

2016

High Point University

Summer Undergraduate Research Program in the Sciences (SuRPS)

Final Research Symposium



Thursday, July 28 & Friday, July 29, 2016
Phillips 120

2016

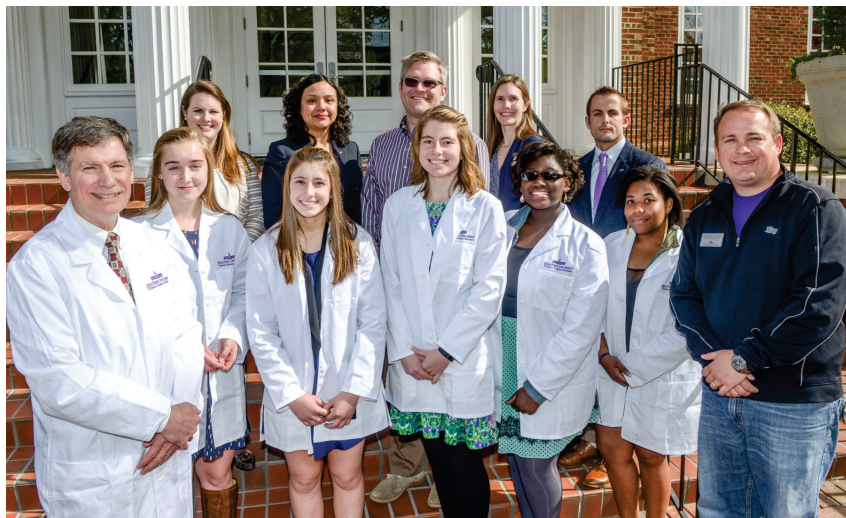
High Point University

Summer Undergraduate Research Program in the Sciences (SuRPS)

2016 SuRPS Students and Faculty



2016 Draelos Scholars and Mentors



SuRPS KEYNOTE SPEAKER

"Soirées in the soil: Bacterial Communication, Cooperation, and Competition in the Rhizosphere"



Sarah Craven Seaton, Ph.D (HPU Chemistry/Biology Class of 2004)
Assistant Professor, Department of Biology
University of North Carolina Asheville

ABSTRACT

In nature, bacteria live in stunningly diverse communities wherein each member must signal, sense, and respond to its neighbors, and such bacterial cell-cell interactions run the spectrum from antagonistic to cooperative. Work in the Seaton laboratory explores cooperation and competition in complex bacterial communities that make their home in the rhizosphere, with a focus on 1) the mechanisms that soil- and plant-associated bacteria use to sense and respond to their neighbors and 2) the interesting phenotypes that emerge when bacteria are confronted with competing species. The seminar will explore two such interaction-induced phenotypes—increased antibiotic production and the emergence of a novel cooperative motility—and discuss the practical applications of exploring bacterial behavior in mixed culture.

BIO

Sarah Craven Seaton is an Assistant Professor of Biology at the University of North Carolina Asheville. She is a graduate of High Point University, earning B.S. degrees in Biology and Chemistry in 2004. She completed her doctoral work in microbiology at the University of Georgia, and then served as a postdoctoral research fellow in the laboratory of Stuart Levy at Tufts University School of Medicine. Dr. Seaton's current work seeks to isolate novel antibiotic-producing bacteria, with the hopes of discovering antibiotics with clinical or agricultural significance.

SuRPS Final Symposium

(Thursday, July 28, 2016, Phillips 120)

Session A: Dr. Michael Grider, Department of Biology, Presiding

	8:30 – 9:05		COFFEE, TEA RECEPTION (Phillips 120)
	9:05 - 9:15	Dr. Brian Augustine	Opening Remarks
Th.1	9:15 - 9:30	Kristen Brokaw	Elucidation of the Efficacy of the Antitumor Quinone, Beta-Lapachone in a BRCA1 Mutant Breast Cancer Cell Line Expressing Elevated NQO1 Levels
Th.2	9:30 - 9:45	Alan Vasquez	The Disappearing Pulsations of the Hot Subdwarf Star CS 1246
Th.3	9:45 - 10:00	Kyra Gillard	Investigation of FDA-Approved Antipsychotics as Modulators of Virulence in Methicillin-Resistant <i>Staphylococcus aureus</i>
Th.4	10:00 - 10:15	Amiras Simeonides	Constructing and Testing a TIRF-FCS Microscope
Th.5	10:15 - 10:30	Sarah Edmark	Characterizing a Potential Mannose-6-Phosphate Receptor Homology Domain in Yeast
	10:30 - 10:45		BREAK

Session B: Dr. Melissa Srougi, Department of Chemistry, Presiding

Th.6	10:45 - 11:00	Daniel Magurno	Does Argon Gas Treatment Affect Cell Viability Following Stroke Model Injury?
Th.7	11:00 - 11:15	Jason Barbaretta	A Novel Application of TEC-disulfide Replacement Showcased in the Synthesis of SFTI-1
Th.8	11:15 - 11:30	Jena Dryden	Effects of Nicotine on Embryonic Development
Th.9	11:30 - 11:45	Spencer Ader	Developing a Self-Driving Car and an Introduction to Computer Vision
Th.10	11:45 - 12:00	Amanda Goodwin	Tat-specific Factor 1's Role in HIV RNA Stability
	12:00 - 1:15		LUNCH BREAK

SuRPS Final Symposium

(Thursday, July 28, 2016, Phillips 120)

Session C: Dr. Veronica Segarra, Department of Biology, Presiding

Th.11	1:15 - 1:30	Jonah Winkler	Utilization of Molecular Dynamics to Examine the Physical Properties of Different Hydrocarbons for Alternative Fuels
Th.12	1:30 - 1:45	Sarah Mastropietro	The Effect of Oxybenzone and Methylparaben on Bone Development and Breast Cancer Cell Growth
Th.13	1:45 - 2:00	Harrison Seitz	Photoenzymatic Repair Capability in the Freshwater Cladoceran Genus <i>Scapholeberis</i>
Th.14	2:00 - 2:15	Kaitlyn Griffith	Construction and Alignment of a Total Internal Reflection Fluorescence Microscope
Th.15	2:15 - 2:30	Max Maurer	Computer Vision Techniques for Obstacle Detection and Tracking in Autonomous Vehicles
	2:30 - 2:45		<i>BREAK</i>

