

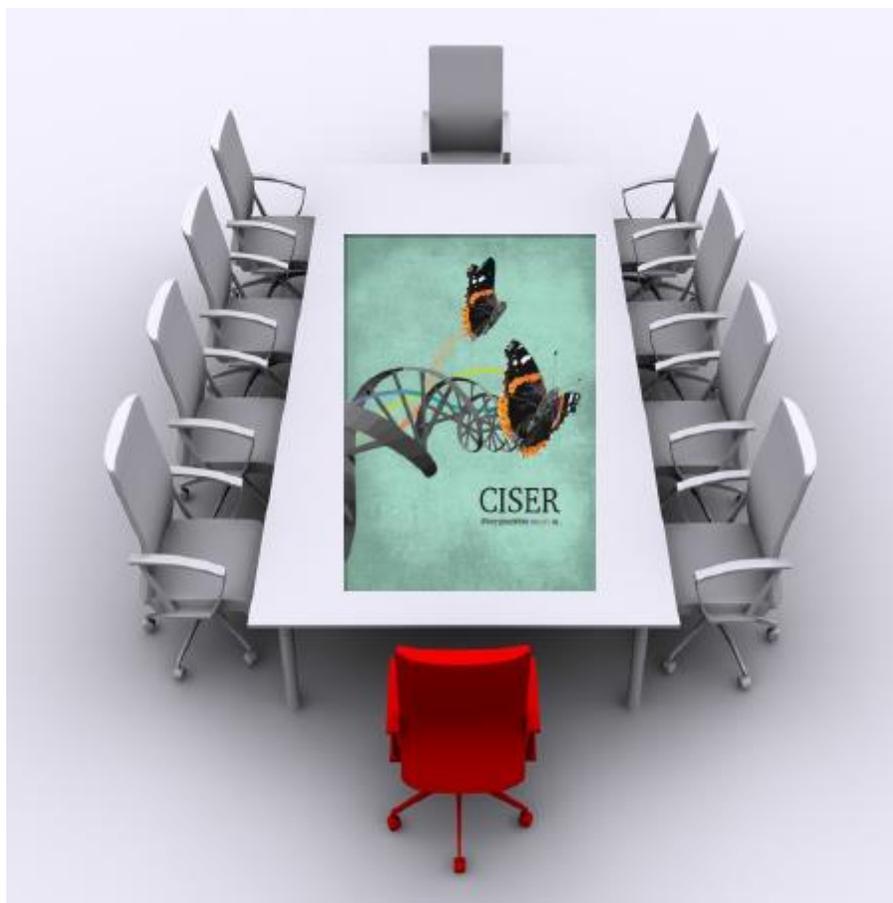


TEXAS TECH UNIVERSITY

CISER: Center *for the* Integration
of STEM Education & Research

2015

Scholar Research Forum



October 17, 2015
Texas Tech University



TEXAS TECH UNIVERSITY

CISER: Center for the Integration
of STEM Education & Research

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Program

Special thanks to Chris and Vickie Rodriguez, and Megan Sovine, for their generous funding of the Christopher Rodriguez Research Presentation Awards. Their support has made the presentation of these awards possible for the first time at a Scholar Research Forum. We deeply appreciate their generosity.

Chemistry 9:00 – 9:25

Synthesis of a [2]Rotaxane via Dynamic Ring-Chain Equilibration

Holden R. Fried

Mentor: Dr. Michael Mayer

Discussant: Mai Dinh

Novel Processing Techniques for Post-Nuclear Detonation Debris Using Ammonium Bifluoride

Belinda Pacheco¹, Chris Leibman¹, Michael Rearick², Kirk Weisbrod³

Los Alamos National Lab: ¹C-CDE, ²EES-14, ³AET-5

Mentor: Dr. Louisa Hope-Weeks

Discussant: Josh Willms

Optical Spectral Imaging of an Ambient Pressure Plasma Jet via Compressive Sensing

John Usala

Mentor: Dr. Gerardo Gamez

Discussant: Jorge Franco

Biological Sciences and Ecology 9:25-10:10

Reevaluating Phylogenetic Relationships Within the *Peromyscus boylii* Group

Elise Wagley

Mentor: Dr. Robert Bradley

Discussant: Dakota Kilcrease

Understanding Species Limits of *Peromyscus mexicanus* Group Using A Genetic Approach

María Núñez

Mentor: Dr. Robert Bradley

Discussant: Carlos J. Garcia

Using Genomic and Bioinformatics Techniques to Determine the Origin and Phylogenetic Distribution of the Zonadhesin Gene in Rodentia

Whitney Watson

Mentor: Dr. Robert D. Bradley

Discussant: Allie C. Smith

Phylogeny of Bats of Genus *Monophyllus*: A Study of Genetic and Morphological Divergence Based On Mitochondrial Cytochrome-b Gene

Marilyn Mathew

Mentor: Dr. Robert Baker

Discussant: Josh Willms

Impact of Human Development on Reptile and Amphibian Biodiversity: Development of a Project

Katherine Crocco

Mentor: Dr. Lou Densmore

Discussant: David Campos

Break 10:10-10:20

Biological Sciences and Medicine 10:20-11:15

Surgical Wound Model

Adam Tsen

Mentor: Dr. Abdul Hamood

Discussant: Timothy Salinas

Vitamin D Regulates Cholesterol Metabolism and Breast Cancer Cell Proliferation

Christopher Ponce

Mentor: Dr. Shaikh Rahman

Discussant: Rachel Aybar

PEDF Induces the Migration, Differentiation and Phagocytic Activity of Macrophages

Dalia Martinez-Marin

Mentor: Dr. Stephanie Filleur

Discussant: Stacy Galvan

CD47 Down-Regulation by PEDF: A New Means of Inducing Tumor Cell Phagocytosis *in vitro*

Nitish Mittal- Urology

Mentor- Dr. Stephanie Filleur

Discussant: Brooke Walterscheid

Terminally Differentiated Sertoli Cells Reinitiate Proliferation After Loss of Cell-Cell Contact

Rachel Dziuk

Mentors: Jannette M. Dufour and Gurvinder Kaur

Discussant: Gregory Smith

Functional Amyloids: A Link between Yeast Reproduction and Mammalian Fertilization

Gage Rowden

Mentor: Dr. Gail Cornwall

Discussant: David Campos

Physics and Technology 11:15-11:40

MALDI-TOF as a New Tool for Quantification of Polyamines in Plants

Aicha Fokar
Mentor: Dr. Masoud Zabet
Discussant: Allie C. Smith

Searching for Low Mass Dark Matter Halo Counterparts to Ultra-Compact High-Velocity Clouds in the Local Group

Maksym Zhelyeznyakov
Mentors: Dr. David Sand and Dr. Elisa Toloba
Discussant: Alex Cardona

Operation Counter Attack: An Offensive Approach to Cybersecurity

Joshua Hernandez
Mentors: Dr. Venkata N. Inukollu and Dr. Joseph E. Urban
Discussant: Paden Ortega

Educational Sciences 11:40-11:55

Evaluating the Television Show Daniel Tiger’s Neighborhood as a Video-based Model to Teach Social Skills to Children with Autism

Marisol C. Alonzo
Mentor: Dr. Wesley H. Dotson
Discussant: Brandon Palomo

Engagement on Daniel Tiger’s Neighborhood Involving Kids with Autism

Amanda Lund
Mentor: Dr. Wesley Dotson
Discussant: Josh Willms

Noon -

Lunch: Special Awards and Presentations

- Fish Missions Update
- Allie Clinton Scholarship
- Christopher Rodriguez Presentation Awards

Synthesis of a [2]Rotaxane via Dynamic Ring-Chain Equilibration

Holden R. Fried

Mentor: Dr. Michael Mayer

Mechanically interlocked compounds such as rotaxanes and catenanes are of significant synthetic interest in pure chemistry as well as in fields such as polymer chemistry/materials science. Polymers of these compounds are also of great interest, owing to the fact that their unique molecular architecture likely possesses a number of interesting properties not found in traditional covalent-only polymers like polyethylene. Catenanes are compounds that consist of two or more interconnected macrocyclic rings that are held together by non-covalent interactions, similar to two links in a chain link. Likewise, a polycatenane would be analogous to a full chain link. Rotaxanes, as shown on the right in the equilibrium below, consist of a macrocyclic ring surrounding an axle, which resembles a dumbbell in shape. Flanking each end of the axle are two large stoppers (shown in red) that act to prevent the macrocycle from

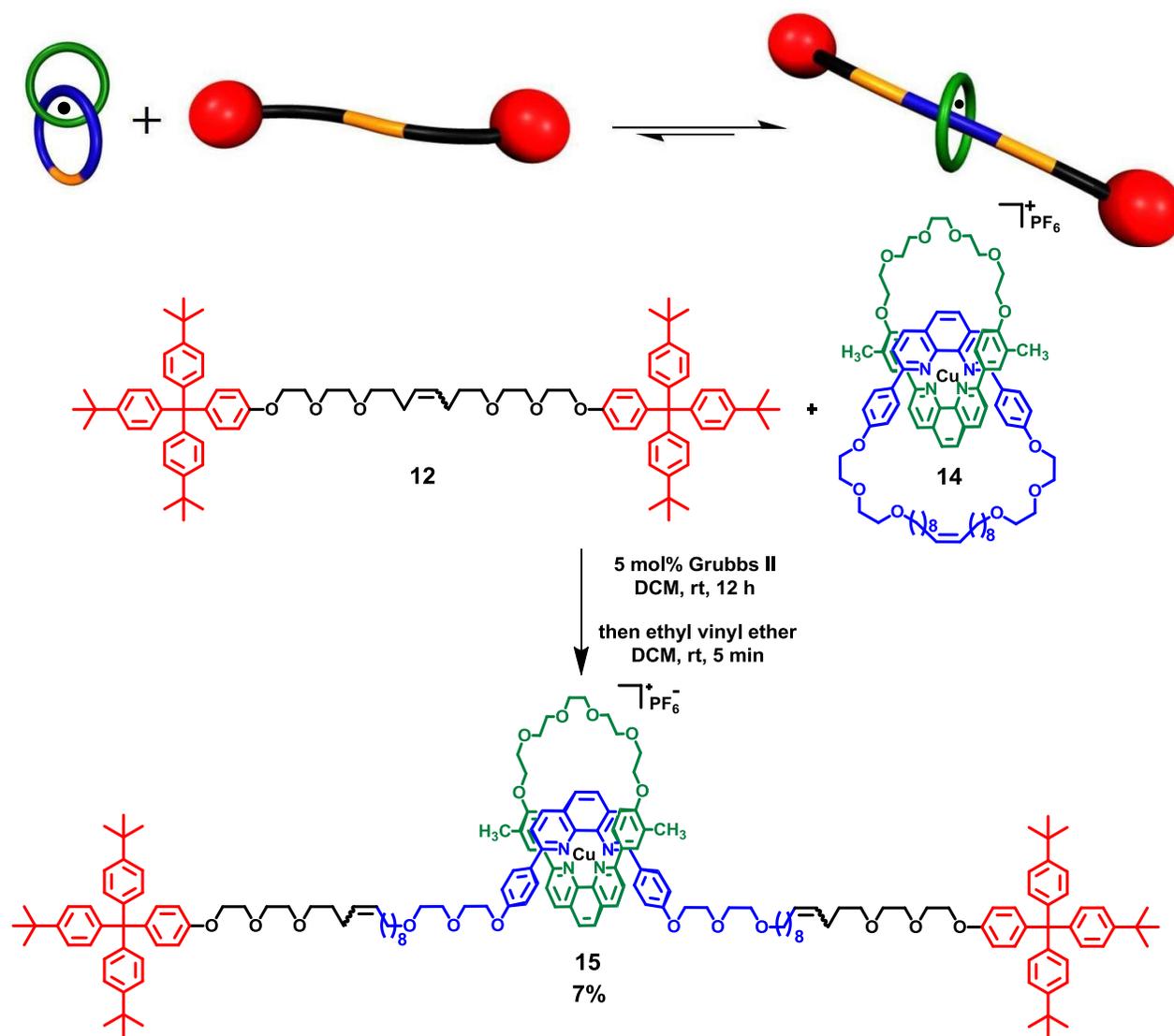


Fig 1—Ball and stick and molecular structures for synthesis of [2]Rotaxane via Dynamic Ring-Chain Equilibration (Colors in ball and stick model are representative of the molecular model)

dissociating from the axle. Current research in our lab involves the creation of a [2]Rotaxane via dynamic ring-chain equilibration, a novel approach to the creation of rotaxanes.

In synthesis of [2]Rotaxanes via dynamic ring-chain equilibration, the approach developed by our lab, we start by creating 2 macrocycles (compound 7 and compound 4). Compound 7 is synthesized as a macrocycle from the beginning. Compound 4 is synthesized first as a non-cyclic diolefin (the olefin group is also known as an alkene) intermediate. We then run a reaction in which we add in tetrakis(acetonitrile)copper(I) hexafluorophosphate ($[\text{Cu}(\text{CH}_3\text{CN})_4]\text{PF}_6$) in dichloromethane (DCM) at room temperature for 2 hours in order to coordinate the two phenanthroline groups of the macrocycles in order to bring them together for catenation. After coordination, to close the diolefin-containing ring and thus form the catenane, we use Grubbs II catalyst to perform a ring closing metathesis (RCM) reaction whereby the two terminal olefin moieties get converted to one internal olefin upon closing the ring. We now have a catenane with an internal olefin on one ring. The black dot in the middle of the catenane in figure 1 represents the copper ion that coordinates the phenanthroline groups of the macrocycles. From here, we add our catenane and di-stoppered olefin (axle)

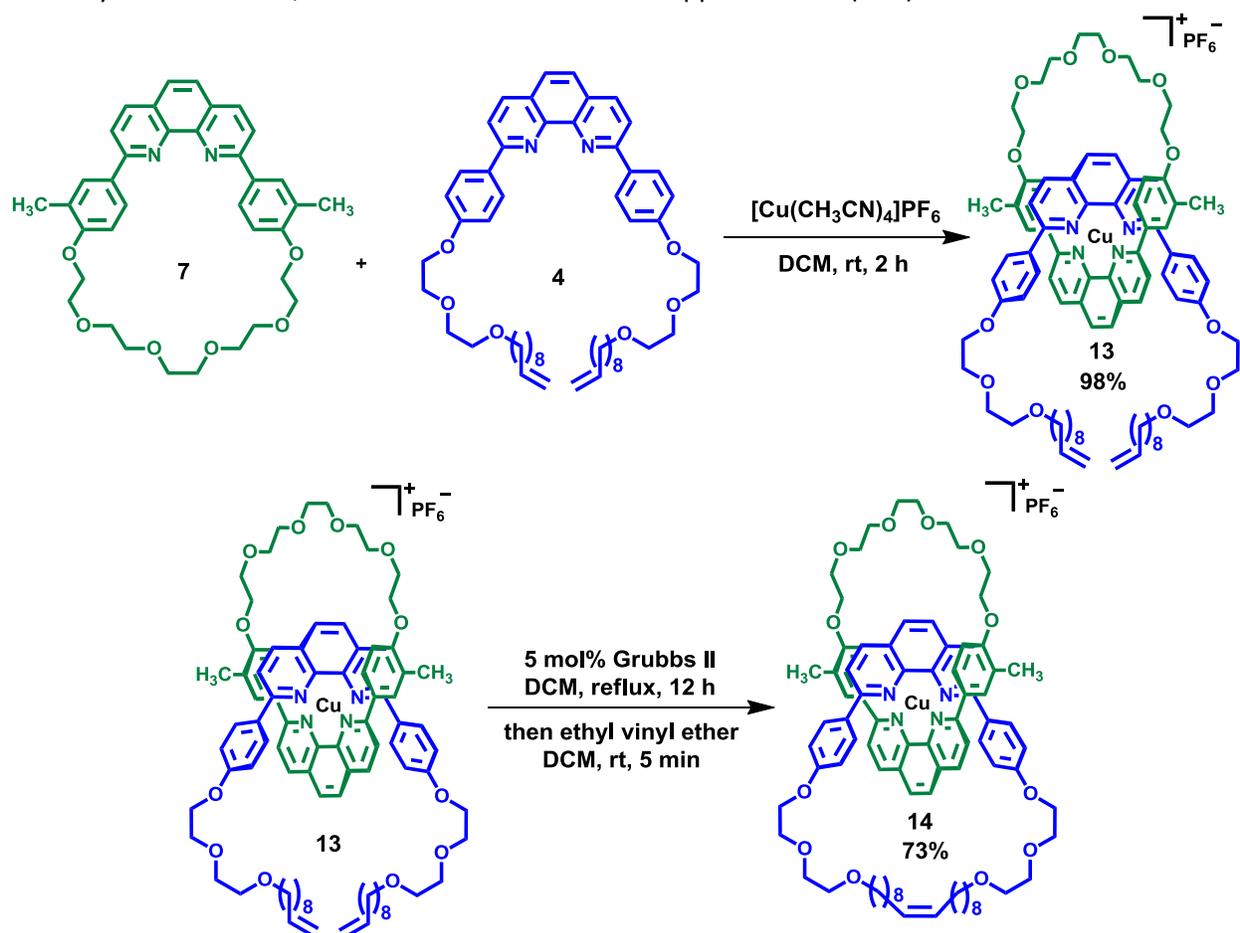
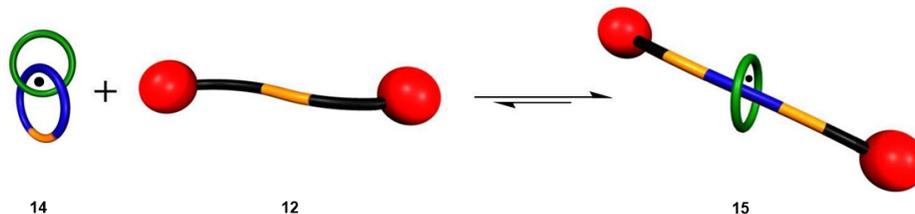


Fig 2—Formation of the Catenane Intermediate

to DCM with 5 mol % Grubbs II catalyst and allow the reaction to run at room temperature for 12 hours. The reaction is then quenched by “killing” the catalyst by addition of ethyl vinyl ether.



entry	14 (equiv.)	12 (equiv.)	cat. (mol %)	catalyst	temp. (°C)	time (h)	conc. (mM)	solvent	yield ^a
1	1	1	5	G-II ^b	rt	12	10	DCM	7
2	1	1	5	G-II	rt	24	10	DCM	13
3	1	1	5	G-II	40	24	10	DCM	23
4	1	1	5	GH-II ^c	40	24	10	DCM	24
5	1	1	5	G-II	58	24	10	DCE	25
6	1	1	5	G-II	40	24	1	DCM	11
7	1	1	5	G-II	40	24	50	DCM	26
8	1	2.5	5	G-II	40	24	10	DCM	73
9	1	5	5	G-II	40	24	10	DCM	81
10	1	10	5	G-II	40	24	10	DCM	88

^aIsolated yields. ^bG-II is Grubbs' second generation catalyst. ^cGH-II is Grubbs-Hoveyda second generation catalyst.

