 (SD=4.08), with only 2% scoring 15 or more cognitive errors. A statistically significant positive relationship was found between the two variables indicating that those with higher levels of cognitive distortions shared higher levels of intervention-related risk factors such as lack of empathy,  $F(1, 731) = 10.33, p = 0.001$ . See Figure 1 for scatterplot of main study variables.

These results support previous findings on cognitive distortions and a lack of empathy, which may impede intervention efforts when working with AISB (Pithers, 1999, Varker et al., 2008). The clinical implications of these results cannot be denied, given that empathy building is used in 94% of treatment programs for AISB (Varker et al., 2008). Furthermore, the endorsement of cognitive distortions can distinguish treated from untreated juvenile offenders in risk evaluations (Eastman, 2004). Thus, reducing cognitive errors and increasing victim empathy together enhances the ability to feel moral emotions which can trigger motivation for change and engagement within the treatment process.

<sup>1</sup>Corresponding author: ekf0014@auburn.edu



**Fig. 1** JSOAP Factor 3 vs. Adolescent Cognition Scale

### Statement of Research Advisor

Emma's work was supported by a more than 20-year public-public partnership between the AU Juvenile Delinquency Lab and the AL Department of Youth Services. She initially presented this data at the 68th Annual Meeting of the Southeastern Psychological Association and will continue interests in this theme as the lab manager in the upcoming academic year.

-Kelli R. Thompson, Psychology

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### Authors Biography



Emma Fisher was a senior year student who recently graduated with a B.A. in Psychology from Auburn University. She was a part of the Juvenile Delinquency Lab and has collected data to help with numerous research projects including her own. Her goal is to receive a PhD in Clinical Psychology. Her interests include personality psychology, recidivism risk, and mental health in forensic settings.



Kelli Thompson is an assistant research professor here at Auburn University. She is head of the Juvenile Delinquency Lab where she assist and guides students in the research process including creating presentations and publications.

# Archaeological Modelling of Ice Patches in the Northwest Territories Using Remote Sensing and GIS

Bethany G. Foust <sup>1</sup>, Mallory Jordan <sup>2</sup>, and Stephanie R. Rogers <sup>3</sup>

<sup>1</sup> Undergraduate Student, Department of Geosciences, Auburn University

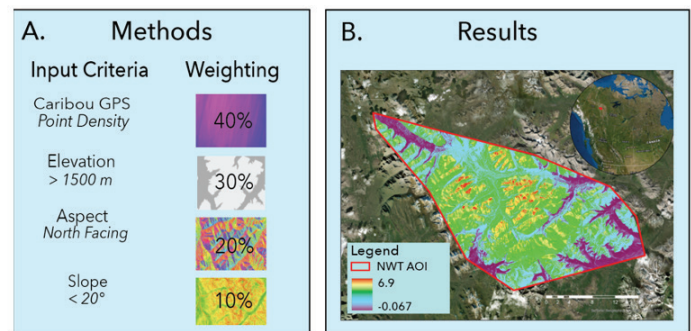
<sup>2</sup> Graduate Student, Department of Geosciences, Auburn University

<sup>3</sup> Assistant Professor, Department of Geosciences, Auburn University

Due to climate change, ice patches, glaciers, and snow are melting at unprecedented rates in high latitude and altitude environments (Ødegård et al., 2017). This phenomenon has exposed archaeological remains across the world, leading to a new field of study called “Ice Patch Archaeology” (Reckin, 2017). This study focuses on ice patch archaeology in the Mackenzie Mountains, located in the Northwest Territories (NWT), Canada (Fig. 1), where ice patches have historically been used by caribou (*Rangifer tarandus*) in the summer to escape from heat and insects (Andrews et al., 2012). Artifacts like bows, arrows, and darts have been uncovered near ice patch margins, which were left behind by native hunters and preserved for thousands of years. This study area is vast and difficult to access, therefore geospatial tools like Geographic Information Systems (GIS) and remote sensing are essential for archaeological prospection site selection. These were used to locate areas of high archaeological potential where artifacts might be freshly exposed from newly melted ice patches.

For this study, a multiscale, GIS-based multi-criteria decision analysis (MCDA) approach was used to find areas with archaeological potential and recently melted ice (Dixon et al., 2005., Rogers et al., 2014). Two models were created: Model 1 was created to first narrow down a very large study area (4,430 km<sup>2</sup>) with a coarse resolution Digital Elevation Model (DEM, 30 m) (ASTER Science Team, 2019), and Model 2 zoomed in on an area of interest to conduct higher resolution analysis using a 2 m DEM (Porter et al., 2018). For both models, the ideal criteria for locating artifacts were determined to be areas with high caribou presence and density, elevation above 1,500 m, N and NE facing slopes, and slope angle which could be easily traversed by humans (Fig. 1A).

Each criterion was classified according to its capacity to contain ice and weighted based on its relative importance to the overall model, with caribou density being most important and slope angle being least important (Fig. 1A).



**Figure 1.** A.) features the ideal criteria input into the MCDA on the left, and how much each layer was weighted in its potential to house archaeological artifacts in ice. B.) shows the resulting map with high potential areas in red, and an inset map of the location of the study area in the Mackenzie Mountains, Canada.

Results from Model 1 enabled us to narrow down the 4,430 km<sup>2</sup> study area to 825 km<sup>2</sup> of high archaeological potential and to conduct more in-depth analyses to further refine the study area and resulting in 60 km<sup>2</sup> of high archaeological potential. Next, Sentinel-2 satellite imagery (European Space Agency, 2015) was used to create a Normalized Difference Snow Index (NDSI) (Gascoin et al., 2020) using green (Band 3) and short-wave infrared (Band 11) bands to locate snow and ice (Equation 1.).

$$NDSI = \frac{(Band\ 3 - Band\ 11)}{(Band\ 3 + Band\ 11)} \quad (1)$$

<sup>1</sup> Corresponding author: s.rogers@auburn.edu



**Equation 1.** Normalized Difference Snow Index used for Sentinel-2 imagery.

Satellite imagery was obtained for September 2018 and September 2021 when there was perennial ice and snow coverage. The 2021 tile was subtracted from the 2018 tile to show areas of recent melt. Recent melt areas were compared to Model 2 results, and overlapping areas were designated as ideal locations (shown in red) for archaeological prospection in Summer 2022 (Fig.1B). Archaeologists will report their findings and we will use this information to further refine the model for the NWT.

### Statement of Research Advisor

Bethany has conducted all associated steps of this research project including the literature review, data collection, data analysis and processing, and interpretation. This research is very important for archaeologists in the NWT, as well as around the world, for recovering culturally significant artifacts that become threatened by decomposition when they are exposed to elements after melting out of environmentally sensitive ice patches.

– Stephanie Rogers, COSAM

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### Authors Biography



Bethany Foust graduated with a B.S. degree in Geospatial and Environmental Informatics at Auburn University. She was an Undergraduate Research Fellow during her senior year in the GeoIDEA Lab. In August, she will start her M.S. degree in Geography and NSF Research Traineeship at Auburn University. Her interests include climate change, water, geography, and cartography.





Mallory Jordan is a research assistant in the GeoIDEA Lab at Auburn University. She recently completed her M.S. in Geography and will commence her PhD in Earth System Science this August at Auburn University. Her research interests revolve around the use of geospatial techniques to address environmental issues, especially those related to water.



Dr. Stephanie R. Rogers is an Assistant Professor of Geographic Information Science in the Department of Geosciences at Auburn University. She studied ice patch and glacial archaeology as part of her PhD research in the Pennine Alps in Switzerland. Dr. Rogers continues to work with archaeologists to better understand how physical characteristics of the terrain can help unlock clues to the past.

# Adverse Effects of Mandated Juvenile Sex Offender Registration Across Treatment

Elena Gagliano<sup>1</sup> and Kelli R. Thompson<sup>2</sup>

<sup>1</sup> Undergraduate Student, Department of Psychological Sciences, Auburn University

<sup>2</sup> Assistant Research Professor, College of Liberal Arts, Auburn University

The purpose of this study was to investigate the adverse effects of mandated juvenile sex offender registration and notification laws. Incarcerated juveniles face many challenges; those adjudicated for sexual offenses face additional stressors with registry requirements upon treatment completion. In 2006, the Adam Walsh Act federally mandated sex offender registration and notification laws in the United States. These laws were expanded to include youth adjudicated for certain sexual offenses with some states mandating public notification.

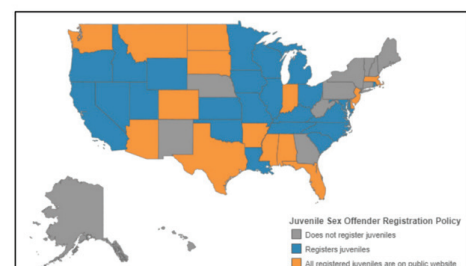
Public juvenile registries are associated with a wide range of unintended negative consequences for youth (Harris et al., 2016; Letourneau et al., 2018). Youth who are required to register report higher rates of harassment, adult solicitation, sexual assault, and suicide attempts than their counterparts who are not required to register (Letourneau et al., 2018). They are also subject to the stigmatized label of “sex offender” which could prove detrimental to healthy identity development (Harris & Socia, 2016). This raises concerns for adolescents navigating their social world after successfully completing treatment (Fix et al., 2020). These public juvenile registries remain in 14 states, despite clear evidence that registries do not reduce incidences of future sexual abuse but do have harmful consequences for youth and families (Chaffin, 2008). See Figure 1 for United States Juvenile Registry Laws by State.

In this study, we followed a group of 35 male adolescents currently in a court-mandated residential treatment program. Youth completed the Concerns About Juvenile Registration and Notification Questionnaire (C-JSORNQ; Fix et al., 2020), a self-report measure on fears and anxieties surrounding registration requirements at three time points across treatment: pre, mid, and post-treatment. It was hypothesized that post-treatment anxiety levels would spike as youth face the real-

ity of registration and notification before re-entering society. A series of independent t-tests were non-significant, indicating youth reported moderate levels of anxiety across treatment without a significant peak at any time point; pre-mid:  $t(27) = 0.06, p = .94$ ; mid-post:  $t(33) = 0.46, p = .65$ ; pre-post:  $t(32) = 0.53, p = .60$ . See Figure 2 for means across treatment.

These findings further strengthen the current scholarly opinion that there are numerous negative consequences associated with juvenile sex offender registration and notification laws. Additionally, these findings illuminate the need for a specific treatment focus on teaching youth to manage anxieties associated with the registry. This may be especially prevalent as they approach reentry to the community.

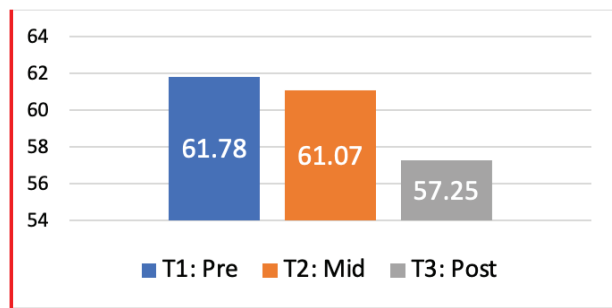
The persistence of anxiety in the face of treatment implies the need to further question wide-ranging repercussions and unintended consequences of juvenile registration policies. This is particularly necessary for youth in treatment who present a very low risk for future reoffending (Letourneau & Miner, 2005). Finally, this raises legal policy concerns for youth in states with public registries who face an additional stressor after successful completion of treatment that youth in other states do not. If it is clear juvenile registries do more harm than good, why should some states still be allowed to implement such policies? The need for federal legislation is quite evident.



Source: [www.theatlantic.com/politics/archive/2015/11/states-slowly-scale-back-juvenile-sex-offender-registry/433473/](http://www.theatlantic.com/politics/archive/2015/11/states-slowly-scale-back-juvenile-sex-offender-registry/433473/)

<sup>1</sup>Corresponding author: [ekg0022@auburn.edu](mailto:ekg0022@auburn.edu)

**Fig. 1.** United States Juvenile Registry Laws by State (2015)



**Fig. 2.** Mean Registry Fears Across Treatment

### Statement of Research Advisor

Elena's research was supported by a more than 20-year public-public partnership between the Alabama Department of Youth Services and Auburn University Department of Psychological Sciences. Her interest in youth registries highlights the important role science plays in civic engagement and public policy.

– Kelli R. Thompson, Liberal Arts

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### Authors Biography



Elena Gagliano is a junior-year student pursuing a B.A. degree in Psychology and a B.S. degree in Human Development and Family Science at Auburn University. She is a 2022-2023 Auburn Undergraduate Research Fellow. Elena has assisted in the identification of gaps in the current understanding of challenges faced by juveniles who have committed sexual offenses.



Kelli R. Thompson is an Assistant Research Professor in the College of Liberal Arts at Auburn University. She is currently the Director of Research for the Alabama Department of Youth Services' Accountability Based Sex Offense Prevention Program.

# A Cross Sectional Study of Temporal Diet Quality in US Adults

Corinne E. Gautreaux<sup>1</sup>, Kristen S. Smith<sup>2</sup>, and Andrew D. Frugé<sup>3</sup>

<sup>1</sup>Undergraduate student, College of Science and Mathematics, Auburn University, Auburn, AL, USA

<sup>2</sup>Ph.D. Student, College of Human Sciences, Auburn University, Auburn, AL, USA

<sup>3</sup>Assistant Professor, Department of Nutrition, Dietetics, and Hospitality Management, College of Human Sciences, Auburn University, Auburn, AL, USA

**Abstract:** In March 2020, the COVID-19 pandemic led to restricted vocational (Voc-PA) and recreational physical activity (Rec-PA) outside of the home. We conducted a nationwide survey in the United States (US) during the mitigation peak of the pandemic (June 2020) to assess health-related changes from the previous year. A diet quality (DQ) assessment tool weighted the relative healthfulness of eating occasions from foods prepared-at-home (Home) and away-from-home (Away). Previously validated instruments assessed PA and demographic variables; height and weight were self-reported to calculate body mass index (BMI). T-tests explored longitudinal, between-sex, and obesity status differences in DQ, PA, and BMI; Pearson correlations explored associations. Of 1648 respondents, 814 valid responses (56.8% female, 81.7% white) were analyzed. Overall and Home DQ was higher for females than males in 2020 ( $p < 0.001$  for both). Respondents increased DQ from 2019 to 2020, primarily from Away ( $p < 0.001$  for both sexes). Total Rec-PA and Voc-PA were higher in males ( $p = 0.002$ ,  $p < 0.001$ ) than in females in 2020; females reported higher other PA ( $p = 0.001$ ). Change in BMI was inversely associated with a change in both DQ and PA ( $p < 0.001$  for both). In this sample of US adults, early adaptations to the COVID-19 pandemic included improved DQ and BMI. Whether these short-term improvements were maintained warrant further investigation.

More details of the research can be found in the authors' published work <https://doi.org/10.3390/ijerph19148289>

**Keywords:** Diet, physical activity, pandemic, COVID-19, body mass index

## Statement of Research Advisor

Ms. Gautreaux began this project with an extensive literature review that allowed her to develop and test her hypotheses regarding health behaviors during a lockdown. Corinne spent countless hours conceptualizing and coding a complex data set in addition to her rigorous analysis. Her findings will be an excellent addition to the healthcare literature as they combine validated and novel methods that advance our understanding of compulsory lifestyle adaptations.

- Andrew Frugé, College of Human Sciences

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## Authors Biography



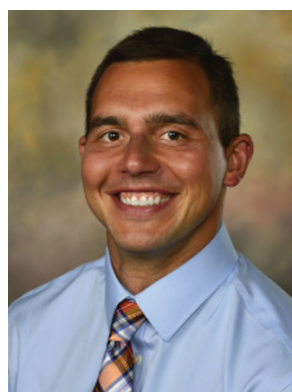
Corinne E. Gautreaux is a senior-year student pursuing a B.S. degree in Biomedical Sciences at Auburn University. She has been involved in the Dietary Chemoprevention, Gut Health, and Aging Lab for the past three years and was an Undergraduate Research Fellow for the College of Science and Mathematics during the Summer 2021 semester. She will be pursuing her professional education at medical school in the Fall of 2022.

<sup>1</sup>Corresponding author: [ceg0054@auburn.edu](mailto:ceg0054@auburn.edu)





Kristen S. Smith is a doctoral student in the Dietary Chemoprevention, Gut Health, and Aging Lab completing a Ph.D. in nutrition at Auburn University. She will continue her research in cancer prevention and survivorship at the Moffitt Cancer Center in Tampa, FL.



Andrew D. Frugé is an Assistant Professor in the Department of Nutrition, Dietetics, and Hospitality Management at Auburn University. He is the Principal Investigator of the Dietary Chemoprevention, Gut Health, and Aging Lab.

# Comparing Training Volumes Between Softball Pitchers with and Without Upper Extremity Pain

Peyton N. Gober <sup>1</sup>, Nicole M. Bordelon <sup>2</sup>, Katherine M. Everhart <sup>2</sup>, and Gretchen D. Oliver <sup>3</sup>

<sup>1</sup>Undergraduate Student, School of Kinesiology, Auburn University

<sup>2</sup>Undergraduate Student, School of Kinesiology, Auburn University

<sup>3</sup>Professor, School of Kinesiology, Auburn University

Windmill softball pitchers are 2.6 times more likely to sustain an upper extremity (UE) overuse injury than positional players [1]. Softball pitching research has focused on identifying the biomechanical parameters associated with upper extremity pain [1-3]. However, data regarding modifiable risk factors such as training load and volume are lacking. Thus, it is necessary to understand which variables related to training volumes are correlated with UE pain in softball pitchers. Youth baseball pitchers with upper extremity pain have higher training volumes and play more months per year than pitchers without pain. Baseball studies have also shown related modifiable risk factors such as pitches thrown in a single game and across a competitive season increased risk of upper extremity pain [4]. Similar research has not been done in softball despite comparable UE injury rates.

The purpose of this study was to investigate the differences between training volumes and months per year playing between softball pitchers with and without upper extremity pain.

Thirty-six youth and high school softball pitchers (14.0  $\pm$  2.0yrs, 58.8  $\pm$  28.5cm, 62.0  $\pm$  19.9kg) who were active on a team roster within the past six months completed an online survey via Qualtrics (Figure 1). Participants were asked Do you currently experience any pain/discomfort in the upper extremity? If they answered “yes” or “no”, they were placed in upper extremity pain (n=11; 15.2  $\pm$  1.9yrs; 164.9  $\pm$  6.7cm; 72.0  $\pm$  22.4kg) or no pain (n=25; 13.5  $\pm$  1.8yrs; 156.2  $\pm$  33.8cm; 57.6  $\pm$  17.4kg) groups. Participants were asked to indicate the number of in and off-season hours per week spent practicing softball, practicing other sports, and strength and conditioning training. Participants were also asked about months per year spent softball training. A

Mann-Whitney U Test was performed to examine the differences between training volumes between softball pitchers with and without UE pain.

**Fig. 1** Survey question asking about softball practice volumes.

There were no significant differences between training volumes and months per year of playing softball between pitchers with and without upper extremity pain (all p-values > .132). Medians and inter-quartile descriptive statistics are presented in Table 1.

**Table 1.** Data are presented as median (inter-quartile range)

Variable	Pain (n=11)	No Pain (n=25)	P
Hours Practicing Softball			
in season	14.0(10.0,20.0)	12.0(8.0,15.0)	.309
off season	8.0(5.0,12.0)	6.0(4.0,11.5)	.398
Hours Practicing Other Sports			
in season	2.0(0.0,7.0)	0.0(0.0,6.0)	.584
off season	3.0(0.0,8.0)	0.0(0.0,3.0)	.132
Hours Strength & Conditioning			
in season	5.0(2.0,8.0)	4.0(2.0,8.0)	.614
off season	7.0(2.0,14.0)	5.0(2.5,6.5)	.324
Months per Year Playing Softball	12.0(11.0,12.0)	12.0(10.5,12.0)	.289

**Figure 2.** Scanning Electron Microscopy Wood with Silver

<sup>1</sup>Corresponding author: png0010@auburn.edu

There were no significant differences between training volumes and months per year of playing softball between pitchers with and without upper extremity pain. UE pain was present in 11 pitchers indicating approximately 31% of pitchers self-reported currently experiencing UE pain. Other factors should be investigated to determine their correlation to UE pain in youth softball pitchers, such as the number of pitches thrown in a game, level of self-satisfaction or arm fatigue during a game, which have been proven to be related to pain in baseball [4]. Once factors are determined that do contribute to this UE pain, regulations such as pitch count that already exist in the realm of baseball can be implemented in softball to work towards injury prevention in this sport also.

The findings show other modifiable risk factors should be examined to identify the susceptibility of UE pain in youth and high school softball pitchers. Results should be interpreted with caution considering the relatively small sample size ( $n=36$ ). Future studies should compare other modifiable risk factors such as throwing volumes, years spent playing competitive softball, and degree of sport specialization between softball pitchers with and without UE pain.

### Statement of Research Advisor

Peyton took the lead in this study. She was an intricate part of survey development, participant recruitment and data analysis. Peyton looks to continue this line of research in her graduate studies.

- *Gretchen D. Oliver, School of Kinesiology, College of Education*

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### Authors Biography



Peyton N. Gober is recent graduate of Auburn University with a B.Ed. degree in Exercise Science. During her four years as an undergraduate student, she served as a research assistant and subsequently a research fellow in the Sports Medicine & Movement Laboratory, where she assisted in research focusing on softball and baseball injury prevention.



Nicole M. Bordelon is a Ph.D. student in Auburn University's Sports Medicine & Movement Laboratory. She received a bachelor's from California State University of San Bernardino and a master's from the University of Wyoming. Nicole's research focus is on softball injury prevention and performance enhancement.



Katherine Everhart is a Ph.D. student in Auburn University's Sport Medicine & Movement Laboratory. She received a bachelors from University of North Georgia and a masters from Georgia College and State university. Katherine's focus is on softball hitting performance and injury prevention.



Gretchen D. Oliver is full Professor and the Director of the Sports Medicine & Movement Laboratory within the School of Kinesiology. She has published over 160 peer-reviewed manuscripts and known nationally and internationally for her work regarding baseball and softball injury prevention and performance enhancement.



# Additive Manufacturing of Braided Structures for Composite Reinforcement

Katherine Griffin <sup>1</sup>, Ajay Jayswal <sup>2</sup>, and Sabit Adanur <sup>3</sup>

<sup>1</sup> Undergraduate Student, Department of Mechanical Engineering, Auburn University

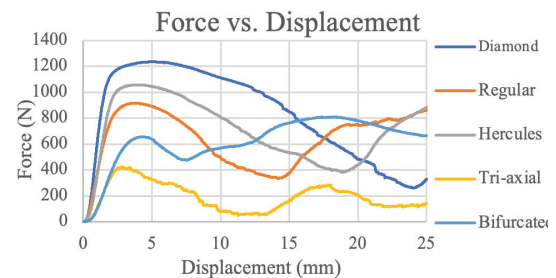
<sup>2</sup> Graduate Student, Department of Mechanical Engineering, Auburn University

<sup>3</sup> Professor, Department of Mechanical Engineering, Auburn University

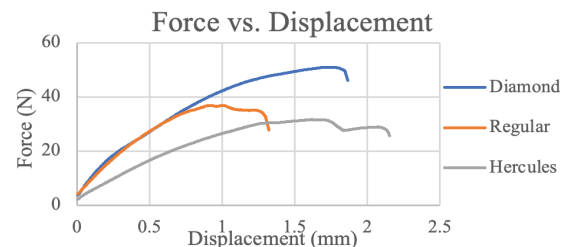
Braided patterns are often used to provide structural reinforcement in other materials. However, traditional methods of fabricating these braided structures are limited in their abilities. These techniques typically involve intertwining spools of material around a mandrel to create a cylindrical braid (Adanur, 1995). However, 3D printing, a form of additive manufacturing, continues to develop and offer many new possibilities in the realm of manufacturing. This can be credited to its process of depositing material in thin layers to gradually build the design from bottom to top (Dai & Hong, 2014). This allows for creating designs that would otherwise be impossible or incredibly difficult to realize.

The objective of this project is to test the abilities of 3D printing technology as it applies to the manufacturing of braided patterns in multidimensional space. Five differently patterned braided cylinders (diamond, regular, Hercules, tri-axial, and bifurcated) have been designed in SolidWorks and 3D printed from polylactic acid (PLA). The diamond, regular, and Hercules patterns have also been printed from thermoplastic polyurethane (TPU) at a 50% scale in size. PLA is a rigid plastic, while TPU is much more flexible (Brancewicz-Steinmetz, Sawicki, & Byczkowska, 2021). To compare the abilities of these structures, a combination of compression and tensile tests has been performed to gain knowledge of a few of their mechanical properties. Results have provided data concerning modulus of elasticity, force at yield, displacement at yield, and other measurements for each of the differently patterned structures and materials. The PLA prints underwent compression tests of the whole structures and tensile tests on separated, individual strands from the diamond, regular, and tri-axial braids. The force vs. displacement plots represent these tests in Fig. 1 and Fig. 2, respectively. Tests performed

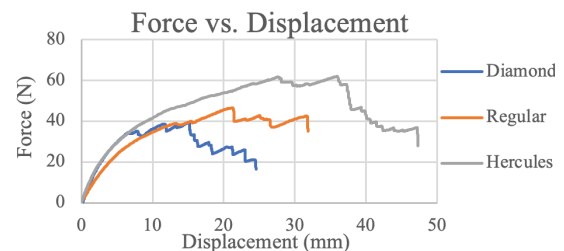
on the TPU prints were of equally portioned strips of the diamond, regular, and Hercules patterns undergoing tensile force at a constant rate, represented by the force vs. displacement plot in Fig 3. The resulting data provides insight into the capacities of both the materials and designs used for experimentation.



**Fig. 1** PLA Compression Tests Force vs. Displacement Graph.



**Fig. 2** PLA Tensile Tests Force vs. Displacement Graph.



**Fig. 3** TPU Tensile Tests Force vs. Displacement Graph.

The results of the PLA compression tests offer data re

<sup>1</sup>Corresponding author: kog0005@auburn.edu

garding the mechanical properties of the structures, such as force at yield, displacement at yield, and the initial slope of the force vs. displacement relation. The forces at yield from highest to lowest are as follows: diamond at 1234.62 N, Hercules at 1056.88 N, regular at 915.16 N, bifurcated at 658.76 N, and tri-axial at 440.21 N. The displacements at yield from highest to lowest are as follows: diamond at 5.04 mm, bifurcated at 4.39 mm, Hercules at 3.89 mm, regular at 3.75 mm, and tri-axial at 3.05 mm. It should be noted that the bifurcated prints began with a larger height than the other prints. The ranking from highest to lowest measured initial slope of the force vs. displacement curve is as follows: diamond at 949.18 N/mm, Hercules at 696.57 N/mm, regular at 525.27 N/mm, bifurcated at 292.76 N/mm, and tri-axial at 259.17 N/mm.

The tensile tests of individual strands of the PLA prints provide data of the maximum force, displacement at maximum force, tensile stress at maximum force, and Young's modulus for diamond, regular, and Hercules patterned braids. The following measurements are averages of the samples tested for each braid pattern. The maximum forces from highest to lowest are as follows: diamond at 52.34 N, Hercules at 47.28 N, and regular at 41.63 N. The displacements at maximum force from highest to lowest are as follows: Hercules at 2.425 mm, diamond at 1.74 mm, and regular at 1.24 mm. The highest to lowest measured tensile stresses at maximum forces are as follows: diamond at 16.66 MPa, Hercules at 15.05 MPa, and regular at 13.25 MPa. The ranking from highest to lowest measured Young's modulus is as follows: diamond at .31 GPa, regular at .30 GPa, and Hercules at .15 GPa.

The tensile tests for the TPU prints provide data of the maximum force, displacement at maximum force, tensile stress at maximum force, and Young's modulus for diamond, regular, and Hercules patterned braids. The maximum forces from highest to lowest are as follows: Hercules at 64.47 N, regular at 44.08 N, and diamond at 40.63 N. The displacements at maximum force from highest to lowest are as follows: Hercules at 33.8 mm, regular at 20.07 mm, and diamond at 13.89 mm. The tensile stresses at maximum force from highest to lowest are as follows: Hercules at 1.08 MPa, regular at 0.88 MPa, and diamond at 0.82 MPa. The ranking from highest to lowest measured Young's modulus is as fol-

lows: diamond at 1.17 MPa, Hercules at 0.87 MPa, and regular at 0.68 MPa.

### Statement of Research Advisor

Ms. Katherine Griffin investigated the design and 3D additive manufacturing of braided structures. First, she used SolidWorks to design various structures such as plain, regular, Hercules, triaxial, and bifurcated braids. Then, using a 3D printing machine, she produced several samples with PLA and TPU. These samples were tested in the lab for tensile strength. The results are very valuable for the advancement of 3D printing of flexible fibrous structures, which has not been done yet.

– Sabit Adanur, Samuel Ginn College of Engineering

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### Authors Biography



Katherine Griffin is a junior-year student pursuing a B.S. degree in Mechanical Engineering at Auburn University. She has studied the design and manufacturing of braided patterns and structures using 3D modeling software and additive manufacturing technology.



Ajay Jayswal completed his BS in Mechanical Engineering from Tribhuvan University, Nepal, in 2017 and his M.S. in Mechanical Engineering from Auburn University in 2021. Currently, he is a Ph.D. Student at Auburn University. He has published 5 peer reviewed journal articles, and 1 peer reviewed conference article. His research interests include additive manufacturing of polymers, computer design of textile-based structures, polymer processing and composites, viscoelasticity, and finite element modeling.



Dr. Adanur is a professor in the Mechanical Engineering Department. He has a BS in Mechanical Engineering, a M.S. in Textile Engineering and Science, and a Ph.D. in Fiber and Polymer Science. His research interests include dynamics of machinery, fibrous structures, composites, nanomaterials, and additive manufacturing of polymeric materials. He has published his research results in numerous books, articles, and conferences.

