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Damsels in Distress: A study of the effect of temperature on damselfly competitive interactions

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**Damsels in Distress: A study of the effect of temperature on damselfly
competitive interactions**

An Honors Thesis submitted in partial fulfillment of the requirements for Honors
Studies in Biology

By

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Spring 2016

Biology

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Abstract:

Temperature has a fundamental role in shaping numerous physiological aspects of organisms, such as growth rate, that determine an organism's performance. Due to global climate change, temperature is projected to keep increasing. As temperature increases, so does an organism's growth rate. Also, as competition increases organisms' growth rate decreases. While studies have examined these factors individually, no experiments have assessed the combined impact of temperature and competition on organism's growth rates. The experiment we conducted manipulated temperature and density in order to project what will happen as a result of rising temperatures.

Introduction:

Climate has a fundamental role in shaping numerous physiological aspects (i.e., metabolism, digestion efficiency, growth rate) that determine a species demographic performance in a local environment and at global scales (Andrewartha and Birch 1954, Angilletta et al. 2004). It is widely recognized that climate is currently changing much faster than many historical records and proxies have shown (Fasullo and Trenberth 2012). An important aspect of climate change is temperature increase. Temperature has increased by .74 °C on average from 1906-2005, and is projected to continue increasing between 1.1 °C to 6.4 °C by the end of the 21st century (IPCC 2007). Even a slight change in temperature can have dramatic impacts on organisms (Kingsolver et al. 2008). For example, temperature may be one of the most important factors that causes climate-related extinctions (Cahill et al. 2012, Sinervo et al. 2010). Freshwater organisms are particularly vulnerable to rising temperature (Heino, Virkkala and Toivonen 2009) via the effects that temperature has on their ability to grow.

Although many factors may affect the growth rates of organisms, temperature is thought to be a major determinant (Angilletta et al. 2004). Growth rate generally increases as temperature increases to an optima (Nilsson-Örtman, Stoks and Johansson 2014). This trend will continue until the temperature is too extreme for the organism to survive (Kingsolver et al. 2008).

As growth rate of organisms' increase, so too does the demand for food, which can increase competition within the species. Consequently, another factor that influences growth rate is density because it effects intraspecific competition (competition between individuals of the same species). There is more competition at

higher densities because of reduced resource availability or increased interference for resources among conspecifics. This results in reduced growth rates and increased mortality rates (Crowley et al. 1987). Density-dependent competition can be assessed by observing a decrease in both growth rate and survival rate with an increase in density (Anholt 1990). Cannibalism is an extreme form of intraspecific competition that is often density and temperature dependent (Crumrine 2010). Organisms often kill others of the same species but do not consume them (Fincke 1994).

While the effects of temperature and competition can both affect growth rates, it is unclear how these two factors may jointly affect growth rates (Lang et al. 2011, Pickup and Thompson 1990). That is, it may be that increases in temperature accentuate or minimize the effects of increased density. For example, if the effects of temperature cause the strength of density-dependence in growth rate to increase, the result is that warmer climates may result in populations being much smaller. This may result in populations being further at risk for extinction via elevated demographic stochasticity because of their small population sizes. Necessary to investigate these possibilities are studies that document the joint contributions of temperature and density on organismal growth rates.

Damselflies are ideal organisms to investigate the impact of rising temperature and its effect on density-dependent growth. Damselfly larvae live in shallow regions of freshwater that are susceptible to the effects of climate change (Hassall 2013). Since their habitat is shallow water, it is more prone to evaporation. As a result, damselfly density would increase which could alter intraspecific competition. We designed an experiment to explore the effect of temperature on intraspecific competition of

damselfly larvae by assessing growth rate and mortality rate of damselflies at different temperatures and densities. We predicted that as temperature increases competitive interactions will also increase and the growth rate will decrease. This trend will continue until a temperature that is too extreme for damselfly survival, thus competition and growth rates will decrease where as mortality rates will increase. This experiment is important for determining what will happen as organisms experience the myriad effects of climate change.

