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Determining the physiological and behavioral aspects of salinity tolerance in the Asian clam, *Corbicula fluminea*

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ABSTRACT

The Asian clam, *Corbicula fluminea*, is an invasive bivalve that now occurs through most of the lower 48 United States. While a significant degree of salinity tolerance has been observed in *C. fluminea*, owing to its estuarine lineage, the physiological and behavioral responses to changes in salinity by these organisms are not completely understood. It was hypothesized that *Corbicula* would initially avoid any salinity stress behaviorally through valve closure, but would eventually have to open to dispel anaerobic waste products and deal with the salinity. To explore this, *Corbicula* were collected and put through a series of experiments at salinity exposures of 0, 2.5, and 5.0 g/L, with tissue water content and hemolymph osmolality being measured. After an initial 96-hour exposure, it was observed that the % tissue water content of clams in 2.5 g/L and 5.0 g/L water dropped 3.3% and 4.2%, respectively, below that of the control groups in 0 g/L at 84.2%. After a 24-hour time course experiment, this change in tissue water was found to largely occur within the first eight hours of exposure for the 2.5 g/L and 5.0 g/L groups. It was also noted that the hemolymph osmolality of both the 2.5 g/L and 5.0 g/L groups rose to approximately 78 mOsm/kg and 148 mOsm/kg, respectively, matching the osmolality of the water in roughly the same time span. The osmolality of the control group did not match the osmolality of the 0 g/L water at 0.5 mOsm/kg, but was held at a constant level around 50 mOsm/kg. In a later experiment measuring the same variables for clams in 10.0 g/L, it was found that the tissue water and osmolality did not begin to change significantly until after 12 hours. A context study was also conducted comparing oxygen consumption and % tissue water between various salinities in a light and dark exposure context. In this experiment, it was observed that clams held in salinities of 5.0 g/L consumed roughly 45.6 mg/L of dissolved oxygen per gram of tissue, whereas clams held in freshwater only consumed roughly 17 mg/L per gram of tissue. These findings suggest that *Corbicula* osmoregulate in purely freshwater but osmoconform at salinities of 2.5 g/L and higher. The data from the context study also suggests that this conformation comes at a significant metabolic cost. Furthermore, and in contrast to the results of some previous studies, a significant level of behavioral avoidance of elevated salinity does not appear to commence until the clams are at a salinity above 5 g/L.

INTRODUCTION

- The Asian clam, *Corbicula fluminea*, is an invasive freshwater bivalve mollusk inhabiting 46 states¹.
- Asian clams cause considerable damage to native mollusk populations and ecosystems and also have significant economic impacts³.
- Although *Corbicula* is a freshwater species, it has been observed in brackish environments. Studies have shown that *Corbicula* are capable of tolerating salinities up to 17g/L¹.
- Corbicula* osmoconform at salinities of 3.0g/L and above, but have been noted to behaviorally avoid salinities above certain levels through valve closure². However, the metabolic and physiological tradeoffs associated with prolonged salinity exposure are not completely understood.
- In initial time course experiments, clams held in the light were observed to conform at a slower rate than clams held in the dark, raising questions about how exposure context relates to salinity tolerance.
- This study seeks to understand the behavioral and physiological methods by which *Corbicula* tolerate elevated salinity levels and the role that light and dark exposure contexts play in salinity tolerance.



Right: Long-term exposure and time course experiments were carried out in isolated glass tanks to produce replicates.



Left: Pegged clams

METHODS

Long-term Exposure

- Clams were held in salinities of 0 g/L, 2.5 g/L, and 5.0 g/L for 96 hours, at which point tissue water content analysis was performed on each individual.
- Tissue water content was measured by dissecting the clams and taking the mass of wet tissue, then drying tissue to produce a water content ratio.
- Some clams were pegged open to eliminate capacity for behavioral avoidance.

