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for Science Students
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Determining the Thermal Tolerance of *Littoraria ardouiniana*

ABSTRACT

This project will discuss how individual thermal physiology enables intertidal gastropods to persist in extreme tropical shores, with particular focus on the species *Littoraria ardouiniana*, from the family Littorinidae. Thermal performance (heart rate vs body temperature) of replicate individuals were plotted to observe their patterns with temperature. In ectotherms, the heart rate should increase with temperature until it reaches a break point called the Arrhenius Break Point Temperature (ABT) and finally dies (zero heart rate = Flatline Temp, FLT). The findings of this project suggest this species of tropical ectotherm may be vulnerable to high heat stress as its thermal tolerance limit is close to the average maximum temperature recorded by temperature loggers deployed in their habitats (mangroves).

INTRODUCTION

Supratidal zones like mangroves (see fig. 1) set forth a range of stressful environmental conditions for marine species living in the area. Conditions such as high temperatures, fluctuating salinity, periodic wet and dry environments, and alternating aerobic and anaerobic conditions ("Mangrove", n.d.). Moreover, tropical shore species are also regarded as especially vulnerable to environmental changes, as their body temperatures, and consequently physiological performances differ keenly with environmental conditions (Marshall et al. 2015).

Figure 1. Mangrove in Tung Chung where specimens were collected

Dong and Williams (2011) concluded that when temperatures exceed the thermal tolerance of these species, a deterioration in cardiac performance and even death is noted. Thus, determining the thermal tolerance of these ectothermic species is important, as it can indicate the thermal environments they can tolerate but also explain why organisms exhibit certain behaviours. Additionally, it appears that for survival in the supratidal zone, it is critical for such organisms to have adaptations to temperature variation (Somero 2002). Therefore, this study aims to investigate the upper limits of thermal tolerance presented by *Littoraria ardouiniana*.

Littoraria ardouiniana on a shrub leaf

MATERIALS AND METHODS

Animal collection

Animals were collected from a mangrove in Tung Chung and maintained in simulated natural conditions in the HOC first-floor aquarium (fig. 2) until assayed. Additionally, the temperature of the organism's habitats (mangrove) were measured using temperature loggers, which were placed at the top and bottom of the shrub branches (figs. 3&4).

Figs 3 & 4. ibutton temperature loggers on branches at the top and bottom of shrub

Heart Rate assays

Heart rate performance was used as an indicator of thermal performance in the species, as it has been shown to be linked to metabolic performance. Following the methods in Burnett at al., (2003) using Blue-Tac or super glue, an infrared sensor was attached onto the organism, directly over the heart, through which variations in the current (light-dependent) generated by the heartbeat was amplified, filtered, and recorded using a PicoScope and Laptop PC (see Burnett et al., 2003). Data was analysed using a customized R-script program.

The organism's heart rate was measured with rising temperatures as it was placed in a beaker, which was then immersed into a water bath (fig.5). This allowed the air temperature in the beaker to be heated up at an ecologically relevant rate (to simulate warming on the shore when the animals are emersed) until the heartbeat was lost.

Fig 2. Snails placed in a tank with branches and leaves from mangrove shrubs

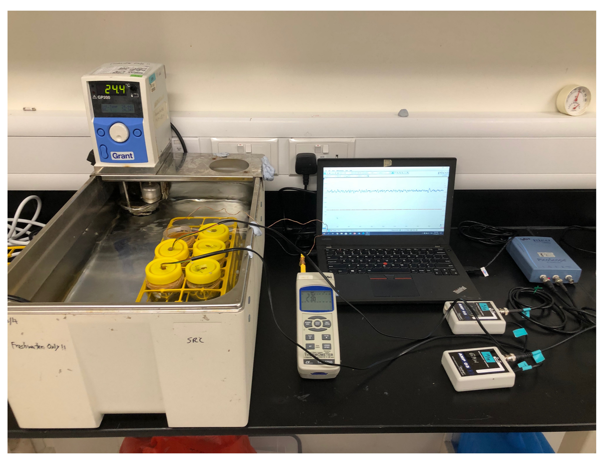


Fig 5. Experiment set-up. From left to right: water bath, beakers in rack, temperature logger with thermocouple, laptop, heartbeat sensors, amplifiers, PicoScope.

Littoraria ardouiniana emerging from their shells after being sprayed with salt water.

RESULTS

The data is separated into three categories: active, moderately active, and inactive, which delineate the activity of the snails at the time of experiment. The snails in each category are also distinguished by the day the experiment was conducted, i.e. "23-06 A" is the first snail in the batch of four which were part of the experiment on 23rd June.

Regarding cardiac performance, all of the snails displayed similar behaviour—they had regular heart beats and heart rates increased in a log-linear fashion with temperature. As the temperature continued to rise, the snails suffered from arrhythmias, thus leading their heart rates to decrease abruptly.