Session D: Dr. Martin DeWitt, Department of Physics, Presiding

Th.16	2:45 - 3:00	Alexandra Sprouse	Relative Contributions of Apoptosis and Necrosis in <i>In-vitro</i> Models of Stroke
Th.17	3:00 - 3:15	Lisa Nguyen	Quorum Sensing and Programmed Cell Death in <i>Escherichia coli</i> and <i>Bacillus subtilis</i>
Th.18	3:15 - 3:30	Joshua Williams	Effects of the Estrogen Mimicking Neutraceutical, Resveratrol on Bone Development
Th.19	3:30 - 3:50	Ambar Khawaja Deanna Lee	Identifying a Mammalian Functional Substitute for Atg27 Identifying Protein Cargo Recognized by Atg27
	3:50 - 4:00	Dr. Kristen Bowey	Recognition of Draelos Scholars

(Note: Friday, July 29 session continued on next page)

SuRPS Final Symposium

(Friday, July 29, 2016, Phillips 120)

Session E: Dr. Meghan Blackledge, Department of Chemistry, Presiding

8:30 – 9:00

COFFEE, TEA RECEPTION (Phillips 120)

Fr.1	9:00 - 9:15	Matthew Warrick	Exploring the Role of Tat-SF1 as an HIV-1 Host Factor
Fr.2	9:15 - 9:30	Olivia Tornow	Towards Further Understanding of Kinase Activity During Oxidative Stress: Synthesis of the Highly Active ERK2 Substrate Sub-D
Fr.3	9:30 - 9:45	Ryan Hegedus	The EREBOS Project: Studying the Effects of Substellar Companions on Stellar Evolution
Fr.4	9:45 - 10:00	Thomas Kylo	Photoenzymatic Repair Capabilities of Diaphanosoma spp. in Response to UV Radiation
Fr.5	10:00 - 10:15	Mallory McKee	The Effects of Nicotine on Zebrafish Development
Fr.6	10:15 - 10:30	Lindsey Palmquist	Evaluating the Efficacy of the Anti-Tumor Agent Beta-lapachone in NQO1 Expressing BRCA2 Mutant Human Breast Cancer Cells
Fr.7	10:30 – 10:45	Ty Carlson	Computer Vision Techniques for Obstacle Detection and Avoidance

10:45 - 11:00

BREAK

Keynote Address: (Introduction by Dr. Dinene Crater, Department of Biology)

11:00 - 11:50	Dr. Sarah C. Seaton (HPU Class of 2004) UNC-Asheville	Soirées in the Soil: Bacterial Communication, Cooperation, and Competition in the Rhizosphere
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11:50 – 12:00 Dr. Angela Bauer Closing Remarks

12:15 - 1:45 *GROUP LUNCH (Wanek Center 2nd Floor Farmer's Market)*

2016 SuRPS Faculty Participation and Projects

Department of Biology

Dr. Kristin Ackerman	“Effects of nicotine on early spinal cord development in zebrafish”
Dr. Neil Coffield	“Regulation of bone ossification in <i>Danio rerio</i> (zebrafish) by estrogenic endocrine disruptor”
Dr. Sandra Cooke	“Comparing the ultraviolet radiation tolerance of freshwater zooplankton populations from transparent and turbid lakes”
Dr. Michael Grider	“Neuroprotective properties of argon gas in ischemic stroke models”
Dr. Veronica Segarra	“Mutagenic characterization of a newly-recognized mannose-6-phosphate receptor domain in Atg27 and its role in autophagy”

Department of Chemistry

Dr. Meghan Blackledge	“Synthesis and evaluation of extracellular death factor peptide analogs to probe SAR of programmed cell death in <i>E. coli</i> and <i>B. subtilis</i> ”
Dr. Keir Fogarty	“Characterization of retroviral gag-cell membrane binding interactions using fluorescence microscopy and spectroscopy”
Dr. Todd Knippenberg	“A computational study of condensed phase properties of hydrocarbon fuels”
Dr. Heather Miller	“Investigating human Tat-specific factor 1’s role in HIV-1 gene expression”
Dr. Melissa Srougi	“Targeting of <i>BRCA1/2</i> mutant human breast cancers overexpressing NQO1 using the antitumor quinone β -lapachone”
Dr. Andrew Wommack	“Photoredox catalysis to probe biological function: employing a redox-inert disulfide bioisostere”

Department of Physics

Dr. Brad Barlow	“Characterizing amplitude variations in the pulsating hot subdwarf star CS 1246”
Dr. Martin DeWitt	“A strategy to enhance visual-based obstacle avoidance routines in unmanned aerial systems”

Special Thanks:

Dr. Joanne Altman, Director HPU Undergraduate Research and Creative Works Program

Dr. Kristen Bowey, Department of Biology Laboratory Manager

Sheena Valenti, Department of Chemistry and Physics Laboratory Manager

Rebecca Smoak, Administrative assistant extraordinaire

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Joint School of Nanoscience and Nanoengineering, Greensboro, NC

Ameritox Corporation, Greensboro, NC

STUDENT ABSTRACTS:

{Note: presenting author is underlined, * denotes faculty advisor(s)}

(Th.9) Developing a Self-Driving Car and an Introduction to Computer Vision

Spencer Ader and Martin DeWitt*

Department of Physics
High Point University

Before a car can be made to drive itself, a single board computer is installed and attached to a camera that serves as the car's eyes. The computer is programmed to control the steering servo and the motor by sending pulses with a specific duty cycle to each motor. With each frame that the camera passes to the computer, the program will send a different duty cycle signal to the servo and motor. A simple program can manipulate the car to do what is desired. The computer language Python and a library of computer vision functions imported from Open Computer Vision were used to help the car see. Using these functions, the car follows a yellow ball, stays on a brick path, recognizes multiple defined objects, and avoids non-specific obstacles. The first task in making the car see was to follow a predefined object. To achieve this, a color histogram of the target in Hue, Saturation, and Value (HSV) was used and then compared to a live image from the car. This isolated the target from the live image and showed the object as black pixels in a background of white pixels. When the target is detected, the steering is corrected to keep the ball in the center of the screen using a proportional function between distance from center and steering angle.