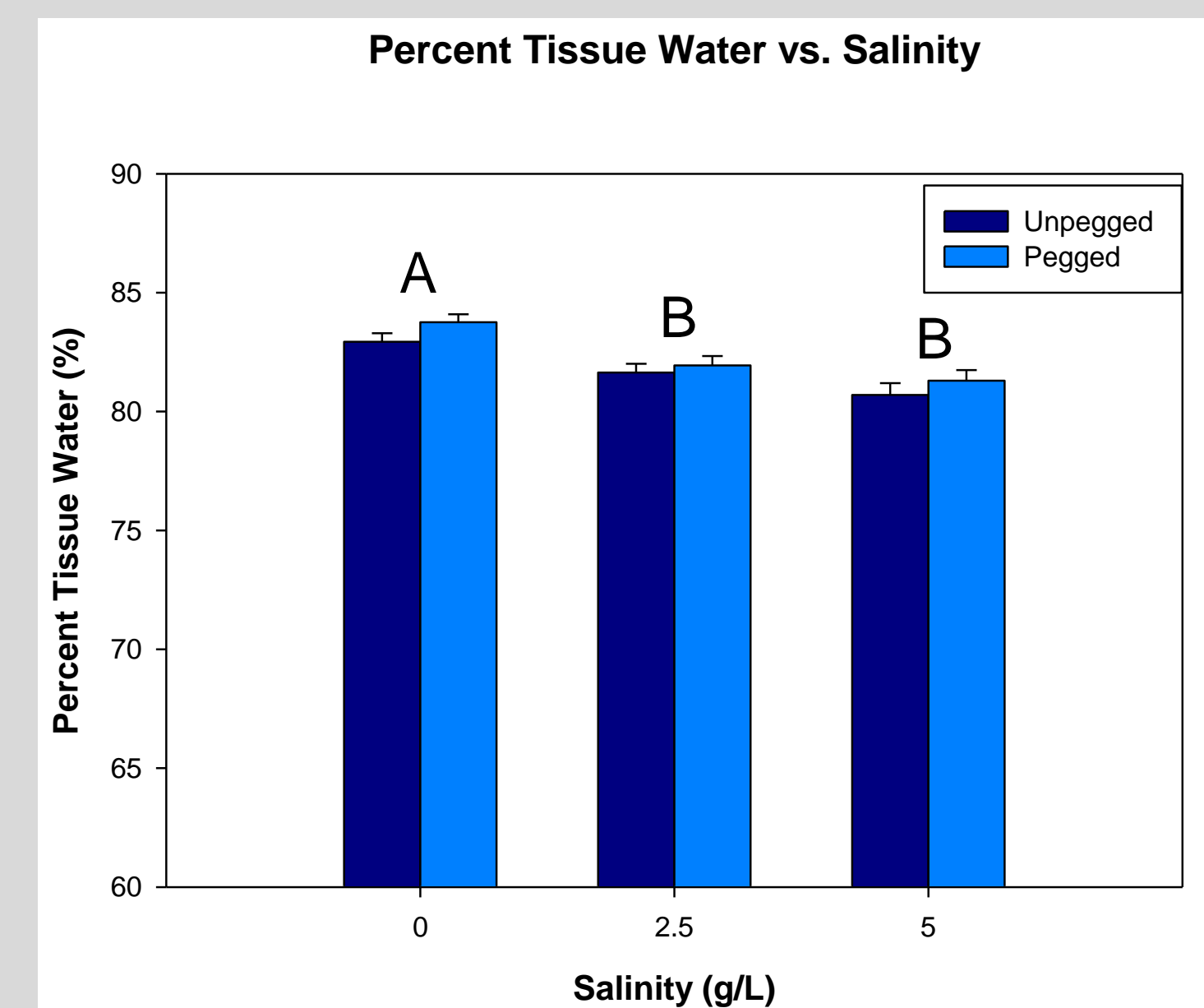
Time Course Exposure

- Clams were exposed to salinities of 0 g/L, 2.5 g/L, 5.0 g/L, and 10 g/L with individuals being sampled at 0, 4, 8, 12, and 24 hours to measure tissue water and hemolymph osmolality.
- Hemolymph osmolality was measured using a freezing point depression osmometer. Hemolymph was collected by cutting the adductor muscles of clams.

