Short notes

Interactive effects of food availability and temperature on wood frog (*Rana sylvatica*) tadpoles

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Temperature and food availability can affect the growth and survivorship of ectothermic vertebrates. We examined how the interaction of temperature and food availability influenced the growth and survival of wood frog (*Rana sylvatica*) tadpoles. Tadpoles experiencing 17 °C survived better than tadpoles experiencing 25 °C. The difference in survivorship between temperature treatments tended to be highest at the lowest food level. Tadpoles in the high food treatment were significantly heavier than tadpoles in other treatments. Tadpoles in the 25 °C treatment were larger than tadpoles in the 17 °C treatment. Tadpoles were substantially heavier in the high food, warm temperature treatment combination than in other treatments. Our results suggest the effects of temperature change on amphibian populations could be mediated by food availability.

Key words: food, growth, survivorship, temperature change

For anuran tadpoles, temperature and food availability can influence a variety of traits. However, the effects can vary among species, or even among populations of the same species. Increased temperature can decrease tadpole or metamorph size in some species (e.g. Álvarez & Nicieza, 2002a,b; Watkins & Vraspir, 2006; Orizaola & Laurila, 2009), whereas other species grow faster at higher temperatures (e.g. Anderson et al., 2001; Arendt & Hoang, 2005; Sanuy et al., 2008). Increased temperature can change developmental rates (e.g. Joly et al., 2005; Sanuy et al., 2008), external morphology (Jorgensen & Sheil, 2008) and gut morphology (Castañeda et al., 2006). Increasing levels of food availability typically increase tadpole growth and mass at metamorphosis, decreasing the time to metamorphosis (e.g. Murray, 1990; Arendt & Hoang, 2005; Peacor & Pfister, 2006).

Few studies have examined how temperature and food availability can interact to affect tadpoles. Laugen et al. (2005) provided unanalysed results (their Table 1) that suggest that in *Rana temporaria*, growth rates of tadpoles are lower at restricted compared to *ad libitum* food levels. However, their results also suggest an interaction between temperature and food availability: depending on the population, a decrease or an increase in growth rate at higher temperatures and restricted food level were observed, although at *ad libitum* food levels all populations showed faster growth rates at higher temperatures (Laugen et al., 2005). Newman (1998) found that changes in food levels, and the timing of food level changes, interact with temperature to influence the time to metamorphosis and metamorph size in *Scaphiopus couchii*. Knowledge of such an interaction may be helpful in understanding geographical variation in tadpole growth rates and size at metamorphosis, and might explain how changes forecast as a result of global climate change may affect amphibian performance, especially if such temperature changes might also affect food levels.

We examined how temperature and food availability interact to influence individual growth and survivorship of wood frog (*Rana sylvatica*) tadpoles. Wood frogs breed in early spring (see Redmer & Trauth, 2005) and thus their tadpoles probably experience relatively cool temperatures (water temperatures in early April for the natal pond for the tadpoles used in this experiment averaged 13.7 °C; Dougherty et al., 2005). Mean preferred temperatures of wood frog tadpoles from Connecticut ranged from 17.6 to 20.4 °C depending on whether the tadpoles were from closed or open canopy ponds (Freidenberg & Skelly, 2004). However, wood frog tadpoles from Ohio can tolerate relatively higher temperatures compared to other areas to the north (Manis & Claussen, 1986).

We hypothesized that growth rates and survivorship would be higher at higher food levels and at warmer temperatures. We also hypothesized that the increase in growth due to temperature would be higher at the higher food levels, which could support both the higher metabolic rate and the higher growth rate found at higher temperatures (see Lindgren & Laurila, 2009).

We collected six wood frog egg masses from a local pond on 27 March 2008. Egg masses were maintained in two large plastic containers in the lab and allowed to hatch. Within each container, hatchlings and tadpoles from multiple egg masses were allowed to mix. For the experiment we created two temperature treatments. One treatment maintained a mean temperature of 17±1 °C (laboratory), and the other maintained a mean temperature of 25±2 °C (greenhouse). Temperatures were maintained via a thermostat. Both treatments were maintained on a 12 h light: 12 h dark photoperiod. We used three food level treatments: low food level (0.0025 g of ground Purina Rabbit Chow per individual), medium food level (0.005 g per individual) and high food level (0.01 g per individual). Food levels were based on Petranka et al. (1994). We used a 2 × 3 fully factorial experiment with eight replicates per treatment combination (a total of 48 experimental units; however, one cooler temperature, high food container was accidentally overfed and was excluded from the analyses). Each experimental unit consisted of a container (21 cm × 14 cm × 5 cm) with five haphazardly chosen tadpoles