(Th.7) A Novel Application of TEC-disulfide Replacement Showcased in the Synthesis of SFTI-1

Jason Barbaretta and Andrew Wommack*

Department of Chemistry
High Point University

Disulfide bonds are critical to tertiary structure and cell signaling in cysteine-containing peptides and proteins. Disulfide bridges form between the thiol groups of two cysteine residues through the process of oxidative folding. Although disulfide bonds have high bond dissociation energy, these bonds are readily liable in reducing environments commonly found in biological settings. Upon replacement of one sulfur atom with a carbon, a thioether bond is formed to deliver a disulfide surrogate that is biologically redox-inert. Thiol-ene coupling (TEC), a radical-mediated reaction between a thiol and an alkene, forms a thioether group and functions as a disulfide replacement. Driven by ruthenium catalysis under blue LED irradiation, this photoredox method efficiently replaces one sulfur atom of the native disulfide with a carbon to deliver the desired thioether functional group. The synthetic product provides improved protease stability while retaining the structural and functional characteristics of the disulfide bond. To showcase this novel TEC application, sunflower trypsin inhibitor-1 (SFTI-1) was prepared by Fmoc solid-phase peptide synthesis. SFTI-1 is a 14-residue disulfide-bridged peptide bicycle (DBPB) and a subnanomolar inhibitor of β -trypsin. As one of the smallest and most powerful naturally occurring plant protease inhibitors, SFTI-1 is an attractive therapeutic candidate. The non-reducible nature of the TEC-disulfide replacement aims to increase SFTI-1 stability in physiologically relevant reducing environments, while retaining powerful protease inhibition. Future directions of this study will examine how TEC-disulfide replacement in SFTI-1 influences its ability to inhibit the serine proteases including matriptase, an enzyme implicated in breast cancer proliferation and metastasis.

(Th.1) Elucidation of the Efficacy of the Antitumor Quinone, Beta-Lapachone in a *BRCA1* Mutant Breast Cancer Cell Line Expressing Elevated NQO1 Levels

Kristen Brokaw and Melissa Srougi*

Department of Chemistry
High Point University

Breast cancer affects 1 in every 8 U.S. women and it is, therefore, important to find specialized treatments since many current approaches kill normal tissues in addition to cancerous tissues. NAD(P)H:quinone oxidoreductase 1 (NQO1) is present in normal cell tissues where it functions as a detoxification enzyme. However, breast cancer cell lines commonly contain elevated levels of NQO1. The ortho-naphthoquinone β -lapachone (β -lap) has been found to be toxic in cells with elevated NQO1. NQO1 reduces β -lap to a highly unstable hydroquinone, which then oxidizes back to the parent form via a semiquinone intermediate, thus creating a futile redox cycle and generating reactive oxygen species (ROS). ROS-induced DNA damage hyperactivates poly(ADP-ribose) polymerase 1 (PARP-1)—a protein involved in DNA repair—resulting in the loss of NAD⁺ and ATP leading to cell death. A subset of breast tumors have mutations in the *BRCA1/2* tumor suppressor genes. Tissues with these mutations have more difficulty repairing damaged DNA. Therefore, we hypothesize that *BRCA1* mutant cell lines expressing NQO1 will be more susceptible to β -lap-induced cytotoxicity. To test this hypothesis, the NQO1 expressing *BRCA1* mutant breast cancer cell line HCC1937 was used. These cells were transfected with the wild-type *BRCA1* gene. Survival assays were performed to test the sensitivity of β -lap and IB-DNQ (a β -lap derivative)—on HCC1937 cell survival. The results from our studies will provide information on the effectiveness of β -lap as a selective therapy for NQO1 expressing, *BRCA1* mutant cancers.

(Fr.7) Computer Vision Techniques for Obstacle Detection and Avoidance

Ty Carlson and Martin DeWitt*

Department of Physics
High Point University

This project involves the development of an autonomous vehicle that is capable of detecting and avoiding obstacles. Multiple strategies for obstacle avoidance based on computer vision techniques will be presented. First, the case in which a vehicle follows a target object while avoiding predefined obstacles is based on the behavior of electrical charges. The vehicle and its target are represented as opposite charges and will attract each other as a result. Obstacles are represented by the same charge as the vehicle, causing the vehicle to be repelled by these obstacles on its path toward the target. The second case deals with obstacles that have not been predefined and focuses on the use of optical flow to detect the displacement of pixels between consecutive images. Optical flow can detect and differentiate between objects that are close to the vehicle from those that are farther away. This method allows the vehicle to identify a wider range of objects as obstacles. The third case deals with a different type of obstacle, large drop-offs, that can be detected using the Canny Edge Detection program. The algorithm relies on noise reduction and the intensity gradient of whatever image the camera is detecting. Although the program was not incorporated into the project, it holds potential for future autonomous vehicles.

(Th.8) Effects of Nicotine on Embryonic Development

Jena Dryden and Kristin Ackerman*

Department of Biology
High Point University

Smoking while pregnant increases the chance of miscarriage, premature birth, and SIDS (Center for Disease Control and Prevention, 2015). Other animal models used for embryonic nicotine exposure, such as rats and mice, show similar gross morphological defects as humans, but these models are complicated and extremely invasive. Although the gross morphological defects within rodents model human defects, the effects of nicotine at the cellular level to cause the malformations remains unclear. Thus, we are utilizing zebrafish (*Danio rerio*) due to their quick development (all organs are present by 3 days post fertilization (dpf)), translucent tissue (which allows for easily visualization of a whole embryo via light microscopy), and external fertilization. In past research, zebrafish embryos have been treated with 10-30 μM nicotine between 6 and 24 hours post fertilization (hpf). In our experiment, the embryos were treated at the 0-1 cell stage (to more closely mimic the effects of smoking during early conception of human fetus) and assayed for different morphological-based parameters such as growth, survival, heartbeat, and motor behavior. With nicotine concentrations of 100 nM, 10 μM , 50 μM , or 100 μM , we are able to observe differences in gross morphology of nicotine-exposed embryos relative to wild-type siblings. Using Polymerase Chain Reaction (PCR), we determined that nicotine does penetrate the embryos as demonstrated by a change in transcript levels of *cfos*, a gene that is typically upregulated within neuronal tissues after noxious stimuli such as nicotine. Preliminary data indicates that nicotine is altering heart rate and embryonic growth.