Methods:

To examine how the strength of competitive interactions and increased temperatures relate to influence damselfly performance, we conducted an experiment where density with temperature was manipulated. We used *E. divagans* for this experiment because it is one of the most common and widespread species of damselfly found in my study area. Samples of *E. divagans* were obtained from Wilson Lake (Fayetteville, AR) and then returned to the lab during November 2016 when most larvae were in 6th or 7th instar.

We used growth rates as a component of performance. We estimated growth rates as the difference in body mass over the course of the experimental period, which was 14 days. Because individuals lose their exoskeleton as they grow, individuals cannot be marked. Thus, we determined the mass of each replicate of each experimental treatment: 1, 2, or 4 damselflies and used the mean to estimate individual growth rate. Damselflies were gently blotted to remove surface water and then weighed to the nearest mg to obtain their initial and final masses.

We performed the experiment in a walk-in environmental chamber set at 20°C on a 12-hour light and dark cycle. Damselflies were housed in test tubes that were secured to a styrofoam float placed in a 37.85-liter aquarium filled with dechlorinated tap water. Each test tube also contained a wooden stick as a perch for damselflies (Figure 1). Water from the aquarium did not circulate into the test tubes. Test tubes were arranged in five groups and each group had a set of four test tubes that received one, two and four damselflies per tube (Figure 2). Aquaria were randomly assigned to one of five temperatures: 20, 22, 24, 26, 28. Heaters in each aquarium controlled the temperature. Each combination of temperature and density was replicated four times. The damselflies were haphazardly assigned to experimental tubes.

Six days per week, damselfly-holding tubes were cleaned by removing 5 ml of water from the bottom of the tube (where waste matter had accumulated) and replenished by adding 5 ml of dechlorinated clean water. The clean water contained a food supply—*Artemia* brine shrimp. Water levels in each aquarium were monitored and fresh water was added to the aquariums when needed in order to maintain a constant water level in all of the aquariums. Each day we recorded the temperature and condition of the damselflies.

The experiment was designed to run for four weeks, but it was terminated at two weeks because of excessive mortality. After we ended the experiment and returned the surviving damselflies to the lab, we measured the final density and final mass. To measure per capita growth rate per tube we used the equation: $(\ln(\text{mass final mg/n}) - \ln(\text{mass initial mg/n})) / \text{duration}$. The duration was 14 days. To measure mortality rate we used the equation: $(-\ln(\text{final density}) - \ln(\text{initial density})) / \text{duration}$



Figure 1: damselflies resting on a perch in a test tube

Photo credit: Dr. Adam Siepielski



Figure 2: 5 aquaria set up in environmental chamber

Results:

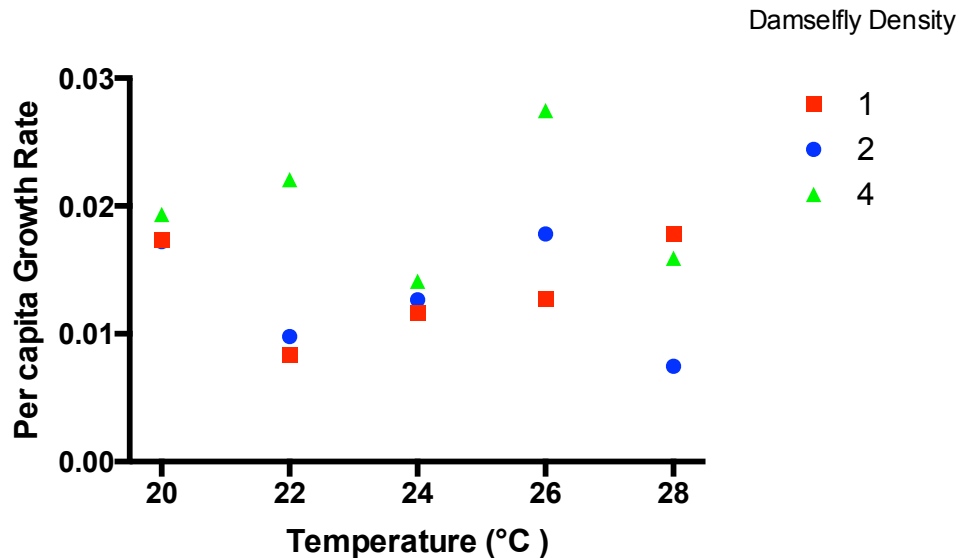


Figure 3. Mean damselfly per capita growth rates in relation to density and temperature. Colors correspond to different damselfly densities.

