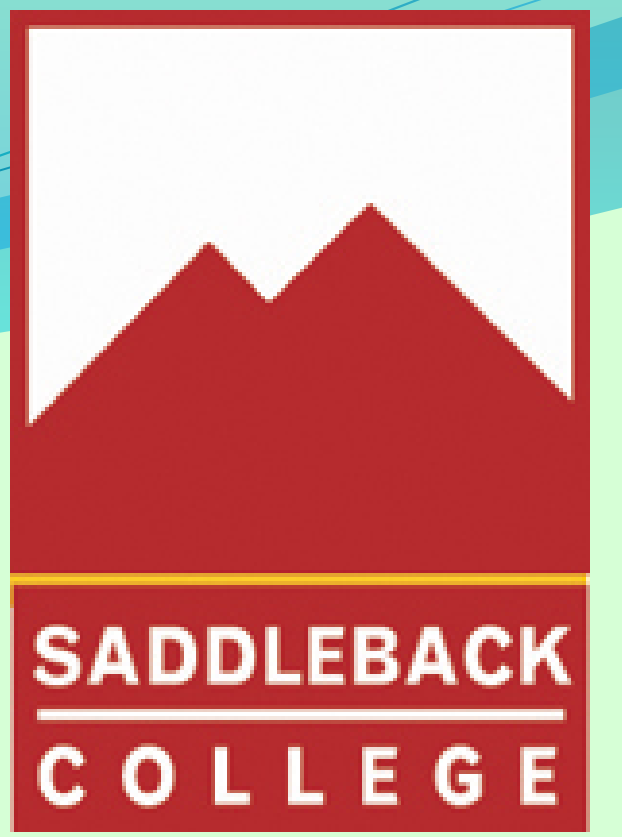


# The Effects of Salinity in the Concentrations of Hemolymph and Hemocyanin in Blue Crab, *Callinectes sapidus*

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## Introduction

Osmoregulation is a process that allows organisms to balance internal water and solute concentration levels. It is an essential process for crustaceans such as *Callinectes sapidus*, because it allows them to adjust to discrepancies in their surrounding environment. Shifts in salinity levels generally cause metabolic changes in crabs, which they counter by regulating their hemolymph osmolytes. Evidence of osmoregulation in *Callinectes sapidus* should present as either hyperosmotic or hypoosmotic hemolymph concentrations in relation to ambient salinity. It is hypothesized that the hemolymph osmolality should be hyperosmotic in relation to the different salinities.

## Materials and Methods

24 female blue crabs *Callinectes sapidus* (Figure 1) were sourced from a local market (99 Ranch) and acclimated to saltwater solutions of varying dilute salinities for 24-hour periods. At each dilution, 20  $\mu$ L samples of hemolymph were collected from each crab using a 27-gauge hypodermic needle and syringe. Collected samples were centrifuged and subsequently tested for concentrations of hemocyanin and osmolality using spectrophotometry and vapor pressure osmometry.



