Modulation of Cardiac Performance in the Blue Crab, *Callinectes sapidus*

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Introduction:

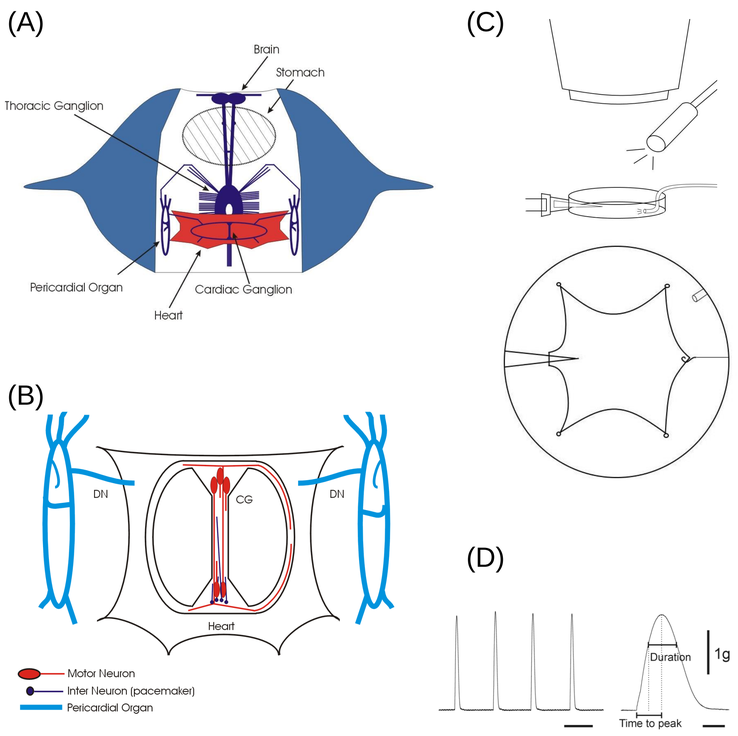
Many rhythmical behaviors such as walking, flying, breathing and chewing are generated by neural circuits. These neurons or “central pattern generators (CPGs)”, produce repetitive, rhythmical bursts of activity that are conveyed to the muscles or “effectors” that produce the behavior. Most behaviors are modified or modulated to adjust the behavior.

The heart of the blue crab, *Callinectes sapidus*, is a simple central pattern generator-effector system. The 9 neuron cardiac ganglion (CG) is a simple CPG located within the cardiac musculature (see Figure 1A and 1B). 4 interneurons act as pacemakers that drive 5 motor neurons that trigger heart contractions. The basic rhythm of the blue crab CG can be modulated by both hormonal and neural signals. Pericardial organs (PO) are neurohaemal structures that flank the heart and release bioactive products into the circulation. The CG is also directly controlled by 3 cardioregulatory fibers that originate in the central nervous system. 1 of these fibers is inhibitory, while the other 2 are excitatory in nature.

Blue crabs are poikilotherms that are exposed to a range of environmental temperatures due to their distribution (western Atlantic from Cape Cod to Argentina) and seasonal variations. Changes in environmental temperature pose physiological challenges to all poikilotherms.

The long term aims of this study are (1) to examine the modulatory effects of two biogenic amines, Dopamine (DA) and Serotonin (5HT), that are released from the PO, on the crab heart, (2) to examine the effect of temperature on cardiac performance, and (3) to determine if the modulation of the crab heart by biogenic amines is altered by temperature.

Figure 1: Schematics of the crab heart and experimental setup. (A) Locations of the heart and pericardial organs in the intact crab. (B) Pericardial organs with dorsal nerve (DN) containing cardioregulatory fibers projecting to the cardiac ganglion within the heart. (C) Experimental setup of cannulated heart attached to force transducer. (D) Sample contractions as recorded with the isometric force transducer.



Methods:

Specimens of *Callinectes sapidus* were purchased locally and housed in aquaria at Valdosta State University. Specimens were covered in ice (20 min) to achieve immobilization prior to dissection. Working heart preparations were dissected following previously developed methods (see Fort *et al.,* 2004). Briefly, hearts were removed intact and cannulated with a modified syringe needle mounted in a sylgard lined petri dish. (See Figure 1C) The heart was attached to the force plates of a Grass FT03 isometric force transducer and placed under a resting load. Perfusion with crab saline was maintained at a constant rate and pressure. Perfusion rate and pressure were maintained when modulator (Dopamine and Serotonin) trials were performed.

Results:

Dopamine and Serotonin both induce excitatory effects in the heart.

