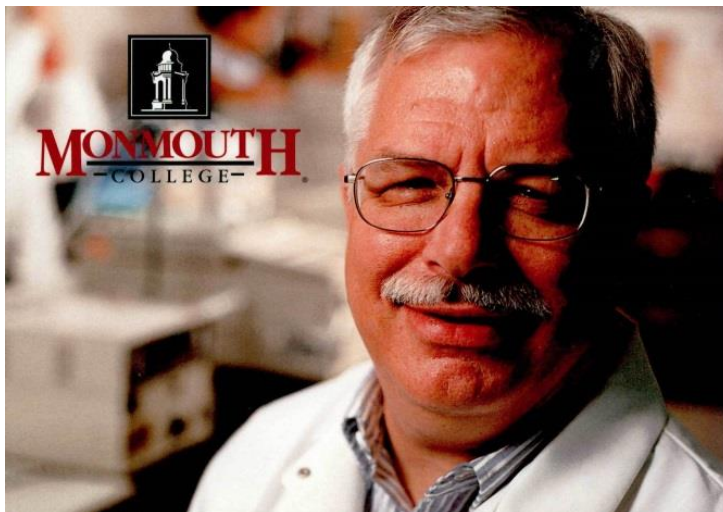


# *Doc Kieft Summer 2019 Undergraduate Research Talks*

July 19, 2019

CSB 100, Monmouth College

starting at 8:45am



**Sam Argubright (MC '20)** – Biochemistry Major – Dr. Brad Sturgeon Lab

**Victoria Burgo (MC '21)** – Biochemistry Major – Dr. Laura Moore Lab

**Alex Cutright (MC '21)** – Biochemistry Major – Dr. Audra Goach Lab

**Logan Evans (MC '21)** – Biochemistry Major – Dr. Michael Prinsell Lab

**Andrew Ferris (MC '20)** – Biochemistry Major – Dr. Brad Sturgeon Lab

**Will Fox (MC '21)** – Biochemistry Major – Dr. Brad Sturgeon Lab

**Tyler Halsey (MC '21)** – Chemistry Major – Dr. Laura Moore Lab

**Trevor Jones (MC '21)** – Biochemistry and Biopsychology Major – Dr. Audra Goach Lab

**Kyle McLaughlin (MC '21)** – Biochemistry Major – Dr. Audra Goach Lab \*talking in the fall at science seminar

**Emily Rein (MC '21)** – Biochemistry Major – Dr. Laura Moore Lab

**Joe Shie (MC '21)** – Biochemistry Major – Dr. Michael Prinsell Lab

**Josie Welker (MC '21)** – Chemistry Major – Dr. Brad Sturgeon Lab

## Schedule of Events

**8:45am-9:00am – Talk Setup – Refreshments**

**9:05am-11:05am – Morning Session of Talks \*talks are 15 minutes with 5 minutes for questions**

9:05am-9:25am – **Logan Evans**

*“Isolation of Alpha and Beta Acid Standards from a Hops Extract”*

*Humulus lupulus* (hops) are a main component in the beer brewing process, being responsible for the bitter flavor, and have many properties that may be useful in pharmaceutical development, such as antibacterial properties and working as inhibitors for cancer causing genes. Hops contain three alpha acids (humulone, cohumulone, and adhumulone) and three beta acids (lupulone, colupulone, and adlupulone). Currently, there are no commercial standards for the individual acids, only a combined standard (ICE-4), or standards of the alpha acids or the beta acids. Through separation by Flash Chromatography, standards could be isolated for the individual acids, benefiting large industrial productions and research into the properties of the individual acids. The purity of the Flash separation can be determined with HPLC and NMR. As of now, the Flash and HPLC chromatographs gathered are in line with previous literature and work on increasing purity is in progress.

9:25am-9:45am – **Alex Cutright**

*“Surface Molecular Interactions and Thermodynamics of Cannabidiol and Phospholipids at the Air-Water Interface via Langmuir Monolayers”*

The lipid bilayer of the cellular membrane is a selectively permeable system composed of various phospholipids, that regulates what can enter and leave a cell. With the new surge of users of this compound comes research that investigates its metabolic pathways and absorption rates. Cannabidiol was analyzed to determine its interactions with phospholipids that play key roles in the bilayer. A substantial difference in phospholipid interactions was shown to occur when cannabidiol was introduced into the system. These findings help for people who use this substance to understand how their bodies intake and process this compound and how it interacts with their body.