# Effects of Prescribed Fire on Water Quality and Stream Dwelling Crayfish (order: decapoda)

Josiah Gullatte<sup>1</sup>, Kaelyn Fogelman<sup>2</sup>, and Jim Stoeckel<sup>3</sup>

<sup>1</sup>Undergraduate student, School of Fisheries, Aquaculture and Aquatic Sciences, Auburn University

<sup>2</sup>Post Doc, School of Fisheries, Aquaculture and Aquatic Sciences, Auburn University

<sup>3</sup>Associate Professor, School of Fisheries, Aquaculture and Aquatic Sciences, Auburn University

Fires are a common disturbance in terrestrial ecosystems, occurring with increased frequency and duration in North America (Higuera & Abatzoglou, 2020). Fires are less volatile in the humid southeast than in the dry western United States, but they are still an important ecological and management issue (Hanby et al., 2012). Alabama's different terrestrial ecosystems are dependent on fire, from the longleaf pine ecosystem to the mixed pine and hardwood forests in the Appalachian Mountains (Varner et al., 2000). Although many studies have investigated the ecological effects of fires, few studies have evaluated the effects of fire on aquatic systems and their fauna. Specifically, no studies have quantified the effects of fire on stream-dwelling crayfish. Prescribed fire causes an increase in nutrient and heavy metal concentrations in water immediately after the fire (Klimas et al., 2020). Heavy metals are of particular concern because crayfish can bioaccumulate metals, including copper, zinc, lead, and others, in their bodies that can cause mortality and interfere with daily activities in high concentrations (Sherba et al. 2002). The brain of the crayfish reflects the environmental availability of the examined heavy metals in water. High zinc, lead, and copper concentrations lead to pathological changes in the crayfishes' olfactory midbrain (Heiba, 2006). This research is particularly important in Alabama, which has the highest crayfish biodiversity in North America, with 99 species (Schuster et al., 2022).

To evaluate how runoff from a wildfire or prescribed burn might affect water quality and stream-dwelling crayfish, we designed two experiments. The first experiment evaluated water quality and compared the effects of burnt versus non-burnt vegetation and woody debris runoff on water quality parameters, including dissolved oxygen, pH, conductivity, total suspended solids, ammonia, nitrite, nitrate, alkalinity, hardness, chloride,

sulfate, aluminum, zinc, copper, and iron. To evaluate<sup>1</sup> the effect of fire exposure on water quality parameters, we collected leaf litter for unburned and burned treatments. We used a concentration of 20 g leaf litter / 1 L of water in all experiments (Duan et al., 2014), mimicking a runoff event through a litter layer into a stream (Figure 1). Each trough used in the experiment was 40 gallons (150 L). This required 3 kg of burned or unburned leaf litter per treatment. Water was exposed to burnt or unburnt litter and was sampled for relevant water quality parameters over 24 hours.

In the first experiment, we found that water exposed to fire debris runoff was higher in dissolved oxygen, pH, total suspended solids, and conductivity over a 24-hour period. Across both treatments, ammonia was high (reaching toxic levels) in the first 6 hours, nitrite was high in the last 4 hours, and nitrate remained zero. Over the entire 24 hour period, there was no difference in aluminum ( $t(18) = 1.80$ ;  $p = 0.089$ ), copper ( $t(18) = -1.05$ ;  $p = 0.306$ ), or zinc ( $t(18) = -0.728$ ;  $p = 0.476$ ) across treatments, but water exposed to burnt litter had significantly higher concentrations of iron ( $t(18) = 3.229$ ;  $p = 0.005$ ; Figure 2). Water exposed to unburnt leaf litter was higher in chloride than water exposed to burnt leaf litter. Repeating this experiment could lead to more statistically significant data. Exposure to high concentrations of heavy metals is known to have sublethal and lethal effects on crayfish. Still, the findings of this initial study reveal that of the four metals tested, only iron was found in higher concentrations in water exposed to burn versus unburnt leaf litter.

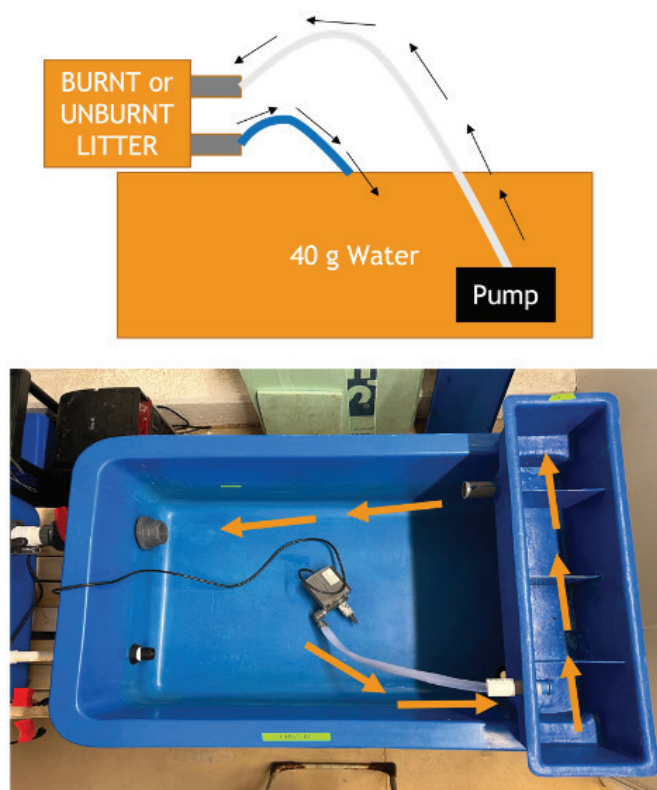
The second experiment evaluates the effects of fire debris runoff on crayfish standard metabolic rates. For this experiment, we trapped and collected crayfish in coordination with US FWS, acclimated and held them

<sup>1</sup>Corresponding author: jmg0121@auburn.edu

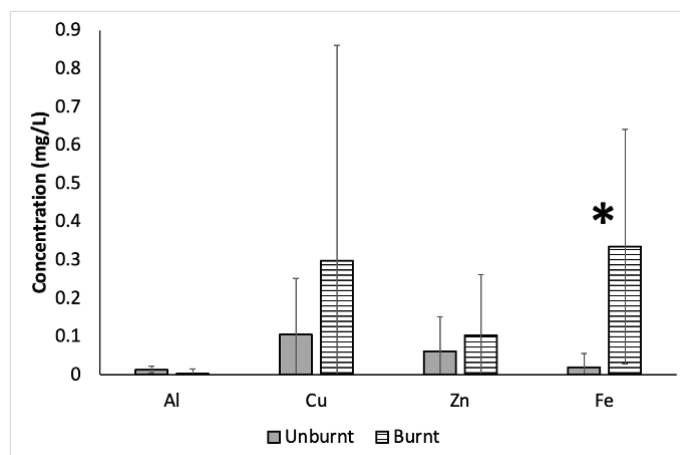


in laboratory aquatic habitats and systems, and cared for them daily. Standard metabolic rate was tested using the same experimental design as the water quality experiments (Figure 1) and used *Loligo* systems intermittent respirometry. Repeating the methods of the first experiment, we collected 50 kg of riparian vegetation, leaf litter, and woody debris to create a 3 kg burnt litter layer for exposure. We ran preliminary experiments to hone a methodology that mimicked a rainfall and runoff event through post-wildfire or post-prescribed burn fire debris. Experiments evaluating the effect of burnt runoff on crayfish metabolic rates are still ongoing due to unanticipated crayfish disease issues.

Fire events are occurring with increasing frequency and duration, and their effects on stream ecosystems and stream-dwelling fauna have not been quantified. This novel research can be used by managers when considering the relationship between fire events, water quality, and crayfish physiology.



**Figure 1:** Side and top view of experimental design simulating a runoff event through a leaf litter layer. Arrows represent the direction of water flow.



**Figure 2:** Concentrations of aluminum (Al), copper (Cu), zinc (Zn), and iron (Fe) in water exposed to 3 kg of unburnt leaf litter (grey) and 3 kg of burnt leaf litter (white and black stripe). Statistical differences in treatments are denoted with an asterisk.

### Statement of Research Advisor

In the wake of the 2020 Australian mega-fires, conversations within the international astacology community have been happening with regard to the need to evaluate the effects of wildfires on crayfish. Fortunately, Josiah had already combined his interests in prescribed fire management and crayfish ecology and proposed these experiments to Dr. Stoeckel and me. While others internationally are doing field-based evaluations of wildfire effects on crayfish populations, Josiah is the first to propose laboratory experiments evaluating the effects of fire on crayfish.

- Kaelyn Fogelman, School of Fisheries, Aquaculture, and Aquatic Sciences

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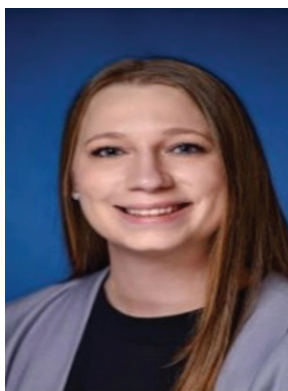
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Kaelyn Fogelman is a post-doctoral research fellow in the School of Fisheries, Aquaculture, and Aquatic Sciences. Her research is on stress physiology and the implications of climate change on imperiled and aquatic invertebrates.



Dr. Stoeckel is an Associate Professor in the School of Fisheries, Aquaculture, and Aquatic Sciences at Auburn University. His lab focuses on crustacean and molluscan ecology with a range of research topics within the fields of conservation, ecotoxicology, ecophysiology, and invasive species.

## Authors Biography



Josiah Gullatte is a senior-year student pursuing a major in Wildlife Ecology & Management with a minor in Fisheries and allied aquaculture at Auburn University. He is a certified burn boss and a technician in the Crustacean and Molluscan Ecology Lab (CAMEL) under Dr. Jim Stoeckel.

# 3D Printed Biodegradable Implants Loaded with Capsaicin for the Treatment of Obesity and its Related Health Risks

Jessica Heard<sup>1</sup>, Manjusha Annaji<sup>2</sup>, Nur Mita<sup>2</sup>, and Jayachandra Ramapuram<sup>3</sup>

<sup>1</sup> Undergraduate Student, College of Science and Mathematics, Auburn University

<sup>2</sup> Graduate Student, Harrison College of Pharmacy, Auburn University

<sup>3</sup> Professor, Harrison College of Pharmacy, Auburn University

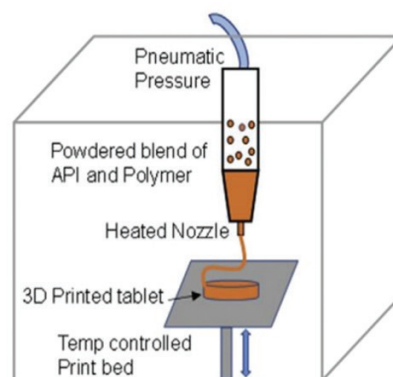
Obesity increases the risk of various health complications, including type 2 diabetes, hyperlipidemia, and cardiovascular disease.<sup>1</sup> The current FDA-approved drug therapies designed to combat obesity are limited and known to have severe side effects. Therefore, there is an urgent need to develop natural drug therapies with reduced side effects.

Research studies have shown that capsaicin, the primary ingredient in chili peppers, is effective in promoting weight loss and reducing obesity-related health conditions.<sup>2</sup> The anti-obesity effects of capsaicin have previously been studied in animal and human models; however, these studies were conducted by administering the drug orally.<sup>3</sup> Oral administration cannot be sustained long-term due to the pungency and poor oral bioavailability of capsaicin. This research project aimed to identify an alternative delivery method for capsaicin.

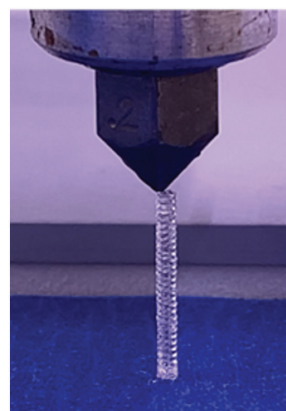
Through an extrusion-based 3D printing method, biodegradable implants were designed to potentially deliver capsaicin directly to the fatty tissue for a prolonged period. Since the implants will be inserted directly under the skin to access the fat regions, the sustained release aspect is vital to reduce the need for multiple invasive procedures and medical appointments. The 3D printing technology may also improve the treatment process as it allows for personalized dosage forms that best fit the needs of clinical patients.

The Cellink BIOX 3D printer prepared the rod-shaped implants through a direct powder extrusion-based printing method. This single-step method allows a blend of the drug and polymer to be inserted directly into the printer, eliminating the need for any initial

manipulation. As shown in Figure 1, the drug-polymer<sup>1</sup> blend is melted within the print head and extruded through a nozzle layer by layer due to the application of high pressure.<sup>4</sup> The final product is depicted in Figure 2.



**Fig. 1** Schematic of extrusion-based 3D printing method.



**Fig. 2** Rod-shaped implant loaded with capsaicin being extruded from the 3D printer nozzle.

The polymer used in creating the implants was chosen for its biodegradable properties and wide usage in

<sup>1</sup>Corresponding author: ljh0025@auburn.edu



controlled release medical devices. Several parameters, including pressure, nozzle diameter, and print bed temperature, were optimized to ensure the implants were well-formed and of the highest quality possible. To maintain the thermal stability of the drug, the printing temperature was optimized to 180°C.

Several studies were conducted to investigate the chemical and physical properties of capsaicin within the implants. An assay was performed to measure the total drug content within each implant, ensuring that the drug maintained thermal stability during the printing process and the drug content of each implant was uniform. To assess the long-term sustainability of capsaicin, an *in vitro* release study was conducted to measure the duration of drug release from the implants under static conditions. A stability study was conducted to ensure the drug remained stable over time under exposure to extreme temperatures. A forced degradation study was conducted to investigate the rate of drug degradation under various chemical conditions.

Overall, the results from this research will determine if 3D-printed devices may be helpful in the site-specific administration and sustained release of capsaicin. Once the implants are thoroughly studied and characterized, the devices will be inserted subcutaneously in a high-fat mouse model to further investigate the effects of capsaicin on diet-induced obesity. All data and findings will be presented in a peer-reviewed journal once this research project is complete.

### Statement of Research Advisor

Jessica Heard has significantly contributed to current ongoing research on capsaicin implants preparation and characterization. She has performed experiments daily and generated data on capsaicin stability and release from the implants.

- Jayachandra Babu Ramapuram, Department of Drug Discovery and Development, Harrison College of Pharmacy

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### Authors Biography



Jessica Heard is a senior-year student pursuing a B.S. degree in Biochemistry at Auburn University. As the 2022 undergraduate research fellow, she has played key research roles in the studies related to capsaicin loaded 3D-printed implants.



Manjusha Annaji is a Ph.D. student at Auburn University, working under the guidance of Dr. Jay Ramapuram. Her research focuses on the fabrication of different 3D printed dosage forms such as orodispersible films, polypills, and polymeric implants for treating various chronic diseases. She is the creator of capsaicin loaded 3D-printed implants and provided the research protocols.





Nur Mita is a Ph.D. student at Auburn University, working under the guidance of Dr. Jay Ramapuram. Her works focus on developing intravenous injections, 3D printed-orodispersible films, and in situ forming implant formulations. She served in the supervisor role and has provided daily guidance on the stability and drug release studies and assisted in data curation and presentation.



Dr. Jayachandra Babu Ramapuram, Professor, Department of Drug Discovery and Development, Harrison College of Pharmacy, Auburn University, is the research mentor. He is involved in conceptualization, study design, and data analysis.

# The Effects of a Storm's First Flush On Nutrient Cycling and the Microbial Assemblage in Parkerson Mill Creek, Auburn, AL

Emma D. Henderson<sup>1</sup> and Ann S. Ojeda<sup>2</sup>

<sup>1</sup> Undergraduate Student, Department of Geosciences, Auburn University

<sup>2</sup> Assistant Professor, Department of Geosciences, Auburn University

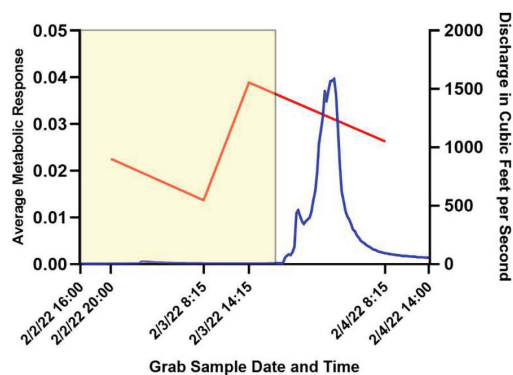
A 'first flush' is the initial surface runoff from a rainstorm, lasting about 24 hours after precipitation begins. First flushes are characterized by the disproportionate loading of contaminants or pollutants into the surface waterbody during the rising limb of the hydrograph when compared to lower loading during the peak or falling limb (Griffin et al., 1980). Recently, Peter et al. (2020) recognized that peak contaminant loading for stream systems occur prior to or coincidently with that stream's hydrograph peak. However, the microbial response to the first flush, and consequent nutrient and pollutant biogeochemical cycling are less understood. Thus, the purpose of this research was to observe how a microbial community responds to a first flush and concurrent changes in water chemistry. We hypothesized that the microbial community would either grow due to an influx of potential nutrient sources in the water brought in from the first flush, or the community would be diluted by the stormwater.

For this research project, water was sampled from Parkerson Mill Creek (PMC) in Auburn, AL, south of Auburn University. PMC is a first order stream that is part of the larger Tallapoosa River watershed. Its headwaters are on the Auburn University campus near the Beard-Eaves Memorial Coliseum, and it flows into Chewacla Creek south of Chewacla State Park. Single, time-representative grab samples from PMC were collected every 6 hours during rainstorms beginning at initial precipitation and lasting for 36 hours.

However, samples were not collected after 10:00 pm or before sunrise, nor were they taken during severe weather. When the grab samples were collected, a YSI EXO2 sonde was deployed to record temperature, pH, dissolved oxygen (DO), turbidity, fluorescent dissolved

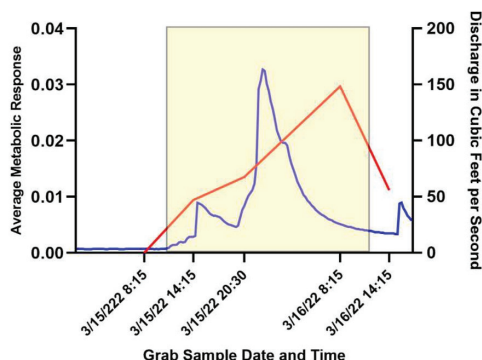
organic matter (fDOM), and specific conductivity (SPC). A total of two storm events were sampled for this study on February 2, 2022, and March 15, 2022.

The microbial community was cultivated from the samples using Biolog Ecoplates™ (Garland, 1997). Biolog Ecoplates™ are well plates that contain 31 different carbon substrates in triplicate, and the microbial community is quantified based on the ability of the community to utilize the array of substrates. Metabolism of the carbon substrate produced a purple color, and the optical density of each plate was recorded every 24 hours to determine the degree of activation in each well. An optical density of 0.25 or higher indicated the carbon substrate had been activated. Once the optical density data were recorded, the average metabolic response (AMR) was calculated for each well plate. The AMR is the average activation level of the carbon source wells against the control (water) and is recorded as a single value, allowing for comparison between Ecoplates (Sigler, 2004). Next, the AMR for each storm event was graphed against PMC's streamflow during respective sample collection time. Figures 1 and 2 show the AMR versus the stream hydrograph for storms 1 and 2 respectively, with the yellow box indicating the first flush 24-hour period.

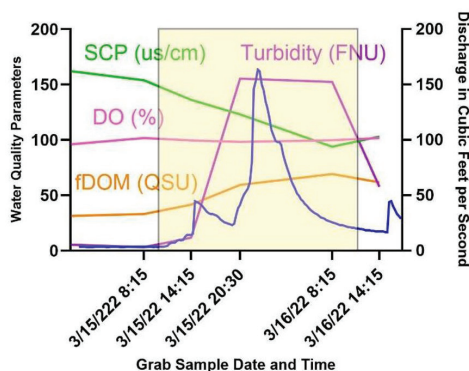


<sup>1</sup>Corresponding author: edh0040@auburn.edu

**Fig. 1** Storm 1: AMR (red line) is plotted against stream-flow (blue line). The yellow box indicates the first flush 24-hour period.



**Fig. 2** Storm 2: AMR (red line) is plotted against stream-flow (blue line). The yellow box indicates the first flush 24-hour period.



**Fig. 3.** Storm 2: Water quality parameters are plotted against streamflow (blue line). The yellow box indicates the first flush 24-hour period.

Both storms 1 and 2 show that the AMR increases during the first flush period after the initial precipitation. The AMR did not decrease again until after the streamflow peak and the first flush window have passed. This data suggests that there is an increase in the concentration of microbial communities during the first flush. We also found that the community metabolic diversity increased during the storm events, but not in a predictable pattern. More frequent sampling to better match the high-resolution data from the hydrograph could help to better understand a more precise relationship between a first flush and the stream's microbial community. In storm 2, we found that turbidity, fDOM, and DO all increased in concentration and SCP was diluted with respect to the first flush. According to Hathaway et al. (2012), these relationships are not the

same for every stream, but could be unique to PMC. Increased turbidity and fDOM are an indicator that nutrient availability (dissolved organic carbon, particulate carbon, and particulate nitrogen) could be also increasing (Snyder et al., 2018). Storm water samples could be further analyzed to identify the phylogenetic diversity of the microbial community present and how contaminants or pollutants change in response to the dynamics of the microbial community.

## Statement of Research Advisor

Parkerson Mill Creek is an impaired waterbody, meaning it doesn't meet water quality standards set by the Alabama Department of Environmental Management. Emma's work helps us understand how microbial communities respond to storm events. Ultimately, her research will better inform nutrient cycling and contaminant fate and transport in this stream.

- Ann Ojeda, COSAM, Department of Geoscience

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## Authors Biography



Emma Henderson is a graduate of Auburn University's class of May 2022 with a B.S. degree in Geology.



Ann Ojeda studies geology, environmental science, and health science to understand how the geosphere controls health outcomes. Her research advances human health and community resilience through (1) the geo-environmental controls on drinking water contaminants and aquifer vulnerability, (2) controls on freshwater resources, and (3) community dynamics and health outcomes from exposure to untreated or undertreated drinking water supplies



# Transport and Physiochemical Properties of Polyether-based Hydrogel Membranes for Ammonia Fuel Cell Applications

Brock J. Hunter <sup>1</sup>, Grayson P. Chapman <sup>1</sup>, and Bryan S. Beckingham <sup>2</sup>

<sup>1</sup> Undergraduate Student, Department of Chemical Engineering, Auburn University

<sup>2</sup> Assistant Professor, Department of Chemical Engineering, Auburn University

Ammonia production is critical to maintaining global fertilizer supplies. The Haber-Bosch process, the production method used today, led to the quadrupling of the global human population with the increased food production capacity. However, this process requires high temperature and pressure to facilitate reaction kinetics. This leads to massive CO<sub>2</sub> production and energy usage totaling to 1-2% of global energy consumption. Researchers are trying to develop methods to produce ammonia through electrochemistry. Such devices can use electricity instead of heat to facilitate the reaction, and they can operate at ambient conditions. The typical setup for this cell can be seen in Figure 1. The focus of developing these cells has been finding a suitable catalyst. However, the membranes currently used these cells to allow a significant amount of ammonia crossover, leading to the oxidation and loss of products. Better membranes are needed than the current standard, Nafion-117, for electrochemical cells to be more efficient than the Haber-Bosch process. Here, new membrane chemistries are explored for applications in electrochemical ammonia fuel cells, and trends in membrane characteristics such as charge and water uptake are correlated to ammonia transport properties.

The analyzed membranes are composed of three main components in a DMSO solution: a crosslinker (methylenebis (acrylamide)), a negatively charged monomer (2-acrylamido-2-methyl-1propanesulfonic acid), and a hydrophobic monomer (either phenyl acrylate (PA) or phenyl methacrylate (PMA)). The solutions were thermally crosslinked and solvent exchanged with water to form thin membranes. The membranes were installed in a H-cell similar to Figure 1. One side of the cell was filled with a 1 M ammonium chloride solution, and the other side contained pure DI water. Conductivity was measured on the water side as ammonia crossed over, and this was used to calculate membrane permeability. The membranes were also tested for water uptake by finding the difference between wet and dried films.

Overall, PMA based membranes showed lower permeability to ammonia than PA membranes. This trend matched the trend in relative water uptake. Increased water content in a film typically means there is more free volume in the membrane which allows for molecular diffusion (i.e., ammonia crossover); however membrane density must be measured before this can be confirmed. There was also a trend seen in the amount of charged monomer in the membrane and crossover. Increased charge content led to increased ammonia crossover. Ammonia is positively charged in solution, so the charged monomer has an affinity for ammonia and aided in its transport. Solubility data would help solidify these findings, as it would probe this thermodynamic affinity. Some PMA membranes showed similar permeabilities to Nafion-117, and other membrane chemistries can be tested to improve performance.

These findings have shown manipulating membrane chemistry for ammonia fuel cells could lead to reduced

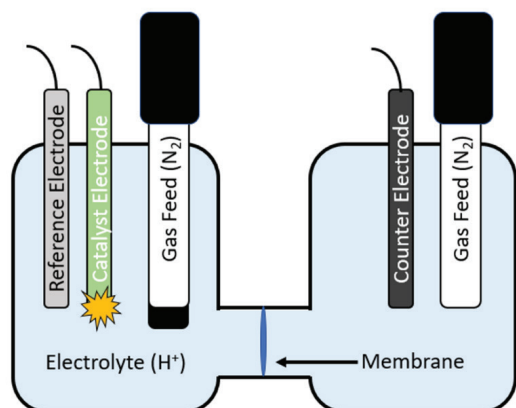


Fig. 1 Ammonia Fuel Cell

<sup>1</sup>Corresponding author: bjh0062@auburn.edu

ammonia crossover, and further studies are needed to solidify membrane objectives for this application. Trends found for membrane charge and water uptake give clear direction to how future membranes in this area should be designed.

### Statement of Research Advisor

Brock has synthesized and characterized a series of new, polymer membranes with an aim at improving the operating efficiency of direct ammonia fuel cells by reducing ammonia crossover. Through his study, he demonstrated the relationship between membrane permeability to ammonia and polymer backbone chemistry, where the addition of a single backbone methyl group per repeat unit had a significant impact on permeability. Additionally, he found that the membrane permeability to ammonia could be tuned through the amount of comonomer incorporated into the membrane. He has set the stage for further investigation of structure-property relationships in this class of polymer membrane and my research group is looking forward to building upon his work towards designing new, improved polymer membranes for the sustainable production of ammonia.

- Bryan Beckingham, Engineering

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### Authors Biography



Brock J. Hunter has recently received his B.S. in Chemical Engineering at Auburn University. He has researched many aspects of nitrogen reduction fuel cells, including membrane design and novel catalysis mechanisms.



Grayson P. Chapman is a junior-year undergraduate studying chemical engineering at Auburn University. He has interests in research pertaining to membrane transport properties.



Bryan S. Beckingham is an assistant professor in the Chemical Engineering Department at Auburn University. He leads a polymer research group exploring polymer chemistry for membranes and 3D printing.

# Sediment Core Evidence of Historical Phosphate Mining and Harmful Algal Blooms in Banana Lake, Florida

*Susan G. Iott<sup>1</sup>, Matthew N. Waters<sup>2</sup>, and Troy L. Clift<sup>3</sup>*

<sup>1</sup> Undergraduate Student, Department of Crop, Soil, and Environmental Sciences, Auburn University

<sup>2</sup> Associate Professor, Department of Crop, Soil, and Environmental Sciences, Auburn University

<sup>3</sup> Graduate Student, Department of Crop, Soil, and Environmental Sciences, Auburn University

## Abstract

While most monitoring programs successfully document eutrophication, the development of eutrophic characteristics often occurred prior to recent monitoring efforts. To reconstruct historic lake changes, paleolimnological techniques and the sediment record can provide data preceding monitoring programs. Here, we measured nutrients (C, N, P) and photosynthetic pigments in a sediment core collected from Banana Lake, a shallow (mean depth 1.2m) and hypereutrophic lake located in Polk County, Florida. In the case of Banana Lake, excess nutrients have allowed cyanobacteria to become the dominant primary producers utilizing lake nutrients and creating a positive feedback loop that intensifies eutrophication. By combining nutrient and pigment data, our results show that cyanobacteria existed in the lake long before monitoring efforts began in the system. Maximum P levels appear to be substantially higher than those of comparable lakes, likely caused by the lake's proximity to phosphate mines and the development of a viable meroplankton community. Additionally, toxins associated with cyanobacteria were found in large quantities in sediments spanning the last 30-40 years. An attempt to manage eutrophication was made in 1991 by removing approximately one million cubic yards of sediment through dredging. Despite these efforts, Banana Lake is still classified as an impaired water body with high sedimentary P, viable meroplanktonic cyanobacteria, and high microcystin concentrations; a fish consumption advisory is also in effect. Management efforts would benefit from coupling monitoring and management efforts with sediment analysis in shallow systems where internal nutrient loading could be the primary driver of persistent eutrophication.

## Introduction

Alterations to land use and urban development around lake ecosystems can lead to the influx of nutrients and the formation of dense algal and cyanobacteria blooms, called cultural eutrophication. These blooms decrease ecosystem services by depleting oxygen and weakening biodiversity while also impacting human health through the production of toxins called cyanotoxins. While most monitoring programs successfully document eutrophication, the development of eutrophic characteristics often occurs decades prior to recent monitoring efforts. To reconstruct historic lake change, paleolimnological techniques applied to sediment cores can be used to provide data preceding monitoring programs. Our study applied these tools to a sediment core collected from Banana Lake; the lake currently exists in an urban setting and maintains a hypereutrophic state. The history of Banana Lake can be divided into three periods based on land management practices and levels of nutrients: an agricultural period, a quiescent period, and a phosphate mining period.