Over the range of concentrations examined (10^{-10}M to 10^{-3}M), both modulators induced excitatory effects on contraction frequency a threshold concentration of 10^{-8}M (see figures 2 & 3) before effects on amplitude were observed.

Maximal excitatory effects on contraction were observed to occur at 10^{-5}M DA and 5HT (not shown).

Summary effects for three concentrations of DA and 5HT (10^{-8}M, 10^{-6}M, & 10^{-4}M) on frequency and amplitude are shown in figure 5.

Phase plots of the rate of change of the force as a function of the force of contraction (figure 4) are similar shapes in control and under test conditions (10^{-6}M drug). This suggests DA and 5HT increase the rate of rise and decay of heart contractions in direct proportion to the increase in the force of contraction.

Figure 2: Effect of Dopamine on heart contractions. Arrow indicates time of drug application. Time bar = 30 sec. Force bar = 1g

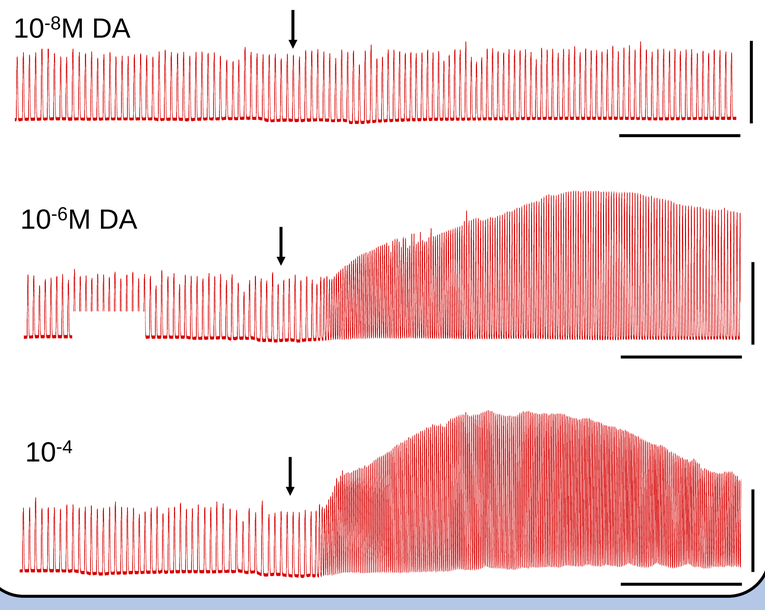


Figure 3: Effect of 5HT on heart contractions. Arrow indicates time of drug application. Time bar = 30 sec. Force bar = 1g

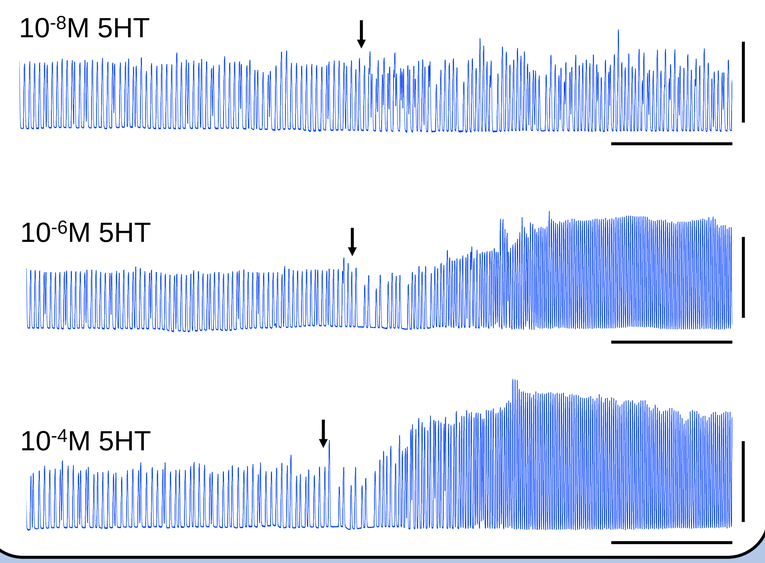


Figure 4: Phase plot of the rate of change of the force (y-axis) as a function of the force of contraction (x-axis). Ten contractions are plotted under control conditions (black lines) and in the presence of 10^{-6}M DA (red lines) and 10^{-6}M 5HT (blue lines). Similar shapes of the phase plots (black vs. color) indicate that the rates of rise and decay of the contractions are both directly proportional to the amplitude of the contraction under the 2 conditions.

