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## INTRODUCTION

This volume provides abstracts of research work performed by some of the undergraduate students working in chemistry during summer, 2007. The majority of these participants worked under the auspices of the Research Experiences for Undergraduates (REU) Program in Chemistry sponsored by the National Science Foundation (NSF). This 10-week program provides undergraduate students an intensive hands-on research experience that involves them in all phases of the research process. In carrying out their projects, REU participants work alongside Stony Brook's faculty, post-docs, graduate students and other undergraduates. This year, nine summer researchers from as many college campuses were selected for the REU program, from a pool of over 200 applicants. The REU summer activities culminate in these research presentations being made to members of the Stony Brook Chemistry Department and the university community on August 10, 2007. The REU students are joined in this symposium by Stony Brook student colleagues.

As you read this collection of the students' abstracts, you will see evidence of their hard work, insight and enthusiasm. You will be impressed at what they have accomplished in only ten weeks. I am confident that these individuals will proceed on to successful research careers.

Robert Kerber  
Distinguished Teaching Professor  
NSF REU Chemistry Site Co-Director  
Stony Brook University

**Melanie Bunda**  
York College

**Synthesis of [Re(CCC<sub>6</sub>H<sub>4</sub>-4-CO<sub>2</sub>C<sub>6</sub>HF<sub>4</sub>)(CO)<sub>3</sub>(dmpz)] as a Pre-Building Block for 3-Dimensional Nano-Frameworks**

Melanie Bunda, *York College, York, NE*; Natalie St. Fleur and Andreas Mayr, *Department of Chemistry, Stony Brook University*

The creation of 3-dimensional nano-frameworks is significant in the areas of single-electronics, solar fuel production, and selective filtration. The frameworks are built by first synthesizing rigid corner structures with specific geometry and selectively piecing them together to form a cube or triangular prism. Once made, the possible uses of these structures are endless. The focus of the current research has been to create the title compound. This compound has an activated ester group and a 1,4-dimethylpiperazine (dmpz) ligand. Both features have been designed such that the molecule is a viable precursor to the corner piece of a 3-D molecular structure.

This novel compound was synthesized through a series of organic and inorganic reactions; purified through column chromatography; and characterized with thin layer chromatography, infrared spectroscopy, and nuclear magnetic resonance techniques. We have successfully substituted the activated ester group for other functional ligands. Future studies will determine the possibilities of replacing the dmpz ligand with ligands designed with the 60° angle needed for the triangular prism, or 90° angle needed for the cube. This research was made possible by a grant from the National Science Foundation through the Research Experience for Undergraduates (REU) Program at Stony Brook University.

**Alex Chameessian**  
Stony Brook University

**Computational analysis of 1,3-Allylic Strain in  $\alpha,\beta$ -unsaturated compounds and their  $\beta,\gamma$  isomers**

Alex Chameessian, *Stony Brook University*; Francis Johnson, *Departments of Chemistry, and Pharmacological Sciences, Stony Brook University*; Nancy Goroff, *Department of Chemistry, Stony Brook University*

1,3-allylic strain has been studied extensively in various chemical systems. We have continued these studies utilizing computational methods to determine the relative stabilities of several  $\alpha,\beta$ -unsaturated compounds and their  $\beta,\gamma$  isomers. The systems under study have been observed experimentally in the early 20<sup>th</sup> century by Reginald Linstead. Using the experimental data gathered by Linstead, our research has aimed at quantifying the contribution of 1,3-allylic strain in determining the equilibrium position of the two isomers. However, our calculations are consistent with the experimental data in some systems and inconsistent in others. In order to explain these inconsistencies we plan to direct our efforts toward gathering new experimental data for comparison with our calculations.

