



TEXAS TECH UNIVERSITY



# **TTU HHMI Scholars Research Log 2013**

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## Table of Contents

TTU/HHMI Scholar	Major	Research Title	Mentor	Page
Banuelas, Victoria	ESS	Effects of Gender on ACL Injuries	James Yang	4
Brown, Taylor	Biology	Drought Tolerance in Adult versus Resprout Oaks in West Texas	Dylan Schwilk	8
Carrillo, Elias	ESS	Assessing the Development of the Standing Long Jump in Children	David Stodden	9
Cueva, Angel	Microbiology Pre-Dental	Is Quorum Sensing Involved in the Pathogenesis of <i>Pseudomonas Aeruginosa</i> in Chronic Wound Infections?	Kendra Rumbaugh	12
Dean, Ryan	Biology	The Removal of an ImpE1 Gene Through the Use of a Minus P Element	Jeffrey Thomas	14
DeLeon, Stephanie	Biology	Population Dynamics of <i>Pseudomonas Aeruginosa</i> and <i>Staphylococcus Aureus</i> in an <i>in vitro</i> Wound Environment	Kendra Rumbaugh	16
Jarrett, Philip	Cell & Molecular Biology	Development of Transgenic Crops to Enhance Abiotic Stress Response and Crop Yield	Hong Zhang	20
Martinez-Marin, Dalia	Cell & Molecular Biology	PEDF Gene Therapy Using Bone Marrow-Derived Macrophages for Advanced Prostate Cancer	Stephanie Filleur	22
Nafees, Saba	Mathematics	Modeling Positions Coupled to F508, Site of Chief CF Causing Mutation	Sean Rice	25
Norton, Alexander	Biology	Effects of Ocean Acidification on Red Drum Larvae Otoliths	Sandra Diamond	28
Nunez, Maria	Zoology	Toxicity of Sediments from Urban Playa Wetlands: <i>Hyallela Azteca</i> Survival	Jonathan Maul	31
Ortega, Graysen	Food Science	Prevalence of <i>Escherichia Coli</i> Non-O157:H7 STECs in Beef in Mexico	Mindy Brashears	34

Ortega, Katelyn	FoodScience	Prevalence of <i>Campylobacter</i> in Retail Ground Beef as Influenced by Packaging Type and Fat Content	Mindy Brashears	38
Pleasant, Stephanie	Chemistry	Methods for Determining the Source of Chemical Threat Agents	Carlos Fraga	43
Rodriguez, Chris	<i>In memorium</i>	Drought Tolerance in Adult versus Resprout Oaks in West Texas	Dylan Schwilk	45
Rojas, Daniela	Microbiology	Effects of Fruit Flavonoid on Steroidogenesis & StAR Gene Expression	Akhilesh Pandey	50
Sparks, Hayley	Microbiology	Role of <i>Clostridium perfringens</i> in Multi-Species Wound Biofilms	Kendra Rumbaugh	53
Stilwell, Jessica	Electrical Engineering	TYRE: An <i>in silico</i> Simulator of Yeast Population Dynamics	Brandt Schneider	57
Suteria, Naureen	Chemical Engineering	Hydrodynamic Resistance of a Train of Confined Microfluidic Droplets	Siva Vanapalli	61
Willms, Joshua	Biology and Math	Effects of Ocean Acidification on Otolith Formation in Red Drum, <i>Sciaenops ocellatus</i>	Sandra Diamond	65
Woollends, Alex	Mathematics	Using a Public Content Management System (CMS) to Mitigate Security Risks	Brian Enderson	68

Effects of Gender on ACL Injuries  
Victoria K .Bañuelas  
Mentor: Dr. James Yang

The anterior cruciate ligament tear is the most common injury occurring to the knee. According to the American Academy of Orthopaedic Surgeons/American Association of Orthopaedic Surgeons, about 200,000 ACL tears occur in the United States each year. The ACL is one of the four major ligaments supporting the knee. It is connected to the femur (thigh bone) and the tibia (shin bone), which serves to provide stability to the knee and prevent the knee from 'giving out'. Harm to this ligament occurs usually during noncontact and pivoting circumstances in athletes.

An increase in females participating in sports in the past two decades combined with the two- to ten-fold higher injury rate in females than in males has raised a recent awareness of both the physical and financial strains this fact has caused (Moul, 1998). About 1 billion dollars goes into treating ACL tears in varsity female athletes alone (Giugliano, 2007). This gender difference in ACL injuries requires further research to address the risk factors contributing to the high volume of female ACL incidences and to developing a preventative strategy for this problem.

Two types of factors contribute to ACL injuries: intrinsic factors, which include anatomical differences in females such as the angle difference in the pelvic girdle and hormonal influences during a females menstrual cycle; and, extrinsic factors, like the environmental conditions in which an athlete is playing a particular sport or biomechanical influences of an athlete's equipment/tools (Silver, 2005).

To investigate these issues, we will construct a knee model and stress the ACL ligament using various approaches specific to the factors mentioned above. Once we have collected the data pertaining to the amount of valgus torque, i.e., the force and impact that the knee can take before tearing the ACL, we will use this to then transition into creating an orthopedic device for ACL prevention in female athletes.

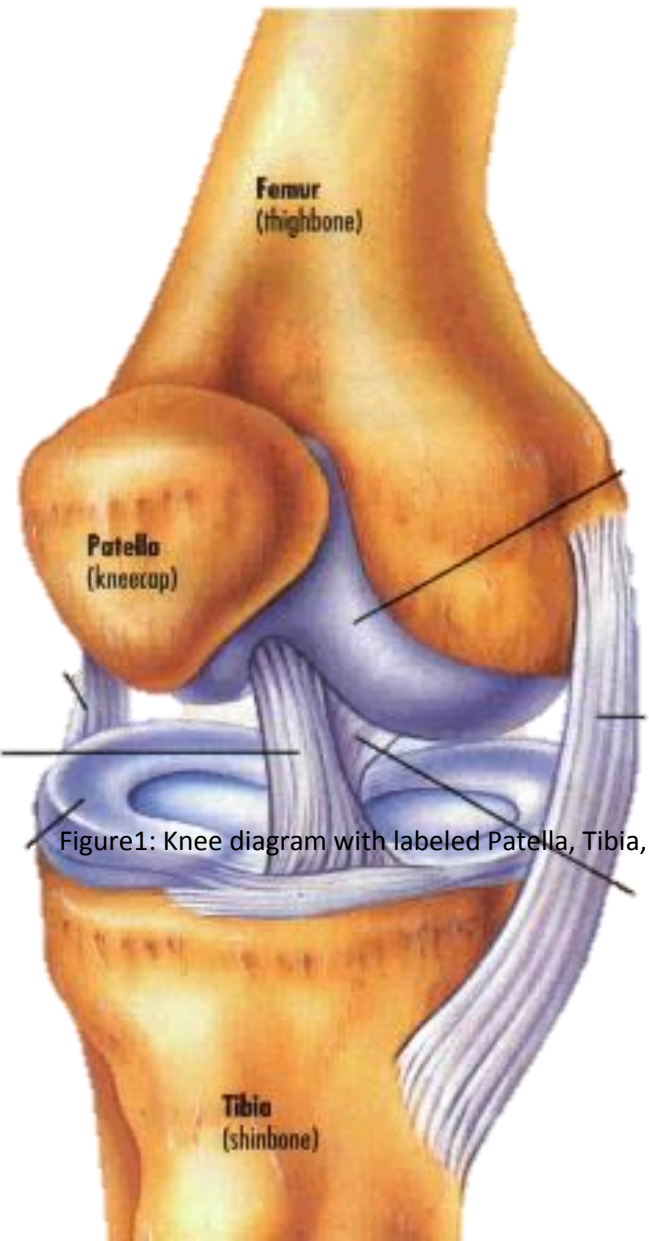
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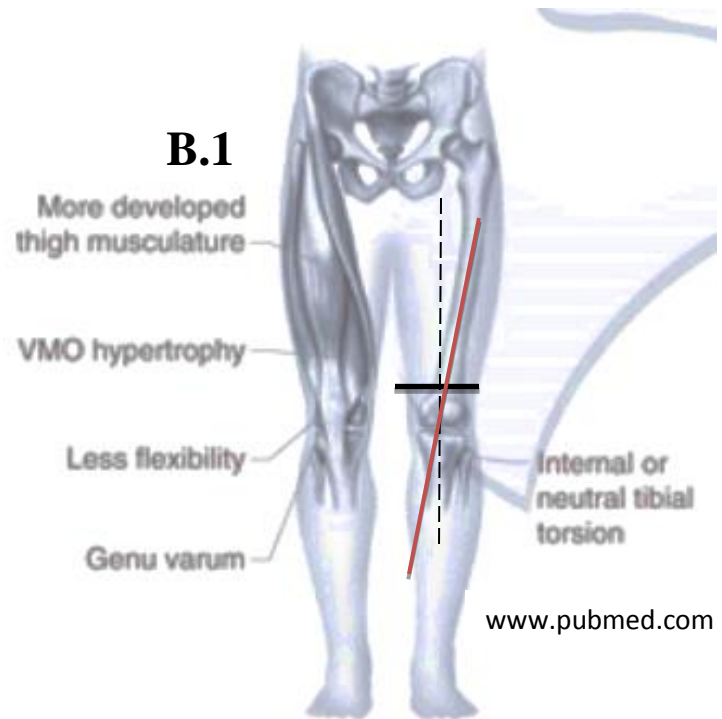
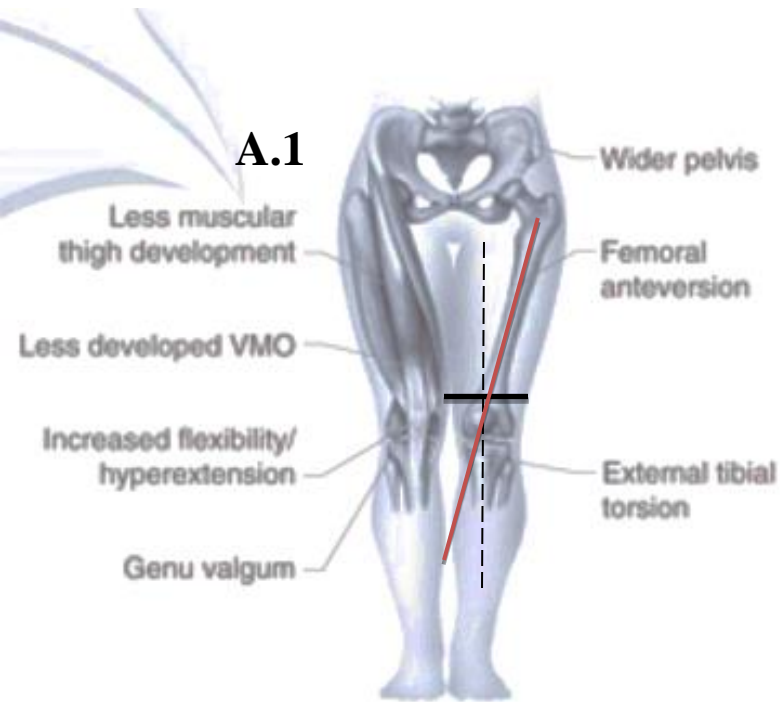
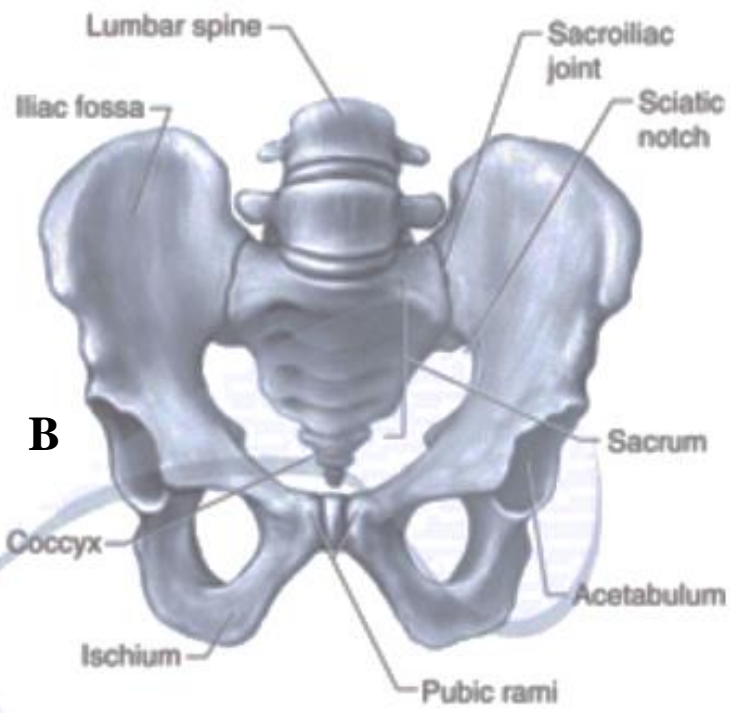
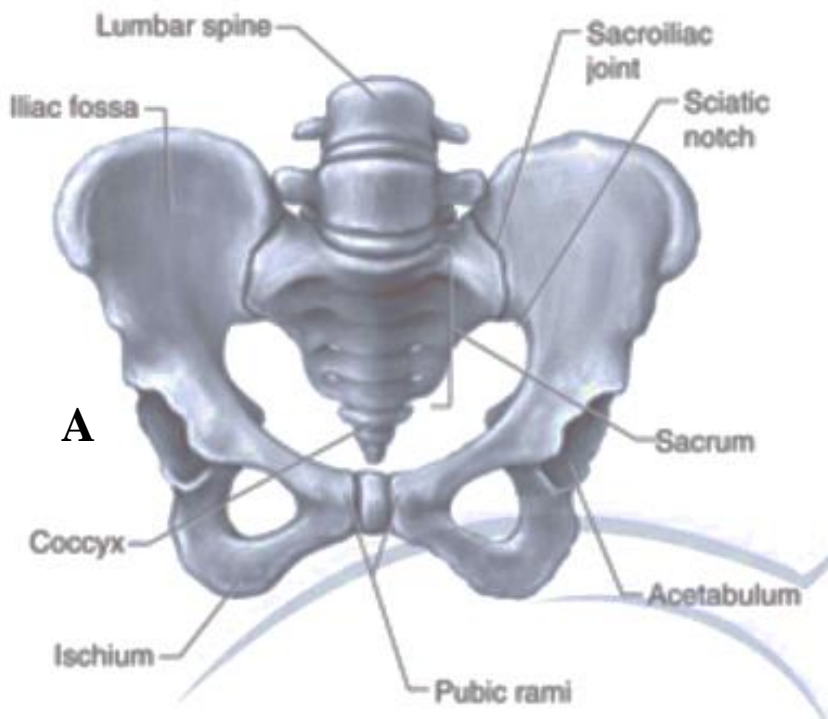
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**(ACL) Anterior Cruciate Ligament**

Figure1: Knee diagram with labeled Patella, Tibia, Femur and ACL.

Health, Medicine and Anatomy  
Reference Pictures



www.pubmed.com

Figure 2: (a) Anterior view of female pelvic girdle (a.1) femoral angle from acetabulum to the tibia  $\geq 16^\circ$ .  
 (b) Anterior view of male pelvic girdle (b.1) femoral angle from acetabulum to the tibia  $\leq 12^\circ$ .

**Victoria K .Bañuelas**

I am a fourth-year student in the Health Exercise and Sports Science department with a double minor in Biology and Chemistry. My hometown is El Paso, Texas and some of my interest include traveling, exercising and dancing. I have worked in a biology lab researching behavioral ecology for the past year and currently in Dr. Jamaes Yang Human-Centric Design Research Lab where I'm heading my own project in developing novel medical devices to improve the quality of life for patients. My future goal is to attend medical school and become an Orthopedic Surgeon and specialize in Sports Medicine.

## Drought Tolerance in Adult versus Resprout Oaks in West Texas

Tailor Brown

Mentor: Dr. Dylan Schwilk

Drought is a deciding factor of tree distribution. Here in west Texas this is a concern. Drought combined with global warming are causing trees to shift distribution and be pushed to their limits. Trees differ in drought susceptibility throughout their different life stages. There is a trade off, at times, when the trees need to invest in either safety or efficiency. Adult trees invest more in safety from drought and fire. Basal resprout trees should invest more in efficiency due to having more root mass underground than material above ground.

To test this model we examined 5 different oak species in the sky islands of west Texas. We used adult trees having pre-fire conditions in 2010, and basally resprouted trees whose adult trees burned in two forest fires from 2011 and 2012. The stems collected were put through a series of steps in order to obtain measurements of conductivity. The first stage hydrates the stem to non-drought conditions. We create a maximum flow rate by pushing a water solution contained in tubing through the stem in order to push out any emboli. The second stage simulates drought using a centrifuge with a custom made rotor. With this we are able to increase specific tensions on the xylem, which is what occurs inside the plant during drought. In the third stage we force the water solution through the xylem using a known pressure and measure the flow rate of solution on a balance. Using programming, statistics and physics we are able to measure conductivity and eventually make a conductivity curve for each stem.

We spent spring 2013 and the first half of summer 2013 trouble shooting and designing methods for every step of our process. During the summer of 2013 we were able to collect and measure 4 individuals across the 5 species of oaks. Thus we have rough data that does not yet have the absolute rates of conductivity. There is still future work that needs to be done statistically in order to finalize our results. The preliminary data when compared to the known adult data shows that the resprouts actually do have a slightly greater conductivity than the adults.

### **Tailor Brown**

I have thankfully completed two years at Texas Tech, and have been researching for several months. I am majoring in biology with medical intentions. My interests range from one end of the spectrum to the other, but I am currently in an ecophysiology lab lead by Dr. Schwilk. Our project works with oak trees across arid mountain ranges here in west Texas. These oaks are used to look at drought responses, plant behavior, and traits. Although I do adore learning in lab and school, occasionally there is personal time to enjoy gardening, eating, sleeping, and building relationships.

## Assessing the Development of the Standing Long Jump in Children

Elias Carrillo

Mentor: Dr. David Stodden

Motor skill competence (MSC) is associated with health related fitness, physical activity, and body composition in children, adolescents and young adults. It was hypothesized in this study that a more comprehensive strategy using both process- and product- oriented assessments would provide a more complete picture of the associations and probable impact of MSC on various health-related outcomes. Research examining the relationships between movement patterns and their outcome in various skills has not been comprehensively addressed. Thus, our understanding of these relationships is lacking and needs to be addressed. The main purpose of this project is to examine the relationship between standing long jump product (jump distance) and standing long jump process (developmental sequences, both take-off and landing) in children ages 4-12.

Standing long jump performance requires an individual to combine developmentally-based movement patterns into one motor skill performance. This includes movements directly involved in preparatory take-off and landing phases of a standing long jump. Performance of certain motor skills is associated with differences in growth (stature) and maturity (proximity to pubescence) in both males and females. SLJ is considered a test of muscular coordination and power. Proper performance requires adequate strength and coordination to project the body forward in both a controlled and explosive manner to maximize displacement between take-off and landing positions. Thus, it is important to acknowledge the influence of stature (height) and weight on product scores. Body weight, when age and stature are controlled, has a negative influence on performance of whole body projection tasks; whereas stature positively correlates with performance. Only one published study has partially addressed the landing phases of SLJ. In efforts to combat this, this project used a recently submitted developmental sequence focused on the landing components of the SLJ that was proposed for children ages 4-12.

We were able to collect our data from three samples that ranged from ages 4-12. Participants have been separated by age into nine groups. The majority of the collection was done in Lubbock ISD schools and is still in an ongoing process. We measured maximum jump distance/height (SLJ) from a total of five trials per participant. Distance jumped was measured to the nearest centimeter to ensure accuracy of measuring. The sum of the modes for both take-off and landing components was used for data analysis to provide a comprehensive assessment of standing long jump movement patterns. Digital video recorders captured each trial from the sagittal plane to allow for analysis of developmental sequence level for each component of the movement. These video clips were then analyzed at 60 frames per second using the *Dartfish* software. The data collection process for the overall study is still an ongoing process.

*Developmental Sequence of the Standing Long Jump Takeoff for Body Components*

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Leg action

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Level 1	Stepping out; a one-footed take-off.
Level 2	Knee extension precedes heels up.
Level 3	Knee extension and heels up simultaneously.
Level 4	Knee extension follows heels up.

Arm action

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Level 1	No arm action; arms remain immobile throughout propulsive phase; may exhibit shoulder girdle retraction "(winging)" close to take-off.
Level 2	Shoulder (flexion only; arms remain immobile during lower extremity flexion. Shoulder flexion occurs with lower extremity extension; some shoulder abduction may be seen.
Level 3	Incomplete biphasic arm action; shoulder hyperextension occurs during lower extremity flexion; shoulder flexion occurs with lower extremity extension; shoulder flexion incomplete (less than 160°) at take-off.
Level 4	Complete biphasic arm action; same as Level III except that shoulder flexion is complete (greater than 160°) at take-off.

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Note: From "A developmental sequence of the standing long jump," by Clark, J.E., & Phillips, S.J. 1985, In J.E. Clark & J.H. Humphrey (Eds.), *Motor development: current selected research*, Vol. 1, pp. 76-85.

*Hypothesized Developmental Sequence for Standing Long Jump of Body Components*

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Shank action	
Level 1	Anterior shank angle at contact is acute or approximately perpendicular ( $\leq 95^\circ$ ) to the ground
Level 2	Anterior shank angle at contact is clearly obtuse relative to the ground

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Foot action	
Level 1	One-footed landing: leg configuration during flight is often asymmetrical
Level 2	Symmetrical neutral: Two-footed landing; ankle is plantarflexed or neutral just prior to contact resulting in a forefoot or flat-footed landing
Level 3	Symmetrical dorsiflexed: Two-footed landing; ankle is actively dorsiflexed at contact resulting in a distinct heel landing

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Arm Action	
Level 1	No reach; arms wing during flight and act only to parachute* during landing
Level 2	Slight forward reach; shoulders flex to bring arms in front of the body during flight but the reach is abandoned as shoulders abduct to parachute; often dropped straight in line with the trunk at contact
Level 3	Full forward reach; shoulder flexion of $\geq 90^\circ$ relative to the trunk is maintained until contact
Level 4	Arms sweep; simultaneous forceful shoulder extension and hip flexion; arms are forcefully thrown down and back into full shoulder extension just prior to contact in order to pitch the legs up

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\*Parachute: to use for balance only

**Elias Carrillo**

I have had the opportunity to be involved in several interesting things while in college. At SPC I became interested in science and began to do undergraduate research at Texas Tech University (TTU). I transferred to TTU in the Fall of 2012. At TTU I continued undergraduate research with my Mentor Dr. Stodden in the Exercise and Sports Science Department before joining Howard Hughes Medical Institute (HHMI). I am currently doing undergraduate research in the ESS department. My future plans are to pursue a career in Motor Behavior and attend graduate school. I will be finishing up my Bachelor's degree in Exercise and Sports Science in May.