(Th.5) Characterizing a Potential Mannose-6-Phosphate Receptor Homology Domain in Yeast

Sarah Edmark, Taylor Cunningham and Veronica Segarra*

Department of Biology
High Point University

Autophagy is a type of membrane transport in eukaryotes in which damaged or unneeded parts of the cell are degraded and recycled. Autophagy is induced when cells are exposed to stressors like starvation or pathogens. Atg27 is a yeast transmembrane protein involved in autophagy and is composed of 3 domains: cytoplasmic, transmembrane, and luminal. While the cytoplasmic domain is known to contain sorting signals for proper Atg27 localization, the function of the luminal domain is unknown. Using structure prediction algorithms, we have identified that the luminal domain of Atg27 contains a mannose-6-phosphate receptor homology (MRH) domain. MRH domain proteins are often found in mammals and are used to recognize N-glycan groups in other proteins. To determine if the luminal domain of Atg27 contains a MRH domain, we used baker's yeast and introduced mutations in the predicted MRH domain that should abrogate its function. These mutants were then tested for normal autophagic function.

(Th.3) Investigation of FDA-Approved Antipsychotics as Modulators of Virulence in Methicillin-resistant *Staphylococcus aureus*

Kyra Gillard, Tyler Wilson and Meghan Blackledge*

Department of Chemistry

High Point University

Patrick Vigueira

Department of Biology

High Point University

Each year over 11,000 people die from methicillin-resistant *Staphylococcus aureus* (MRSA) infections. MRSA is particularly difficult to treat because it employs virulence factors such as biofilm formation and antibiotic resistance that allow it to evade many common therapies. MRSA, like all bacteria, mediate their behavior through a process called quorum sensing (QS). QS is a chemical signaling process that bacterial cells use to communicate with each other. In bacterial cells QS modulates bacterial virulence, including the formation of biofilms and regulation of enzymes involved in antibiotic resistance. Interrupting QS signaling pathways to prevent virulence is an attractive target for novel therapeutics. Researchers have been exploring compounds looking for QS modulators capable of acting as anti-biofilm therapies and antibiotic adjuvants capable of re-sensitizing MRSA to existing common antibiotics. Three FDA-approved antipsychotics, amoxapine, clozapine and loxapine, were investigated for their ability to inhibit and disperse biofilms and for the ability to resensitize MRSA to common β -lactam antibiotics. The antipsychotics' ability to inhibit biofilm formation for antibiotic resistant strains of *S. aureus* was evaluated using standard crystal violet absorbance assays. Data from the crystal violet absorbance assays and β -lactam antibiotic resensitization assays will be presented. By developing new adjuvant therapies, new methods of treating MRSA may be developed.

(Th.10) Tat-specific Factor 1's Role in HIV RNA Stability

Amanda Goodwin and Heather Miller*

Department of Chemistry

High Point University

Human immunodeficiency virus, HIV, is a retrovirus that while vastly studied, lacks complete understanding by the scientific community. Investigators are just beginning to identify the large number of host factors that HIV utilizes to propagate. In this study, we focused on the Tat specific factor 1, Tat-SF1, a human protein originally thought to play a role in viral transcription. However, recent findings demonstrate that Tat-SF1 may instead play a role in RNA stability. We hypothesize that if Tat-SF1 levels are knocked down in human cells, then altered stabilization of HIV RNA isoforms will be detected. An HIV-expressing plasmid was transfected into HeLa cells after Tat-SF1 was knocked down by RNA interference. Then, actinomycin D was added to stop transcription. Next, the treated cells were lysed at 0, 2, and 4 hour time-points and the RNA was purified for analysis. The RNA was reverse transcribed into cDNA and HIV RNA levels were analyzed by quantitative PCR. Preliminary data shows that Tat-SF1 knockdown affected the stability of HIV RNAs. Further exploration of this mechanism could be used toward future HIV drug therapies.

(Th.14) Construction and Alignment of a Total Internal Reflection Fluorescence Microscope

Kaitlyn Griffith, Emma Welter, Simeon Simeonides, and Keir Fogarty*

Department of Chemistry

High Point University

An instrument was constructed that was capable of achieving total internal reflection fluorescence (TIRF) microscopy. This type of microscopy only allows for fluorophores very near the surface of a coverslip to be seen due to its 100 nm excitation field depth. TIRF excels at observing surface interactions, like interactions that may occur at the cell membrane. The process of constructing the instrument began by coupling an optic fiber with the laser. Then through a series of mirrors and lenses, the beam size and direction was manipulated. Later in the light path the beam met up with a periscope, which is a tool that allows for the beam's height to be adjusted. After being raised to the appropriate height, the laser beam was focused through a lens into the back aperture of a 60x objective. Movable stages were situated in such a way that lenses and mirrors could be moved to alternate between TIRF and laser spectroscopy. TIRF requires the laser beam to enter at the back side-edge of the aperture, while spectroscopy requires the beam to travel straight through the center of the objective. Once TIRF was in place fluorescein and fluorescent nanospheres were used in proof of principal experiments that confirmed the functionality of the laser excitation field. Having the ability to use both methods of excitation will add to the versatility of the instrument.

(Fr.3) The EREBOS Project: Studying the Effects of Substellar Companions on Stellar Evolution

Ryan Hegedus and Brad Barlow*

Department of Physics
High Point University

HW Vir systems are binary systems containing a hot subdwarf star and a smaller companion red dwarf star. Previously, there were only 15 known HW-Vir binaries until light curves from the OGLE Survey revealed 36 new such systems. The smaller, cooler companion in each of these systems has survived engulfment by a nearby red giant star. Thus, analyzing the companion mass distribution of HW Vir systems could reveal whether brown dwarfs or even planets can survive engulfment by a red giant. The EREBOS Project was started to obtain follow-up observations of the new binaries discovered by OGLE. The ultimate goal of EREBOS is to find a lower mass limit for these systems. Using the Goodman Spectrograph (SOAR 4m telescope) and the SMARTS 0.9m telescope, we were able to collect time-series photometry and spectra for four of the EREBOS targets. We wrote photometry and spectral analysis code in Python to extract light curves and orbital velocities. The data were then modeled with the software Binary Maker in order to obtain all stellar parameters, most importantly the companion mass. Our initial results imply that brown dwarfs may be able to survive red giant engulfment.

