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Henry L. and Grace Doherty Charitable Foundation Coastal Studies Research Fellowship

Role of Specific Amino Acids in the Function of Allatostatin Peptides in the American Lobster

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My project this summer was focused on investigating what part of the structure of AST-C peptides is responsible for differences in the responses to similar peptides. Peptides are short chains of amino acids and can modulate neural circuits, which in turn can control rhythmic movement. The family of peptides that I focused on is the AST-Cs. The three isoforms, peptides with different variations in their overall similar amino acid sequences, we looked at were AST-CI, AST-CII, and AST-CIII (See Figure A) in the American lobster (Dickinson et al., 2018).

The lobster heart is neurogenic, meaning that it is controlled by the cardiac ganglion, a group of neurons in the heart (Cooke, 2002). Thus, we can remove the lobster heart from the lobster body and as long as it is maintained in conditions that mimic those in the lobster, it can keep on beating for hours. The heart was isolated, cannulated and perfused with saline. I then tied the anterior arteries to a force transducer and recorded the amplitude and frequency of the heart contractions, that is, how hard the heart was beating and how fast it was beating (Dickinson et al., 2018). We then perfused the different AST-C peptides through the heart by adding low concentrations of the peptides to the saline running through the heart; this enabled us to record how the lobster heart beat was modulated.

While perfusing each of the three peptides through the heart in the past, the Dickinson lab found that AST-CI and AST-CIII caused similar changes in both amplitude and frequency of the heartbeat, but both differed from the responses caused by AST-CII. Concluding that AST-CI and AST-CIII cause more similar responses in the heart and that AST-CII causes a different one, we looked at the amino acid sequence to try to understand the root of this difference.

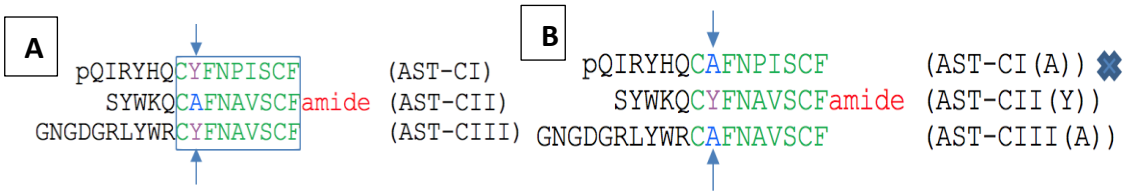

Figure A depicts the amino acid sequence of the AST-C peptide isoforms AST-CI, AST-CII, and AST-CIII. These are normally produced in the lobster heart. The colored part of the sequence represents the conserved region of the sequence in lobsters. Observing this sequence, we noticed that both AST-CI and III had a tyrosine (marked by Y) while AST-CII had alanine (marked by A) in that specific position of the conserved region of the AST-Cs. Alanine is non-polar while tyrosine is polar, making them very different (Dickinson et al., 2018). We hypothesized that this might be the cause of the similarity in the modulations caused by AST-CI and AST-CIII and the different modulations caused by AST-CII. To test this hypothesis, we generated peptides with altered amino acid sequences. Figure B depicts the artificially generated peptides with AST-CII(Y) now having tyrosine and AST-CIII(A) now having alanine. We predicted that these generated isoforms would modulate the heart differently than their original counterparts, that those with tyrosine (Y) would modulate the heart similarly to one another, and that those with alanine (A) would modulate the heart similarly as well.

The responses of the heart to this peptide family vary across individuals, so comparisons require a large sample size. The sample size of 32 lobsters we gathered to date are generally consistent with my hypothesis that the identity of the alanine or tyrosine in the amino acid sequence is an important cause of the differences in responses to similar peptides, but we only have about half the sample size needed to draw clear conclusions. I look forward to continuing this research in the years to come.

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