# Is Quorum Sensing Involved in the Pathogenesis of *Pseudomonas aeruginosa* in Chronic Wound Infections?

Angel R. Cueva

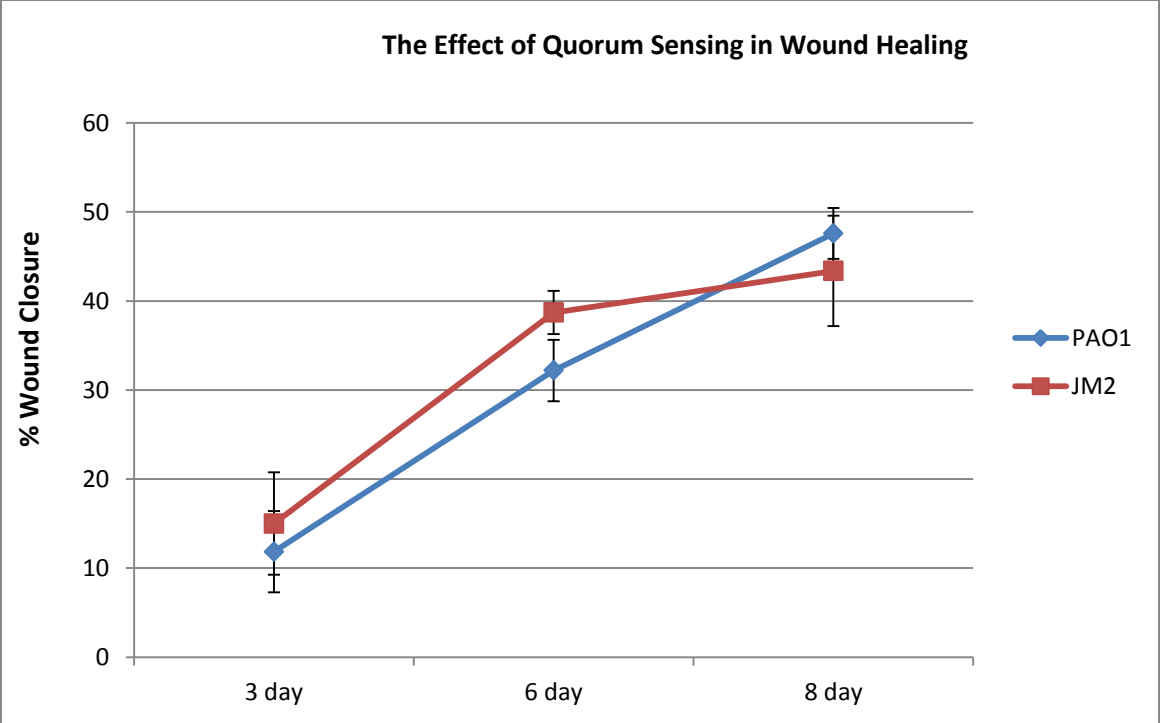
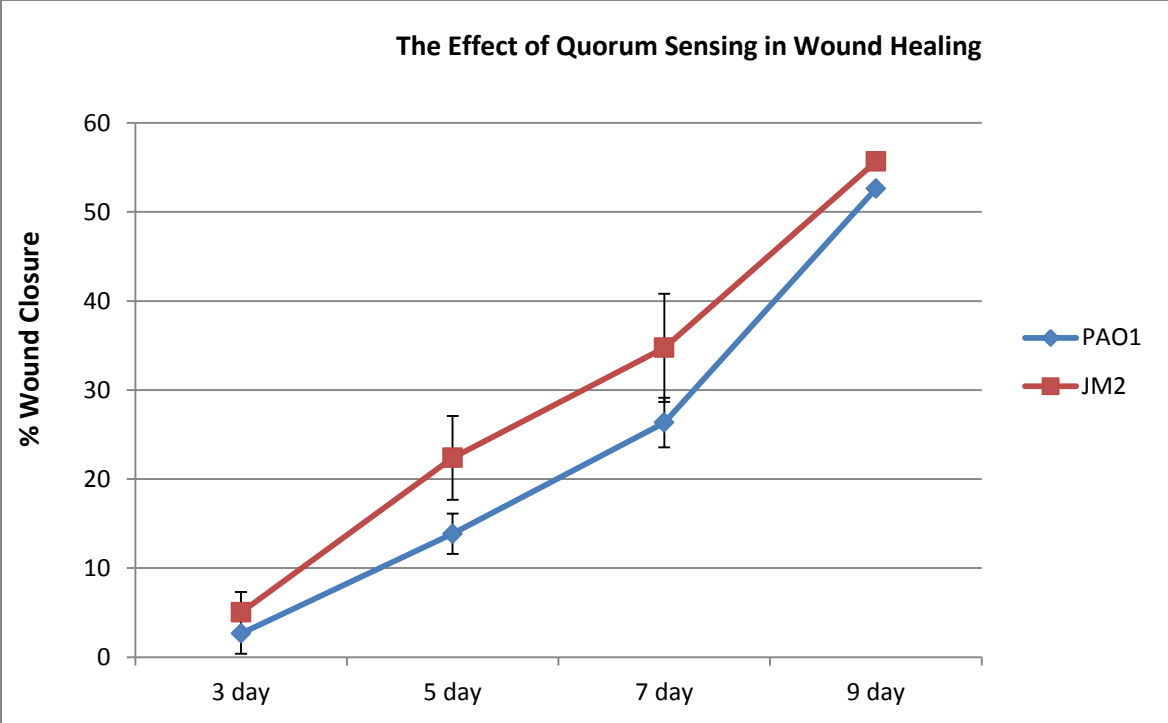
(with contributions from Andrew Armstrong)

Mentor: Dr. Kendra Rumbaugh

*Pseudomonas aeruginosa* is a gram negative bacterial species which is ubiquitous and a common environmental contaminant, and can be a transient colonizer of human skin. *P. aeruginosa* is associated with a number of acute and chronic infections, such as burn wounds, cystic fibrosis, pneumonia, and diabetic foot ulcers which contribute greatly to high rates of morbidity and mortality globally. The production of some virulence factors that contribute to the pathogenesis of *P. aeruginosa* is regulated by cell to cell signaling, or quorum sensing (QS). *P. aeruginosa* QS involves three distinct regulatory systems which alter gene expression in response to the amount of specific chemical signals, or autoinducers, within an environment. We have previously demonstrated that QS is essential to the pathogenesis of *P. aeruginosa* in burn wound infections; however, little has been elucidated about the role of QS in the chronic wound environment. Likewise, the efficacy of QS inhibiting compounds as effective treatments for chronic wound infections has not been investigated.

Our role is to investigate QS in the chronic wound environment. We utilize a chronic wound mouse model for in vivo experiments. To create the chronic wound, the mice are first anesthetized with an intraperitoneal injection of Nembutal, their backs are shaven, and the remaining hair is removed with Nair. The mice are then given a localized injection of Lidocaine, then a full-thickness surgical excision of less than 2cm is given. A surgical op-site bandage is applied to the wound through which either *P. aeruginosa* wild-type strain PA01 or PA quorum sensing mutant JM2 is injected. The progression of the chronic wound is monitored as is the healing of the wound itself. Healing progression is determined by percent wound closure over time.

Our most recent results show that the quorum sensing negative mutant JM2 has similar percent wound closure to the wild type PA01. This data could imply that quorum sensing may not play a significant role in chronic infections, unexpected based on our results of QS in burn wounds. There is unpublished data in our lab that shows blood proteins, present in chronic wounds, can alter and possibly suppress QS. In addition, there is evidence in the literature that QS inhibitors in vivo are not effective, despite high efficacy in vitro. This gives more evidence to the possible decreased role of QS in the chronic wound environment



**Angel R. Cueva**

A fourth-year student in microbiology, I previously worked in Dr. Kai Zhang’s lab (two summers and a school year), but switched to Dr. Kendra Rumbaugh’s lab at the beginning of August. I am a pre-dental major and my general interests are in pathology. I am likely to pursue a career in science, as a Doctor of Dental Surgery or a research scientist. My other interests are the outdoors.

The Removal of an ImpE1 Gene by Imprecise Excision of a Minos Element  
Ryan Dean  
Mentor: Dr. Jeffrey Thomas

*Ecdysone-inducible gene E1 (ImpE1)* is a gene with a role in the embryonic development of the cephalic furrow in *Drosophila melanogaster* (S. Russell, JH Thomas). Located on the left arm of the 3<sup>rd</sup> chromosome, this large protein coding gene's molecular function remains unknown. Research on the *ImpE1* gene's molecular function will help characterize the gene itself and lead to further understanding of *Drosophila*'s cephalic furrow formation. Transposon insertions in *Drosophila* are suitable for gene tagging, gene disruption and many other genetic engineering applications, making it an appropriate medium for *ImpE1*'s investigation (Bachmann et al., 2008). The P-Element transposon is a favorite for gene disruption projects among many biologists studying *Drosophila* (AC. Spradling et al). Although there are no P-element insertions in the *ImpE1* gene, there are two other transposon insertions in *ImpE1*, a PiggyBac insertion and a Minos element insertion. The PiggyBac insertion disrupts *ImpE1* and causes a defect in the cephalic furrow formation (S. Russell, B. Siddiqi, JH Thomas). However, the PiggyBac insertion does not completely disrupt *ImpE-1* gene activity (S. Subedi, JH Thomas). The Minos Element insertion is in an intron and does not cause a cephalic furrow defect. We will create a deletion in the *ImpE1* gene via imprecise transposon excision of the Minos transposon insertion (Metaxakis et al. 2005).

To do this, the transposon must be crossed with a transposase in order to be excised. Most of the excisions will be precise, and will not disrupt the gene. Others will be imprecise and create a deletion in the *ImpE1* gene when part of the genome is removed along with the transposon. After excision, the *ImpE1* gene will be amplified using PCR to determine whether the excision was imprecise and if so, to indicate the extent of the deletion of *ImpE1* genomic DNA. After PCR, sequence data will be compared with that of the wild type to determine whether an exon has been deleted.

We have identified nine excisions, none of which were imprecise. It is expected that the probability of the Minos transposon's imprecise excision from the genome is low. In addition, the probability of the entire *ImpE1* coding sequence being excised is even lower. Thus, this investigation requires several hundred replicates in hopes of yielding a fly with the desired mutant sequence.

Minos Transposon (without Transposase) × P Element Transposon (Engineered Minos Transposon)

#### References

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Turner, F.R. and Mahowald, A.P. (1979). Scanning electron microscopy of *Drosophila melanogaster* embryogenesis. III. Formation of the head and caudal segments. *Dev. Biol.* 68: 96-109



Figure 1. The formation of the cephalic furrow in a stage 7 *Drosophila melanogaster*

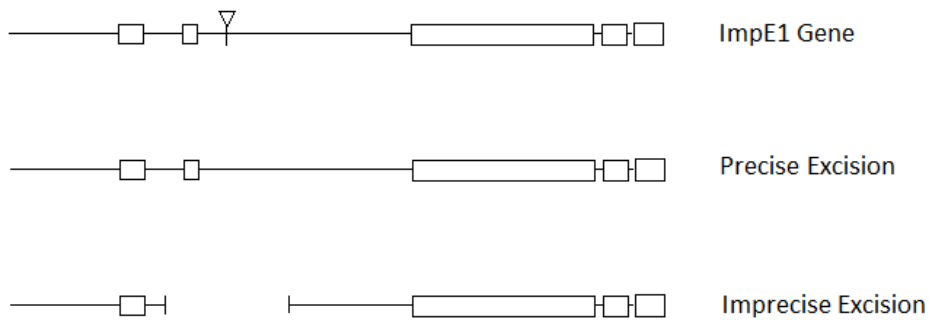


Figure 2. Comparison of the ImpE1 gene with and without precise transposon excision

**Ryan Dean**

I am a fourth-year student majoring in Biology at Texas Tech University. I have been researching in Dr. Jeffrey Thomas's lab for approximately 9 months, concentrating on the fields of genetics and developmental biology. I am pursuing a future in medicine and I look forward to incorporating the valued principles of research into my practices as a physician. I am passionate about outdoor activities, golf and the Boston Red Sox.

Population Dynamics of *Pseudomonas Aeruginosa* and *Staphylococcus Aureus* in an *in vitro* Wound Environment

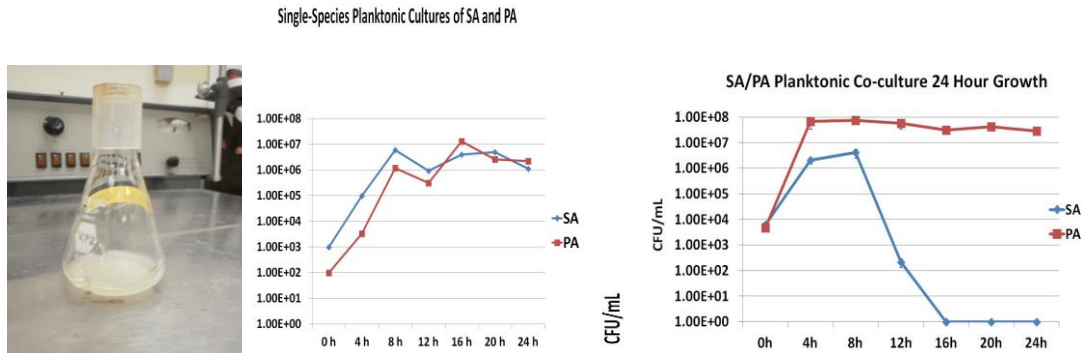
Stephanie DeLeon

Mentor: Dr. Kendra Rumbaugh

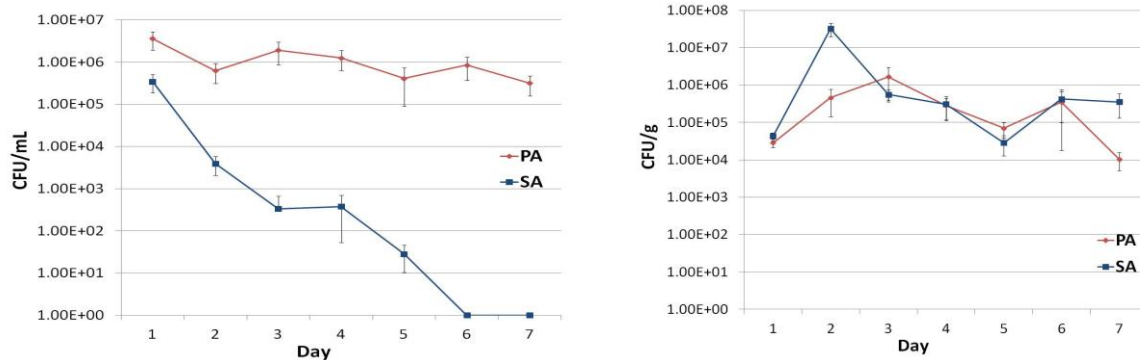
Two of the most prominent and notoriously resistant bacterial species found together in wounds are *Pseudomonas aeruginosa* (PA) and *Staphylococcus aureus* (SA). However, few studies have examined the interactions of these two species in the context of wound environment, primarily because PA quickly kills SA when the two species are cocultured in standard lab media. Recently, a simple method to grow polymicrobial biofilms *in vitro* was established, which uses a chopped-meat and blood plasma-based media. This model reliably supports the growth of polymicrobial biofilms, which accurately reflect the composition of human wound infections. We used this model to examine the interspecies relationship between SA and PA in an *in vitro* 'wound-like' environment. We examined the population dynamics of the two species in this wound biofilm model and compared the antimicrobial tolerance of planktonic cells to that of biofilm-associated cells. Our results demonstrated that: 1. When PA and SA were grown in planktonic cell co-cultures, PA quickly killed SA; however, when they were grown in our wound biofilm model, PA did not kill SA. 2. SA alone was readily able to form a host-matrix-associated wound biofilm. 3. PA was not able to form its own biofilm, but was able to colonize the biofilm made by SA, when the two were grown together. 4. Imaging analysis of the two species within the biofilm demonstrated many single-species microcolonies that were often located in close proximity to each other. 5. Strikingly, while no SA planktonic cells survived gentamicin treatment, 54% ( $\pm 13.5$ ) of SA cells in wound biofilms remained viable after treatment, and 91% ( $\pm 5.8$ ) of SA cells from SA/PA dual-species biofilms remained viable after treatment. Taken together, these data indicate that within the wound environment SA and PA may have synergistic interactions that allow them to coexist within a host-matrix-associated biofilm, which results in the benefit of increased antimicrobial tolerance for both species.

In preparation of planktonic cultures, SA and/or PA were grown, shaking at 250 rpm, in Luria Bertani (LB) broth or wound-like media (WLM) at 37°C. Samples were taken at the indicated time points, serially diluted and plated on *Pseudomonas* and/or *Staphylococcus* isolation agar to determine the number of colony forming units (CFU). *In vitro* wound-like model: WLM is made up of 45% Bolton's Broth, 50% bovine plasma, and 5% horse-laked red blood cells. After inoculation with SA and grown under static conditions the media coagulates and bacteria are surrounded by a host-derived matrix (HDM). Coagulation typically occurs within 10-12 hours of SA inoculation. *Note*: uninoculated media does not coagulate, nor does WLM media inoculated with PA. Antibiotic tolerance assay: Pellets of planktonic cells or sections of HDM were suspended in 200  $\mu\text{g}/\text{mL}$  gentamicin, 32  $\mu\text{g}/\text{mL}$  ciprofloxacin, 15  $\mu\text{g}/\text{mL}$  tetracycline (in 50% ethanol) or PBS for 5 hours. Antibiotics were neutralized in 1 ml DE neutralizing broth, and then cells were vortexed, serially diluted and plated on *Staphylococcus* and/or *Pseudomonas* isolation agar to quantitate CFU. The percentage of cells viable after antibiotic treatment was determined by dividing the number of cells that survived antibiotic treatment, by the number of cells in the PBS treatment, multiplied by 100.

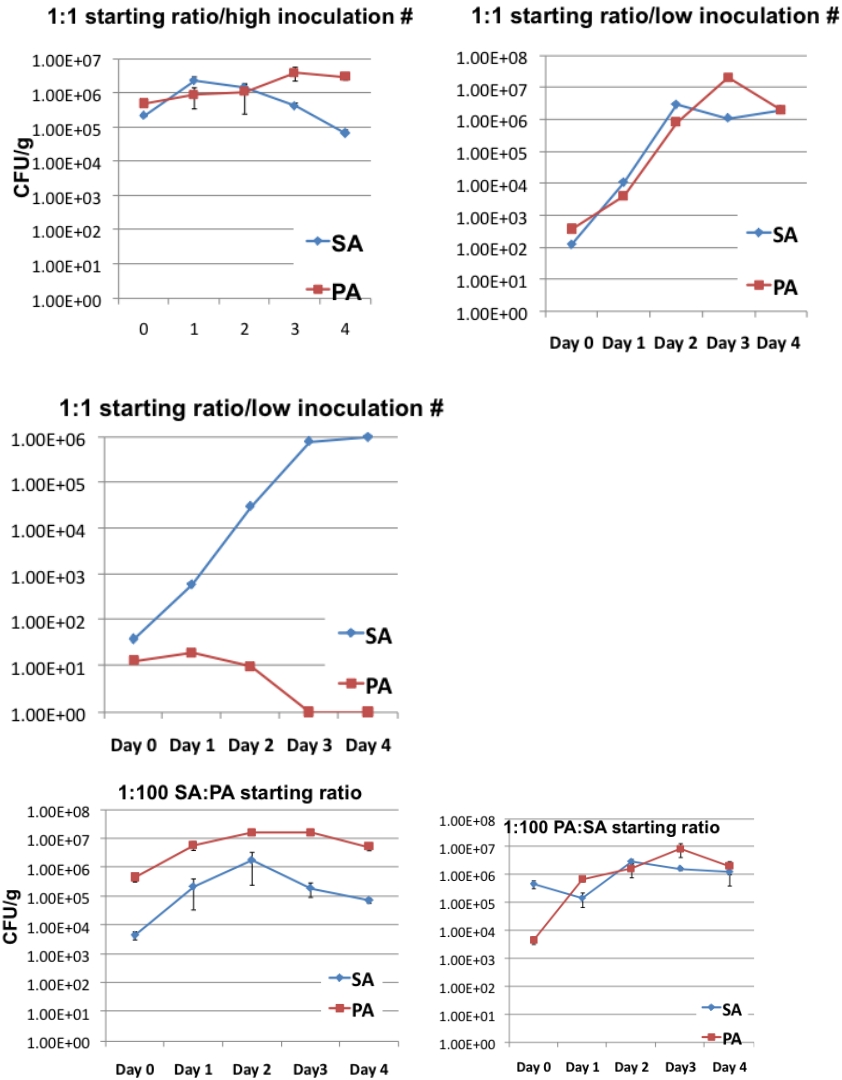
In conclusion, while PA quickly eradicates SA in planktonic co-cultures, these two species are able to exist stably together in our *in vitro* wound model, as they do in actual wounds. SA and PA appear to reach an equilibrium in the wound model, which is relatively independent of their starting inoculum. The composition of WLM makes SA significantly more susceptible to gentamicin than when it's grown in LB. However, this susceptibility is negated if the WLM is allowed to coagulate. PA and SA were more tolerant to gentamicin and tetracycline when they were together. Future studies will investigate the mechanism of this enhanced tolerance.



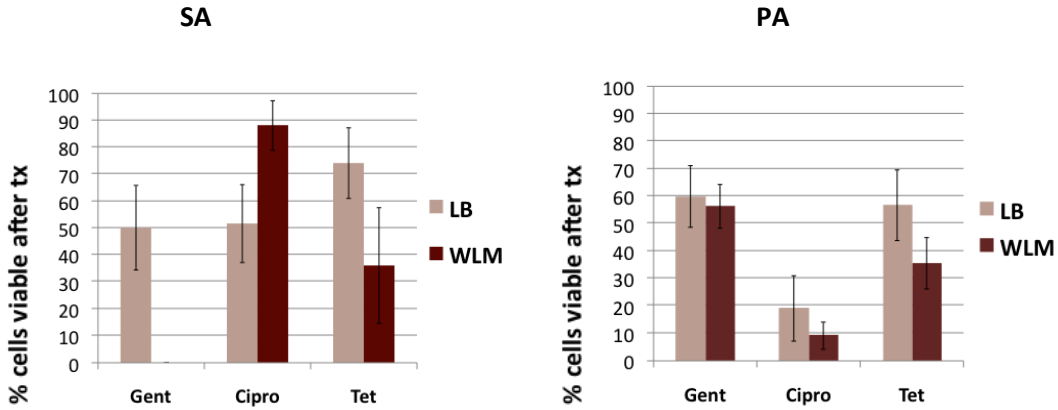
**Figure 1. PA and SA grew at similar rates when shaking at 250 rpm in LB medium at 37°C. However, when PA and SA were grown in a planktonic co-culture PA quickly took over after 16 hours of growth.**



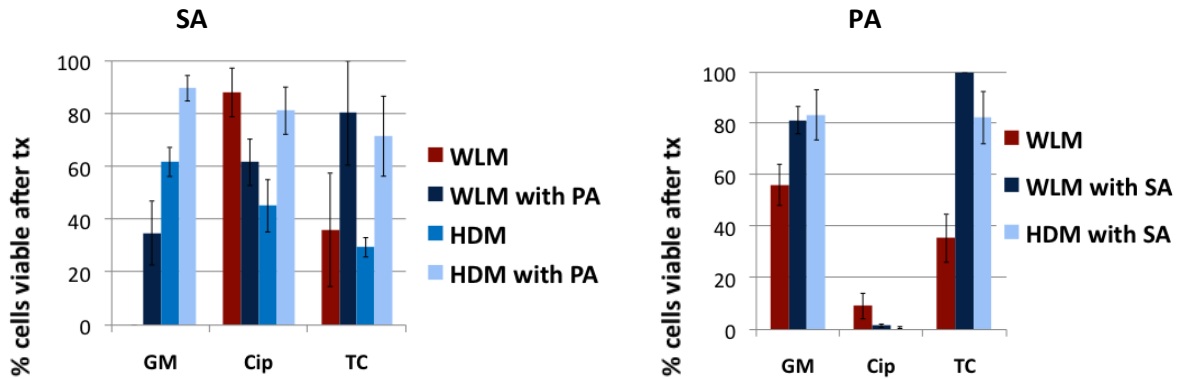
**Figure 2. PA and SA were grown in static conditions for 7 days in either LB or WLM. PA eventually killed off SA in LB but not in WLM.**



**Figure 3. PA and SA were inoculated into WLM at different starting ratios, and co-cultures were grown in static conditions for 4 days.**



**Figure 4. Planktonic mono-cultures were grown overnight in LB or WLM and tolerance to gentamicin, ciprofloxacin, and tetracycline was determined. One-way Analysis of Variance (ANOVA), followed by Tukey-Kramer Multiple Comparisons Test was used to determine the difference between groups (n=7-10/group).**



**Figure 5. Planktonic (WLM) or static (HDM) mono or co-cultures were grown overnight and tolerance to gentamicin, ciprofloxacin and tetracycline was determined.**

**Stephanie DeLeon**

I am a fourth-year student majoring in Biology. I have been in the lab since the summer of 2011. My research in microbiology consists of interactions of dual-species in-vitro biofilms. I am likely to pursue a career in science and medicine, and eventually hope conduct research while practicing medicine. Other things that I am passionate about include traveling, medical missions, and volunteering.