**Rikki Enzor**  
Judson College

**Recombinant Generation of pBAD-NTL9, For Production of NTL9-F5<sub>FCN</sub> for Protein Folding Studies**

Rikki Enzor, Judson College, Marion, AL; Juah Chung and Humeyra Taskent, Department of Chemistry, Stony Brook University; Isaac Carrico and Daniel Raleigh, Institute of Chemical Biology & Drug Discovery and Department of Chemistry, Stony Brook University, Stony Brook, NY

Many studies seek to determine the principles that direct protein folding. A complete understanding of these principles will enable the development of treatments for diseases caused by protein misfolding and the genetic engineering of proteins for any specific biological function. Several unnatural amino acids have been introduced into proteins; their novel properties enable study of protein folding mechanisms.

The unnatural amino acid *p*-cyanophenylalanine (F<sub>CN</sub>) has several unique properties that make it a useful fluorescent probe for studying protein folding. It has a strong absorption where tyrosine (Y) and tryptophan (W), the two naturally occurring amino acids that generate fluorescence, do not absorb significantly. Also, the intensity of the F<sub>CN</sub> fluorescence changes significantly as its environment changes between hydrophobic and aqueous. These properties make F<sub>CN</sub> an excellent probe for studying how the hydrophobic core of a protein forms and what drives its formation.

This research project involves using molecular cloning and recombinant DNA techniques to generate pBAD-NTL9 F5F<sub>CN</sub>, a mutant form of the ribosomal protein NTL9 in which F5 is replaced by the unnatural amino acid F<sub>CN</sub>. Several molecular genetics manipulations were performed to transfer the NTL9 gene from a pET-3a vector to a pBAD vector. PCR was performed to remove the NTL9 gene from pET-3a, amplify it, and add NcoI and NdeI restriction sites to either of its ends. To prepare a pBAD Myc-His vector for ligation with the NTL9 gene, the pBAD's second NdeI restriction site, located on the opposite side of the vector from the NdeI in its cloning site, was removed by QuikChange mutagenesis. Corresponding sticky ends were produced on the NTL9 fragment and the pBAD vector by restriction enzyme digest with NcoI and NdeI. The NTL9 insert was ligated into the pBAD vector backbone. When the NcoI restriction site was introduced at the beginning of the NTL9 sequence, one base pair was altered; thus another QuikChange mutation had to be performed to correct the NTL9 sequence. In further experiments, the F5 codon will be mutated to TAG, the amber suppressor codon. A specially developed pSup vector that contains regions coding for tRNA's possessing a loop region that binds F<sub>CN</sub> has been obtained from the laboratory of Peter Schultz of Scripps Institute. A dual-vector system will be produced by transforming pBAD-NTL9 F5F<sub>CN</sub> and pSup, and then protein expression will be induced to generate NTL9 F5F<sub>CN</sub>.

Special thanks to Ruchi Gupta for valuable discussions contributing to this research. Support for this study was provided by the National Science Foundation through the Research Experiences for Undergraduates program.

## Benjamin Gowen Centre College

### **Synthesis of a Tumor-Targeting Drug Conjugate Using a Strategically Designed Self-Immolative Disulfide Linker**

*Benjamin Gowen, Centre College; Danville, KY; Shuyi Chen, Xianrui Zhao, Department of Chemistry, Stony Brook University; and Iwao Ojima, Department of Chemistry, Stony Brook University; Institute of Chemical Biology & Drug Discovery, Stony Brook, NY*