9:45am-10:05am – **Andrew Ferris**

*“Perfection of Free Radical isolation techniques and analysis via Electron Paramagnetic Resonance (EPR)”*

Free radicals play a vital role in the human body, acting as intermediates and byproducts of various metabolic and enzymatic reactions necessary for life. The overabundant presence of radicals has also been linked to a plethora of human illness spanning from Alzheimer’s disease to Cancer; and possibly even to the process of aging itself. However, the specific mechanisms and behaviors of these free radicals are poorly understood. In order to understand the interactions between free radicals and the complex molecules within the human body, their behavior with simpler molecules such as polycyclic aromatic hydrocarbons must first be evaluated. Initially, the radicals must be properly isolated from potential contaminants, and analyzed via EPR Spectroscopy. The behavior of free radicals on such molecules has previously been studied, and their results will be tested against those derived from the techniques utilized in this experiment. Once the technique’s validity has been confirmed via data comparison, it can then be applied to more complex molecules.

10:05am-10:25am – **Trevor Jones**

*“Analysis of Flavonoids in American Propolis”*

Propolis is one of the many products extracted by honeybees, and has gained popularity recently due to publications of its health benefits. It has been shown to be anti-oxidative, anti-carcinogenic, and anti-inflammatory to name a few. The main focus of past research has been on Brazilian propolis while propolis products from other countries have been neglected. This research investigates American propolis with Gas Chromatography-Mass Spectrometry to determine the phenolic content and to identify the major flavonoids. Future work for the project includes further purification of the propolis extract using column chromatography, and analysis with a high resolution Liquid Chromatography- Tandem Mass Spectrometer housed at Western Illinois University.

10:25am-10:45am – **Emily Rein**  
“*Purification of Ovoperoxidase*”

Peroxidases are enzymes that catalyze oxygenation with hydrogen peroxide. There are many types of peroxidases that carry out specific functions in the organisms they reside in. Ovoperoxidase is a peroxidase that is secreted from the purple sea urchin (*S. purpuratus*) eggs at time of fertilization. This enzyme is responsible for hardening the fertilization membrane that will prevent polyspermy, or multiple fertilizations to a single egg. The goal of my research project is to purify this protein, ovoperoxidase. To obtain this goal of purification, the eggs are to be lysed by means of an acetic acid wash as well as an ammonium sulfate precipitation followed by column chromatography for further purification. When purification is achieved, I hope to compare ovoperoxidase to other peroxidases such as horseradish peroxidase to determine if others are able to carry out various functions more efficiently.

10:45am-11:05am – **Will Fox and Josie Welker**  
“*Enzymatic Oxidation of Biophenols*”

Acetaminophen is an active ingredient in many over-the-counter and prescription painkillers, such as Tylenol and Percocet. Evidence supports that the mechanism for acetaminophen-induced liver injury involves a one electron oxidation. 4-Hydroxyphenylacetic acid (HPA) is a promising new hepatoprotective drug which can prevent this type of liver damage. The oxidation of these two substrates can be accomplished by using horseradish peroxidase (HRP) and hydrogen peroxide. Horseradish peroxidase catalyzes the reaction of the decomposition of hydrogen peroxide into water and oxidation products in the presence of a substrate, which increases the decomposition rate. The High Pressure Liquid Chromatography (HPLC) data shows that there are four main products formed from the reaction with the substrate, HRP, and hydrogen peroxide. When this reaction is run with 4-hydroxyphenylacetic acid (HPA) or Acetaminophen, it was found that increasing the concentration of hydrogen peroxide resulted in less starting material and more oxidation products. The goal is to determine the identities of the four oxidation products. Biochemical kinetic simulation programs can help simulate the data and elucidate the identity of the four oxidation products by determining the rates of the reactions.