Figure 1. Blue crab, *Callinectes sapidus*

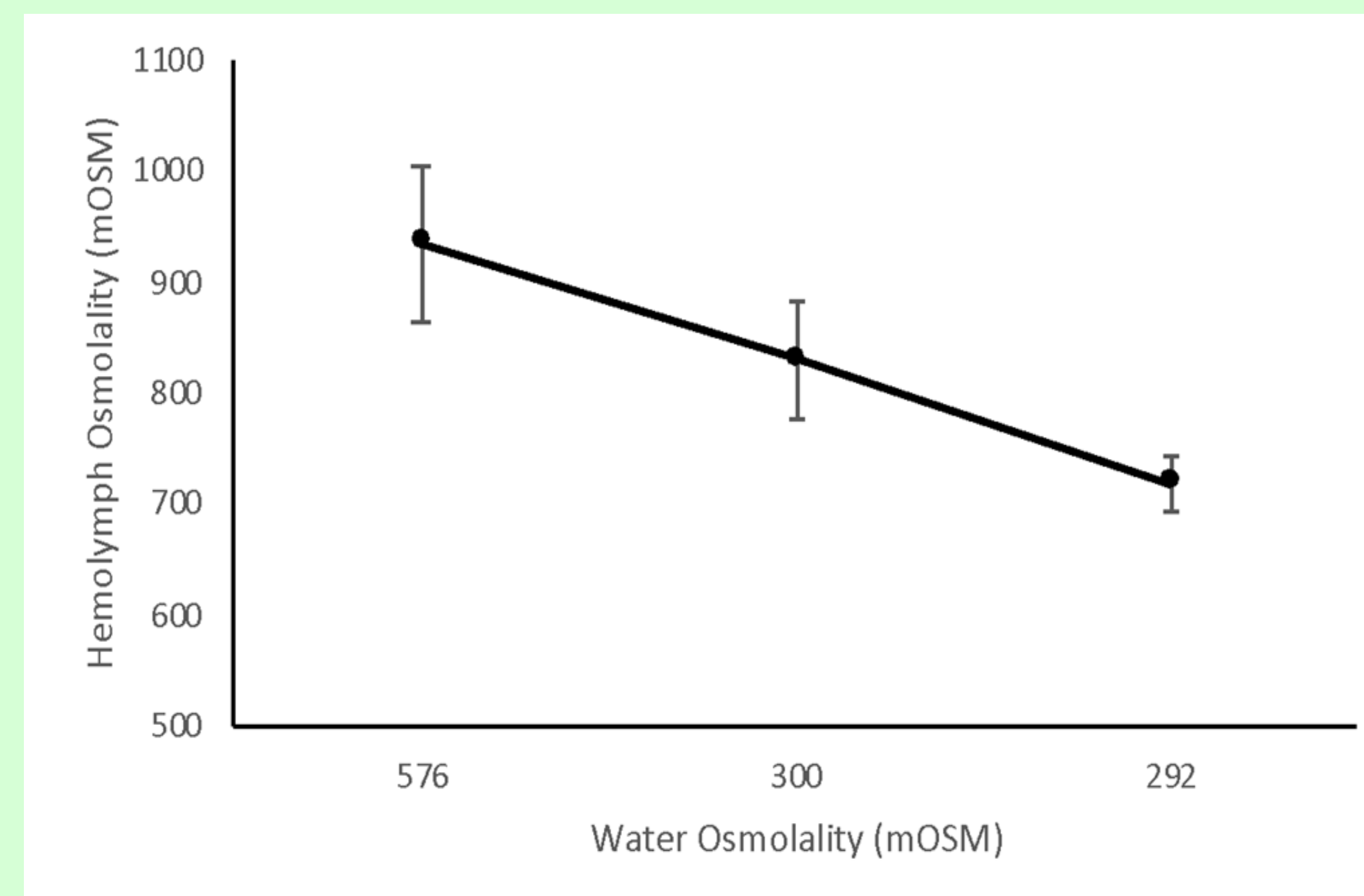


Figure 2. Mean hemolymph osmolality of *C. sapidus* as a function of water osmolality. Significant changes in hemolymph osmolality were noted ( $p < 0.05$  using a two tailed t-test,  $N=14$ ). Error bars indicate SEM ( $\pm 70, 53, 24$  mOSM respectively).