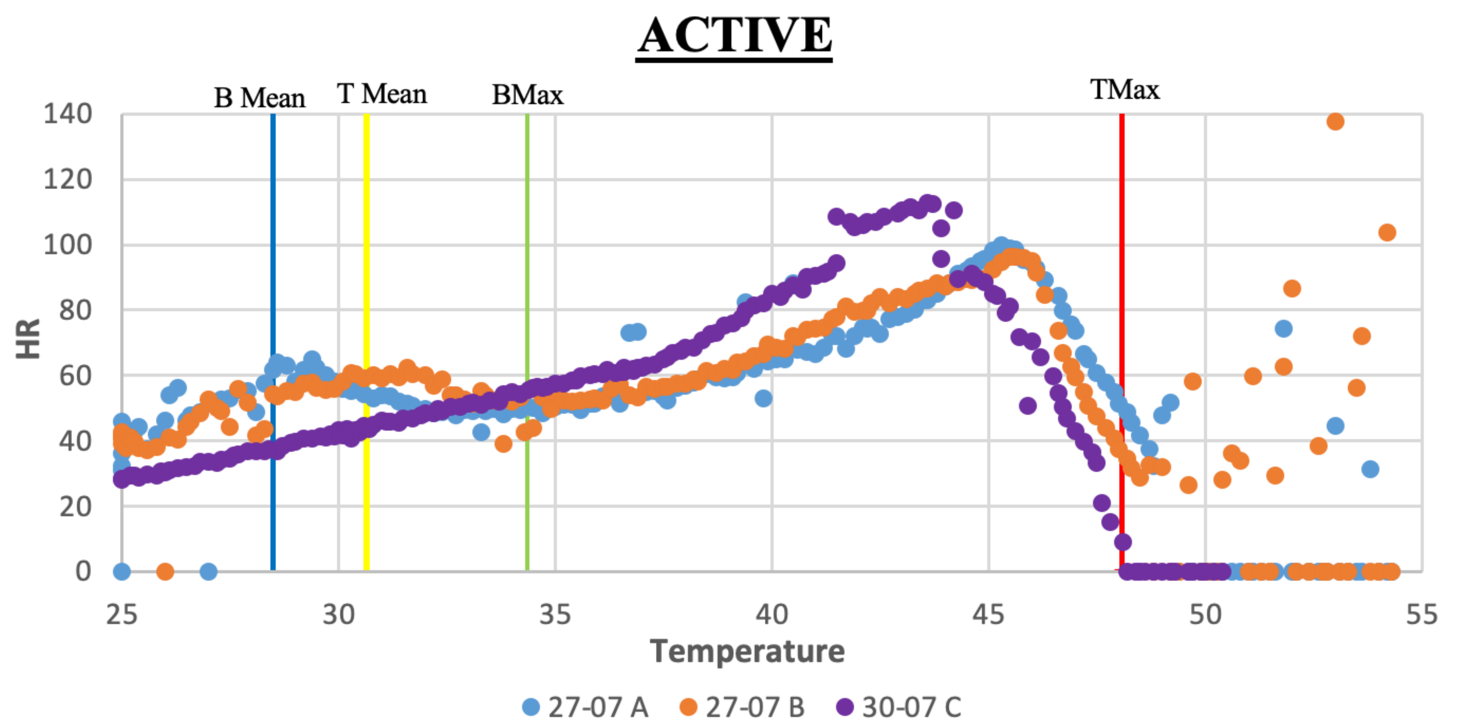


Fig 6. Thermal performance of three active snails (HR vs Body Temp.)