Figure 3—Optimization of [2]Rotaxane Formation

This final reaction initially provided extremely low yields (~7%) until optimization studies were done, after which our lab obtained an excellent isolated yield of 88% for the reaction as shown in Figure 3.

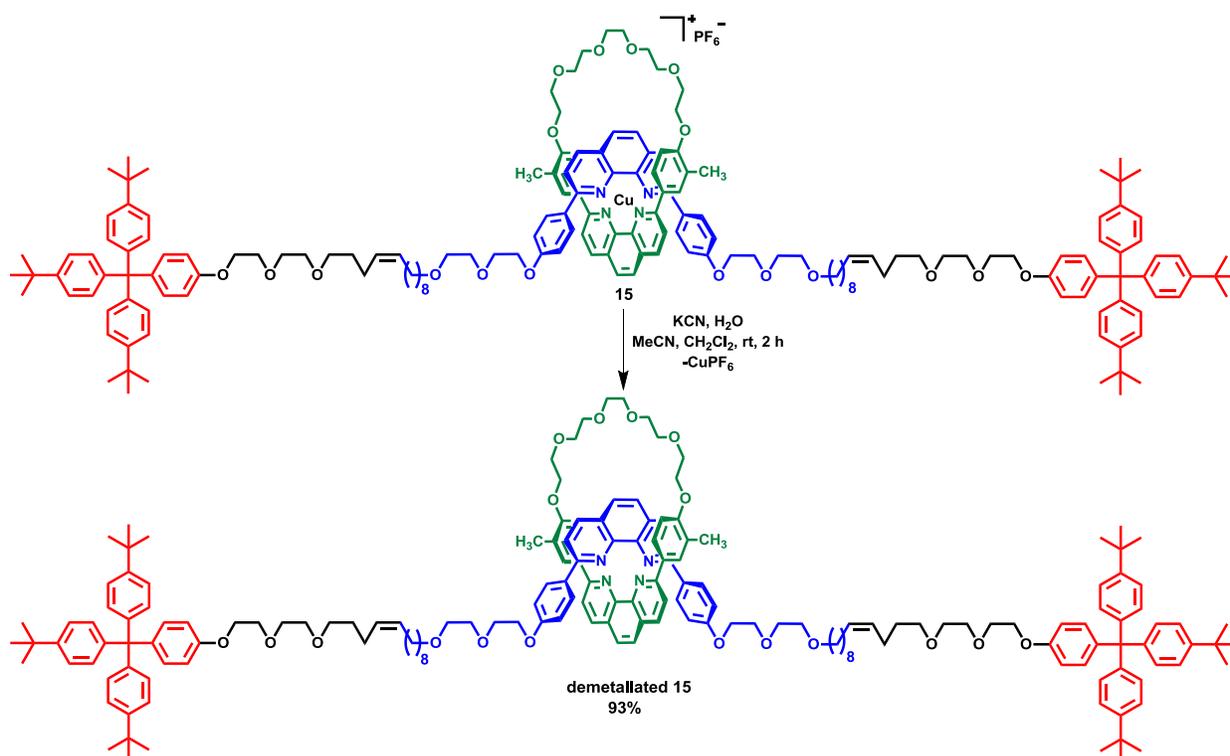


Figure 4—Demetalation of the [2]Rotaxane

Following formation of the [2]Rotaxane, the copper ion can be removed and the macrocycle will remain on the di-stoppered olefin due to the bulky stoppered end groups that prevent dissociation of the macrocycle. We call the removal of the copper ion demetalation, and achieve this reaction by using potassium cyanide among other reagents. After demetalation, the macrocycle is free to move along the axle without dissociating from it. Because of this property, it is theorized that rotaxanes can act as molecular machines, particularly as a molecular switch. This application would likely utilize two (or more) different functionalities on the axle that can switch in response to certain stimuli, such as changes in acidity, light, etc.

Further studies are needed to assess these applications; the primary focus of this research is solely to outline an effective synthetic technique to create [2]Rotaxanes.

Holden Fried

I am a senior biochemistry major working in organic chemistry. I plan to transfer to organic chemistry for graduate school, but I would also be interested in working for a lab that integrates both organic chemistry and biochemistry. Although I am a senior, I am taking an extra year, namely because of the large amount of coursework from my three minors: biology, music, and math. I have been in my current lab, working for Dr. Mayer studying entangled polymers since the fall of 2014, and plan to stay there until I graduate. In my spare time I like to play piano, compose music, read, and learn a variety of different things.

Novel Processing Techniques for Post-Nuclear Detonation Debris Using Ammonium Bifluoride

Belinda Pacheco¹, Chris Leibman¹, Michael Rearick², Kirk Weisbrod³

Los Alamos National Lab: ¹C-CDE, ²EES-14, ³AET-5

LA-UR-15-25873

Mentor: Dr. Louisa Hope-Weeks

Analysis of post-detonation nuclear debris is of critical importance for the elucidation of nuclear device design and origin. Existing methods for sample preparation material use large quantities of concentrated mineral acid fluxes and are lengthy (6 to 72+ hours). A novel approach being investigated for nuclear debris processing utilizes solid ammonium bifluoride (NH₄)HF₂ for the dissolution of fused glass samples containing actinides and fission products. In this study, various sample digestion procedures that all employ ammonium bifluoride (ABF), were evaluated for efficiency, ease of use, and safety.

Previous thermodynamic modeling conducted by our group was applied for the development, exploration, and comparison of methods for digesting fused glass samples with ABF. Using OLI Systems Software, we were able to demonstrate that a majority of the metal oxides in the fused glasses will form fluoride or ammonium fluoride complexes when treated with ABF. Furthermore, ABF reacts with the major component of glass, SiO₂, to produce SiF₄ gas. This means that at least 50% of the glassy sample will evaporate into a gaseous form rather than being incorporated in aqueous waste. Similarly, our modeling established optimal experimental parameters for the three methods under currently under investigation: microwave assisted digestion, conventional oven dissolution, and hot plate fluxing.

Experiments using National Institute of Standards and Technology glass standard reference materials (SRMs) have been concluded for both conventional oven dissolution and hot plate fluxing. The products of these procedures have been characterized and quantified by XRD and ICP-MS respectively. Overall, ABF digestion with subsequent 2-3M nitric acid aliquots was successful at dissolving and recovering a majority of the alkali and alkaline earth metals, rare earth metals, lanthanides, and actinides present in the SRMs used. Additionally, the conventional oven method appeared to be the most versatile and efficacious method explored producing near quantitative results as early as two hours into the digestion period. Relatively lower elemental recoveries were observed for the hot plate method, though the utilization of a copper well plate helped improve results. Furthermore, preliminary experiments have been conducted using an Ethos EZ microwave which, under increased temperature and pressure, would allow for the digestion SRM glass in 1 hour or less. Several mechanical obstacles are currently being addressed to successfully implement this method for comparison.

By using solid ammonium bifluoride we have been able to significantly improve the efficiency of glassy digestion by reducing sample preparation time by more than 60% in comparison to conventional techniques. Similarly, we have circumvented the need for highly concentrated mineral acids by using solid ABF which considerably increases the safety and simplicity of sample preparation procedures. In all, this approach would allow for more efficient sample processing for the purposes of rapid actinide isotope ratio analysis and nuclear forensics.

Belinda Pacheco

My name is Belinda Pacheco and I am a senior chemistry major with plans to graduate in May 2016. I was born and raised in the small town of Muleshoe, TX (1 hour Northwest of Lubbock) which has a large population of approximately 5,000 individuals. My passion and knack for chemistry developed very early on in my high school career. Having a Texas Tech alumni as my mentor and chemistry teacher I was motivated to attend Tech and pursue my aspirations of a research centered career. At Texas Tech I have been able to develop and evolve as a person and as a researcher with fantastic opportunities this institution has given me. As a PRISM Scholar, I was first introduced to undergraduate research which served to propel me into my first and ongoing internship working on analytical chemistry projects at Los Alamos National Lab. Now, as an Undergraduate Research Scholar and a member of the Scholars' Service Organization, I have been paired with a great mentor in the Department of Chemistry and Biochemistry where I am currently working on producing new photocatalytic materials via sol-gel synthesis. In the coming years I aim to attend graduate school on a Ph.D. track with the ultimate goal of specializing in inorganic or nuclear chemistry to be applied to national security.

	Mineral Acid Method	Alkali Fusion Method	Proposed Method
Reagents Used	90% HNO ₃ , 70% HClO ₄ , 50% HF	Alkali fluxes	ABF
Reagent amount/ Sample size	Liters/10g	Varies/<0.5g	40g/10g
Temperature	180-200°C	1200°C	120°C
Processing Time	1-3+ days	6-12 hours	< 2 hours

Figure 1: Comparison of established techniques and the ABF method.¹

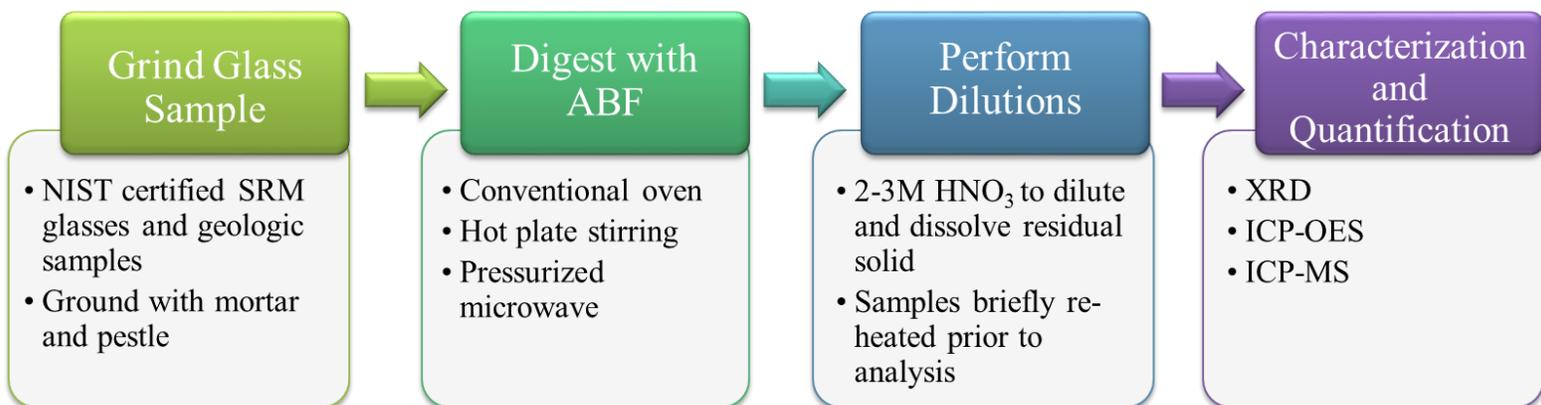


Figure 2: Generalized method taken for all ABF digestions



Figure 3: Final powdered product from the reaction of ABF and SRM glass which would subsequently undergo dissolution in 2-3M nitric acid for elemental analysis

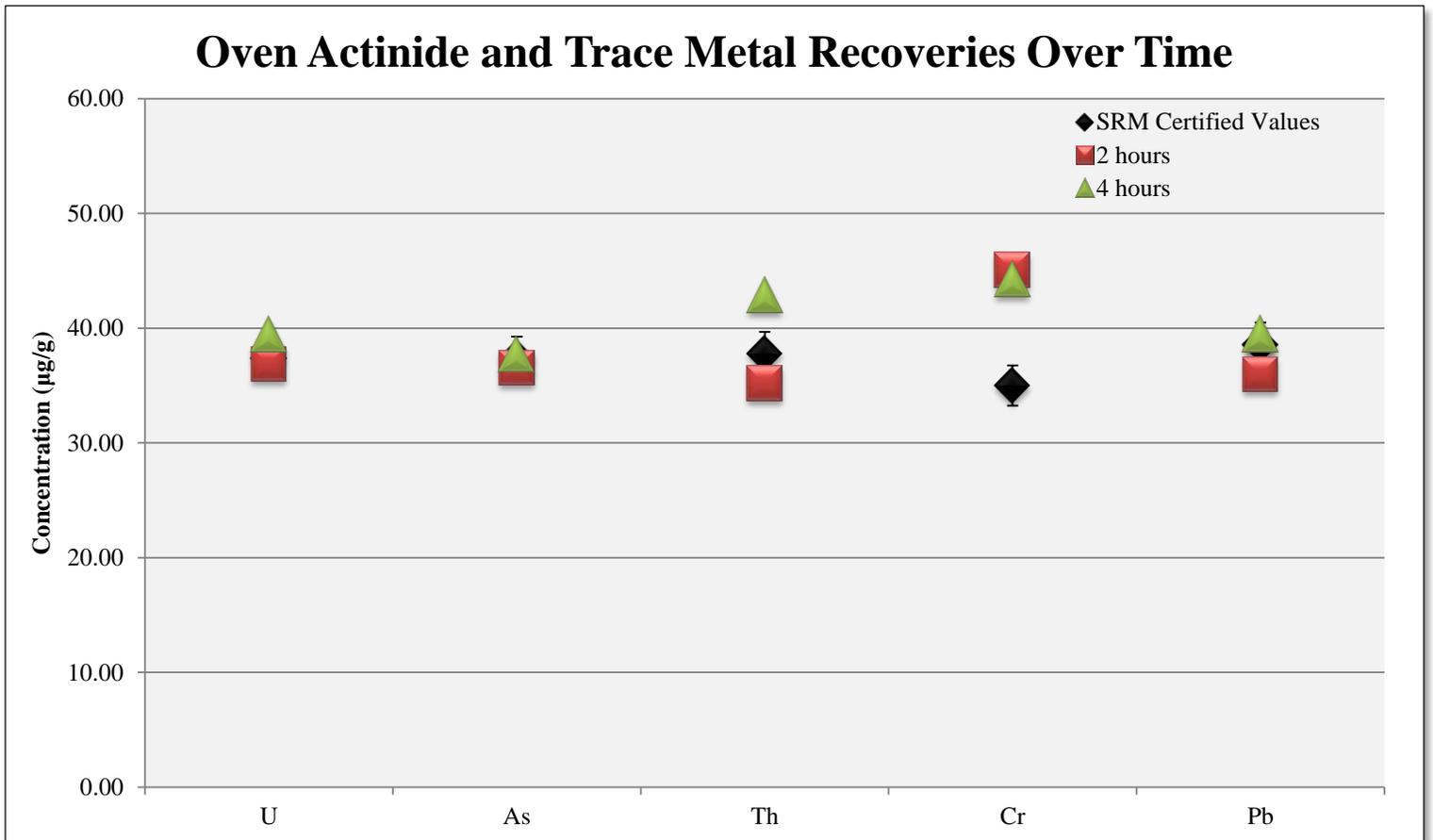


Figure 4: Recoveries from SRM 612 oven digestions over 2 hour intervals. Data points after 4 hours were excluded from this figure but were similar to that of 4 hours.

Optical Spectral Imaging of an Ambient Pressure Plasma Jet via Compressive Sensing

John Usala

Mentor: Dr. Gerardo Gamez

Optical spectral imaging is the process of obtaining a picture of an object along with its different light components at each spatial position. The spectral image allows one to get information about the chemical composition of such object. As such, spectral imaging is important for agricultural, health, food processing and materials characterization applications. In this study, spectral imaging is performed on an ambient pressure plasma jet (APPJ). APPJs are used in a variety of fields, from chemical analysis to materials processing to medicine. Nevertheless, their underlying mechanisms are still not completely understood. Herein, compressive sensing (CS) spectral imaging of APPJs will be used to better understand their fundamental chemical processes.

Spectral imaging is currently achieved with expensive array detectors, most commonly Charge Couple Devices (CCDs). Further, current spectral imaging acquisition techniques require large sets of stored data that pose a memory burden, and the resolution of the acquired image is limited by the number of samples that need to be taken. This research project circumvents these problems by utilizing a relatively new acquisition technique called compressive sensing. A proper understanding of CS necessitates a grasp of traditional imaging.

Traditional imaging (like the cameras on our phones) focuses the object as an image onto an array of pixels. The response of all the pixels is electronically transmitted, compressed, and then reconstructed to give an image. This works well, but in order to increase resolution one must have both, more and smaller pixels. This is expensive. Further, as each pixel's response must be recorded, a massive amount of memory is needed to store each value. In order to make this computationally feasible, the recorded signal, after acquisition, must be compressed. Familiar compression files include JPEG, TIFF, BMP, GIF, along with many others. This compression process uses a mathematical transform to store a smaller set of data, and then the most important parts of the original image may be reconstructed with an inverse transform. In the end though, the traditional compression process acquires more data than is needed, as the compression process proves that the image is "smaller" than the amount of acquisitions.

CS is unique because it performs compression during the process of signal acquisition. In this spectral imaging application, the image is focused onto an array of many tiny mirrors called a digital micro mirror device (DMD). Each mirror of the array either reflects light to a single detector or away from the detector. The total magnitude of the response is recorded along with the pattern of the DMD. After enough measurements have been taken, a reconstruction process yields the image. The sampling period involving the DMD is similar to a weighing process that puts multiple objects on a scale at once and in the end determines the weight of each component by solving a system of linear equations.