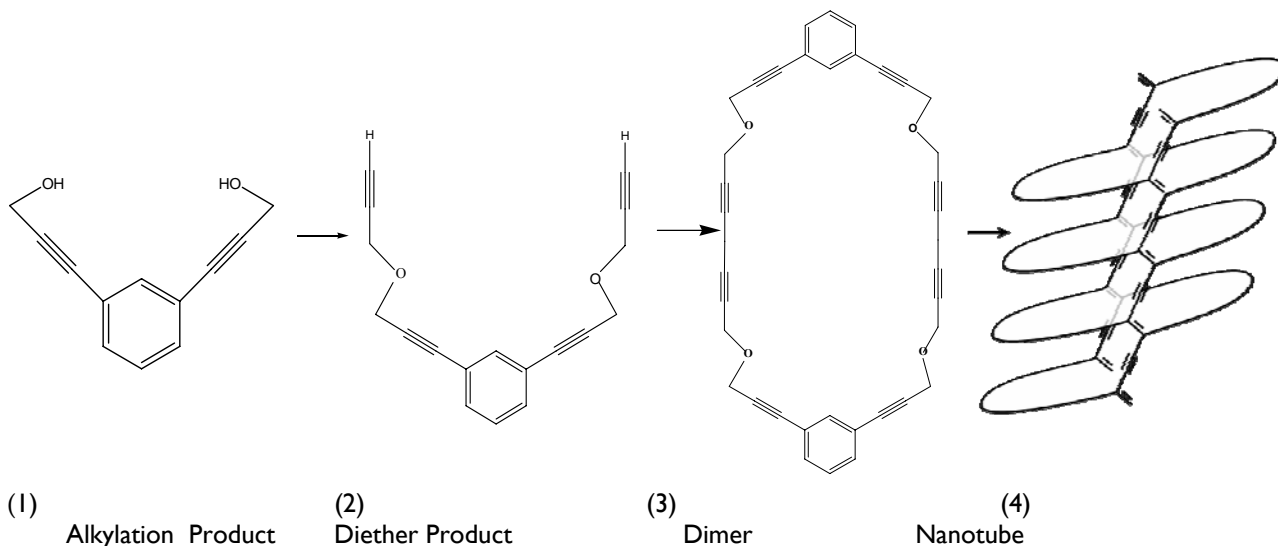
Conventional chemotherapy drugs lack the ability to distinguish tumor cells from healthy cells. This lack of specificity leads to systemic toxicity, causing many harmful side effects. The Ojima group has designed and synthesized tumor-targeting drug conjugates consisting of a cytotoxic drug “warhead” connected to a tumor-targeting moiety by an appropriate cleavable linker. It is important that the chosen linker be stable in the circulatory system, but readily releases the original drug warhead once internalized into tumor cells. In this project, a strategically designed self-immolative disulfide linker was successfully synthesized and attached to the drug warhead paclitaxel, also known as Taxol®. Compounds were characterized by NMR and mass spectrometry. The drug-linker conjugate will be attached to biotin, a tumor-targeting moiety, followed by cytotoxicity assays on the fully assembled conjugate. Additionally, a drug conjugate bearing biotin directly connected to the warhead by an ester moiety was synthesized for comparison with the self-immolative disulfide linker. This work was supported by the National Science Foundation Research Experience for Undergraduates (NSF-REU) 2007 Summer Research Grant as well as a grant from the National Cancer Institute.

## Rebekah Guevara Eastern Nazarene College

### Synthesis of a Macrocyclic Diacetylene System to Form Organic Nanotubes

Rebekah Guevara, Eastern Nazarene College, Quincy, MA; Steven Chow, Frank Fowler and Joseph Lauher, Department of Chemistry, Stony Brook University, Stony Brook, NY

Synthetic organic nanotubes are attracting considerable interest because of their potential applications in biological, chemical and material sciences. The diacetylene macrocycle was designed for organization into an open tubular network, where it could be polymerized to produce a covalent organic tube containing a conjugated polymeric wall, analogous to the carbon nanotubes. Specifically, the research of macrocyclic diacetylene systems has been recently investigated by incorporating pyridine rings into their design. The goal of this research project is to synthesize a macrocyclic diacetylene dimer. Compound **1** was synthesized by Sonogashira coupling reaction starting with *m*-diiodobenzene. Compound **1** was then alkylated via the Williamson ether synthesis to produce diether **2**. The macrocyclic diacetylene was produced by the Hay coupling with **2**. Preliminary results indicate the successful acetylene coupling to the *m*-diiodobenzene as well as the alkylation leading to the production of a diether. Currently studies are being conducted to test the dimerization of the diether to give the macrocyclic diacetylene.



This study was supported by the Research Experience for Undergraduate Program sponsored by the National Science Foundation.

