Neuropeptide Identification and Modulatory Impact of Diet in Three Crab Species Emily Herndon '23

This summer my research focused on determining whether ten different neuropeptides were present in the brains of three crab species. This topic was based upon prior research in the Dickinson and Stemmler Labs which demonstrated that some neuropeptides traditionally found in other crustaceans were incapable of eliciting a response in the nervous system of the *Pugettia producta* aka Kelp Crab (Dickinson et al, 2008). The Kelp Crab differs from other crustaceans because of its exclusively herbivorous diet of kelp. This has been the foundation for my research hypothesis— is the brain of the Kelp Crab lacking the presence of specific neuropeptides found in other crabs with omnivorous diets? To better explore this question, I compared the Kelp Crab to two additional crab species: *Libinia emarginata* and *Chioenecetes opilio* (the Spider Crab and Snow Crab). Both species have a traditional crustacean omnivorous diet and are within the same genus as the Kelp Crab.

The Dickinson Lab initially compiled a list of ten neuropeptides (included below) to research whether the peptides elicited responses in the nervous systems of the three crabs. In correlation with this ongoing work, I used the dissected brain samples from these same crabs to determine if the ten neuropeptides were present utilizing a mass spectrometry instrument. Determining the presence is critical for this work because it offers an explanation as to why a nervous system may be unresponsive—it could either be lacking a specific peptide or contain the peptide without its necessary receptor.

To evaluate the presence of the neuropeptides I followed two different protocols. First, I utilized the crabs' transcriptomes which contain their genetic information in the condensed form of coded letter sequencing. I was able to search this database by using bioinformatics software and eventually determine how the amino acid sequence of the peptides (the chemical makeup of the compounds) would appear in each crab. Understanding the chemistry of a peptide allows us to create a prediction about how the compound will appear on a mass spectrometry reading. Alongside this work, I followed a chemical procedure to extract a solution of neuropeptides from the brain samples. Finally, I ran the mass spectrometry instrument on the samples to confirm whether the neuropeptide samples were present as specific peak values.

I have included a table for the ten neuropeptides below, alongside my determinations for the presence of each peptide within the three different crab brains. This data will be utilized in collaboration with the work of Dan Powell and the Dickinson Lab to evaluate the connections between neuropeptide presences and responses in different diets. Furthermore, I plan to continue my research in the form of a Chemistry Honors Project this year and broaden my exploration into the chemical properties of crustacean brains.

	Pugettia	Libinia	C. Opilio
1. CabTRP I	DETECTED	DETECTED	DETECTED
2. CCAP	DETECTED	NOT DETECTED	NOT DETECTED
3. Myosuppressin	DETECTED	DETECTED	DETECTED
4. Proctolin	DETECTED	DETECTED	DETECTED
5. RPCH	DETECTED	DETECTED	DETECTED
6. G-SIFamide	DETECTED	DETECTED	DETECTED
			in both forms
7. ACP	NOT DETECTED	NOT DETECTED	NOT DETECTED
8. NRNFLRFa	DETECTED	DETECTED	DETECTED
9. HIGS peptide	DETECTED	DETECTED	DETECTED
10. CDLDH	DETECTED (WEAK)	DETECTED (WEAK)	NOT DETECTED

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References

Dickinson, P.S., Stemmler, E.A. and Christie, A.E. (2008) 'The pyloric neural circuit of the herbivorous crab Pugettia producta shows limited sensitivity to several neuromodulators that elicit robust effects in more opportunistically feeding decapods', *Journal of Experimental Biology*, 211(9), pp. 1434–1447. doi:10.1242/jeb.016998.