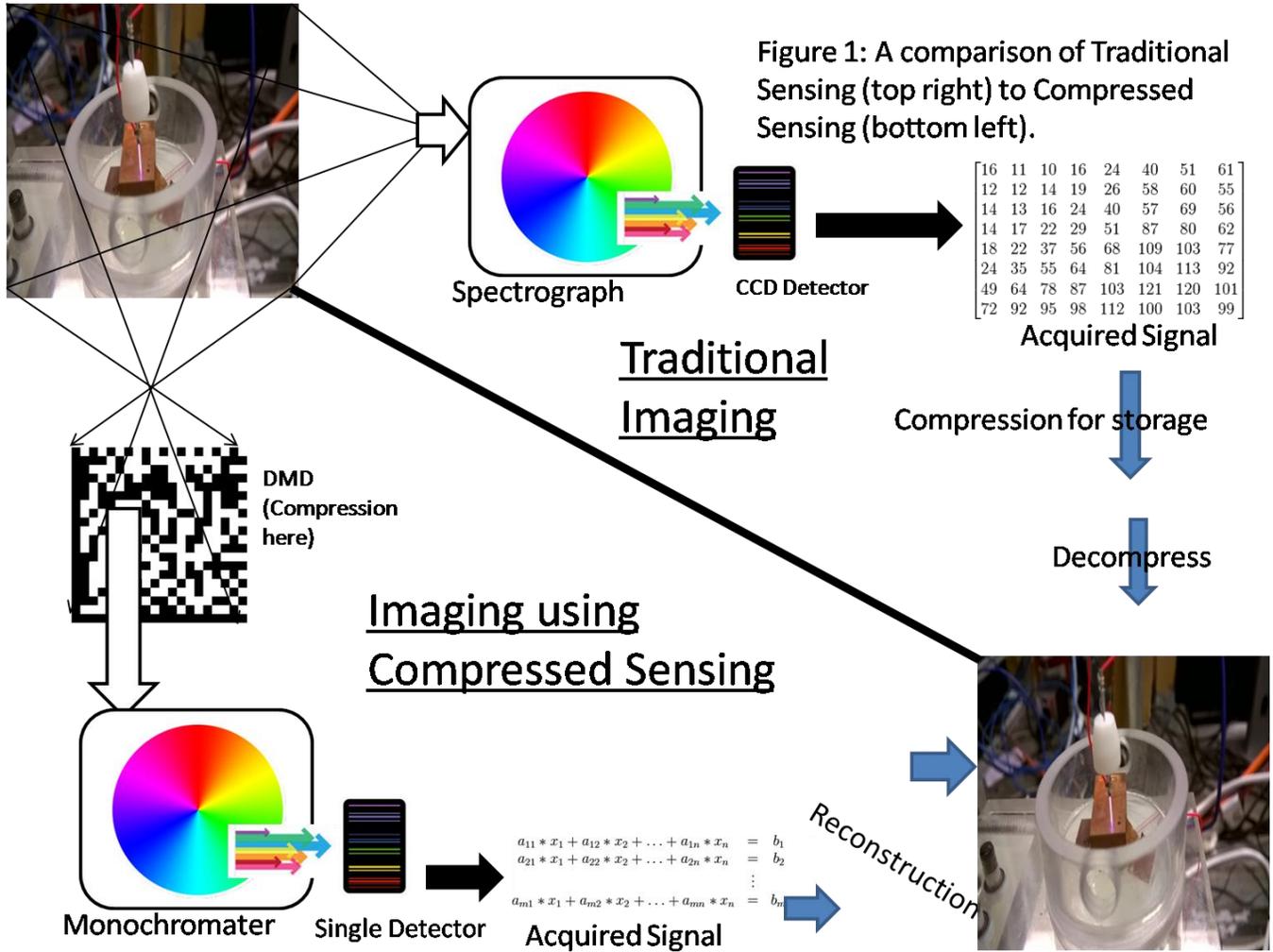
CS in this project is beneficial not only because one may take fewer measurements, but also because the instrumentation needed is more than an order of magnitude less expensive. Specifically, in lieu of a CCD (containing a million or more tiny pixels), a single pixel detector is used. Next, the experimental design allows for novel light shaping with respect to the entrance of the monochromator, and this may enhance spectral resolution with respect to traditional imaging systems.

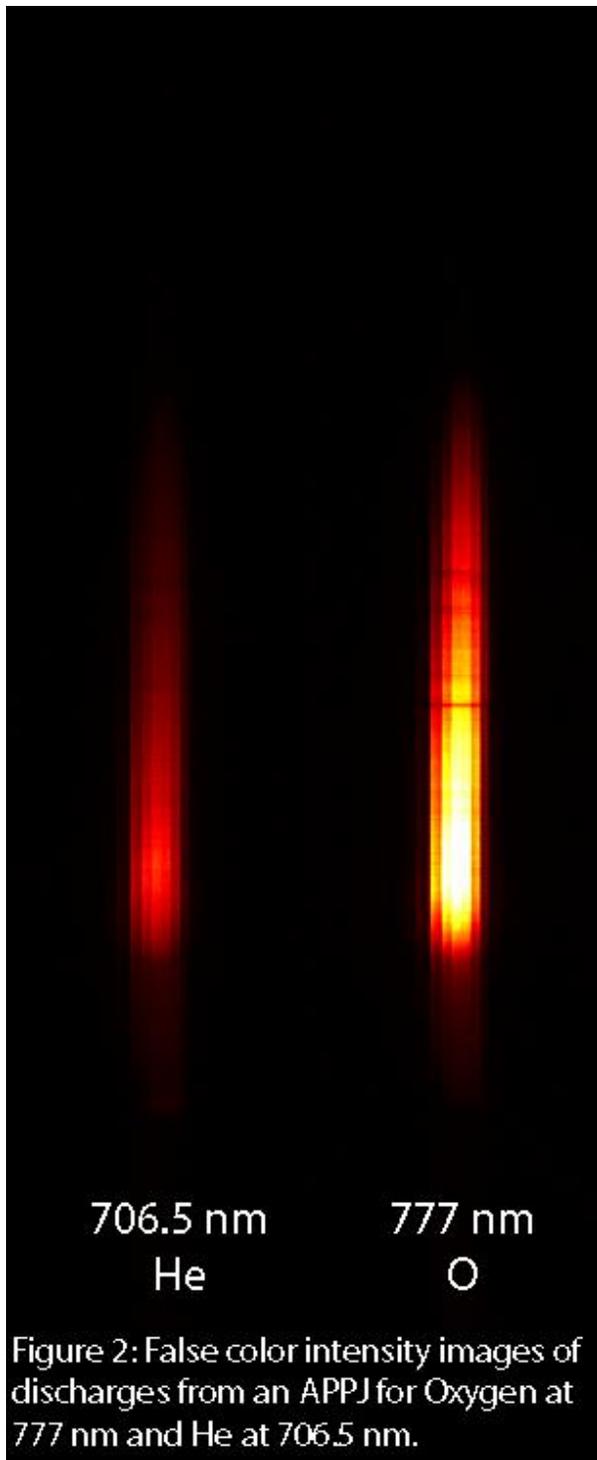
In order to spectrally image the APPJ via CS and thus gain knowledge of its chemical mechanisms, I have begun by determining sparsifying basis representations for our experimental images, finding viable measurement matrices, and testing various computationally feasible reconstruction algorithms. I have

performed simulation measurements to approximate both the resolution and time of reconstruction with the various measurement parameters. I am currently trying to utilize the High Processing Computing Center on campus in order to perform more computationally expensive reconstructions. Finally, I have helped an electrical engineer colleague set up the optics of our system. He has been indispensable in interfacing the ideas electronically and performing adept signal and instrumental analysis.

John Usala

I am a Senior at Texas Tech majoring in Chemistry. I have worked in the lab of Dr. Gamez since October 2014. I have worked on, and still assist with, the Glow Discharge Optical Emission Spectrometry (GDOES) project, and now I am developing a spectral imaging instrument utilizing a single pixel detector. I want to study Analytical Chemistry in graduate school. I spend my free time with my family and my dog.





Reevaluating Phylogenetic Relationships Within the *Peromyscus boylii* Group

Elise Wagley

Mentor: Dr. Robert Bradley

The genus *Peromyscus* has been the focus of many systematic studies because of the diversity that exists within this group of rodents. This genus can be divided into 13 species groups, the specific arrangements of each being a topic for evaluation. The *Peromyscus boylii* species group is of special interest because of the speciation that has occurred within this group. This is due to the diverse environmental and geographical isolation that has followed in the regions of Mexico in which these species occur. Previous studies have examined morphological, allozymic, mitochondrial and nuclear DNA sequences, and karyotypic data to try to resolve species level relationships within this group.

Herein we propose to utilize a combined dataset consisting of one mitochondrial (*Cytb*) and five nuclear genes (*Adh1-12*, *Fgb-17*, *Dmp1*, *GHR*, *Rbp3*) to attempt to resolve relationships within the *boylii* species group. DNA sequences from these genes will be analyzed in a phylogenetic context (Bayesian and likelihood methods) in order to determine the evolutionary relationship of species within the *boylii* group. Although this is a relatively standard methodology, the goal of our study is to use the most complete data to represent the species group thereby offering the most accurate analysis to date.

The *boylii* group is one of the largest species groups in *Peromyscus*, only preceded by the *mexicanus* group (Kirkland 1989). The relationships of species classified within the *boylii* group have been the focus of much debate pertaining to their speciation and evolution. The unique and isolated terrain in which these species occur have allowed for much diversity and speciation (Bradley 2014).

Although many studies have evaluated the relationships among this species group, incomplete taxonomic sampling has prevented the development of any unified decision as to the arrangements within the *boylii* species group. Other difficulties arise because of the morphological similarities among the species leading to different phylogenetic interpretations (Smith 1990). Further still, nomenclature discrepancies have also hindered research (Carleton 1977). The data obtained from this study will offer the most complete and holistic analysis of the group. Studies before this have lacked representative samples for each species and create inaccurate and misleading results.

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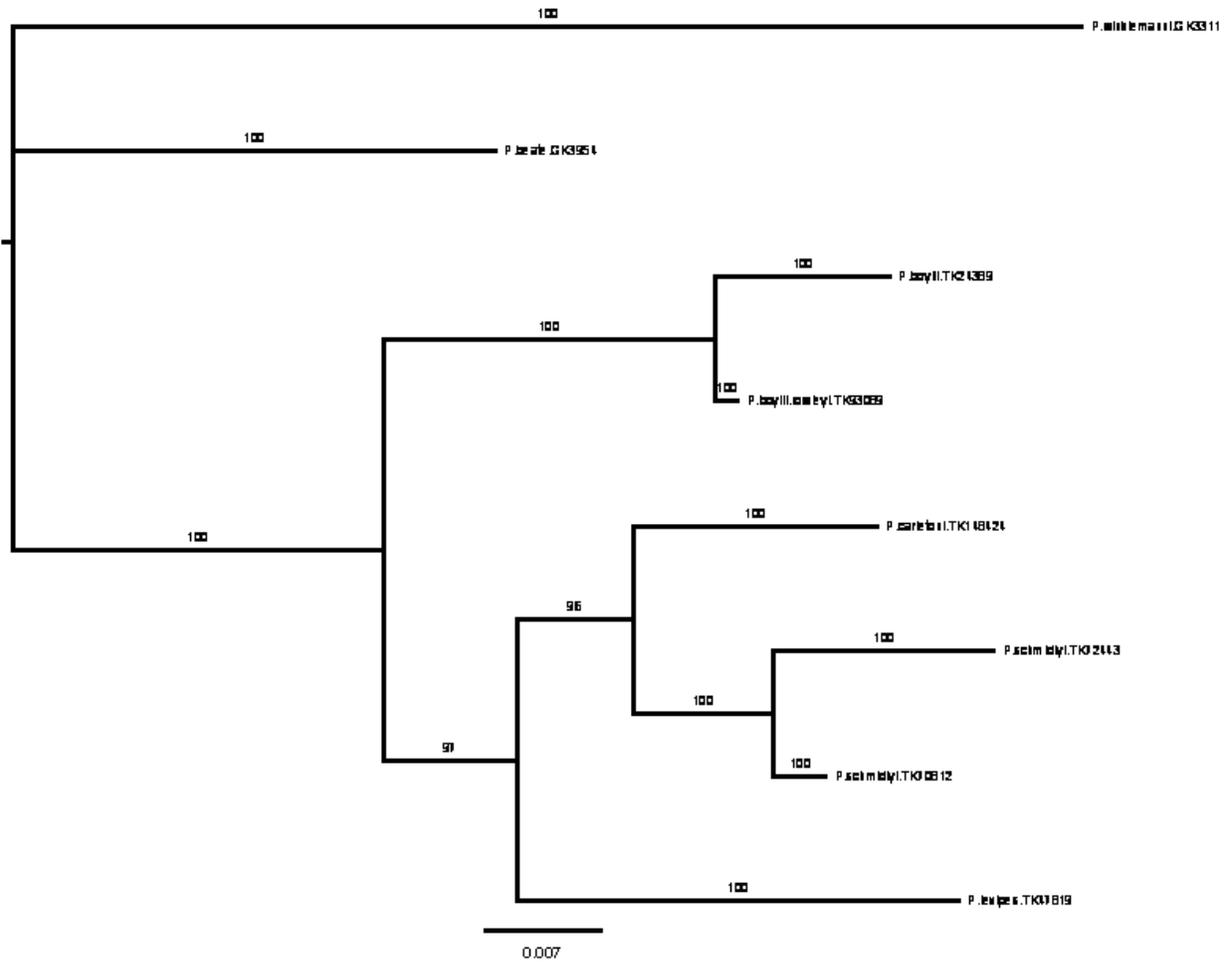


Table 1

Elise Wagley

I am a junior majoring in Cellular and Molecular biology. I work for the biology department as an undergraduate researcher. This will be third semester working in Dr. Robert Bradley’s lab with several other CISER members. Surprisingly enough, although I was born and raised in Austin, I am a die-hard red raider! One of the things I truly admire about Texas Tech is their genuine student support. I am hoping to pursue a dual M.D/PhD program in the future, pairing medical work as a general physician with conservation biology research.

Understanding Species Limits of *Peromyscus mexicanus* Group Using A Genetic Approach

María Núñez

Mentor: Dr. Robert Bradley

The genus *Peromyscus* deer mice is one of the most widely distributed mammalian taxa in North America. Its range extends from the Atlantic to Pacific coast, east to west, and from Canada to Panama, north to south. Due to their wide distribution, this genus consists of more than 50 species and several subspecies. Currently, *Peromyscus* is divided into 13 species groups. Before the introduction of molecular analysis, most relationships were determined using morphology to resolve which organisms were of the same species or which species were more closely related to one another. Current genetic studies provide another way to determine phylogenetic relationships. Here, DNA sequence data is used to determine the phylogenetic relationships of the *P. mexicanus* species group. *Peromyscus nudipes* is one of the species of this group, and it is found in southern Costa Rica and northern Panama. *P. nudipes* systematic relationships to the other *P. mexicanus* species has not been studied, therefore, this will be the objective of this study.

For this study, 56 Cytochrome-b (cytb) sequences were used, 28 of these samples were obtained from GenBank and the other 28 were obtained from samples at the Museum of Texas Tech University. Mitochondrial DNA was extracted with standard DNA Extraction methods. Standard Polymerase Chain Reaction (PCR) procedures were followed using primers LH14115 and H15288 for amplification.

The laboratory work includes PCR cleaning, and cycle sequencing using the same primers. Cycle sequencing reactions were purified using isopropanol clean up protocols, and purified products were sequenced, with an automated sequencer. Resulting sequences were aligned and proofed using sequencer 4.0 support; Chromatograms were examined to verify all base changes. Cytb sequences obtained in this study will be deposited in GenBank. We are going to use different phylogenetic software to generate the phylogenetic tree and the Kimura 2-parameter model of evolution to calculate genetic distances among samples to assess levels of genetic divergence of the species in the *P. mexicanus* group.

References

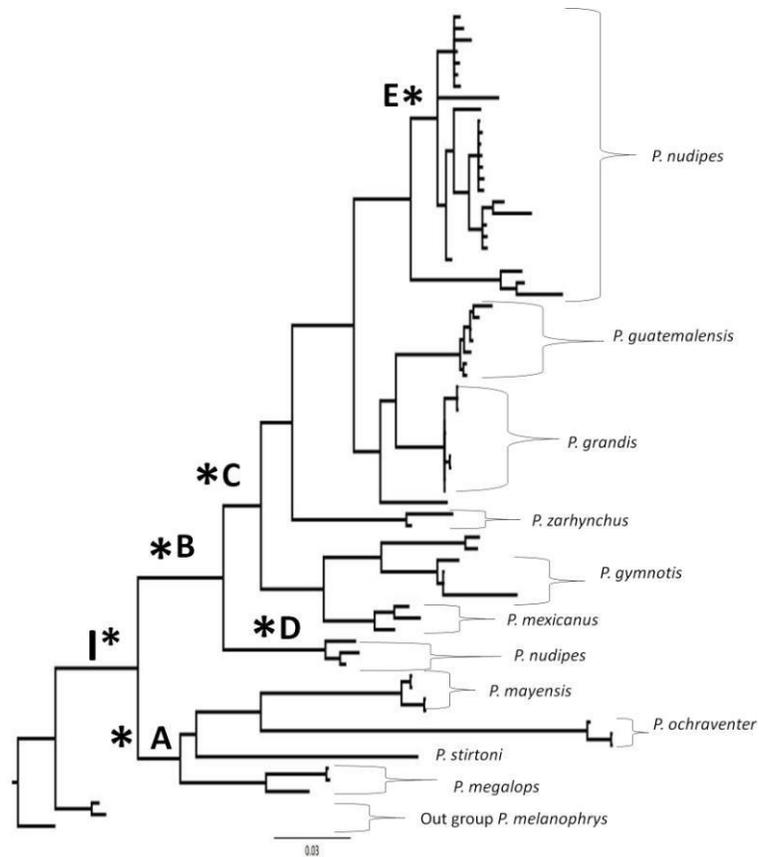
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Figure 1. Distribution of genus *Peromyscus* in North America and depicted in blue *Peromyscus mexicanus* group distribution in Middle America.



Figure 2. Picture depicting *Peromyscus grandis*.



Maria Nuñez-Tabares

I'm a senior majoring in Zoology. I have been involved in research for about six years, since I joined the Plains Bridges Program while attending South Plains College. I am currently working in Dr. Robert Bradley's lab in the Biology department. My goal is to become a teacher and help inspire the next generation to become interested in the STEM fields. My research interests are varied, but for the most part I like projects that involve researching how we as humans affect our environment either directly or indirectly.

Using Genomic and Bioinformatics Techniques to Determine the Origin and Phylogenetic Distribution of the Zonadhesin Gene in Rodentia

Whitney Watson

Mentor: Dr. Robert D. Bradley

Proteins involved in reproduction have been shown to evolve at a higher rate in comparison to the proteins not associated with this particular system. A reproductive protein called zonadhesin exists and it is crucial in species-specific fertilization. This means that during the mating process between two species, the species-specific attributes of this molecule might prevent hybrids from forming at the stage of gamete recognition. In some respects, the functionality of this protein may serve as a post-mating, pre-zygotic isolating mechanism. In addition, even if these hybrids are viable; the protein might also affect their degree of fertility.

Zonadhesin (ZAN) is a multi-domain, transmembrane protein that is utilized in the binding of the egg's zona pellucida layer (ZP) to the spermatozoa. ZAN's structural domains have been studied in other mammalian taxa, but wild rodent systems have been under-utilized in reproductive isolation studies. There are three distinct domains of the protein, one of which has variable and conserved residues among taxa.

Some rodent species are known to rapidly radiate, evolve quickly and develop a variety of isolating mechanisms. Portions of ZAN have been duplicated in the recent past that might be phylogenetically informative. The protein's ability to radiate quickly may facilitate its function of preventing genetically different species from hybridizing.

My undergraduate research role is to investigate representative species from the five suborders within the order Rodentia, each of which displays sufficiently variable ZAN sequences, allowing for a comparative molecular study. The nucleotide differences in the gene can be analyzed phylogenetically and describe the sub-ordinal relationships among rodents.

A related study, which I have been actively working on with my mentor, involves a bioinformatics investigation of this gene among all of Mammalia. In addition, portions of this gene are currently being sequenced in several species of rodents allowing for a broad comprehensive comparison of rodent taxa. As a whole, the goal of this zonadhesin research is to determine if the gene is an effective molecular marker to determine phylogenetic relationships among mammals, specifically those which radiate quickly. Other molecular and morphological datasets utilize multiple markers (genes like cytochrome-b, Rbp3, among other morphological measurements), whereas we can use a single gene to describe those relationships with statistical support.