Effects of density and temperature on growth rates

My experiment did not provide a clear correlation between growth rate, temperature and damselfly density (Figure 3). Given the unexpectedly high mortality damselflies experienced during the experiment, the results obtained precluded firm conclusions. The lack of replication and resulting highly unbalanced design also prevented the use of a statistical hypothesis-testing framework. Instead, I present some of the results that hint at possible patterns that need to be further explored in a new experiment.

Density 4 tubes experienced the most growth, with the exception of the tubes at 28°C. Density 2 tubes started with a high growth rate at 20°C and decreased at 22°C and then continued to steadily increase until dropping at 28°C. Density 1 tubes followed the

same pattern as density 2 until 28°C and the growth rate dropped. Density 1 tubes experienced the lowest overall growth rate, except at 28°C. The growth rates for all the densities were most similar at 20°C and 24°C. Thus, overall there is no clear association between density and temperature. Further studies are needed to test the hypothesis we developed.

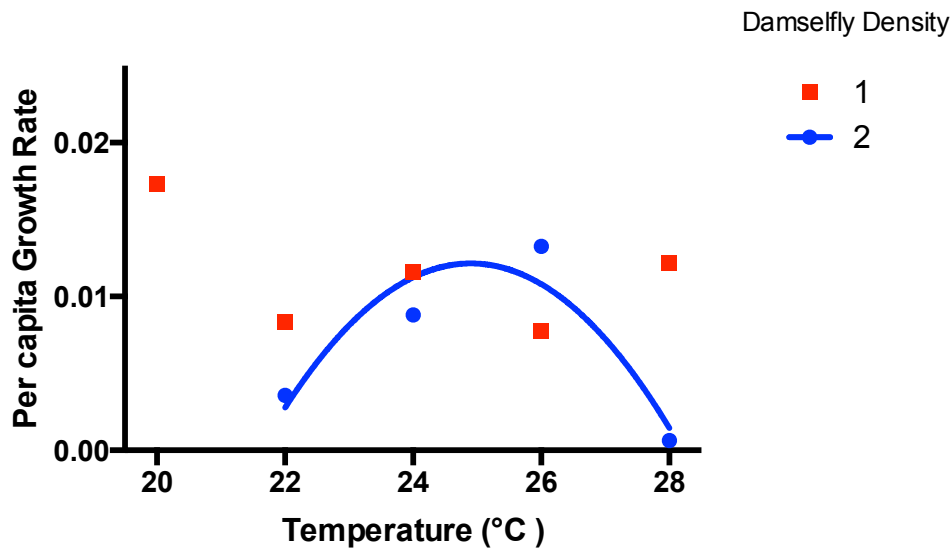


Figure 4. Damsselfly per capita mean growth rates in relation to density and temperature excluding tubes with mortality. Colors correspond to different damsselfly density. Density 4 is not accounted for because there was mortality in every tube. There is a quadratic fit to the density 2 treatments.

Effects of density and temperature on growth rates with no mortality

Figure 4 shows the per capita growth rate without including the tubes that had mortality. Density 4 tubes had mortality at all temperatures, so it was not included. Density 1 tubes were the only treatment that had at least one tube with no mortality at all temperatures. Density 1 had the highest growth rate at 20°C and similar growth rate at other temperatures. Density 2 had a steady increase in growth rate between 22-26°C

and then minimal growth at 28°C. Because of this trend I included a quadratic fit to the density 2 treatments. Thus, overall these results suggest that increasing density may result in an intermediate temperature (24-26°C) resulting in maximal growth rates, whereas treatments with no density effect cause growth rates to linearly decline. This pattern for the single individual density treatment is unexpected. It is necessary to do additional experiments to further examine this relationship.

