Doc Kieft Summer 2018 Research Program

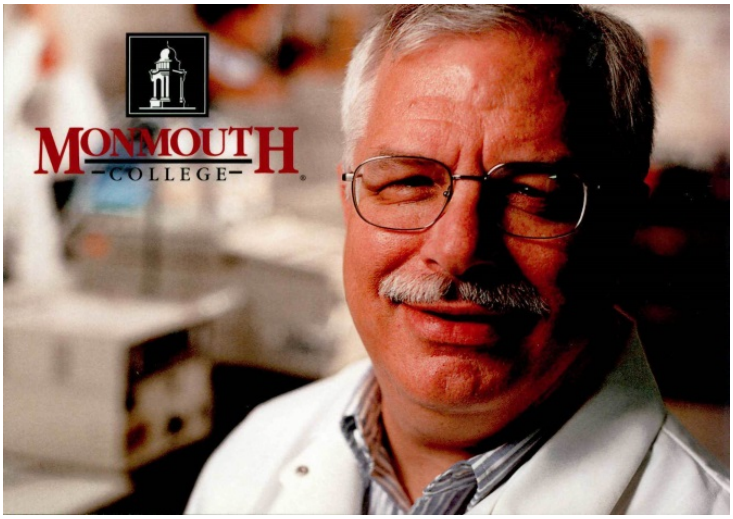
Undergraduate Research Talks

July 20, 2018

Pattee Auditorium

Monmouth College

8:30am-1:00pm



Brandon Allen (MC ’19) –Chemistry Major – Sostarecz Lab

AJ Angellotti (MC ’20) – Biochemistry Major – Prinsell Lab

Seth Croslow (MC ’21) – Chemistry Major – Sostarecz Lab

Jennista Eastman (MC ’19) – Biochemistry Major – Moore Lab

Tyler Halsey (MC ’21) – Chemistry Major – Moore Lab

Modou Lamin Jarju (MC ’20) – Biochemistry Major – Sostarecz Lab

Jake McCrery (MC ’20) – Biochemistry Major – Prinsell Lab

Katie Richter (MC ’21) – Biochemistry Major – Sturgeon/Moore Lab

Julia Sterr (MC ’21) – Biochemistry Major – Moore Lab

Zelinda Taylor (MC ’20) – Biochemistry Major – Sturgeon Lab

Lexi Tibbs (MC ’19) – Biochemistry Major – Sturgeon/Prinsell Lab

Schedule of Events

8:30am-9:00am – Talk Setup - Refreshments and Opening Remarks (8:45am)

9:00am-1:00pm – Session of Talks

9:00am-9:20am – **Tyler Halsey**
Method Development for Analysis of Volatile Molecules in Bread Aroma through GC/MS

Bread aroma has been studied by analyzing volatile compounds after baking. Bread studied in this research was made with a simple recipe. Multiple methods of sampling and extraction were tested, emphasis in study was placed on Soxhlet extraction and steam distillation. Effectiveness of methods was evaluated through Gas Chromatography/Mass Spectrometry. Preparation of standards allows for the study of each method's ability to extract organic compounds theorized to be present in bread aroma. Soxhlet extraction was evaluated to be the most practical and effective of methods tested. Using this method vanillin and eugonol were identified in Sa-04 yeast.

9:20am-9:45am –**Brandon Allen**
A Brewster Angle Microscope for the Visualization of Langmuir Monolayer Films

Brewster Angle Microscopy is a useful technique for the analysis of surface characteristics of films at the air-water interface in a Langmuir trough due to its ability to obtain images while simultaneously collecting isothermal data. The goal of this research was to construct a relatively low-cost Brewster Angle Microscope (BAM) using LEGO™ Mindstorms, a laser, a 10x microscope objective, and a CCD camera. This BAM was then used to obtain images of dipalmitoylphosphatidylcholine (DPPC) films and DPPC/Cholesterol films during compression analyses. Software to control the microscope and automate image processing was developed using LabVIEW and ImageJ. The basics of Langmuir monolayer films and the principles of Brewster Angle Microscopy will be discussed as well as some of the basic software that was developed over the course of this research project.

9:45am-10:05am – **Seth Croslow**
The Aggregation of Insulin at the Air-Water Interface

Diabetes is a disease that occurs when the pancreas produces little to no insulin and affects more than 370 million people in the world. Though widely studied, insulin's aggregation mechanisms are not fully understood. It is known that insulin aggregates to form inactive hexamers inside the body in the presence of Zn^{2+} ions in the pancreas. Using a *Kibron Langmuir Trough*, divalent metal cations, such as Mn^{2+} and Cu^{2+}, were added to the trough's subphase and analyzed at the air-water interface to determine if the insulin hexamer would be induced. Since diabetic patients inject insulin subcutaneously, insulin's interactions with common lipids found in the cell membrane such as dipalmitoylphosphatidylcholine (DPPC) were also studied since interactions could render the insulin less effective. Images of insulin monolayers were also taken using a Brewster Angle Microscope. It was found that divalent metal cations other than Zn^{2+} also induce insulin hexamers and that there are only minor interactions with common cell membrane lipids.

10:05am-10:35am – **Zelinda Taylor and Alexandria Tibbs**
Synthesis and Oxidation of Lignin Monomers using Electron Paramagnetic Resonance Spectroscopy

Lignin and lignan synthesis occurs via oxidative coupling of three basic monomers: p-coumaryl alcohol, coniferyl alcohol, and sinapyl alcohol. The alcohol-forms of these monomers occurs naturally, but the acid-forms: p-coumaric acid, ferulic acid, and sinapic acid, are commercially available. In order to study these oxidative coupling reactions, the synthesis of the alcohol monomers from the acid monomers has been investigated with products characterized by high performance liquid chromatography (HPLC) and nuclear magnetic resonance spectroscopy (NMR). The oxidative coupling reaction is a result of the formation of a radical intermediate. To detect these short-lived radicals, the lignin monomers were oxidized by an enzymatic system containing horseradish peroxidase (HRP) and H_2O_2. Products were evaluated using HPLC and radical intermediates were detected using immobilized enzyme-ESR spectroscopy (IE-ESR). Current results will be presented.