(Th.19) Identifying a Mammalian Functional Substitute for Atg27

Ambar Khawaja¹, Thomas Moss² and Veronica Segarra^{2,*}

¹High Point Central High School
High Point, NC

²Department of Biology
High Point University

Eukaryotic cells undergo a process called autophagy when experiencing stressors like starvation. Autophagy helps cells survive and maintain homeostasis by allowing for the recycling of materials such as defective and unnecessary organelles and proteins. A yeast transmembrane protein called Atg27 functions in autophagy. Atg27 is composed of three domains; the transmembrane, the luminal domain, and the cytoplasmic tail. While the function of the luminal domain is unknown, the role of the cytoplasmic tail is to properly localize Atg27. In mammals, there are proteins with the overall same domain organization as Atg27 called Lysosome-Associated Membrane Proteins (LAMPs). To determine if LAMPs are the mammalian equivalent of Atg27, we generated mutant strains of baker's yeast containing the human LAMP1 gene in the ATG27 locus.

(Fr.4) Photoenzymatic Repair Capabilities of Diaphanosoma spp. in Response to UV Radiation

Thomas Kyllö, Harrison Seitz, Kayla West and Sandra Cooke*

Department of Biology
High Point University

The impact of fluctuating ultraviolet (UV) radiation on freshwater organisms as a result of climate change has a range of both direct and indirect consequences to freshwater ecosystems. UV-B has a damaging effect on freshwater zooplankton, which means the organisms must have certain ways of combating that damage in order to survive. One of these coping mechanisms could include the use of photoenzymatic repair (PER), which is a process that uses UV-A and visible light to repair the damage done by UV-B. The focus of this study was to determine the effects of UV radiation, including the use of PER, on *Diaphanosoma* spp., a widely distributed cladoceran species. The UV sensitivity and PER capabilities of *Diaphanosoma* spp. are relatively unknown, however based on similar species we hypothesized that *Diaphanosoma* spp. use PER, and the degree to which they use it varies with temperature. A *Diaphanosoma* spp. population from City Lake in North Carolina was subjected to UV-B and photo repair radiation (+PRR), and UV-B without photo repair radiation (-PRR), along with dark controls. Each trial was done using 4 replicate dishes, and performed at 15, 25, and 30° C in order to determine the ideal temperature for PER. The effect of PER was determined by monitoring survival and reproduction of the two treatments and control over 72 hours. It was determined that *Diaphanosoma* spp. had the highest PER capabilities at 15° C. Future research will include examining different temperatures, including 10 and 20° C.

(Th.19) Identifying Protein Cargo Recognized by Atg27

Deanna Lee and Veronica Segarra*

Department of Biology
High Point University

Transport vesicles allow organelle-to-organelle transport of protein molecules to their designated locations. During transport vesicle formation, sorting-signal containing protein cargo are selectively recognized by receptor proteins. These receptor proteins ultimately allow for the incorporation of specific cargo into the appropriate transport vesicle. Atg27 is a baker's yeast transmembrane protein that plays a role in autophagy, a process in which damaged or unneeded cellular components are recycled. We have identified Atg27 as a possible receptor for protein cargo. Using genetic and biochemical assays, we aim to identify the protein cargo recognized by Atg27.

(Th.6) Does Argon Gas Treatment Affect Cell Viability Following Stroke Model Injury?

Daniel Magurno, Alexandra Sprouse and Michael Grider*

Department of Biology
High Point University

Ischemic strokes are one of the most common forms of death and long-term injury, affecting fifteen million people worldwide every year. Among those affected, five million die, and another five million are permanently disabled. Yet, there are currently no approved therapeutic treatments to lessen the neural damage in those affected by strokes. Although Xenon gas has neuroprotective effects following a stroke-like injury, it is not a viable candidate for treatment of humans because it is prohibitively expensive and provides an undesired and unsafe anesthetic effect. Recently, similar neuroprotective effects were demonstrated with argon gas treatments. However, the mechanism(s) of argon-mediated neural protection is unknown. In this study, we use rat PC12 cells that are induced to neuronal phenotype by serum withdrawal and the addition of growth factors. We quantified survival of cells incubated with or without argon gas, under control conditions and under each of the following injury conditions: glucose deprived, oxygen deprived, or oxygen and glucose deprived. We are applying the findings of this study to further investigate the molecular signaling mechanisms of argon-mediated neuroprotection.

(Th.12) The Effect of Oxybenzone and Methylparaben on Bone Development and Breast Cancer Cell Growth

Sarah Mastropietro, Anastasiya Melnytska, Kristen Bowey* and Neil Coffield*

Department of Biology
High Point University

Estrogen is a sex hormone that controls many processes in the developing embryo including bone development. In addition to responding to endogenous estrogen, cells within an organism can also respond to a number of chemicals that are structurally similar to estrogen. Many of these chemicals can be found in commercially available personal care products. Two known estrogen mimics in particular, oxybenzone and methylparaben are commonly found as the active ingredients in sunscreens and cosmetics, respectively. To test the estrogenic activity of these compounds on early bone ossification, our lab utilized the zebrafish embryo model system. Dpf 0 (days post fertilization) embryos were immersed in oxybenzone at concentrations of 10⁻⁵, 10⁻⁶, or 10⁻⁷M or methylparaben at concentrations 10⁻⁴, 10⁻⁵, 10⁻⁶ M. The embryos were exposed to the treatments through dpf 14. Two fish were harvested per day starting at dpf 7 and continuing to dpf14. The fish were stained with the fluorescent calcium binding dye Calcein and imaged using a fluorescence dissecting microscope. The number of calcium positive vertebrae at each time point were blindly scored by at least two students.

(Fr.5) The Effects of Nicotine on Zebrafish Development

Mallory McKee and Kristin Ackerman*

Department of Biology
High Point University

Each year, over 1,000 babies in the U.S. die because their mothers smoked while pregnant (American Pregnancy Association, 2016). In addition to increased mortality, nicotine exposed fetuses are at risk of premature birth, SIDS, and low birth weight. Various animal models are used to examine nicotine-exposed embryos, but mice and rats yield few pups per birth, the procedures to nicotine-treat are complicated, and extraction methods to move the pups out-of-utero are extremely invasive. Thus, we are utilizing zebrafish (*Danio rerio*) as a model to study vertebrate development because of their high reproduction rates (hundreds of embryos per mating) and their transparent external development. External fertilization allows pharmacological studies to begin at the one-cell stage, while tissue transparency allows for examination of development in real time via light microscopy and relatively easy tracking of changes in gene expression within the whole embryo. Past studies exposed zebrafish embryos to nicotine between 6-24 hours past fertilization (hpf) with 10-30 μM nicotine. To more closely mimic the human experience, we exposed embryos at the 0-1 cell stage with a concentration range at 100 nM, 10 μM , 50 μM , or 100 μM nicotine and assayed for survival, growth, heart beat, and motor behavior. To ensure that nicotine penetrated the embryonic tissue, we used a Polymerase Chain Reaction (PCR)-based approach and observed an upregulation of the early immediate gene, cFos, which has been shown to be increased in the nervous system of other nicotine-exposed animals. Preliminary data indicates that nicotine effects behavior and overall zebrafish development.