**Avinash Khanna**  
Stony Brook University

**Design of Bio-available InhA Inhibitors with Activity against Drug-Resistant Strains of *Mycobacterium tuberculosis***

Avinash Khanna, *Stony Brook University*; Yelizaveta Gegina, Hua Xu, Nina Liu, Christopher amEnde, Peter J. Tonge, *Department of Chemistry, Stony Brook University*

Every year Tuberculosis claims approximately 1.7 million lives, and the disease continues to spread every second. A physician can cure a patient who is diagnosed with drug sensitive tuberculosis in about 9 months, and much longer if the strain of *Mycobacterium tuberculosis* (MTB) is drug resistant. The treatment for multi-drug resistant MTB would cost the patient about a quarter of a million dollars. The treatment for drug resistant tuberculosis can be made much more time and cost efficient. InhA is an enzyme in the fatty acid synthesis (FAS-II) cycle that synthesizes the precursor to mycolic acids, a vital component of the cell wall of *M. tuberculosis*. Isoniazid (INH), a potent InhA inhibitor, is the current front line drug against tuberculosis. INH is a prodrug that needs to be activated within the mycobacteria by a catalase-peroxidase enzyme called KatG before it can inhibit InhA. About 80% of INH resistance is caused by mutations in the KatG enzyme. To bypass this undesirable activation step, we have synthesized compounds which are direct nanomolar inhibitors of InhA. These compounds are based on triclosan which has a  $K_i$  of  $12.5 \pm 0.0$ , and a  $Mic$  value of  $12.5 \pm 0.0$  for the enoyl reductase (FAB I) enzyme. Despite these novel compounds having excellent inhibition properties *in vitro*, in animal models they showed low bioavailability. Therefore, current work focuses on increasing solubility of the inhibitor in order to help the drug reach a stage of preclinical trials for the treatment of patients infected with drug resistant tuberculosis. This research is supported by the grant NIH 1057765-1-407000, and AI 070J8J, and by the Beckman Foundation's Beckman Scholars Program.

**Jessica Levin**  
Cornell University

**Nanofiltration Membrane Based on Electrospun Nanofibrous Polyacrylonitrile Scaffolds and Crosslinked Polyethyleneglycol and Sodium Alginate Coating**

Jessica Levin, *Cornell University*; Kyunghwan Yoon, Benjamin S. Hsiao and Benjamin Chu,  
*Department of Chemistry, Stony Brook University*

Clean water is becoming a commodity. At least 400 million people today suffer the consequences of water shortages. Water issues are not restricted to landlocked people: sewage and bilge water disposal is a major issue on both cruise and military ships alike. Efficient methods of recycling “dirty” water are desirable to remedy water shortage difficulties as well as sewage control. This study’s objective is to examine a new method to fabricate nanofiltration membranes that have both high flux and good rejection rates, as well as low fouling rates. Nanofiltration (NF) membranes can filter materials as small as viruses, synthetic dyes, and multivalent salts from water. The method includes the use of electrospun nanofibrous scaffold based on polyacrylonitrile with a thin coating of UV-crosslinked polyethylene glycol mixed with sodium alginate (SA) gel. Permeation evaluation of this NF composite membranes was carried out using dead-end filtration and scanning electron microscopy (SEM). SEM results indicated that a thin topcoat (thinner than 1 $\mu$ m) has formed on the surface of electrospun scaffold. Dead-end filtration of Congo red, a synthetic dye, was used to test the rejection rates and permeation flux. The permeation results also indicate that the performance of this membrane system is comparable typical NF membranes.

**Marie Majkut**  
Washington & Jefferson College

**The effect of self-guided Langevin dynamics and temperature on the folding of tryptophan zipper 2**

Marie Majkut, *Washington and Jefferson College, Washington, PA*; Carlos Simmerling, *Department of Chemistry, Stony Brook University, Stony Brook, NY*

Molecular dynamics simulations have made it possible to visualize the folding of proteins under various conditions. The information gathered through simulations can be used to complement experimental data and improve future experiments. Molecular dynamics allow us to determine kinetic and thermodynamic properties of proteins and other larger systems. Though the simulations provide important data, they are often slow and involve the availability of a large amount of computational power. Self-guided Langevin dynamics introduces a guiding force in order to enhance the rate at which a protein or biomolecular system converges to its native state. The strength of the guiding force is based upon the guiding temperature and time step, which can be independently set for each simulation.