Beginning in the early 1820s, Creek Indian chief Oponay maintained a large plantation east of Lake Hancock that grew to encompass the area between Lake Hancock and Banana Lake. However, the plantation and the black and native settlements were destroyed during the Second Seminole War from 1835-1842. Banana Lake was left relatively undisturbed until the Southern Phosphate Corporation began mining phosphate on the west side of the lake around 1924. The historical trophic state of Banana Lake, as well as urbanization and land management practices, were investigated as possible causes for the lake's eutrophication. Eutrophication and the presence of toxin-producing cyanobacteria have led to the

<sup>1</sup>Corresponding author: [sgi0002@auburn.edu](mailto:sgi0002@auburn.edu)



lake's fish consumption advisory and impaired status. While most monitoring programs successfully document eutrophication, the lake's eutrophic characteristics were present prior to recent monitoring efforts. As a result, the primary objective of our study was to determine when the hypereutrophic conditions began in time and to determine if one of the primary land use alterations triggered the decrease in lake health.

## Methods

A sediment core was collected from the middle of Banana Lake and sectioned into 4-cm increments. Each section was freeze-dried and ground with a mortar and pestle. Ground sediments were analyzed for nutrients (carbon, nitrogen, phosphorus), photosynthetic pigments, and cyanotoxin, microcystin. Carbon and nitrogen content was determined through elemental combustion analysis, and phosphorus and other elements were determined through Inductively Coupled Plasma analysis. Pigments and microcystin were extracted and measured with High-Performance Liquid Chromatography and spectrophotometry, respectively. Photosynthetic pigment analysis measures a variety of chlorophylls and carotenoids capable of diagnosing total primary producer abundance, phytoplankton community structure, and cyanobacteria presence through time.



**Fig. 1** Collection of a sediment core using a core barrel.

## Conclusions

Cyanobacteria pigments occurred throughout the sedi-

ment core, indicating that Banana Lake has been hypereutrophic as far back as a few hundred years and encompassing the period during which human activities included the historic agricultural period. Surprisingly, the cyanobacteria community has remained dominant and constant despite multiple dramatic changes to nutrient inputs into the lake through time. Whereas the cyanobacteria community has remained dominant, phosphorus data emerged as the most interesting aspect of the lake's history. At approximately 52 cm in depth along the core, total phosphorus dramatically increased beyond levels previously observed in similar lake systems. Nearby Lake Apopka, a lake known for its problematic cyanobacteria community, contains sedimentary phosphorus concentrations around 2 mg/g while the most recent period of P concentrations in Banana Lake are around 30 mg/g. Phosphate mines are the most likely cause for such a large and dramatic shift, with the two mines that would become Stahl Lake and Little Banana Lake being established around 1924. The phosphorus concentration from 52-cm to 8-cm depth is so large that no amount of agricultural input from local orange groves could account for it. While a wastewater treatment plant began discharging effluent into Banana Lake at around the same time, it is unlikely to cause such an increase. Effluent standards became stricter over time, and there is no decreasing trend of phosphorus from 52 cm to 8 cm.

Management decisions have primarily been based on the last ten years of monitoring data. However, understanding the long-term drivers of eutrophication, rather than the endpoint, will better inform such efforts. Our data suggest that Banana Lake has experienced eutrophic events throughout its recent history linked to dynamic changes in lake management and surrounding landscapes. Pigments also show a community shift between species of cyanobacteria; however, this shift does not correlate with the extreme differences in phosphorus and other nutrients. In the presence of unprecedented phosphorus and fluctuating nutrients, the cyanobacterial community in Banana Lake appears relatively unaffected, suggesting a possible ecological threshold was achieved through minimal land use practices of the past. Microcystin, on the other hand, increases logarithmically in the top 32 cm of the sediment. Toxin increases combined with constant cyanobacteria and continued impairment status signal that



the dredging of Banana Lake in 1991 was largely ineffective in controlling eutrophication. Attempts to stop or reverse eutrophication may be fruitless in the context of a naturally hypereutrophic lake near phosphate deposits.

## Statement of Research Advisor

Susan's work has been extremely extensive in analyzing her sediment core from Banana Lake. Her results have demonstrated evidence of extreme impacts on urban lake systems with a particular focus on environmental analysis of phosphate mining in this area of Florida. In addition, Susan has performed an extensive investigation into historical documents to establish the timeline for changes in her data. This timeline resulted from Susan connecting with local stakeholders and historians in the area, which was above and beyond the expectations of the project. Finally, by coupling her phosphate mining impacts with Banana Lake being in a continual harmful algal bloom, Susan's work is beginning to establish ecological thresholds linking landscape processes with aquatic responses.

- Matthew Waters, *Crop, Soil, and Environmental Sciences*

## Acknowledgments

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## Authors Biography



Susan G. Iott is a senior-year student pursuing a B.S. degree in Environmental Science at Auburn University. As an undergraduate research fellow and student worker in the Auburn Paleoenvironmental Laboratory, she has contributed to research in eutrophication and cyanotoxin production in sub-tropical lakes.



Matthew N. Waters is an Associate Professor of Environmental Sciences at Auburn University, where he leads the Auburn Paleoenvironmental Laboratory. Trained as a classic limnologist and paleolimnologist, Dr. Waters and his students attempt to develop and apply geochemical analysis on sediment cores collected from natural lake systems to understand better the ecological dynamics of cyanobacteria and the triggers of cyanotoxin production. His studies focus on the SE USA and Mesoamerica.



Troy L. Clift is in the second year of his Crop, Soil, and Environmental Science master's program at Auburn University. Originally from Virginia, he attended Longwood University, where he was awarded a B.S. in Integrated Environmental Science. His research investigates the drivers of cyanotoxin production in sub-tropical lakes in central Florida, USA.

# Integration of Bioactivity – Global Natural Product Social Molecular Network (GNPS): Proof of Conception Açai

Madeline L. Kunze<sup>1</sup>, Kabre L. Heck<sup>2</sup>, and Angela I. Calderón<sup>3</sup>

<sup>1</sup> Undergraduate Student, Department of Chemical Engineering, Auburn University

<sup>2</sup> Graduate Student, Department of Drug Discovery and Development, Harrison School of Pharmacy, Auburn University

<sup>3</sup> Associate Professor, Department of Drug Discovery and Development, Harrison School of Pharmacy, Auburn University

*Euterpe oleracea* Mart, commonly known as açai, is a fruit present in botanical dietary supplements (BDS) frequently taken by cancer patients. The açai extracts have cancer-related properties: antioxidant, antiproliferative, cytoprotective, and anti-inflammatory. The goal of my research is to prove that Global Natural Product Social Molecular Network (GNPS) can be integrated with a bioactivity tool and produce a visual molecular network for açai and identify known and unknown compounds related to these anti-cancer properties. GNPS is a cutting-edge software program which has been integrated with bioactivity in our laboratory.

This method compares two different commercially available açai supplements prepared using acidic methanol. The extraction procedure consisted of maceration of the capsule powder, followed by sonication and evaporation of the solvent. Liquid chromatography-mass spectrometry (LC-MS) was used to identify the mass of compounds from the two different acidic methanol extracts of açai formula 1 (F1) and formula 2 (F2). Parallel artificial membrane permeability assay (PAMPA) mimics a human gastric intestinal track and shows how much of the extract is passively diffused, on the acceptor side and non-passively diffused, on the donor. PAMPA was done at a range of concentrations (1,000-1.95 µg/mL), which accounts for human relevant doses (4.494-6.557 µg/mL) and potential overdose. CYP3A4 is a major liver enzyme that metabolizes many anticancer drugs; the donor and acceptor site compounds have been tested for inhibition of hepatic CYP3A4. The CYP3A4 inhibition assay was analyzed through LC-MS of each extract formulation by measuring the amount of

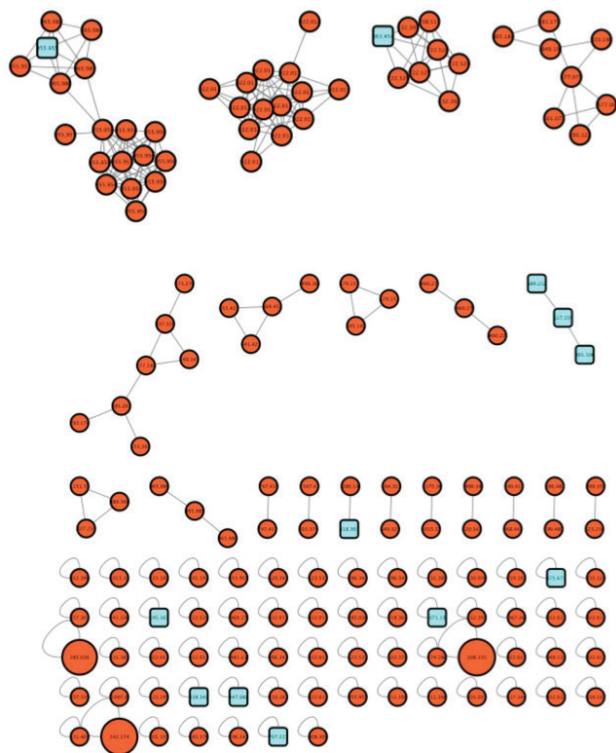
metabolite made in the presence of açai extract. The last step is to compile all the data collected from LC-MS and MS/MS and upload it to GNPS. GNPS is the core computer program used, but other software programs also aid in the process (MZmine, Rstudio, and Cystoscope) of creating a visual bioactive molecular network.

The LC-MS identified which compounds were present in each extract formulation and compared to a chemical fingerprinting library of all açai compounds previously identified. The two formulations, even though both aqueous extracts of açai, had very different chemical compositions. BDS are commonly not standardized or strictly regulated. CYP3A4 inhibition was anticipated for F1, since we had also observed CYP3A4 inhibition previously with the methanol extracts. Inhibition by F2 was not as well observed; the acceptor side had no inhibition and the donor side had very slight inhibition. Figure 1 shows how all the data collected can be integrated to generate a bioactive visual molecular network for açai acidic methanol extracts. This figure identifies the mass of each compound, along with if it is known or unknown, and the ones that have high bioactivity and low bioactivity.

This research shows that Integrated Bioactivity – Global Natural Product Social Molecular Network (GNPS) can differentiate known and unknown natural products from a botanical extract and aid in the creation of a visual molecular network of bioactive açai constituents. In the future, the GNPS-Bioactivity tool can be used for other botanical dietary supplements and the isolation of high bioactive compounds.

<sup>1</sup>Corresponding author: mlk0040@auburn.edu





**Fig. 1.** Visual molecular network produced with GNPS, blue squares are known & orange circles are unknown.

### Statement of Research Advisor

Madeline has learned to perform LC-MS analysis of botanical dietary supplements and analyze data using Integrated Bioactivity – Global Natural Product Social Molecular Network (GNPS) for identification of CYP3A4 inducers in açai. Madeline developed strong natural products chemistry literature reading and problem-solving abilities. Her contribution was critical to providing an example of a shorter and less laborious process to identify bioactive compounds in complex botanical extracts.

- Dr. Angela I. Calderón, Harrison College of Pharmacy

### Authors Biography



Madeline Kunze is a junior-year student pursuing a B.S. degree in Chemical Engineering at Auburn University. She has played key research roles in integration of GNPS with the bioactivity tool and in writing a review paper about the benefits of traditional medicine for anticancer treatment.



Kabre L. Heck is a Graduate Teaching Assistant from the Harrison College of Pharmacy in the Department of Drug Discovery and Development. Kabre received her undergraduate degree in Medical Studies from Lenoir-Rhyne University in 2019 and is currently pursuing a PhD in Pharmaceutical Sciences with an emphasis in Medicinal Chemistry. In the Natural Products Laboratory of Dr. Calderón, Kabre investigates potential interactions between pharmaceuticals and botanical dietary supplements and utilizes liquid chromatography-mass spectrometry for quantitation of metabolites and performing structural elucidation.



Angela I. Calderón, Ph.D. is an Associate Professor in the Department of Drug Discovery and Development, Harrison School of Pharmacy, Auburn University. She received her degrees in Pharmacy and Pharmacognosy from the University of Panama, Panama, the University of Illinois at Chicago, and the University of Lausanne, Switzerland. Dr.



Calderón specializes in natural drug products chemistry. She works to apply mass spectrometry to natural products drug discovery, and quality and safety assessment of botanical dietary supplements. Dr. Calderón enjoys educating graduate and undergraduate students as the next generation of natural products researchers. with unique skills in mass spectrometry.

# Trauma Exposure and Impulsivity Symptoms in Court-Involved Youth

Madysen E. Maggio <sup>1</sup> and Kelli R. Thompson <sup>2</sup>

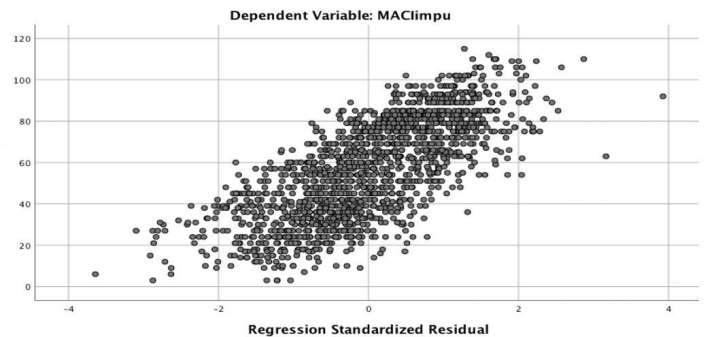
<sup>1</sup> Undergraduate Student, Department of Psychological Sciences, Auburn University

<sup>2</sup> Assistant Research Professor, College of Liberal Arts, Auburn University

In juvenile justice settings, adolescents with complex trauma histories often develop a wide range of mental health consequences such as depression, anxiety, poor impulse control, or aggression; and are often labeled with potentially unwarranted diagnoses as a result (Ford, Chapman, Connor, & Cruise, 2012). Many symptoms of attention deficit disorders (e.g., inattentiveness, daydreaming, drowsiness, and impulsivity) may be the result of trauma-related internalizing disorders, thereby complicating the primary diagnosis (Becker & Langberg, 2012). Adolescents adjudicated for illegal sexual behavior report higher rates of trauma-related psychopathology, with one study reporting 77.5% of the sample experienced at least one traumatic event (Newman et al., 2018). Once incarcerated, the cumulative effect of complex trauma and poly-victimization has a considerable impact on the cognitive, behavioral, and mental outcomes of these youth (Alexander, McCallum, & Thompson, 2020). Therefore, understanding how trauma impacts the development of these behavioral symptoms can improve accurate diagnosis and subsequent treatment for either attention deficit-hyperactivity disorder (ADHD) or post-traumatic stress disorder (PTSD).

The current study examined the role trauma exposure played in symptoms of impulsivity in a sample of adjudicated male youth. Participants included youth ( $n = 1,597$ ) in a court-mandated residential treatment facility. The Millon Adolescent Clinical Inventory (MACI; Millon & Davis, 1993, 2003) was used to assess impulsive propensity, family discord, and childhood abuse. Linear regression analyses were used with the child abuse and family discord scales entered as predictors and the impulsivity scale as the dependent variable. The overall model was significant,  $F(2, 1596) = 690.78$ ,  $p = .000$ , indicating a positive association between trauma

predictors and impulsivity. Family discord primarily<sup>1</sup> drove the association ( $\beta = .68$ ,  $p = .00$ ), accounting for 42.3% of the variance. This relationship is depicted in Figure  $\beta$  The child abuse scale was not a significant predictor ( $\beta = .02$ ,  $p = .42$ ) of impulsive symptoms.



**Fig. 1** Scatterplot of main findings.

These results highlight the need for adequate trauma assessment and screening when working with court-involved youth. Those witnessing indirect trauma in homes characterized by high family conflict and tension were positively associated with higher rates of impulsive and aggressive behavior. These external behavioral symptoms are common to ADHD, but also to childhood PTSD. These results imply that inadequate trauma assessment in youth could increase the chance for misdiagnosis. These results also speak to the socialization of adolescent males and the potential harms associated with toxic forms of masculine expressions. An inability to properly name and express trauma-related emotions may lead to explosive behaviors and impulsivity, which could lead to law enforcement or subsequent court involvement (Mulvey and Iseline, 2008). Furthermore, studies of cortisol reactivity have found multiple response patterns for youth in high-stress environments, such as those with high family or neighborhood conflict (Guidice, Ellis, & Shirtcliff, 2011). Those

<sup>1</sup>Corresponding author: mem0214@auburn.edu

with mild to moderate trauma showed high reactivity patterns characteristic of the impulsive symptoms seen here; while others with more severe trauma showed a numbing pattern not displayed here. Future studies are needed to truly understand the various neurological impacts of trauma exposure.

## Statement of Research Advisor

Madysen's research highlights some of the excellent undergraduate scholarship from the Juvenile Delinquency Lab in the Department of Psychological Sciences. The lab is supported by a more than 20-year public-public partnership with the Alabama Department of Youth Services. She will expand on this theme as an upcoming 2022-2023 College of Liberal Arts Undergraduate Research Fellow.

- Kelli. R. Thompson, *College of Liberal Arts*

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## Authors Biography



Madysen E. Maggio is a senior-year student pursuing a B.A. degree in Psychology at Auburn University. She has played key research roles in the misdiagnosis of ADHD in the juvenile population. She is a double minor in social work and counseling. She plans to work with adolescents in the Foster Care System and address early onset trauma.



Kelli. R. Thompson is an Assistant Research Professor and Director of the Juvenile Delinquency Lab in the Department of Psychology at Auburn University. She received her B.S. in Human Development and Family Studies from Auburn in 2005. She received her M.A. in Theology from Fuller Theological Seminary in 2008 and her Ph.D. in Applied Developmental Psychology from the University of New Orleans in 2016.

# Acquisition and Development of the Gut Microbiome in Lizards

Taylor W. McKibben <sup>1</sup>, Kaitlyn M. Murphy <sup>2</sup>, and Tonia S. Schwartz <sup>3</sup>

<sup>1</sup> Undergraduate Student, Department of Mechanical Engineering, Auburn University

<sup>2</sup> Ph.D. Candidate, Department of Biological Sciences, Auburn University

<sup>3</sup> Associate Professor, Department of Biological Sciences, Auburn University

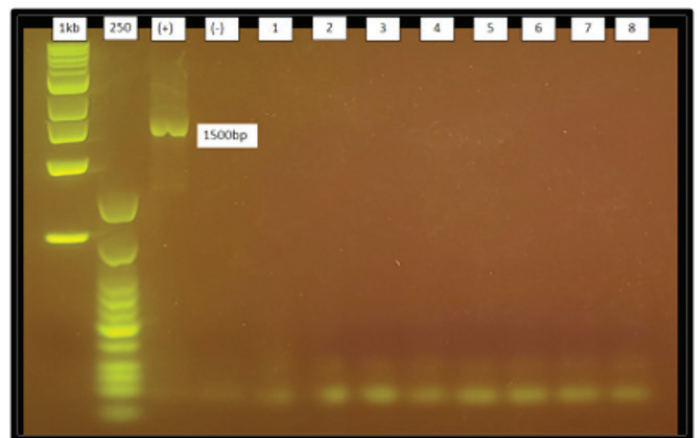
The gut microbiome plays a pivotal role in the health of an animal. While studies have addressed the acquisition of the gut microbiota in placental mammals, limited research has been conducted on the origin of reptilian gut microbiota, especially for oviparous species. Dietary changes in early life have also been shown to play a major role in determining the composition of mammalian gut microbiota; early development of the gut microbiota has positive lifelong health repercussions<sup>1-4</sup>. In this study, we address two pivotal questions in an emerging model of an oviparous lizard (*Anolis sagrei*, brown anole): (1) is there microbial colonization in the embryonic gut, and (2) how does the diversity of the diet shape community composition in early life.

To answer these questions, we collected eggs from a captive laboratory colony of *A. sagrei*, extracted DNA from embryonic and hatchling gut tissue, and analyzed the resulting 16S rRNA gene sequences of each sample using the Oxford Nanopore minion 16S sequencing kit.

We first established a breeding colony of 76 wild-caught brown anoles from Palm Coast, Florida. The breeding colony's living conditions and the incubation conditions were similar to previous experiments except that our experimental eggs were being sanitized through bleach immersion. These animals were kept in breeding pairs and fed a diet of crickets and watered daily. The eggs were collected every 2 to 3 days, bleached in a 1:5 bleach to DI water solution to remove external bacteria, and placed in sterile petri dishes with autoclaved vermiculite within an incubator. Eggs were randomly assigned to one of three groups: (1) dissected at an embryonic stage, (2) hatchlings fed a control diet, (3) hatchlings fed a diverse diet. Eggs assigned to the embryonic timepoint were dissected approximately five days before hatching. Eggs assigned a dietary treatment

were fed their diet for approximately 5 weeks. Dietary<sup>1</sup> treatments were either a control diet of only fruit flies or a diverse diet consisting of fruit flies, cockroaches, and crickets.

After dissecting all embryos and hatchlings to procure their intestinal tracts, the tissues were processed to isolate DNA using Zymo Quick-DNA Fecal/Soil Microbe Minikits. Eluates were amplified utilizing PCR with the Oxford Nanopore 16S whole gene amplicon primers, and the products were visualized on an agarose gel. The gel containing PCR product from embryonic guts contained no band at the ~1500bp region compared to a positive control of microbial DNA (Fig. 1).



**Fig. 1** Gel of the 16S PCR products of embryonic gut DNA. The first two lanes are ladders, the third lane is a positive control from a five-week-old hatchling, the fourth lane is the negative control showing only primer-dimer. Lanes 5-12 are embryonic samples showing no 1500 bp 16S PCR product.

Results from the PCR amplification of gut DNA suggested that bacteria were not present; thus, those samples were not sequenced. Additional validation was ob

<sup>1</sup>Corresponding author: tss0019@auburn.edu



tained through qPCR which showed that the amount of bacterial DNA in the embryonic gut samples were 1/159th the amount of DNA from a positive control sample and had an average Qc of 38 in a 40-cycle run that was not distinguishable from the negative controls.

DNA extracted from the gut for both dietary treatments (n=10/treatment) along with four adults, was processed through the minION 16S gene sequencing protocol. After sequencing the 24 samples, over 1.4 million reads were base-called and classified using Oxford Nanopore's minION and Epi2me software. The data will be checked for quality and analyzed for future publication. Given the gene sequencing data we have collected, we are confident that our analysis will provide insight into the influence of dietary variation in gut bacterial community composition during early life.

While we cannot say for certain that there is no bacterial DNA in embryonic guts, the comparison between embryonic samples and controls suggests that if any are present the amount is biologically irrelevant. While poultry papers<sup>5</sup> showed colonization of the gut in ovo even in sterilized conditions, we did not see similar levels of colonization in our samples. Our results contrast research<sup>6</sup> utilizing field-acquired embryonic samples from both birds and lizards, which suggested maternal transmission but those studies utilized whole yolk samples rather than dissected intestinal tracts<sup>6</sup>. The addition of our research may alter the perception of microbial transmission in lizards, whereby the development of gut microbiota may occur following hatching rather than during embryonic stages.

### Statement of Research Advisor

Microbiome research is a rapidly growing field and how the microbiome develops is a key question. Taylor has designed and implemented a study that will be foundational in our understanding of microbiome development in a reptile model system. Upon final analysis, Taylor will be submitting his findings for peer-review publication in a scientific journal.

- Tonia Schwartz, Department of Biological Sciences.  
COSAM

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### Authors Biography



Taylor McKibben is a senior pursuing a B.S. degree in Microbiology. He has played key research roles in experimental design, literature review, and manuscript writing. He will be continuing his education at Auburn University as a masters student in the Department of Biology.



Kaitlyn Murphy is a 5th year Ph.D. Candidate under Drs. Daniel Warner and Mary Mendonça in the Department of Biological Sciences. She is interested in the influence of environmental variation on microbiota associated with reptiles, particularly in maternal transmission among oviparous species. She served as a graduate mentor to Mr. McKibben and will assist with the downstream analyses of bacterial sequences.



Dr. Tonia Schwartz is an Associate Professor in the Department of Biological Sciences. Her Functional Genomics Lab addresses questions about how animals respond to environmental stressors. She served as a mentor to Mr. McKibben as he conducted his research on the development of the microbiome in brown anoles.

# Identifying Manual Material Handling Tasks using wearable sensors with Machine Learning

*Daniel D. Michael<sup>1</sup>, Howard Chen<sup>2</sup>, and Duha Ali<sup>3</sup>*

<sup>1</sup> Undergraduate Student, Department of Mechanical Engineering, Auburn University

<sup>2</sup> Research Assistant Professor, Department of Mechanical Engineering, Auburn University

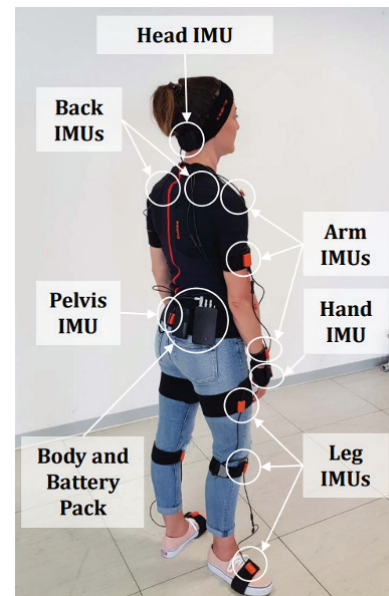
<sup>3</sup> Graduate Student, Department of Industrial and Systems Engineering, Auburn University

In the United States, work-related musculoskeletal disorders (WMSDs) such as lower back pain and carpal tunnel syndrome account for about 30% of all non-fatal workplace injuries resulting in days away from work each year. Assessing the risk associated with various jobs is the first step toward mitigating WMSDs. This is typically accomplished by observing workers in their occupational setting, which is imprecise and consumes much research time. Wearable sensors, specifically Inertial Measurement Units (IMUs), represent a promising new data collection method that can measure worker kinematics. Accurately and automatically identifying work tasks using these machine learning models will play a vital role in achieving accurate, fast, and non-invasive risk assessment.

This research attempted to investigate the use of deep learning to distinguish between one-handed and two-handed manual material handling tasks. An additional goal was to create a deep learning model that was easily trainable and generic enough to fit future ISE research.

Relevant research was found published by Bassani, which informed the initial machine learning models and provided an initial dataset. The data was generated using 17 IMU sensors, 14 subjects, and 37 trials consisting of 7 task types. The IMU sensor placement is shown in Figure 1. The undergraduate research developed classification convolutional neural network models using TensorFlow Keras API for machine learning. An example of model architecture is shown in Figure 2. The models consistently produced an accuracy greater than 95% on the provided dataset, as shown in Figure 3. Because the (training) accuracy did not increase beyond the validation accuracy, the model was shown to not overfit the training data.

<sup>1</sup>Corresponding author: ddm0016@auburn.edu

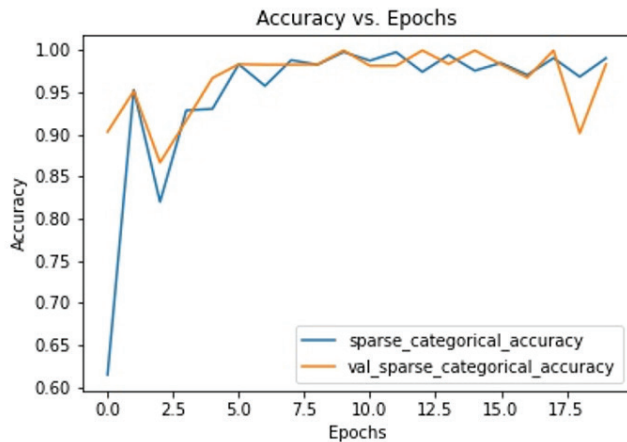


**Fig. 1** Sensors were placed at 17 locations in Bassani's dataset.

Layer (type)	Output Shape
conv1d_36 (Conv1D)	(None, 1998, 64)
conv1d_37 (Conv1D)	(None, 1996, 64)
dropout_10 (Dropout)	(None, 1996, 64)
max_pooling1d_17 (MaxPooling)	(None, 998, 64)
flatten_17 (Flatten)	(None, 63872)
dense_34 (Dense)	(None, 100)
dense_35 (Dense)	(None, 7)
Total params: 6,542,311	
Trainable params: 6,542,311	
Non-trainable params: 0	

**Fig. 2** Convolutional neural networks require feature extraction through Conv1D layers and neural node cre





**Fig. 3** Accuracy increases during the training process.

After developing successful data preparation methods and machine learning models, IMU data was collected using the researcher as the subject, as shown in Figure 4. The data was generated using 6 IMU sensors. The subject repeated a trial 30 times which consisted of 7 task types. The trial was performed with both hands, the right hand and the left hand. This data was collected to investigate the use of machine learning and IMU sensors to distinguish between right-handed, left-handed, and two-handed tasks. Models besides CNN were investigated using generic settings for more than 15 classification architectures. This research is still ongoing, but initial results allude to accuracies greater than 75%. The ultimate goal is to perform this classification with only one sensor.



**Fig. 4** IMU data was collected using the researcher as a subject.

## Statement of Research Advisor

One-handed manual material handling is an under-researched field of study that contributes to workplace injuries. Sensor systems exist for measuring the motion patterns of a worker throughout the day. But these systems are lacking in their ability to categorize specific tasks, including one-handed material handling. Mr. Michael developed a machine learning algorithm to classify various manual material handling tasks using wearable sensors using a combination of open source and lab-collected data.

- Dr. Howard Chen, Department of Mechanical Engineering

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## Authors Biography



Daniel D. Michael is a senior-year student pursuing a B.S. degree in Mechanical Engineering at Auburn University. He has completed a co-op at Southern Research Institute in the Space Propulsion and Systems Department. He plans to pursue a Ph.D. in Mechanical Engineering at Duke University in the Fall.



Howard Chen is a Research Assistant Professor at Auburn University in the Mechanical Engineering Department. He completed his Ph.D. at the University of Iowa in Industrial Engineering and has performed key research in sensor-based ergonomics research.





Duha Ali is a graduate student pursuing a Ph.D. in Industrial and Systems Engineering at Auburn University. She received her B.S. degree in Electrical Engineering from Jordan University of Science and Technology. She has performed key research in Occupational Safety and Health.