(Th.15) Computer Vision Techniques for Obstacle Detection and Tracking in Autonomous Vehicles

Max Maurer and Martin DeWitt*

Department of Physics
High Point University

Cars with autopilot, unmanned aerial vehicles, and other autonomous vehicles are becoming increasingly popular. A serious concern with autonomous vehicles is how to insure they are capable of going where they are intended while avoiding unpredictable obstacles that could cause major disasters. Our goal was to turn an ordinary remote control car into a self-driving autonomous vehicle using various computer vision analysis methods. This was achieved by installing a raspberry pi microcomputer equipped with a small USB camera onto our remote control car. The computer was given full control of the cars driving functionality. The package Open Computer Vision in the Python programming language was utilized to aid us in creating code that could analyze the scene in front of the car. Two techniques from this package were utilized. The first filtered the colors in the image with the use of a color density histogram taken from the colors of desired object. The second technique, camshift tracking, located a desired object from the filtered image and followed its location from frame to frame. The method can even be adapted to detect multiple targets simultaneously. With this information the car was able to respond as desired to certain events. With this functionality we were able to implement code that allowed our car to complete tasks such as follow a yellow kickball, and drive along a brick pathway while controlling both the steering and speed of the vehicle completely unassisted.

(Th.17) Quorum Sensing and Programmed Cell Death in *Escherichia coli* and *Bacillus subtilis*

Lisa Nguyen, Rebecca Ulrich and Meghan Blackledge*

Department of Chemistry
High Point University

Bacteria can communicate intra-species and interspecies by the use of quorum sensing (QS) signaling molecules. Both *Escherichia coli* and *Bacillus subtilis* contain a QS molecule called the extracellular death factor (*EcEDF* and *BsEDF*, respectively). *EcEDF* and *BsEDF*, acting as interspecies QS molecules, can lead to programmed cell death (PCD) in dense cultures of both bacteria during times of stress. The amino acid sequences of the QS molecules and the amino acid residues important for activity have been elucidated. Specifically, the asparagine residue in the *BsEDF* hexa-peptide RGQQNE has been found to be required for activity. However, the structure-activity relationship of *BsEDF* is not yet fully known. For this project, *BsEDF* analogues were made by manual solid-phase peptide synthesis and were purified using high-pressure liquid chromatography (HPLC). The analogues will be used in biological assays of *E. coli* to learn more about how the structure of *BsEDF* affects cell death. Details concerning the synthesis and purification of the *BsEDF* peptides, as well as preliminary data of the biological assays, will be presented.

(Fr.6) Evaluating the Efficacy of the Anti-Tumor Agent Beta-lapachone in NQO1 Expressing *BRCA2* Mutant Human Breast Cancer Cells

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Currently, there are few specialized treatments targeting breast cancer cells with *BRCA1/2* mutations, that cause little to no harm to normal tissues. Beta-Lapachone (β -Lap) is an antitumor quinone that is selectively bioactivated in the presence of NAD(P)H:quinone oxidoreductase 1 (NQO1). NQO1 is overexpressed in solid tumors compared to normal tissues. In the presence of NQO1, β -lap undergoes a two-electron reduction forming a hydroquinone, which can be further reduced to a semiquinone intermediate. The semiquinone reverts back to the parent quinone resulting in futile redox cycling and production of reactive oxygen species (ROS). ROS-induced DNA damage hyperactivates the DNA repair protein poly (ADP-ribose) polymerase-1 (PARP-1) leading to cell death. Of interest, cancers with *BRCA1/2* mutations are unable to efficiently repair double-stranded DNA breaks. As a result, we hypothesize that *BRCA1/2* mutant, NQO1 positive cells will be highly sensitive to β -lap, which will result in irreparable DNA damage resulting in cell death. To test this hypothesis, the breast cancer cell line HCC1428, a *BRCA2* mutant, was used. These cells were transfected with wild type *BRCA2* to compare *BRCA2* status vs sensitivity to β -lap. Cellular survival experiments were used on these cells to determine the toxicity of β -lap and IB-DNQ (a β -lap derivative) at different doses over various periods of time. Our research provides information on the pathway of β -lap-induced cell death in NQO1 expressing, *BRCA2* mutant breast cancer cells.

(Th.13) Photoenzymatic Repair Capability in the Freshwater Cladoceran Genus *Scapholeberis*

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Recent decreases in stratospheric ozone have allowed more ultraviolet radiation to penetrate the atmosphere than in years past. Ultraviolet-B (UV-B) radiation contains enough energy to damage an organism's cell's DNA, which may lead to cell death or skin cancer. However, some species of freshwater zooplankton can use ultraviolet-A (UV-A) radiation for a process called photoenzymatic repair (PER) to undo damage done by UV-B radiation. UV-B radiation causes cyclobutane pyrimidine dimer lesions in the DNA, but photoenzymatic repair occurs when UV-A radiation activates a photoreactivating enzyme, photolyase, which creates a cyclobutane bridge to heal the lesion. The cladoceran zooplankton genus *Scapholeberis* has been well-studied in its usage of protective pigments to shield itself from UV-B radiation, but its use of PER is not well-documented. Our experiments studied the UV sensitivity and PER capabilities of *Scapholeberis* to survive UV-B radiation, and subsequently recuperate using UV-A radiation. We set up three different treatments of *Scapholeberis*. Two received 15 minutes of exposure from a 312 nm UV-B lamp, followed by either 24 hours of exposure from a 365 nm UV-A lamp (+PRR), or no subsequent UV exposure (-PRR). These two then joined the final treatment (Control) in a darkly covered box, absent of any UV exposure. Survival was measured every 24 hours for 72 hours. We ran trials at 15°C, 25°C, and 30°C to determine the effect of temperature on survival rates. *Scapholeberis* mortality was highest at 30°C, while specimens exhibited the greatest PER rates at 15°C.