The effect of self-guiding on the folding of tryptophan zipper 2 into the native structure was studied in comparison to the folding using standard molecular dynamics. Thousands of simulations were run under various combinations of self-guiding parameters for the SGLD and differing temperatures for the standard MD. The folding rates and corresponding populations were calculated in order to determine if self-guiding helped to locate the native structure faster, if there was any affect on the population, and if self-guiding was better than simply increasing the temperature under standard MD.

The folding rates and corresponding populations were determined with respect to the root-mean-square-deviation between the trajectories and the native structure, as well as with respect to the hydrogen bonds formed. Based upon the rates and populations, self-guided Langevin dynamics with certain parameter values enhances the folding of tryptophan zipper 2 into its native conformation. These results can be applied to simulations involving larger systems such as HIV-protease.

This work was supported by the National Science Foundation through their Research Experience for Undergraduates program.

**Taamee Pak**  
Stony Brook University

**H-NOX Protein Family as a Prokaryotic Nitric Oxide Sensor**

Taamee Pak, *Stony Brook University*; Elizabeth M. Boon, *Department of Chemistry, Stony Brook University*

Biochemical sensing generally involves the binding of a signal to a receptor causing an intracellular cascade which eventually leads to a cellular response. There are many different types of signals such as hormones, neurotransmitters, and small gas molecules. Nitric oxide (NO) is one such gaseous signaling molecule involved in the immune, circulatory, and nervous systems. The NO receptor in eukaryotes is an enzyme called soluble guanylate cyclase (sGC). sGC shares a considerable percentage of homology to the Heme Nitric Oxide and/or Oxygen (H-NOX) binding family in prokaryotes. We propose that the H-NOX domain is a prokaryotic NO-sensing protein that induces cellular response, most likely through regulation of histidine kinase, a protein encoded by a neighboring section of the genome.

To test our hypothesis, we amplified the H-NOX gene from *Pseudoalteromonas atlantica*, a primary biofilm bacteria, and cloned it into an *E. coli* expression plasmid. We then expressed and purified the H-NOX protein from the *E. coli*. The H-NOX protein will now be characterized via NO-binding and kinetics experiments which will indicate whether or not H-NOX has the properties of an NO sensor.

Supported by a URECA summer fellowship.

## Rohit Repala

### Stony Brook University

**Design, Synthesis and Mechanism Study of Taxane Based Tumor-targeting Anticancer Agents**  
 Rohit Repala, Stony Brook University; Jin Chen and Iwao Ojima, Department of Chemistry, Institute of Chemical Biology & Drug Discovery, Stony Brook University

Currently, Taxol® is the most effective chemotherapy drug used to treat cancers such as ovarian, breast, and non-small cell lung cancer. Taxol® and other traditional cancer drugs work on the principle that rapidly dividing cells are more likely to be killed than normal cells. The shortcoming of this methodology is that healthy tissue is also attacked due to the lack of specificity of current cytotoxic drugs causing undesirable side-effects. Therefore, there is an urgent need for the development of new drugs that are capable of increased selectivity for tumors.

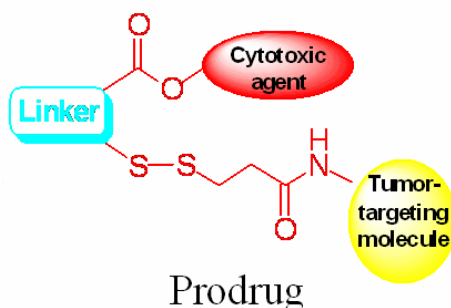
Growing tumors have an enhanced metabolism and overexpress many tumor-specific receptors, which can be used as targets for many tumor-targeting cytotoxic agents. One promising strategy involves linking a cytotoxic agent (warhead) to a tumor-targeting moiety through a suitable linker (Figure 1). This "Prodrug" would remain systemically non-toxic until it reaches the target site. Once bound to the target cell, the drug is internalized and is restored to original potency.