Exposure Context

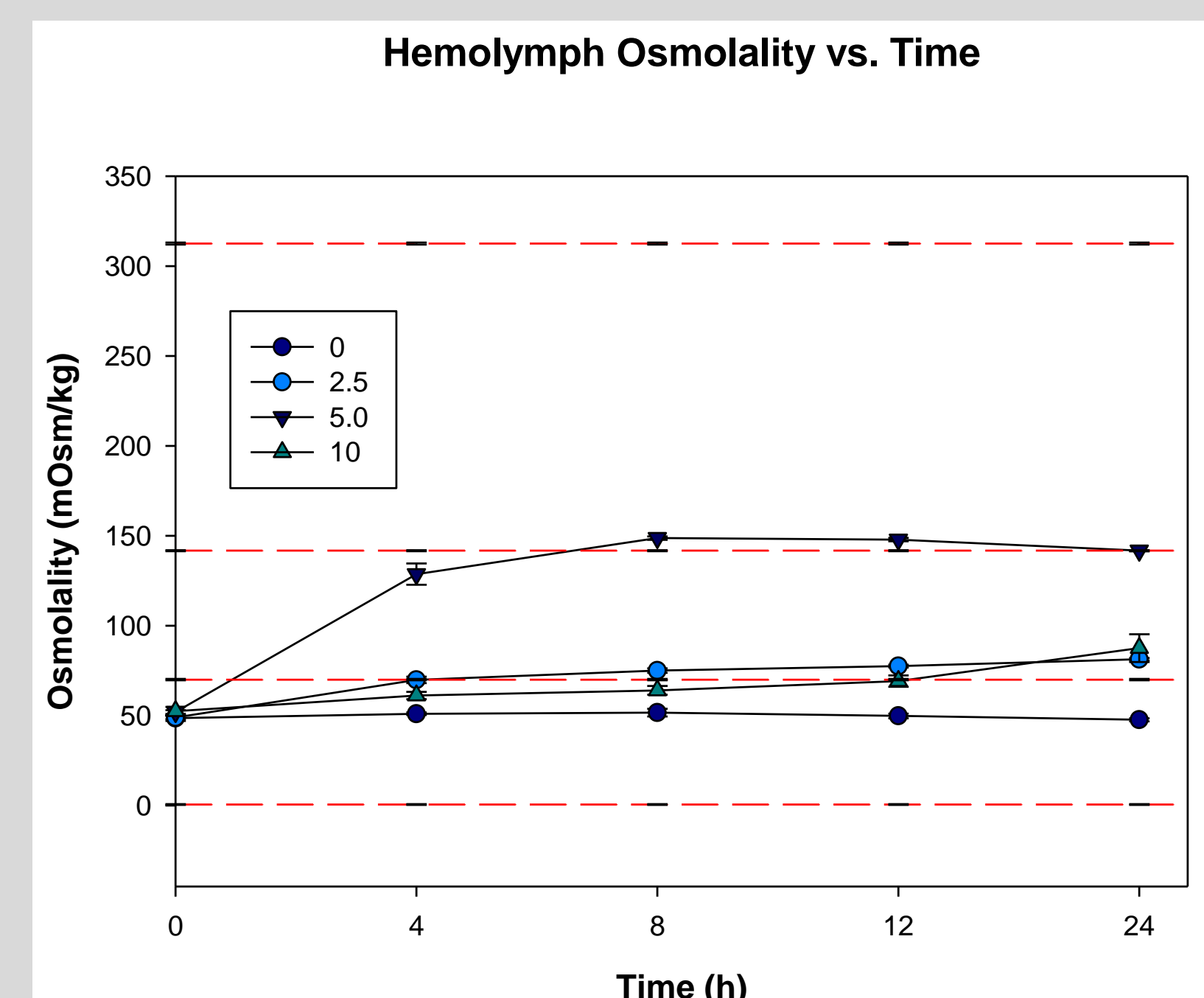
- Clams were exposed to salinities of 0 g/L and 5.0 g/L in a light and dark context.
- Clams were held in BOD bottles with initial and final dissolved oxygen being measured to monitor metabolic rate.