(Th.4) Constructing and Testing a TIRF-FCS Microscope

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This project focuses on the construction of a Total Internal Reflection Fluorescence Correlation Spectroscopy (TIRF-FCS) apparatus. This apparatus uses a high-power laser to create a very small excitation field that will create bursts of light as fluorescent-labelled molecules travel through it. In addition to the process of aligning the laser using mirrors and lenses, a Single Photon Counting Module (SPCM) was aligned to detect output fluorescence. A LabVIEW program was written to collect data from the SPCM using a Data Acquisition (DAQ) card, interface the DAQ to a PC to adjust parameters such as sampling rate and number of samples collected, and display and save the data to the PC. FCS uses a statistical technique called autocorrelation to analyze the bursts of light collected by the DAQ into data like molecule mobility and concentration. A Python program was written to display the photon count data and perform a multiple-tau autocorrelation, then save the autocorrelated data. This data was then fitted with an autocorrelation curve using a program called PyCorrFit to determine parameters such as average particle brightness and time of diffusion.

(Th.16) Relative Contributions of Apoptosis and Necrosis in *In-vitro* Models of Stroke

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Ischemic stroke, the blockage of blood and oxygen flow to the brain, is the fifth leading cause of death in America, killing one American every four minutes. There are currently no therapeutic treatments specifically aimed at promoting survival of neurons following ischemia. We selected an in-vitro model of ischemic stroke that allowed more control of experimental conditions. We culture neuron-like PC12 cells under oxygen-glucose deprivation (OGD) conditions to model a lack of blood flow to neurons following a stroke. Additionally, in order to better elucidate the relative contribution of oxygen deprivation OR glucose deprivation to neural injury, we include injury groups deprived of only oxygen or only glucose. We seek to identify the specific pathways through which these injuries damage the cells. For example, cell death in response to glucose deprivation has traditionally been demonstrated to occur by apoptosis, programmed cell death. However, our data suggests that, compared to controls, glucose deprivation for 24 hours also significantly increases necrosis, the unregulated destruction of cell components. We use flow cytometry to quantify the relative participation of necrosis or apoptosis in response to each of the previously listed injuries. Through this technology, we aim to develop therapeutic treatments directed towards the specific apoptotic and necrotic pathways, as appropriate to each aspect of an ischemic stroke. Specifically, we have initiated studies to examine the potential for argon gas to attenuate cell death in response to these conditions.

(Fr.2) Towards Further Understanding of Kinase Activity During Oxidative Stress: Synthesis of the Highly Active ERK2 Substrate Sub-D

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Extracellular signal-regulated kinase 2 (ERK2) is a member of the mitogen-activated protein kinase family and is involved in the regulation of cell development, growth, and differentiation. The current project focus is to further study ERK2 activity in response to cellular oxidative stress. ERK2 has two ligand recruitment sites, the D-recruitment site (DRS) and the F-recruitment site (FRS), to which peptide and protein substrates bind and become phosphorylated. In a previous computational study of small molecule binding interactions, two novel peptides, Sub-D and Sub-F, were modeled in binding to the DRS and FRS within ERK2, respectively. The *N*-terminus of Sub-D consists of the highly polar amino acid sequence FQRKTLQRRNLKGLNLNL to facilitate DRS docking. A flexible hydrophobic linker, made up of three 6-aminohexanoic acids, connects this polar docking sequence to the consensus, serine-containing phosphorylation site on the *C*-terminus, TGPLSPGPF. Using a semi-automated, flow chemistry approach, peptide Sub-D was successfully synthesized using Fmoc-based solid-phase peptide synthesis. The identity and purity of Sub-D was confirmed through HPLC purification and mass spectrometry data. In collaboration with the Poole Lab at Wake Forest School of Medicine, initial data indicates synthetic Sub-D is a highly active ligand for ERK2.

(Th.2) The Disappearing Pulsations of the Hot Subdwarf Star CS 1246

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Hot subdwarf stars were once main sequence stars, like the sun, that deviated from normal stellar evolution due to binary interactions and evolved into extreme horizontal branch stars. Several of these stars exhibit rapid pulsations driven by iron opacity instabilities. CS 1246 is a rapidly pulsating hot subdwarf discovered in 2009 that is dominated by a single 371 second pulsation. At the time of its discovery, the pulsational amplitude was one of the largest known, making CS 1246 an ideal candidate for follow up studies. We present six years of photometry and two nights of spectroscopy that confirm the decay in amplitude of CS 1246. After fitting multiple models to the amplitude change, our data shows consistency with an exponential decay that is reminiscent of a damped harmonic oscillator. We know CS 1246 pulsates through the kappa-mechanism whereby an iron layer in its atmosphere periodically heats up and cools down causing a changing opacity which drives the pulsations. If this hot subdwarf is behaving like a damped harmonic oscillator then we predict the iron is no longer present in the right position to drive these pulsations. However, if the iron layer were to radiate back into the optimal region, we may see a return in these once strong pulsations.

(Fr.1) Exploring the Role of Tat-SF1 as an HIV-1 Host Factor

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Tat specific factor 1 (Tat-SF1) is a human protein that functions as a transcription and splicing factor. This protein has also been identified as an HIV host factor and is being investigated for its hypothesized role in viral gene splicing. Tat-SF1 has been found to alter the levels of HIV RNAs, though the exact mechanism by which this happens is unknown. It is possible, however, that Tat-SF1 is involved in HIV alternative splicing, RNA stability, or export. If associated with any of these processes, then an interaction with HIV RNA would be expected. To test this hypothesis, RNA immunoprecipitation (RIP) was utilized. The RNA molecules that Tat-SF1 interacts with were then isolated for further analysis. Total RNA was reverse-transcribed into its respective cDNA, which could later be amplified through quantitative polymerase chain reaction (RT-qPCR). Data from these experiments will reveal if Tat-SF1 interacts with the viral RNA, and if so, at what location. By exploring the specific Tat-SF1 binding sites on the HIV genome, more insight can be gained into the complete role of this protein for the virus.

(Th.18) Effects of the Estrogen Mimicking Neutraceutical, Resveratrol on Bone Development

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Our research this summer aims to explore and define the effects of an estrogen activating neutraceutical compound on the rate of embryonic bone ossification. The zebrafish (*Danio rerio*) was chosen due to its size, cost, translucency and its ability to respond to exogenous human estrogen. Zebrafish embryos were exposed to increasing concentrations of resveratrol, a neutraceutical commonly found in the red wine, which is a known estrogen activator. Embryos were collected, separated into individual petri dishes, treated with the compound and monitored. The numbers of hatching fish, numbers of surviving fish, along with any gross anatomical changes were documented on each day of the fourteen day experiment. Calcein dye, a calcium binding compound, was used to monitor vertebrae ossification in the living embryos. The rate of ossification was measured by counting the number of visible vertebrae at each day during the experiment.