Messenger RNA and/or complementary DNA sequences will be used to ensure the phylogenetically informative protein-coding regions of the protein are incorporated into the analysis. These sequences will be taken from GenBank, genome databases, or sequenced in the Bradley lab to be utilized in our phylogenetic study. The nucleotides will be scanned for specific domains of interest and aligned using Muscle in MEGA 6. Then Bayesian and Maximum Likelihood analyses will indicate if this protein is phylogenetically informative in Rodentia.

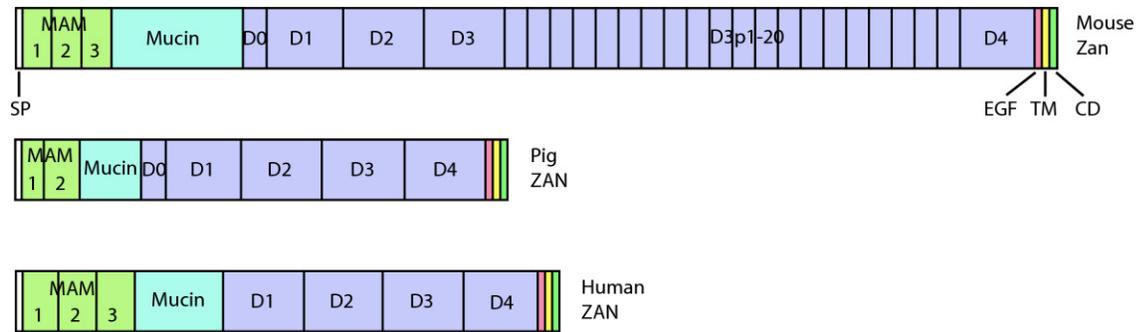


Figure 1. This is a schematic diagram and comparison of mouse, pig, and human zonadhesin. The differences in the structural domains of the proteins can be seen. The mouse has additional D3p domains, which might contribute to the rapid evolution of the gene in that order of mammals (Koop et al. 2005).

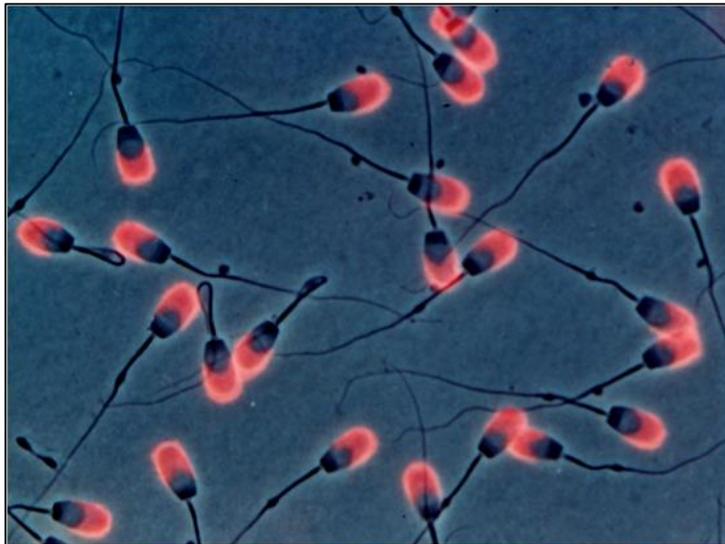


Figure 2. This is a immunofluorescent assay of zonadhesin, located on the apical membrane of the sperm cell (Hickox et al. 2011).

Whitney Watson

I am a senior majoring in Microbiology and minoring in Chemistry and Spanish. I have been involved in research for only one semester and am currently partaking in my second semester in Dr. Bradley’s lab. My future goals are focused on attending P.A. school and becoming a Physician’s Assistant in a field concentrated on pediatrics. My hobbies include reading, spending time outdoors, and watching movies. I love to spend my free time volunteering through the Texas Tech Therapeutic Riding Center.

Phylogeny of Bats of Genus *Monophyllus*: A Study of Genetic and Morphological Divergence Based On Mitochondrial Cytochrome-b Gene

Marilyn Mathew

Mentor: Dr. Robert Baker

The genus *Monophyllus* is distributed throughout the Caribbean Islands. Two species of *Monophyllus* are recognized, *M. redmani* and *M. plethodon*, and are sympatric in Puerto Rico. Observations of morphological differences within each species group were noted and led to two hypotheses. Both hypotheses, based on the Morphological Species Concept, suggested multiple species among both *M. plethodon* and *M. redmani*.

Molecular data generated from the mitochondrial cytochrome-b gene were used to evaluate whether genetic differentiation was congruent with morphological variation. Genetic differentiation between the two recognized species of *Monophyllus*, *M. redmani* and *M. plethodon*, is approximately 11% based on the entire mitochondrial cytochrome-b gene (1140 base pairs). Applying the Genetic Species Concept, genetic differentiation greater than 5%, using the cytochrome-b gene, can lead to the hypothesis of more than one species. Our data do not support the hypothesis of more than one species of *M. plethodon* (genetic distance < 5%). The experimental data generated from specimens of *M. redmani* suggests that there could be an unrecognized species on the island of Puerto Rico (genetic distance > 5%). The type locality of *redmani* is Jamaica, which would mean that specimens with greater than 5% genetic differentiation found elsewhere may constitute an unrecognized species. Genetic differentiation values of the cytochrome-b gene are compatible with standards used to determine taxonomic status, subspecies versus species versus genus and emphasize that 5% cyt b differentiation suggests species level differences in taxonomic studies in mammalian species. Multiple species concepts exist and not all can be applied. However, application of multiple species concepts provide more confidence in conclusions drawn from the data. Based on our data, there is no indication for the existence of more than three species in the genus *Monophyllus*.

The Morphological Species Concept predates the Genetic Species Concept and the Morphological Species Concept is probably the most widely applied species concept in taxonomy. Three reasons the Morphological Species Concept is so prevalent is because genetic data was not always available, many specimens were collected prior to collecting tissue for genetics, and collecting morphological data is not destructive to the specimen. Regarding the Genetic Species Concept, empirical data was used to suggest that 5% genetic differentiation could constitute separate species. This is not a rule; it is only a suggested starting point when using mitochondrial Cytochrome-b data.

I am currently conducting further research to analyze the cytochrome-b gene, by completing sequencing. In addition, we plan to sequence a nuclear gene which could determine if gene flow is occurring among the two groups classified as *M. redmani*. Ultimately, the goal of this project is to continue our search for unrecognized biodiversity among specimens of *M. redmani* and *M. plethodon*.



Figure 1: Specimen from the Texas Tech University Museum from which morphologically differences between the species were studied.

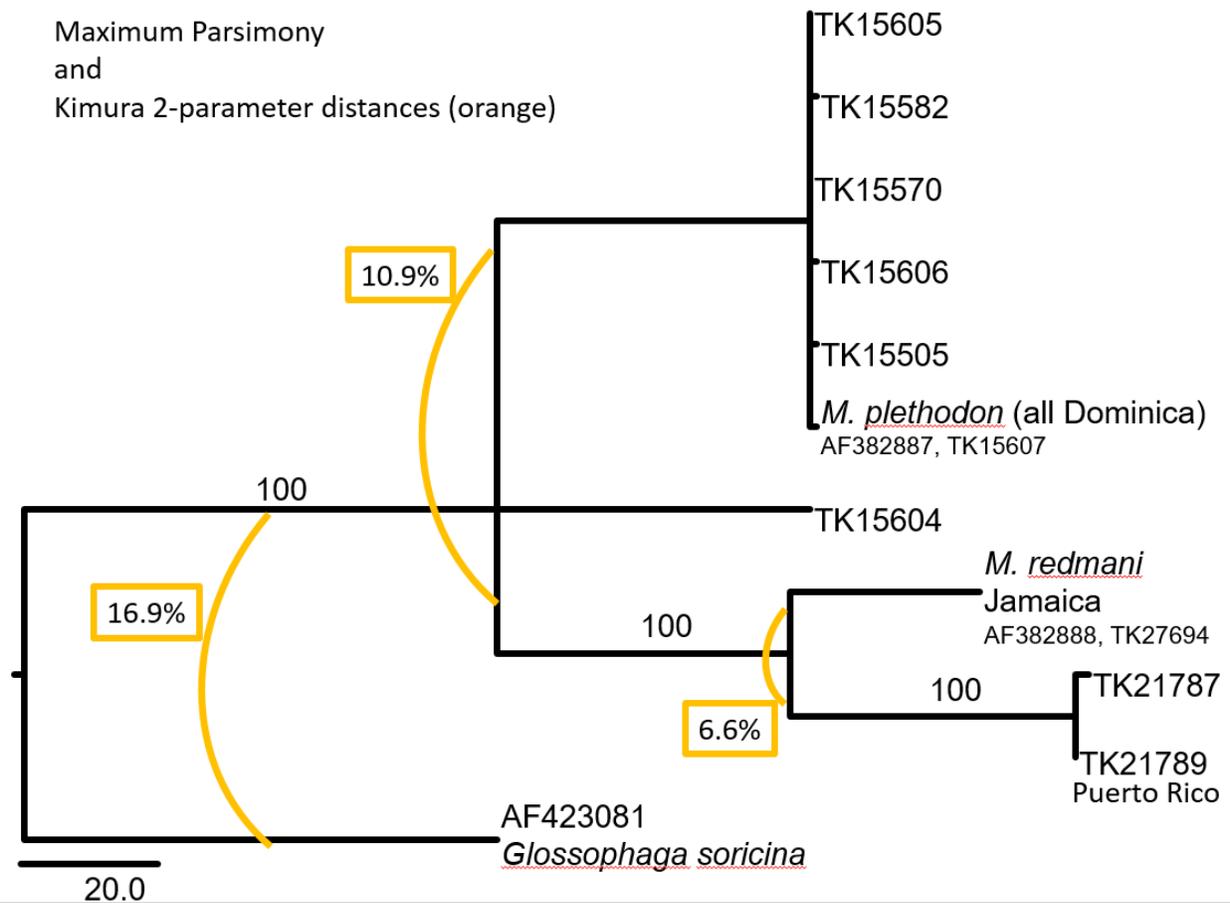


Figure 2: Phylogenetic tree representing the data compiled showing deviation between Redmani from Jamaica and Puerto Rico

Marilyn Mathew

I am a sophomore in Cell and Molecular Biology. I began research in the summer preceding my junior year of high school and have since been involved in research across three separate fields. My primary interests lie in medicine and clinical research, specifically pediatric oncology. I am currently studying the genomics of Monophyllus bats in the Caribbean Islands focusing on the processes for DNA extraction and sequencing to hopefully be applied to other fields in my future. My interests outside of academia include dance, cooking, and just relaxing with good company.

Impact of Human Development on Reptile and Amphibian Biodiversity: Development of a Project

Katherine Crocco

Mentor: Dr. Lou Densmore

With the development and construction of human-populated areas comes a costly price; that is, the destruction of species habitat and populations. There have been many studies on the effect of anthropogenic impact on species' biodiversity in urban areas, but very few specifically centralized on small areas such as college campuses in the United States. Likewise, there is no past or current information on the biodiversity of reptile and amphibian species on Texas Tech Campus. For us to be able to better plan our development as to not disturb or harm the habitats of important reptile and amphibian species, and for us to landscape in a way that will encourage the healthy migration and success of any threatened or endangered species in the area, we must first know what species live here and how our progress affects their population density and richness.

For the purpose of species preservation, it was my aim to create an inventory of the reptile and amphibian species living on Texas Tech campus assessing its population ecology. I was going to then analyze the data to explore how reptile and amphibian species diversity (richness and evenness) is affected by the change in landscapes at Texas Tech University. After that, it was my plan to outline the benefits of coexisting with these animals and bring public awareness on the issue of Texas Tech herpetofauna conservation. My main hypotheses were that 1) reptile and amphibian species richness will be low on Texas Tech campus due to the generality of high human traffic and poor habitat resources for species success; 2) the species I will encounter on Texas Tech campus are the common species commonly seen in urban areas in the Lubbock County, as determined by larger herpetological surveys of the area; and 3) the herpetofauna abundance and diversity on Texas Tech campus will be fragmented due to the adverse affects of university development on the natural habitat of reptiles and amphibians in the area.

I used ArcGIS to create a map of sectors that I would survey of all areas of Tech campus, including main campus, construction areas, and other sites owned by TTU. I researched surveying techniques, and constructed my own for animal surveying. Some of these included pitfall and cover board traps. With these techniques, I planned on conducting the survey on different sectors for 2 weeks per month until the first cold front of the year.

It was my aim to provide a detailed inventory of reptile and amphibian species located on Texas Tech campus for the purpose of conservation. With this inventory, information on species diversity and richness would have been acquired and provided for use in future studies. The conservation status of each species recorded would have been obtained, and it would be known if Texas Tech campus is home to any endangered or threatened species. Landscapes and habitats of any animals found would be recorded, and this would assist the University when providing accommodations for reptile and amphibian preservation in the area.

Surgical Wound Model

Adam Tsen

Mentor: Dr. Abdul Hamood

Although patients see doctors and surgeons with the hope of improving their health, some actually come back after surgery worse through contamination and diseases. Doctors and nurses are exposed to a numerous amount of pathogens in the hospital setting, and occasionally these bacteria can infect a patient undergoing surgery. *Methicillin-Resistant Staphylococcus Aureus (MRSA)* is a well-known gram-positive bacteria that infects people with open wounds, and in this case, an open surgical wound. *MRSA* mode of infection follows most of its relatives as the gram-positive bacterium first synthesizes a biofilm and then proceeds to proliferate and spread across the body via the wound.

The goal of this experiment is to observe and investigate the effects of *MRSA* in a created surgical wound on mice, along with the inhibitory effects of a wash created by NextScience on *MRSA*. For this experiment, we will create the wound by following standard procedures: anesthetizing the mice, shaving their backs, removing any hair leftover, and finally making a designated incision. Our incision will lead to a separation of the muscle and skin layers to effectively create a "pouch" as to insert the *MRSA* on one group of mice. There will 3 groups of mice in this experiment. The first group of mice will be the control group, they will have the incision made and the *MRSA* implanted inside wound. The following two groups will have the *MRSA* placed on the "pouch" as before but one will be treated with PBS, following a previous outlined protocol, and the other with the NextScience Wash. The mice will not be touched for a controlled amount of time. Upon completion, we will observe the effects of the *MRSA* on the mice.

Our experiment is still in its initial stage, hence, we currently do not have any results. However, we hope to further the experiment after collecting the initial sets of data of the inhibitory effects of the NextScience Wash. Furthering the research can be accomplished in numerous methods, by varying the time elapsed between the injection of *MRSA* into the mice, changing the dosage and concentrations of PBS and also by the NextScience Wash, to name a couple.

Adam Tsen

My name is Adam Tsen and I am a senior majoring in General Studies with my three concentrations: biology, chemistry, and Spanish. I have recently joined CISER and have joined Dr. Hamood's lab at the Health Science Center. I plan to attend the Texas Tech Medical School after I graduate and hope to continue research throughout med school, whenever possible. My current project is a Surgical Wound Model involving mice and *MRSA* and observing the inhibitory effects of a wash provided by NextScience.

Vitamin D regulates cholesterol metabolism and breast cancer cell proliferation

Christopher Ponce

Mentor: Dr. Shaikh Rahman

Obesity is a major health problem and a risk factor for breast cancer. Obesity is also strongly associated with estrogen receptor positive breast cancer in postmenopausal women. Breast cancer is the second most-common cancer related death in the USA. Cancer cells undergo metabolic changes during progression. Several studies have revealed cholesterol as an important regulator of breast cancer development though the molecular mechanisms remain unknown.

Vitamin D is an essential component for bone and calcium metabolism. A growing body of evidence from animal and human studies shows that vitamin D improves peripheral insulin action, suppresses the renin-angiotensin system, decreases systemic inflammatory mediators of vascular disease, and prevents foam cell formation. In addition, studies have shown lower levels of serum vitamin D in breast cancer patients. By contrast, women with higher levels of serum vitamin D levels are at a lower risk to develop breast cancer. However, whether vitamin D regulates cancer cell proliferation by regulating cholesterol metabolism is unknown. We hypothesize that vitamin D prevents cancer cell proliferation by regulating cholesterol homeostasis.

MCF 7 (an estrogen receptor (ER) positive cell line) cells were grown in culture medium and treated with (1 μ M and 2 μ M) and without (control) vitamin. D (calcitriol) for 24 hours. Cells were harvested and lysed with lysis buffers and nuclear and cytosolic fractions were prepared. Protein concentration was measured by Bio Rad protein assay kit using a microplate reader. Equal amounts of proteins were separated by 10% SDS-PAGE and transferred to PVDF membranes. The membranes were blocked with blocking buffer for one hour and subsequently probed using the primary antibodies. After washing three times in TBST, the blots were incubated with HRP-conjugated anti-rabbit secondary antibody or HRP-conjugated anti-mouse secondary antibody for 1 h at room temperature. The membranes were then scanned and visualized using a LI-COR ODYSSEY CLx. Vitamin D treatment significantly increased the expression of ATP-binding cassette transporter A1 (ABCA1), a protein involved in cholesterol efflux and AMPK (a regulator of energy metabolism, inhibits inflammation) activity as evaluated by phosphorylation of AMPK (P-AMPK) and cleaved caspase 3 (which is involved in apoptosis) proteins.

In conclusion, Vitamin D treatment increases cholesterol efflux and apoptosis in MCF7 cells. Our preliminary data indicate that vitamin D increases apoptosis by increasing phosphorylation of AMPK and by increasing cholesterol efflux. We will further analyze the expression of genes involved in cholesterol synthesis, uptake and efflux by quantitative polymerase chain reaction. We will also measure the levels of cholesterol and cholesterol ester in vitamin D treated cancer cells. Furthermore, we will analyze cell viability and will perform cell proliferation assay.

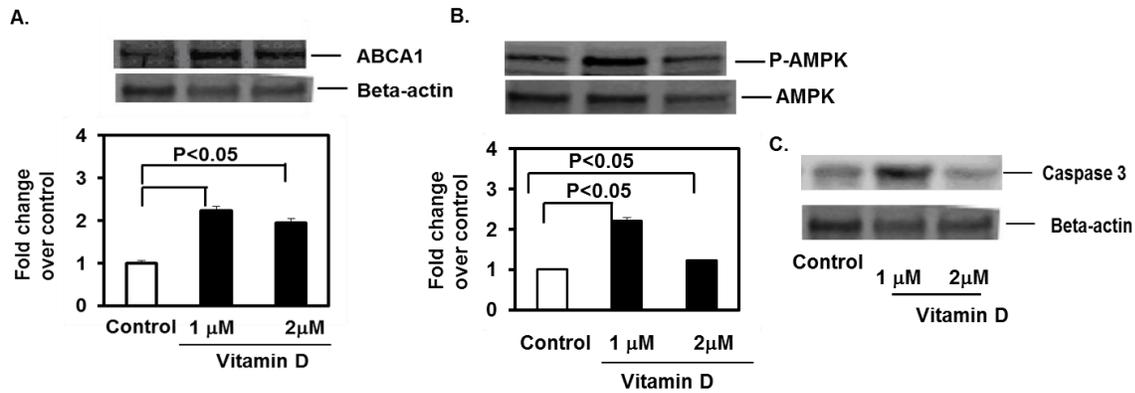


Figure 1. A. Immunoblot and densitometric values for ABCA1. B. Immunoblot and densitometric values for P-AMPK. C. Immunoblot for caspase 3. Representative vblots are shown. Data are presented as mean±SEM. n=3.