Development of Transgenic Crops to Enhance  
Abiotic Stress Response and Crop Yield  
Philip C. Jarrett  
Mentor: Hong Zhang

Crops are commonly grown under unfavorable environmental conditions that inhibit full expression of desirable phenotypes. The most prominent stresses that affect crop yields are imbalances of salts, extremes of temperature, and low availability of water. Due to the constant rise in human population and loss of agricultural land, the need for genetically engineered plants is more prevalent than ever before. A substantial increase (50%) in yield of crops such as maize, wheat, and rice is required to meet the expected global population by the year 2050. An anticipated 69.1% of potential crop yield is lost annually to abiotic stresses, the most common of which is drought. Approximately one-third of the planet's arable land suffers from chronic drought conditions and virtually all agricultural regions are susceptible to periodic episodes of drought. As a result, recent plant biotechnology research has focused heavily on the development of crops that may be easily maintained under drought conditions.

Molecular cloning techniques were used to develop genetic constructs that possess genes known to confer heat, drought, and/or salt tolerance in plants. Genes of interest include all of the following:

- Arabidopsis Vacuolar <sup>+</sup>H-Pyrophosphatase 1 (AVP1) – Salt and Drought Tolerance
- Arabidopsis Sodium-<sup>+</sup>H Antiporter 1 (AtNHX1) – Salt Tolerance
- Rubisco Activase (RCA) – Heat Tolerance
- *Oryza sativa* SUMO E3 Ligase (OsSIZ1) – Heat Tolerance
- Salt Overly Sensitive 1 (SOS1) – Salt Tolerance

Wild-type *Arabidopsis thaliana* (AT) plants were transformed with various combinations of the above genes then grown for several generations to isolate homozygous lines. High expression lines among these were distinguished by the use of Northern Blotting Analysis with probes specific to the transgenes. The resulting overexpression lines will undergo physiological characterization assays to discern the phenotypic consequence of the overly expressed genes on heat, drought, and salt tolerance.

Prior studies have shown that overexpression of single genes listed above may confer heat, drought, or salt tolerance in plants. The combined overexpression of multiple genes should, as a result, confer tolerance to multiple abiotic stresses. The resulting plants should, therefore, possess an improved genetic potential to yield greater returns in the field. Plants will be tested for tolerance to individual abiotic stresses as well as to combinations of multiple stresses. Prior experiments have shown that, under 200 mM salt treatment conditions, AT plants overexpressing SOS1 or AtNHX1 grow as much as 100% taller than wild-type plants under the same conditions. Additionally, creeping bentgrass plants overexpressing OsSIZ1 and AT plants overexpressing RCA have both shown significant heat tolerance compared to their wild-type counterparts. Lastly, cotton plants overexpressing AVP1 have been shown to produce ~28% more fiber than their wild-type counterparts under drought conditions. We anticipate that AT plants developed to overexpress more than one of these genes will possess significantly greater yield potential than those plants that overexpress only one of the interest genes.

**Philip Jarrett**

I am a third-year student in cell and molecular biology. I began research in the summer following my high school graduation and have since been involved in research across four separate laboratories. My primary interests lie in medicine and clinical research, but I am currently developing transgenic crops that possess resistance to abiotic stresses such as heat, drought and salt. I am pursuing medical school and hope to work someday as a physician. My other interests include personal fitness, meditation and education.

PEDF Gene Therapy Using Bone Marrow-Derived Macrophages for Advanced Prostate Cancer  
Dalia Martinez-Marin  
Mentor: Dr. Stephanie Filleur

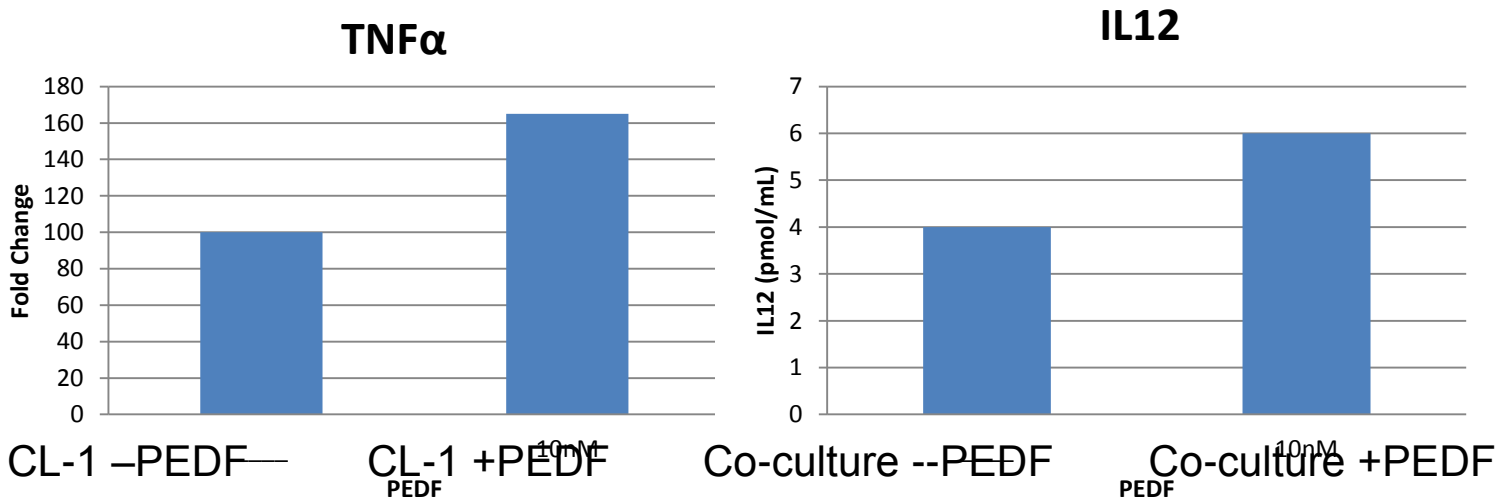
Macrophages have been described as one of the main inflammatory components in tumor growth, including in prostate cancer. Pigment Epithelium-Derived Factor (PEDF) is a secreted glycoprotein, which blocks angiogenesis, promotes neuronal survival and differentiation, and was recently suggested as an immune-modulating 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.

To test our hypothesis, the techniques 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. Tumor progression promotes a phenotype switch to M2 macrophages, which promotes 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 quantitative real-time PCR (qRT-PCR). By using ELISA we were able to determine whether particular proteins, in this case the M1 and M2 specific markers, were present and to determine how much was present in our samples. Western blotting also allowed us to look at the protein level for the different expression levels of different concentrations of PEDF in our samples. qRT-PCR allowed us to quantitatively determine how much each of these genes were expressed. In these techniques, we studied M1 type specific markers iNOS, TNF $\alpha$ , and IL12, and M2 type specific markers Arginase 1 and IL10. In addition, of looking at the expression of PEDF in macrophages, we also looked at PEDF when macrophages and cancer cells were cultured together.

Using qRT-PCR we found that there was a significant increase in TNF $\alpha$ , M1-type specific marker, in human macrophages, and a significant decrease in IL10, a M2-type specific marker, in both mouse and human macrophages. Using western blotting we found that an increase in PEDF expression up-regulates the expression of iNOS, a M1-type specific marker, and down-regulates the expression of Arginase 1, a M2-type specific marker. These data show 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*. We have also found that PEDF increases the engulfment of tumor cells by macrophages. To investigate this we co-cultured PCa (CL-1) cells (which have been engineered to express the DsRed-Express florescent protein) and macrophages (RAW 264.7)

together (ratio 1:40) and observed them using confocal imaging microscopy. In order to assure that what we had found in the vacuoles of the macrophages were indeed the cultured PCa cells we used spectral imaging microscopy and we were able to verify that the DsRed-Express protein was exhibited in the vacuoles of the macrophages are a match to that in the PCa cells.

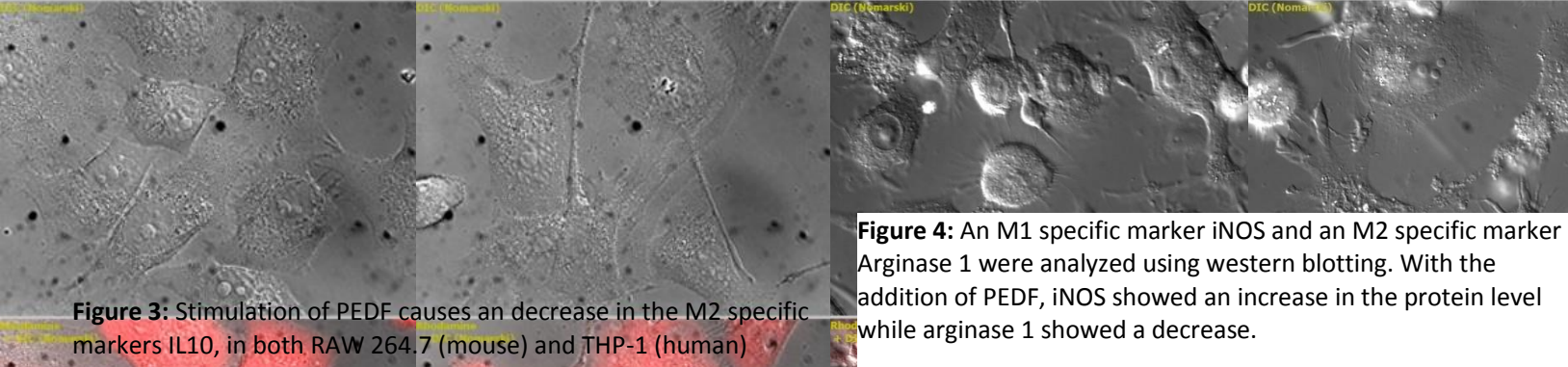
The results of our study are of importance as they suggest that macrophages may play a key role in PEDF anti-tumor effects. The next step is to test our findings using *in-vivo* models as well as further developing an effective and efficient method to use BMDMs as delivery systems for PEDF. A better understanding of PEDF may help lead to the further development of PEDF based anticancer therapy or to the improvement of alternatives to chemotherapy for prostate cancer.



**Figure 1 and 2:** Stimulation of PEDF causes an increase in the two M1 specific markers: TNFα (analyzed using RT-qPCR) and IL12 (analyzed using ELISA)



### iNOS & Arginase 1



**Figure 3:** Stimulation of PEDF causes a decrease in the M2 specific markers IL10, in both RAW 264.7 (mouse) and THP-1 (human) macrophages.

**Figure 4:** An M1 specific marker iNOS and an M2 specific marker Arginase 1 were analyzed using western blotting. With the addition of PEDF, iNOS showed an increase in the protein level while arginase 1 showed a decrease.

**Dalia Martinez-Marin**

I am a third year Cell and Molecular Bio major. I started doing research the summer of my junior year in high school and have done research in four labs here at TTU. I am very interested in medical research specifically in cancer research. After graduation I am hoping to pursue an MD/PhD and continue working in cancer. My other hobbies is playing in the TTU orchestra and playing video games.

## Modeling Positions Coupled to F508, Site of Chief CF Causing Mutation

Saba Nafees

Mentor: Dr. Sean Rice

Cystic fibrosis is a genetic disease that arises due to misfolding of the protein *CF Transmembrane Conductance Regulator*, CFTR, and is predominantly caused by deletion of a single amino acid, Phe508, along with many other mutations. Although our understanding of Cystic Fibrosis has increased dramatically over the last few decades, currently there are no known mechanisms that explain how common mutations act and interact to inhibit CFTR protein folding. It is known that multiple mutations join to have a coupling effect on the CFTR protein folding machinery, impacting both its processing and folding. Proper and deep analyses of the mutation coupling data could illuminate the way the various mutations act independently or dependently to produce disease. This information could provide insight into CFTR protein folding mechanics and enable us to pharmacologically target the protein folding process to ensure better treatment for CF patients.

Modeling the coupling effect to uncover its impact on the whole system is a profound mechanistic problem. Studying interactions between different mutations requires fitting nonlinear functions to the data. A typical way to do this would be to use multivariate polynomial regression. However, in standard polynomial regression, the coefficients of each term are a function of the degree of the polynomial that we choose to fit. This is because the different terms (first order, second order, etc.) are not independent of one another. Therefore, the coefficients do not have independent biological meaning, and it is impossible to use polynomial curve fitting and obtain a function that precisely reveals the properties of the ambiguous data set. We thus must identify functions that are independent of each other and can be analyzed separately to give meaning to the coefficients. This is the purpose of Orthogonal Polynomials and, for multivariate data, the use of dual basis. The observed distribution of mutations defines a covariant basis and a contravariant basis. Projecting different phenotypic responses, such as protein folding and processing, into these and taking the covariance would yield their interrelation.

With regards to CF, our goal is to build functions that capture the intrinsic properties of the chief mutations of CFTR, as depicted in Figure 1, so that we can assess each mutation's coupling interaction with other mutation(s). We can define a basis per mutation and their covariance will indicate their interaction. This will provide insight into the mechanism of the mutation-caused misfolding problem. In general, the orthogonal polynomial and dual spaces approach, summarized in Table 1, conserves intrinsic biological properties of any phenomenon being tested while simultaneously capturing its quantitative properties. This approach also allows one to work with nearly any kind of data (univariate, multivariate, continuous, discrete, etc.). This means that the models can be expanded to include all types of causal factors, both genetic and environmental. We aim to illuminate the interactions between not only different variables in genetic diseases, but to a wide variety of biological phenomena, including but not limited to, interactions of different cancer drugs and parent/offspring phenotype relationships.

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"Cell - Requirements for Efficient Correction of  $\Delta F508$  CFTR Revealed by Analyses of Evolved Sequences." *Cell - Requirements for Efficient Correction of  $\Delta F508$  CFTR Revealed by Analyses of Evolved Sequences.*

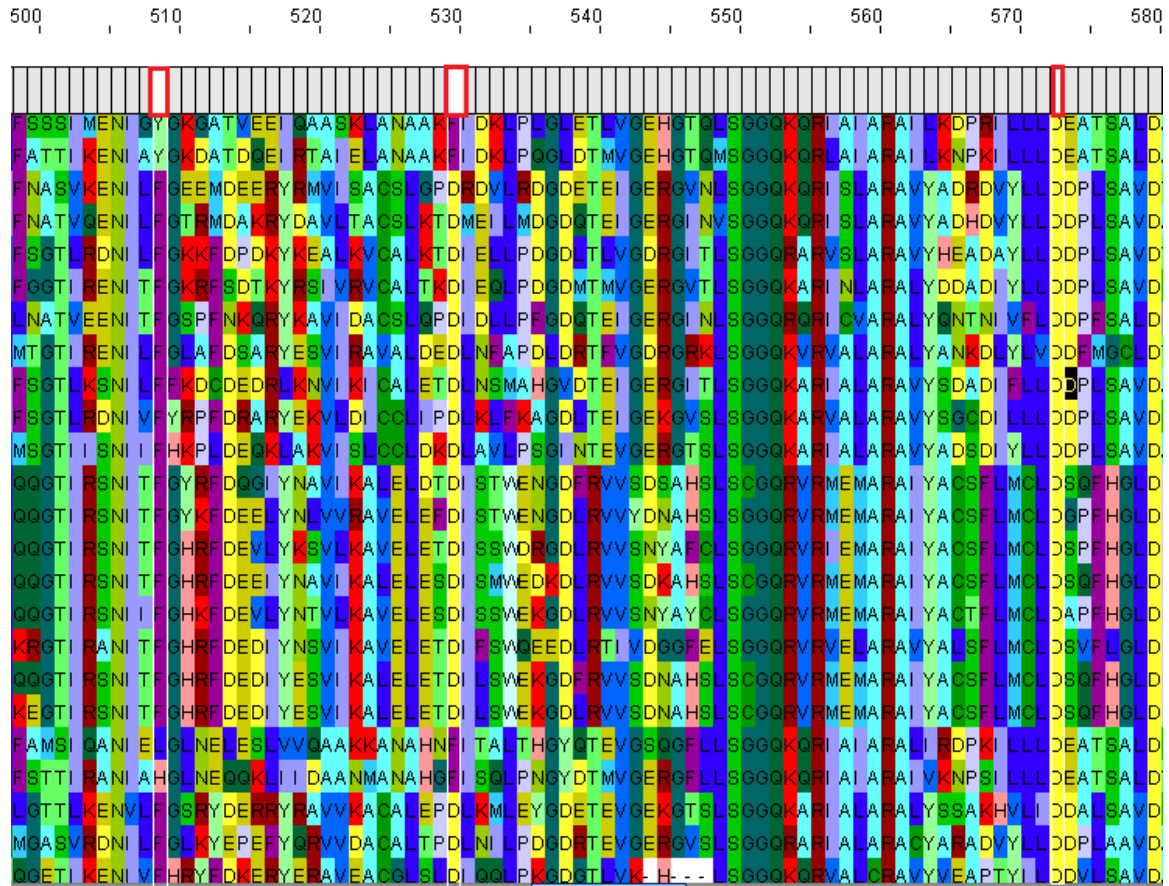


Figure 1. A subset of Multiple Sequence Alignment of the protein CFTR, with highlights of amino acids at positions 508, 529 & 573, with annotations done in pfaat program. The most conserved are Phenylalanine (denoted as F) on the 508 spot and Aspartate (denoted as D) on both the 529 and 573 spots. Mutations on these 3 locations are the specific focus of this research as these are known to increase protein folding yield.

Color scheme: all shades of blue and purple are the nonpolar hydrophobic amino acids (G,V,L,I,M,M,F,W,P). All shades of green are polar hydrophilic amino acids (S,Y,T,N,C,Q). All shades of yellow are electrically charged acidic amino acids (D, E). Pink (H), maroon (R), and red (K) are electrically charged basic amino acids.

Covariant	Contravariant
<p>First Order</p> $\mathbb{P}^1 = \phi_1$ $\mathbb{P}^2 = \phi_2$	$\mathbb{P}_{\bullet}^1 = \phi_1 - \begin{bmatrix} \phi_1 \\ \phi_2 \end{bmatrix} \phi_2$ $\mathbb{P}_{\bullet}^2 = \phi_2 - \begin{bmatrix} \phi_2 \\ \phi_1 \end{bmatrix} \phi_1$
<p>Second Order</p> $\mathbb{P}^{11} = \phi_1^2 - \begin{bmatrix} 2\phi_1 \end{bmatrix} - \mathcal{P}^{[1]} \{ \phi_1^2 \}$ $\mathbb{P}^{12} = \phi_1 \phi_2 - \begin{bmatrix} \phi_1, \phi_2 \end{bmatrix} - \mathcal{P}^{[1]} \{ \phi_1 \phi_2 \}$ $\mathbb{P}^{22} = \phi_2^2 - \begin{bmatrix} 2\phi_2 \end{bmatrix} - \mathcal{P}^{[1]} \{ \phi_2^2 \}$	$\mathbb{P}_{\bullet}^{11} = \mathbb{P}^{11} - \begin{bmatrix} \phi_1^2 \\ \mathbb{P}_{12}^{22} \end{bmatrix} \mathbb{P}_{12}^{22} - \begin{bmatrix} \phi_1^2 \\ \mathbb{P}_{12} \end{bmatrix} \mathbb{P}^{12}$ $\mathbb{P}_{\bullet}^{12} = \mathbb{P}^{12} - \begin{bmatrix} \phi_1 \phi_2 \\ \mathbb{P}_{11}^{22} \end{bmatrix} \mathbb{P}_{11}^{22} - \begin{bmatrix} \phi_1 \phi_2 \\ \mathbb{P}_{11} \end{bmatrix} \mathbb{P}^{11}$ $\mathbb{P}_{\bullet}^{22} = \mathbb{P}^{22} - \begin{bmatrix} \phi_2^2 \\ \mathbb{P}_{11}^{12} \end{bmatrix} \mathbb{P}_{11}^{12} - \begin{bmatrix} \phi_2^2 \\ \mathbb{P}_{11} \end{bmatrix} \mathbb{P}^{11}$
<p>Third Order</p> $\mathbb{P}^{111} = \phi_1^3 - \begin{bmatrix} 3\phi_1 \end{bmatrix} - \mathcal{P}^{[2]} \{ \phi_1^3 \} - \mathcal{P}^{[1]} \{ \phi_1^3 \}$ $\mathbb{P}^{112}$ $\vdots$	$\mathbb{P}_{\bullet}^{111} = \mathbb{P}^{111} - \begin{bmatrix} \phi_1^3 \\ \mathbb{P}_{122}^{222} \end{bmatrix} \mathbb{P}_{122}^{222} - \begin{bmatrix} \phi_1^3 \\ \mathbb{P}_{112}^{122} \end{bmatrix} \mathbb{P}_{112}^{122} - \begin{bmatrix} \phi_1^3 \\ \mathbb{P}_{112} \end{bmatrix} \mathbb{P}^{112}$ $\mathbb{P}_{\bullet}^{112}$ $\vdots$

Table 1: The polynomials on the left form a (non-orthogonal) basis that is dual to the basis defined by the polynomials on the right.

### Saba Nafees

I have now embarked on my third and senior year at Texas Tech! I have been studying under the direction of Dr. Sean Rice, who is a mathematician and evolutionary biologist, since May of 2012 when I was admitted into the HHMI program. My general research interest focuses on the utilization of mathematical tools to analyze patterns and changes inherent to biological phenomenon. Currently, I'm working on analysis of the genetic disease Cystic Fibrosis. I am starting my graduate studies this semester as part of the dual BS/MS program in Mathematics. I'm likely to pursue an M.D/PhD with my PhD in Mathematics and be a surgeon while continuing to research. My other activities/interests include classical piano, tennis, singing, languages, tutoring/teaching, and above all, journeying to discover and learn!