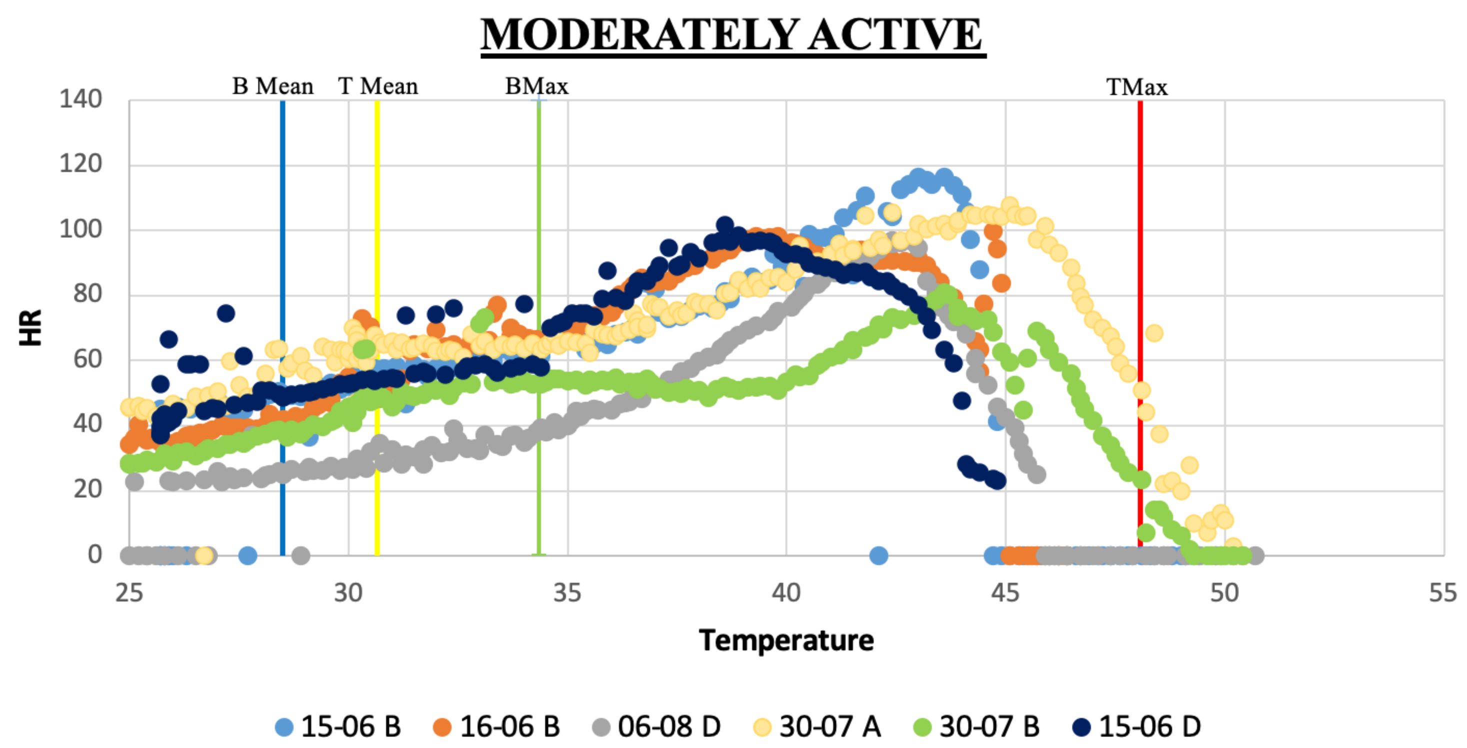


Fig 7. Thermal performance of six moderately active snails (HR vs Body Temp.)

