

This summer I had the opportunity to work in the Biology Department at Sewanee, under the supervision of Dr. Elise Kikis. The internship began May 13<sup>th</sup> and ended August 15<sup>th</sup>. This internship was an opportunity to expand my previous work during the school year of 2012 – 2013. It allowed me to finish incomplete projects from the previous semesters. The work ran from Monday through Friday and sometimes Saturdays. The hours were flexible for each individual intern, as it was dependent on the tasks set for each person. As a lab, we conducted experiments using *C. elegans* as the genetic model system to address questions relating to the aggregation/toxicity of human neurodegenerative disease-associated proteins. Currently, the two proteins that we are observing are the *atx-3* and huntingtins.

Throughout the summer, I was expected to carry out my own experiments as well as to assist those in my lab with their projects too. As a team, we all had to help maintain the equipment and the lab space. In addition to maintaining the workspace as individuals, it was our responsibility to update our lab members with data relevant to our individual work. Therefore, every Tuesday beginning in June, I attended lab meetings throughout the internship period. Prior to our meetings we were assigned to read papers relevant to our work, as it revealed useful information to conduct our own research. At lab meetings we would discuss these papers and ask each other any questions that were relevant to our experiments. Lab meetings were also a time for us to ask each other questions concerning our own projects and any issues concerning failures and successes.

In addition to these responsibilities, I was accountable for the Huntington's project. The scientific question that I studied was "what is the effect of the polyQ expansion in the context of the huntingtin protein (Htt)." Specifically, I used molecular cloning techniques to express the Htt protein tagged with yellow fluorescent protein in *C. elegans* neurons. Essentially, this allowed me to explore a number of questions relating to protein accumulation, localization, and toxicity which inevitably will help determine if there are any genetic factors contributing to the aggregation and toxicity of the Huntington protein. To do this I had to create the necessary gene constructs. To obtain these constructs, I began with a procedure called Polymerase Chain Reaction (PCR). I started with a template/ pCI-neo vector/ plasmid which I then infused with polyQ expansion length of 15 nucleotides of Huntington's DNA. The appropriate forward and reverse primers were also added to this solution. The purpose of PCR is to amplify the region of DNA. In my case, I set my reactions for PCR so that I could make copies of the DNA sequence containing the Huntington's region. After producing the necessary copies of DNA fragments, the recombinant DNA was introduced into bacterial *E. coli* cells. After incubating the *E. coli* cells at room temperature, I plated them onto kanamycin-resistant plates, which I allowed to grow overnight. This allowed me to grow individual colonies. To test whether or not these colonies actually acquired the Huntington PolyQ15 DNA, I took a microscopic sample of each individual colony and incubated them separately in LB broth solution with kanamycin. I was able to collect each colony and use a

procedure called *MiniPrep* by *Qiagen* to obtain the DNA from the bacteria. At the end of the internship, I was then able to send my samples to the Yale DNA Sequencing Center to see if I indeed produced the necessary constructs to insert into the *C. elegans* gonads. The sequencing will enable me to determine if the *E. coli* cells have acquired the DNA fragments I had amplified from my PCR.

Upon the establishment of these lines, I will be able to further my research in the next semesters here at Sewanee. By producing my gene constructs I have initiated a much larger project that will need additional help from other students. Ultimately, I have helped open a new opportunity for students interested in research. In the larger science community, I initiated a new project that will further aid other scientists studying neurodegenerative diseases, specifically Huntington's disease. Throughout this internship, I not only worked on my own project but I contributed my efforts to lab members working on the Ataxin-3 gene. I helped by keeping their lines healthy by feeding their *C. elegans* and by making plates with food necessary to keep them alive. In addition, I helped produce several western gel blots to indicate the levels of protein with different PolyQ lengths in the worms.

In my experience working in Dr. Kikis' lab, I learned how to work with other scientists in a professional manner. While working in the lab I also learned how to work high-functioning and high-maintenance machines that required patience and care. This is very important in my desired future field of work because knowledge of these machines will help me carry out tasks in different labs. Some

of the high points of this research was the opportunity to constantly learn something new every day and not only learn from the instructor but from my lab members, too. The lab meetings were also educational opportunities because it allowed me to understand other aspects in neurodegenerative diseases. In one lab meeting we learned and reviewed the ethics of laboratory work. I believe that was the most informative and most relevant to everyone in the lab. One of the low points I experienced was the failure of certain experiments, which followed in many trials of the same experiments, which caused some days/ weeks to be redundant. However, I cannot always expect my experiments to work, so I remained patient. This research opportunity has been very satisfying and complementary to my future goals as a scientist. I plan to continue this research and plan to publish a thesis and hopefully a full manuscript. If not, this research has given me an opportunity to expand this research further into my graduate studies.