## Effects of Ocean Acidification on Red Drum Larvae Otoliths

Alexander Norton

Mentor: Dr. Sandra Diamond

An increased concentration of atmospheric carbon dioxide as a result of global climate change has significantly lowered the pH of the earth's oceans. If the current amount of CO<sub>2</sub> emissions is maintained, atmospheric carbon dioxide concentrations could reach anywhere from 730-1020 ppm by the year 2100 (Meehl, et al. 2007). This could cause the pH of the oceans, including the Gulf of Mexico, to fall 0.4-0.5 units below ambient levels, and also reduce the amount of available carbonate ion by as much as half compared to pre-industrial revolution values (Caldeira and Wickett, 2005). Marine calcifying species, such as mollusks and tropical corals, have already been observed to have very seriously compromised growth and development capabilities as a result of ocean acidification (Doney, et al. 2009), but the potential effects of a lowered ocean pH has not been thoroughly studied on fish and other non-calcifying species (Munday, 2011).

In this experiment, we will study the effects of lowered pH and increased atmospheric carbon dioxide concentrations on the morphology of red drum (*Sciaenops ocellatus*) otoliths. We are using a larval fish species because early life stages, such as red drum larvae, are likely to be the most vulnerable to global climate change and ocean acidification because their relatively small body size and their physiological functionality and homeostasis might not be fully developed (Bignami, 2013). The otolith, an inner ear bone of fish species, is of particular interest in this experiment because it is an organ that can reveal a large amount of information about a fish's life history. Composed of calcium carbonate aragonite, otoliths can indicate a fish's age, growth patterns, and the environmental conditions in which it lived. Additionally, otoliths help fish detect movement, sound, and orientation, which makes otoliths an absolutely critical organ in a fish's three-dimensional environment.

For our proposed methodology, we will utilize 40 red drum larvae and randomly separate them into two groups of 20. Each experimental group will be exposed to differing levels of atmospheric CO<sub>2</sub> within a growth chamber but held to identical levels of temperature, salinity, and dissolved oxygen. One group of red drum will be placed under 404 ppm of atmospheric CO<sub>2</sub> (ambient conditions, pH 8.15), and the other group will be placed under 1050 ppm of atmospheric CO<sub>2</sub> (predicted levels in 2100, pH 7.8). Each red drum larval group will be held under their respective conditions in the carbon dioxide chamber for 8 days, and immediately afterwards, both their right and left otoliths will be extracted. We hypothesize that the group of red drum larvae exposed to 1050 ppm of atmospheric CO<sub>2</sub> will have different otolith morphology than red drum larvae exposed to 404 ppm of atmospheric CO<sub>2</sub>. Any statistically significant morphological differences between otolith groups could cause biases in the estimates of age and growth of the fish under future climate conditions. In addition, differences between the right and left otoliths on the same fish will be examined, as fluctuating asymmetry in the morphology or skeletal arrangement of an organism can be an indicator of stress. This experiment will help pave the way to a better understanding of the effects that global climate change has on marine life.

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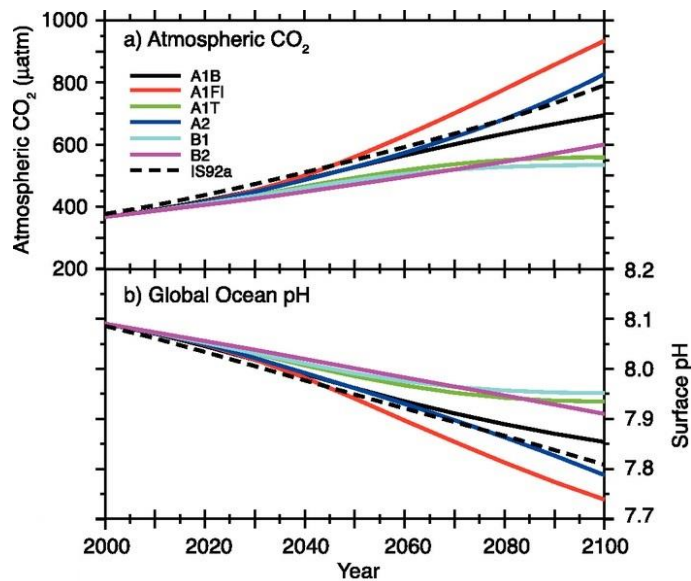


Figure 1: Predicted atmospheric CO<sub>2</sub> concentration and global ocean pH by the year 2100.

Source: [http://www.ipcc.ch/publications\\_and\\_data/ar4/wg1/en/ch10s10-4-2.html](http://www.ipcc.ch/publications_and_data/ar4/wg1/en/ch10s10-4-2.html)



Figure 2: Red drum larvae.

Source: <http://www.utmsi.utexas.edu/people/staff/gjholt/field-studies/red%20drum%20meg%202.htm>

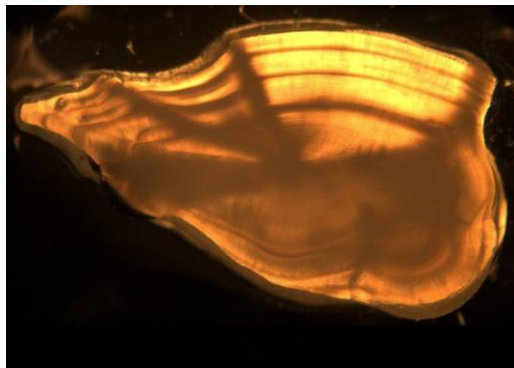


Figure 3: Red drum otolith.

Source: <http://people.uncw.edu/scharff/Students/Tayloe/tayloe%20index.htm>

**Alexander Norton**

I am a third-year biology student from Spring, Texas, and Fall 2013 is the second semester that I have worked under Dr. Sandra Diamond in marine biology research. My general scientific interests are in global climate change, chemistry, and evolution. After I graduate from Texas Tech University, I plan to pursue a career in science, medicine, and writing. My other interests include writing, reading, and sports.

## Toxicity of Sediments from Urban Playa Wetlands: *Hyalella Azteca* Survival

Maria Nunez

Mentor: Dr. Jonathan Maul

The area of the United States known as the Southern High Plains is known for its high temperatures and little rainfall. In cities found in this area, such as Lubbock, Texas, the little water available there is found in small depressional areas known as playas. Because water is scarce, these playas are important resources for the organisms that may live in them and the organisms that depend on them for survival. Since most of the playas found in the Lubbock area are surrounded by businesses and residential areas, they often receive the runoff water that comes from these neighborhoods. This water may contain contaminants, such as pesticides and insecticides. These contaminants can be harmful to the aquatic organisms living in playas. The overuse of these contaminants for lawn care and landscaping can cause a large buildup in the sediment which some aquatic invertebrates live in. Temperature could have an effect. Temperature could possibly increase the toxicity of the contaminants found in playa sediments. Temperature in playas has a tendency to fluctuate rapidly throughout the day, going anywhere from 31 °C to as low as 10 °C. This is important because some contaminants can have higher toxicity at lower temperatures. We therefore hypothesized that possible sediment toxicity can be determined by observing *Hyalella azteca* survival when exposed to sediments at various temperature regimes.

For this study sediment 2-liter samples were taken from 12 playas in the Lubbock area. These were frozen to eliminate any biological growth in them. Three temperature regimes were selected: a control of 24 °C, a low of 10 °C, and diurnal cycle treatment of 21-31 °C. The sediments were then thawed and 18 100 ml samples from each individual playa sediment were placed in 1-L jars, filled with 175 ml of mod hard water. Ten *Hyalella azteca* were added to each jar. The jars were then split within the temperature treatments along with another set of jars containing the control sediment. The exposure lasted ten days and then *Hyalella* survival was assessed and recorded.

When analyzing the overall survival for each playa individually, when compared to the control, sites 2-4 had higher mortality rates and site 1 had a higher survival rate than the control. Comparing the survival for each individual temperature treatment we found that, as expected, mortality was the highest at 10 °C out of the three, followed by the 21-24 °C treatment, with the control (24 °C) having the highest survival. For each playa the survival between the three temperature treatments was compared. The results showed that survival for site 1 was the same in all temperatures, for sites two and three mortality was higher at the 10 °C treatment, while site 4 had high mortality at both the 10 °C and the 21-31 °C. The diurnal temperature had the lowest survival. These results strongly indicate that contaminants may be present in some of these test sites, especially at lower temperatures. Sediment samples will be chemically analyzed at a later date to determine exactly what chemicals are found in each individual playa.

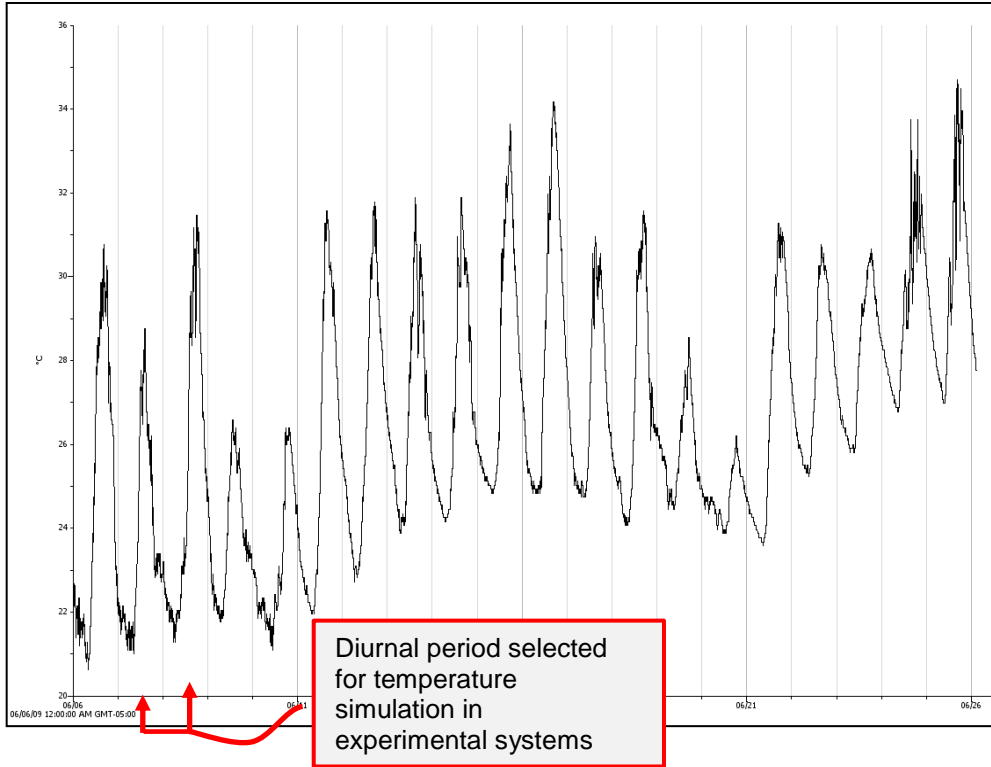


Figure 1. Diurnal water temperature fluctuation in an urban playa within the city of Lubbock, TX from 06/06/2009 to 06/26/2009. The diurnal cycle selected for diurnal temperature modeling in laboratory-based experimental systems were the lows between June 8th and 9th, 2009.

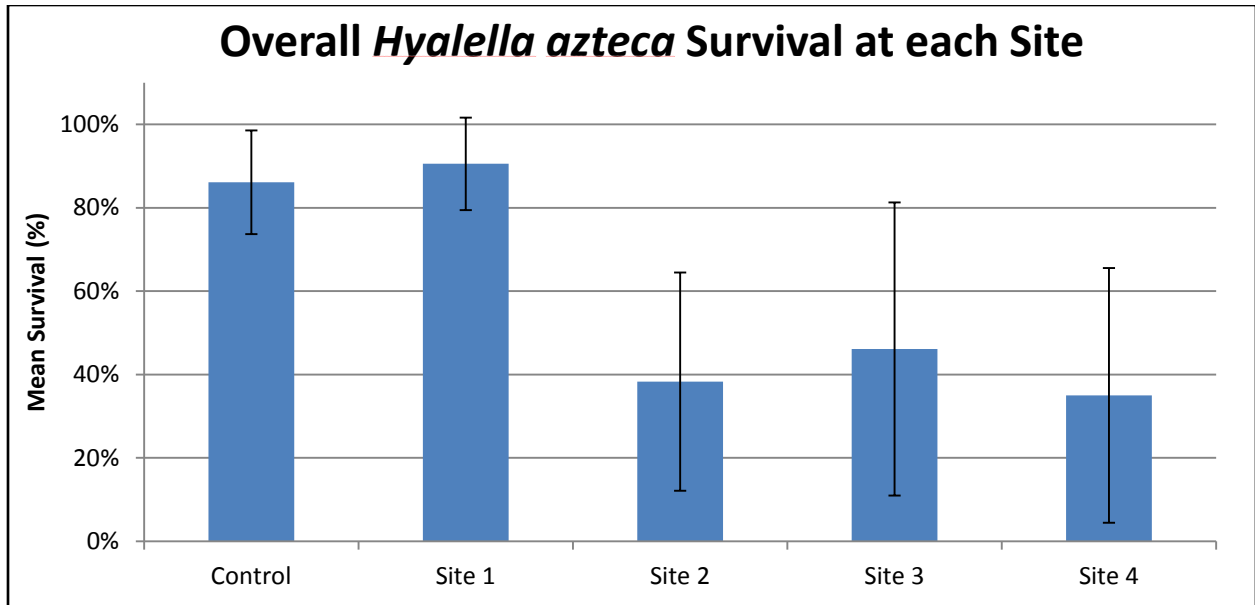


Figure 2

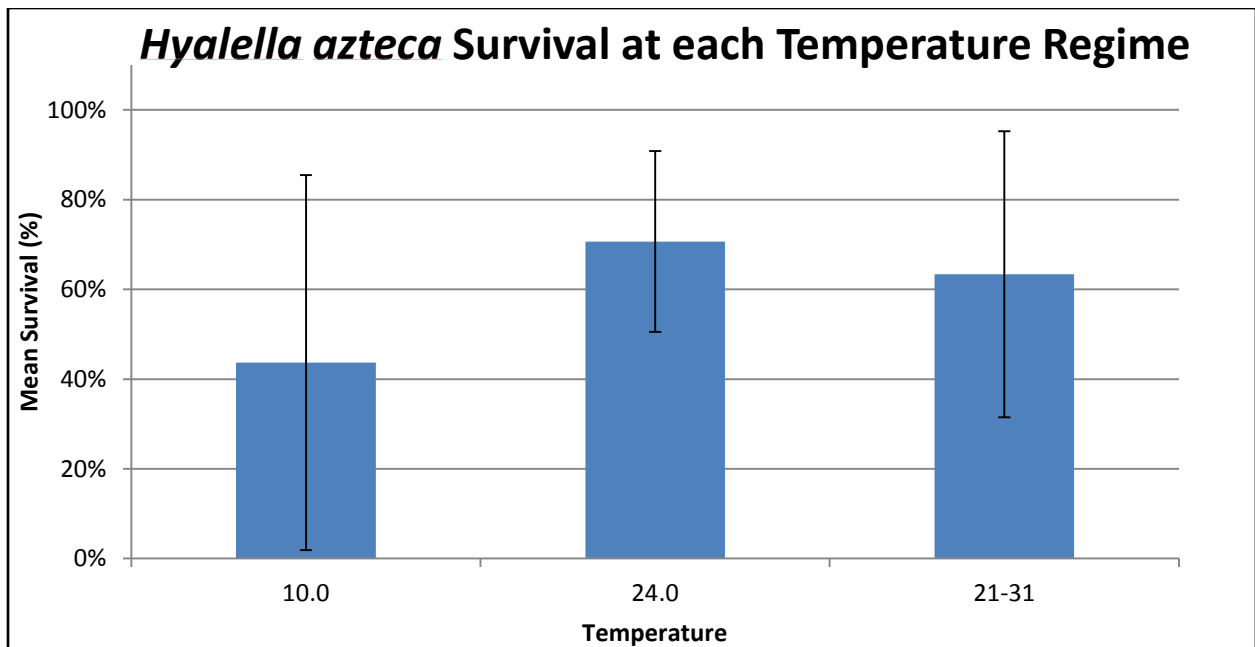


Figure 3

**Maria Nunez**

I am a fourth year student in Zoology minoring in Spanish. Prior to Texas Tech I had been in Dr. Jonathan Maul's aquatic toxicology lab for two summers. My interests are in studying how changes in the lower levels of an ecosystem affect it as a whole and how many of the changes that can occur are caused by humans. I hope to have a job where I can teach and do research, though one of my major hopes is to be able to participate in outreach and help those from minorities to pursue careers in science.

Prevalence of *Escherichia coli* non-O157:H7 STECs in Beef in Mexico  
Graysen Ortega  
Mentor: Dr. Mindy Brashears

Six strains of pathogenic *Escherichia coli* non-O157:H7 STECs, commonly known as “the big six” have recently been labeled as adulterants by the USDA (FSIS 2011). Relatively little is known about these pathogens compared to *E. coli* O157:H7. Research is currently being conducted at Texas Tech University to determine the prevalence of *E. coli* non-O157:H7 STECs in beef in the United States, as well as long term studies on the prevalence of *Salmonella* in Mexico. *E. coli* non-O157:H7 STECs are dangerous pathogens that pose a substantial risk to public health. A baseline prevalence must be established in order to effectively validate interventions and modifications to the beef production system in the future. The purpose of this study is to determine the prevalence of *E. coli* non-O157:H7 STECs in beef in Mexico.

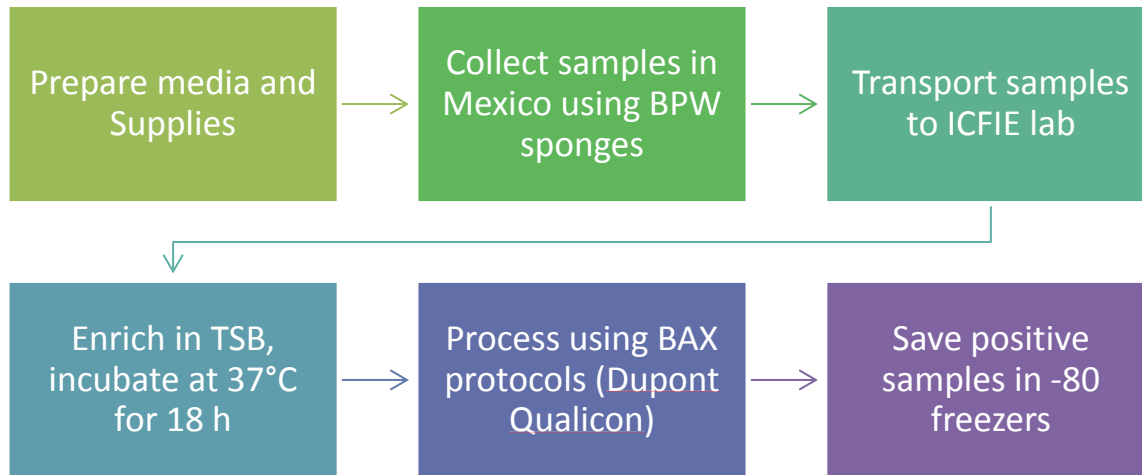
Samples were collected at beef processing plants in three major cities in Mexico. In addition to market samples collected in one city, hide, pre-evisceration, and post-evisceration samples were collected using sponges hydrated with buffered peptone water. All samples were then enriched in Tryptic Soy Broth (TSB), incubated at 37°C for 18 h, and processed in accordance with standard BAX protocols (Dupont Qualicon).

The hide, pre-evisceration, and post-evisceration prevalence varied by city with rates in city one being 96.9% (n=65), 76.9% (n=65), and 80.8% (n=65), respectively; city two 100% (n=25), 50% (n=25), and 0% (n=25); and city three 100% (n=20), 100% (n=20), and 75% (n=20). The prevalence in market samples from city three was 5.7% (n=105). Serogroups O121, O126, and O103 were the most common, with prevalence rates as high as 96.7% in city two’s hide samples, while O111 was the least common serogroup with prevalence rates of 0%-30%.

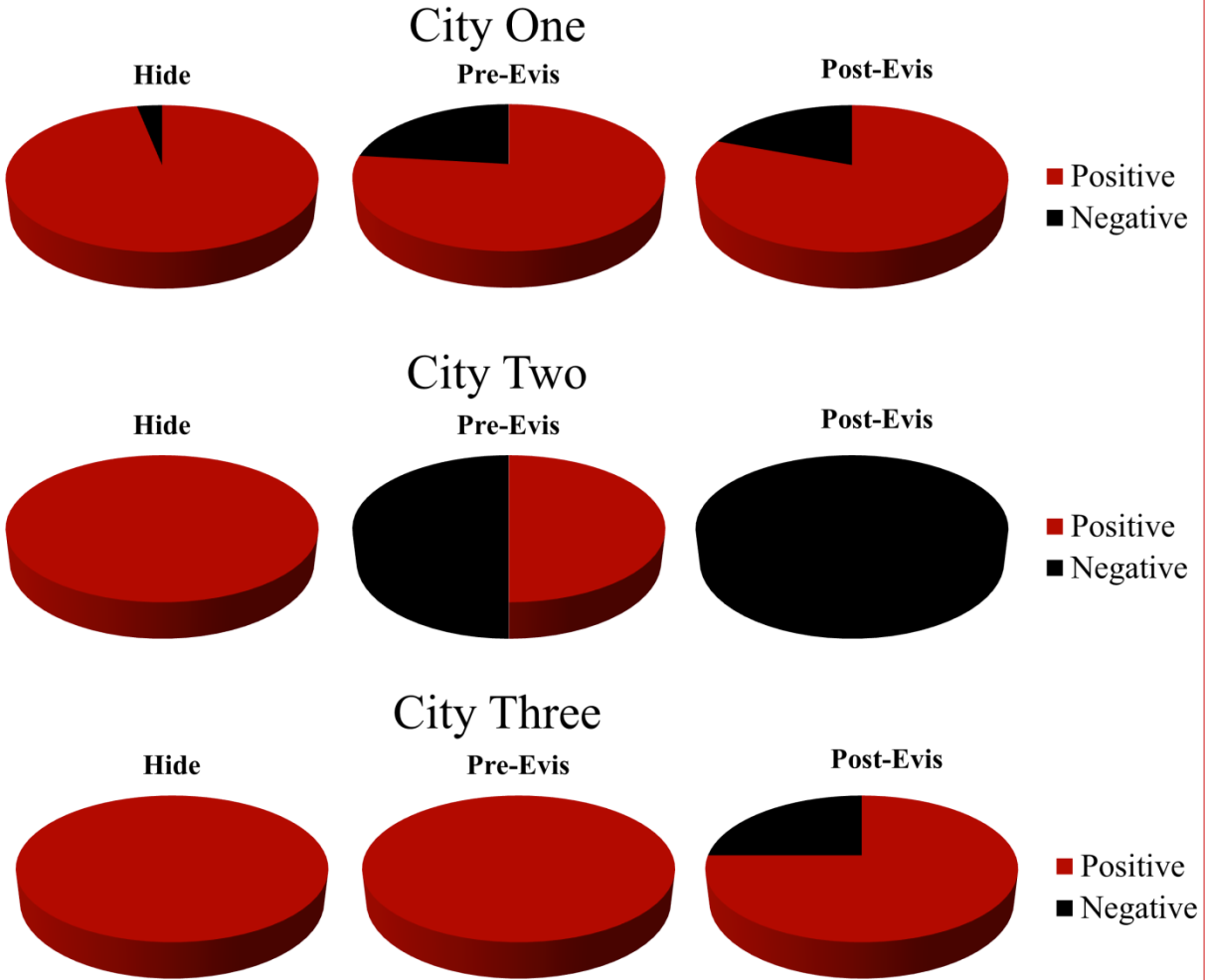
Contamination of food animals and food products with *E. coli* non-O157:H7 STECs can pose an important public health risk, especially to populations most susceptible to *E. coli* infections. While *E. coli* O157 has been reported at low prevalence in Mexico, this study effectively establishes that other STECs need to be addressed and controlled in this country.