Christopher Ponce

I am a third-year student majoring in mathematics and minoring in chemistry. I began my first small research project last fall. My research interest focuses on finding and understanding metabolic pathways that induce breast cancer cell growth. Currently, I am working on measuring the effects of a treatment on breast cancer cells at different stress levels. I am interested in attending medical school and hope to one day become a radiation oncologist. My other interests include hiking, road trips, and shooting.

PEDF Induces the Migration, Differentiation and Phagocytic Activity of Macrophages

Dalia Martinez-Marin

Mentor: Dr. Stephanie Filleur

Macrophages have been described as one of the main inflammatory components in tumor growth, including in prostate cancer. Pigment Epithelium-Derived Factor (PEDF) is a secreted glycoprotein, which blocks angiogenesis, promotes neuronal survival and differentiation, and was recently suggested as an immune-modulating factor. PEDF also acts as an inflammation-modulating factor in prostate cancer. Prior studies have shown that PEDF expression increased the recruitment of tumor-cytotoxic M1-type macrophages into orthotopic MatLyLu rat prostate tumors suggesting a new way through which PEDF curbs prostate cancer growth. The totality of these data reinforces the anti-tumor properties of the PEDF gene in human castration-refractory prostate cancer (CRPC). It also emphasizes the critical need to develop a specific and efficient delivery system for PEDF gene. The objective of the present study is to investigate PEDF gene therapy using bone marrow-derived macrophages (BMDMs) as a novel therapeutic modality for advanced CRPC, using immunohistochemistry and in-vitro migration assay. Our central hypothesis is that the expression of PEDF will induce the migration and differentiation of BMDMs into a tumor-cytotoxic phenotype and, as a corollary, will block tumor growth and metastases formation, and prolong survival. We have formulated this hypothesis on the basis of our preliminary data that showed that PEDF stimulates the migration of monocytes/macrophages in-vitro and our observation that PEDF expression levels correlate with macrophage density in human prostate specimens.

In addition, we are looking at the mechanisms at which PEDF induces the migration and differentiation of macrophages. To test our hypothesis we used two established cell lines, RAW 264.7 monocytes and THP-1 monocytes. We studied different markers for two (M1 and M2) macrophage phenotypes. M1 macrophages are tumor-cytotoxic, and tumor progression promotes a phenotype switch to M2 macrophages, which promote tumor growth, survival and metastasis. As explained earlier, PEDF expression has been shown to be associated with an increase in density of M1 macrophages. We evaluated the effect of PEDF expression on monocytes and macrophages by using different concentrations of PEDF (0nM; 0.5nM; 1nM; 5nM; 10nM). We studied the different concentrations by using Enzyme-linked immunosorbent assay (ELISA), western blotting, and real-time PCR (RT-PCR). In these techniques, we studied M1 specific markers iNOS, and M2 specific markers Arginase 1. We found that an increase in PEDF expression up-regulates the expression of iNOS and down-regulates the expression of Arginase 1. This shows us that PEDF is associated with an increase of macrophages of an M1 phenotype. Our data of PEDF expression correlates to our previous findings that PEDF induces the migration of monocytes and macrophages in-vitro. As a result of their differentiation, we have found that PEDF stimulates the phagocytosis of tumor cells, which suggest another mechanism by which PCa growth is halted.

We are looking at PEDF mechanisms by investigating the expression levels of PEDF receptors (ATP5B, PNPLA2, and LRP6) in RAW 264.7 macrophages and BMDMs. So far we have found that the ATP5B and PNPLA2 are the two main receptors expressed in both cell types. While all three receptors in RAW 264.7 increased, only ATP5B and PNPLA2 receptors increased in BMDMs. This is being studied using the same techniques as the macrophage markers such as western blotting and real-time PCR. Using immunocytochemistry we were able to see an increase in expression of both ATP5B and PNPLA2 in cells treated with PEDF than those without PEDF. In order to determine the specific activity of PEDF receptors we are using nucleofection to transiently transfect the RAW 264.7 macrophages with siRNA (ATP5B and PNPLA2) to knock-down receptor activity. Concurrently we are using (S)-BEL and angiostatin to inhibit the activity of PNPLA2 and ATP5B on macrophages in the presence/or not of PEDF in a co-culture system. Our

results suggest that ATP5B is involved inactivation of the phagocytic pathway, whereas PNPLA2 regulates the differentiation while maintaining macrophages' phagocytic activity.

We are also investigating the PEDF-derivative synthetic 18-mer peptide and its mechanism of action on macrophages. P18 has been shown to block endothelial cell chemotaxis and induces apoptosis in vitro, and has been shown to be more effective than its parental 34-mer peptide in blocking growth and angiogenesis in prostate cancer. We were able to demonstrate using western blotting, RT-qPCR, as well as immunocytochemistry that macrophages treated with P18 in comparison to PEDF (parental 34-mer peptide), shows a higher efficacy of PEDF expression. Using confocal microscopy we were also able to demonstrate a much larger rate of phagocytosis of prostate cancer cells by macrophages than compared to PEDF.

Finally, we will assess PEDF gene therapy using bone marrow-derived macrophages (BMDMs) as a novel therapeutic modality for advanced PCa. This project may lead to development of improved therapeutic approaches to treat PCa. Our central hypothesis is that PEDF expression will induce the migration and differentiation of BMDMs into a tumor-cytotoxic phenotype and, as a corollary, will block tumor growth and metastases formation, and prolong survival. The results of our study are of importance as they suggest that macrophages may play a key role in PEDF anti-tumor effects. A better understanding of PEDF may help lead to the further development of PEDF based anticancer therapy or improvement of alternatives to chemotherapy for prostate cancer.

Dalia Martinez-Marin

I am a senior getting a B.S in Cell and Molecular Biology. I started doing research the summer of my junior year in high school and have done research in four labs here at TTU-TTUHSC. I am very interested in medical research specifically in cancer research. After graduation I plan on pursuing a PhD and continue doing research in cancer. My other hobbies are playing in the TTU symphony orchestra and playing video games.

Figure 1: PEDF treatment led to the differentiation of macrophages towards the M1/tumor-cytotoxic pathway. PEDF treated mouse macrophages led to an increase in M1-type specific markers in RAW 264.7 IL12 (left) and iNOS (center), and led to a decrease in M2-type specific marker IL10 (right).

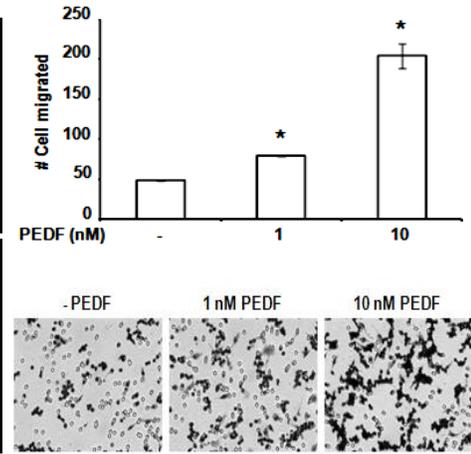
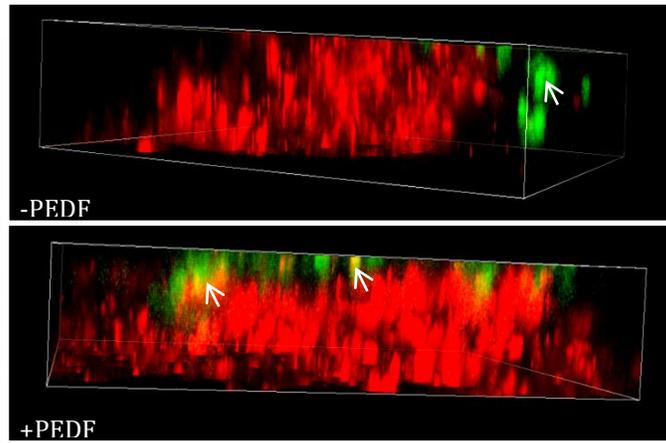
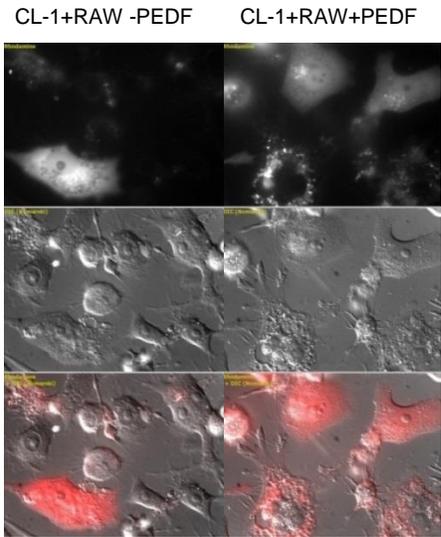
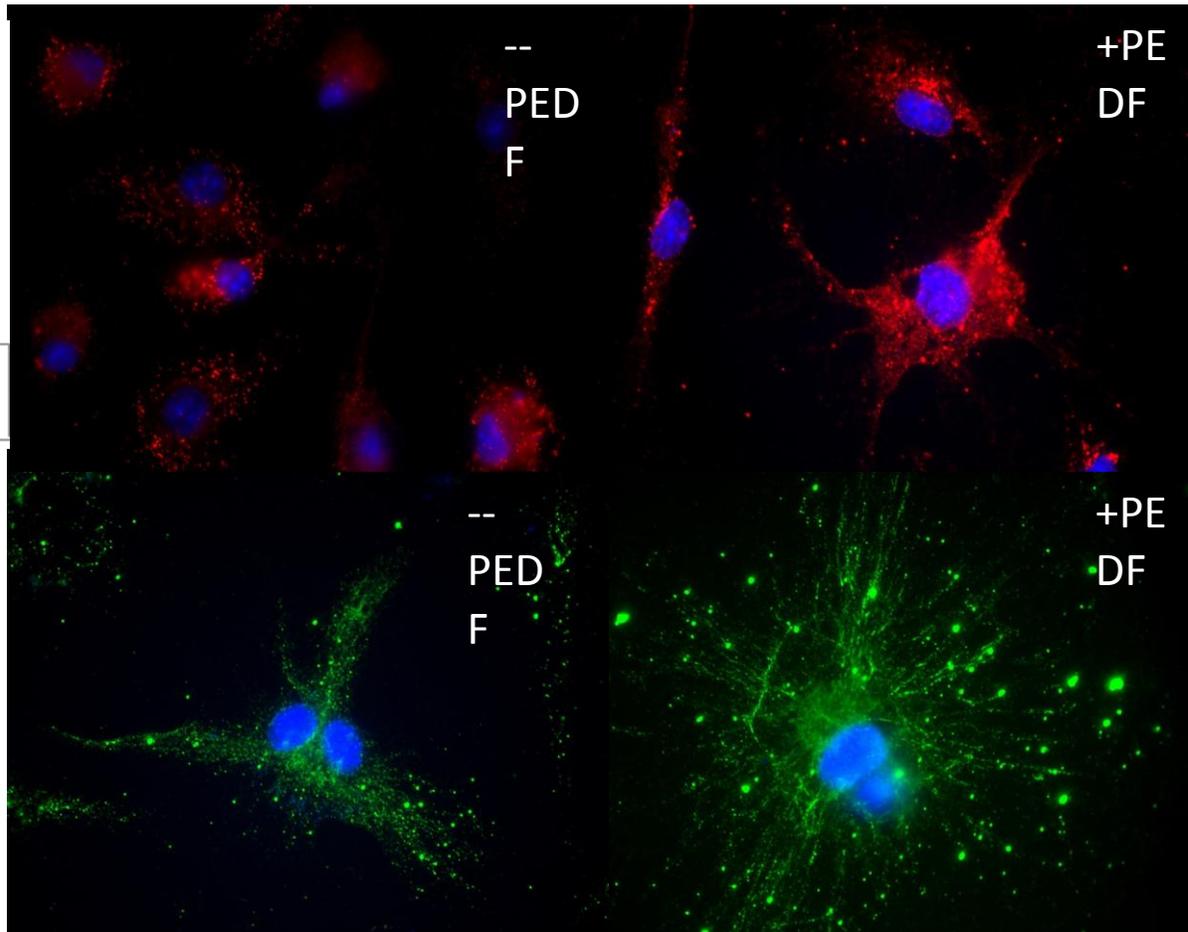
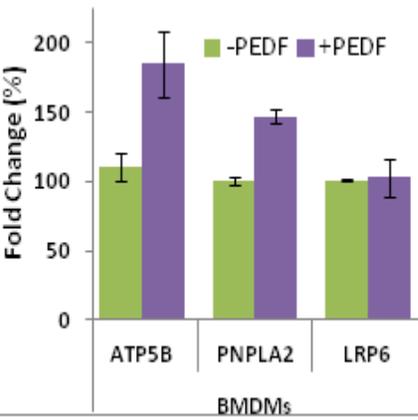


Figure 2: (Left) PEDF increases the engulfment of tumor cells by macrophages. **(Center)** Using confocal imaging we were able to determine that PEDF leads to migration of macrophages towards the CL-1 spheroid. **(Right)** PEDF induces the migration of macrophages in vitro.



CD47 down-regulation by PEDF: a new means of inducing tumor cell phagocytosis *in vitro*

Nitish Mittal- Urology
Mentor- Dr. Stephanie Filleur

Prostate cancer is the second leading cause of cancer related death in men in western countries. Currently there is no cure for advanced or metastatic prostate cancer. Our lab focuses on investigating the functions of the pigment epithelium derived factor (PEDF) in prostate cancer. PEDF is a secreted glycoprotein, which promotes neuronal survival and differentiation, and blocks angiogenesis, the formation of new blood vessels from pre-existing ones. In previous studies, we have shown that PEDF also curbs prostate cancer growth, and enhances the migration and phagocytic activity of macrophages leading to tumor cell phagocytosis. The present project focuses on studying the role of CD47, a cell surface protein, in PEDF-induced phagocytosis. CD47 is an integrin associated protein that has been shown to play a role in several cellular processes such as apoptosis, proliferation, adhesion and migration. CD47 has also been identified through its interaction with SIRP- α as a “don’t eat me signal” protecting tumor cells from phagocytosis. CD47 is ubiquitously expressed in human cells and has been found to be overexpressed in many different tumor cells, including prostate cancer. In contrast, SIRP- α is expressed mainly by macrophages and dendritic cells. Recently, the lab has demonstrated that PEDF reduces the expression and cell surface localization of CD47 in prostate cancer cells, suggesting that CD47 down-regulation could be responsible for PEDF-induced phagocytosis. In the present project, we propose therefore to: 1) validate CD47 down-regulation by PEDF using several prostate cancer established cell lines; and 2) evaluate the role of CD47 in PEDF-induced phagocytosis. This project could lead to better understanding of the role of PEDF in the tumor microenvironment and may result in the identification of new therapeutic target for metastatic prostate cancer. We are going to use RT-qPCR, FLOW cytometry, western blotting, and confocal imaging to demonstrate our hypothesis: *PEDF down-regulates CD47 expression in prostate cancer cells allowing for the phagocytosis of tumor cells by macrophages.*

In this experiment, we are using several cell lines as experimental models: RAW 264.7 monocytes/macrophages, and prostate cancer that express (CL-1 PEDF and PC3 PEDF) or do not express PEDF (CL-1 E10 and PC3 E19). Hence, during this project, we are trying to determine whether PEDF helps macrophages in the process of phagocytosis of cancer cells by down regulating CD47.

Cell lines. RAW 264.7 monocytes, and prostate tumor cells were cultured according to ATCC protocols. All cells were cultured in a 75 cm² tissue culture flask and place in a CO₂ (5%) incubator at 37° C. CL1-E10/PC3 E19 and CL-1 PEDF/PC3 PEDF were transfected with an empty vector and a PEDF expressing vector, respectively. Before experimentation RAW 264.7 monocytes were treated for 48hrs in Phorbol 12-myristate 13-acetate (PMA) to induce differentiation into macrophages.

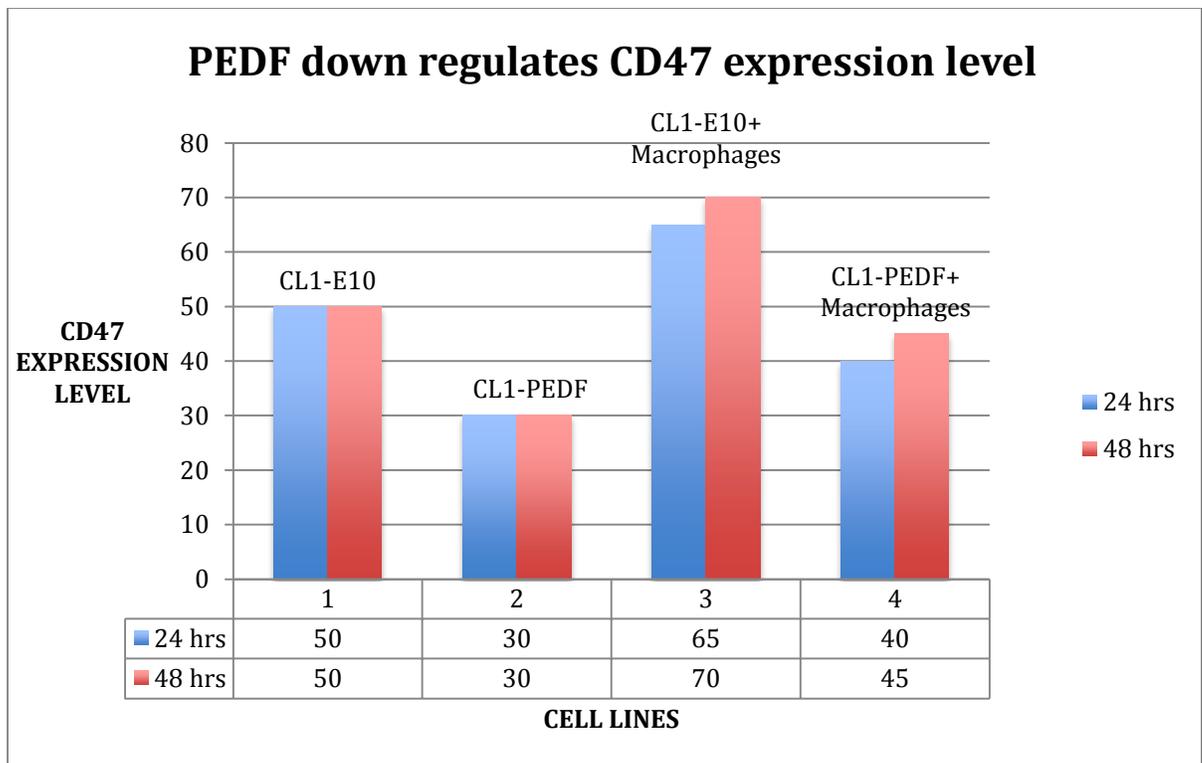
RT-qPCR. Until now, I am working on the first experiment. During this experiment, we seeded monocytes(RAW 264.7) in cell culture dishes. After 24hrs, we added PMA in complete medium for 48hrs. We then seeded CL-1 E10 and CL-1 PEDF cells alone into different cell culture dishes or in co-cultures with the macrophages in the presence of PMA to prevent macrophages from going back to monocytes. We collected RNA, after 24 and 48hrs seeding. Then using reverse transcriptase we converted the RNA into cDNA. We then run a qPCR with a set of CD47 human primers to determine the expression level of CD47 in our mono and co-cultured cells.