# Confrontation with the Abject after Experiences of Sexual Interpersonal Violence: An Analysis of Contemporary Horror Films

Regan A. Moss <sup>1</sup> and Allison Vandenberg <sup>2</sup>

<sup>1</sup> Undergraduate Student, Department of Psychological Sciences, Auburn University

<sup>2</sup> Instructor, Women's and Gender Studies, Auburn University

Sexual violence is pervasive, directly impacting 1 in every 59 adults in the United States every year (Basile et al., 2007). This form of violence is even more prevalent amongst young adults: reports highlight that 1 in 4 women and 1 in 6 men experience sexual interpersonal violence during collegiate years (Ruane, 2014). The recognition of this public health concern is sweeping across a variety of media outlets. However, public narration through such outlets has many associated harms. Some argue that the atrocity of sexual violence becomes normalized because of the frequency at which it is reported to the public. Depictions may even give rise to new forms of sexual interpersonal violence (Powell, & Henry, 2017). The variance in sexual violence depiction leads to differential – and often harmful – outcomes on viewer perceptions of rape and the victim: perpetrator. (Burt, 1980; Henry, & Powell, 2018). Importantly, media coverage “perpetuates myths and stereotypes about rape, rapists, and rape victims” (Burt, 1980). Often, such myths build upon gender stereotypes (Nagar, 2016). Horror contrasts other forms of media, subverting the typical to propose an alternate narrative. The messages conveyed through horror films and the methods through which such messages are conveyed have vital public health implications. Results will inform policies surrounding public messaging related to sexual interpersonal violence. Additionally, findings will highlight outlets and methods for productive discourse.

The purpose of this study is to investigate common themes that emerge in depictions of sexual violence in popular horror films airing after 2000. Films were analyzed through three iterations of thematic analysis to answer three research questions: 1) How is the subject separated from the soul (i.e., made an abject)? 2) What is the function of social structures to purify the abject?

3a) How does the subject confront the soul after sexual violence/reconcile with the maternal body, and 3b) how does the reconciliation undermine symbolic gender relations? After analyzing the films through thematic analysis, several themes emerged. Themes of silencing, objectification of the female body, and symbolic and corporeal power emerged in the making of abjects. Institutional purification was done through symbolic power, familial dismissal, and medical pathologizing. Confrontation (i.e., reconciliation) was reached through social revelation, physical separation, reordering of physical power, and sexual constriction of the perpetrator.

In these films, sexual violence was facilitated through symbolic and physical power, such as physical restrictions, financial restrictions, social isolation, and symbolic reinforcements of such. These positions were enforced by further silencing within multiple layers of social and relational realms. These films revealed the dismissal of victim-narrated reports in social and built environments (e.g., family, economic system, law enforcement, direct care providers), leading to the purification of the abject and the perpetuation of abjection-enforcing acts. Institutions purified the process of abjection and the objects of abjection through report dismissal and the silencing of victim narratives, mental health sequelae, and fears. Victims of sexual violence confronted their abuser, perpetrator, and social and relational positions through a variety of individual means. Horror films provoked investigation of the means and platforms through which sexual violence occurred that may otherwise remain unchallenged (e.g., family relations, mental health care providers). In doing so, they provide a constructive perspective on how social responses to sexual violence can be altered in order to

<sup>1</sup>Corresponding author: ram0083@auburn.edu

better serve the victim and survivor.

## Statement of Research Advisor

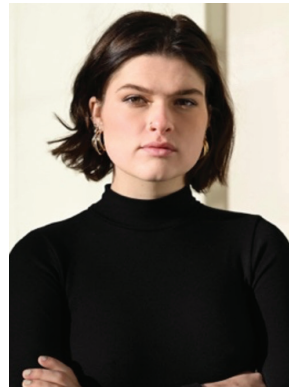
The purpose of the study is to better understand the common depictions of sexually based violence in media and the social norms these depictions reflect in order to aid in the development of regulatory practices that counteract potential harms flowing from such depictions. The project that she and I embarked upon has the potential to build upon both her remarkable academic achievements and her inspirational activist work relating to raising awareness of sexual violence.

- Allison Vandenberg, *College of Liberal Arts*

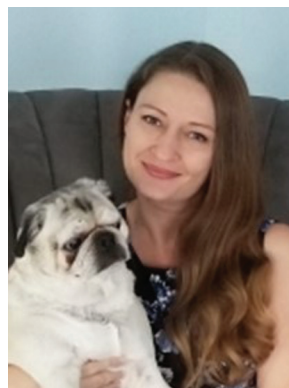
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## Authors Biography



Regan Moss (she/her/hers) graduated from Auburn University with a BS in Neuroscience. As a Women's and Gender Studies Minor, Regan received the Samia I. Spencer Award-- the first undergraduate to have ever done so. Regan is presently a research associate and consultant at Yale University Child Study Center and the Department of Obstetrics, Gynecology and Reproductive Sciences at Yale University Medical School. She is pursuing her Master of Public Health at Columbia University. Her research interests include inter- and trans-generational trauma (e.g., sexual violence and sexual abuse), motherhood, maternal-infant relations, and menstrual & reproductive health.



Allison Vandenberg is an instructor of women's and gender studies in the College of Liberal Arts at Auburn University. She received her Ph.D. in gender studies with a concentration in sexualities, desires, and identities and a minor in history from Indiana University in 2016. Her work focuses on gendered engagement in bodily practices, embodiment, and phenomenology. Previous publications include "Toward a Phenomenological Analysis of Historicized Beauty Practices," which appeared in the Spring/Summer 2018 issue of *Women's Studies Quarterly*, and "The Somatechnics of Hair Straightening: Technology, Transformation, and Social

Change,” which was published in the edited volume (Re)Possessing Beauty: Politics, Poetics, Change. Her regular course offerings include Introduction to Women’s and Gender Studies; Feminist Theory; Gender, Beauty, and Culture; Women in European History; and The Virgin Always Lives: Gender, Sexuality, and Horror.



# Re: SMP – Recycled Shape Memory Polymers

Margaret A. Nelson<sup>1</sup>, Midhan Siwakoti<sup>2</sup>, Rylee Cardon<sup>3</sup>, Eldon Triggs<sup>4</sup>, and Russell W. Mailen<sup>5</sup>

<sup>1</sup> Undergraduate Student, Department of Aerospace Engineering, Auburn University

<sup>2,3</sup> Graduate Student, Samuel Ginn College of Engineering, Auburn University

<sup>4</sup> Lecturer, Samuel Ginn College of Engineering, Auburn University

<sup>5</sup> Associate Professor, Samuel Ginn College of Engineering, Auburn University

Shape memory polymers (SMPs) are active materials with growing potential for use in aerospace applications, including as actuators for deployable structures and roll out solar arrays. SMPs change shape in response to external stimuli, such as heat, and retain their deformed shape after removal of the stimuli. Unfortunately, SMPs tend to be single-use materials due to the nature of the shape change. Therefore, there exists an abundance of unutilized SMP waste, which demands a reevaluation of the material infrastructures currently in place. In-situ resource utilization (ISRU) and recycling promote the use and integration of sustainable material infrastructures on earth and throughout long-duration space missions. Mechanical recycling is the most viable alternative material processing system to establish SMPs as a multi-use material. However, material degradation is inevitable with recycling. Therefore, it is imperative that material properties of recycled SMPs be quantified. In this study, we investigate the effects of recycling on the thermomechanical properties and shape memory performance of SMP sheets. As a result, recycled SMPs are viable throughout six extrusions and support the concept of highly functional, recycled SMPs to promote sustainable aerospace practices and applications.

The sustainably sourced shape memory polymers utilized in this study were discarded polystyrene CD cases. Polystyrene is a low-cost and highly accessible SMP, so it is commonly used in the production of plastic products, including CD cases. Then, we implemented a laboratory-scale mechanical recycling process to recycle the polystyrene CD cases. First, the material was mechanically broken down by hand. The polymer pieces were then fed into a single-screw Filabot EX2 Filament Extruder. The extruder breaks down and melts the material into a continuous profile. This process produced a polymer resin that was then spiraled by hand. The polymer resin spiral was then processed through

compression molding in a Wabash Genesis Hydraulic<sup>1</sup> Press. Then, the material was removed from the plates and inspected for imperfections, namely air bubbles and shearing. After pressing into flat sheets, samples for characterization were cut from the material. After evaluating the recycled material, the sample was returned to the bulk recycled material, which was then mechanically broken down again and the recycling process, as seen in Figure 1, was repeated as necessary. This process was used to produce material samples ranging from zero to six extrusions. Recycled SMPs were characterized in a TA Instruments HR-20 Hybrid Rheometer/Dynamic Mechanical Analysis (DMA). Samples were subjected to temperature sweep tests and shape recovery tests. The shape recovery tests required an initial pre-strain sequence using the thin film fixture in the DMA. Then, samples were subjected to constrained recovery or free recovery tests described below.

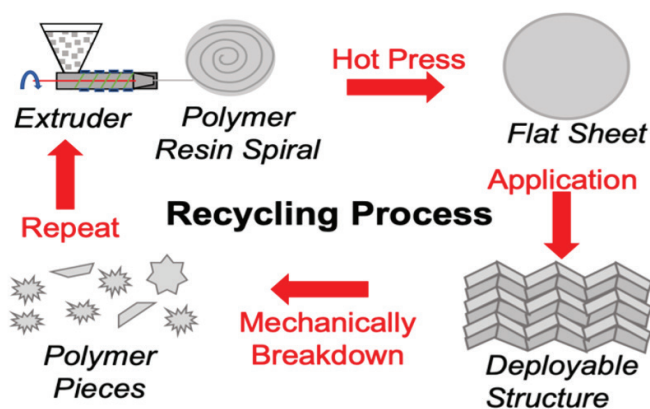
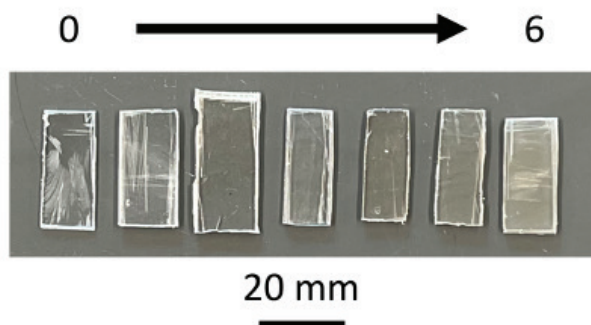


Fig. 1. Polymer Recycling Process applied to SMP.

Samples ranging from zero to six extrusions were produced using the in-house, mechanical recycling process and compared for signs of physical degradation. Figure 2 shows changes to the physical appearance of the samples. Degradation resulting from recycling is increasingly obvious when working with a transparent and colorless material like polystyrene<sup>[1, 2]</sup>. In fact, phys

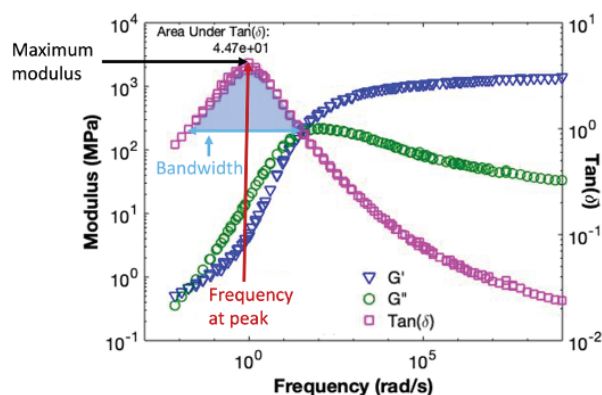
<sup>1</sup>Corresponding author: man0045@auburn.edu

ical degradation was the most obvious sign of degradation for the up to six times recycled SMPs. With each successive extrusion, the material became increasingly cloudy and yellow. These changes in the appearance of the material signify a breakdown at the atomic level.



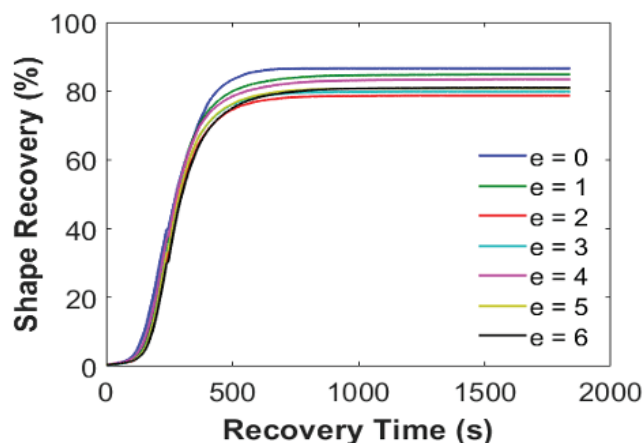
**Fig. 2.** Untested DMA Samples of Recycled Polystyrene.

Viscoelastic master curves obtained from DMA results were compared for samples ranging from zero to six extrusions; representative results are shown in Figure 3. This figure has been annotated to show a variety of parameters which will be compared for each dataset. In the results, the storage modulus, loss modulus, and phase angle agree with expected trends<sup>[3, 4]</sup>. When analyzing the viscoelastic master curves, changes in the  $\tan(\delta)$  curve were observed specifically as this property is a ratio of the loss and storage moduli. Therefore, changes to both properties are accounted for in this singular analysis. These results indicate that there is no clear trend to the way the recycled SMPs are degrading. Therefore, degradation is minimal and does not prevent recycled SMPs from functioning properly in aerospace applications.

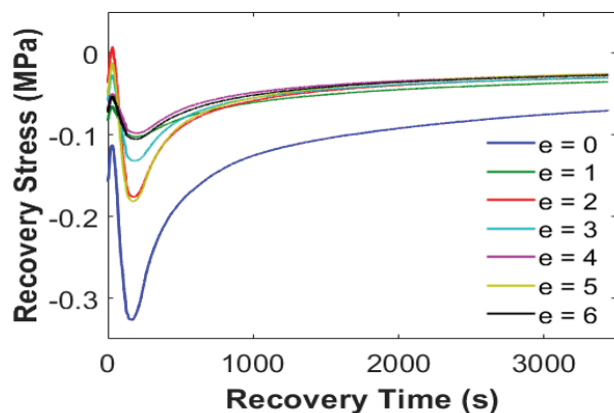


**Fig. 3.** Representative Visoelastic Master Curve for Once Extruded Polystyrene.

Shape recovery tests were developed to determine the extent to which the material retains its shape memory properties after recycling. Herein, we considered free recovery and constrained recovery tests, the extremes of possible use cases. Free recovery tests use zero load conditions after pre-straining to provide a measure of the maximum shape recovery<sup>[5]</sup>. Through the developed testing procedure, the SMP samples were subjected to a pre-straining conditioning process so that shape recovery behavior could occur. The material was then allowed to recover fully within the tension test fixture of the running test. The representative results, shown in Figure 4, illustrate the gap during shape recovery when subjected to zero axial force. By observing the changes that occurred from the free recovery results for zero to six extrusions, it is evident that recycling had a negligible effect on the free recovery behavior. Therefore, the material can still perform predictably and efficiently as a SMP. Constrained recovery tests restrict the shape memory property of the material by holding the gap of the fixture constant. This information reveals the effectiveness of the shape recovery of the material when subjected to a constant gap<sup>[6]</sup>. Observing the results of these DMA tests, as represented in Figure 5, the most significant change in recovery stress occurs after the first recycling but there is minimal variation for subsequent recycling sequences. The minimal changes to the axial force occur in no particular pattern. Therefore, it can be assumed that the ability of the material to recover is minimally degraded by recycling as revealed through constrained recovery testing.



**Fig. 4.** Free Recovery of Zero to Six Times Extruded Polystyrene.



**Fig. 5.** Constrained Recovery of Zero to Six Times Extruded Polystyrene.

In this paper, we investigated the effects of recycling on the thermomechanical properties and shape memory performance of SMP sheets. We implemented a desktop recycling process to extrude recycled polystyrene and produce uniform sheets. Degradation as a result of mechanical recycling was assessed through DMA. It was demonstrated that recycled SMPs are viable for aerospace applications for up to six extrusions as resulting degradation is minimal, occurring most obviously in the physical appearance. Due to the minimal changes in material properties, it is concluded that degradation of recycled SMPs does not significantly impact material performance. Therefore, recycled SMPs are viable for long-duration space missions throughout the six tested extrusions.

### Statement of Research Advisor

Maggie started working with my research group Fall 2020, and she immediately demonstrated an incredible potential to conduct transformative research. Her passion for sustainable engineering practices led her to quickly immerse herself in a project related to recycling of polymers. She is a quick learner and a hard worker and I am so proud to see her progress.

- Russell W. Mailen, Samuel Ginn College of Engineering

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### Authors Biography



Maggie A. Nelson is a junior-year student pursuing a B.S. degree in Aerospace Engineering at Auburn University. She has played key research roles in the development of an effective and sustainable material infrastructure for polymers in aerospace. Nelson has been named the 2021 and 2022 Astronaut Scholar for Auburn University due to her involvement and dedication to developing research endeavors. She plans to continue conducting research in both aerospace and



materials engineering departments.



Midhan Siwakoti is a graduate student pursuing a Ph.D. in Aerospace Engineering at Auburn University. He has fabricated several procedures for the study of thermomechanical behavior of polymers.



Russell W. Mailen is an assistant professor in the Department of Aerospace Engineering at Auburn University. He has earned a B.S. in Aerospace Engineering (Kansas, 2007), a M.S. in Mechanical Engineering (Baylor, 2012), a Master of Materials Science and Engineering (North Carolina State, 2016), and a Ph.D. in Mechanical Engineering (North Carolina State, 2017). He worked in the aerospace industry as a liaison engineer and structural analyst at Hawker Beechcraft Corporation and L-3 Communications. His research interests include multiphysics finite element analysis, smart materials and structures, mechanical metamaterials, polymer composites, and the thermo-mechanical behavior of polymers.



Rylee Cardon is a graduated student from the Samuel Ginn College of Engineering with an M.S. degree in Aerospace Engineering. She conducted extensive research on the recycling and processing of polymers.



Eldon Triggs is a Lecturer in the Samuel Ginn College of Engineering at Auburn University. He is interested in the manufacturing and use of polymers and competition rocketry.



# The Influence of Urbanization on Stream Quality and Fish Health

Kate R. Norrid<sup>1</sup>, B. Graeme Lockaby<sup>2</sup>, and Dennis R. DeVries<sup>3</sup>

<sup>1</sup> Undergraduate Student, College of Forestry, Wildlife, and Environment, Auburn University

<sup>2</sup> Professor, College of Forestry, Wildlife, and Environment, Auburn University

<sup>3</sup> Professor, School of Fisheries, Aquaculture, and Aquatic Sciences, Auburn University

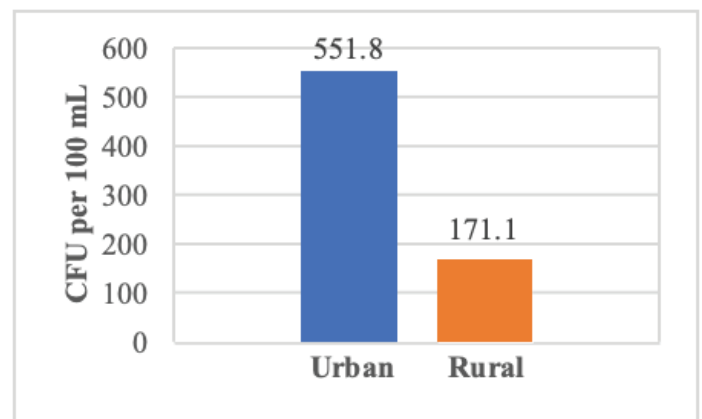
As urbanization increases, stream quality is affected due to the manipulation of waterways to promote fast-draining channels that prevent flooding. As this occurs, stream hydrology is negatively impacted, and it causes unnatural changes in flow frequency and magnitude (Walsh et al. 2005). In addition, human waste runoff into streams pollutes the water, leading to higher levels of pollutants such as *E. coli* and suspended solids. The negative relationship between water quality and the degree of urbanization has been well documented; however, there is relatively little information on how urbanization-related water degradation affects the health and overall physiology of resident aquatic biota.

A small number of studies have been conducted that correlate stream quality and overall fish health (Morse 2005, Menvielle 2006, Helms et al. 2009, Sayer 2012, Nagrodski et al. 2013, Wedge et al. 2015, Iwanowicz et al. 2016), but there remains little information on this critical topic for the lower Piedmont physiographic region of the southeastern United States, a region that is rapidly urbanizing (Wear 2013, Curl and Bell 2021). Our research represents an initial attempt to quantify the specific effects of urbanization-related water degradation on overall fish health within our focal region.

Six total streams, 3 urban and 3 rural, were selected in and around Auburn and Opelika, Alabama. Streams were either 2nd or 3rd order and were associated with watersheds that ranged in size from 130 to 5700 hectares. Indices of fish health and water quality were quantified at each site. Measured water quality metrics included dissolved oxygen, nitrate, phosphate, total suspended solids, and *E. coli*. To quantify fish condition, we collected sunfish (*Lepomis* spp.) from each site using seine nets. These fish were selected for our study due to their abundance in the region and widespread cosmopolitan distribution. Physiological health indices collected

from the fish included length-weight ratios (calculated<sup>1</sup> from fish length and weight), hepatosomatic index (calculated from liver weight and fish weight), and gonadosomatic indices (calculated from gonad weight and fish weight). In addition, we extracted otoliths from each fish to determine age, allowing us to quantify population age structures in rural and urban stream systems.

Preliminary results have shown stark differences in *E. coli* levels between land use types (Figure 1). Water and fish data compilation is still ongoing, with statistical analysis in progress. We hypothesize that fish from urban degraded streams will show lower fish condition measures relative to those from forested reference streams and that the urban systems will have altered population dynamics (i.e., younger-biased age structure); however, the extent of these differences is yet to be determined. Once finalized, our results will be of significance to the field of stream conservation because of the evidence linking urbanization, poor water quality, and negative health implications for native aquatic organisms. In addition, they will help fill the current knowledge gap on this subject in the region and give local insight.



**Fig. 1** Means of *E. coli* levels (colony forming units per

<sup>1</sup>Corresponding author: krn0009@auburn.edu

100 mL) by land use type.

## Statement of Research Advisor

This study, including the hypotheses and general approaches, represents original ideas of Kate Norrid that she developed independently after reviewing pertinent literature. She clearly identified a key gap in the scientific literature pertaining to urbanization effects of surface water and has been responsible for implementing each stage of the study with advice from myself and Dr. DeVries. Kate's insight and creativity on this project show an unusually high level of research aptitude.

- Graeme Lockaby, College of Forestry, Wildlife, and Environment

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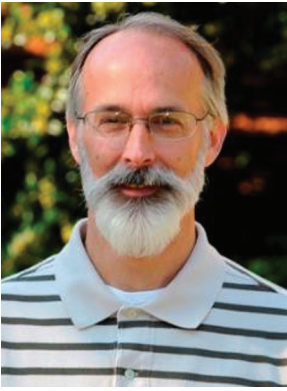
## Authors Biography



Kate R. Norrid is a senior-year student pursuing a B.S. degree in Wildlife Ecology and Management at Auburn University. She is interested in studying health from a holistic environmental perspective and plans to work in stream restoration and mitigation following graduation.



Dr. Graeme Lockaby is a professor in the College of Forestry, Wildlife, and Environment at Auburn University. He earned his B.S. and M.S. in Forestry at Clemson University and his Ph.D. in Agronomy-Soils at Mississippi State University. Lockaby's program focuses mainly on the biogeochemistry of forested floodplains and relationships between wetlands and human health.



Dr. Dennis DeVries is a professor and assistant director for research programs in the School of Fisheries, Aquaculture, and Aquatic Sciences at Auburn University. DeVries's program broadly focuses on the interface between basic fields of ecology and applied fields of fisheries and natural resource management.

# Audiological Screening Protocol Using Novel Tablet Application: A Validation Study

*Anna O'Donell<sup>1</sup>, Ashlyn Wheat<sup>2</sup>, Cheryl Seals<sup>3</sup>, and Aurora Weaver<sup>4</sup>*

<sup>1</sup> Undergraduate Student, Department of Speech, Language, and Hearing Sciences, Auburn University

<sup>2</sup> Graduate Student, Department of Speech, Language, and Hearing Sciences, Auburn University

<sup>3</sup> Associate Professor, Department of Computer Science and Software Engineering, Auburn University

<sup>4</sup> Associate Professor, Department of Speech, Language, and Hearing Sciences, Auburn University

Close to 15% of children in the U.S. exhibit some level/type of hearing loss of at least 16 dB HL by the time they enter kindergarten (Niskar et al., 1998). Research supports that access to regular hearing screening programs is not readily available across all populations (Brennan-Jones et al., 2015), which may exacerbate the negative impact of hearing loss on an individual's quality of life – particularly regarding proper academic development and success of school-aged children. This thought process led to the development of the Auburn Graded Early Skills application (AGESapp), which is a user-friendly application that screens hearing in a fast and efficient manner for patients in a variety of settings (i.e., schools, clinics, rural areas). The purpose of this research study is to determine the validity and reliability between the gold-standard four-frequency pure tone hearing screener with American Speech-Language-Hearing Association's (ASHA) guidelines (ASHA, 2005) (e.g., portable audiometer), the AGESapp hearing screener, and selected auditory processing skills test battery which assesses individual auditory skills and cognition.

Data was collected by using an engaging “train station” activity to screen peripheral hearing in children and adults. The AGESapp was designed to mimic the clinical approach to hearing screening, and measured hearing at the following frequencies: 500 Hz, 1000 Hz, 2000 Hz, and 4000 Hz. Each stimuli uses novel frequency mixture of pure-tone (sinusoidal) and noise stimuli in order to stimulate each center frequency region of the cochlea. Stimuli in the AGESapp begins at 50 dB HL, and then decreases after conditioning to a screening level of 20 dB HL for the stimuli with a center frequency at 1000 Hz. Overall, the app randomly screens the remaining test frequencies for each ear at 20-25 dB HL. A mixture of 89 participants (34 adults and 55 children)

between the ages of 3-65 years from Auburn University<sup>1</sup> and surrounding community daycares and pre-schools were tested using both the gold standard and AGESapp.

Out of the 89 participants screened, 5 children were re-screened a second time due to instrumental issues and/or participant motivation. The app proves to be ecologically valid for children 6 years of age and younger with a certified professional. Adults and school-age children also prove to be ecologically valid to self-administer the application. We ran the stats in SPSS software to compare the sensitivity and specificity of the AGESapp versus the gold standard portable hearing screener. Overall, results indicate that the AGESapp screening was sensitive to referrals on a hearing screening in 67-80% in adults and 43% in children. The AGESapp screening was 100% specific for passing participants who passed the gold standard hearing screening.

This study reveals that the AGESapp is an effective tool to screen hearing to increase efficiency of community and school-based hearing screenings. The work will inform iterations of the application, to increase engagement and ease of use for a wide range of ages (3-65 years old). Further developments of the application are underway, which will test additional hearing, speech, and language skills. AGESapp successfully gives audiological screenings to potential patients in an inexpensive and easy access way. Thus, successfully fulfilling our initial goal.

## Statement of Research Advisor

Anna Grace O'Donell has worked as an undergraduate research assistant in the AMP lab (2020-2021), and as an undergraduate research fellow (2021-2022). She has contributed to the design and development of a nov

<sup>1</sup>Corresponding author: ago0006@auburn.edu



el App in my lab and has been key in progressing the project goals forward. She shows promise for a clinical research career, and drive, as her career aspirations will incorporate elements of grant writing and community outreach in the field of audiology. She contributed to the data analysis for the present study and the interpretation of the study results for publication. Anna Grace has presented these results at the Student Research Symposium at Auburn University, as well as a state conference (Speech Hearing Association of Alabama's 2022 convention).

-Aurora J. Weaver, *Speech, Language, and Hearing Sciences*

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## Authors Biography



A.G. O'Donnell is a senior-year student pursuing a B.S. degree in Speech, Language, and Hearing Sciences at Auburn University. As the 2021-2022 URF she assisted in design, development of the AGES app, research design, data collection, management, analysis, preparation, as well as dissemination of the finding at a state professional conference. She has ac-

cepted a position in University of Louisville's clinical doctorate of audiology 2022 incoming class, where she will continue to pursue a career in Audiology and looks forward to opportunities to use her user her knowledge and skills in clinical research during her graduate studies.



A. Wheat is a first-year doctoral student pursuing a clinical doctorate of Audiology at Auburn University. She obtained her undergraduate degree in Speech, Language, and Hearing Sciences from Auburn and works as a graduate research and teaching assistant for Dr. Aurora J. Weaver. Her contributions to the Audiological Screening Protocol study consists of facilitating data collection, project management and manuscript preparation.



Dr. Cheryl Seals is an associate professor in Auburn University's Department of Computer Science and Software Engineering. Her research areas of expertise are human computer interaction, user interface design, usability evaluation and educational gaming technologies. Seals also works with outreach initiatives to improve computer science education at all levels. The programs are focused on increasing the computing pipeline by getting students interested in STEM disciplines and future technology careers.



Dr. Weaver is an Associate Professor for the Department of Speech, Language Hearing Sciences. She received her Bachelor's degree in Hearing, Speech and Language Sciences, her clinical doctorate in Audiology (Au.D.), and her Ph.D. in Hearing Science from Ohio University, in Athens Ohio. She is the Lead Researcher in the Auditory and Music Perception Lab where she studies the following areas: Developmental Psychoacoustics, Central Auditory Processes & Diagnostic Audiology. Her responsibilities include oversight of all aspects of the fellowship project.

# Identifying Species of Bacteria that Assist in Survival in Environments with an Extremely High Salt Content or High Temperature

Walker R. Olive<sup>1</sup> and Kathy Lawrence<sup>2</sup>

<sup>1</sup> Undergraduate Student, Department of Entomology and Plant Pathology, Auburn University

<sup>2</sup> Professor of Entomology and Plant Pathology, Auburn University

Plant-growth promoting rhizobacteria assist plants in mitigating stress from not only pathogens but heat and salt stress<sup>1,3</sup>, which is becoming more present in Earth's changing climate. High salinity levels directly impact biochemical reactions, limit photosynthesis capabilities, decrease transportable iron, as well as decreasing turgor<sup>2</sup> (the level of rigidity in the cells). High temperatures, leading to inadequate water supply for plants, can limit nutrient bioavailability in the soil.<sup>1</sup> To tackle the above concerns, identification of certain bacterial specimens from the selected environments have been collected and are currently being examined for evidence of their role in plant growth promotion. In our research, examining the resilience of selected bacteria on plated media as well as in the greenhouse can provide the information needed to possibly pursue new species identification of those same bacteria.