The first part of my project involved the synthesis of the Prodrug using DHA as the tumor-targeting moiety, SB-T-1214, as the cytotoxic warhead and a disulfide containing linker. DHA, a naturally occurring polyunsaturated fatty acid (PUFA) is used as the tumor-targeting moiety because growing tumor cells use PUFAs as biochemical precursors and for energy sources. Previous studies have proven that the new second generation taxoids (including SB-T-1214) have 1-3 orders of magnitude higher potency than Paclitaxel, the active compound in Taxol®. A disulfide containing linker is attractive for this DHA-taxoid conjugate because it accounts for the higher concentration of glutathione (GSH) in tumor cells. Glutathione cleaves the linker and releases the original taxoid from the conjugate.

The second part of the project involves elucidating the mechanisms of internalization, drug release and real-time distribution of the conjugate inside cancer cells using fluorescent microscopy. We will utilize fluorescence microscopy, to elucidate the interaction between the tumor-targeting molecule and its receptors, the internalization of the conjugate, real-time distribution of the conjugate inside the cell, and the release of the drug. Understanding the mechanism of internalization and release of the taxoid will give us the foundation to conduct further studies to develop potent and tumor-specific cytotoxic agents.

This research is supported with funding from the Beckman Scholars Program.

Figure 1.



**Megan Sikowitz**  
St. Mary's College of Maryland

**Enzymatic Synthesis of Steroid Substrates for *M. Tuberculosis Rv1106c* Using Cholesterol Oxidase**

Megan Sikowitz, *St. Mary's College of Maryland*; Xinxin Yang and Nicole S. Sampson, *Department of Chemistry, Stony Brook University*

*Mycobacterium tuberculosis* is an opportunistic infection estimated to affect 30% of the world's population. *Mtb* organisms resistant to front line medications and now second line medications have emerged. The emergence of extremely drug resistant *Mtb* has raised the importance of understanding the pathways of infection and finding new drug targets. The *Mtb* gene *Rv1106c* has recently been characterized as a  $3\beta$ -hydroxysteroid dehydrogenase. The dehydrogenase activity in *Mtb* is important in the infection pathway, although it is not entirely yet clear where in the pathway this activity is important. One possibility is that the oxidation of cholesterol in the membrane changes the structure of the cell membrane and alters its permeability. A second possibility is that the dehydrogenase is required for biosynthesis of steroidal immunomodulators.

In order to further understand and characterize *Rv1106c*, steroid substrates were synthesized utilizing the enzyme activity of cholesterol oxidase from *Streptomyces* that was heterologously expressed in *E.coli*. The synthesis of these molecules will be of use in determining the substrate specificity of *Rv1106c*. This enzyme-mediated synthesis involved the expression and purification of the choA protein, then enzymatic synthesis, extraction and purification of the steroid products. Three sterols were used as substrates in the enzymatic reactions with both wild type and mutant E361Q ChoA to yield the product and intermediate. Funding for this project was made possible by the National Science Foundation as part of the Research Experience for Undergraduates grant and grants from the National Institutes of Health.

**Nathaniel Webber**  
Truman State University

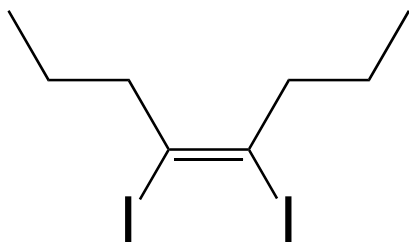
**Experimental Study of the  $^{13}\text{C}$  NMR of Cis and Trans-4,5-diiodo-4-octene in Lewis Basic Solvents**

Nathaniel T. Webber, Truman State University, MO; Nancy S. Goroff, Department of Chemistry, Stony Brook University, NY