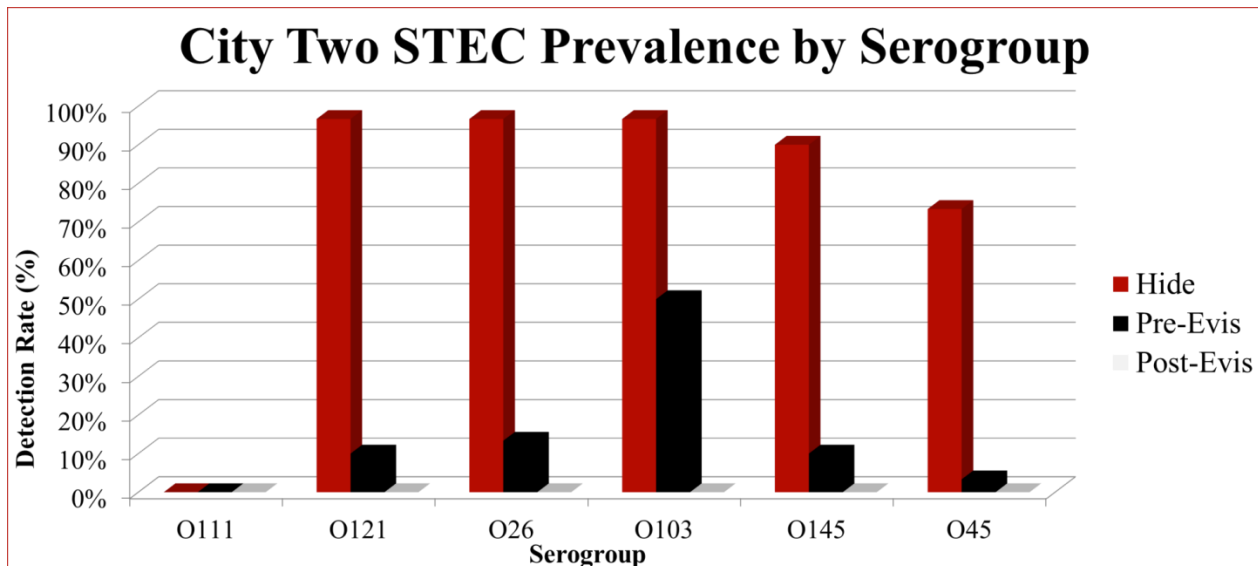
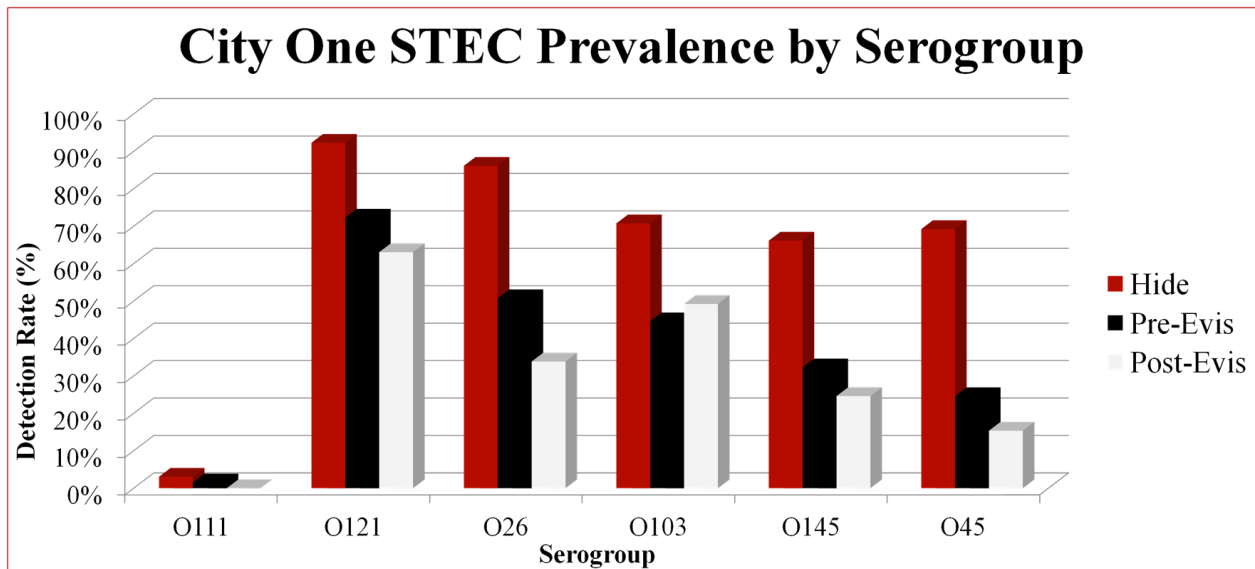
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# STEC Prevalence in Processing Plants by City





**Graysen Ortega**

I am a senior food science major from Lubbock, Texas. I have been involved in food safety research with Dr. Mindy Brashears since high school. My general interests are in studying food-borne pathogens to find ways to create a safer food supply. My focus has been on increasing safety in Mexico and I have been able to take several trips to Mexico to collect data. I plan on attending law school after I finish my degree in food science. I am a passionate supporter for agriculture and the integration of science into feeding a growing world population. My other interests include: traveling, camping, and reading.

## Prevalence of *Campylobacter* in Retail Ground Beef as Influenced by Packaging Type and Fat Content

Katelyn Ortega

Mentor: Dr. Mindy Brashears

*Campylobacter* is a gram negative, non-spore-forming bacilli shaped bacteria that requires a microaerophilic environment (5% oxygen, 10% carbon dioxide, and 75% nitrogen). *Campylobacter* spp. causes more than 2.4 million cases of human illness annually in the United States. It is an extremely fastidious organism, which makes it difficult to culture in a lab setting. It is a mystery as to how this pathogen is the most common cause of gastroenteritis in the United States. It is also a wonder as to why *Campylobacter jejuni* can cause a sister syndrome known as Guillain-Barre syndrome, which is an autoimmune disease that causes paralysis. Victims wake up paralyzed and would have never known that it was caused by consuming a food borne pathogen. Little is known about the prevalence of contamination in ground beef. The purpose of this study is to determine the prevalence of *Campylobacter* in retail ground beef as influenced by packaging type and fat content.

Ground beef samples were collected from 7 stores across Lubbock varying in fat content with multiple package types. Samples were enriched in Bolton broth, incubated for 60 hours at 42°C within candle jars with Campy gas packs, then processed in accordance with standard BAX protocols. The positive samples were plated onto MCCDA and R&F Campy agar, incubated for 60 hours at 42°C within candle jars with Campy gas packs. After incubation the growth was confirmed growth by Schimdex Campy agglutination, then streaked onto MCCDA and R&F Campy agar, incubated for 60 hours at 42°C within candle jars with Campy gas packs. Finally, isolates were collected and frozen for serotyping.

Prevalence varied based on fat content, package type, and store. The overall prevalence for *Campylobacter* within the sample set (n=189) was 30%. In regards to packaging type, 29% of the overwrap (n=144), 35% of the chub (n=40), and 33% of the over-the-counter (n=3) samples tested positive for *Campylobacter*. The prevalence of *Campylobacter* across fat content showed 35% of 73:37 (n=20), 39% of 80:20 (n=56), 18.5% of 85:15 (n=27), 21% of 90:10 (n=19), 23% of 93:7 (n=43), and 58% of 96:4 (n=12) were positive. The 7 stores where the samples were collected showed Store A with 20% (n=35), Store B with 37% (n=27), Store C with 48% (n=48), Store D with 16.7% (n=18), Store E with 40.7% (n=27), Store F with 0% (n=8), and Store G with 13% (n=26) testing positive for *Campylobacter*. The most common package type proved to be overwrap packaging, which had the highest amount of positives. However, the chub packaging had a higher percent positive. The fat content sample sizes varied greatly, which showed 80:20 and 93:7 with the most samples collected. The 96:4 and 80:20 ratios showed the highest percent positives. Store C, by far, had the highest prevalence of *Campylobacter*.

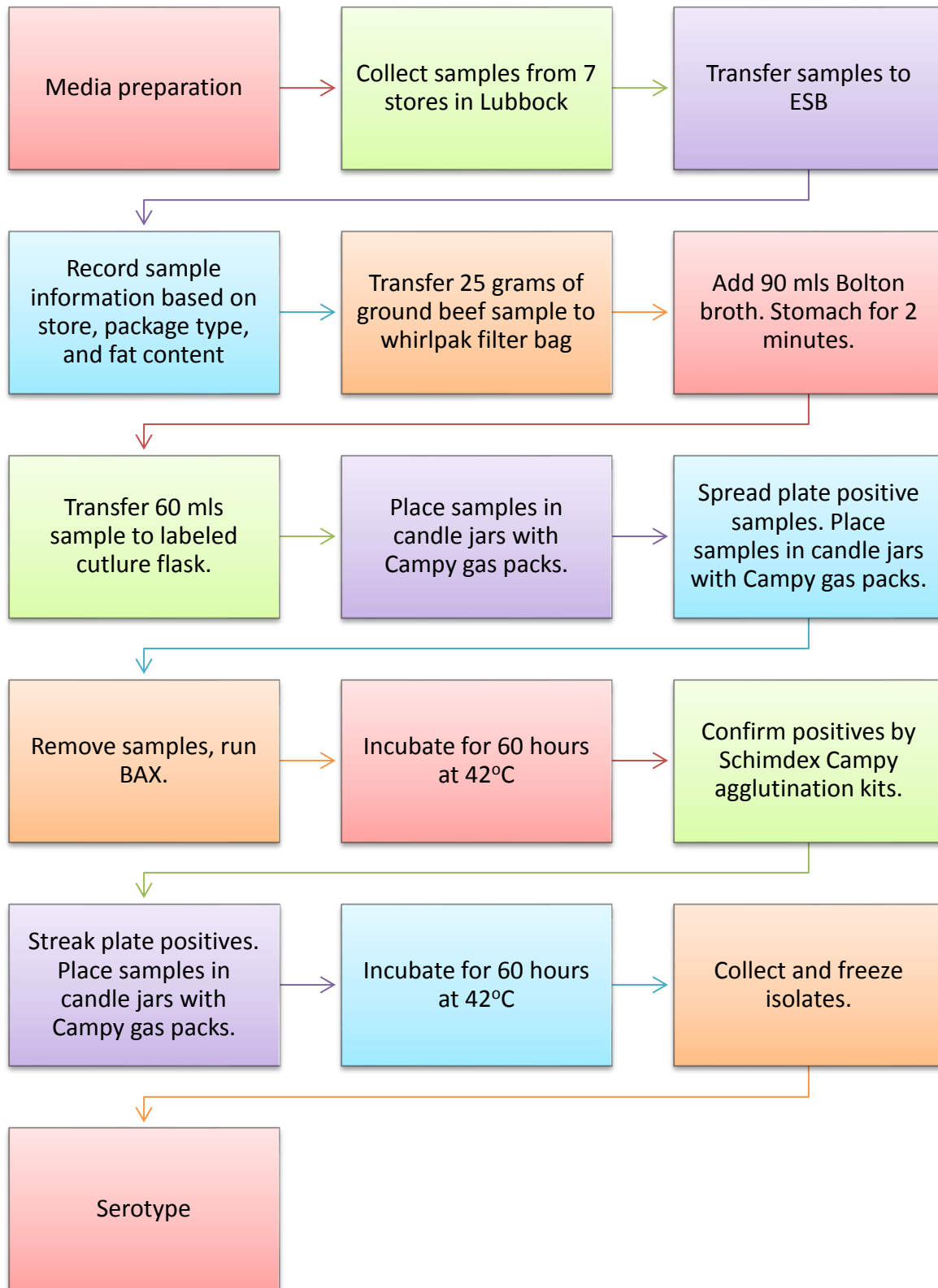
*Campylobacter* has proven itself as a problem in the food industry causing more illness annually than *Salmonella* and *Shigella* combined. Poultry has always been the prime suspect for *Campylobacter* contamination, but little is known about *Campylobacter* prevalence in ground beef. Previous studies at Texas Tech University have shown 7% prevalence of *Campylobacter* across the United States. This study showed an overall prevalence of 30%, which is significantly higher than previous findings. *Campylobacter* contamination could be seasonal or regional; whatever the reason, *Campylobacter* has

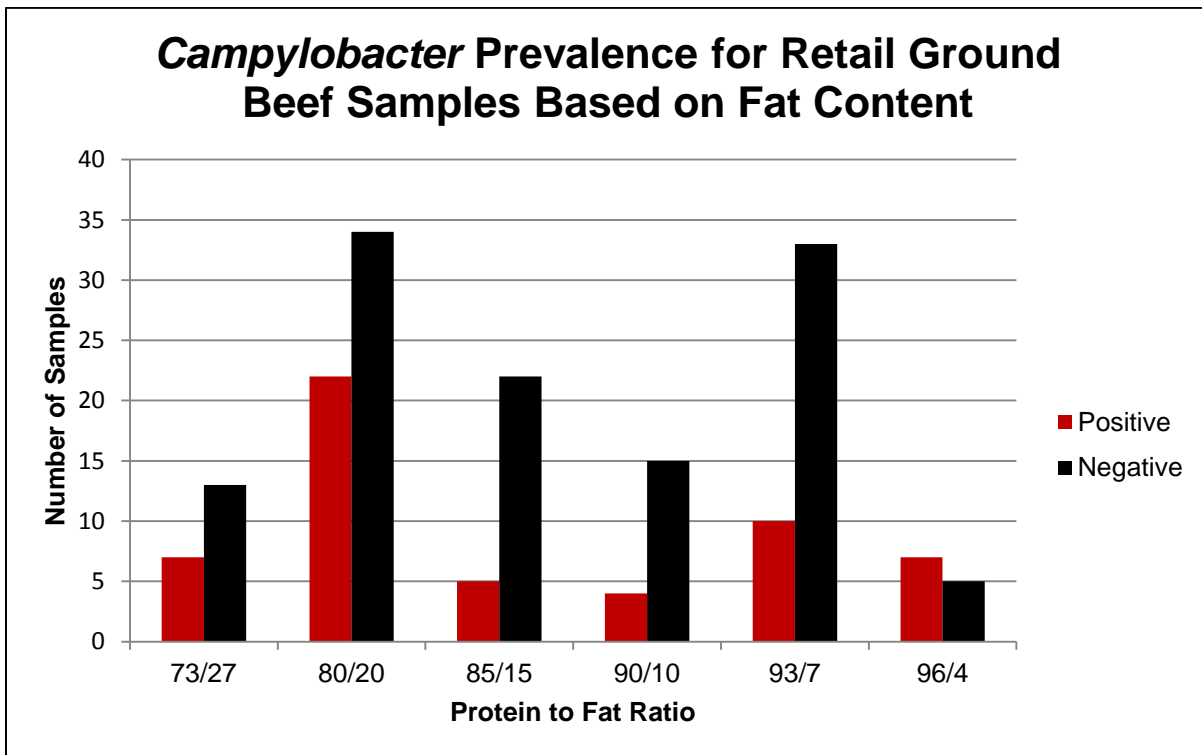
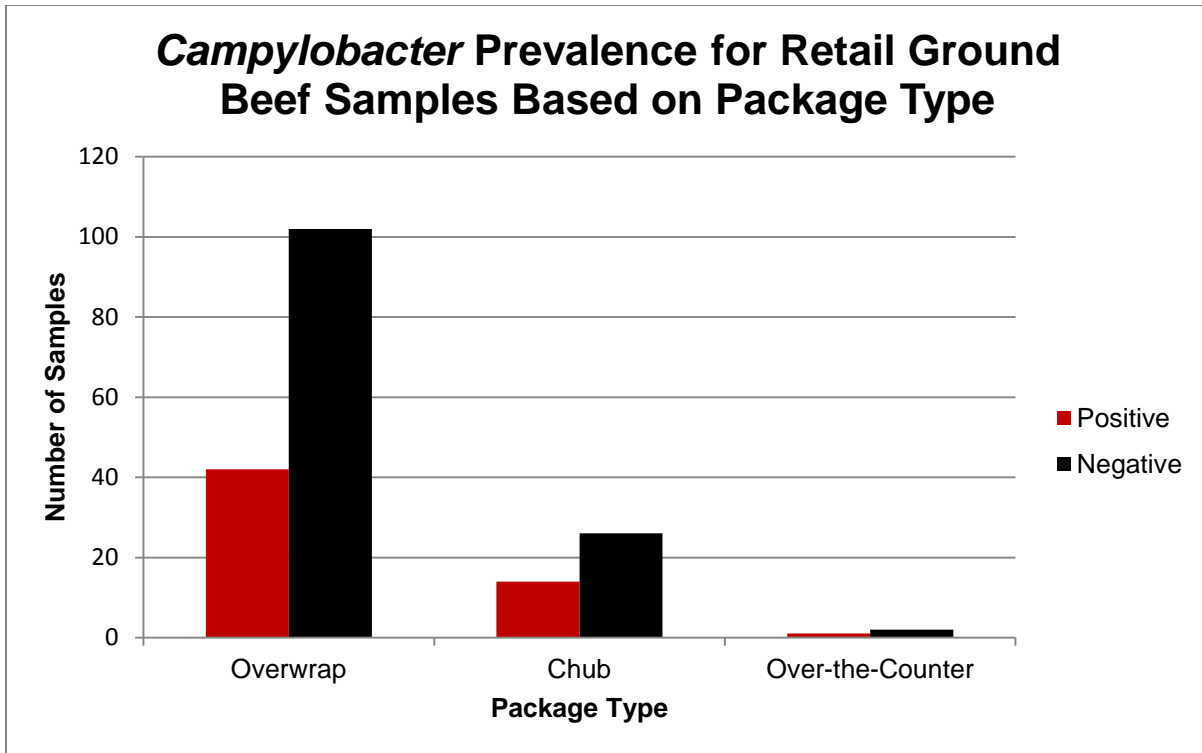
proven to be a prevalent and dangerous pathogen affecting millions of people. It is always important to remember to cook ground beef thoroughly until it reaches the proper internal temperature of 160°F. This study will continue by serotyping the isolates collected and frozen to find which strains are the most common in the region tested.

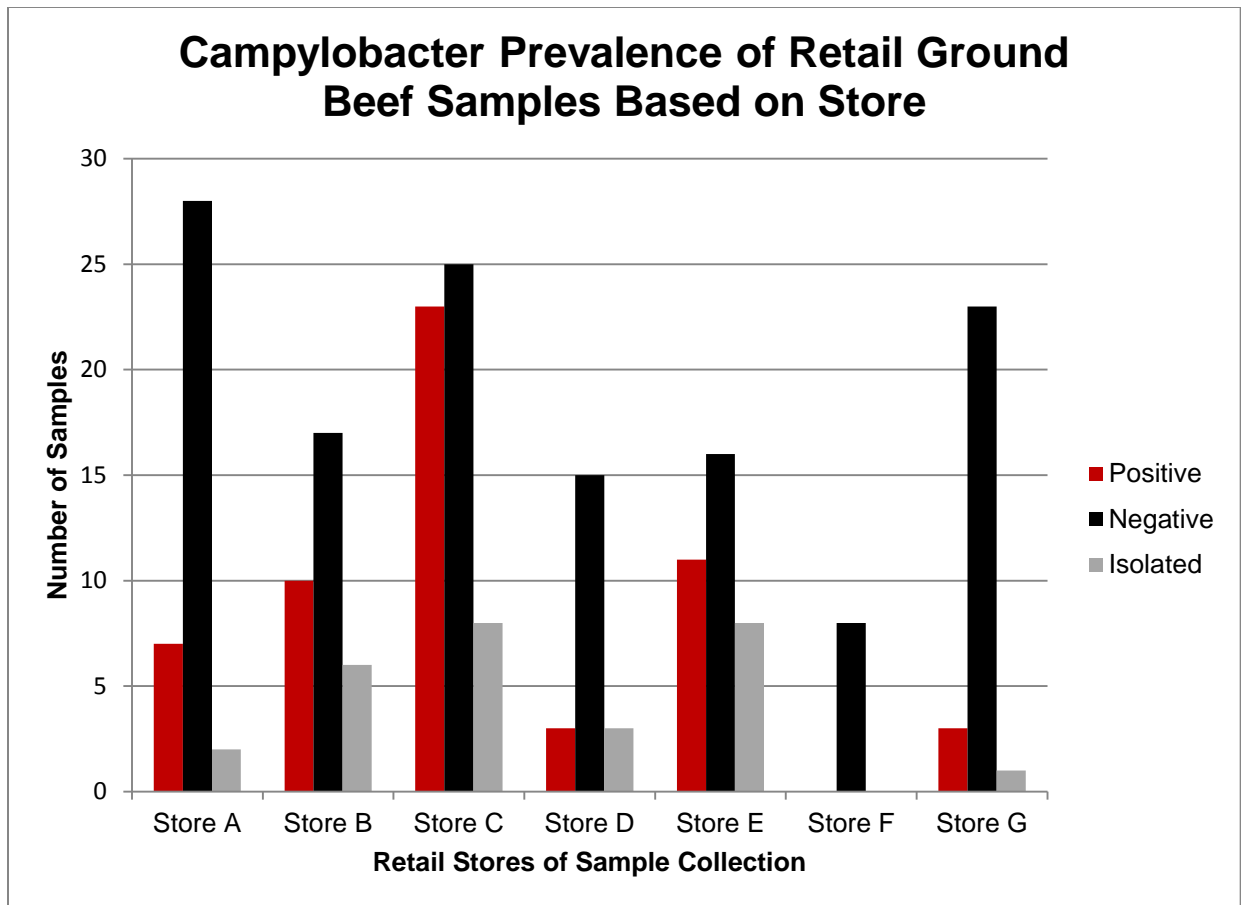
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**Katelyn Ortega**

I am a senior from Lubbock, Texas majoring in food science. I have participated in food safety research with Dr. Mindy Brashears since my junior year of high school. I have become very passionate about food safety, and I am interested in learning about food-borne pathogens in order to discover ways to make the food supply safer. My research has involved the pathogen Campylobacter, and I have been fortunate enough to participate in other studies regarding several prevalent food-borne pathogens across multiple nations. I plan to continue research under Dr. Brashears by attending graduate school at Texas Tech University. I want to be a part of making the world a safer place to consume food. My other interests include: dancing, singing, and traveling.

## Methods for Determining the Source of Chemical Threat Agents

Stephanie Pleasant

Mentor: Dr. Carlos G. Fraga

Pacific Northwest National Laboratory (PNNL) is on the forefront of national security in preventing terrorist attacks across the world that involve chemical threat agents (CTAs). CTAs are used in chemical attacks that have deadly side effects even in small quantities. Recent events such as the potential use of CTAs in Syria have added to heightened security and call for the prevention of future attacks by developing a capability for tracing CTAs to their source. Like a fingerprint, each CTA has its own unique profile that allows it to be traced back to its specific starting ingredients or place of origin. Three different subtasks were executed in this project to understand the different components of matching CTAs to their source: (1) derivatizing degradation products from the nerve agent sarin—methylphosphonic and isopropyl-methylphosphonic acid, (2) synthesizing ammonium nitrate (AN) for matching it back to its original reagent stock from four different countries of origin using High-Performance Ionization Chromatography, and (3) discovering chemical attribution signatures through the analysis of chemical data collected on a CTA simulant using LECO ChromaTOF software.