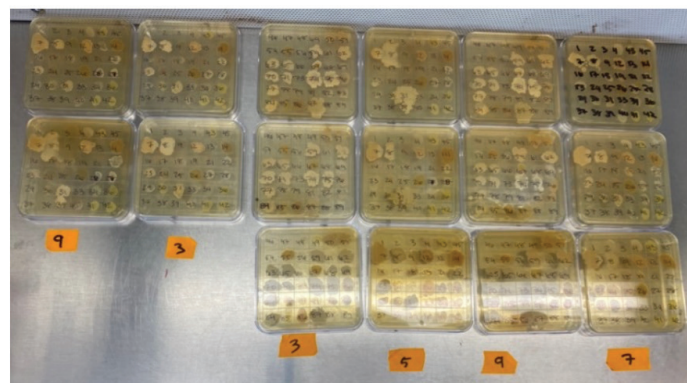
Samples for the salt water-based portion of the experiment were obtained in or near Mobile, AL, and the drought/heat tolerant varieties were found in New Mexico. Root sections around 1.0 cm in length are extracted using a sterile scalpel and stored in test tubes filled with sterile water. These sections would be vortexed and serially diluted, and plated onto tryptic soy agar. More dilutions would be made after the root tissue was macerated with a tissue grinder.

The resulting plates were incubated for 5 days at 28°C while colonies were selected based on morphology determined by a college at days 2 and 5. For storage, the samples were prepared with a tryptic soy broth amended with 30% glycerol and placed in cryopreservation.

Polymerase chain reaction (PCR) was run on all samples before plating to ensure bacterial presence and checked

with gel electrophoresis. The potentially salt-tolerant<sup>1</sup> bacteria were plated on a gradient of plates ranging from 2-10% NaCl in tryptic soy agar.<sup>3</sup>

Specimens from the drought-prone areas are currently being plated on a mixture of tryptic soy agar and concentrations of 3g, 5g, 7g, or 9g per liter of polyethylene glycol, testing osmotic resistance.<sup>5</sup>



**Fig. 1.** The image above displays an example of incubated plates with varying concentrations of polyethylene glycol in the media.

Both the salt and drought-tolerant specimens were allowed to incubate at 30 °C for at least one day. Bacteria that grew through every gradient of salt concentration were sent to the MCLab for sequencing. The drought-tolerant varieties are currently undergoing greenhouse trials. The root systems of corn plants have been inoculated with a bacterial solution of a set turbidity standard in yellow 150cm<sup>3</sup> containers filled with pure sand. Inoculated plants will not be watered for 7 days, as watering will wash away the new bacteria and not create proper drought conditions.

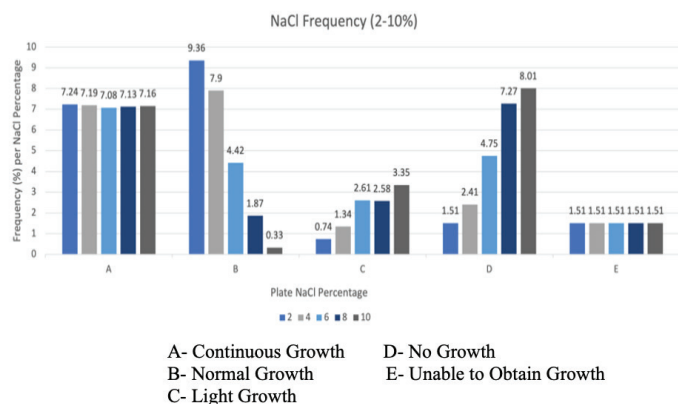
<sup>1</sup>Corresponding author: wro0003@auburn.edu





**Fig. 2 & Fig. 3.** Image 1 (left) shows plants that were inoculated with the bacterial solution after receiving no water for 7 days. The second image (right) shows five of the same plant species that did not receive any bacteria before not receiving water for 7 days. The figure on the right shows obvious wilt and stunted growth from water loss.

PCR was successful on most samples and helped to determine samples with sufficient DNA content for further examination, with only a few not producing bands in gel electrophoresis, even after DNA extraction. The salt-tolerant varieties that grew on every gradient and shipped off to MCLab for sequencing were analyzed using BioEdit, with the top 40 most promising strains currently starting assessment for new species identification. (See Figure 4 for more information) Plates with drought-tolerant varieties are finishing the gradient plating process with signs of resilient strains in the collection. The last batch of samples from the drought-tolerant specimens is being run through the greenhouse at this time. Previous tests show a distinct difference between the corn plants with the bacteria and without after 7 days of no watering. (See Figures 1 and 2) Due to this being an ongoing research project, the results are not yet complete.



**Fig. 4** Above displays the percent occurrence of each category of growth (A-E) per NaCl % concentration.

Differing concentrations of NaCl helped to identify the strongest strains collected.

Results from the greenhouse trials, although not complete, demonstrate that the collected species of bacteria serve as a plant growth-promoting agent when it comes to drought conditions. This information, however, does not try to claim that only a specific species or plant works with this growth-promoting relationship. Sequencing the most resilient strains from the salt-tolerant batch of bacteria shows enough genetic variation to investigate as a potentially new species. Continued plating on a TSA/polyethylene glycol gradient shows potential for bacteria that resists osmotic stress.

### Statement of Research Advisor

Beginning in the Fall of 2020, Walker has provided contributions to our studies of bacteria from high-salt and from arid environments. Her role, in addition to the upkeep and maintenance of bacterial cultures, was to conduct the Polymerase Chain Reaction (PCR) protocol on 500+ bacterial strains so that they could subsequently be identified by 16S rRNA gene sequencing. Before she sent her gene-products for sequencing, she had to confirm her results by gel electrophoresis.

-Dr. Kathy Lawrence, Entomology and Plant Pathology

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## Authors Biography



Walker R. Olive is a senior-year student pursuing a B.S. in Applied Biotechnology at Auburn University. She has contributed a year and a half to the project and has a passion for research and work with bacteria.



Dr. Kathy Lawrence is a professor and researcher in the department of Entomology and Plant Pathology. Areas of expertise are in soil borne and foliar fungal diseases, specifically plant parasitic nematodes and fungi attacking field crops, vegetables, and ornamentals, with emphasis on nematode and fungal pathogen interactions and host-pathogen relationships in the environment.

# Development of PID Controller for Controlling the Phase Difference Between Coaxial Rotors

*Andrew H. Parker<sup>1</sup> and Vrishank Raghav<sup>2</sup>*

<sup>1</sup> Undergraduate Student, Department of Mechanical Engineering, Auburn University

<sup>2</sup> Assistant Professor, Department of Aerospace Engineering, Auburn University

Coaxial rotors have two rotors on top of each other that rotate in opposite directions, which provides a distinct advantage over a conventional single-rotor helicopter. It offers better stability and maneuverability since it removes the tail rotor on a helicopter. However, the two rotors in the coaxial rotor result in high aerodynamic interactions, which are not well studied across different conditions.

I have been working in the Applied Fluids Research Group (AFRG), which has the objective of studying and understanding the aerodynamics of coaxial rotors across different conditions. The lab has a medium to small-scale coaxial rotor setup for studying their aerodynamics and performances at various Reynolds number conditions. The medium-scale and small-scale coaxial rotor setup are similar to the standard coaxial rotor design with a medium-scale rotor on top and a small-scale rotor on the bottom. During my time at AFRG, I have learned that the lab uses an experimental technique known as Particle Image Velocimetry (PIV) to study the aerodynamics of the coaxial rotor system. The technique involves the use of lasers, smoke particles, and high-speed cameras to find the velocity field at a specific 2D plane.

Throughout the course of their experiments, the lab has identified that the phase difference between the two rotors is an important issue that affects the flow field below the rotor. The phase difference can be represented by the angle between the blade of the upper rotor to the bottom rotor. The current setup has two rotors that are mechanically uncoupled, such that the individual performance of each rotor can be studied. Therefore, electronic control over the phase difference between the rotors is required. The rotors are actuated using a brushless DC motor (BLDC motor) which can function efficiently at high speeds but is not equipped with positional feedback for controlling the phase difference.

I began my research with a small-scale single-rotor BLDC motor and an IR sensor. My initial objective was to create a control system using a feedback loop with these two devices to be able to control the RPM of the motor using a user-defined setpoint. The objective of this task was to gain a basic understanding of control theory and the program LabView and eventually use this knowledge to control the phase difference between two motors.

The next objective was to gain a mathematical representation of the physical system. I created a simulation within Simulink that used the data from the physical system to accomplish this. The speed of the BLDC motor is determined by a value between 0 and 1, called the duty cycle. First, I determined the relationship between the inputted duty cycle and the outputted RPM of the motor. I then determined the time it took for the motor to reach the expected RPM at different duty cycles. Using the data from these tests, Simulink generated a second-order function that represented the system.

However, numerous objectives remained unobtained. The phase difference between the two test case rotors still needs to be conducted, which will be done so by using a high-precision encoder. This will allow positional feedback from the BLDC motor actuating the rotors. Once a robust coaxial test system has been constructed on this test setup, the current technique will be employed in the medium and small-scale coaxial rotor setups in the AFRG lab.

## Statement of Research Advisor

With the increasing rate of adoption of drones in civilian and military applications, it is very important to understand the aerodynamic interactional forces between the two rotors of a coaxial rotor. This study is a starting point to identify the physical interactional mechanisms of the aerodynamic loading on coaxial rotors of small-

<sup>1</sup>Corresponding author: ahp0019@auburn.edu

scale rotors. Synchronizing these rotors is very important since that will enable an improved understanding of aerodynamic loads. In the future, the plans are to adopt synchronization techniques to study aerodynamic interactions.

– *Vrishank Raghav, Aerospace Engineering*

## Authors Biography



Andrew H. Parker is a senior-year student pursuing a B.S. degree in Mechanical Engineering at Auburn University. He conducted research on control systems and implemented what he learned into the physical system through computer coding.



Vrishank Raghav is an Assistant Professor in the Department of Aerospace Engineering at Auburn University. He is the principal investigator of the Applied Fluids Research Group, where his research interests are centered on the theme of unsteady fluid dynamics with applications across multiple disciplines.

# Understanding Natural Product Biosynthesis in Blueberry

Emma M. Peacock<sup>1</sup>, Lovely Mae F. Lawas<sup>2</sup>, and Courtney P. Leisner<sup>3</sup>

<sup>1</sup> Undergraduate student, Department of Biological Sciences, Auburn University

<sup>2</sup> Postdoctoral Fellow, Department of Biological Sciences, Auburn University

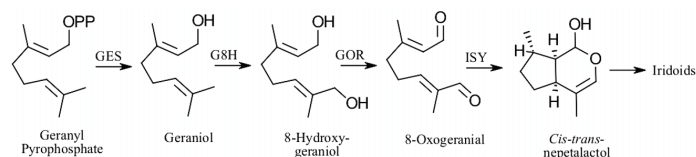
<sup>3</sup> Assistant Professor, Department of Biological Sciences, Auburn University

Blueberries (*Vaccinium* spp.) are widely known for their flavor and human health benefits due to the production of specialized metabolites with potential positive health aspects. Iridoids are a class of specialized metabolites in plants that have known human health benefits, including anti-inflammatory and anti-cancer properties (Dinda et al., 2011). Recent work has found that blueberries make iridoid compounds, with a subset of wild and cultivated blueberry species making the specific iridoid glycoside monotropein (Leisner et al., 2017). While the biosynthetic pathway of iridoid compounds is well studied in other plants, including the model medicinal plant *Catharanthus roseus* (Kellner et al., 2015), the genes involved in monotropein biosynthesis in blueberry are not known. Additionally, the human health benefits of monotropein in blueberry are not well studied. Elucidating genes in the monotropein biosynthetic pathway would not only increase our understanding of this important metabolic pathway but create avenues for future research on understanding the human health impacts of iridoid production in blueberry. For this work, the key enzyme geraniol synthase (GES) was analyzed. GES has been identified as a key enzyme associated with iridoid biosynthesis (Mint Evolutionary Genomics Consortium, 2018), catalyzing the conversion of geranyl pyrophosphate (GPP) to geraniol (Fig. 1) in the plastid during one of the earliest steps of the iridoid biosynthetic pathway. The purpose of this study was to isolate and sequence this key gene in the iridoid biosynthetic pathway and to perform functional enzyme characterization to understand and validate its specific role in iridoid production in blueberry.

To isolate GES, RNA was extracted from various tissues of the blueberry cultivar Ornablu which has been shown to produce the iridoid glycoside monotropein (Leisner et al., 2017). The extracted RNA was treated with DNase and reverse transcribed into cDNA which

was used as a template for polymerase chain reaction using gene-specific detection primers. The GES gene, which was amplified in floral tissues, was purified and sent for sequencing. The resulting sequence of the amplified GES was aligned with the computationally derived sequence of GES used to design the detection primers. When aligned, the amplified GES with high-quality sequencing peaks covered 33% of the computationally derived sequence but with 95% identity, indicating its similarity to the computationally derived gene. A GES gene construct was then designed, commercially synthesized, and cloned into *E. coli* BL21 (DE3) competent cells for heterologous expression.

This research is ongoing, and current work is focused on expressing the GES gene in *E. coli* cells and optimizing the protein purification step to increase the yield of the isolated GES protein. Once purified, the protein along with its substrate GPP will be used in enzyme assays and analyzed by gas chromatography-mass spectrometry to functionally characterize the GES enzyme in blueberry. Overall, results from this study will provide new insights into the function of a key gene within the iridoid biosynthetic pathway, which will allow for future work into the function of these iridoid compounds in blueberry, and lead to a better understanding of the human health benefits of blueberries.



**Figure 1.** Early iridoid biosynthetic pathway. GES, geraniol synthase; G8H, geraniol 8-hydroxylase; GOR, 8-hydroxygeraniol oxidoreductase; ISY, iridoid synthase (Adapted from Lichman et al., 2020).



## Statement of Research Advisor

This project is part of a larger effort to identify all biosynthetic pathway genes for monotropein biosynthesis in blueberry. GES has been shown to be a key enzyme associated with plants that make iridoid compounds across a wide phylogeny of the plant kingdom. Therefore, the work done in this project provides critical information as to why some cultivars of blueberry do and do not make iridoid compounds. This paves the way to understand more about the genetic regulation of this pathway, which makes this type of natural product research more amendable to future clinical evaluation.

-Courtney P. Leisner, *Biological Sciences*

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## Authors Biography



Emma M. Peacock received a B.S. degree in Microbial Biology (Cell/Molecular) with a minor in Mathematics at Auburn University in the spring of 2022. Starting in the fall of 2022, she will be a graduate student within the Interdisciplinary Graduate Program at Vanderbilt University. During her time at Auburn, her research was focused on identifying key genes involved in the iridoid biosynthetic pathway in blueberry.



Lovely Mae F. Lawas is a Post-doctoral Fellow in the Department of Biological Sciences at Auburn University. She received her B.S. degree in Biology and M.S. degree in Molecular Biology and Biotechnology from the University of the Philippines Los Baños. She worked as a Researcher at the International Rice Research Institute prior to obtaining her doctoral (Dr. rer. nat.) degree in Molecular Plant Physiology from the University of Potsdam and the Max Planck Institute of Molecular Plant Physiology. Her research interest is centered on plant metabolism and adaptation of crops to abiotic stresses, with her current research focusing on plant specialized metabolites.



Dr. Courtney P. Leisner is an Assistant Professor in the Department of Biological Sciences at Auburn University. She received her B.S. from The College of William and Mary, M.Sc. in Botany from Washington State University and her Ph.D. in Plant Biology from the University of Illinois Urbana-Champaign. Following graduate school Dr. Leisner was a Ruth L. Kirschstein National Research Service Award Postdoctoral Fellow funded through the National Institute of Health. Dr. Leisner's research group focuses on climate change impacts on plant development and metabolism, with the goal to engineer a more sustainable future food supply.

# United States Zero Hunger Pathways Project: Profile of Food Insecurity in America

Cat Powers<sup>1</sup>, Claire Davies<sup>2</sup>, Amanda Scott Rogers<sup>3</sup>

<sup>1</sup> Undergraduate Student, College of Human Sciences, Auburn University

<sup>2</sup> Undergraduate Student, College of Human Sciences, Auburn University

<sup>3</sup> Undergraduate Student, College of Human Sciences, Auburn University

*The United States Zero Hunger Pathways Project: Profile of Food Insecurity in America* details a situational analysis on the state of hunger in the United States. The Zero Hunger Pathways Project was created by the United Nations Sustainable Development Solutions Network USA to unite research institutions and work towards the United Nations Sustainable Development Goal 2: Zero Hunger. This goal seeks to eradicate hunger by 2030, though food insecurity has exacerbated as a result of the COVID-19 pandemic.

The purpose of the situational analysis is to apply a systematic approach to ending hunger, develop a baseline report that describes the current state of hunger, and create the foundation for an actionable plan to end hunger in the United States. The working group that led the project includes Alicia Powers, Bassel Daher, Asma Lateef, and Caroline Fox. The working group sought to incorporate four core values that reflect the future of the United States food system: sustainability, health, equity, and resiliency. The four values directed the organization of the situational analysis.

The situational analysis is a baseline report describing the current state of hunger in the United States. Articles gathered for analysis were chosen based on three criteriums: applicability to the topic, credibility of the source, and relativity to the current state of hunger. The chosen articles were synthesized to demonstrate the interconnectedness of the multidimensional aspects of hunger. The multidimensional aspects under review include poverty, race/ethnicity, education, unemployment, homelessness, and COVID-19. The draft was then presented in a Dialogue Series that sought feedback from experts in varied fields.

The situational analysis demonstrated that food insecurity disproportionately affects specific populations

within the United States, particularly Black and Latino Americans. The review also demonstrated the extensive cost of food insecurity. Though the monetary cost can be quantified to \$29 trillion USD, the true cost of hunger is much deeper and is expressed through impacts such as attention deficits, loss in economic productivity, and mental health conditions (Hendricks, et al., 2021). The feedback from Dialogue Series participants included clearly differentiating COVID-19's impact on food insecurity in comparison to the previous prevalence of food insecurity in the United States. In addition, the citation of more literature was suggested.

The situational analysis is only one tenet of the Zero Hunger Pathways Project. In addition to this baseline report, the working group conducted other Dialogue Series that addressed various topics related to food insecurity and food systems. These include Supplemental Nutrition Assistance Program Recommendations, Trade-Offs Evaluations and Science-Policy Communications, and Taking Innovations to Scale. These Dialogue Series, in addition to the situational analysis, support the claim that to achieve Zero Hunger in the United States by 2030, research and policy must seek to make food systems equitable, resilient, healthy, and sustainable.

## Statement of Research Advisor

Cat provided tremendous leadership to drafting, seeking feedback, and finalizing the situational analysis for inclusion on the United States Zero Hunger Pathways Project final report. Initially, Cat led a group of peers in conducting a comprehensive and critical review of literature to support the first draft of the situational analysis. Cat then facilitated receipt of feedback through conduct of a dialogue that many food security experts throughout the United States attended. Following re

<sup>1</sup>Corresponding author: cep0065@auburn.edu

ceipt of feedback, Cat then revised and finalized the situational analysis.

-Alicia Powers, *Hunger Solutions Institute*

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## Authors Biography



Cat Powers is a recent graduate with a B.S. degree in Global Studies at Auburn University. She served as the First Author for this situational analysis.



Amanda Scott Rogers is a recent graduate with a B.S. degree in Global Studies at Auburn University.



Claire Davis is a recent graduate with a B.S. degree in Global Studies at Auburn University.



# The Effect Of Teacher Habit Formation On Preschoolers' Activity Levels

*Camille M. Preston<sup>1</sup>, Katherine E. Spring<sup>2</sup>, and Danielle D. Wadsworth<sup>3</sup>*

<sup>1</sup> Undergraduate Student, Neuroscience - College of Liberal Arts, Auburn University

<sup>2</sup> Doctoral Candidate, School of Kinesiology, Auburn University

<sup>3</sup> Associate Professor, School of Kinesiology, Auburn University

Physical Inactivity is arguably the greatest public health crisis of the 21st century <sup>[1]</sup>, and although the benefits of physical activity (PA) are well established, most children fail to meet PA recommendations <sup>[2,3]</sup>. Establishing PA behaviors in the early years may increase adherence in PA patterns and health benefits across childhood, adolescence, and adulthood <sup>[4]</sup>.

Therefore, interventions aimed at increasing and maintaining PA are essential to curb this public health crisis, particularly in young children. Only 50% of preschool age children meet the physical activity recommendations for their age. Children from high socioeconomic status, urban areas, and preschool age boys are more likely than others to meet these recommendations <sup>[5]</sup>.

Data also shows that preschool years may be a critical period for the development of obesity later in life <sup>[6]</sup>. Obesity, diabetes, and heart disease disproportionately affect individuals from low-income families and are evident even in young children. This health disparity may be ameliorated, in part, by increasing PA at an early age <sup>[7]</sup>.

Physical inactivity habits begin in childhood and continue to develop with age. As with most habits, it is important that the establishment of PA behaviors begin in the early years of a child's life, as this might promote positive trajectories in the patterns of PA throughout childhood, adolescence, and adulthood.

Research shows that teacher-led interventions aimed at increasing PA are effective in the short term but tend to lack long-term success. Preschool teachers serve as the foundation of PA during the preschool years, and teachers' behaviors and PA practices are important to target. Establishing teachers' PA habits may be a sus-

tainable solution to provide PA opportunities in young children.

Researchers gathered height, weight, sex, PA, fundamental motor skills (FMS), and body composition fat mass (FM) and fat free mass (FFM) of 46 preschoolers. PA was assessed with accelerometers placed on preschoolers' wrists during the school day. The preschoolers' baseline FMS assessed object manipulation, locomotion, and stationary skills.

Teachers' habit formation was assessed with the automaticity index in relation to implementing certain types of physical activities, both inside and outside, throughout the day.

Linear regressions examined the relationship between baseline PA levels and body composition to stationary, locomotor, and object manipulation skills. At the end of the 8 week intervention, all measures were retested to see if any improvements had been made.

Our initial results show that body composition in terms of FFM are associated with higher levels of FMS. In particular, stationary and object manipulation scores are associated with greater levels of FFM. Currently, we are analyzing changes in PA, FMS, and body composition over the course of the intervention, as well as the role of teachers' PA automaticity.

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<sup>1</sup>Corresponding author: cmp0082@auburn.edu



**Fig. 1.** Assessing how well a child can walk up a set of steps.

**Table 1.** Participant Data

Variable	Mean	Standard Deviation
Weight	15.56 kg	2.08 kg
Height	100.41 cm	5.70 cm
BMI %	15.50 kg/m <sup>2</sup>	1.15 kg.m <sup>2</sup>
Body Fat %	20.87%	3.01%
Fat Mass	3.34 kg	.83 kg
Fat Free Mass	12.48 kg	1.50 kg
Stationary Skills	48.16	6.46
Locomotor Skills	145.8	27.73
Object Manipulation Skills	34.88	6.46
Gross motor Quotient	96.32	12.57

Notes: SS, LS, OMS, are means of raw scores which are then used to calculate GMQ. There is not a unit of measure for these items.

## Statement of Research Advisor

This project is one of the few studies to examine body composition in young children, as well as, the roles of active play, physical activity and fundamental motor skills in childhood obesity. Furthermore, examining teacher's automaticity in providing physical activity opportunities as the mechanism of change provides a method for sustainable solutions to increase physical activity.

– Danielle D. Wadsworth, School of Kinesiology, College of Education

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Danielle D. Wadsworth is an Associate Professor at Auburn University, and the director of the Exercise Adherence and Obesity Prevention Lab. Her research focuses on understanding the psychological, social, environmental, and behavioral underpinnings of long-term exercise behavior across the lifespan and translating these findings to effective evidence-based interventions.

## Authors Biography



Camille M. Preston is a senior student pursuing a B.S. in Neuroscience at Auburn University. Throughout this project, she has assisted in the data collection for physical activity, weight, height, teacher automaticity, and body composition. She has also assisted in observing teacher and preschoolers' physical activity practices in the classroom.



Katherine E. Spring is a Doctoral Student in the Exercise Adherence and Obesity Prevention Lab at Auburn University. Her research focuses on the effects of physical activity and play have on physical, behavioral and learning outcomes in young children. These evidence-based outcomes are used to design and implement based interventions at the individual, interpersonal and/or organizational level of the socioecological model.

# Investigating Plasma Inflammatory Cytokine Levels' Relationship to Pregnancy Outcomes in *Bos Taurus* Heifers

Gavin L. Rankins<sup>1</sup>, Rachel A. Hollingsworth<sup>2</sup>, Erin E. Mahoney<sup>2</sup>, Anna G. Holliman<sup>2</sup>, and Paul W. Dyce<sup>3</sup>

<sup>1</sup> Undergraduate Student, Department of Animal Sciences, Auburn University

<sup>2</sup> Graduate Student, Department of Animal Sciences, Auburn University

<sup>3</sup> Associate Professor, Department of Animal Sciences, Auburn University

Anytime a heifer (a bovine female from weaning until the first parturition event) or cow (bovine female which has already calved at least once) fails to calve, it is costly to the individual producer and the beef industry as a whole. Open beef heifers are one of the costliest inefficiencies on an operation because the producer has already expended resources to raise her, and the heifer will fail to produce progeny. If a producer pregnancy checks their herd at weaning, an open cow is less costly compared to an open heifer because the cow can be culled before any extra feed is invested in her. However, the producer still incurs the cost of replacing the cow and many producers do not pregnancy check their cattle. Only 34.5% of all beef producers in the United States use pregnancy detection as a management tool to make culling decisions (Lamb et al, 2014). For nearly two-thirds of all United States' beef producers, open cows are first diagnosed at the end of the subsequent calving season. While the producer can opt to cull open cows at the end of the calving season, the cost of maintaining her without a return has already been incurred for a year (Lamb et al, 2014). In the Southeast, it typically takes a raised replacement heifer six successful parturition events to pay for the cost of her development and maintenance in the herd. If a cow fails to calve in one of her first six opportunities and the producer opts to keep her, it takes nine parturition events to pay for the cost of her development and maintenance in the herd (Boyer et al, 2020). The time it takes to receive a profitable return from an individual beef cow emphasizes the importance of fertility in beef production. In 2014, Lamb et al estimated the total cost of infertility to the entire United States beef industry to be about \$2.8 billion annually (Lamb et al, 2014).

The goal of this research project is to test possible biomarkers as predictors for future reproductive potential in beef heifers. The ultimate goal is to find a reliable biomarker in the plasma and a test which accurately quantifies that biomarker. The test could then be used as a predictor of future reproductive success, or lack thereof, in beef heifers. The specific objective of this project is to investigate the relationship between plasma inflammatory cytokine levels and pregnancy results in heifers.

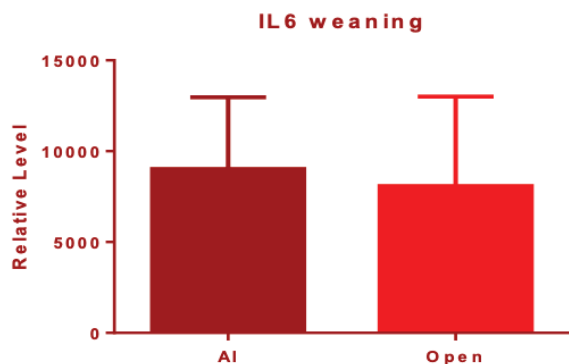
Estrous synchronized Angus and Angus-cross heifers were utilized for this project. Following estrous synchronization, fixed-time artificial insemination (FTAI) was performed on the heifers and fourteen days later a fertile bull was introduced for three consecutive estrous cycles. Heifers conceiving from FTAI were labelled as high-performing heifers, while those heifers which remained open following both FTAI and bull exposure were labelled as low-performing heifers. Previous research conducted in our lab identified metabolites linked to inflammation in the blood plasma which were at different levels at weaning in high-performing and low-performing heifers (Phillips et al, 2018). Furthermore, mRNA transcript levels for the inflammatory cytokines, tumor necrosis factor-alpha (TNF- $\alpha$ ) and interleukin-6 (IL-6), differed in the white blood cells between these two groups (Phillips et al, 2018). In the current project, we evaluated the levels of TNF- $\alpha$  and IL-6 proteins in the plasma using Western blotting. The levels of the two proteins were compared in high-performing (n=9) and low-performing (n=6) heifers in blood samples drawn at weaning and at the time of AI. Target proteins were quantified using ImageJ densitometry.

<sup>1</sup>Corresponding author: glr0018@auburn.edu



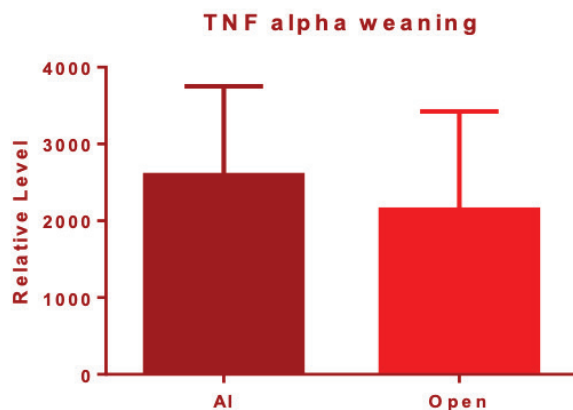
etry and compared using a t-test in GraphPad Prism software.