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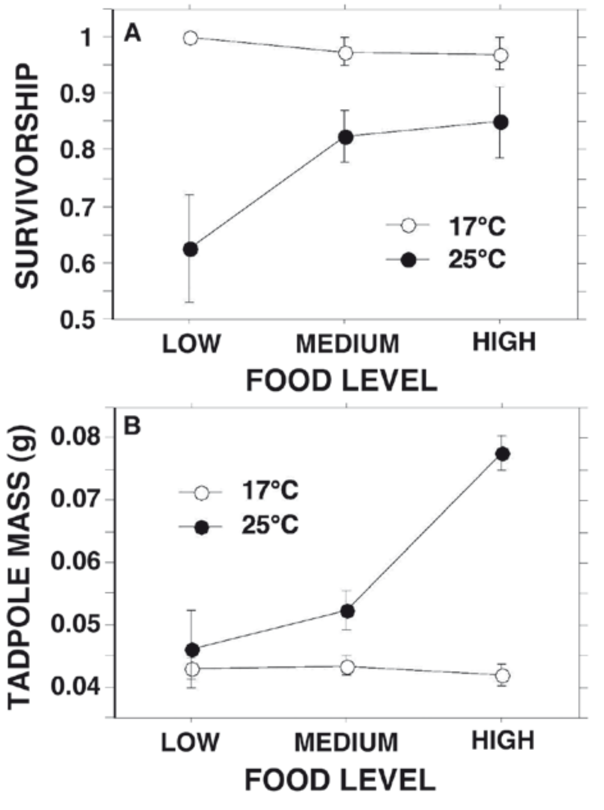

Fig. 1. The effects of the interaction of food level and temperature treatment on A) survival and B) tadpole mass at the end of the experiment on wood frog tadpoles (*Rana sylvatica*). Means are given ± 1 SE.

(Gosner stage 25; Gosner, 1960). We fed tadpoles every third day. At each feeding, water level was refreshed and excess food and faeces were removed from the container. We also rotated the location of the containers to minimize any effects of undetected gradients (e.g. light, temperature, etc.). Eleven days after the tadpoles were introduced into the experiment, we recorded the number of tadpoles surviving and weighed the surviving tadpoles to the nearest 0.001 g using an electronic balance after blotting the tadpoles dry to obtain a mean tadpole mass for each container. We analysed the effects of temperature and food level on tadpole final mass and survivorship (arcsine-square root transformed) using separate univariate ANOVAs for each dependent variable.

Survivorship was not significantly affected by food level (Fig. 1A; $F_{2,41}$=1.28, P=0.29). Tadpoles experiencing 25 °C had significantly lower survivorship than tadpoles experiencing 17 °C (Fig. 1A; $F_{1,41}$=30.61, P<0.01). The interaction of food level and temperature was not statistically significant; however, there was a tendency for the difference in survivorship between temperature treatments to be greatest at the lowest food level (Fig. 1A; $F_{2,41}$=2.97, P=0.06).

Tadpoles in the high food treatments were significantly heavier than tadpoles in the low and medium level treatments (Fig. 1B; $F_{2,41}$=11.22, P<0.01; Fisher's PLSD: high vs low, P<0.01; high vs medium, P<0.01; low vs medium, P=0.31). Tadpoles in the 25 °C treatment were larger than tadpoles in the 17 °C treatment (Fig. 1B; $F_{1,41}$=34.52, P<0.01). There was a significant food level by temperature treatment interaction (Fig. 1B; $F_{2,41}$=13.31, P<0.01). Tadpole mass was substantially higher in the high food, 25 °C treatment combination than all other treatment combinations, although the difference between 25 °C and 17 °C treatments increased with food level in general.

Our results show clearly that both food availability and temperature influenced the growth rate of wood frog tadpoles. Of particular interest was the interaction between temperature and food availability – tadpole growth was dramatically higher in the high food, high temperature treatment than in any other treatment. This is similar to the pattern found in some populations of *R. temporaria*, where increasing temperature had a negative effect on growth rate at restricted food levels, but a positive effect at *ad libitum* food levels (Laugen et al., 2005). The underlying mechanism for these results probably lies in the effect of temperature on metabolism and growth rates. Tadpoles in higher food treatments are able to grow faster at higher temperatures because the larger amount of food available will permit both the elevated metabolic rate associated with higher temperatures (e.g. Lindgren & Laurila, 2009) and a faster growth rate, whereas growth rate may be more constrained at lower temperatures and lower food levels.

Temperature also influenced survivorship, with more tadpoles surviving at cooler temperatures. While not statistically significant, there appeared to be a potential interaction between temperature and food level, with the effects of temperature on survivorship being highest at low food levels. Our results contrast with Sanuy et al. (2008) and Orizaola & Laurila (2009), who found that survivorship was higher at warmer temperatures in *Bufo calamita* and *R. lessonae*, respectively. The differences may relate to adaptations for specific temperatures. *Rana sylvatica* is an early spring breeder (late March) and thus its tadpoles may be better adapted to cooler temperatures, compared to *R. lessonae* and *B. calamita* that breed in early summer or in warm water (Bregulla, 1988; Sjögren et al., 1988; Sanuy & Joly, 2009). Indeed, the lower temperature treatment (17 °C) we used is within the range of preferred temperatures found for wood frog tadpoles by Freidenburg & Skelly (2004) and the higher temperaure treatment (25 °C) we used is near the maximum tolerated temperature found by Manis & Claussen (1985) for Ohio tadpoles acclimated at 15 °C. Thus, the higher temperature treatment we used may approach the upper limit for wood frog tadpoles and the higher mortality in this treatment is likely to reflect this fact.

Taken together, our results suggest that the effects of any temperature change, whether associated with global climate change or with local manipulations of the environment, could be mediated by food availability. Thus, predictions about the role of temperature effects on amphibian individuals and populations will need to take into account the effects of both temperature and food availability to provide a better understanding of potential consequences.

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