I am not able to produce all the results, but I want to demonstrate that PEDF has a significant effect on CD47 and as a result, we can induce phagocytosis by macrophages, to help lead in the further development of PEDF-based anticancer therapy.

If my research is successful, it means that macrophages can act as a key component in PEDF anti-tumor effects. This would lead to better understanding of the role of PEDF and hopefully, to the development of new alternatives in the treatment of the prostate cancer.

I failed a few times during my research and I tried to figure out the reasons behind it.

- In one case, I seeded my RAW 264.7 (macrophages) and added PMA. Then, I seeded CL-1 through co-culturing and checked under the microscope. While everything sounds alright, the next day, all the cells were dead. We tried to find out the reason: I took a long time for seeding the cells which could have led to an important stress level for the cells resulting in their death.
- In other case, during the cell culture process, I was getting ready for seeding CL-1 with RAW 264.7 (macrophages), but I found out that cells were not confluent enough. So, I had to wait for 3 extra days to achieve the proper confluency to seed the cells.
Reason: Probably, I made a mistake during the seeding process of RAW 264.7 in cell culture dishes such as (improper counting of the cells. These two failures made me realize the carefulness and precision required during the research to produce good results in time. While these attempts may be disappointing, I am happy to be involved in a research lab where everyone is willing to help at any given point of time.



Nitish Mittal

I am a second year student in Chemistry. I am an international student and my first year at Texas Tech University was a great experience. My dream is to get into a good medical school and become a successful doctor and help the society in a best possible way. I have been working in the lab of Dr. Stephanie Filleur for past few months and it has been a great learning curve. For leisure activities, I like playing lawn tennis and badminton.

Terminally Differentiated Sertoli Cells Reinitiate Proliferation After Loss of Cell-Cell Contact

Rachel Dziuk

Mentors: Jannette M. Dufour and Gurvinder Kaur

Sertoli cells (SCs) are immune privileged somatic cells that line the seminiferous tubules of the testis where they create a unique microenvironment for developing germ cells. During puberty, hormonal changes induce major gene expression changes in SCs. For example, SCs cease proliferative activity and form the dynamic blood testis barrier (BTB) through adjacent SC-SC tight junction interactions.

According to dogma, SCs are terminally differentiated after puberty, where their adjacent interactions in the BTB allow for successful spermatogenesis. However, new research findings like our previous study contradict the conventional dogma and suggest SCs may be capable of re-initiating proliferation or reverting back to de-differentiated, immature states. Previously, terminally differentiated adult Lewis rat SCs were transplanted under the kidney capsules of adult Lewis rat or non-obese diabetic, severe combined immunodeficient (NSG) mice. Graft bearing kidneys were removed at days 5, 10, 14, 19, and 50, paraffin embedded, and immunostained for SC marker, Wilms' Tumor (WT) 1, and cell proliferation marker, KI67. Graft analysis revealed mature SCs reinitiated proliferation after transplantation for approximately 14 days [1]. Maximal SC proliferation was observed at day 10, so adult Lewis rat SCs were transplanted as previously described. Animals received daily injections of 5-bromodeoxyuridine (BrdU), and their graft bearing kidneys were removed at day 10. Analyses also revealed SCs arranged in tubule-like structures proliferated significantly less than SCs not arranged in tubule-like structures. Multiple factors involved in transplantation may influence SC proliferation. Because we observed a correlation between proliferation and the ability for SCs to form tubule-like structures, we hypothesize the loss of cell-cell contact between mature SCs influences their proliferation after transplantation.

Using the same animal model as before, day 10 graft bearing kidneys were double immunostained for SC marker, WT1, and tight junction protein marker of the BTB, Claudin-11. High Claudin-11 expression was observed in SCs arranged in tubule-like formations. The total area of SCs within tubules and outside of tubules along with the total area of Claudin-11 were determined. Interestingly, Claudin-11 expression was significantly higher in SCs arranged in tubules compared to SCs outside of tubules in NSG mice grafts.

No significant difference was observed in Claudin-11 expression in Lewis rat grafts. Speculated reasons for this observation stems from the nature of the animals. NSG mice are immunodeficient. It has been reported that inflammatory molecules like TNF- α associate with and down-regulate tight junction protein Claudin-11 [2]. A non-specific inflammatory response generated against transplanted SC grafts in Lewis rats could explain the decreased formation of SC tubules and why Claudin-11 was non-significantly expressed within tubules.

Future studies will analyze and compare the inflammatory responses generated against Lewis rat grafts to that of NSG mice. Additionally, future studies may also examine the functionality of the observed SC-tight junction formations. Collectively, these data may uncover novel control mechanisms of proliferation and potential dedifferentiation of SCs.

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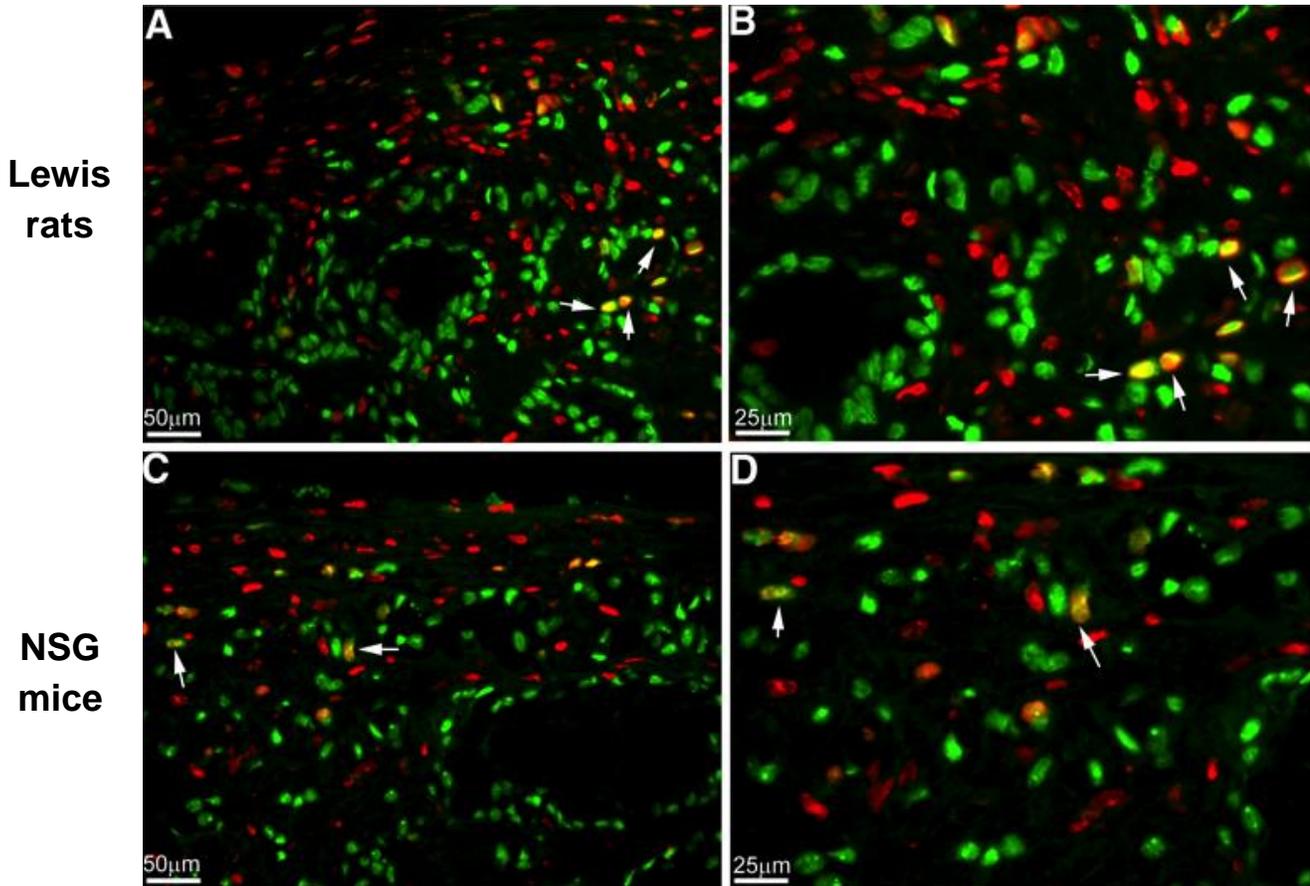


Figure 1: Preliminary data: Mature SCs resume proliferation after transplantation. Graft bearing kidneys sections were stained with SC marker WT (green, A-D) and Proliferation marker, 5-Bromo-2-Deoxyuridine (BrdU) (red, A-D). Double positive proliferating SCs were yellow. Results: Mature SCs resumed proliferation after transplantation. Majority of the BrdU+WT1+ cells were located outside the tubules i.e. randomly present throughout the graft [1].

Recipients	% of BrdU+ SCs outside of tubules	% of BrdU+ SCs within tubules	Total % of BrdU+ SCs
Mice (n=4)	9 (4-21)	2* (0.8-6)	11 (5-27)
Rats (n=4)	11 (4-34)	6 (2-15)	17 (6-49)

Table 1: SCs arranged in tubules proliferate significantly less compared to those outside of tubules. *p ≤ 0.05 compared to outside of tubules [1].

NSG Mice

Lewis Rats

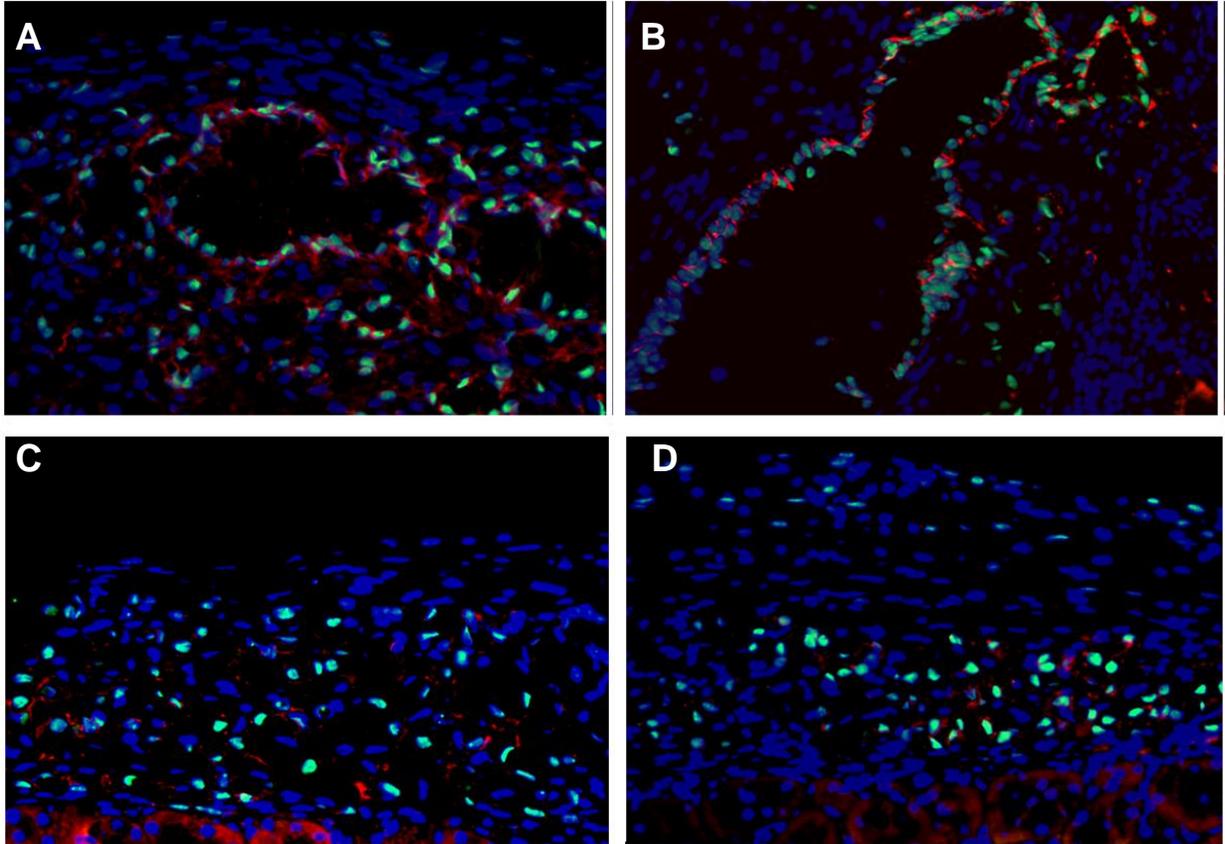
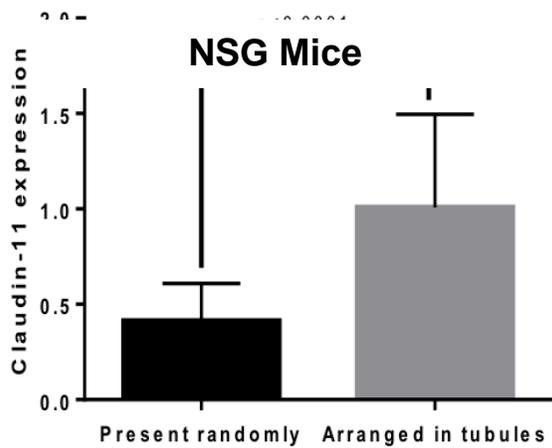


Figure 2: Immunofluorescence staining reveals high Claudin-11 expression within SC tubules and low Claudin-11 expression outside of SC tubules. Coral/pink: Claudin-11 tight junction protein, Aqua blue: WT1 for SCs, deep blue: DAPI nuclear stain. Grafts A and B showed high Claudin-11 expression in the tubule formations. Grafts C and D showed very little tubule formation and decreased Claudin-11 expression by SCs.



Lewis Rats

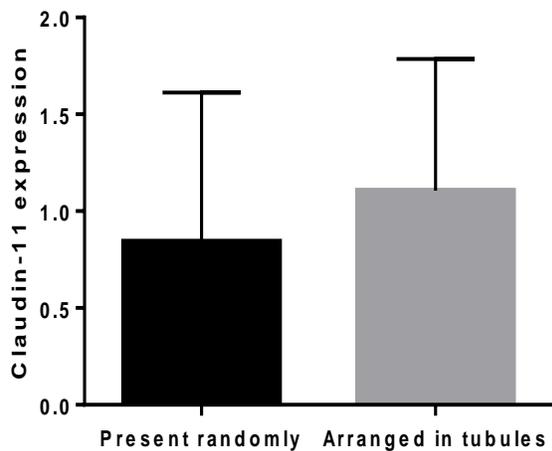


Figure 3: Claudin-11 expression was significantly higher within SCs tubules in NSG mice grafts.

Rachel Dziuk

I am a senior cell and molecular biology major. I entered the Dufour lab in June of 2013 as a Summer Accelerated Biomedical Research Intern and found my passion for biomedical research. My projects investigate Sertoli cell immune-privilege mechanisms after transplantation. Understanding Sertoli cell mechanisms will advance transplantation success or the potential novel use in gene therapy to treat diseases like type 1 diabetes. I will pursue my Ph.D. in immunology or cell biology to explore my interest in autoimmunity. My hobbies include enjoying outdoor adventures, cooking, and playing piano.

Functional Amyloids: A Link between Yeast Reproduction and Mammalian Fertilization

Gage Rowden

Mentor: Dr. Gail Cornwall

Amyloids are proteins that self-aggregate and form cross- β sheet fibrils. They are typically thought of as pathogenic structures such as the A β aggregates and neurofibrillary tangles in Alzheimer's disease, or those involved in transmissible spongiform encephalopathies. However, in a previous study it was found that amyloids serve a functional purpose in the mouse sperm acrosomal matrix, and the zona pellucida (ZP) surrounding the mouse oocyte. This suggests amyloids may play a role in sperm-egg recognition.

The mouse ZP consists of 3 proteins, ZP1, ZP2, and ZP3, which form a fibrillar matrix around the oocyte. An analysis of these proteins using AmylPred 2, which predicts amyloidogenic sites, revealed an important hydrophobic conserved region in the ZP-N domain of ZP3 which is also present in the α -agglutinin/Sag1p protein in *Saccharomyces cerevisiae* (Baker's yeast). The conserved region was also found in mollusk, fish, frog, quail, and human ZP3 proteins. Sag1p is known to be involved in sexual recognition between Mat-A and Mat- α mating types. This suggests yeast sexual mating, as in mice, may be mediated by an amyloid-amyloid interaction. Discovering a similar mechanism for sexual recognition between complex multicellular animals and single-celled eukaryotes would illustrate remarkable conservation through hundreds of millions of years of evolution.

Using Congo Red (CR) to stain for cross- β sheets (structures characteristic of amyloid), we observed the appearance of CR staining localized to the shmoos after induction with mating pheromone in both A- and α -mating types. Shmoos are extensions of the cell wall which allow for the mating bridge to form. The presence of CR fluorescence in these extensions would imply amyloid is involved in yeast sexual reproduction; however, further research is required.

MALDI-TOF as a New Tool for Quantification of Polyamines in Plants

Aicha Fokar

Mentor: Dr. Masoud Zabet

Polyamines (PAs) are straight-chained C3-C15 aliphatic hydrocarbons substituted with amine groups and are considered a major secondary metabolite regulating cellular growth and development. In plants, polyamines have been implicated in a wide range of processes including responses to biotic and abiotic stress, cell elongation, senescence and hormonal signals. Despite the large understanding of their ubiquitous role, their exact function remains unclear. This is partially due to the difficulties associated with measuring natural polyamines in plants. In an effort to identify the exact functions of these molecules, we are looking to discover an accurate and reproducible means by which we can measure these natural polyamines.

Direct analyses of PAs is difficult because of their simple aliphatic structure, and thus they are usually derivatized. To date, high performance liquid chromatography (HPLC) is a well-established method in analyzing polyamines. The purpose of this study is to develop a new, fast and reliable method to quantify polyamines to further understand the role of polyamines in signal transduction, plant development and responses to stress.