The first subtask related to the analysis of degradation products of sarin (GB), methylphosphonic acid (MPA) and isopropyl-methylphosphonic acid (IMPA). The specific task was to confirm that the entire GB sample had been converted to MPA. Due to the low volatility of MPA and IMPA, established methods were used to generate volatile derivatives and analyze these derivatives by Gas Chromatography-Mass Spectrometry (GC-MS).

The purpose of the procedure was to be able to analyze the sample further by using another analytical technique that would allow for the matching of MPA samples back to their original stock solutions of methylphosphonic dichloride (DC). It was learned that the real-world samples were not pure MPA, and the initial hydrolysis reaction of GB in 10% NaOH did not go all the way to MPA because the half-life of IMPA to convert to MPA takes 1,900 years (at pH 7), much longer than expected. Future work will be geared toward full conversion of IMPA in the GB samples to MPA by altering the pH.

The second project involved analysis of ammonium nitrate (AN) to discover potential forensic signatures. The precursor nitric acid was used as a model compound to demonstrate how the impurity profile of CTAs can be used to match a CTA back to a specific stock of that CTA after the synthesization occurs. This project required obtaining multiple different samples of nitric acid with the specific tasks of identifying and ordering the samples and then generating a chemical process permit.

Due to the lack of time, the AN experiment could not be conducted. It is anticipated that synthesis and analysis of the AN samples will obtain reliable forensic signatures through the use of anionic impurity profiles obtained from HPIC for sample matching of  $\text{NH}_3$  samples according to their source. The project is ongoing and will be completed at a later time.

The third project was based on previous studies of the effects of real-world factors on the recovery and exploitation of forensic impurity profiles of the CTA dimethyl methylphosphonate (DMMP). The discovery of chemical attribution signatures through the analysis of chemical data collected on DMMP was obtained from LECO ChromaTOF software. The work conducted on LECO ChromaTOF was a continuation of previous work that used a new approach at the data analysis in order to discover new attribution signatures.

The goal was to find a new novel approach using principal component analysis that would allow for the separation of different stocks. Although there were no new significant attribution signatures discovered using the LECO ChromaTOF data analysis, the group will continue to work towards this goal using the same software or another chromatograph method source.

Forensic chemistry and signature matching is an important and newly developing field. Successful matching CTAs to a source will aid law enforcement officials in deterring and apprehending culprits of WMD-related crimes ultimately fighting terrorism and helping make the world a safer place.

#### **Stephanie Pleasant**

I am a senior chemistry major and will graduate in spring 2014. I have been a member of the TTU-HHMI family since August of 2011 and have had endless opportunities and support through this program such as my research experiences at: TTU, a national laboratory, and one of the top genome institutes in the country. My plans after graduation are to apply to graduate school and obtain my PhD. My ideal job would be working for the government conducting meaningful research. In my spare time I love camping, hiking, salsa dancing, and practicing jiu-jitsu.

Drought Tolerance in Adult versus Resprout Oaks in West Texas  
Chris Rodriguez, Joshua Willms, and Tailor Brown  
Mentor: Dr. Dylan Schwilk

The Davis Mountains are a unique mountain range that lies in West Texas. Surrounded by the Chihuahuan Desert, the Davis Mountains rise up forming a “Sky Island” forest which houses one of the most biologically diverse and scenic regions in Texas (Poulos & Camp 2010; Poulos et al., 2007; Whittaker & Niering 1965). There are two main factors we believe are important controls on the distributions of tree species in the Davis Mountains: stress due to drought in the summer and stress due to freezing combined with drought throughout the spring. The temperate forest of the Davis Mountains maintains a moderate climate but sees intense diurnal fluctuations and temperature diversity between high and low elevations. Generally, the assumption is made that higher elevations contain cooler climates, but in the Davis Mountains there is cold air drainage that creates a cooler climate with more frequent and severe short-duration freezing events at lower elevations. One of the goals of this project is to determine a connection between the differences in water stress adaptations of oak species and their distributions in the Davis Mountains. Understanding plant species distribution can help scientists understand the possible geographical locations certain species of plants can survive. The Intergovernmental Panel on Climate Change is predicting a warmer, more arid climate for the Trans Pecos region in the future, and it will be important to predict the changes in species distribution while this occurs (IPCC, 2007).

Drought-induced xylem cavitation (embolism) is an important mechanism of mortality in desert plants. When evaporative demand exceeds water supply, xylem cavitation can result in catastrophic hydraulic failure via the aspiration of air bubbles into the xylem water from the surrounding tissues (Zimmerman, 1983). This study will examine the adaptations each oak species has to resist embolism from drought and freezing. Drought and freezing cause embolism in two different ways in a plant, but both can lead to the death of the plant through xylem cavitation (Sperry and Sullivan 1992). Both freezing and drought are related to plant distribution but are rarely studied together (but see Willson & Jackson, 2006). It is important to see how freezing and drought affect xylem embolism in the four oaks. It is also relevant to determine whether or not there is a relation between the distribution of each species and their vulnerability to cavitation. If freezing acts as a major control on distribution in these species, then it is possible for a warming climate to result in downhill movement of species because low elevations experience more severe freezing stress, although a downhill shift is a pattern rarely predicted. Throughout the summer of 2013 the methodology for determining stem conductivities has been modified and improved. Four individuals were tested across five oak species utilizing a hydraulics system for simulating the flow of water through stems and centrifugation to simulate tension due to drought stress. Our preliminary data suggest that resprouts have slightly greater conductivity than adults of the same species, but further analysis is necessary to finalize results and interpretations.

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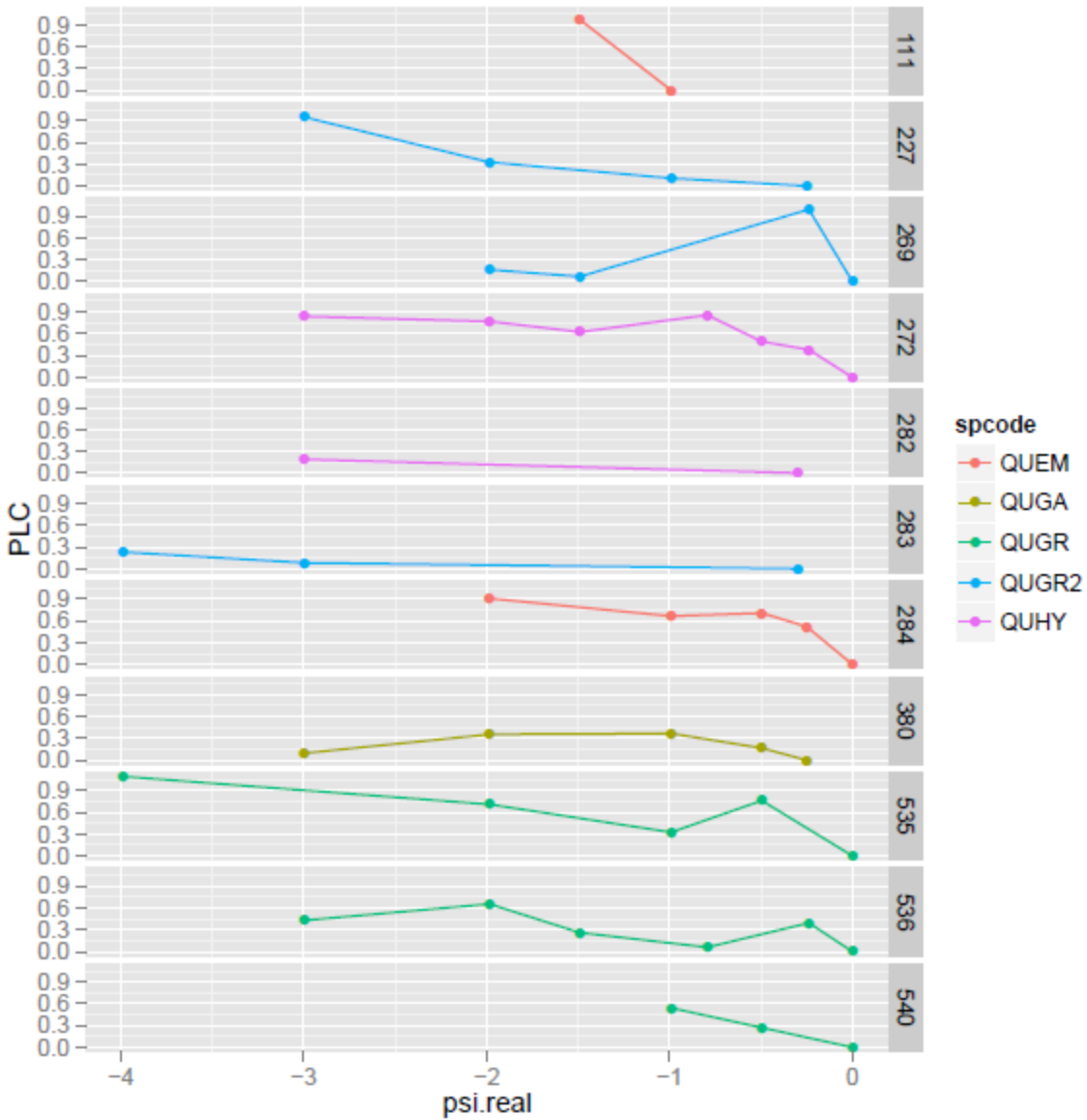


Figure 1. Vulnerability curves for five species of oaks. Percent Loss Conductivity (PLC) is graphed against tension-induced drought simulation (psi.real). As PLC increases, the percentage of stems which are closed off by embolism increases.

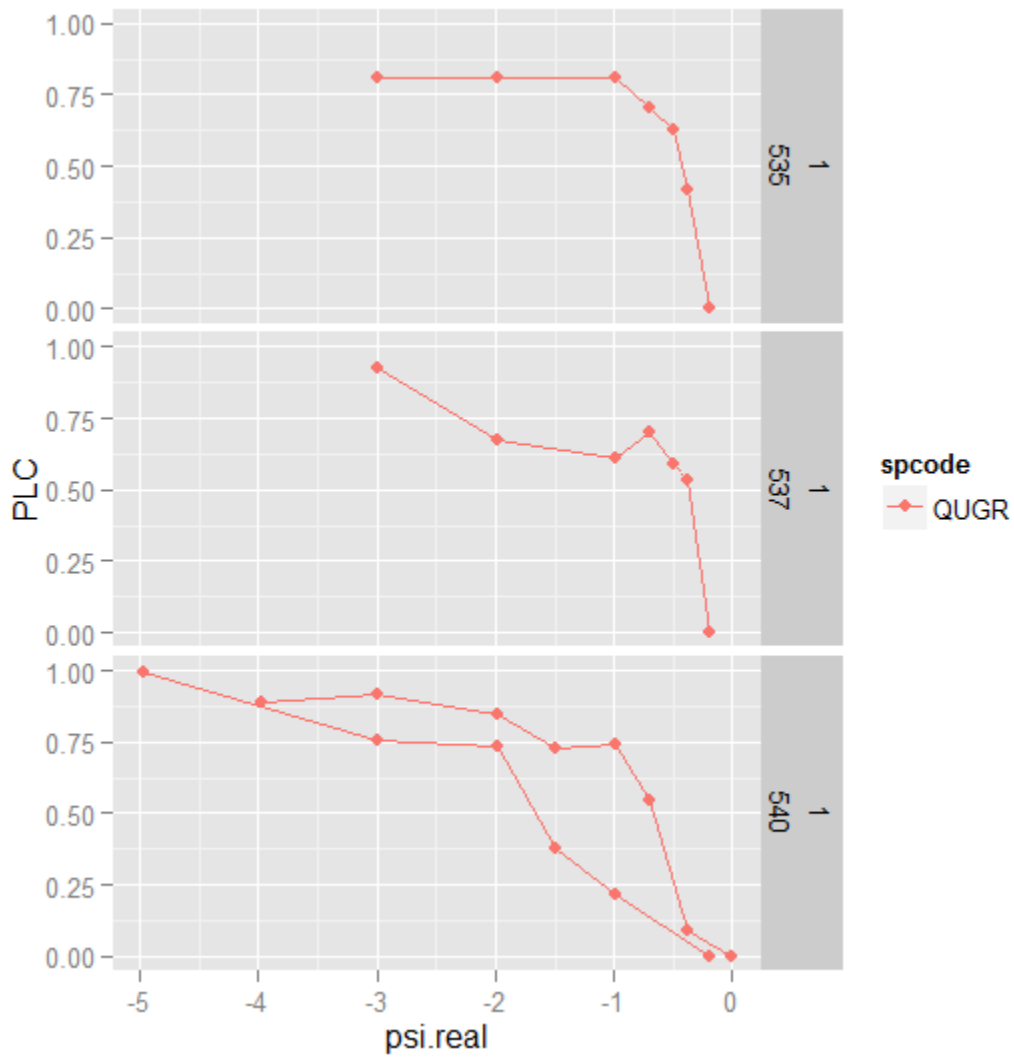


Figure 2  
 Preliminary vulnerability curves for *Quercus grisea* resprouts. Percent Loss Conductivity (PLC) is graphed against tension-induced drought simulation (psi.real). As PLC increases, the percentage of stems which are closed off by embolism increases.

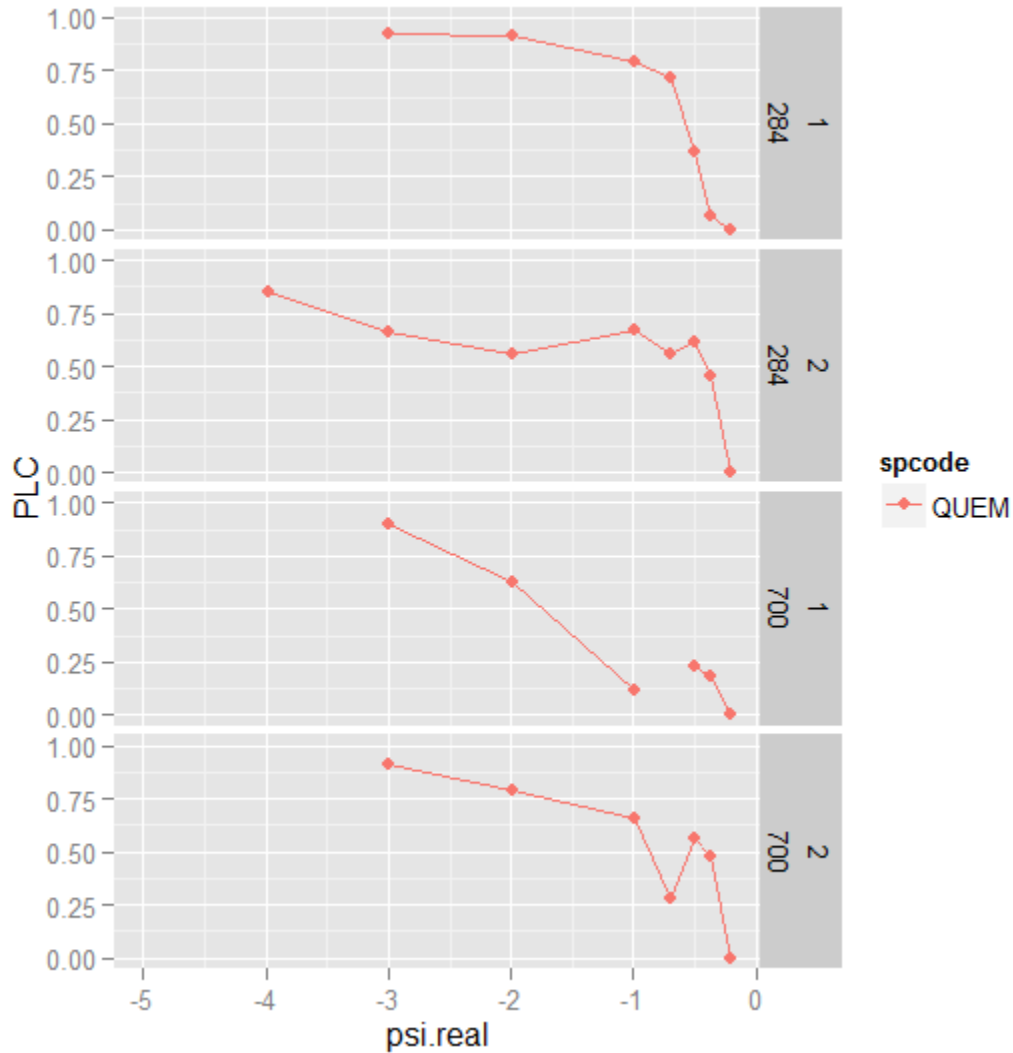


Figure 3  
 Preliminary vulnerability curves for *Quercus emoryi* resprouts. Percent Loss Conductivity (PLC) is graphed against tension-induced drought simulation (psi.real). As PLC increases, the percentage of stems which are closed off by embolism increases.

**Christopher Rodriguez** (*in memorium*)

Christopher Rodriguez was a young man and scholar whose spirit overflowed with joy, whose almost tangible friendliness touched all who surrounded him, and whose brilliance and intelligence gleamed through his research and studies. His dedication to his work was unmatched and the passion with which he pursued his goals remains an inspiration for all of us. We miss you Chris and will remember you always.

## Effects of Fruit Flavonoid on Steroidogenesis & StAR Gene Expression

Daniela Rojas

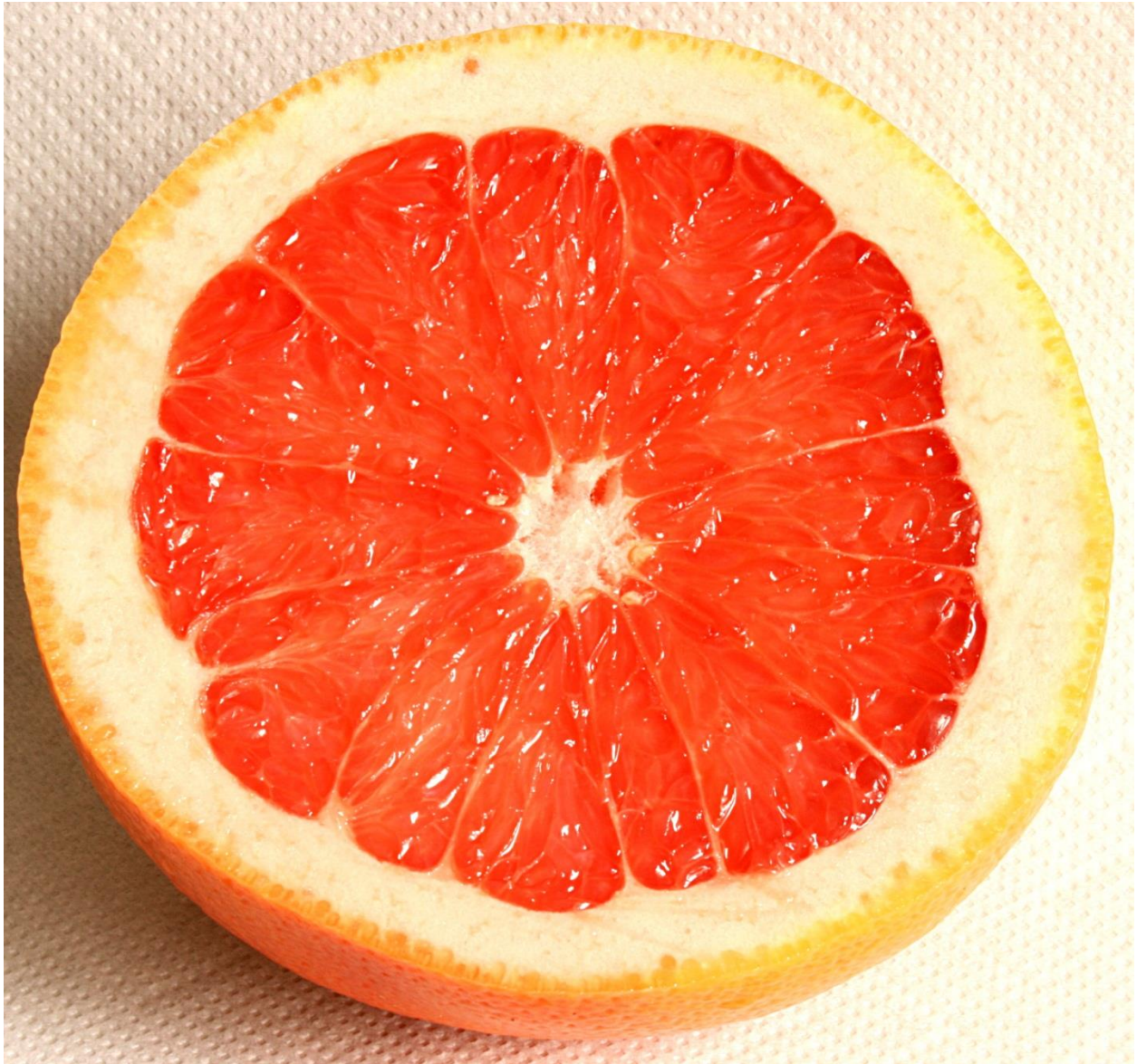
Mentor: Dr. Akhilesh Pandey

The current ongoing project serves to further expand our understanding of Steroidogenic Acute Regulatory (StAR) gene expression and sex hormone production by murine hippocampal neuronal. The hippocampal area is closely related to memory retention. Cell lines, such as HT22, contain estrogen receptors<sup>1</sup>. The goal in this project is to screen different natural flavonoids and their effect on the neuronal cell lines in terms of estradiol production and StAR protein production. We want to test these hormone levels with the enhanced performance of the StAR protein, as estradiol/testosterone production can be linked to physiological well-being, and age-related decrease in these sex hormones can have implications leading to susceptibility for neurodegenerative diseases and lower mental performance<sup>2</sup>. If a relationship is found, we want to find the molecular mechanism in which maximized levels of StAR gene expression might enhance hippocampal estradiol levels. This way we can relate intake of Natural Flavonoids with possible memory retention during aging at a neuronal level, and possible decrease in memory loss symptoms of patients with neurodegenerative diseases<sup>3</sup>.

The first step of this project is establishing the relationship between brain estradiol levels and StAR levels in a culture medium. This will be done through the use of ELISA kits to quantify the levels of these hormones in different hippocampal HT22 cell lines. A control will be screened for a baseline quantity of estradiol production. Another HT22 cell line culture will be exposed to StAR gene expression levels and screened for Estradiol levels. As soon as this relationship is established, we plan to screen the effects of different natural flavonoids on StAR gene expression via luciferase assays, StAR gene mRNA expression via reverse transcriptase semi quantitative Polymerase chain reaction (PCR), and Real time quantitative PCR and protein expression by western blot/Dot blot technique. This last stage of testing will bring about an indirect relationship between the intake of products, such as natural fruit flavonoids, with the enhancement of sex hormone levels, healthier aging, and memory retention.

The overall outcome we want to achieve with this project is to find relationships between natural products and possible improvement of brain and physical function for aging individuals. A successful project will bring about possible innovations in the treatment of age-related diseases, and also natural ways to enhance health for aging individuals. Natural Flavonoids can be found in products such as fruits and vegetables, so we want to test and succeed in this project as it will depict the importance of the intake of healthy products in terms of healthier aging and delaying of neurodegenerative disease symptoms.