Previous experiments have shown that iodoalkynes and iodoalkenes act as Lewis acids when in Lewis basic solvents. This acid-base interaction can shift the  $^{13}\text{C}$  NMR by as much as 15 ppm. The role of stereochemistry has not been examined in previous studies. Calculations suggest that regio and stereoisomers of diiodoalkenes will exhibit significantly different NMR solvent effects. To test this possibility, we set out to prepare cis and trans-4,5-diiodooctene (Figure 1 and 2). The cis isomer has been prepared according to literature procedures, but the synthesis of the trans isomer has been elusive. The  $^{13}\text{C}$  NMR of the cis isomer has been measured in chloroform, pyridine, and DMSO.

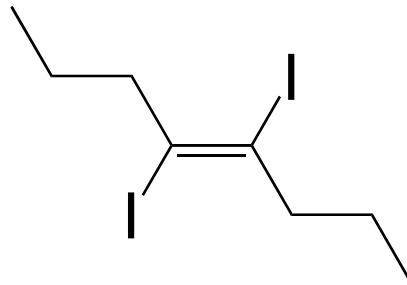
Acknowledgement: This research was supported by the NSF-REU at Stony Brook and ongoing research is support by NSF.

**Figure 1.**



**cis-4,5-diiodo-4-octene**

**Figure 2.**



**trans-4,5-diiodo-4-octene**

**Beth Zucconi**  
Shippensburg University

**Detection of Inhibition of Menaquinone-9 Biosynthesis in *Mycobacterium tuberculosis***

Beth Zucconi, Shippensburg University, PA; Rong Zhou, Peter Tonge, Huei Jiun Li, Varun Talanki, Tonge Lab – Department of Chemistry, Stony Brook University, NY

Tuberculosis, once thought to be well-controlled, has again become a medical threat with the proliferation of multi-drug resistant strains (MDR) and extreme resistant strains (XTR) of *Mycobacterium tuberculosis* as well as with increased susceptibility due to the immunocompromised state of multitudes of individuals afflicted with HIV. This calls for novel drug development. A prime target is the menaquinone (MK-9) biosynthesis pathway. This lipid (MK-9) is an indispensable part of the electron transport chain needed for the bacteria to produce energy for life. It is not present in humans who have Coenzyme-Q10 in its place. Therefore a drug hindering the synthesis of MK-9 would have minimal effects on human metabolism while killing the bacteria. A method of detecting MK-9 absence is necessary to prove the blocking of this specific pathway. *Bacillus subtilis* and *Mycobacterium smegmatis*, non-pathogenic bacteria similar to *M. tuberculosis*, were grown and lysed with heat. Cellular debris was removed by centrifugation while the lipids were extracted from the supernatant using Bligh and Dyer method. These lipids were concentrated and placed on a solid matrix for MALDI mass spectroscopy. The resulting spectra from *M. smegmatis* showed small peaks at 785.274 m/z and 789.344 m/z, consistent with the mass of oxidized and reduced MK-9 respectively and well as with its low cellular concentration. Menaquinone-9 was then synthesized organically to provide a standard for testing and to form an isotope to affirm its identity on mass spectra when the isotopic labeled intermediate in the pathway is fed to growing bacteria. 2-methyl-1,4-diacetyl-naphthalene was directly deacetylated with  $K_2CO_3$  to form the respective diol. The isoprene chain from the respective alcohol solanesol was then coupled to the naphthalene using  $BF_3 \cdot Et_2O$  in an anhydrous reaction dissolved in dry dioxane. Isotope formation required monodeacetylation under the same conditions, variant proportions, and identical isoprene coupling. A dimethoxy-naphthalene was then formed using NaH and  $CH_3I$ . Ceric ammonium nitrate will be used with  $O^{18}$  labeled water to form the oxidized MK-9. This will then be run on mass spectrometry to determine behavior of MK-9 under such testing conditions. This research was funded by the National Science Foundation Research Experience for Undergraduates and the National Institutes of Health.

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