MALDI-MS has been used for the analysis of a variety of small molecules. In this case, we applied MALDI-TOF for the qualitative and quantitative analysis of dansylated PAs. First, the standard calibration curves were obtained for each PA followed by the quantification of the results through utilizing an established method of HPLC.

This will be required in gaining a meaningful understanding of polyamine homeostasis in the plant cell. In this study, we investigate the application of MALDI-TOF as a new tool for qualitative and quantitative analysis of polyamines using model plants *Arabidopsis thaliana* and Tobacco (*Nicotiana tabacum*). Further, we will apply this method to analyze polyamines extracted from various mutant lines with lesions in plant hormone biosynthesis and signaling genes.

Aicha Fokar

I am a junior studying cell and molecular biology. I have been involved in research since my junior year in high school. I recently worked in the lab of Dr. Richard Strauss where we used mathematical equations to model scale patterns on catfish. I am now joining a biotechnology lab with Dr. Zabet Hamoud. I look forward to delving into the biotech field, and later pursue a career in pharmacology and toxicology. My interests include research, political science and art.

Searching for Low Mass Dark Matter Halo Counterparts to Ultra-Compact High-Velocity Clouds in the Local Group

Maksym Zhelyeznyakov

Mentors: Dr. David Sand, Dr. Elisa Toloba

The λ CDM model, which describes the evolution of the universe, predicts the existence of a large number of low mass halos in the local group. The number of dwarf galaxies that have been discovered however, is magnitudes smaller than the simulations predict. This has been dubbed as the missing satellites problem (Klypin et. al. 1999, Moore et. al. 1999).

An early solution developed to resolve this problem has been looking for galaxy counterparts linked to high-velocity HI clouds. Several low surface brightness dwarfs have been discovered using this technique. The increased spatial resolutions of the ALFALFA and GALFA HI surveys have identified a number of ultra-compact high-velocity HI clouds (UCHVCs), which could be linked to low mass, low surface brightness, gas rich galaxies, that reside in the Local Group, that would otherwise be difficult to detect in other optical surveys. I present a search for dwarf counterparts to selected (30) UCHVCs, using data collected by CTIO F5 DECam, Clay Megacam, APO spicam, as has data taken from the CFHT and GALEX archives.

I visually looked for UV-bright, diffuse, and blobby structures within 1 arcminute of each one of the UCHVCs. Two possible dwarf candidates were found. One near 290.37+66.23-115 (Figure 1), another near 8.88+62.16+281 (Figure 2).

In order to check whether or not these objects were dwarfs, I created color-magnitude diagrams of them (Figure 2). In order to do this, I used the SEXTRACTOR routine to pick out all stars in an image. In order to get picked, a star had to be 2 standard deviations brighter than the background of the image. I then took all the stars within 3 arcminutes of our UCHVC center coordinates. I then corrected these stars for milky way extinction, and plotted them on a $(g - r)_0$ vs r_0 diagram to get the 'Astronomical color' of these stars. In order to test for accurate stellar populations, I plotted isochrones at different distances. No clear stellar populations were found. I concluded that these two objects are most likely not in the local group.

Another technique I used to look for dwarfs was one outlined in Janesh et. al. 2015. I used the SEXTRACTOR routine to extract stars within 15 arcminute radius around each one of our UCHVCs. I then created a CMD filter using an isochrone for 13.5 Gyr, with $[Fe/H] = -2.49$. I made isochrones for different distances ranging from 0.25 Mpc to 3 Mpc in 0.25 Mpc increments, and picked isochrone that matched with the largest number of stars. In order to select candidate stars, I ran them through the CMD filter. A star would pass the filter if it or its error bars were consistent with the isochrone. Using the updated, filtered list of stars, I created spatial distribution diagrams. I binned the stars on to a bin size of 7 x 7 (250 by 250 bins total), and plotted them on to a ra vs dec diagram. I then smoothed the image by filtering it through a 1 Gaussian kernel. I then calculated a new image S, such that

$$S(x, y) = \frac{A(x, y) - \bar{A}}{A_\sigma}$$

where \bar{A} is the mean of image A, and A_σ is the standard deviation of image A, producing an image in units of standard deviations σ . Several regions did not have any stars that passed the CMD filter, so I left

them out. I made a colorbar cut at 5σ for clarity's sake. I then convolved the image with a 1σ gaussian kernel, in order to be able to see if there truly could be a clear dwarf galaxy resolved by this process. Nothing of significance was found however.

So far, a few low surface brightness dwarf galaxy counterparts to UCHVCs have been found. Even fewer objects have been confirmed to reside in the local group. There is currently about a 1 magnitude discrepancy between the theoretical prediction for the number of low mass dark matter halos, and the observed number of dwarfs in the local group.

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Janesh, W., Rhode, K.L., Salzer, R.J., et. al. 2015, ApJ, 844, 1

Maksym Zhelyeznyakov

I am a second year Physics student at Texas Tech University. I was born in Kryzhopil, Ukraine, where I lived until I was 11 years old, after which I moved to Germany for 2 years, and finally to El Paso, Texas at age 13. During my first year at Texas Tech, I was involved in Dr. David Sand's lab, doing research in Astrophysics. This fall I will be working with Dr. Luis Grave-de-Peralta doing research in quantum optics. I plan to pursue a PhD in physics, and do research at the university or industry level. In my spare time I like writing computer programs, reading, and building things.

1 Possible Dwarfs

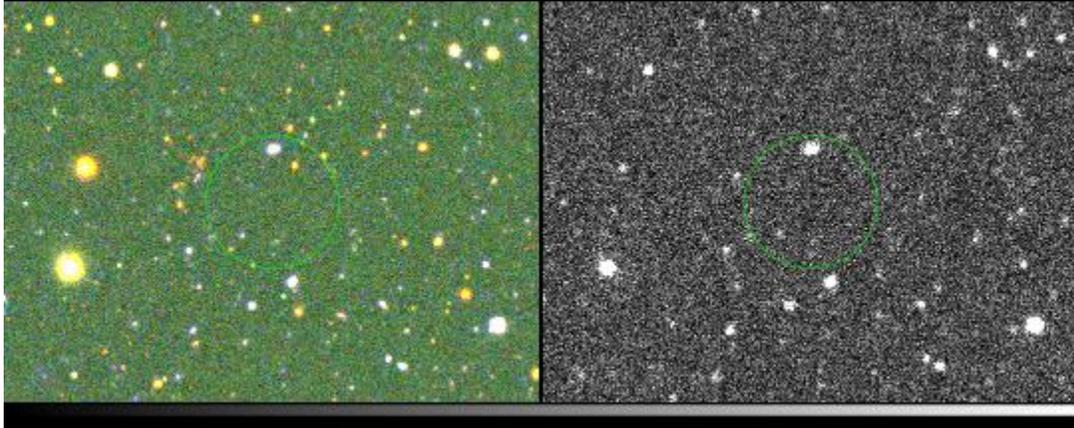


Fig. 1: HVC290.37+66.23-115. The image on the left was created by combining g , r , and UV bands. The g and r bands are taken using the CLAY F5/MEGACAM at 3600 s and 1800 s exposures on 2015-03-20 UTD. The depth of these images are $g \sim 24.9$ and $r \sim 24.4$ mags. The UV band (right) is was taken from the GALEX archive. The exposure time for the near field UV band is 2335 seconds. The object center is at RA 14:35:31.7 DEC 13:31:26 (J2000 sexadecimal coordinate system). This object is not likely associated with the UCHVC due to having a velocity inconsistent with the UCHVC.

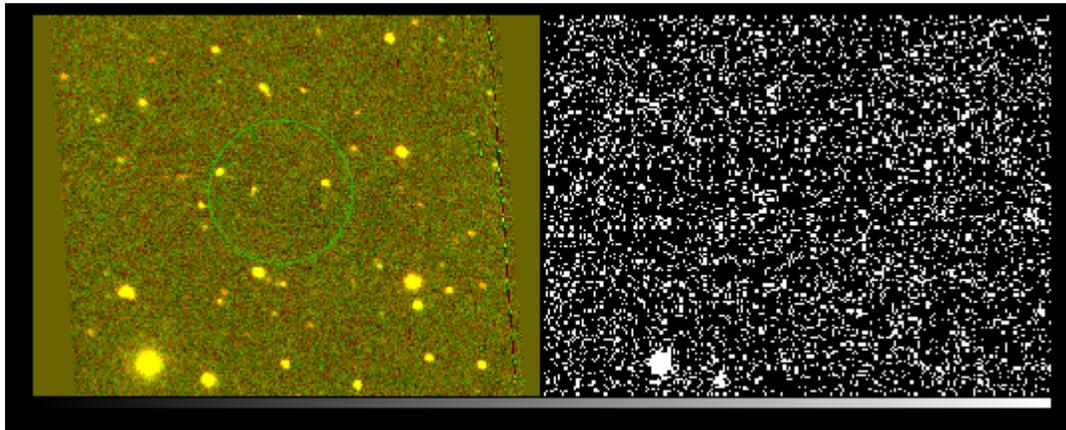


Fig. 2: HVC8pt88+62.16+281. The image was created by combining g and r from APO 3.5m/Spicam. The images were taken at 1200 s exposures with a depth of $r \sim 24.1$ and $g \sim 23.4$ mags. The UV image for this object was taken from the GALEX archive at a 103.05 s exposure. Both of these objects were selected for their blobby and diffuse nature, and because they are bright in the UV band. The green circles represent a 1 arcminute uncertainty in the position of the UCHVC. The center of this object is positioned at RA 14:35:31.7 DEC 13:31:26 (J2000 coordinate system). Although spectra were not taken for this object (yet), it is unlikely to be a dwarf due to the lack of a characteristic stellar population.

2 Selected Color Magnitude Diagrams

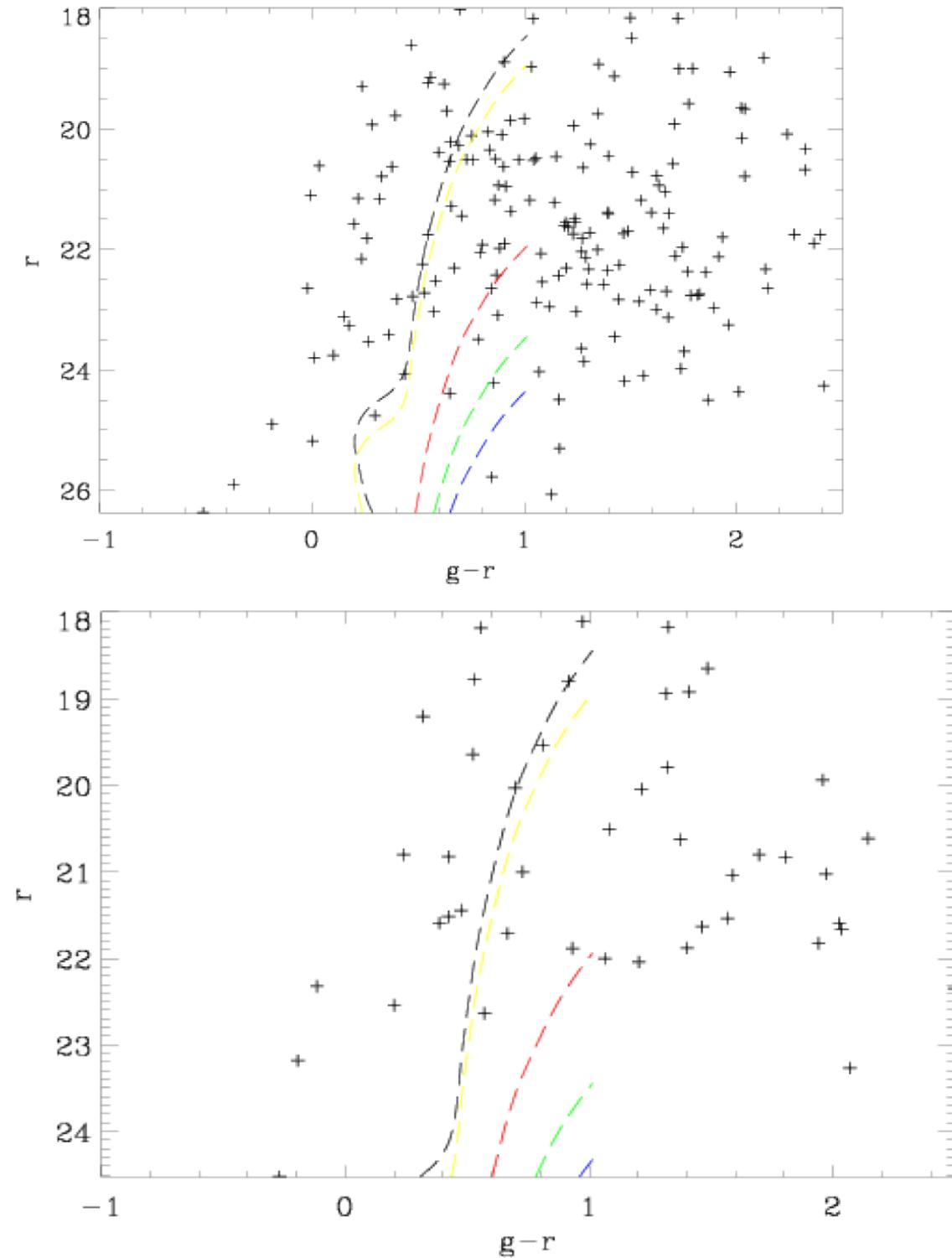


Fig. 3: Top: HVC290.37+66.23-115, Bottom: HVC8pt88+62.16+281. The lines correspond to isochrones at different distances. Black = 200 kpc, Yellow = 250 kpc, Red = 1 Mpc, Green = 2 Mpc, Blue = 3 Mpc. These isochrones correspond to a metallicity of -2.50 and an age of 13.5 Gyr.

Selected Stellar Maps

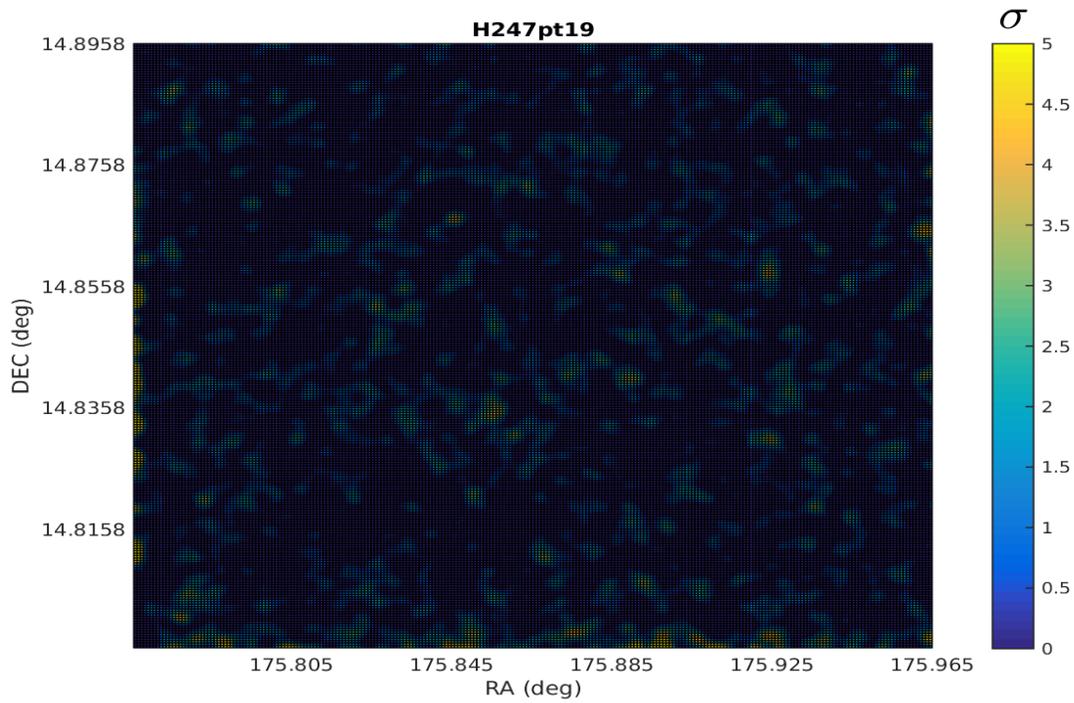
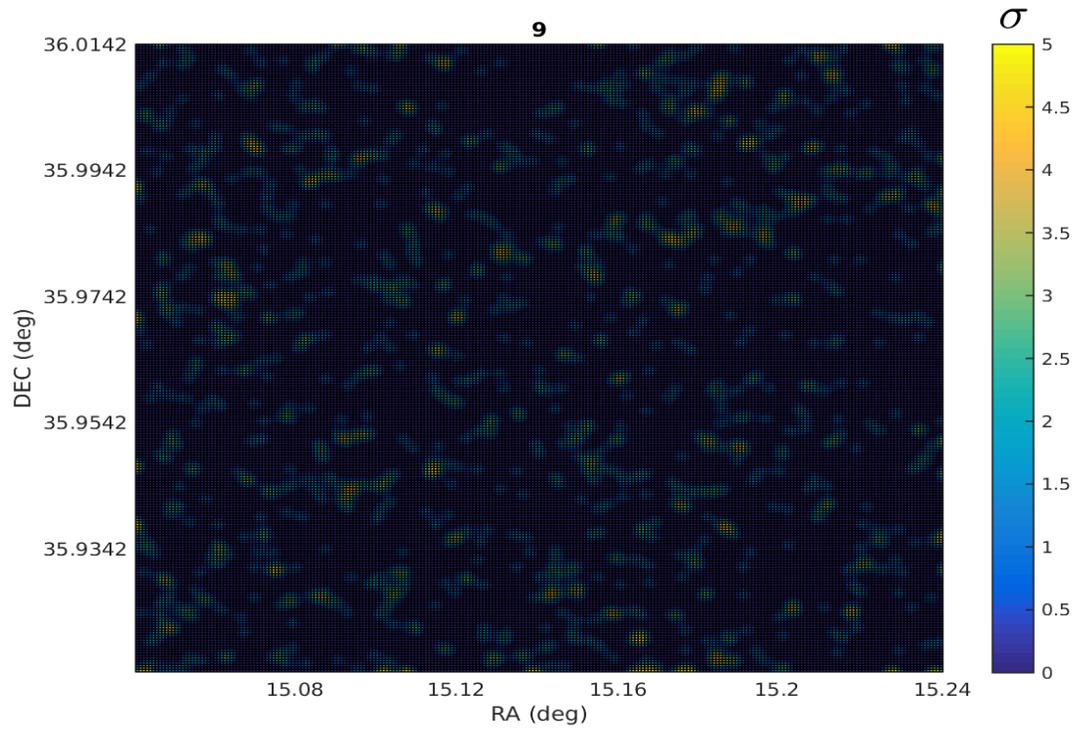


Fig. 4: Top: Stellar map of UCHVC at RA 01:08:29.6 DEC 37:45:00. Bottom Stellar map of HVC 247pt19.