Based on the previous findings in our lab of significantly different mRNA transcript levels for IL-6 and TNF- $\alpha$  in the white blood cells between high-performing and low-performing heifers (higher transcript levels in the low-performing heifers) (Phillips et al, 2018), the hypothesis was the low-performing heifers would have higher plasma protein levels for IL-6 and TNF- $\alpha$ . However, no statistical difference was found in either of the cytokine levels in the samples taken at the time of weaning (Figures 1 and 2) or the samples taken at the time of artificial insemination. Several possible explanations could explain this observation. The obvious conclusion to draw from this study is neither IL-6 nor TNF- $\alpha$  are adequate as a potential predictive biomarker for future reproductive success in beef heifers. An explanation for the difference in the transcript levels and the protein levels could be a post-transcription regulation factor. Oftentimes, mRNAs function as a regulatory factor by preventing the translation of mRNA to protein. There are many other regulators of translation as well, and one or more of these regulatory factors could potentially cause the differences seen in this protein study compared to the white blood cell transcript study. While this study focused on comparing the cytokine levels based on a per volume of plasma basis, the protein levels could also be compared on a total plasma protein basis. Work was devoted to this thought during the project; however, running the Western blots on an equal total protein load did not result in any significant difference in the cytokine levels between the low-performing and high-performing heifers.



**Fig. 1** Comparison of the relative levels of IL-6 in equal volumes of blood plasma at weaning in high-performing (bar labelled AI) and low-performing (bar labelled

Open) heifers.



**Fig. 2** Comparison of the relative levels of TNF- $\alpha$  in equal volumes of blood plasma at weaning in high-performing (bar labelled AI) and low-performing (bar labelled Open) heifers.

### Statement of Research Advisor

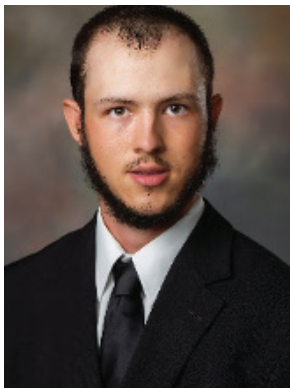
Over the past year Gavin has worked to establish and test a replicable method in the lab to compare inflammatory cytokines, in bovine blood plasma. This assay will permit examination of other blood parameters in heifers at various stages of development.

- Paul Dyce, College of Agriculture

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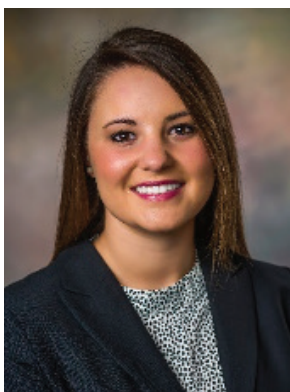
## Authors Biography



Gavin L. Rankins is a May 2022 graduate of Auburn University with a B.S. degree in Animal Sciences – Animal/Allied Industries. He has been member of the Auburn University Reproductive Biology Lab from May 2021 until graduation with a research focus on proteins.



Anna G. Holliman is a graduate student in the College of Agriculture at Auburn University. She received her B.S. degree at Mississippi State University in Animal and Dairy Sciences and Biological Sciences. She joined the Reproductive Biology Lab in Fall 2021. Her research focus is on proteins.



Rachel A. Hollingsworth is pursuing an M.S. degree in reproductive biology in the Department of Animal Sciences of the College of Agriculture at Auburn University. She received a B.S. degree in Animal Sciences at Auburn University. Her current research is focused on utilizing molecular biology tools and techniques to validate potential targets which might serve as indicators of beef heifer fertility.



Dr. Paul W. Dyce is an Associate Professor of Reproductive Biology, Department of Animal Sciences, College of Agriculture at Auburn University.



Erin E. Mahoney is a May 2020 graduate of the University of Tennessee at Martin. She received a B.S degree in animal science and a B.S degree in veterinary technology at UTM. She is in her second year of her masters studying reproductive biology and bovine fertility. She hopes to receive a spot at a U.S. vet school upon graduation.

# Parenting Behavior in Two Distinct Juvenile Populations

Elise M. Reach<sup>1</sup> and Kelli R. Thompson<sup>2</sup>

<sup>1</sup> Undergraduate Student, Department of Psychological Sciences, Auburn University

<sup>2</sup> Assistant Research Professor, Department of Psychological Sciences, Auburn University

Research comparing adolescents with illegal sexual behavior (AISB) and adolescents with general delinquent behavior (AGDB) finds the two distinct populations to differ in many ways while also sharing some unique features (Seto & Lalumière, 2010). In particular, the groups share similarities in relationships with family and attachment styles, whereas the AISB group often scored higher on childhood victimization (Seto & Lalumière, 2010). It is important for clinicians working with this population to understand the family relationships and attachment styles of the youth client (ATSA, 2017). Caregiver attachment mediates the relationships between childhood victimization and later self-disclosure of illegal sexual behavior, highlighting the importance that childhood victimization may play in one's ability to trust and engage in the treatment process (Harrelson et al., 2017). Vulnerability is a critical element in a young offender's life, sometimes resulting from poor attachment bonds between the child and parents and leading to low self-esteem, poor social skills, or lack of empathy for others (Marshall, 2010). Thus, evidence suggests attachment problems, intimacy deficits, and loneliness are significant features of justice-involved youth, particularly AISB. Given how attachment deficits may interfere with the treatment process, the purpose of the current study was to investigate differences in parenting behaviors in a sample of detained youth.

Archival data were used from a high-security residential facility serving male adolescents ( $n = 48$ ) adjudicated for serious delinquent behavior, like homicide, rape, robbery, aggravated assault, and kidnapping. A series of one-way ANOVAs were used to analyze the relationship. The group variable was the type of offense, general delinquent behavior vs. illegal sexual behavior (AISB;  $n = 28$  and AGDB;  $n = 20$ ). The outcome variables were parenting behaviors measured by the Alabama Parenting Questionnaire (APQ; Frick, 1991). The

APQ is a well-validated self-report scale of parenting<sup>1</sup> behaviors across five key domains: positive parenting, corporal punishment, inconsistent discipline, parental involvement, and supervision/monitoring (Zlomke, 2015). Results indicated the groups differed significantly on poor monitoring,  $F(1, 46) = 8.53$ ,  $p = .005$ , while inconsistent discipline was approaching significance,  $F(1, 47) = 3.34$ ,  $p = .075$ . The overall group means poor monitoring was 27.70 ( $SD = 6.64$ ). As seen in Figure 1, the AGDB group scored higher than the overall group mean and the AISB group.

These results suggest the need for ongoing parental involvement for youth in residential treatment settings. These youth may often return to homes characterized by poor monitoring and inconsistent discipline at a critical time post-treatment when the youth need more support than ever. Just as youth have learned new skills for life improvement, clinicians working with these youth may also want to ensure caregivers have improved skills. This would align with prior research indicating a link between certain parenting behaviors, such as caregiver consistency, and risk for later reoffending (Henggeler et al., 2009; Seto & Lalumiere, 2010). This is a challenge for clinicians working in residential settings, where contact with caregivers may not always be easy.

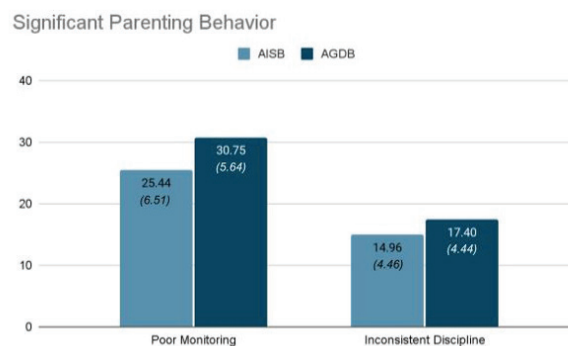


Fig 1. Means & Standard Deviations of Main Study

<sup>1</sup>Corresponding author: emr0046@auburn.edu



Variables.

3159-3169, (2015). **Journal Paper**

**Note:** AISB = Adolescents with Illegal Sexual Behavior; AGDB = Adolescents with General Delinquent Behavior.

### Statement of Research Advisor

Elise first started this research question as a directed readings course credit project with the Juvenile Delinquency Lab in Spring 2022. She stayed with the project and first presented the data at the 41st Annual Research Conference for the Association of the Treatment of Sexual Abusers in Los Angeles, CA, in Fall 2022. She continues to see this project through publication as she looks toward graduate school.

- Dr. Kelli Thompson, College of Liberal Arts

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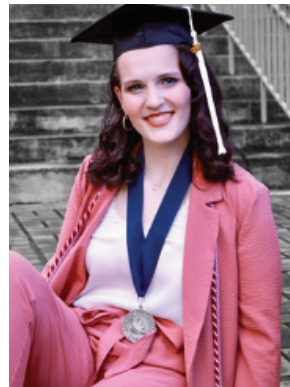
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### Authors Biography



Elise M. Reach is a graduating senior pursuing a B.A. degree in Psychology at Auburn University. She has played key research roles in parenting behavior and juvenile delinquent behavior. Elise is planning on continuing her education to Master and Ph.D. She has applied for different psychology/counseling master's programs around the United States. She has a real passion for understanding the mind and why people do the actions they do.



Kelli R. Thompson is an Assistant Research Professor in the Department of Psychological Sciences. Her lab is supported by a more than a 20-year public-public partnership with the Alabama Department of Youth Services. Her research interests include the assessment and treatment of adolescents with illegal sexual behavior. She teaches a comprehensive healthy sexuality group at the Mt. Meigs facility. Undergraduate research mentoring remains a personal and professional focus.



# A Measurement Device for Quantifying Neuromuscular Impairment in Hands

Mark A. Roces<sup>1</sup>, Chandler T. Stubbs<sup>2</sup>, and Chad G. Rose<sup>3</sup>

<sup>1</sup> Undergraduate Student, Department of Mechanical Engineering, Auburn University

<sup>2</sup> Graduate Student, Department of Mechanical Engineering, Auburn University at Montgomery

<sup>3</sup> Assistant Professor, Department of Mechanical Engineering, Auburn University

Neuromuscular impairments such as cerebral palsy, stroke, and spinal cord injuries limit the ability to perform activities of daily living, decreasing the quality of life of those affected. Soft exoskeletons have been proposed to address this need by assisting hand motor function for those with motor impairment to improve their overall quality of life. Before these devices can be commercialized and implemented in the home and clinic, their performance must be validated. To test the effectiveness of hand devices, there have been efforts to create models of the human hand. An instrumented hand developed recently has displayed near identical range of motion (ROM) and joint stiffness to a healthy hand [1]. However, impaired hands can be very different from healthy hands, with changes in stiffness and ROM (e.g., hypertonia). To test the effectiveness of soft devices on impaired hands, a model needs to be developed to simulate the range of joint stiffness and ROM seen in impaired hands. To build this model, designers first require accurate measures of impaired hand properties. The goal of this project is to develop a device that can accurately measure joint stiffness and ROM for testing on impaired subjects to make modifications to the instrumented hand and assist in the validation of soft exoskeletons. Validation of this device rests on producing the same double-exponential relationship between stiffness and ROM as seen in prior research [2]. The device, shown in Fig. 1, uses a motor-driven arm to rotate the subject's finger, which is attached to a load sensing component via a splint.

device reads force measurements through a LabVIEW script.<sup>1</sup> Each increment is held for 10 seconds, and the average force reading is recorded. Fig. 2 shows the joint torque results from initial measurement runs, accounting for finger and splint linkage lengths, along with a double-exponential trend line.

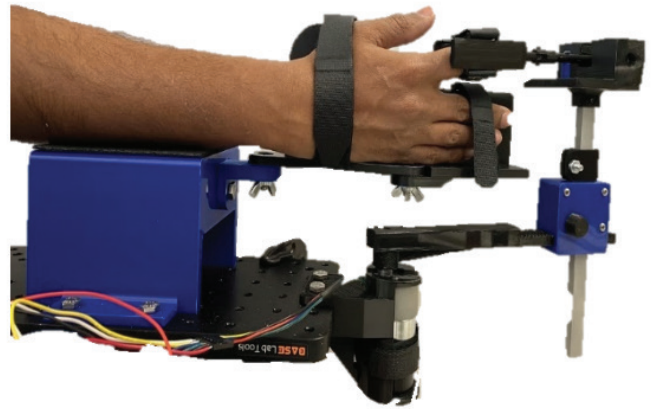


Fig 1. Joint stiffness measurement device.

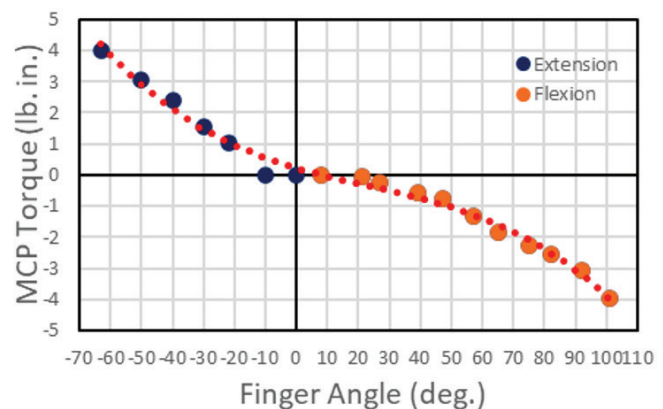


Fig 2. Joint torque results plotted against finger angle.

The readings show a range of motion of  $-65^\circ$  in extension to  $100^\circ$  in flexion, with a joint stiffness of 4 in-lb. to -4 in-lb., respectively. The results show double-expo

The subject's palm and idle fingers are held in place at a preset angle which can be adjusted. An encoder built into the motor tracks joint angle and interfaces with an Arduino microcontroller to increment finger angle by  $10^\circ$  in flexion and extension. A 50 lb. load cell connected to a National Instruments data acquisition

<sup>1</sup>Corresponding author: mar0083@auburn.edu

nential relationship between the two measured quantities, validating the device for further measurements. Modifying the device further will allow for more comprehensive measurements to be taken of hands of individuals with neuromuscular impairment. The gathered biomechanical data then be applied to the instrumented hand to validate soft exoskeletons.

### Statement of Research Advisor

Mark, through his independent research in my group has addressed a bottleneck in the measurement of biomechanical joint properties of individuals with neurological disorders or injuries, by developing a testbed which will serve a crucial role in future studies. Further, the adjustability of his testbed will enable investigations into the impacts of interjoint couplings between the wrist and fingers, further contributing to the field.

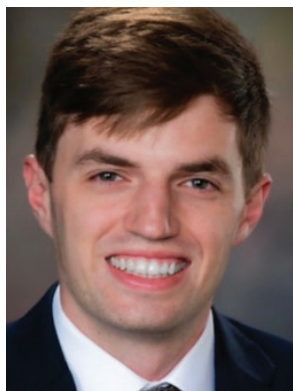
- Chad G. Rose, Samuel Ginn College of Engineering

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### Authors Biography



Mark Rocés is a senior-year student pursuing a B.S. degree in Mechanical Engineering at Auburn University. Following graduation, he will be working as a Manufacturing Engineer for Eaton Corporation.



Chandler Stubbs is a Graduate Student pursuing an MS degree in Mechanical Engineering at Auburn University. He received a B.S. degree in mechanical engineering at Auburn University. He provided mentorship and guidance in the development of the device. His interests include robotics and human-robot interaction.



Chad G. Rose, PhD is an Assistant Professor in the Department of Mechanical Engineering. He holds a bachelor's degree in Mechanical Engineering from Auburn University and a master's degree and Ph.D. in Mechanical Engineering from Rice University. Dr. Rose's primary research focus is on the design and control of robots to rehabilitate, assist, or augment human motor and sensory function.

# High Speed Event Based Camera Modeling for Fluid Flow Synthetic Image Processing

*Nick Rush<sup>1</sup>, Michael C. Hamilton<sup>2</sup>, and Brian Thurow<sup>3</sup>*

<sup>1</sup> Undergraduate Student, Dept. of Electrical and Computer Engineering, Dept. of Aerospace Engineering, Auburn University

<sup>2</sup> James B. Davis Professor, Department of Electrical and Computer Engineering, Auburn University

<sup>3</sup>Department Chair, Department of Aerospace Engineering, Auburn University

Fluid modeling for hypersonic applications is currently a field of great interest, though these models are currently limited without empirical data to verify the models on. Typical high-speed wind tunnels often impart geometric constraints along with harsh flow environments, making observation of the flow difficult. To improve models, high resolution and high frame rate imaging systems are required in order to capture the behavior of the flow. With higher resolution and frame rates, there is a requisite increase in data throughput, which further exacerbates the geometric constraints present in most wind tunnels. To decrease required data throughput, various methods of data compression have been proposed: multi-level processing to determine pixel relevancy <sup>[1]</sup>, in pixel analog-to-digital conversion <sup>[2]</sup>, and even removal of redundant pixels through a bio-inspired solution, typically known as event-based camera operation.

Event cameras differ from conventional cameras in that each pixel is read out asynchronously, depending on whether there have been any changes in light detected in the pixel. Due to this, only new image data is transmitted from the sensor, dropping redundant pixels that have not changed in the observed scene. The primary benefit to asynchronous sampling for event cameras is that for scenes that have primarily still scenery with motion in the foreground, the area of motion is sampled at a much higher rate than the still background. For high speeds characterizing supersonic and hypersonic flows, this focus on motion is paramount.

Due to the difficulty of integrating a new camera system, this work utilized a simulation-based approach to determine the efficacy of event cameras for high-speed fluid flows. Synthetic images were generated at varying

particle densities to simulate a particle image velocimetry (PIV) experiment, and a model of an event camera was created to process these generated images. The highest particle density trial can intuitively be seen to be the trial that would create the most events. As each particle moves through the simulated scene, it generates events, so more particles entail more events for a given flow field.

With the highest density trial, on average, only 30% of pixels between generated images showed change in lighting significant enough to warrant registering an event, indicating that the majority of data collected by a standard frame camera for the same scene would be redundant from frame to frame.

With these benefits come certain restrictions. First, particles used for PIV measurements need to be carefully selected so that they reflect enough light to trigger an event in a real event camera system. Second, high speed PIV applications rely on pulsed light sources to help mitigate motion blur, though pulsed light sources may generate false events in a real event camera system. Both limitations must be weighed carefully before implementation, which is not an insurmountable situation.

Event cameras are continually evolving and becoming more effective at sampling scenes with high motion content and pose a serious opportunity for more effective fluid flow measurements, particularly showing promise for PIV experiments. Other fluid flow experiments, such as oil flow visualization and pressure sensitive paint methods, may also experience benefits from event cameras. Simulations can be conducted for these types of experiments in future work, leveraging the re

<sup>1</sup>Corresponding author: nvr0001@auburn.edu

sults of the efforts of this project.

## Statement of Research Advisor

This undergraduate research project that Mr. Rush has undertaken, regarding creating new methods to tackle the big data problem for large camera arrays, is very timely, important, and relevant to ongoing interdisciplinary projects between my group in ECE and Dr. Thurow's group in AE.

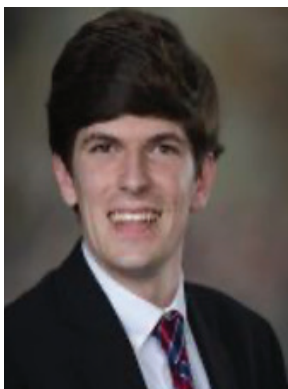
- *Michael C. Hamilton, Electrical and Computer Engineering*

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## Authors Biography



Nick is a senior majoring in electrical and aerospace engineering at Auburn University. He performs research with the Auburn Nanosystems Group over various topics including nano-films, cryogenics, and imaging system hardware design. He was chapter president of Sigma Gamma Tau at Auburn University, and has previously interned with Uhnder Inc., where he worked on RTL automation scripts. He is currently interning with The MITRE Corporation, working on anti-tamper digital hardware designs.



Dr. Michael C. Hamilton received the B.S.E.E. degree from Auburn University in 2000, and the M.S.E.E. and Ph.D. degrees in electrical engineering from The University of Michigan, in 2003 and 2005, respectively. From 2006 to 2010, he was a member of Technical Staff at MIT-Lincoln Laboratory. In 2010 he joined the Electrical and Computer Engineering Department at Auburn University, where he is now a James B. Davis Professor. In addition to his research group (Auburn Nanosystems Group) at Auburn University, he is the Director of the Alabama Micro/Nano Science and Technology Center (AMNSTC). In 2022, he joined the Google Quantum AI team as a Visiting Faculty Researcher. His research interests include superconducting electronics technologies, microwave superconductivity and packaging/integration technologies for extreme environments (including cryogenic and quantum systems). He has served as the Auburn University IEEE Student Chapter Faculty Advisor, is Chair of MTT-7 Technical Committee on Microwave Superconductivity and Quantum Technologies, and is an IEEE JMW Topic Editor in the MTT-7 area.





Dr. Brian Thurow is the Department Chair and W. Allen and Martha Reed Professor in the Department of Aerospace Engineering at Auburn University. He received his B.S. (1999), M.S. (2001) and Ph.D. (2005) from The Ohio State University and established the Advanced Flow Diagnostics Laboratory at Auburn University in 2005. Dr. Thurow has taught classes in aerodynamics, propulsion, light-field imaging, optical diagnostics, compressible fluid dynamics and turbulence. His research program is focused on the development and application of advanced image-based diagnostics for aerothermal measurements with a focus on multi-spectral and 3D imaging techniques using plenoptic cameras or multi-camera arrays. His research has been sponsored by the Army Research Office (ARO) and Air Force Office of Scientific Research (AFOSR), both of which started with Young Investigator Program grants, as well as the Office of Naval Research (ONR), Air Force Research Laboratory (AFRL), National Science Foundation (NSF), NASA Langley Research Center, Sandia National Laboratories and several DOD SBIR/STTR awards. Dr. Thurow has received numerous awards including an NDSEG Graduate Research Fellowship; the SGA Outstanding Faculty Member Award in 2006, 2008, 2009, 2012 and 2013; the AIAA Most Outstanding Faculty Member Award in 2009, 2012 and 2013; the William F. Walker Teaching

Award for Excellence in 2009; the Auburn Alumni Engineering Council Research Award for Excellence in 2009; the Provost's Award for Supporting Graduate Scholarship in 2011 and the 2015 Konrad Dannenberg Educator of the Year Award.

# Determining the Ability of Polyphosphate Accumulating Organisms to Use Organic Compounds in Algal Photosynthate

Justus Smith<sup>1</sup>, Qichen Wang<sup>2</sup>, and Brendan Higgins<sup>3</sup>

<sup>1</sup> Undergraduate Student, Department of Biosystems Engineering, Auburn University

<sup>2</sup> Postdoctoral Researcher, Department of Biosystems Engineering, Auburn University

<sup>3</sup> Assistant Professor, Department of Biosystems Engineering, Auburn University

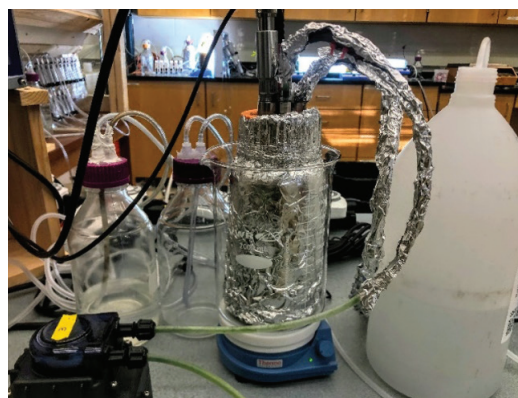
In current methods of wastewater treatment, aerobic bacteria play a vital role in the degradation of pollutants and the purification of waste. However, these bacteria require high levels of oxygen to operate, leading most treatment plants to synthetically aerate their systems. This process is costly and adds significant expense to a system that already incurs deficit spending to create.

In searching for an alternative to oxygenate these bacteria, photo-activated sludge systems have recently emerged as a possible solution (Mohamad et al., 2021). These systems use cultures of microalgae to oxygenate the aerobic bacteria rather than artificially, reducing the need for artificial aeration. As well, the presence of microalgae increased the abundance of certain bacteria, namely Polyphosphate Accumulating Organisms (PAOs). It is apparent that the algae and PAOs form a symbiotic relationship due to the algae's oxygenation, but little is known about the effect of algal secretions, also known as Algal Photosynthate (AP), on this relationship. Since PAOs are known to require carbon compounds, namely Volatile Fatty Acids (VFAs), and since similar carbon compounds to these are found in AP, it is possible that the interchange between these compounds significantly contributes to the relationship between the organisms.

Before work could be done to determine the effects of AP on PAO abundance and productivity, it was first important to develop a consistent source of PAOs for further use in the experiment. To do this, a sample of activated sludge was first taken from a local wastewater treatment plant in Columbus, Georgia, and added to a continuous reactor system developed specifically for this project (Fig. 1). This 1L seeding reactor would

continuously cycle every 8 hours through a 1:4 ratio of VFAs and Artificial media, exporting half of its volume at the end of each cycle to allow for continuous nutrient uptake by the bacteria. In the 8 hour cycle, the reactor would consist of 5 phases: Feeding (hours 0-0.5), Anaerobic Operation (hours 0.5-2.5), Aerobic Operation (hours 2.5-5.5), a settling phase (hours 5.5-7.5), and a waste removal phase (hours 7.5-8). The reactor was also covered completely to prevent algal growth.

A unique component of PAOs that makes them especially useful for wastewater research is their ability to uptake phosphate in aerobic environments yet release phosphate in anaerobic environments. This oxic-anoxic synthesis and release allow researchers to manipulate phosphorus removal from wastewater by controlling the vacillation of these conditions. As well, since PAOs can operate in both presence and absence of dissolved oxygen, they can become the dominant bacteria in activated sludge where these cycles are present. To simulate aerobic-anaerobic cycling in the PAO seeding reactor, a fish tank aerator was attached to humidifying device and cycled on and off.

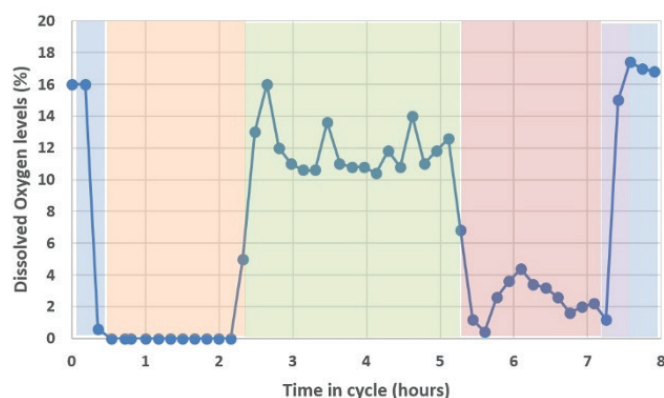


**Fig. 1** PAO seeding reactor in operation.

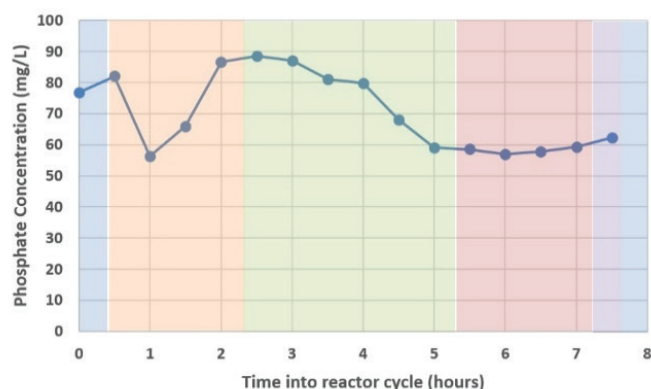
<sup>1</sup>Corresponding author: Justus@auburn.edu

After the AftPAO seeding reactor was operated continuously for 1 month, a kinetic study was performed in which the reactor would be sampled every 30 minutes for one full cycle of the reactor. These samples would then be analyzed for pH, Optical Density, Dissolved Oxygen, and Total Phosphate levels to gauge favorable PAO growth conditions and the presence of PAOs.

Results of this kinetic study indicated that the reactor was truly operating under aerobic and anaerobic conditions (Fig. 2). As well it also showed concentrations of Phosphate increasing in the anaerobic phase of the cycle, and decreasing during the aerobic phase, indicating the presence of PAOs within the system (Fig. 3).



**Fig. 2** Dissolved Oxygen Levels from Kinetic study on PAO seeding reactor.



**Fig. 3** Phosphate Concentrations from Kinetic study on PAO seeding reactor.

In conclusion, PAOs can indeed be found in a wastewater treatment plant local to the Auburn area. Further kinetic studies will now be done to verify preliminary testing, and pending significant results, the seeding reactor will be used to provide PAOs for future experiments to observe the relationship between these valu-

able microorganisms and Algal Photosynthate.

## Statement of Research Advisor

Understanding how algae interact with polyphosphate accumulating organisms (PAO) is critical to designing photosynthetically-powered biological phosphorus removal systems. Optimizing such systems can improve wastewater treatment while reducing energy and greenhouse gas emissions. Justus's research project resulted in the setup of a PAO reactor in our laboratory that has been operating nearly continuously for nearly six months. This is an important first step to developing replicated continuous PAO reactors.

- Dr. Brendan Higgins, Department of Biosystems Engineering

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## Author Biography



Justus Smith is a sophomore year student pursuing a B.S. degree in Biosystems Engineering at Auburn University. He has played key research roles in the culturing of specific heterotrophic bacteria from a local wastewater treatment plant for further experiments on wastewater remediation.



Qichen Wang is a post-doctoral researcher in the Department of Biosystems Engineering. His research is about developing biological processes for nutrients and carbon recycling from wastewater. Qichen received his first bachelor's degree in Food Engineering at Dalian Ocean University. He received

his second bachelor's degree in Biology and his master's degree in Marine conservation and policy at Stony Brook University. He received his Ph.D. in Biosystems Engineering at Auburn University.



Dr. Brendan Higgins is an Assistant Professor in the Department of Biosystems Engineering at Auburn University. His research encompasses applications in bioenergy, waste remediation, water quality, and production of high-value nutraceuticals. He continues to study combined algal-bacterial processes with a particular focus on mechanisms of interaction.