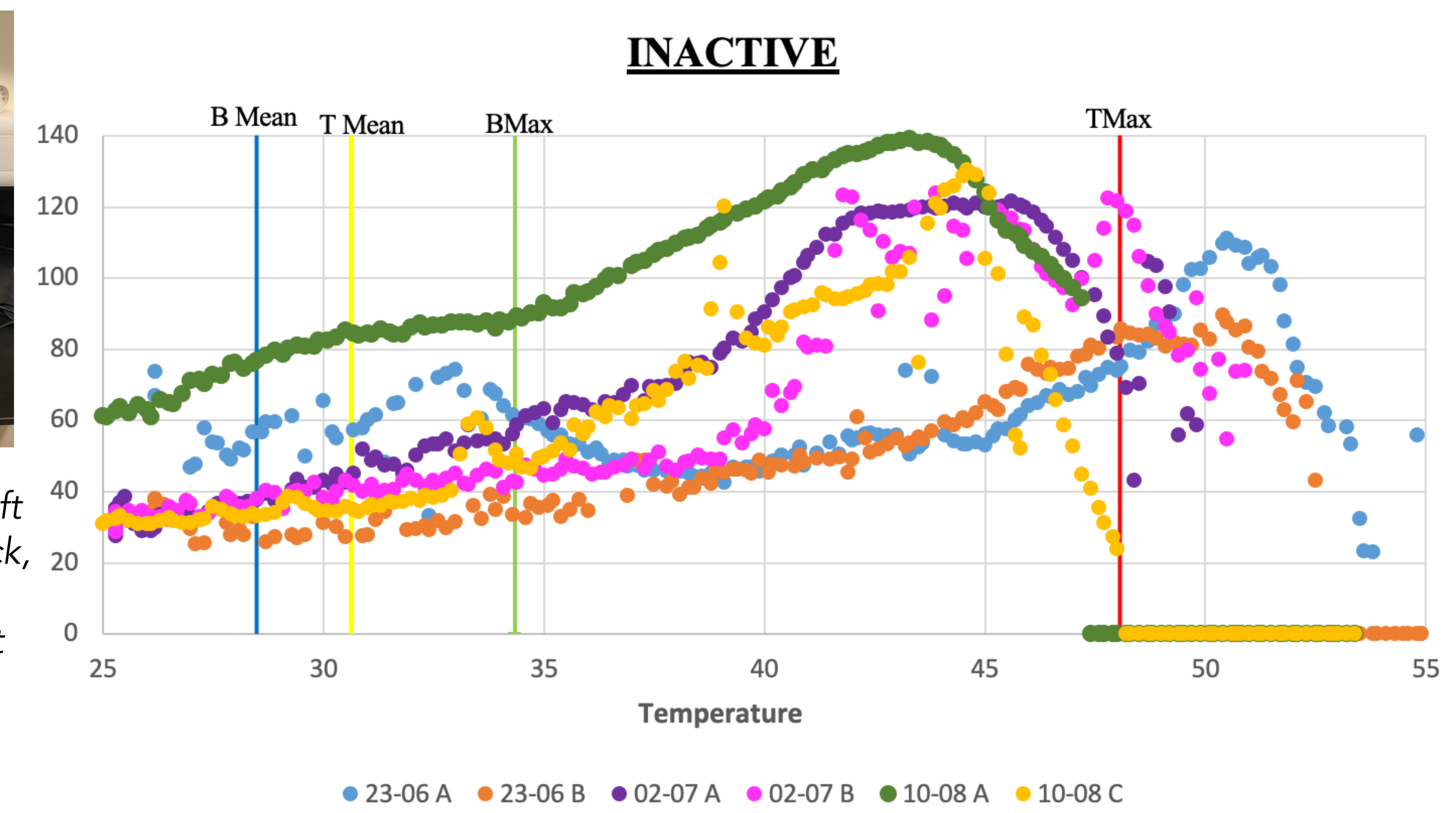
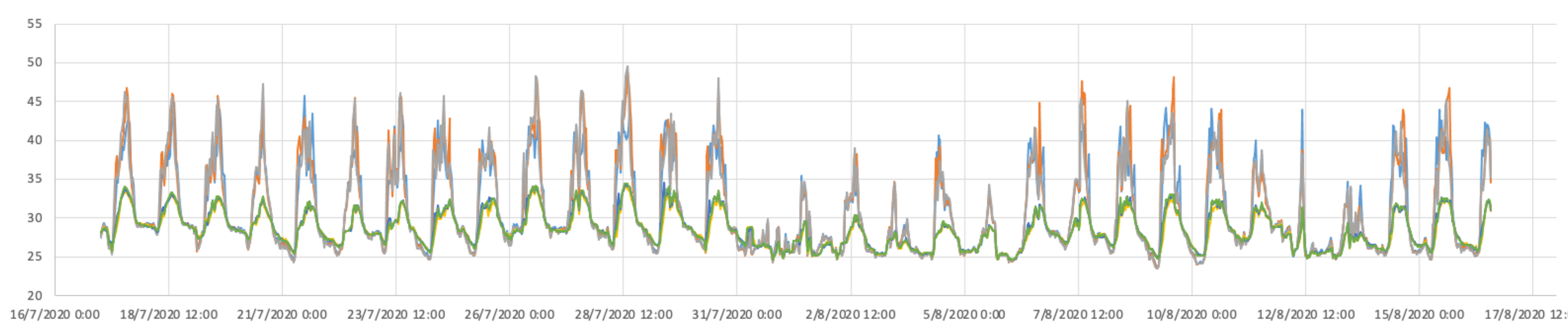


Fig 8. Thermal performance of six inactive snails (HR vs Body Temp.)



Data from ibutton temperature loggers from 16/07 – 17/08; logging temperature of mangrove surroundings where Littoraria ardouiniana live.

DISCUSSION

Living in a supratidal zone, *Littoraria ardouiniana* encounters different thermal stresses. Figures 6,7, & 8 suggest the snails can, on average, withstand temperatures much higher than those registered by the top (TMean) and bottom (BMean) temperature loggers. Although, it can be seen that a majority of the snails would be unable to withstand the maximum temperature registered by the top temp. logger (TMax), which was ~48.1°C. Amongst the active snails, they could still maintain normal cardiac function until their internal temperatures reached an average of ~44.8°C, whereas for the moderately active and inactive snails, this was at ~42.2°C and ~47°C respectively.

The lower average ABT of the active snails suggests, in comparison to the other two categories, an earlier impediment of aerobic performance and oxygen transportation as a result of the heart's incapacity to function past this critical temperature. As dictated by Marshall et al. (2015), snails are susceptible to desiccation during active movement and choose instead to retract into their shell and stay inactive during high and possibly taxing surrounding temperatures. Thus, when they are inactive, they can withstand a more varied range of temperatures.

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