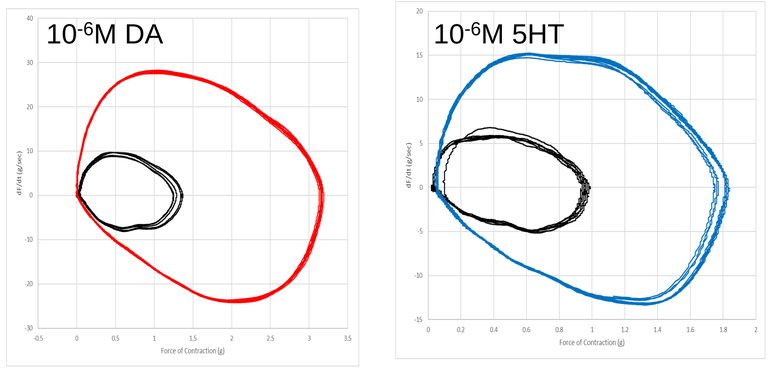
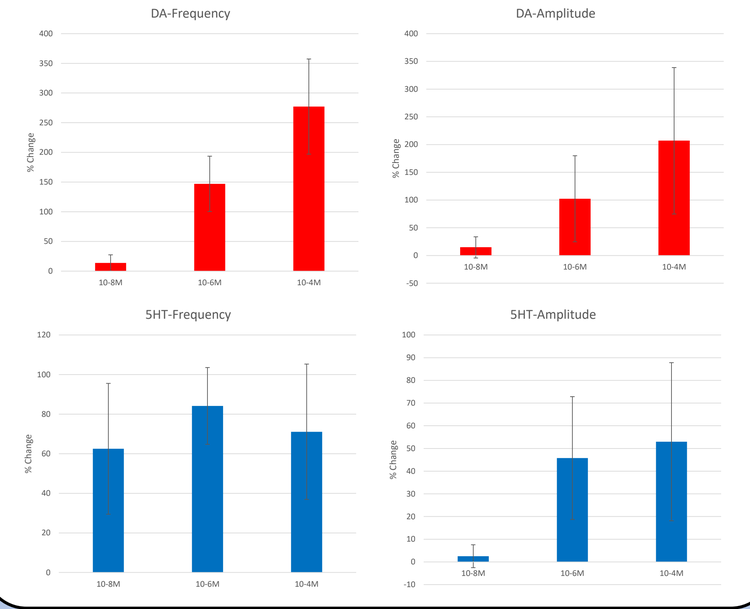


Figure 5: Effects of Dopamine and Serotonin on contraction frequency and amplitude. (n=3)



Discussion:

Dopamine and Serotonin induced positive chronotropic and inotropic effects in the heart of the blue crab (Figures 2 & 3).

The threshold of these effects for the modulators is consistent with the concentration of circulating neurohormones in the intact crab (10^{-8}M) (Fort *et al*, 2004).

Of the two biogenic amines, dopamine appears to be a more potent modulator of cardiac activity than serotonin. At a concentration of 10^{-6}M, dopamine is approximately twice as potent as serotonin in inducing chronotropic and inotropic effects (Figure 5).

The main effect of both modulators on contractile parameters appears to be on contraction amplitude. Phase plots of rate of change of force as a function of force, for both modulators, show similar shapes. This indicates that the rates of rise and decay of the contractions are both directly proportional to the amplitude of the contraction under the 2 conditions (Figure 4).

While phase plots are a good indicator, further analysis of contractile parameters under control and modulator conditions is required. Measurements of parameters such as time to peak, contraction duration, rate of rise and rate of decay are required for a more complete understanding of the effects of the modulators.

These experiments have illustrated that Dopamine and Serotonin, at concentrations consistent with neurohormones in the intact crab (10^{-8}M – 10^{-6}M), modulate cardiac activity. Concentrations of 10^{-6}M Dopamine and Serotonin will be used in future experiments when examining how temperature effects the cardiac system.

Future Work:

(1) To increase the number of working heart preparation experiments examining the effects of DA and 5HT.

(2) To start trials of working heart preparations at different experimental temperatures in the absence and presence of DA and 5HT.

(3) To examine the combined effects of temperature and modulators on the isolated cardiac ganglion (nerve recording only preparation) and the semi intact working heart preparation (nerve and muscle recording preparation).

References:

Fort, T.J., Brezina, V., & Miller, M.W. 2004. Modulation of an integrated central pattern generator-effector system: Dopaminergic regulation of cardiac activity in the blue crab *Callinectes sapidus*. *J. Neurophysiol.* 92: 3455-3470

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