# Probing Mechanical Heterogeneity of Additively Manufactured Stainless Steel Using Digital Image Correlation

John Snitzer<sup>1</sup> and Xiaoyuan Lou<sup>2</sup>

<sup>1</sup> Undergraduate Student, Department of Materials Engineering, Auburn University

<sup>2</sup> Associate Professor, Department of Materials Engineering, Auburn University

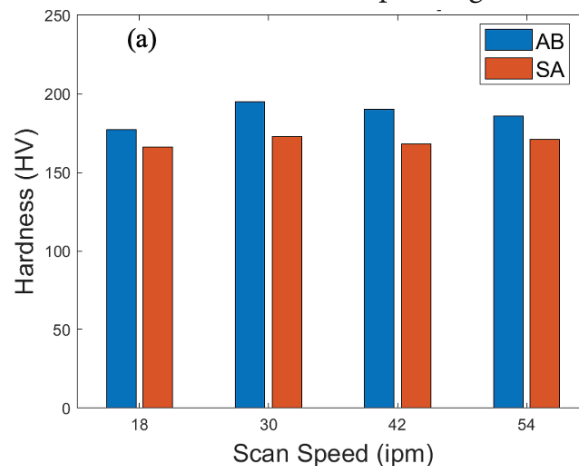
The purpose of this work was to investigate the microstructural and mechanical heterogeneity of additively manufactured (AM) 316L stainless steel. AM is a novel technology facilitating the production of geometrically complex parts that could not be produced otherwise. Parts can be made with short lead time and low upfront cost. The layer-by-layer approach of manufacturing produces near-net shape parts with minimal need of post-processing. The production of AM parts generally produces columnar grains with smaller dendritic cells forming within grains causing anisotropic mechanical properties [1]. Grain and cell properties are heavily dependent on process parameters including scan speed, laser power, hatch spacing, and layer height [1-3]. Microstructural variation impacts mechanical properties such as hardness, yield strength, and tensile strength [3].

An Optomec LENS 500 DED machine was used to manufacture samples with commercially available 316L powder. Given the nature of AM, parts can be made with varying process parameters by editing specific lines of the printing file. The different processing parameters tested in this work were laser scan speed and laser power. All samples were manufactured with a hatch distance of 0.015 inches and a layer height of 0.015 inches. Initially, a parameter study was performed to ensure parts would be manufactured with a density upwards of 99%. It was found that the range of scan speed with a laser power of 400W was 18 to 54 inches per minute (ipm), and the range of laser power with a scan speed of 30 ipm was 300 to 700 W.

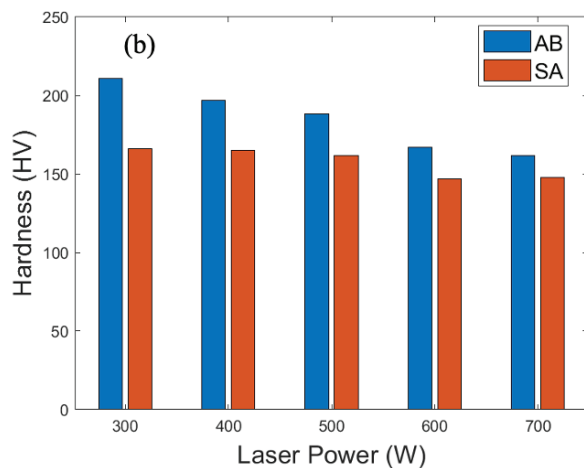
After determining the parameter range to be tested, microstructural characterization samples were manu-

factured with the gradient parameter ranges outlined<sup>1</sup> above. The samples were then cut parallel to the build direction. A solution annealing (SA) heat treatment was then performed (1121°C for 2 hours) on one of the cut samples to compare the as-built (AB) and SA conditions. It was found that varying scan speed did not significantly impact the microstructure in terms of cell and grain size. When varying laser power, a significant change in microstructure was observed. As the laser power increased, the cell and grain size exponentially increased.

The hardness of each sample was measured using Vickers hardness (HV) and can be found in Fig. 1. It was found that the SA heat treatment reduced the hardness by about 20 HV on average for both the gradient power and gradient speed samples. No significant change was observed when varying the scan speed which corresponds well to the lack of change in microstructure. Laser power on the other hand showed a significant drop in hardness as laser power increased. This result was expected due to the higher laser power having higher residual heat and causing an annealing effect in the heat affected zone outside of the melt pool region.

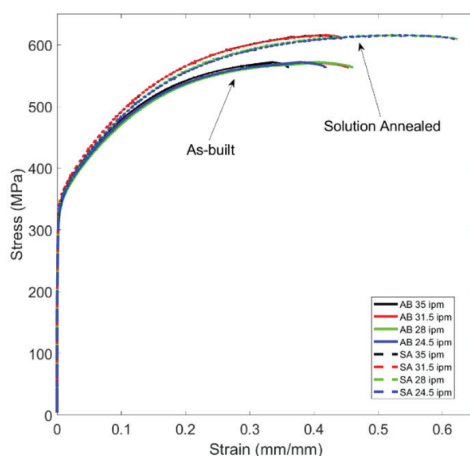


<sup>1</sup>Corresponding author: jas0209@auburn.edu

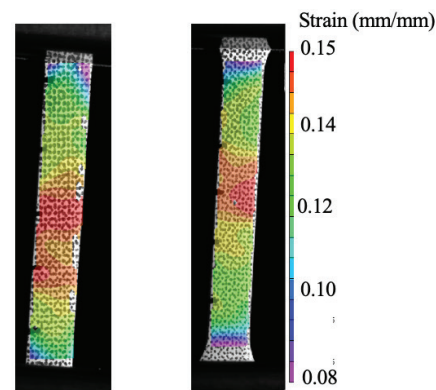


**Fig. 1.** Hardness (HV) vs (a) laser scan speed and (b) laser power.

Tensile bars with gradient parameters were also manufactured to be tensile tested using digital image correlation (DIC). DIC allows for local strain tracking under global stresses. Two samples, AB and SA gradient speed, were tested in this work. The tensile test was performed at a strain rate of  $6.6 \times 10^{-4} \text{ s}^{-1}$  to failure. The tensile test results can be found in Fig. 2. It was observed that the SA samples had higher ductility compared to the AB samples which was expected due to the decrease in hardness and reduction of residual strain. Interestingly, however, the solution annealed sample had a higher tensile strength. This result was not expected given that hardness has been found to be an indicator of tensile strength. It was also observed that strain accumulated along the interface between parameter changes. Further investigation showed a decrease in interfacial hardness.



**Fig. 2.** Stress vs strain of gradient speed AB and SA samples.



**Fig. 3.** DIC mapping of (a) AB at 500MPa and (b) SA at 500MPa where purple represents low strain and red represents high strain.

This work has proved that DIC can be used to determine localized strain under global stresses on AM samples as well as gradient parameter samples. Future work is planned to test the gradient power samples as well as investigate the decrease in interfacial hardness and increase in tensile strength of the SA condition. Future publications are being written in regards to this work.

## Statement of Research Advisor

John developed a strategy to fabricate and validate the use of microstructurally graded specimen to study microstructure- property relationship for additively manufactured steel. The developed method is valuable for increasing the testing throughput to qualify additive manufactured alloys for high- temperature extreme environments in a range of energy and aerospace applications.

- Xiaoyuan Lou, Materials Engineering

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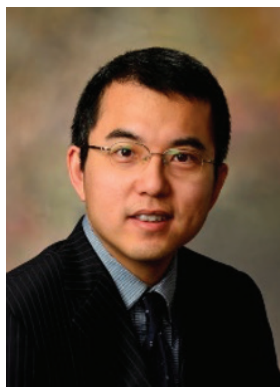
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## Author Biography



John Snitzer is recent graduate from the department of Materials engineering. He has designed gradient specimens to be tested using DIC to reduce the qualification time for AM materials in critical applications. He will be attending Purdue University for a graduate degree in Nuclear Engineering as a DOE-NE graduate fellow.



Dr. Xiaoyuan Lou is an Associate professor in the department of Materials Engineering. He oversaw the work performed and provided perspective into industry applications. He focuses on developing advanced manufacturing methods for applications in extreme environments, where components see high temperature, corrosion and oxidation, irradiation, etc.

# How Parental Relationships Influence Adolescents During the Transition to College

Gabby Thompson<sup>1</sup>, Angel Perry<sup>2</sup>, Ashley Bean<sup>1</sup>, and Sara Driskell<sup>3</sup>

<sup>1</sup> Undergraduate Student, Department of Psychological Sciences, Auburn University

<sup>2</sup> Graduate Student, Department of Counseling, Family & Consumer Sciences, and Kinesiology, University of Montevallo

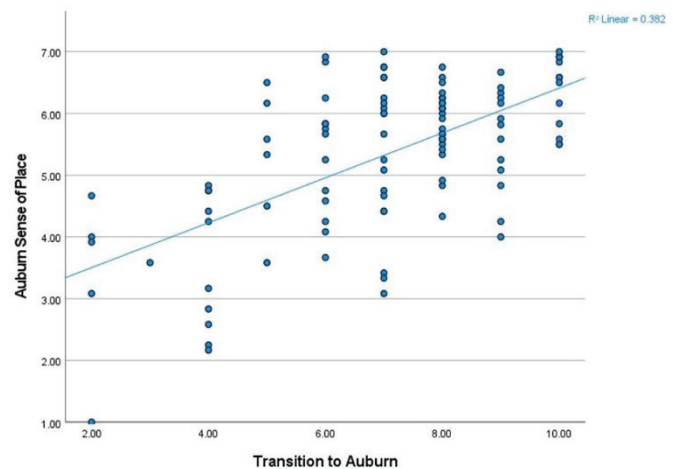
<sup>3</sup> Visiting Assistant Professor, Department of Psychological Sciences, Auburn University

Social relationships are an integral part of college life (Kennedy, 1999). Before leaving home, parents continuously shape how their children interact with others. At college, adolescents have a new opportunity to seek out individuals and relationships in their own way. Because early childhood attachments are incredibly meaningful for adolescents, these past relationships may ultimately shape how they connect with new people on campus (Nijhof & Engels, 2007).

In this survey, we aimed to explore new Auburn students' views on their transition to college and connections to home. We collected data from 106 undergraduate freshman or transfer students, who were surveyed through the Psychology Department's participant pool (83% Women, 22% Men, 0.9% Non-Binary/Gender-Non-conforming; 84% White American, 5.7% Asian/Asian American, 7.5% Biracial/Multiracial, 2.7% Other). We surveyed them on how negative to positive their overall transition to Auburn was, how easy or difficult it was to make friends at Auburn compared to home, how they coped with stress at Auburn, and how their relationship with their guardians had changed since coming to Auburn, what they communicated about with their guardians and their feelings of being at home at Auburn.

Participants ranked their transition to Auburn very positively even during a commonly difficult transition period ( $M=7.05$ ,  $SD=2.10$  on a 0-10 scale). In addition, this transition was strongly correlated with their sense of place at Auburn ( $M=5.35$ ,  $SD=1.24$  on a 1-7 scale),  $r=.62$ ,  $p<.001$ . In turn, sense of place was also strongly correlated with students' current sense of well-being ( $M=3.84$ ,  $SD=0.80$  on a 1-6 scale),  $r=.48$ ,  $p<.001$ . Although many participants said it was easier making friends at Auburn (47.47%), a similar number said it

was easier at home (40.40%); 12.12% said it was the same. This shows that as incoming students have a better transition, they are also feeling more connected to Auburn, and they are doing better overall.



**Fig. 1.** Scatter plot with fit line illustrating the positive correlation between how positively students rated their transition to Auburn and their feelings of a sense of place at Auburn.

Participants reported that they talked with their guardians about many major stressors, including finances ( $M=5.08$ ,  $SD=1.80$ ), academic stressors ( $M=5.75$ ,  $SD=1.44$ ), life decisions ( $M=5.63$ ,  $SD=1.46$ ), and feeling unsure about their decisions ( $M=5.32$ ,  $SD=1.80$ ), all on a 1-7 scale. 46.2% of participants reported that connecting with guardians is one of the main ways that they cope with stress and deal with homesickness. This shows how important these connections to home still are for many students.

We found that incoming Auburn freshmen and transfer students still have positive experiences during this transition period, which relates to their sense of place

<sup>1</sup>Corresponding author: driskell@auburn.edu



at Auburn and sense of well-being. We also found that their ongoing conversations with guardians imply students still need and value support and guidance for the everyday stressors of colleges. This research is important for understanding what factors are correlated with student struggles in a new environment and their ability to have positive first-year experiences and social interactions. Although starting college is an exciting time to be independent and away from guardianship, college students often rely on their parents for support and comfort when embarking on this new journey full of highs and lows.

### Statement of Research Advisor

Gabby conducted an expansive correlational study examining numerous important factors for understanding when incoming students struggle and when they thrive in their new social environments at Auburn. Learning more about these factors can help students have more positive transitions to Auburn as well as more fulfilling student life at Auburn. Gabby's research also has implications for the broader academic, health, and overall personal concerns of first year and transfer undergraduate students at Auburn University.

- Sara Driskell, Department of Psychological Sciences

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### Authors Biography



Gabby Thompson is pursuing a degree in the College of Liberal Arts, specifically in Psychology. During her undergraduate research experience, she was mentored by Dr. Sara Driskell.

# The Removal of Phosphorus from Agricultural Runoff Using Biochar

*Sarah Tyndall<sup>1</sup>, Hossein Jahromi<sup>2</sup>, Sushil Adhikari<sup>3</sup>*

<sup>1</sup> Undergraduate Student, Department of Biosystems Engineering, Auburn University

<sup>2</sup> Assistant Research Professor, Department of Biosystems Engineering, Auburn University

<sup>3</sup> Professor, Department of Biosystems Engineering, Auburn University

Algal blooms and eutrophication in runoff water deposits are problems that many landowners are facing due to the leaching of phosphorus from land applied fertilizers. Not only does this release an unwanted smell to the environment, but it also leads to the starving of oxygen and nutrients from these bodies of water, resulting in the death of these ecosystems.

Biochar is a charcoal like substance that is made by exposing biomass, or organic wastes, to high pressures and temperatures. The high porosity and number of functional groups in biochar prove to be a good candidate of phosphorus absorption. This study analyzes the effectiveness of phosphorus absorption through biochar as well as the effectiveness of phosphorus absorption through biochar impregnated with metal ions.

The first steps taken to analyze biochar absorption were characterization tests. These tests were done in order to obtain a higher level of understanding of the structure and chemical makeup of biochar. It was found that biochar has a very high carbon content, specific surface area, and cation exchange capacity. Carbon has been proven to be very effective in previous absorption studies (Almanassra 1, 2021), as well as samples with high specific areas. The high cation exchange capacity proves to show that when exposed to the metal cations in the impregnation step, the biochar has a higher chance to actually bond with these metals.

The first experiments conducted were phosphorus leaching tests. These tests were performed to determine whether or not the biochar, after washing and drying for 24 hours, still had traces of phosphorus. It was found that biochar was still releasing phosphorus, but not at a level that would affect any samples.

Nonmodified biochar was simply washed and dried<sup>1</sup> before phosphorus testing. In this study biochar was impregnated using the same method with 4 different metals, Aluminum, Calcium, Iron, and Magnesium. Using a 5wt% metal loading, a solution was prepared by mixing the selected metal chloride salt with 100mL of de-ionized water on a stirring plate. The biochar was then added once the metal salt was completely dissolved and allowed to stir for 3 hours. After the solution was stirred for 3 hours, the solution was immediately placed in the 105°C oven for the complete removal of water. The dried biochar was then placed into a 530°C muffle furnace for ten minutes to finish the metal impregnation.

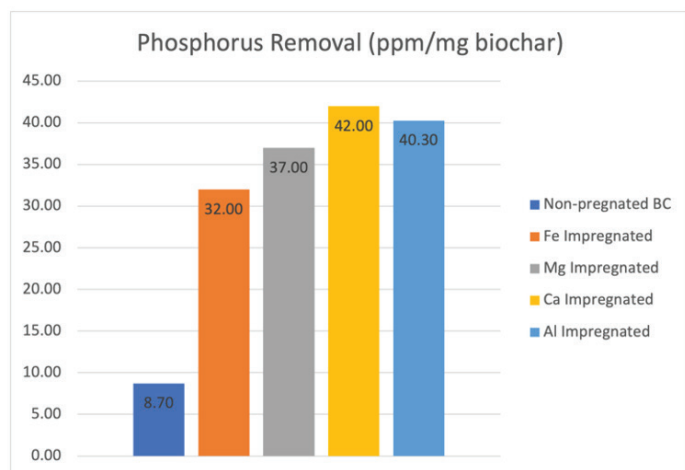
A 5% phosphorus solution was then made by mixing monobasic potassium phosphate with deionized water. All biochar samples, including non-modified, and 4 different impregnated biochars, were then added to 50mL centrifuge tubes at a concentration of 2g biochar/ 20 mL phosphorus solution. These centrifuge tubes were allowed to shake at 350rpm and 40°C for three hours. All samples were then filtered using a vacuum filtration system, separating the solid biochar from the liquid phosphorus solution, and then analyzed using Hach's high range total phosphorus kit (Hach, Loveland, CO, 2013). This kit comes with prepared reagent tubes. The addition of the sample and premade chemical reagents to this tube results in a color change and this color change is measured by the DR900 to determine phosphorus levels in the sample.

The recorded data from one full phosphorus test including non-modified biochar and all four modified biochars can be seen in Fig. 1. It was found that biochar with no modifications removes a small amount of phosphorus per gram of biochar, showing that it does have

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<sup>1</sup>Corresponding author: set0033@auburn.edu

the capability to remove large amounts of phosphorus from runoff water when biochar is applied in larger quantities. It was also found that all of the biochars impregnated with metals removed a much larger amount of phosphorus, making them a better candidate for phosphorus removal in larger applications. This means that, when applied to areas with high levels of phosphorus runoff, biochar holds the capability to effectively remove phosphorus from runoff water, resulting in fewer cases of eutrophication and algal blooms as well as a healthier environment overall.



**Fig. 1.** Results from one complete P-removal test

### Statement of Research Advisor

Sarah performed literature review, developed experimental design, collected data and analyzed. Sarah also worked extremely well interacting with other members of the group.

- *Sushil Adhikari, Biosystems Engineering*

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### Authors Biography



Sarah E. Tyndall is a senior-year student pursuing a B.B.S.E. degree in Biosystems Engineering at Auburn University. She has played key research roles in phosphorus removal using biochar, and hydrothermal liquefaction aqueous phase projects in her lab. She is also an avid lover of music and was a member of the Auburn University Marching band for 3 years.



Hossein Jahromi is an assistant research professor at Auburn University. Hossein received his PHD from Utah State University in 2019 and is an expert in Heterogeneous catalysis, reaction engineering, thermochemical conversion, reaction kinetics, hydrotreatment, material characterization, biofuels, and biomass.



Sushil Adhikari is a professor in the Department of Biosystems Engineering at Auburn University. He is currently the director at the Center for Bioenergy and Bioproducts in Auburn, Alabama. He is an expert in thermochemical conversion processes mainly biomass gasification, fast pyrolysis, hydrothermal liquefaction of algae and upgrading for liquid transportation fuels.



# Superconducting Materials and Devices for Future High-Performance Non-von Neumann Computers

*Harrison A. Walker<sup>1</sup> and Michael C. Hamilton<sup>2</sup>*

<sup>1</sup> Undergraduate Student, Department of Materials Engineering, Auburn University

<sup>2</sup> James B. Davis Professor, Department of Electrical and Computer Engineering, Auburn University

Current computational systems have been primarily based on industry-dominant complementary metal-oxide-semiconductor (CMOS) technology for many decades. Most computers are organized according to the von Neumann architecture in which the processor and memory must communicate with one another to process information. This architecture experiences a bottleneck in information flow that restricts the speed of computation. In the age of big data, this presents a limiting factor for the many computational tasks rooted in machine learning. Alternative platforms being investigated must be highly scalable, demonstrate lower power consumption, and perform at higher levels of speed and efficiency than current systems. Neuromorphic hardware takes inspiration from the human brain and is organized with processing and memory distributed throughout the whole system, aiming to reduce the inherent latency in von Neumann-like systems.

A Von Neumann (or Princeton) architecture has been the basis of most computational systems since its conception. <sup>[1]</sup> This architecture employs a central processing unit (CPU) that works alongside a dedicated memory that stores data and instructions together. The processor and memory must communicate with each other to process information, requiring the movement of data and instructions, leading to an information flow bottleneck that provides one limitation for the speed of computation. Non-von Neumann computing attempts to distribute both processing and memory throughout the whole system with the goal of reducing the inherent latency found in von Neumann-like systems. Neuromorphic computing attempts to mimic the human brain and is organized with both processing and memory distributed among the system. It has been claimed that efficient emulation of scalable biological

neural networks could allow for computation that negates the information bottleneck associated with von Neumann-like architectures and provide a low-power platform more apt for neural networks and parallel processing. <sup>[5]</sup> Novel neuromorphic circuit elements based on superconducting niobium nitride nanowires have been proposed for use in the implementation of physical neural networks. <sup>[3] [2]</sup>

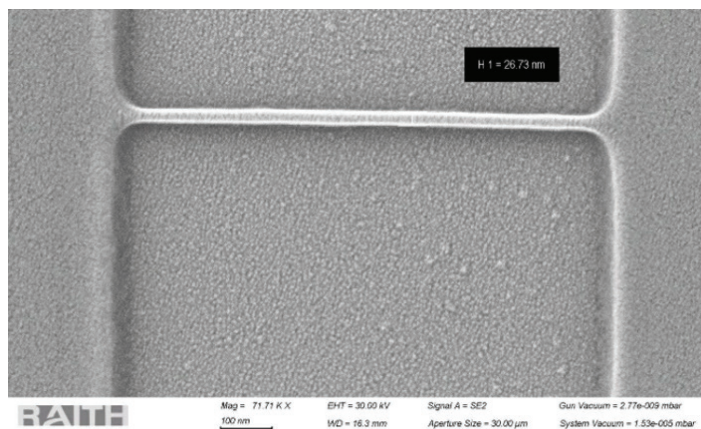
In a superconducting 1D nanowire, quantum phase slip (QPS) causes the wire to demonstrate an insulating, zero-current state when an applied voltage is below a critical value and to exhibit resistive behavior when above. <sup>[6]</sup> Quantum phase slip junctions (QPSJs) are promising superconductive electronic devices for applications in high-speed and low-power neuromorphic computing. Coherent quantum phase slip events can be leveraged by overdamping of QPSJs to create individual quantized current pulses, which are analogous to neuron spiking events in the human brain. Through the implementation of synapse circuitry, non-volatile memory can be realized through spike-timing-dependent plasticity, a biological learning mechanism. <sup>[4]</sup> Simulation of these circuit elements shows promise, but fabrication challenges have hindered the realization of useful devices. To advance these technologies, this undergraduate research fellowship project explored process development for superconducting NbN nanowire fabrication.

NbN thin films were grown on intrinsic silicon substrates using electron beam evaporation deposition techniques. The photoresist PMMA 950K A2 was then spin-coated onto the wafer. Nanowires were patterned into the resist using an electron beam lithography (EBL) system. Nanowires of lengths ranging from 0.38  $\mu\text{m}$  to 1.84  $\mu\text{m}$  and widths of 60 nm and 70 nm were

<sup>1</sup>Corresponding author: haw0022@auburn.edu

patterned to test fabrication capabilities. After EBL, unfocused argon ion milling was used as a physical etching process to remove exposed NbN and leave behind the nanowires. Parameters such as argon flow rate, acceleration voltage, time, and angle of incidence were varied to determine optimal parameters. Etching was characterized by utilizing atomic force microscopy and scanning electron microscopy.

Although difficulties such as disconnections, surface roughness, and polymer crosslinking posed issues during fabrication, the smallest nanowire achieved was 26.73 nm. The nanowire is pictured in figure 1. Electron beam lithography and ion milling as a fabrication process show promise for the manufacturing of ultrathin nanowires for use as quantum phase slip junctions.



**Fig. 1** Fabricated NbN Nanowire.

### Statement of Research Advisor

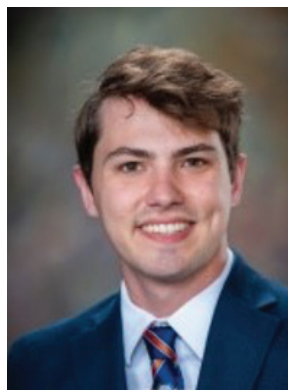
The year-long undergraduate research fellowship project that Mr. Walker has described, focusing on a relatively new type of superconductive electronic device, called a quantum phase slip junction (QPSJ), is very exciting and has significant implications for future computing technologies. Superconducting computing has shown promise of exceptionally high-speed and exceedingly low power dissipation compared to conventional computing. Mr. Walker has performed experimental work to help us to better understand the materials and fabrication processes needed to create useful QPSJ devices for future superconductive electronics.

-Michael C. Hamilton, Electrical and Computer Engineering

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### Authors Biography



Harrison Walker is an Auburn graduate with a degree in Materials Engineering. He has utilized the multidisciplinary nature of materials science to research within the College of Electrical and Computer Engineering, fabricating nanowires to aid in the realization of superconducting neuromorphic hardware. He plans to continue his education by pursuing a PhD at Vanderbilt University in interdisciplinary materials science.



Dr. Michael C. Hamilton received the B.S.E.E. degree from Auburn University in 2000, and the M.S.E.E. and Ph.D. degrees in electrical engineering from The University of Michigan, in 2003 and 2005, respectively. From 2006 to 2010, he was a member of Technical Staff at MIT-Lincoln Laboratory. In 2010 he joined the Electrical and Computer Engineering Department at Auburn University, where he is now a James B. Davis Professor. In addition to his research group (Auburn Nanosystems Group) at Auburn University, he is the Director of the Alabama Micro/Nano Science and Technology Center (AMNSTC). In 2022, he joined the Google Quantum AI team as a Visiting Faculty Researcher. His research interests include superconducting electronics technologies, microwave superconductivity and packaging/integration technologies for extreme environments (including cryogenic and quantum systems). He has served as the Auburn University IEEE Student Chapter Faculty Advisor, is Chair of MTT-7 Technical Committee on Microwave Superconductivity and Quantum Technologies, and is an IEEE JMW Topic Editor in the MTT-7 area.



# Characterization of Ore from the Island Queen Iron Deposit in Puerto Rico

Sam Warren<sup>1</sup>, and Laura Bilenker<sup>2</sup>

<sup>1</sup> Undergraduate Student, Department of Geosciences, Auburn University

<sup>2</sup> Assistant Professor, Department of Geosciences, Auburn University

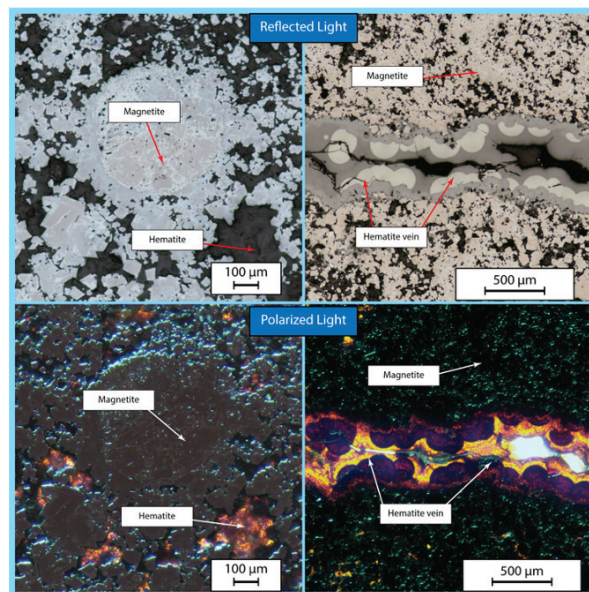
The purpose of this project was to characterize the magnetite ore from the Island Queen iron deposit in Puerto Rico. By using a suite of geochemical methods and microscopic observation, we can determine how the Island Queen deposit formed. Studying metal-rich deposits increases our understanding of how and where Earth's resources are created. This knowledge is significant because metals like iron are necessary for the construction of renewable energy sources and many everyday items.

Island Queen is located on the eastern side of Puerto Rico and is classified as an iron skarn deposit based on the minerals observed in its rocks (Krushensky and Schellekens, 2001). Skarns form when magma releases heat and fluid into a rock layer rich in carbonate, usually limestone (Meinert et al., 2005). This results in new minerals, potentially economic ore, and changes in the physical characteristics of the area surrounding the intrusion. This new cooked and altered rock can contain higher concentrations of metals, and in the case of Island Queen, iron is present in the form of the minerals magnetite ( $\text{Fe}_3\text{O}_4$ ) and hematite ( $\text{Fe}_2\text{O}_3$ ).

To study the deposit before major field work, samples were collected by our collaborators at the University of Puerto Rico, Mayagüez. Four samples of ore were made into thin section slides, which were analyzed using a petrographic microscope. Mineral proportions were estimated and spatial relationships between different minerals were described. This helped us identify the most common minerals and their potential sequence of formation. Petrographic analysis showed that the Island Queen ore rocks are mostly magnetite, hematite, and quartz (Figure 1). There are also trace amounts of epidote and garnet, which are common skarn minerals.

Next, the elemental compositions of the ore minerals

(magnetite, hematite) were measured using laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS).



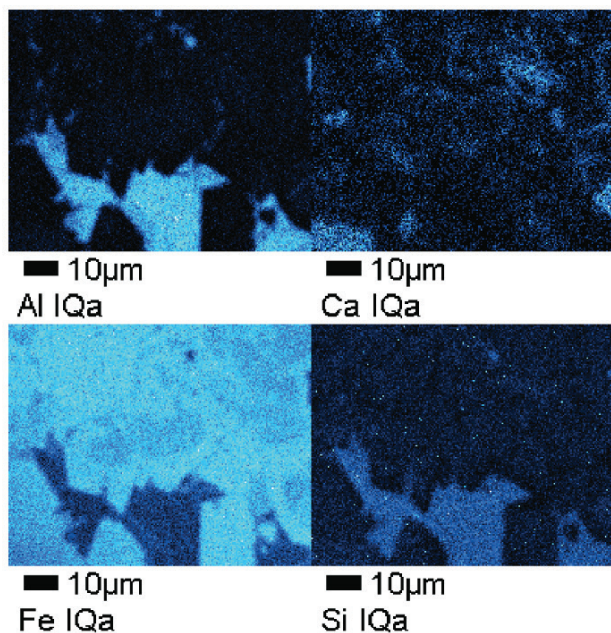
**Fig. 1** Island Queen iron ore minerals under the microscope. The upper two photos show magnetite and hematite under reflected light and the same two photos below in transmitted polarized light. The photos on the right show a vein of hematite that has formed in between the magnetite crystals.

