Variable responses to the neuropeptide C-type allatostatin (AST-C), by the cardiac neuromuscular system of the American lobster, *Homarus americanus*

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Central pattern generators (CPGs) are neural networks that control rhythmic motor systems in organisms by producing consistent, patterned outputs to activate muscle tissue. CPGs serve critical functions in the organisms that they are found. For example, the cardiac ganglion found in crustaceans is responsible for creating the pattern of action potentials necessary for a constantly rhythmic heartbeat. Chemical compounds such as neuropeptides act as modulatory signals that influence central pattern generators to generate a variety of motor outputs. However, the mechanisms that cause central pattern generators to produce different motor outputs are not yet well understood. Since it is known that neuropeptides can influence the central pattern generator to generate different motor outputs, it is useful to study the roles and physiological impact of these neuropeptides in individuals so that we then can learn about the principles behind pacemaking and modulation of central pattern generator.

C-type allatostatin (AST-C), a neuropeptide, has been identified to modulate the CPG in *H. americanus.* Recent studies by the Dickinson lab have revealed a great deal about the impact AST-C has on cardiac output. It is clear that in all lobsters the application of AST-C into the CPG results in an overall decreased heart rate, and it was also shown that AST-C alters contraction amplitude (Wiwatpanit et al, 2012). What is most interesting is that the response by the CPG to AST-C was not constant across individuals. Instead, AST-C caused either an increase or decrease in contraction amplitude across individuals. We hypothesized that the differences in the expression of AST-C receptors among individuals may be regulated by the molt cycle. Because the total hemolymph (blood) volume increases during the molt cycle, a lobster likely must adjust the force with which its heart contracts depending on the total hemolymph volume. Moreover, it is hypothesized that some molt-related protein may actually alter the expression of AST-C receptors, and thus alters the heart's response to AST-C application. Thus far the Dickinson lab has identified four AST-C receptors, however the mechanisms that underlie how they are interact with AST-C peptides are not yet well understood.

We plan to study the relationships between response to AST-C, receptor expression, and molt cycle using transcriptomics, which will allow us to directly examine expression levels of proteins of interest using transcriptomes. A transcriptome is a snapshot of all the mRNA sequences being expressed at a certain time within a specific type of tissue. We spent this past summer first collecting physiological data on whole heart responses to the different isoforms of AST-C, and then isolated RNA from key tissues in each lobster tested. We grouped RNA samples based on physiological response, ie. whether the heart responded to AST-C with an increase or decrease in contraction amplitude. By using a bioanalyzer, we were able to ensure all RNA samples were of good quality as well as quantity. We now have sufficient amount of RNA samples that we hope to send off for sequencing this fall.

In our future study, we will continue to measure the physiological response of the whole heart and continue to compile our RNA samples. Additionally, we will isolate RNA from juvenile lobsters and generate transcriptomes from these samples. The transcriptomes of these juvenile lobsters will be used as a fingerprint to determine the molt stage of the adult lobsters. We predict that we will find a correlation between the molt stage of the lobsters, receptor expression and physiological response to AST-C.

Faculty Mentor: Patsy Dickinson
Funded By:NSF [IOS-1354567], NIH [Maine INBRE, P20GM0103423 from NIGMS]
References
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