As this project is still in the planning stages, there are no results or conclusions to be made. Yet, we are very hopeful that this project will bring about beneficial innovations in the realm of aging and age-related disease research. The success of this project will mean further investigation of natural substances that can delay aging, aging symptoms, and age-related diseases. It will also provide a mechanism and molecular level of understanding of how hormone levels impact aging.



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**Daniela Rojas**

I am a Microbiology Texas Tech Undergraduate student in my senior year. I wish to pursue Medicine, especially Oncology, which is a subject that my family can relate to. I am originally from Silver Spring, Maryland, but fell in love with Texas and its culture. Research has been very important to me throughout my time at Texas Tech University because it has opened up a lot of options and knowledge concerning scientific phenomena that I can always go back to in my future career. Outside of school and research, I love to dance, sing, and write.

## Role of *Clostridium perfringens* in Multi-Species Wound Biofilms

Hayley Sparks

Mentor: Kendra Rumbaugh

A common problem in bacterial wound infection is the formation of multi-species biofilms. Biofilms are aggregations of bacteria that are encased in a polysaccharide matrix. In the case of human infection, these biofilms often exist to shield bacteria from several components of the immune system and may play a role in antibiotic tolerance. Current research has mainly focused on the roles of aerobic bacteria in these wound biofilms. However, recent investigations have shown that anaerobic bacterial species actually make up the majority of the population in these biofilms. One of the most common anaerobic species found in these infections is *Clostridium perfringens*, a strict anaerobe bacterium that produces the toxin which causes gas gangrene in wounds. My current research is attempting to determine if *C. perfringens* participates in biofilm formation. Further if it does participate under what conditions does it do so? And finally, what effect does this bacterium have on other species present in the biofilm?

*In vitro* models are currently being used in this investigation. These consist of inoculating a 45% meat peptone broth, 5% laked horse blood, and 50% bovine plasma media with the desired bacterial species. This media mimics the environment found in most chronic, burn, and abscess wounds, making it an ideal model for studying these biofilms in an *in vitro* environment. As *P. aeruginosa* (strain PA01) and *S. aureus* (strain SA31) are the two most common species of aerobic bacteria found in wound biofilms, they were included in the biofilm studies, as well as *C. perfringens*. All three species were cultured at 37°C overnight from cryosamples using 10 mL of appropriate liquid media. PA01 and SA31 were shaken in beveled flasks during this time while *C. Perfringens* was incubated in an anaerobic chamber and was not shaken. After incubation, the optical density of the cultures was measured and the cultures were then diluted to approximately equate the amount of bacteria being used from each culture. These samples were then serially diluted and plated on appropriate agar to determine the colony forming units (CFU) of the starting samples. The 10<sup>-2</sup> dilutions of these bacterial samples were then used to inoculate the formerly described wound-like media in micro test tubes which were incubated at 37°C for 24 hours. The resulting biofilm samples were extracted, washed in PBS, homogenized, diluted, and plated on isolation agar to collect the CFU of each species in the biofilm. To determine antibiotic tolerance, the above process was followed but resulting biofilms were exposed to a 0.2 mg/mL solution of gentamicin or 1mg/mL solution of tetracycline for 5 hours and were then neutralized for 10 minutes using Dey-Engley broth. The resulting material was then rinsed and homogenized in PBS and the serially diluted and plated on appropriate isolation agar to collect the CFU of the samples.

Using these methods, our data has indicated that *C. perfringens* is able to grow in the biofilm environment when co-cultured with PA01 and SA31, even when cultured aerobically. However, when cultured by itself in wound-like media *C. perfringens* does not form a biofilm and will only grow in such an environment when incubated anaerobically. It was also seen that *C. perfringens* will grow aerobically in wound-like media when co-cultured with SA31 or PA01, indicating that *C. perfringens* may require the presence of aerobic bacteria in order to infect a wound. Investigations have been conducted to

determine if the presence of *C. perfringens* in biofilms increases the antibiotic tolerance of SA31 or PA01 when the biofilms are exposed to the antibiotic tetracycline or gentamicin. Current results indicate that while *C. perfringens* does not appear to increase gentamicin tolerance, it may increase tetracycline resistance, especially that of PA01. Currently our research is focused on determining the physical location of *C. perfringens* in biofilms as this may be a factor in its ability to grow in an aerobic environment, for example, there may regions within the biofilm that are particularly hypoxic. *In vivo* investigations may be pursued with this project in the near future as well as expansion to include other infectious anaerobic species. When published, the findings of this project may encourage other investigators to begin examining the roles of often overlooked anaerobic bacteria in wound infections and the effect they may have on current treatments.

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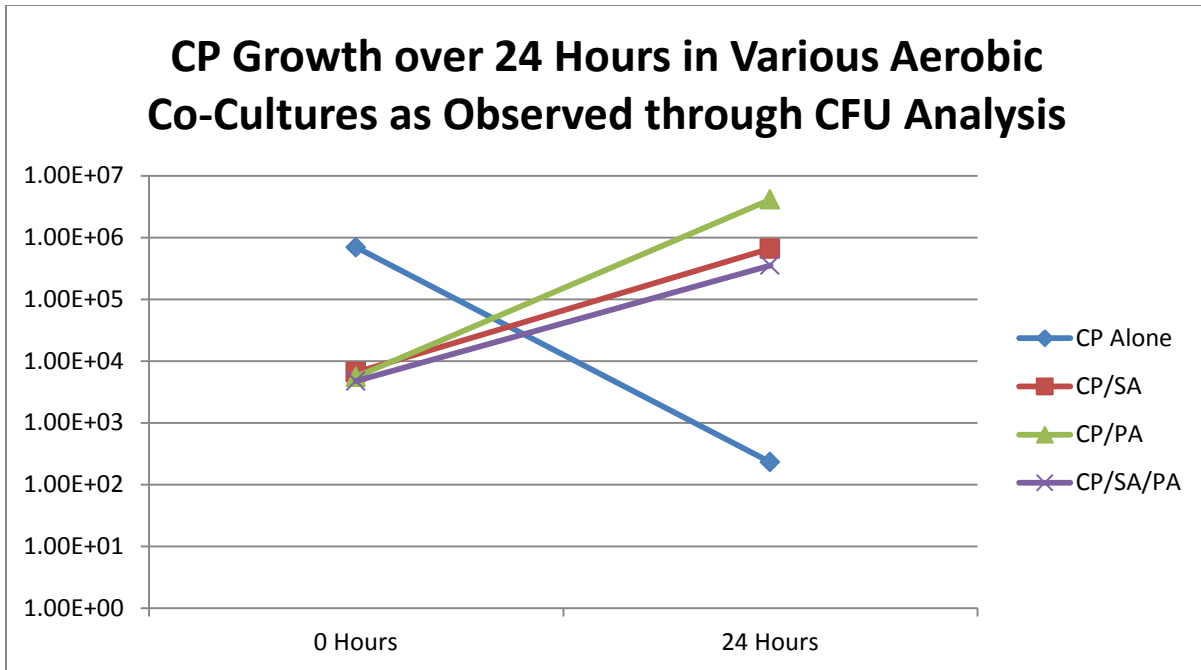


Figure 1. *C. perfringens* (CP), a strict anaerobe will grow aerobically in wound-like media when co-cultured with either SA31, PA01, or a combination of the two. However, CP does not grow aerobically in wound-like media by itself. This indicates that CP requires co-infection in order to grow in aerobic wounds. This figure shows the inoculating CFU count of CP compared to the resulting CFU count after 24 hours of incubation in wound-like media.

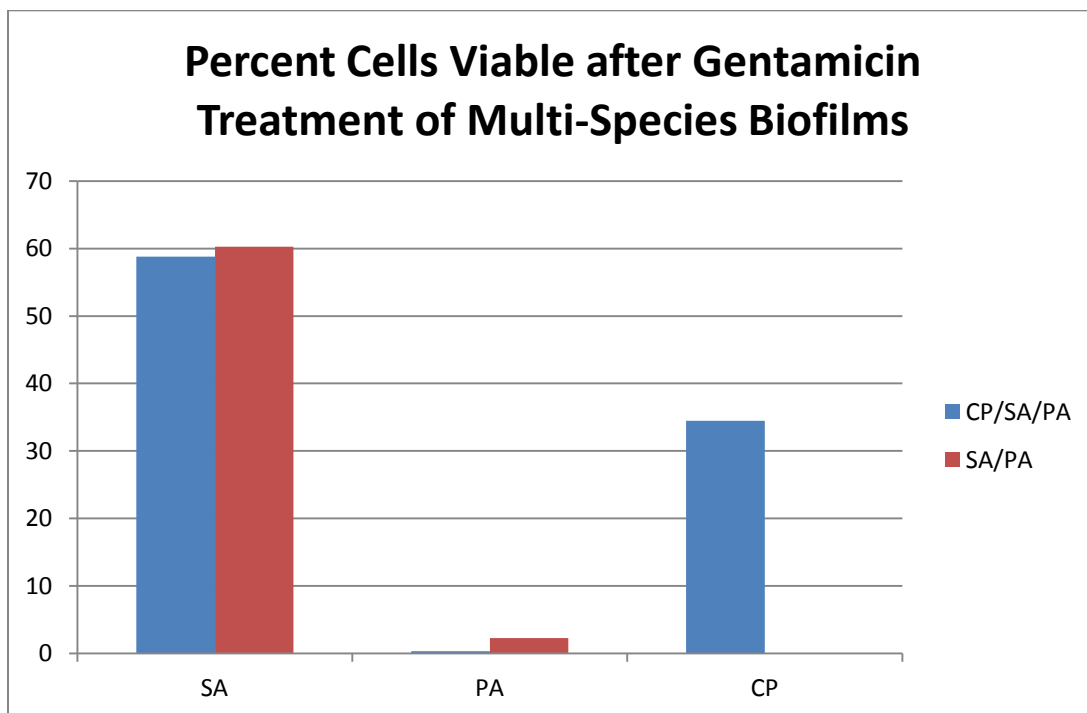


Figure 2. The effects of the presence of CP on the gentamicin tolerance of SA31 and PA01 were observed in relation to the tolerance measured in SA31 and PA01 co-cultured biofilms. The presence of CP in the

biofilm seems to have little or possibly detrimental effects on the gentamicin tolerance exhibited by SA31 and PA01.

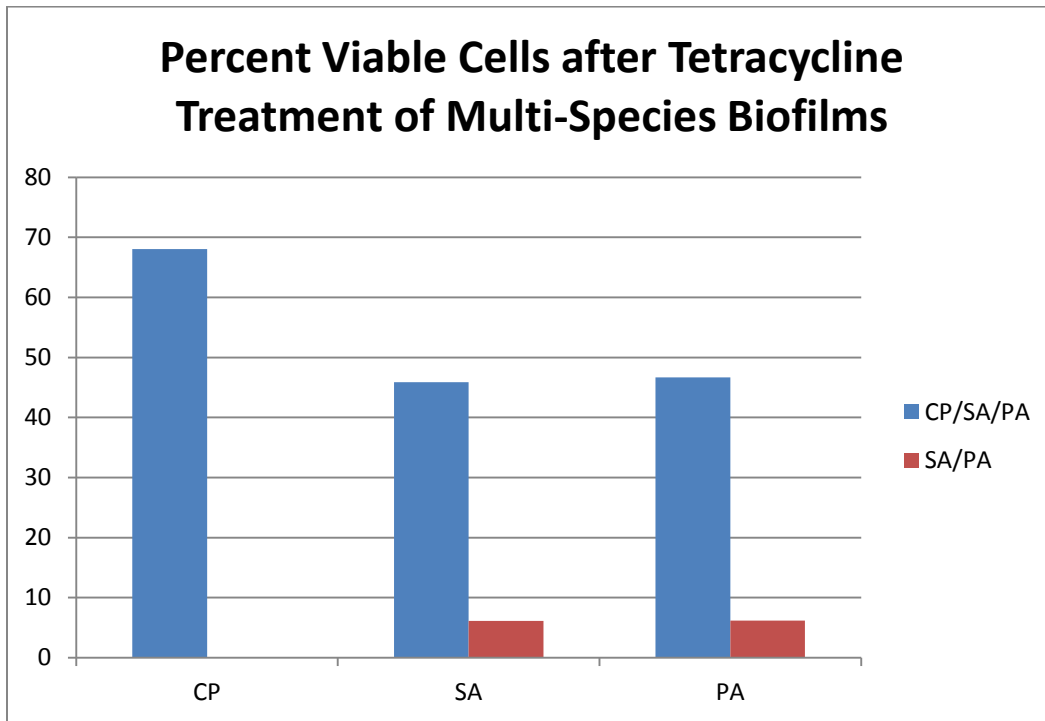


Figure 3. The effects of the presence of CP on the tetracycline tolerance of SA31 and PA01 were observed in relation to the tolerance measured in SA31 and PA01 co-cultured biofilms. Data from this assay have consistently shown that the presence of CP in the biofilm significantly increases the tolerance of SA31 and PA01 to tetracycline.

### Hayley Sparks

I am a senior at Texas Tech majoring in Microbiology. I have been participating in undergraduate research for a year now. My own personal scientific interest lies in the spread and control of infectious diseases, specifically those caused by viruses. I am currently preparing to apply to doctorate programs focusing on Virology or Epidemiology. I plan to pursue a career in epidemiology, working to contain and control disease outbreaks. My other interests include reading and bird watching.

TYRE: An *in silico* Simulator of Yeast Population Dynamics  
Jessica Stilwell  
Mentor: Dr. Brandt Schneider

Aging may be inevitable, but man has been seeking a formula for eternal life since the beginning of time. Researchers are actively searching for ways to prolong life, prevent disease, and restore worn-out or damaged tissues. It has long been observed that there is a clear relationship between cell size, proliferation, and lifespan. Dr. Brandt Schneider's lab at TTU HSC has been attempting to elucidate the genetic pathways by which these three characteristics are regulated in the yeast *S. cerevisiae* with the hope of drawing conclusions regarding the biological mechanisms by which mammals age. My role in Dr. Schneider's lab is the development of a Theoretical Yeast Replicator Emulator (TYRE) in MATLAB to model yeast population dynamics utilizing various user-input parameters and mathematical growth equations. It is our goal to ensure the reliability of the model within a certain statistical confidence interval and from there, to make generalizations about how variables such as initial cell size, critical size, and mass doubling time work together to generate yeast populations.

It has been noted that every strain of yeast produces a characteristic curve of cell size distributions. These graphs are generated using a Coulter counter, which sorts cells based on their volumes and graphs the size versus frequency of the population. Each strain's curve is consistently reproducible. With this in mind, we developed a simulator which grows a population from one initial cell and produces a graph comparable to Coulter curves obtained *in vivo* in addition to reporting the mean size, mode, and number of cells in the resultant population. The simulator's output curve is determined by eleven user-input parameters in addition to a mathematical formula for either linear or exponential growth.

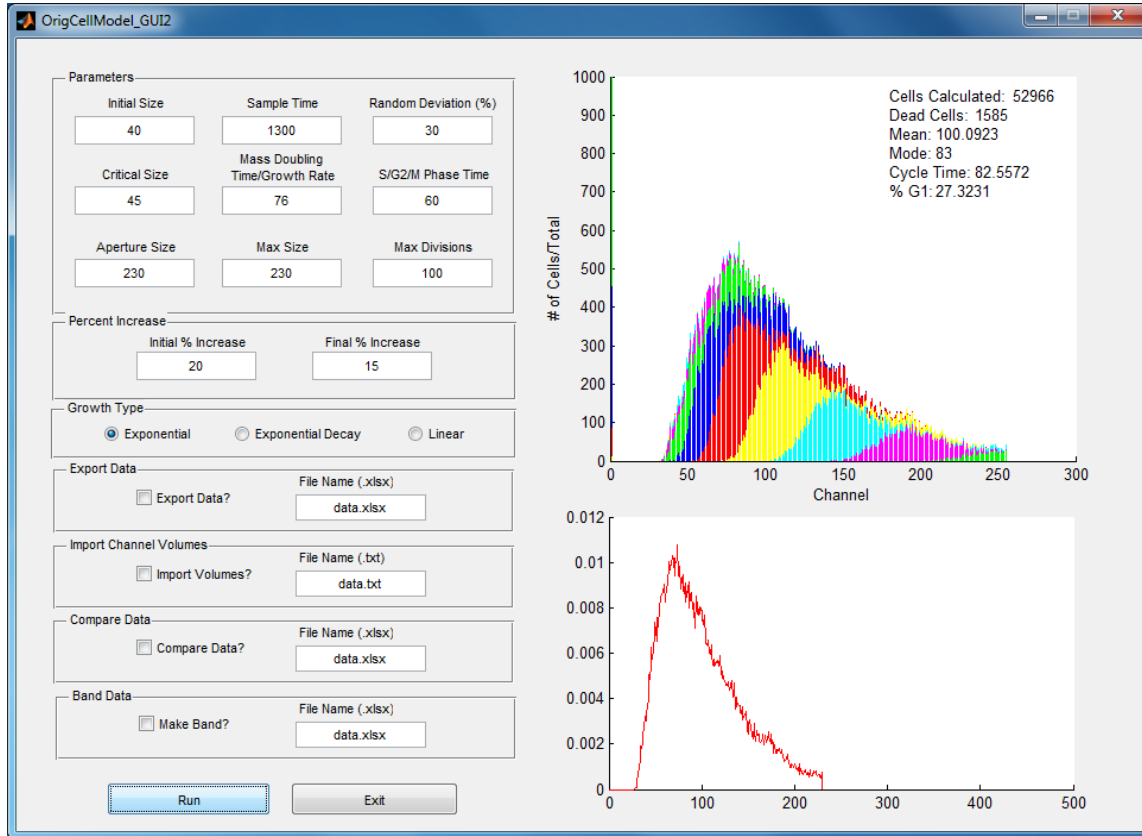
The variables available for the user to adjust are initial cell size, critical cell size, percent of random deviation, mass doubling time, S/G2/M phase time, sample time, maximum cell size, aperture size, maximum divisions, and initial and final percent growth. Among these parameters, some affect the output more than others. For example, the mass doubling time, defined to be the amount of time it takes for a cell to double in volume, is a major determinant of the mean size of the population in addition to the number of cells produced within the sample time. However, initial cell size merely serves as a starting point for the model and has little effect on the population as a whole.

In addition to the eleven parameters that can be adjusted, there are three modes of growth built into the simulator. The first is time-dependent linear growth, which merely grows each cell a set amount each time step, and the second is size-dependent exponential growth which increases the volume added to the cell exponentially each time step. The capability of the model to grow populations according to different mathematical formulas is essential to helping us understand how cells gain mass. There has been a long-standing debate among cell biologists regarding whether cells grow exponentially or linearly, and it is our hope that the simulator may shed some light on the subject.

We have successfully matched the Coulter curves of many different strains of large and small yeast in addition to some on dietary restriction. However, a visual confirmation that the Coulter graph and the simulator output are similar will not suffice in publication. Therefore, we have developed a statistical test based on the Monte Carlo method. First, we will make a band of *in vivo* curves from each strain to be matched. From this, we will determine how often the data overlaps *in vivo*. This percentage will be the threshold for how many points within a simulated curve must fall within the band in order to be considered a statistically significant match.

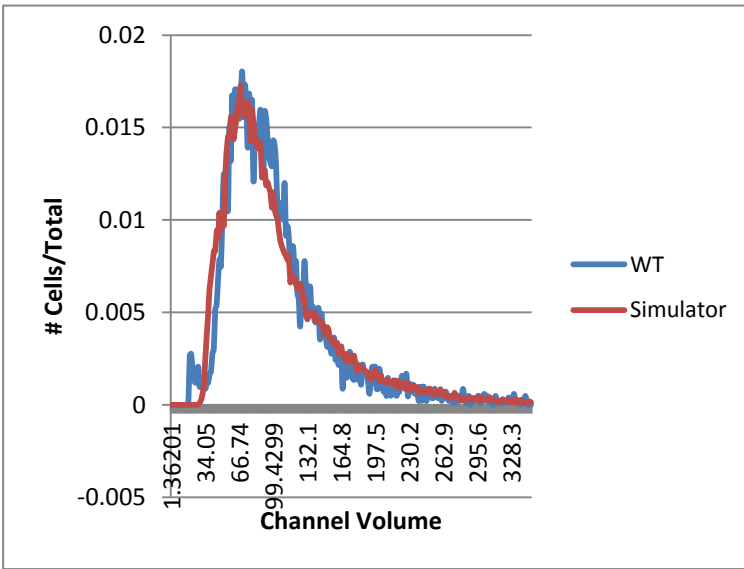
The genetic pathways determining cell size and their relationship to lifespan have yet to be elucidated. However, it is clear that understanding the cellular aging process will have implications for the treatment of cancer, Alzheimer's, and other diseases. We have developed TYRE using computer programming techniques and mathematics that suggest some potential answers to the puzzle, and we are confident that our model will help guide us towards the truth and further the study of cell biology.

**Figure 1**



**Theoretical Yeast Replicator Emulator (TYRE).**

Figure 2



**WT**

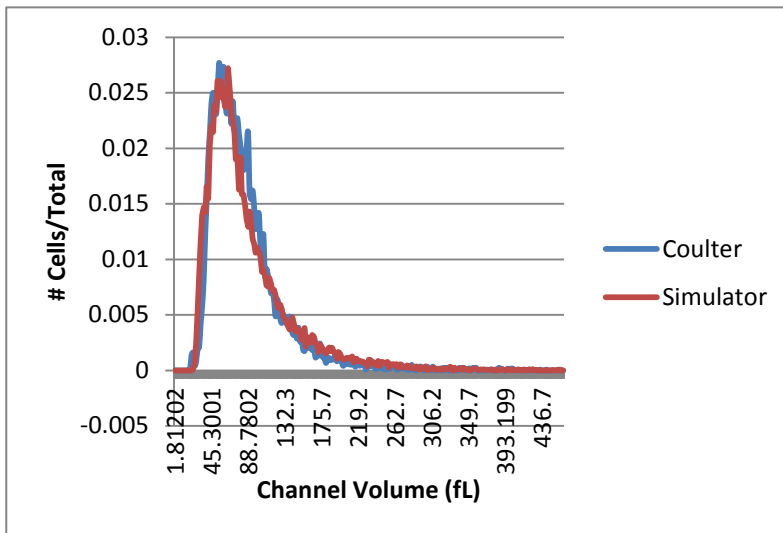
Mean: 102.1  
Mode: 70.15

**Simulator**

Mean: 102.17  
Mode: 68.04

A curve made by the simulator (red line) that matches a diploid wildtype Coulter Counter curve (blue line).

Figure 3



**BY4743 2%**

Mean: 85.46  
Mode: 57.08

**Simulator**

Mean: 87.3  
Mode: 65.22

A curve made by the simulator (red line) that matches a haploid wildtype Coulter Counter curve (blue line).

**Jessica Stilwell**

I am a senior with a major in electrical engineering and a minor in mathematics. I have had the pleasure of working in Dr. Brandt Schneider's lab since June 2011, and I am currently in the process of developing a simulator in MATLAB which will model yeast population dynamics. In the future, I would like to pursue a graduate degree in electrical engineering, and ultimately, I would like to start my career in analog hardware design, research and development, or robotics. In my free time, I enjoy singing, playing guitar, and songwriting.