10:35am-10:55am –**Katie Richter**
Exploring the Effects of Lignin Monomers and Their Oxidation Products on Gut Microbiota

The microbiome is comprised of the estimated 100 trillion bacteria that inhabit each human body. These microorganisms, gut bacteria specifically, play a large role in determining health and cognitive function. Lignans, dimers made up of two lignin monomers, are a class of natural products with a vast expanse of important biological activities that differ based on their structure. By growing various types of gut bacteria in the presence of lignin monomers and their oxidation products, a substance could be discovered that will act as a prebiotic to gut bacteria.

Break 11:00am – 11:15am

11:15am-11:45am –**Jake McCrery and AJ Angellotti**
Formation of N-N Bonds as a Tool for Synthesis and Drug Development

Molecules containing a nitrogen-nitrogren bond are extremely uncommon naturally occurring compounds, several of which have demonstrated biological activity. The current method of synthesizing nitrogen-nitrogen bonds is accomplished by using hydrazine or a hydrazine derivative as the starting material and adding side chains one at a time. This route can result in low overall yields due to the linear nature of the synthesis and contains starting materials that are not suitable for use in an undergraduate laboratory. A new method of synthesizing nitrogen-nitrogen bonds using a nucleophilic nitrogen and an electrophilic nitrogen with an attached leaving group is being developed. This method would utilize a bimolecular substitution reaction in the key step, which would result in higher overall yields due to the convergent synthesis and would avoid the use of dangerous starting materials. Once achieved, this method could be used to test the biological activity of known compounds and unnatural derivatives.

11:45am-12:05pm- **Jennista Eastman**
Investigating the Oxygen Stability of the L28 Mutations in the FNR Transcription Factor

Fumarate Nitrate Reductase (FNR) is the protein that controls the switch from aerobic to anaerobic respiration in *Escherichia coli* bacteria. In the presence of oxygen, the oxygen-sensitive [4Fe-4S] cluster is degraded spontaneously to a [2Fe-2S] cluster and forms a monomer, making the protein inactive. In the absence of oxygen, FNR forms a dimer, binds to DNA and expresses genes needed for anaerobic respiration. Previous studies suggest that a hydrogen bond between the L28H variant and the Q183 residue forms a certain conformation that prevents the entrance of oxygen, prohibiting the degradation of the iron-sulfur cluster and resulting in a protein that is active under aerobic conditions. The activity of FNR under aerobic conditions was measured after switching the Glutamine at the 183 position to an Alanine. This mutation had about the same activity as the L28H mutation, suggesting that the hydrogen bond between these residues is not the cause of the cluster stability

12:05pm -12:30pm – **Modou Lamin Jaraju**
A closer look at the Lipid-Protein interactions of a myelin sheath model membrane

The myelin sheath membrane is an integral component of the central nervous system that is wrapped tightly around the axons segments of nerve cells. It functions as an insulator that speeds up the impulses through the nerve fibers. The membrane is comprised of phospholipids, cholesterol and proteins. The proteins and phospholipids strongly interact via a conformation change. In this study, the Langmuir Monolayer technique is utilized to investigate the interactions that transpire between Myelin Basic Protein and Langmuir films of anionic and neutral phospholipids. The lipids used were a Brain Lipid Extract, which was composed of various anionic lipids and a Brain Sphingomyelin. The Langmuir Monolayer technique was used as an experimental model of the lipid membrane, and through the observation of the changes in surface pressure and molecular area, the degree of order or fluidity was inferred. Experiments were carried out using a Langmuir trough; the lipids were deposited onto water and Phosphate Suffered Saline sub-phases. A comparison of the results indicated that monolayers were more fluid on the PBS sub-phase than they were on water. Ultimately, from the ideal mixing of the lipids and protein, it was observed that the monolayer was more ordered and fluid when the MBP was added along to the lipids. This may be indicative of the MBP ability to strongly interact with the lipid membrane.

12:30pm-12:50pm – **Julia Sterr**
Isolating Genes from a Metagenomic Library to Increase the Toxin Tolerance of E. coli to Produce Biofuels More Efficiently

Biofuels are a clean, renewable energy source that is produced through fermentation of pretreated lignocellulosic biomass. Yeast is the organism most commonly used in the fermentation process; however, *Escherichia coli* is a more flexible

organism and is able to use 5-carbon sugars to produce biofuels. Even so, *E. coli* is not a great catalyst, and this makes biofuel production more costly and less effective. Biofuels are produced after lignocellulose is pretreated and fed to organisms, yielding ethanol and byproducts. Some byproducts from pretreatment and fermentation are toxic to *E. coli*; therefore, increasing the tolerance of these toxins could allow the bacteria to perform better as a catalyst and increase ethanol yield and cost effectiveness. In this summer research project, a metagenomic library, was inserted into *E. coli*. The library, Azure, contains genes from organisms living in extreme environments. Extremophiles have genes that are not found in more "normal" organisms and may allow them to better tolerate toxins. The *E. coli*, containing Azure, was challenged against the toxins vanillin and ferulic acid. Through this, a gene was discovered in the metagenomic library that appears to improve the toxin tolerance of *E. coli* and may aid the bacteria in producing biofuels.