Operation Counter Attack: An Offensive Approach to Cybersecurity

Joshua Hernandez

Mentors: Dr. Venkata N. Inukollu and Dr. Joseph E. Urban

Currently, the implementation of cyber attacks would be no different than the usage of a nuclear attack [1] resulting in one outcome, catastrophe. Unfortunately, the implementation of cyber attacks is inevitable. The digital world makes any states' nuclear weapons vulnerable to cyber attacks. Nuclear weapon codes are kept in a database, hence, being placed in the digital world. Thus, a successful cyber attack could render a state's ability to launch a nuclear attack, an example of this would be Iran with the usage of Stuxnet [2]. The outcome would then make this country vulnerable and unable to defend itself. The first state to create and implement a cyber attack towards nuclear weapons could essentially conquer other nations if not all of them, assuming that this was the states original intention.

The research provided was conducted through a time frame of two and a half months. The limited time forced a different approach to research itself. This approach was taking two already applied researches and then combining them into one counter attack. The outside perspective that brought this approach was that most researchers tend to start work from scratch or build off what they or someone else has already done. None, from what was noticed, ever approach research by combining multiple applied research and that is what was done here. Next, the research that was going to be done needed to revolve around a type of attack. Cybersecurity mostly revolves around defensive procedures, thus lead to the thought of a counter attack. This type of attack would occur only from a defensive stand point. This allows for a cautious and limited side towards a Cyberattack that would only be dangerous to the intruder.

The cyber attack that is being proposed will only be implemented as a counter attack. The counter attack will activate upon being a victim of an attack. The use of a Honey Pot (HP) [3] will prevent any damage from being taken and to understand the attacker's intentions. Next, a Botmaster Trace (BT) [4] will be implemented in order to insure that the counter attack is successful and accurately initiated by the defender.

The HP is designed for deception with the sole objective of allowing the attacker to believe that they have been successful with their objective [3]. The HP will notify if there is suspicious activity occurring. Once the notification is acknowledged, the BT will then be implemented to track where the attack is being placed [4]. Once the attacker's location is compromised, the counterattack will begin. The objective is to catch the attacker off guard by putting them on the defensive and additionally, making them aware that they are not the only ones with offensive capabilities.

With the advancement of technology, there will always be a way to infiltrate a network. Unfortunately, there is no ultimate defense and hackers are figuring out new ways to attack network systems. Additionally, the viewpoint of trying to prevent cyber attacks narrows the ability to portray other options. Though, one effective defensive option would be the HP. The HP would appear legitimate by being a mirror image of a production server of a corporation though filled with fake information [3]. This approach would allow attackers to view the server as something valuable and easily accessible. Thus, the strategy of using AAIHDP (An Architecture for Intrusion Protection using Honey Pot) will be used [3]. AAIHDP uses HP as an IP address trap [3] making it the perfect defensive protocol to catch an attacker.

Once an attacker is caught inside the HP, the BT will then be implemented to find their location. The login time and the lifetime of the IP address is traced from the service provider and if it crosses the

threshold it is grouped as a suspicious botnet [4]. The BT has been proven to only have an extremely small chance of failure [4], thus making it a trustworthy source. Additionally, the use of the BT will ensure that the attack is sent to the correct person. The necessity of the BT is imperative. Most cyber attackers have many ways to conceal their locations, the BT prevents them from hiding.

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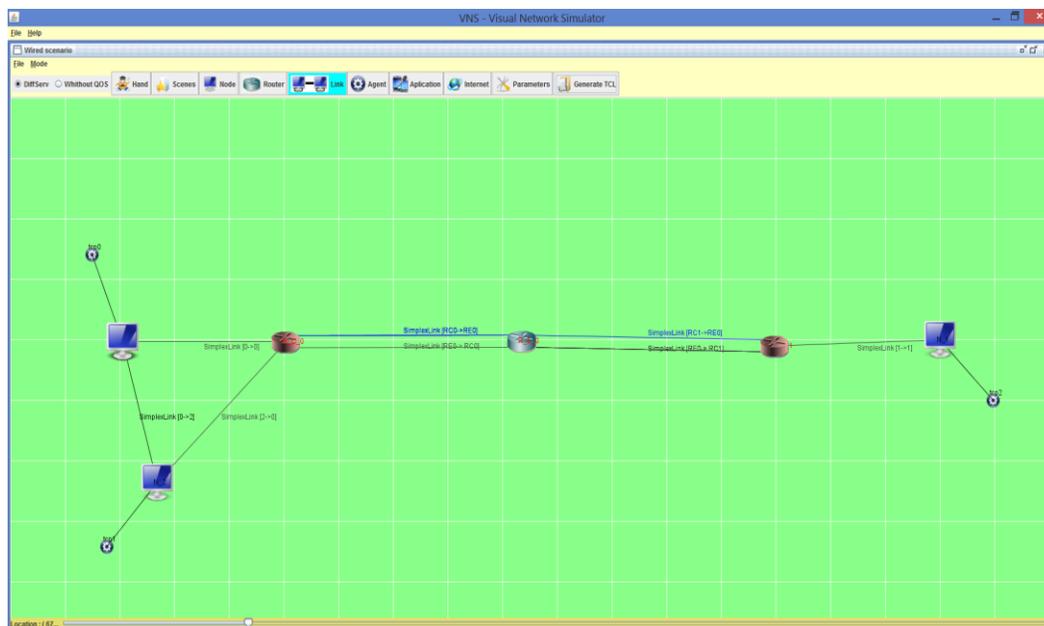


Figure 1: presents a basic format of the simulator.

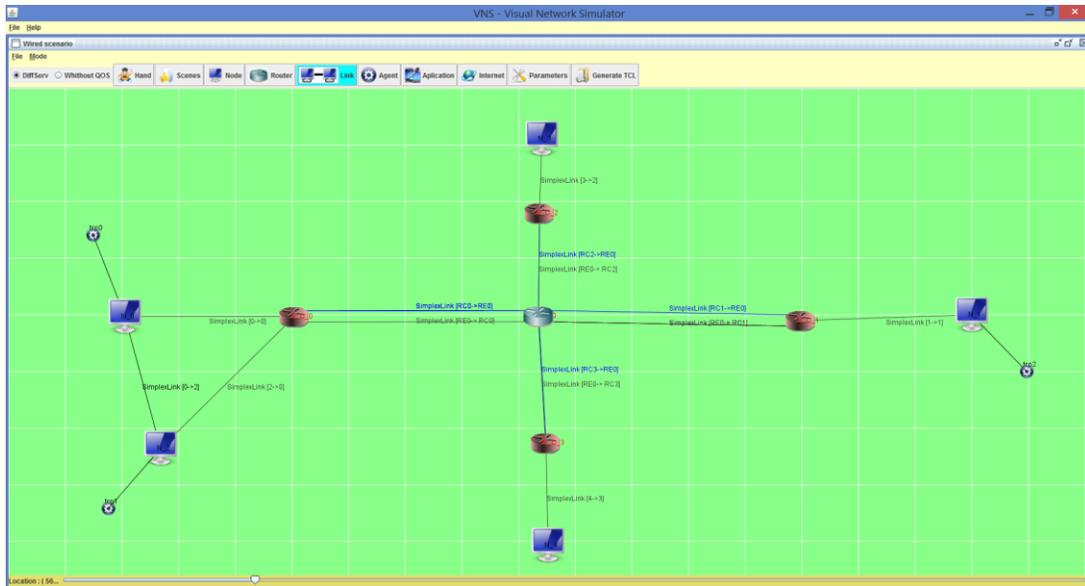


Figure 2: presents a more detailed presentation of the simulator.

Joshua Hernandez

I am a senior at Texas Tech University. I was first introduced to research this summer when I participated in the Research for Undergraduate Experience(REU) program this summer at Texas Tech University. I was able to do research on Cybersecurity, which is what I have always wanted to do since I was in high school. After graduation I plan to pursue the STEM MBA program at Texas Tech University. Anything after that is too far in the future for me to predict. In my free time, I like to go to the gym, listen to music while I write, or socialize with friends.

Evaluating the Television Show Daniel Tiger’s Neighborhood as a Video-based Model to Teach Social Skills to Children with Autism

Marisol C. Alonzo

Mentor: Dr. Wesley H. Dotson

Children with Autism Spectrum Disorders (ASD) have deficits in social communication and patterns of restrictive and repetitive behaviors, which results in difficulty adapting to social situations (DSM-V, 2013). Early behavioral intervention can effectively address these deficits (e.g., Eldevik, et al., 2010; Reichow, 2012). Interventions, such as video modeling and direct instruction, have been used to successfully teach social skills to children with ASD. The PBS show Daniel Tiger's Neighborhood includes many elements such as labeling and breaking down a targeted skill, modeling appropriate behaviors, and encouraging practice of the skill (Odom, et al, 2010). Daniel Tiger’s Neighborhood is a widely-watched children’s educational television show that incorporates many of the elements of effective video modeling into episodes.

The purpose of this study was to evaluate whether watching Daniel Tiger’s Neighborhood episodes can help children with ASD learn social skills taught in the show. Targeted social skills include sharing, calming down, time to stop playing, and trying new foods. Skills will be taught through direct instruction provided by the cartoon characters, along with a facilitator such as a teacher or parent. The percent of each skill performed correctly is measured by using a multiple baseline across skills design replicated across more participants with more skills. The study includes three phases: baseline, intervention, and maintenance.

The sessions for this study will be conducted in 6 x 10ft therapy rooms with one-way mirrors. The naturalistic probes will be conducted in room full of preferred toys for five minutes of free play. The intervention is conducted in another room with a laptop, video, table, and chairs. The intervention is 20 minutes, where the experimenter will present to prompt engagement with the Daniel Tiger video. The session will end with five minutes of free play to score naturalistic probes after the intervention, looking for changes in behavior. The participants for this study are young children with ASD from 3 – 7 years of age.

The results from the pilot study suggest that Daniel Tiger’s Neighborhood episodes can help children with ASD learn social skills. Effective interventions to teach social skills to children with ASD have included video modeling interventions, have been effective in teaching social skills to children with ASD (Ayres & Langone, 2005; Shukla-Mehta, et al., 2010).

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doi:10.1177/1088357609352901.

Marisol Alonzo

I am a senior in Human Development and Family Studies. I was introduced to research through the Plains Bridges over the Baccalaureate Program. I am going on my second year of research at the Burkhardt Center for Autism Education and Research. I am working with Mentor Dr. Wesley Dotson. The research is evaluating the television show Daniel Tiger's Neighborhood as a Video-based Model to Teach Social Skills to Children with Autism. I plan on pursuing a Master's Degree in Behavior Analysis, to receive a BCBA, to continue with a Ph. D, and autism research. My other interests include family time, church, and sports activities.



Figure 1

Waiting

- **Avoid whining, crying, arguing**

- **Avoid approaching the desired object**

- **Avoid touching the desired object**

- **Engage in another activity such as singing**



Figure 2

Calming Down

- **Avoid whining, crying, arguing when told it is time to calm down**
- **Hug/Squeeze self**
- **Inhale a deep breath**
- **Exhale a deep breath**



Figure 3

Trying New Foods

- **Touch the offered food item**
- **Put the food item in mouth**
- **Chew the food item**
- **Swallow the food item**





Figure 4

Stopping Play

- **Say “ok” or some equivalent acknowledgement**
- **Avoid whining, crying**
- **Stop playing within 15 seconds of the statement**
- **Walk to the experimenter**



Figure 5

Sharing

- **Child says “sure” or “yes”**
- **Child hands toy to asker within 10 seconds**
- **Child avoids whining, crying, screaming**
- **Child avoids taking toy back from asker**



Engagement on Daniel Tiger’s Neighborhood involving kids with Autism

Amanda Lund

Mentor: Dr. Wesley Dotson

Children on the Autism Spectrum struggle socially. Various interventions have been developed to help them overcome this social issue. One form of intervention is video modeling. The popular PBS show “Daniel Tiger’s Neighborhood” shows many elements of video modeling including targeting social skills and providing models of the skill. Such skills from this TV program include stopping play when it’s time to go, waiting, trying new foods and sharing. Prior pilot research has evaluated this show and indicated that children with Autism Spectrum Disorder can acquire targeted social skills by watching the episodes.

The current study examined the specific behavior of the children with Autism Spectrum disorder who participated in the pilot study (Dotson, et al., in review) described above. There were two goals of the study: 1) to determine what behaviors a child engaged in while watching the program and 2) determine whether those behaviors predicted rates of the children learning the targeted skill. Four behaviors were measured using a 15 second partial interval recording system. The presence or absence of engagement, orientation, off-task, and challenging behaviors were scored every 15 seconds for the duration of each episode.

Two 5yr old children with ASD participated in the study. During the episode, both participants were generally engaged with the episode and remained oriented towards it while it was playing. Higher rates of engagement with “Daniel Tiger’s Neighborhood” were positively correlated with quicker acquisition of targeted skills. These results support the conclusion that watching the episodes of Daniel Tiger’s Neighborhood was likely responsible for the learning of the children because as they were more engaged in the episodes their rate of learning increased.

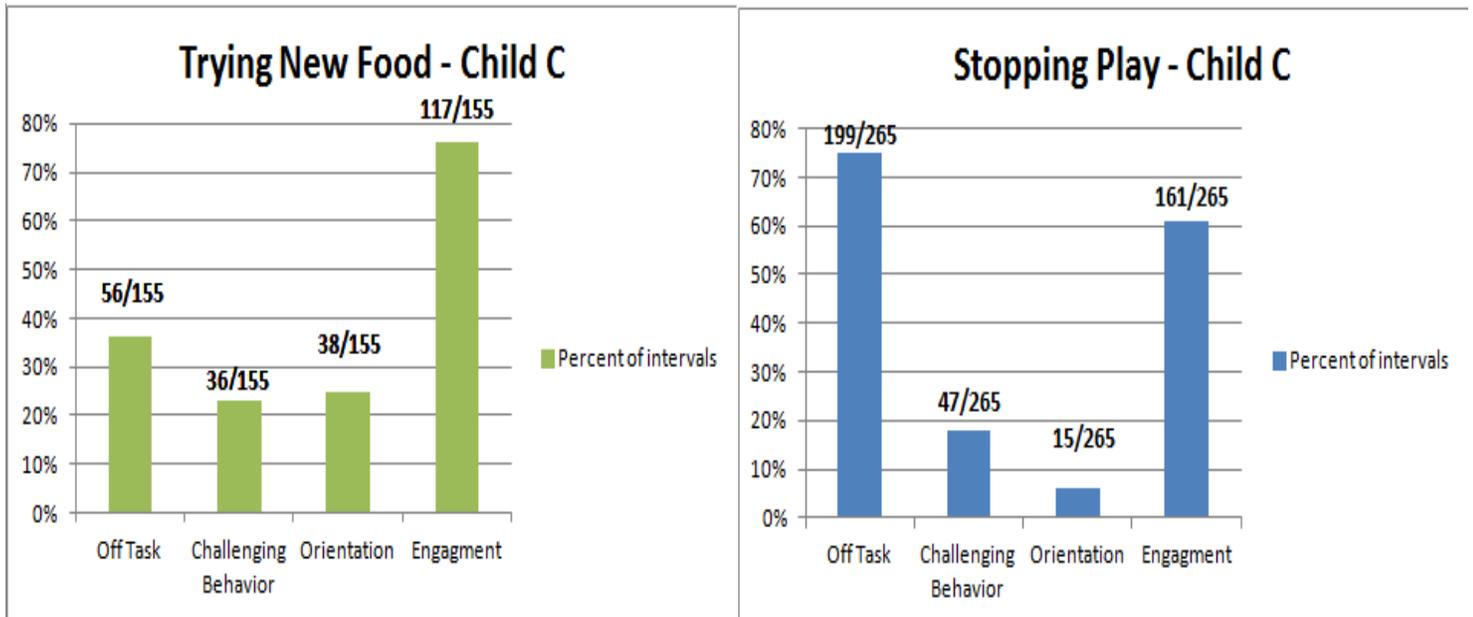


Figure 1: Behaviors of child C while watching episodes of Daniel Tiger’s Neighborhood.

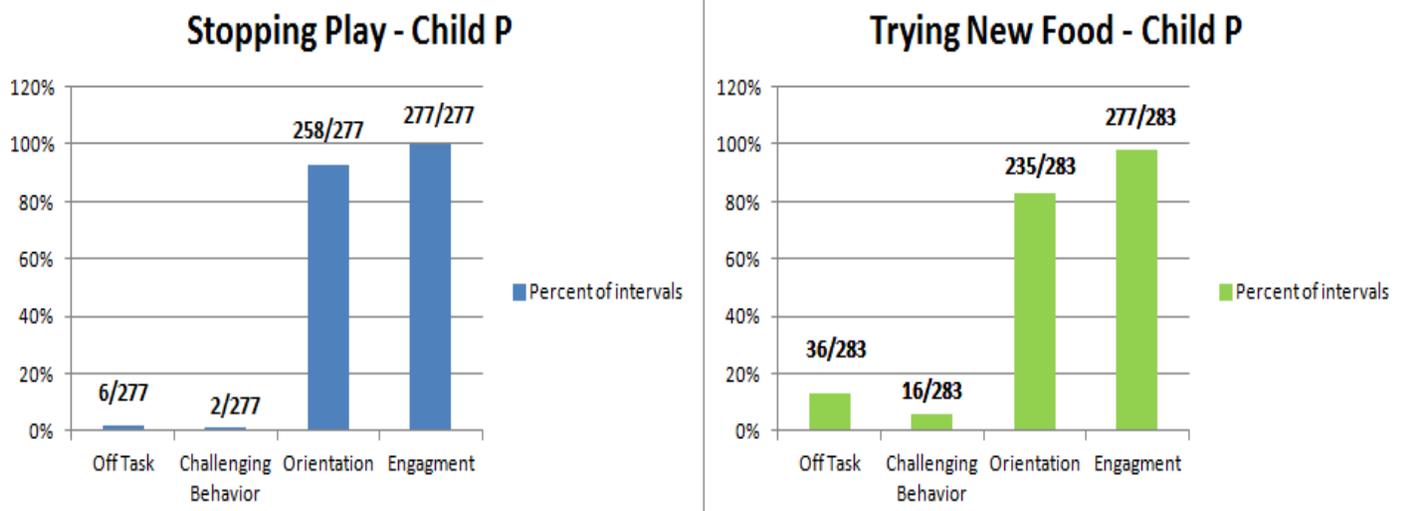


Figure 2: Behaviors of child P while watching episodes of Daniel Tiger’s Neighborhood.

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Amanda Lund

I am a junior majoring in Multidisciplinary Studies with a focus in Special Education. I have been involved with the Burkhardt Center for Autism Education and Research since the fall of my sophomore year. I am very interested in research involving social skills among kids with autism. My goal in life professionally is to give kids with special needs a better quality of life in order to succeed in the work force.