**11:10am-12:10pm – Lunch in the Nutrition Lab**

**12:15pm – Group Picture in front of CSB**

**12:30pm-2:10pm – Afternoon Session of Talks**

12:30pm-12:50pm – **Victoria Burgo**  
“*Acetic Acid Tolerance of Escherichia coli*”

Sustainable production of biofuels will require efficient utilization of lignocellulosic biomass. Acid stress, however, inhibits efficiency due to the accumulation of acidic metabolites during the pretreatment and fermentation processes. Organic acids, such as acetic acid, are among the most concentrated toxins released during pretreatment. Acetic acid is a weak organic acid that exerts toxic effects to most microorganisms by decreasing their intracellular pH and metabolic disturbances by the acetate anion. Although the addition of a base to the medium could relieve acid stress, it would be more efficient and cost effective to design acid tolerant strains of bacteria. The goal of this project was to isolate genes from metagenomic libraries that will promote survival of *E. coli* under acid stress. This was done by transforming the libraries into *E. coli* and performing directed evolution through acid challenging.

12:50pm -1:10pm – **Joe Shie**  
“*Development of a N-N Bond Forming Reaction*”

Natural molecules containing nitrogen-nitrogen (N-N) bonds have exhibited promising biological activity. The most common method of synthesizing these molecules is linear, sequentially derivatizing a protected hydrazine by adding functional groups. This method is not efficient due to the number of reaction steps needed to be performed to obtain the desired N-N bond containing product. The proposed method involves synthesizing an electrophilic nitrogen compound and reacting it with a deprotonated nucleophilic nitrogen compound via an  $S_N2$  reaction to form the desired N-N bond. This method forms the N-N bond in the last step and is more convergent, reducing the number of linear steps, resulting in a better overall theoretical yield than the current linear methods.

1:10pm-1:30pm – **Tyler Halsey**

*“Investigating the Effect of Multiple Interactions on the Dimerization of the Fumarate Nitrate Reductase Transcription Factor in E. coli Bacteria”*

Facultative anaerobes, like E. coli, contain FNR, a protein that controls gene expression for anaerobic respiration. In the absence of oxygen FNR forms a dimer containing an iron-sulfur cluster, and is then able to regulate gene expression. In the presence of oxygen the iron-sulfur cluster degrades and causes the dimer to unzip. E. coli FNR’s crystal structure has not been isolated, so A. fischeri FNR is used as a reference structure to assist in interpreting the impacts of amino acid interactions on the stability of FNR. Our investigation focused on the 130 and 140 position and the speculated salt bridge formed between these positions across the dimer. Additional studies are going to focus on the 142 position.

1:30pm-1:50pm – **Sam Argubright**

*“3-D Printing of EPR Flow Cells”*

Electron Paramagnetic Resonance (EPR) is a spectroscopy technique that detects unpaired electrons. When these unpaired electrons exist with a molecular structure these are referred to as radicals. Radicals are highly reactive with very short life spans. The highly reactive nature of the radicals makes it difficult to detect using EPR. The necessity to mix reactions solutions quickly and efficiently has motivated us to design a flow cell that takes advantage of fluid dynamics. By designing my own flow cells and can have a greatly reduce cost for lab equipment and have better product generation using flow cells.

1:50pm-2:10pm – **Alex Cutright and Trevor Jones**

*“Advancements of Open-Source Microscopy through the construction of a fluorescence microscope and updates to the Brewster Angle Microscope”*

Brewster Angle Microscopy is a useful tool for the analysis of surface characteristics of films at the air-water interface in a Langmuir trough. A Brewster Angle Microscope has the capability to obtain images of the film while isothermal compression data is collected, making it an ideal instrument for the research of Langmuir monolayer films. The goal of this research project was to construct a BAM stage using metal, replacing the previous LEGO Mindstorms pieces, as well as to design and 3D print objective and laser mounts. This ensures the structural integrity and longevity of the microscope for years to come. Future research includes upgrading various critical parts of this microscope such as the stepper motors and optics, which will provide more accurate and clear images. Another project in the Goach lab is a fluorescence microscope, which will be used to image lipids on the Langmuir monolayer trough. This is done by fluorescing fluorophores that attach to the lipid’s head group and emitting light in accordance to a set wavelength. The goal for this microscope is to utilize multiple microscopy techniques in order to understand how a lipid’s ability to compress is changed with added molecules, such as CBD oil or flavonoids. Further research includes creating a fluorescence library within the microscope and setting up the optics required.