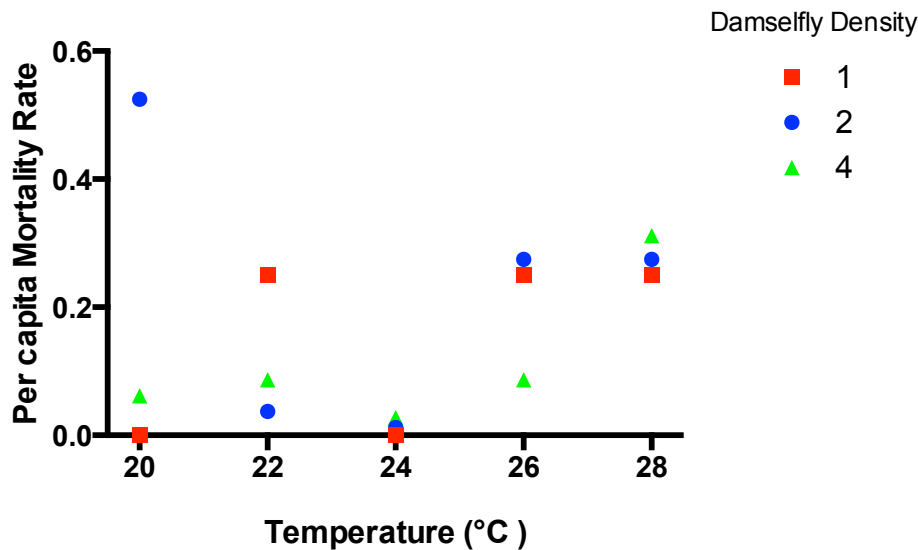


Figure 5. Mean damselfly per capita mortality rates in relation to density and temperature. Colors correspond to different damselfly density.

Effects of density and temperature on mortality rates

Per capita mortality was largely invariant to temperature and density, although it does appear that an intermediate temperature (24°C) resulted in the lowest consistent mortality across densities (Figure 5). Density 2 had the highest overall mortality rate at all temperatures except 22°C and 28°C. Density 4 had roughly similar mortality rates as density 2 treatments at 20-28°C and the highest overall mortality at 28°C. Density 1 had lowest mortality rate at 20°C and 24°C and the same mortality rate at 22, 26 and 28°C.

All three densities had the lowest mortality rate at 24°C, therefore making 24°C the optimal temperature. All 3 densities had high mortality at 28°C, thus making it not an ideal temperature for damselfly survival.

Discussion:

Results of the experiment did not allow me to determine if there was an effect of temperature on the strength of damselfly intraspecific competitive interactions. Overall, I found no consistent effects of either temperature or density on per capita growth rates. Other studies have found that both of these factors can individually be important in affecting damselfly growth rates: Growth rate generally increases as temperature increases (Nilsson-Örtman, Stoks and Johansson 2014) and growth rate generally decreases and density increases (Crowley et al. 1987).

However, one important result can be seen in Figure 4 (average mean growth rate without mortality). Damselfly density 2 growth rate steadily increases from 22°C-26°C and then drops at 28°C. The optimal temperature for growth is between 24 and 26°C. This pattern contrasts with the single density treatment that showed a consistent decline in growth rates with an increase in temperature. This suggests that increasing the strength of competition can modify the effect of temperature on growth rates. However, caution is required in interpreting this pattern given the unexpectedly high mortality observed.

Another important result can be seen in Figure 5 (mean per capita mortality rate). Although per capita mortality rate was generally invariant to temperature and density, mortality rates at temperatures of 24°C and 28°C were similar among densities.

At 24°C, all three densities have the lowest mortality. Thus, suggesting 24°C the optimal temperature for damselfly survival regardless of density. At 28°C, all 3 densities had high mortality, therefore making it an unsuitable temperature for damselfly survival irrespective of density. However, 28°C was not too extreme of a temperature because there were still surviving damselflies. An important factor to consider in mortality rate is cannibalism, which is often density and temperature dependent (Crumrine 2010). During the experiment I noticed evidence of cannibalism especially in the higher density tubes. This was evident when I saw no carcass or part of a damselfly carcass remaining at the bottom of the test tube because often organisms will kill but not consume (Fincke 1994). However, it is impossible to accurately determine how many deaths were results of cannibalism because it was possible for the damselflies to escape their test tube.

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