Electron microprobe analysis (EMPA) was performed to measure iron concentrations to calibrate the LA-ICP-MS data and create element concentration maps using backscattered electron (BSE) imaging. In the maps of Figure 2, relative concentrations of aluminum (Al), calcium (Ca), iron (Fe), and silicon (Si) are displayed across mineral grains. Brighter blues indicate higher concentrations of the element measured. Lastly, all the compositional data were compared to other deposits around the world, whose origins are well-studied based on the approach of Nadoll et al. (2014).

<sup>1</sup>Corresponding author: scw0051@auburn.edu



The composition and properties of Island Queen ore is consistent with what is expected for iron skarn deposits. The element concentration map created using EMPA showed some mineral grains had higher levels of aluminum than expected from the minerals observed. This may be due to later natural alteration by fluids that traveled through the area.



**Fig. 2** Elemental concentration maps of Island Queen minerals. The brighter blue indicates higher elemental composition counts.

The calcium content of the ore was relatively low in the analyzed grains, which means that this sample formed relatively close to the magma intrusion.

These observations and measurements lay the important groundwork for characterizing the Island Queen iron deposit, especially to determine sampling and investigation strategies for future field work. This project also refined the analytical methods that we will use to study more Island Queen samples. Future data will provide insight into the exact formation processes that resulted in the Island Queen deposit, which can be applied to the other iron deposits in Puerto Rico and locations around the world.

### Statement of Research Advisor

Sam's semester-long fellowship began with him tackling the obscure geological literature of Puerto Rico and papers on iron mineral geochemistry, which was not a

simple task. For his research, Sam utilized three diverse analytical methods (microscopy, EMPA, LA-ICP-MS) and successfully learned the pertinent skills in a short period of time. He was also active in our Economic Geology & Geochemistry research group meetings, contributing two presentations and regularly engaging in discussion. Sam's characterization of the properties and composition of Island Queen ore will play an important role in field work strategy this summer and in future research on new samples.

- Laura Bilenker, Department of Geosciences

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### Author Biography



Samuel Warren is a recent graduate of Auburn University as of May 2022 with a B.S. Degree in Geology.



Laura Bilenker is an Assistant Professor in the Department of Geosciences and heads the Economic Geology & Geochemistry research group.

# Abuse, Sexual Discomfort, and Victim Typology in Adjudicated Youth

Jenna M. Wettstein<sup>1</sup> and Kelli R. Thompson<sup>2</sup>

<sup>1</sup> Undergraduate Research Fellow, College of Human Sciences, Auburn University

<sup>2</sup> Assistant Research Professor, Department of Psychology, Auburn University

Adolescent sexual offending is a focal public safety issue. Nearly half of all adults with sex offenses reported engaging in sexually abusive behavior beginning in adolescence (Righthand & Welch, 2001). However, intervention is important for adolescents adjudicated for illegal sexual behavior (AISB) because it breaks the cycle through treatment aimed at deterring further victimization of children and adults (Burkhart & Cooke, 2010).

Victim typologies provide important information for assessment and identify important treatment-related factors. For example, child and mixed-age victim offenders are more likely to have been victims of sexual abuse (Kemper & Kistner, 2010). Those with mixed-age victim patterns reported the highest rates of child abuse and most severe family problems (Link & Losel, 2021.) AISB with child-age victims exhibit greater anxiety and internalizing problems than other victim-age patterns (Faniiff & Kolko, 2012). Youth with child-age victims tend to be less sexually experienced and socially isolated, suggesting a higher level of sexual discomfort (Leroux et al., 2016). Additionally, youth with peer-aged victims are more likely to have poor parental monitoring and are less susceptible to parental discord (Faniiff & Kolko, 2012).

The current study sought to define the relationship between a history of sexual abuse and victim age in a sample of male AISB. Data was collected from 916 participants in a court-mandated residential treatment facility. A series of ANOVAs were used where the grouping variable was based on victim-age pattern (1=child age victim, 2=peer or older age, 3=mixed age). These groups were compared on the Millon Adolescent Clinical Inventory (MACI; McCann, 1999) Scales for family discord, childhood abuse, and sexual insecurity. The family discord scale is a measurement of tension and conflict with parents (McCann, 1999). The child abuse

scale is a measurement of the severity of abuse, including physical, sexual, and/or emotional abuse.

The sexual insecurity scale is a measurement of discomfort regarding issues of sexuality and awareness of evolving sexual feelings.

Regarding the family discord scale,  $F(2, 915) = 3.42, p = .03$ , the group with peer-age victim patterns ( $n = 236$ ) had the lowest mean scores compared to the other victim-age groups. Means and standard deviations are presented in Figure 1. On the child abuse scale,  $F(2, 915) = 9.60, p = .00$ , the group with peer-age victim patterns also had the lowest mean scores. For sexual insecurity,  $F(2, 915) = 6.03, p = .00$ , the group with child-age victim patterns ( $n = 585$ ) had the highest mean scores compared to the other victim-age groups. These results add to the existing literature on victim typology and shed light on the victim-offender cycle. The results suggest that youth with more complex trauma histories and mental health needs, such as those with child-age victim patterns, may need more resources and time to achieve the same treatment gains as those with fewer risk factors. Overall, AISB are not a “one size fits all” group (Newman et al., 2018). Treatment programs should be individualized to meet these needs.

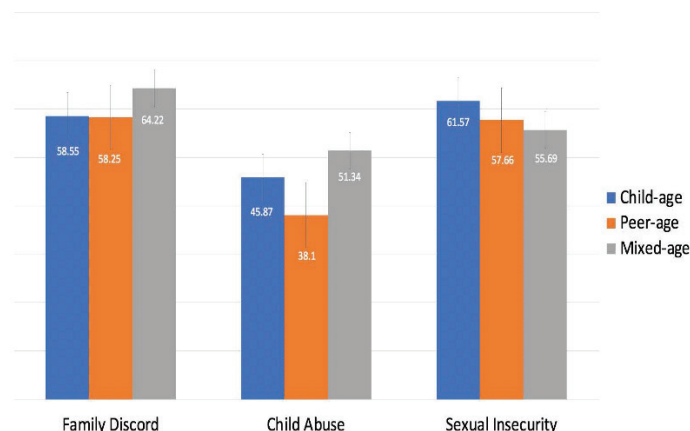


Fig. 1. Means and standard errors of the variables of

<sup>1</sup>Corresponding author: kelli.thompson@auburn.edu

interest.

### Statement of Research Advisor

Jenna's study was presented at two regional conferences, Southeastern Psychological Association and the AU Student Research Symposium, where she won awards at each. This included the Psi-Chi Regional Research Award and an Outstanding Presenter Award. Jenna will continue this victim typologies theme with new research presented at the Association for the Treatment of Sexual Abusers in Los Angeles, CA, in October.

- Kelli R. Thompson, Ph.D., College of Liberal Arts

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### Authors Biography



Jenna Wettstein is a senior-year student pursuing a B.S. degree in Psychology at Auburn University. Her research interests include improving resources for children and adolescents exposed to trauma. Jenna is very passionate about promoting the recognition of etiological factors that contribute to victimization. In addition to her work with the Department of Psychological Sciences, she is a 2022-2023 College of Human Sciences research fellow. She is using her fellowship to investigate trauma-informed care in the foster system.



Kelli R. Thompson is an Assistant Research Professor in the Department of Psychological Sciences. Her lab is funded by a more than 20-year public-public partnership with the Alabama Department of Youth Services. She has shown a particular interest in undergraduate research mentoring in recent years. In the spring 2022 semester, more than a dozen original undergraduate research projects were completed by students. Most have gone on to publish in undergrad journals such as this.



# Effects of the Acoustic Field Generated by a Single-Axial Rotor

Emily Wilson<sup>1</sup>, Lokesh Silwal<sup>2</sup>, and Vrishank Raghav<sup>3</sup>

<sup>1</sup> Undergraduate Student, Department of Aerospace Engineering, Auburn University

<sup>2</sup> Graduate Student, Department of Aerospace Engineering, Auburn University

<sup>3</sup> Assistant Professor, Department of Aerospace Engineering, Auburn University

The purpose of this study was to understand the sources of noise generated by small-scale rotors, specifically to reduce this noise in unmanned aerial vehicles (UAVs) or drones. Drones are utilized to serve a variety of different purposes. However, the noise generated by their rotor blades creates limitations for their adaptation to both private and public utilization settings [1]. Therefore, a study was conducted on the sources of this noise and how to reduce it.

The sources of sound produced from a single-axial rotor can be allocated into three categories, monopole, dipole, and quadrupole. Monopole noise, or thickness and high-speed impulsive noise, is radiated in all directions. Dipole noise is characterized as loading noise, which is caused by the changes in blade loading due to a change in rotor thrust. There is also quadrupole loading or blade-vortex interaction noise due to the aerodynamic interactions occurring between the blades in the aerodynamic field [3].

In this study, two microphones were used to measure the acoustic field on a small 15-centimeter diameter rotor. For initial testing, the rotor was clamped to the table with one microphone placed level and the second microphone placed slightly below the rotor. Further testing will include placing the rotor on a stand to attain a more accurate reading. The microphones are connected to data acquisition for both a numerical and visual representation of the acoustic field generated by the rotor. The rotor was operable at different speeds, and the blades on the rotor were interchangeable for experimentation.

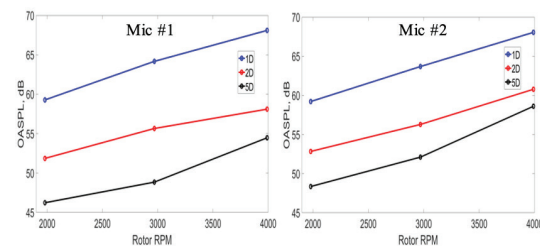
Experiments were conducted under unique conditions to attain the most accurate representation of the gen-

erated acoustic field. First, the rotor was operated at three different rotor speeds, 1980, 2970, and 3990 RPM. The varying rotor speeds represent a better understanding of the acoustic field changes. Second, the distance between the rotor center and the microphone was varied, with an experiment conducted at 1, 2, and 5 rotor diameter distances. This was done to study the drone acoustic noise propagation. The data was sampled at a frequency of 25,000 Hz.

To analyze the results, the data collected was interpreted through a series of MATLAB codes. This allowed for the calculation of the overall sound pressure level (OASPL) at each unique testing condition. The equation for this calculation is in Eq. (1).

$$OASPL = 10 \times \log \left( \int_{20}^{20000} \frac{PSD(f)df}{P_{ref}^2} \right) \quad (1)$$

It was observed that the OASPL increased in all incidences where the rotor RPM was increased. This result was expected due to the increase of dipole noise produced by a greater thrust. Also, it was observed that the OASPL decreased as measurements were taken at further distances from the rotor center. This observation was also expected due to the dissipation of monopole noise into the surrounding environment. These results can be seen below in Fig. 1.



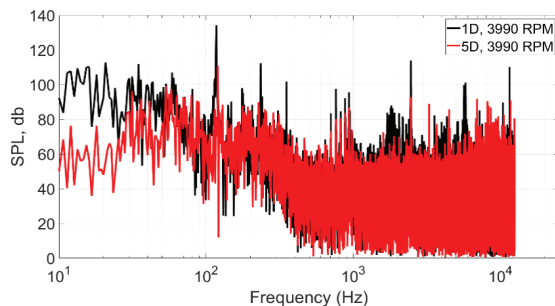
**Fig. 1.** OASPL measurements for microphones 1 & 2. A frequency spectrum was required for analyzing the

<sup>1</sup>Corresponding author: ejw0043@auburn.edu

sound pressure level at different frequencies. A fast fourier transform was used to transform the data from the time domain to the frequency domain. This conversion is done using Eq. (2). The sound pressure level (SPL) is analyzed across the audible frequency range.

$$SPL = 20 \times \log \left( \frac{P_{rms}}{P_{ref}} \right); P_{ref} = 2 \times 10^{-5} \quad (2)$$

It was observed that lower frequency sound levels were higher when measured closer to the rotor and lowered as measurements were taken further away. The monopole and dipole sound sources can be accounted for this phenomenon. It was also observed, however, that high-frequency sound levels showed similar characteristics across all distances from the rotor center. These higher frequency sounds are of concern due to the irritation and damage they can cause to human ears. This is due to the quadrupole sound source. These results can be seen in Fig. 2.



**Fig. 2.** SPL measurements in frequency domain at 1 & 5 diameter distances.

Previous studies have shown to reduce this noise, and modifications can be made to the blade design. A common way to reduce this noise is to incorporate trailing edge serrations on each blade [2]. Although there is a consensus that this will reduce the noise generated, the fluid dynamics remain unknown. To explore this solution, experiments will be conducted using both smooth trailing edge and serrated trailing edge blades to measure their effects on the acoustic field generated by the rotor. The accuracy of this data will be improved upon by including data collected from eight microphones to attain a more complete reading of the acoustic field, and by utilizing strain gauge loadcells in the rotor stand to quantify whether the rotor is performing appropriately.

## Statement of Research Advisor

With the increasing rate of adoption of drones in civilian and military applications, the noise produced by the rotors will be a significant shortcoming to their widespread adoption. This study is a starting point to identify the physical aeroacoustic sources of the noise produced by small-scale rotors. In the future, the plans are to optimize the wing planform to reduce the magnitude of noise produced.

- Vrishank Raghav, *Department of Aerospace Engineering, Samuel Ginn College of Engineering*

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## Authors Biography



Emily Wilson is a junior-year student pursuing a B.S. degree in Aerospace Engineering at Auburn University. She has worked in the Applied Fluids Research Group for two years with responsibilities for the development of experimental testing designs.



Lokesh Silwal is a Ph.D. candidate at Auburn University. He is working in Applied Fluids Research Group advised by Dr. Vrishank Raghav. He specializes in the aerodynamic interactions of next generation multi-rotor helicopters, plume-surface interactions, and experimental flow diagnostic techniques.



Vrishank Raghav is an Assistant Professor in the Department of Aerospace Engineering at Auburn University. He is the principal investigator of the Applied Fluids Research Group where the research interests are centered on the theme of unsteady fluid dynamics with applications across multiple disciplines.

# The Politics of Perception: How the White House Manipulates National Crisis Narratives

*Jordan A. Windham*<sup>1</sup>

<sup>1</sup> Undergraduate Student, Department of Political Science, Auburn University

Since its institution, the office of the President of the United States has expanded its power, prestige, and attention with the sitting president coming to symbolize the nation (Pika, Maltese, and Rudalevige, 2019). The sitting president has also become perceived as responsible for the state of the nation (Pika, Maltese, and Rudalevige, 2019). With this responsibility, a presidential administration's ability to maintain a positive perception of its performance is key as public approval is an indicator of both its political capital (Christenson and Kriner, 2020) and its reelection prospects (Lewis-Beck and Rice, 1982). With public attention focused on the White House, an administration's success in shaping how the public perceives it can make or break its future, particularly during crises when people look to the president for safety and reassurance (Pika, Maltese, and Rudalevige, 2019). We also considered partisanship, which ensures a stable attachment to a party and shapes how individuals respond to information (Campbell, et al., 1960), and polarization, the increasing divergence in partisan ideology (Bartels, 2002), as significant influences on public approval.

We analyzed the Trump administration's attempts to shape public perceptions of the COVID-19 pandemic, tracking the administration's statements from the virus' public emergence in January 2020 to the presidential election of November 2020. We then compared the Trump administration's narrative strategies to those of the Reagan administration during the Iran-Contra Affair to better understand how different narrative approaches and audiences shape outcomes related to how presidential administrations manage (or not) a crisis of national significance.

President Trump's use of Twitter to communicate directly with the US public offered a unique and unprecedented window into a president's attempts to influence

public perception of a national crisis. In this paper, we investigate the Trump administration's narrative manipulation efforts primarily through President Trump's tweets, which are supplemented with relevant public statements to contextualize the Trump administration's strategies. Tweets were filtered for the keyword's "coronavirus", "COVID", "COVID-19", and "pandemic" to maintain content relevance.

We used public approval ratings to assess the administration's success in shaping public opinion, as well as the administration's situation as they cast their narratives. As we only needed general data trends, we used public approval data from Gallup.com, which provided the Trump administration's approval ratings in approximately 2-week intervals. We have selected and analyzed representative tweets from each public approval data window, which were typically the first two weeks and the last two weeks of each month. Tweets that did not fall inside a defined public approval window are grouped with the next data set. We classified the Trump administration's approaches into four categories: (1) downplaying the virus and reassuring the public, (2) diverting attention, (3) blaming others, and (4) the no spin zone.

As the virus emerged in January, Trump leaned into the first three narratives, dismissing and distracting from the virus. When WHO declared a pandemic in mid-March, the No Spin Zone narrative was used in an attempt to unite the public behind his administration with an honest, nonpartisan approach that took the virus seriously. Trump also continued to use his other narratives, with the only adaptation being that he refrained from casting Democrats as the opposition but tried to unite them against "common enemies" like WHO and China.

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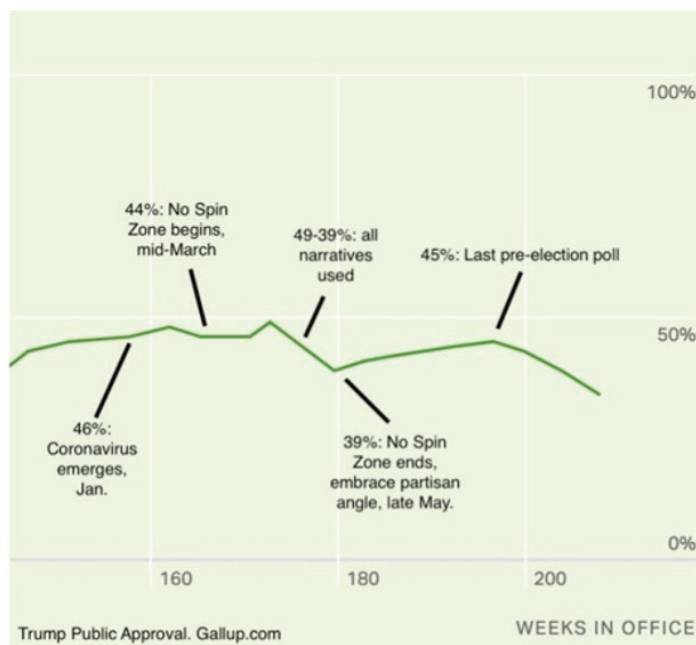
<sup>1</sup>Corresponding author: jaw0127@auburn.edu



- Downplaying and reassuring
  - "It's one person coming in from China. We have it under control. It's going to be just fine," (CNBC, 2020).
- Diverting attention
  - "The Fake News Media refuses to discuss how good the Economy and Stock Market, including JOBS under the Trump Administration, are doing. We will soon be in RECORD TERRITORY. All they want to discuss is COVID 19, where they won't say it, but we beat the Dems all day long, also!!!" (Trump Twitter Archive).
- Blaming others
  - "First of all, we have done a great job. We've gotten the governors everything they needed. They didn't do their job. Many of them didn't and some of them did. ... But the governors do it. We gave them massive amounts of material" (Sanford, 2020).
- No Spin Zone
  - "It's absolutely critical for the American people to follow the guidelines for the next 30 days. It's a matter of life and death," (WION 2020).

**Fig. 1** Examples of Tweets exemplifying narrative categories.

This non-partisan mixed strategy ended in late May, when, after weeks of stagnant approval, Trump's approval dropped 10 points (Gallup, 2021). More tellingly, Trump's Republican approval dropped 7 points to 85 percent—the only time during the pandemic that his support among Republicans was under 90 percent (Gallup, 2021). After stagnant approval and copartisan wavering, Trump returned to partisan narratives and his support among Republicans recovered. For the rest of the time before the election, Trump used all four narratives as his approval recovered (Gallup, 2021).



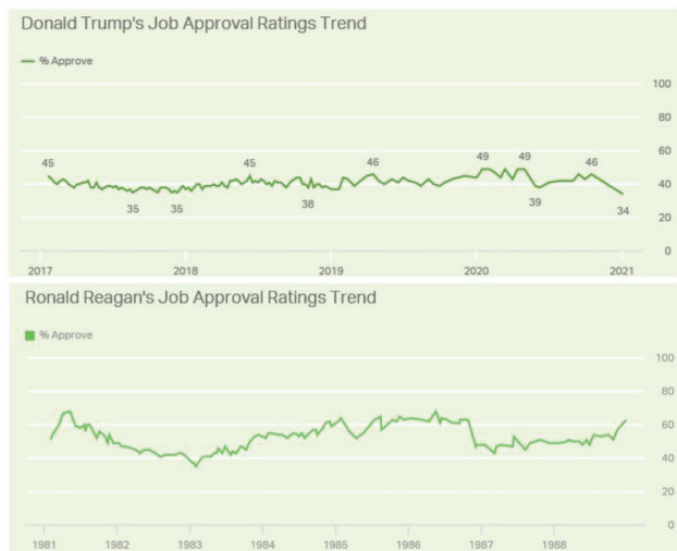
**Fig. 2** Timeline of Trump administration's public approval (Gallup, 2021).

Reagan's approach to the Iran-Contra Affair differed distinctly. The Iran-Contra affair was when the Reagan administration secretly sold arms to Iran and used the

proceeds to illegally fund the Contras, a Nicaraguan paramilitary organization. Initially, Reagan denied the scandal's existence. When the story came out and this narrative was no longer viable, Reagan, while insisting on his personal innocence, took responsibility, apologized, and publicly supported multiple independent investigations. After this narrative, Reagan's public approval eventually made a full recovery. Reagan stuck to a single narrative despite the risk of committing to a losing one. We found this a risk worth taking when compared to Trump's alternative: inconsistently employing multiple disparate narratives to try to mobilize public opinion and retreating to his base when these attempts failed.

It is possible that polarization would have kept Trump's approval at the same levels regardless. Yet, it is still worth exploring the strategic differences in presidents' strategies for what this can tell us about presidents' narrative responses to crises moving forward. The polarized environment also provides context for Reagan and Trump's approaches and success. Reagan consistently and Trump for a time used non-partisan narratives. While Reagan made a full recovery, Trump was unable to unite even half of the public. We found that our now hyper-polarized electorate made it easier for Trump to shape the perceptions of his co-partisans, as evidenced by his consistently high support from Republicans.

Yet, the same polarization that united Republicans made it difficult for Trump to find a message that he could use to unite Democrats and Republicans. After failing to realize this support and even losing support from his own party, Trump fell back on partisan narratives and consolidated support among his base. This supports the idea that, at a minimum, narrative manipulation will be more difficult in an era of polarization. This also lends support to the idea that, in hyper-polarized political environments, presidents will focus their narrative manipulation attempts on consolidating and mobilizing their party's half of the electorate, rather than risk this support by reaching out to members of the opposition party.



**Fig. 3.** Comparison of the Trump and Reagan administrations' public approval trends (Gallup, 2021).

### Statement of Research Advisor

The results of this project add to the growing literature and collective understanding of how leaders, most notably US presidents, strategically wield communication to attempt to shape, control, and/or manipulate the narrative regarding a crisis of national significance. In comparing and contrasting the cases of the Reagan Administration's response to the Iran-Contras Scandal and the Trump Administration's response to the Covid-19 pandemic, we now have a clearer understanding of the communication tactics and strategies that are used by American leaders and which tend to be most effective at shaping or manipulating a national crisis narrative and which ultimately have proven ineffective.

-Matthew Clary, College of Liberal Arts, Political Science

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[5] Pika, Joseph A., John Anthony Maltese, and Andrew Rudalevige. *The Politics of the Presidency*. CQ Press, 2019. **Book**

### Authors Biography



Jordan Windham graduated summa cum laude in spring 2022 with a B.A. in Political Science at Auburn University. Her undergraduate research has focused on public opinion and electoral politics. She is a 2022 Teach For America corps member teaching special education in Idaho.

# Posttraumatic Stress Symptoms in Adolescent Males Adjudicated for Illegal Sexual Behavior

*Abigail P. Winskowitz<sup>1</sup> and Kelli R. Thompson<sup>2</sup>*

<sup>1</sup>Undergraduate Student, Department of Psychological Sciences, Auburn University

<sup>2</sup>Assistant Research Professor, Department of Psychological Sciences, Auburn University

Trauma symptoms can vary between individuals for a multitude of reasons. Depending on the levels of exposure, types of trauma, and even the individual's coping mechanism, children and adolescents will interpret and respond to trauma in unique ways (Cohen, Mannarino, Deblinger 2016). Adolescents faced with trauma symptoms experience a vast change in personal worldview, as well as life trajectory if left untreated (Cohen, Mannarino, Deblinger 2016). Trauma symptoms can be triggered for a variety of reasons but often result in disruptions to caregiver attachment, as well as to therapeutic processes (Harrelson et al., 2017). In a recent meta-analysis, adolescents adjudicated with illegal sexual behavior (AISB) experienced higher rates of victimization, with 46% reporting some history of sexual abuse, compared to 16% of non-sex offenders (Seto, Lalumiere 2010).

Trauma exposure has been linked with the high rates of posttraumatic stress symptoms, indicating a need for trauma-informed care when working with this population (Copeland, Keeler, & Angold, 2007). Additionally, AISB had a higher prevalence of psychopathology, specifically symptoms of anxiety, with social anxiety being significantly higher, when compared to a non-sex offending population (Seto & Lalumiere, 2010). When screened for symptoms of traumatic stress disorders, nearly half a sample of AISB indicated at least one item related to suicidal ideation (Everhart Newman et al., 2018). Traumatic events are generally linked more with internalizing psychopathologies, such as anxiety and depressive disorders (Copeland, Keeler, & Angold, 2007). However, some links to externalizing symptoms have been found with some victimized groups experiencing more behavior problem scores (Tricket, 1998).

The current study examined differences in posttraumatic stress symptoms between a sample of AISB compared

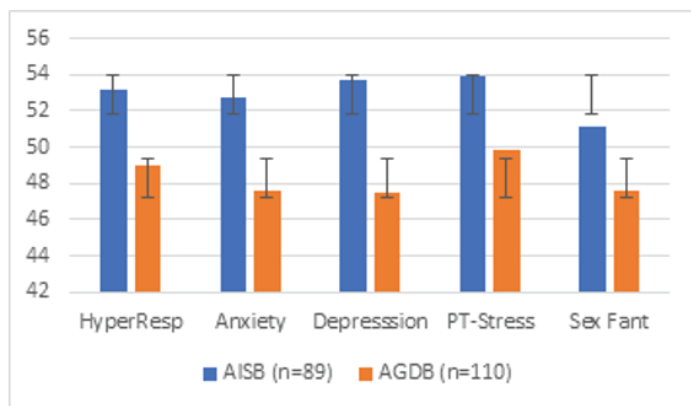
to a sample of adolescents adjudicated for general delinquent behavior (AGDB). The participants (n = 199) were juveniles receiving court-mandated services at a secure residential facility. The AISB group was younger (mean age = 15.81 and 8th grade modal level) than the AGDB group (mean age = 17.20 and 9th grade modal level). The AISB was more likely to have witnessed domestic violence (31%) compared to the AGDB group (22%). The Trauma Symptom Checklist for Children (TSCC; Briere, 1996) was used to measure posttraumatic stress symptoms during a pre-treatment screener evaluation. A one-way ANOVA was used to test for significant mean differences on the TSCC scales across the grouping variable AISB versus AGDB.

Results, as seen in Table 1, indicated that AISB displayed more internalizing symptoms of posttraumatic stress disorder, such as depression, anxiety, and hyperresponsivity. Additionally, AISB scored higher on scales of sexual fantasy related to trauma symptoms suggesting some type of defense mechanism unique to this group. Means and standard errors are displayed in Figure 1. Interestingly, while not statistically significant, the AGDB group scored higher on several measures of sexual concern and stress. These results highlight the importance of understanding and screening for the different ways trauma symptoms show up in individuals. Particularly for those that work with justice-involved youth, these differences can be used to model different treatment approaches for individual clients.

<sup>2</sup>Corresponding author: [kelli.thompson@auburn.edu](mailto:kelli.thompson@auburn.edu)

**Table 1.** Results of one-way ANOVA analyses.

TSCC Subscales	<i>F</i> (1, 198)	<i>p</i> -value
Under Responsivity	0.78	0.38
Hyper Responsivity*	5.84	0.02
Anxiety*	12.05	0.00
Depression*	14.41	0.00
Anger	0.27	0.60
Post-Traumatic Stress*	6.04	0.02
Dissociation	2.71	0.10
Overt Dissociation	2.79	0.10
Sexual Fantasy*	6.06	0.02
Sexual Concerns	2.24	0.14
Sexual Preoccupation	1.92	0.17
Sexual Distress	0.74	0.39

**Fig 1.** Means and standard errors of the main study variables.

### Statement of Research Advisor

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- Kelli R. Thompson, *Juvenile Delinquency Lab*

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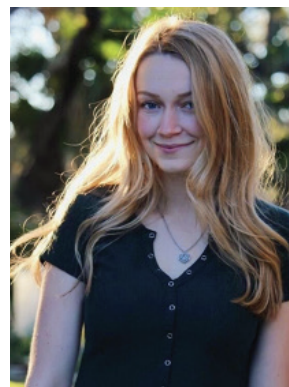
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### Authors Biography



Abigail Winskowicz is a senior-year student pursuing a B.A in Psychology at Auburn University. She hopes to continue her education in the field of psychology in graduate school. She is currently a research assistant in The Juvenile Delinquency Lab at Auburn University.





Kelli R. Thompson is an Assistant Research Professor in the Department of Psychological Sciences. Her research interests include the assessment and treatment of adolescents with illegal sexual behavior. She teaches a comprehensive healthy sexuality group at the Mt. Meigs facility. Undergraduate research mentoring remains a personal and professional focus.



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Office of Undergraduate Research  
206 Cater Hall  
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