## Hydrodynamic Resistance of a Train of Confined Microfluidic Droplets

Naureen Suteria

Mentor: Dr. Siva Vanapalli

Microfluidic devices have become increasingly popular in the fields of science and engineering due to their ease of fabrication. These devices have significant potential for a wide range of applications including biomedical diagnostics and biosensing. Many of these applications involve the transportation, sorting and storage of droplets.

When a single-phase fluid flows in a confined channel, there is a resistance to flow. Two-phase fluid flow, which includes droplets, introduces even more resistance to flow, known as hydrodynamic resistance of the droplets. The hydrodynamic resistance of droplet is the main key factor that controls the behavior of the droplets in a channel. Researchers have tried to determine the parameters that may affect the hydrodynamic resistance of droplets in a microfluidic channel. However, their studies have limited domain of parameters. The purpose of my research is to measure the hydrodynamics resistance of droplet over a wider range of parameters, which include capillary number ( $Ca$ ), drop length ( $L$ ), intra-drop spacing ( $\lambda$ ), viscosity ratio and surface tension between the inner and outer phase.

The benefit of using a microfluidic device is that the fabrication process is fairly quick and cost effective. After a design has been made into a photomask, soft lithography techniques are used to create a master mold on a silicon wafer. A polymer, polydimethylsiloxane (PDMS), is then cured on the mold to create the device. The device must be bonded to a glass slide or a thin layer of PDMS before it can be used. The microfluidic device that I use has two key components. One is a comparator, which is used to measure the resistance of a droplet. It has 2 parallel channels where it directly compares a reference channel with known resistance to a parallel test channel containing droplets of unknown resistance. Upstream from the comparator is the other key component, a cross-junction mechanism. It is used to create the water-in-oil droplets and control the drop size and intra-drop spacing.

Before the resistance of the drop can be measured, I first needed to make sure that the device was producing drops with consistent size and spacing. Figures 1 and 2 show the drop size and spacing as a function of the oil flow rate. There were 6 different oil-to-water flow rate ratios used to generate the droplets. The drop sizes are fairly consistent, all within 5% of the average for each condition. There is a bit more scattering with the drop spacing, which is caused by oil leaking around the edges of the channel walls. Even with this disturbance, there is still only about a 10% error for the data.

The resistance of the drop, normalized with the resistance of the reference channel, is plotted as a function of the capillary number of the oil in figure 3. From this data, it can be inferred that as the drop size increases, the resistance of the drop increases. This is supported by intuition that increasing the volume of a fluid will increase its resistance in a confined channel. Another important observation about this figure is that for a fixed capillary number (or flow rate) the resistance increases as drop size increases.

Lastly, the effect of oil leaking around the drop was analyzed, in figure 4. This figure shows the velocity of the drop as a function of the velocity of the oil. Typically, we would assume that the oil and drop are flowing at the same rate, since they are in a confined channel and their flow is controlled by syringe pumps, however, the droplet is moving significantly faster than the carrier oil. This is due to the fact that the walls of the channels are hydrophobic, and thus repel the water droplet. This allows oil to move in the opposite direction on the walls and corners and thus has an accelerating effect on the droplet.

This research project is not yet complete, as many parameters have yet to be analyzed. One important aspect is to decouple the drop size and spacing and to see what effect spacing has on the resistance without changing the drop size. Also, a surfactant was used to coat the water droplets, which changes the surface tension between the fluids, and it's effect also needs to further investigated.

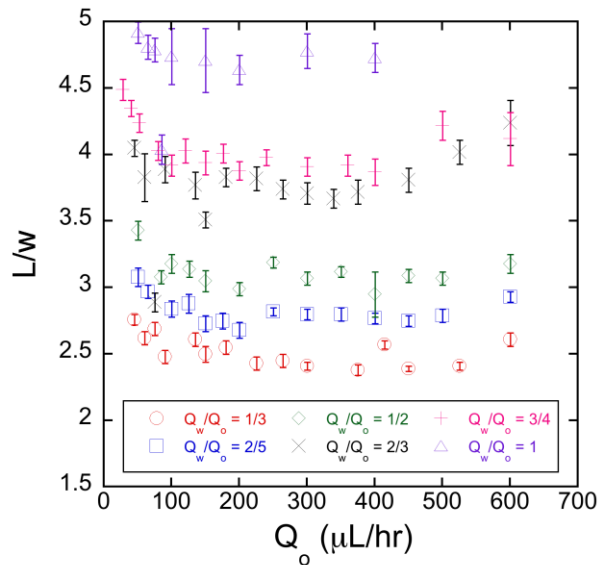


Figure 1: Drop size as a function of oil flow rate.

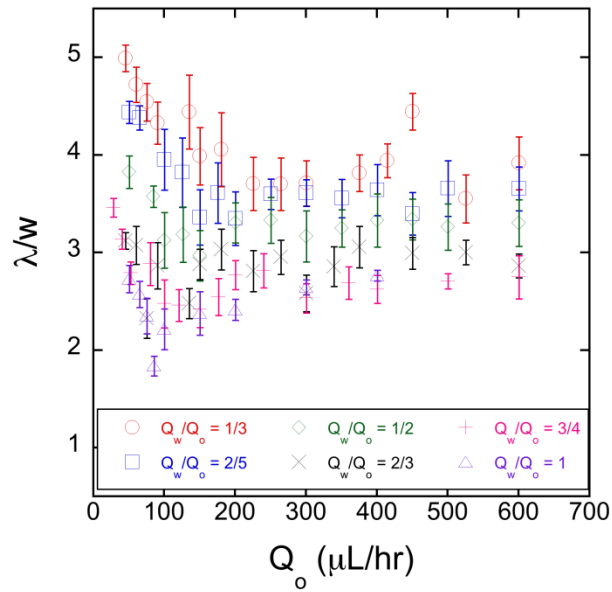


Figure 2: Intra-drop spacing as a function of oil flow rate.

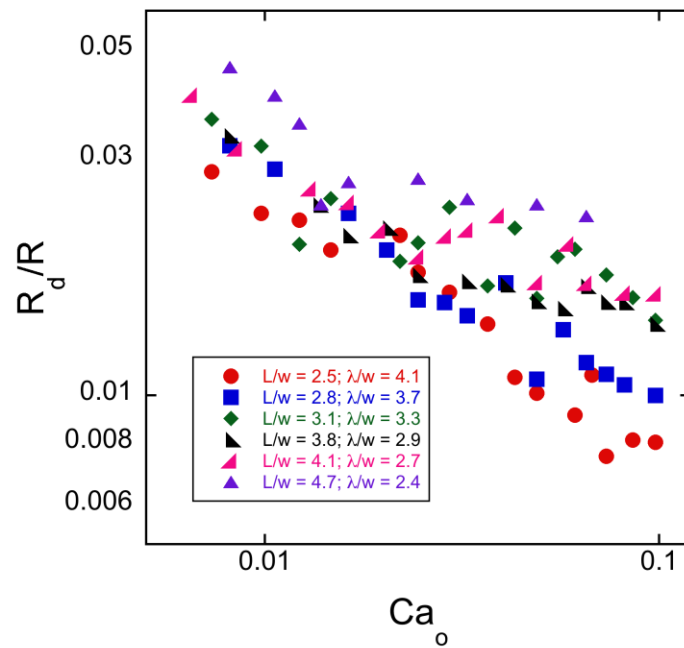


Figure 3: The resistance of a confined drop as a function of capillary number plotted on a log-log scale

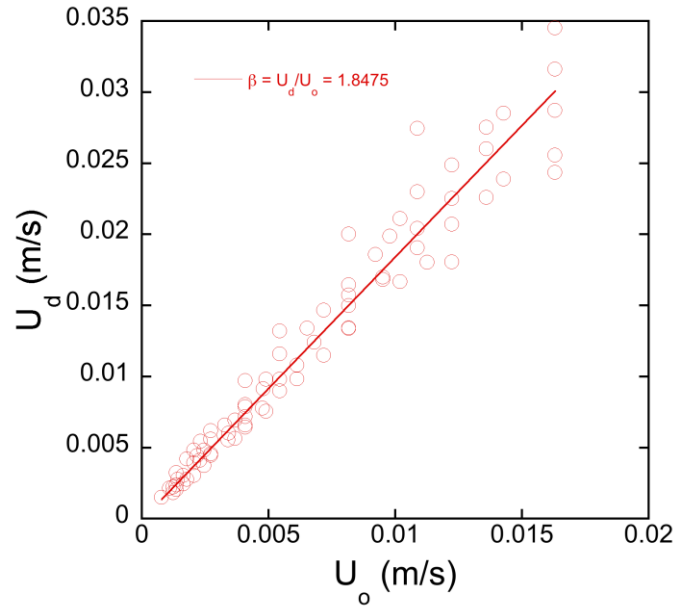


Figure 4: The velocity of a drop as a function of the fluid velocity.

**Naureen Suteria**

I am currently a senior studying Chemical Engineering at Texas Tech University. I have been working in Dr. Vanapalli's research lab for the past year. My research interests include droplet-based microfluidics, which I hope to explore more in depth as I work on obtaining my Master's degree in Chemical Engineering from TTU. In my spare time I enjoy expressing my creativity by painting and creating new recipes in the kitchen.

Effects of Ocean Acidification on Otolith Formation in Red Drum, *Sciaenops ocellatus*  
Joshua Willms  
Mentor: Dr. Sandra Diamond

Red drum, *Sciaenops ocellatus*, is a common species of bony fish that live in the Atlantic Ocean and the Gulf of Mexico and are a primary target of commercial and recreational fisheries. Information concerning the ecological sustainability of red drum, as well as other bony fish, is essential for effective fisheries management. One environmental factor that could drastically influence the distribution and abundance of red drum is ocean acidification. The ocean acts as a 'sink' for CO<sub>2</sub> emissions, which results in a global decrease in surface water pH<sup>2</sup>. As the pH decreases, it becomes increasingly difficult for calcifying organisms to uptake calcium from the environment<sup>2</sup>. This compromises the structural integrity of shells and skeletons in shellfish, coral, and other marine organisms that rely on calcium for physiological structures. The effects of ocean acidification on bony fish, however, are poorly understood at the present time. Because no established methodology has been developed for examining the effects of pH and temperature on bony fish, our hope is to design an experimental procedure so that the effects of ocean acidification can be examined across a suite of bony fish.

Otoliths are bones found in the ears of fish. Fish regularly lay down layers of bone onto otoliths, and elements from the environment are incorporated as this takes place. By examining these elements the location of fish over time can be determined. The integrity of this tracking method can be compromised by changes in water chemistry, such as lower pH, changes in dissolved oxygen concentration (DO), and increases in temperature. By 2100, surface ocean pH will be 0.4-0.5 units lower than preindustrial values and the DO in parts of the Gulf of Mexico will be below 2 mg/L. Temperature change will vary depending on geographical location, but will generally increase. While ocean acidification, hypoxia, temperature change, and other modifications in water chemistry have been shown separately to affect elemental incorporation in marine organisms, combinations of parameters have not been examined together in a controlled setting. The purpose of this study is to examine potential synergistic or antagonistic effects of simultaneously lowered pH and increased temperature.

In this study red drum will be exposed to pH and temperature values projected for Gulf of Mexico in 2100. Changes in elemental incorporation based off of climate change scenarios will be used to modify techniques so that they will be applicable for future studies. Preliminary results show that when pH is lowered, otoliths increase in size. This result goes against expectation, because bony fish are succeeding in incorporating more calcium into their otolith structures, whereas other calcifying organisms are experiencing the opposite effect.

#### References

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- Jacobson, M. Z. (2005), Studying ocean acidification with conservative, stable numerical schemes for nonequilibrium air-ocean exchange and ocean equilibrium chemistry, *J. Geophys. Res.*, 110, D07302, doi:10.1029/2004JD005220.

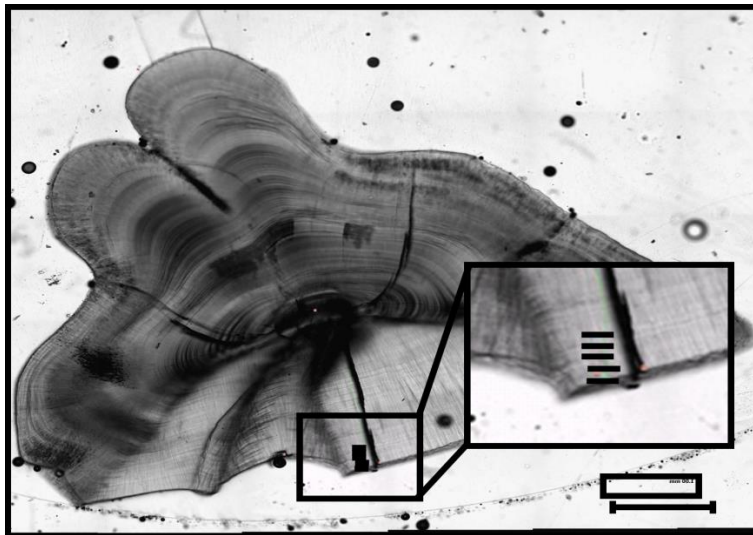


Figure 1.  
Growth Rings Bony fish add a ring of bone to their otoliths daily, analogous to the way trees add growth rings (Source of image: Sullivan 2009).

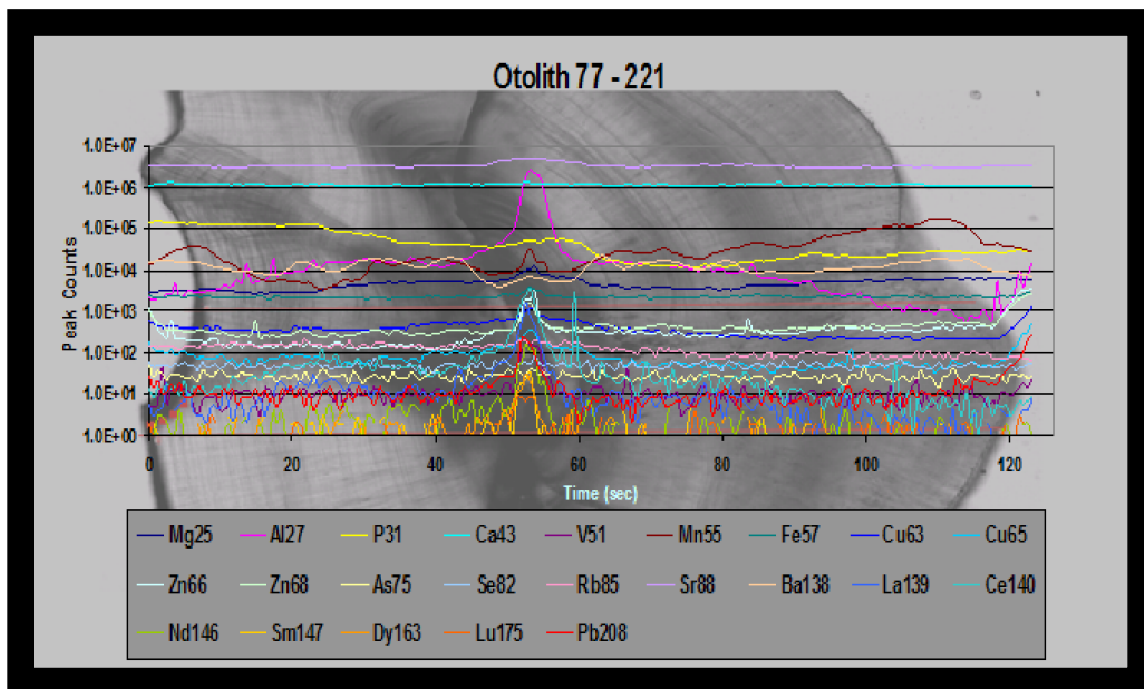


Figure 2.  
Laser Ablation ICPMS: LA-ICP-MS element signatures were superimposed over an otolith picture, corresponding to the raster path. (Source of image: Sullivan 2009). Counts of elements are shown in colored lines.

**Joshua Willms**

I am a senior majoring in Biology and Classics (with an emphasis on Greek language). I work in both the Diamond and Schwilk labs, and I am conducting an honors thesis outside of my majors in physics and cosmology. My ultimate goal is to research neurology or genetic engineering through a joint MD/PhD program. Outside of school, I train and compete in Brazilian JiuJitsu on a regular basis.

## Using a Public Content Management System (CMS) to Mitigate Security Risks

Alex Woollends

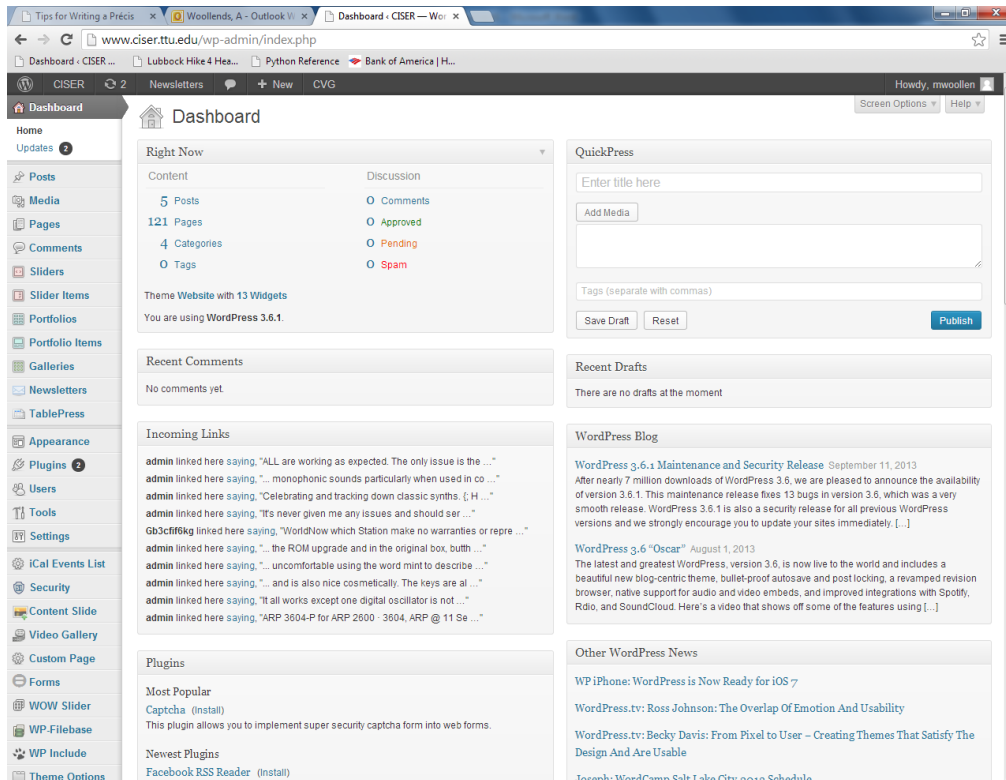
Mentor: Brian Enderson

Public-facing websites attract attention, and some of this attention is unwarranted. Secure information such as phone numbers, email addresses, or even physical addresses of persons can be obtained, even though this information might be available to only administrative users on the site. By creating a public site and backing up the information in another content management system or database, such as Microsoft SharePoint, we can more effectively 'hide' the secure information from public attention. If the public-facing website should go down, the data can be uploaded relatively quickly. My mentor, Brian Enderson, and I, are creating a public-facing website for the CISER webpage while managing SharePoint services and secure data on a private site. The original idea of creating a public-facing site for non-secure content was Brian's.

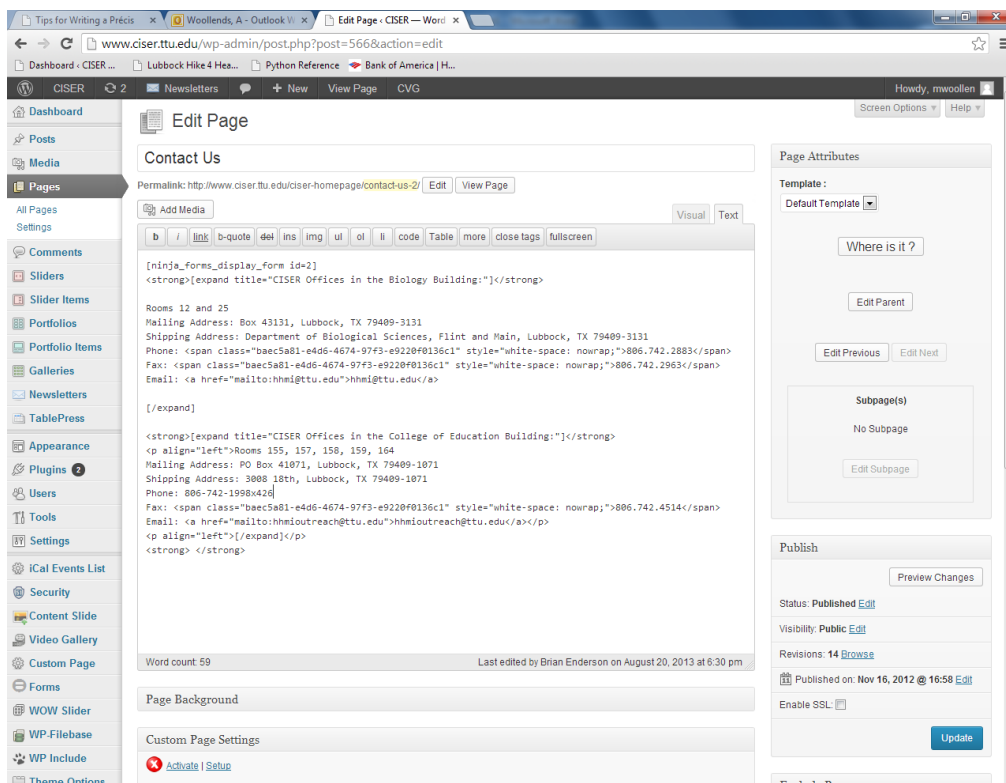
The project consists of a private SharePoint 2010 site and a public facing one running on the Content Management System from WordPress. WordPress is a quick and easy to use CMS that uses a Graphical User Interface (GUI) for its interface. The GUI enables quick and easy content uploads and editing.

There is the ability to customize the appearance of the site by modifying the HTML (Hypertext Markup Language) and CSS (Cascading Style Sheets) code. WordPress also has a robust online community, and is used by millions. Because of its heavy usage and large community, compatibility problems are often fixed relatively quickly. Plugins, or widget like software, install in the main template of the WordPress site that you work with to give additional functionality.

This functionality can be in the form of additional security, rich text editing, slide-shows, calendars, and RSS feeds. The modular design of WordPress and its easy to use interface made it a good choice for our CMS. While modifying HTML and CSS code has been necessary in some instances to achieve a certain look, reliance on programming and/or modifying code has been kept to a minimum thanks to the robust GUI. This means that future Undergraduate Research Scholars can take over the site management without having to understand programming languages and the code left behind by others. This means that development can continue relatively smoothly for other web administrators.



The main 'Dashboard' of WordPress. Notice the GUI interface and clean design.



An example of the HTML editor in WordPress – notice the 'text' tab near the visual. This demonstrates the two facets of the design – one GUI, the other code.

**Alexander Woollends**

I am senior majoring in Mathematics, with a minor in philosophy. I work in the Biological Sciences Building with Julie Isom as the webmaster for the CISER and Hike4Health website (the former is still 'under construction'). I have not decided what I want to do with regards to my undergraduate degree, but I feel as though graduate school is in my future. My interests and hobbies include gaming and philosophy, especially with regards to epistemology and ethics. A Catholic, I invest a significant amount of time researching theology and its intersection with ethics and epistemology.