(Th.11) Utilization of Molecular Dynamics to Examine the Physical Properties of Different Hydrocarbons for Alternative Fuels

Jonah Winkler and Todd Knippenberg*

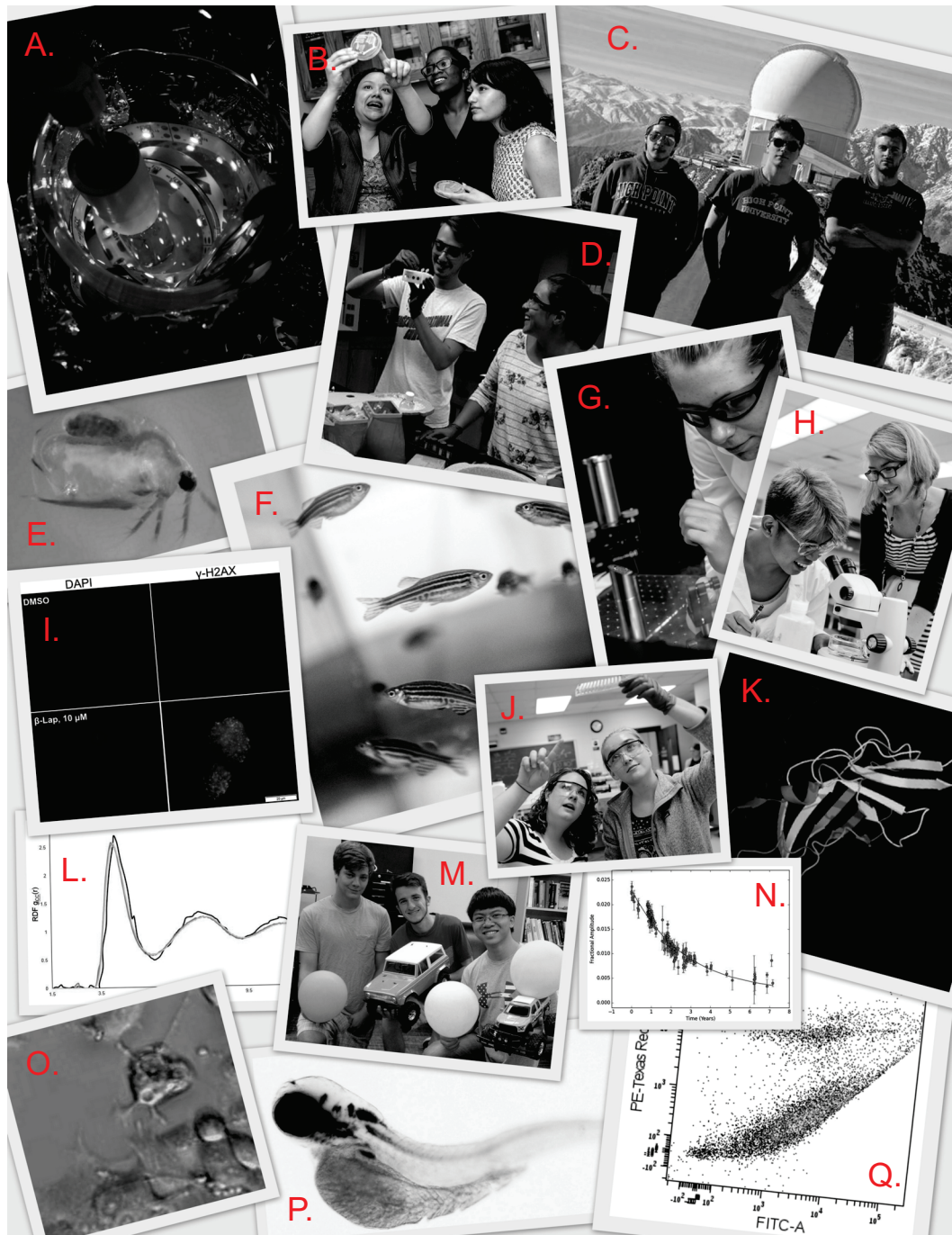
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The discovery of new alternative fuels has become a growing interest in recent years due to massive fuel consumption. The main objective of this project begins comparing known experimental values of physical properties, such as density, to results from computer simulations. If these results are acceptable, then it might be possible to explore fuels that have not been tested experimentally yet. Molecular Dynamic simulations are used in this study based on the AIREBO (adaptive intermolecular reactive empirical bond order) potential, which is used to calculate the inter- and intra-molecular interactions between the atoms in a system. The AIREBO potential was used to simulate the hydrocarbon compounds known as tetralin, decalin, and hexadecane in hopes to accurately predict their densities at temperatures ranging from 293.15K to 373.15K. Multiple trials for each system at each different temperature were simulated using molecular dynamics and it was found that the results were in good agreement with the experimental densities of each molecule. The radial distribution function (RDF), which is used for determining how the atoms in a system are packed in the liquid phase describes \was calculated for methane, benzene, and ethane. These results from the RDF also showed good agreement between the simulation and corresponding experimental values. In addition to the utilization of the AIREBO potential, bomb calorimetry was used to calculate combustion enthalpies of octane and hexadecane, which resulted in close relation to experimental values.

MORE SCENES FROM SuRPS 2016





(A.) Blue LED initiated-reaction in Wommack lab; (B.) Segarra lab; (C.) Barlow lab in Chile observatory; (D.) Miller lab; (E.) *Scapheloberis spp.* from Cooke lab; (F.) *Danio rerio* fish from Coffield lab; (G.) Laser alignment in Fogarty lab; (H.) Ackerman lab; (I.) Fluorescent microscope images from Srougi Lab; (J.) Blackledge lab; (K.) Protein model from Segarra lab; (L.) Molecular dynamics model from Knippenberg lab; (M.) DeWitt lab; (N.) Star data from Barlow lab; (O.) Microscopy from Grider lab; (P.) *In-situ* RNA study from Ackerman lab; (Q.) Fluorescence data from Grider lab.