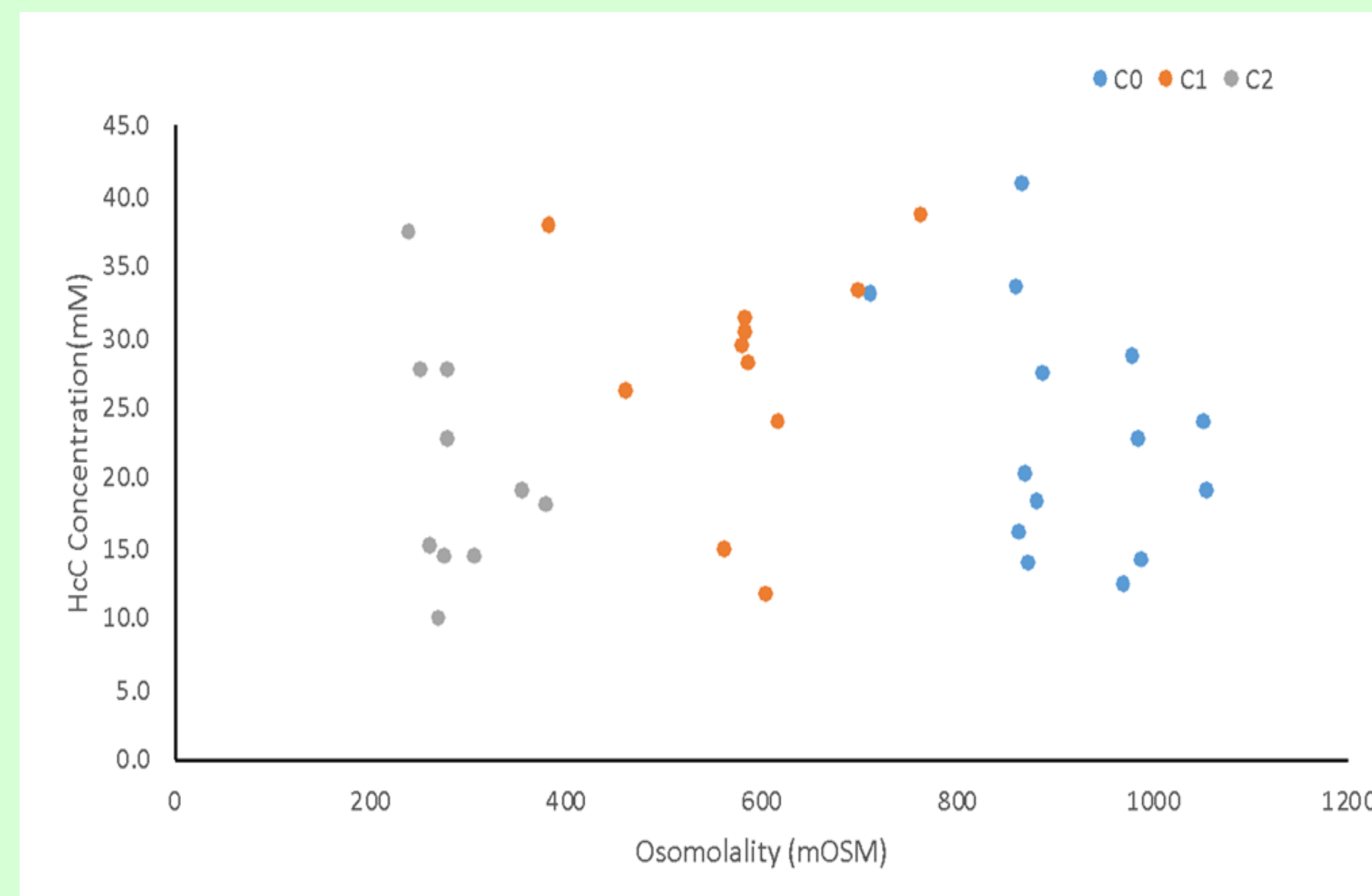


Figure 3. Concentrations of *C. sapidus* hemocyanin acclimated to varying salinities. C0, C1, C2 denote dilution iteration. No correlation was observed

## Results

Changes in the osmolality of crab hemolymph was noted after 24 hours of acclimation to a dilute solution (Figure 2). Hemolymph osmolality changed from a mean of  $935 \pm 70$  mOsm/kg to  $830 \pm 53$  mOsm/kg and to  $718 \pm 24$  mOsm/kg at successive dilutions. No further testing was conducted past the third dilution. These changes represent the crab's significant ability to osmoregulate ( $p < 0.05$ ).

Concentrations of hemocyanin were spectrophotometrically measured to determine any correlation between blood protein levels and environmental conditions (Figure 3). Absorbance data showed no significant correlation ( $p > 0.05$ ) between changes in ambient osmolality and hemocyanin concentrations.

## Discussion

The results of this experiment suggest that *C. sapidus* exhibit the ability to osmoregulate. Blue crabs typically inhabit estuaries that contain brackish waters of varying salinity. In order to survive in such diverse environments it would reason that the crabs have the capability to change their physiology accordingly which is supported by the significant changes in collected data. These results are like those found by Lovett et. al. (2006) that concluded that *C. sapidus* hyperosmoregulates in seawater concentrations below its isosmotic point.

Similarly, we correlate the crab's ability to osmoregulate with the lack of significance in changes to the concentrations of hemocyanin.

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