RESULTS

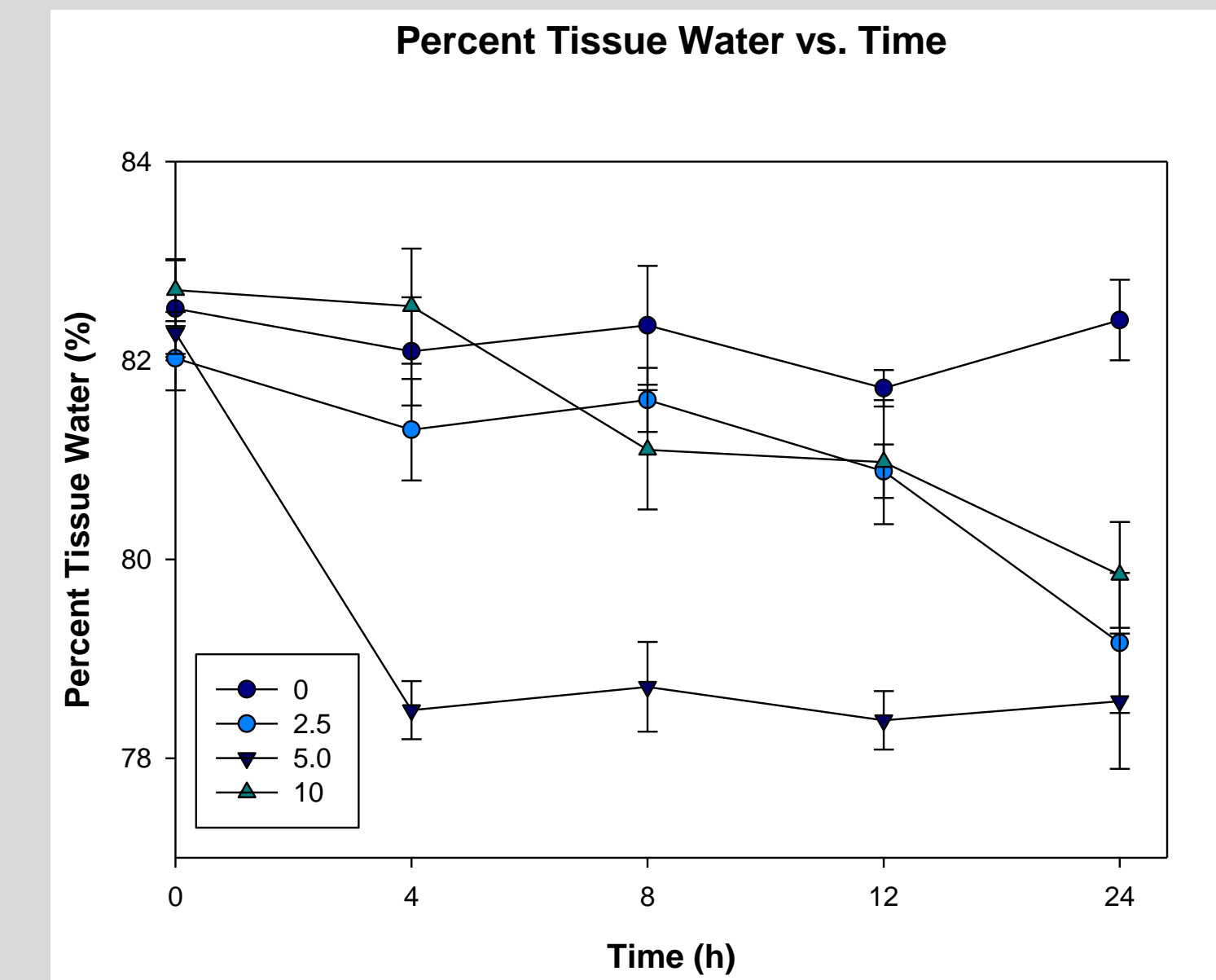


Left: Clams exposed to salinities of 2.5 g/L and 5.0 g/L for 96 hours showed significantly lower percent tissue water than control (Holm-Sidak Method, P<0.05). No significance was noted between pegged and unpegged treatments. Non-matched letters indicate a significant difference.

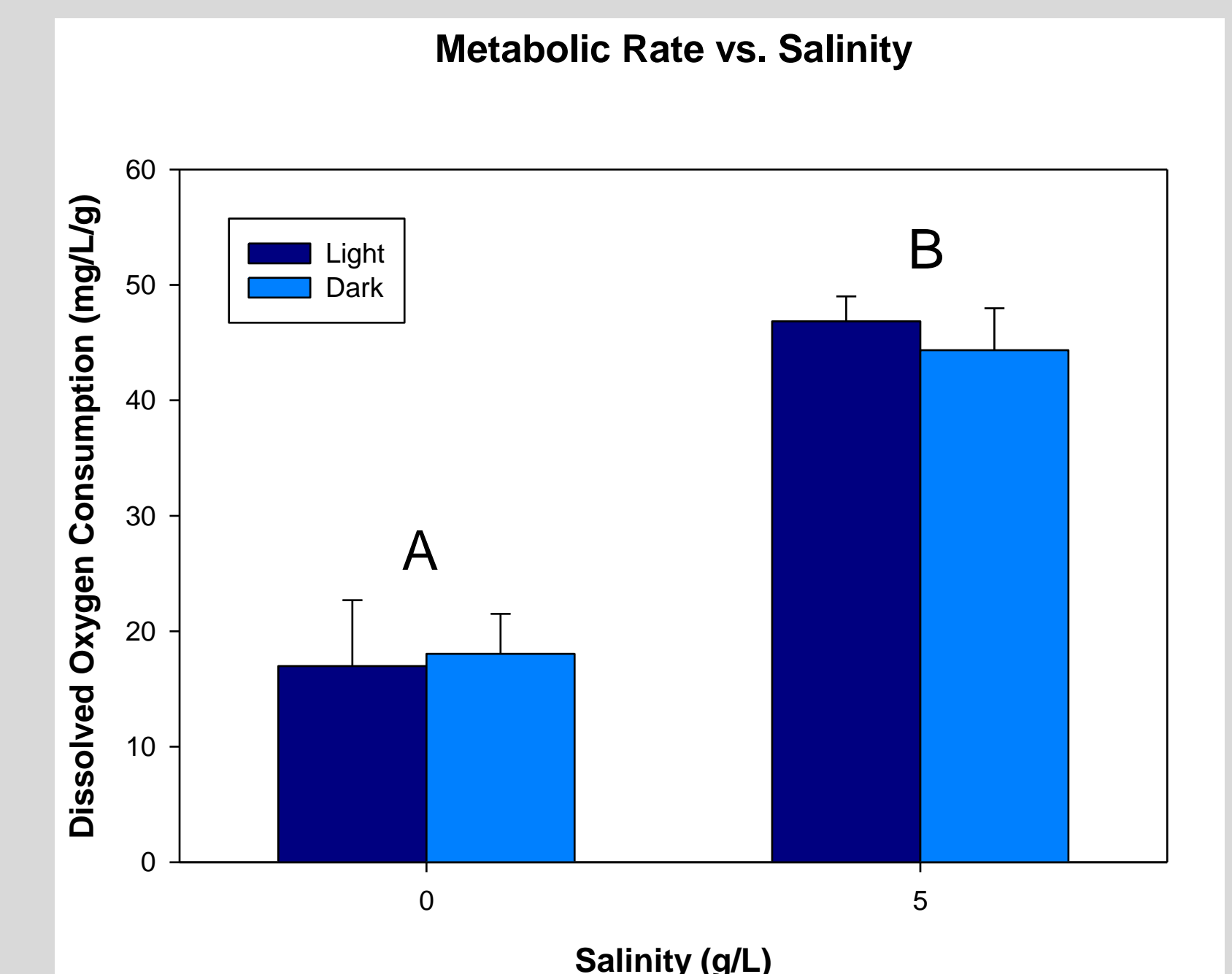
Below: Clams exposed to 2.5 g/L and 5.0 g/L showed significant increase in hemolymph osmolality within the first 8 hours of exposure, completely conforming to the osmolality of the water within this time. Clams held in 10 g/L showed significant increase over the whole exposure period, but still had not fully conformed after 24 hours. Red dashed lines indicate osmolality of treatment water.



Right: For clams exposed to 5.0 g/L, significant decreases in percent tissue water occurred within the first four hours of exposure. Clams held in 2.5 g/L and 10 g/L did not show any significant decrease in percent tissue water until 24 hours.



Below: Clams exposed to 5.0 g/L displayed a significantly elevated metabolic rate over control after 24 hours of exposure (Holm-Sidak Method, P<0.05). No significant difference in metabolic rate was observed between clams held in the light and those held in the dark.



CONCLUSIONS

- Corbicula* osmoregulate in freshwater, but osmoconform in salinities of 2.5 g/L and greater.
- Corbicula* quickly conform to salinities of 2.5 g/L and 5.0 g/L, but behaviorally avoid 10 g/L salinity, indicating that the physiological costs of avoidance outweigh those of conformation at salinities up to 5.0 g/L.
- Conformation comes at a significant metabolic price, raising questions about the nature of the physiological tradeoffs that these clams face in tolerating elevated salinities.

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