The Effects of Temperature and Salinity on Wood Frog (Lithobates sylvaticus) Tadpole Growth and Survival

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Abstract

Increased levels of road salt runoff in combination with increased temperatures earlier in the spring could create stressful environments for wood frog (*Lithobates sylvaticus*) tadpoles. We examined the effects of salinity and temperature as stressors, both independently and jointly. We used three concentrations of NaCl (control, low, and high) and two temperature treatments. Higher temperature resulted in significantly decreased survivorship, but did not affect mean tadpole mass. Salinity did not have any significant effects, nor did the interaction of salinity and temperature. These results suggest warming trends may have greater effects on this population than changes in salinity.

It is common for northern temperate regions to have salt-contaminated freshwater habitats due to the frequent use of road deicing compounds (e.g., Kaushal et al., 2005). Such salt-contamination can cause stunted growth, slower rates of metamorphosis, and decreased survival of anuran larvae (Dougherty and Smith, 2006; Collins and Russell, 2009; Langhans et al., 2009), however some species or populations of anurans are relatively tolerant of salt-contamination (e.g., Dougherty and Smith, 2006; Karraker, 2007; Collins and Russell, 2009). Indeed, some species appear to be prevented from occupying ponds contaminated with road salt, whereas other species appear to be able to occupy them (Collins and Russell, 2009).

Temperature is another factor that can affect the performance of anuran larvae. However, the effects of temperature on anuran tadpoles can be variable, with increased temperatures shown to decrease tadpole growth in some species (e.g., Álvarez and Nicieza, 2002; Orizaola and Laurila, 2009) and increase tadpole growth in other species (e.g., Sanuy et al., 2008; Castano et al., 2010). Temperature can also affect survivorship in tadpoles, with higher temperatures sometimes resulting in lower survivorship (e.g., Castano et al., 2010) or higher survivorship (e.g., Sanuy et al., 2008; Orizaola and Laurila, 2009).

Wood frogs (*Lithobates sylvaticus*) are a common and widespread frog in northeastern North America that inhabit vernal pools (Redmer and Trauth, 2005). This is a region where salinization of freshwater is important (Kaushal et al., 2005). Given that wood frogs are early spring breeders (Redmer and Trauth, 2005), they are likely to be affected by both road salt contamination and any warming trends associated with global climate change. Previous studies have found that wood frogs are susceptible to the effects of salt-contamination, but the concentrations that increase mortality or affect growth can vary among populations (e.g., Collins and Russell, 2009; Langhans et al., 2009; Petranka and Doyle, 2010). Road salt can also have significant demographic effects on wood frog populations (Karraker et al., 2008). Considering temperature, Castano et al. (2010) found that the survivorship of wood frog tadpoles from Ohio was better at 17°C than at 25°C; however, they found that tadpoles at 25°C were larger than tadpoles at 17°C.

To our knowledge, no previous study has examined the potential interaction between salt contamination and temperature on the performance of wood frog tadpoles. Such information may allow us to better understand the potential impacts of these environmental stressors on wood frogs.

To this end, we examined how the combination of increased salinity and increased water temperature may affect the growth and survivorship of wood frog tadpoles.

Materials and Methods

We collected wood frog egg masses (N = 6) from a local pond within 24 h of oviposition. We incubated the eggs in the laboratory until hatching. Upon hatching, tadpoles were maintained in large plastic containers where tadpoles from the different clutches were allowed to mix. We began the experiment once tadpoles reached Gosner Stage 26 (Gosner, 1960) and had a mean mass of 0.023 ± 0.001 g (N = 10).

The experiment was a 3 x 2 fully factorial design with three salinity treatments (control, low, and high) and two temperature treatments (25°C and 30.5°C) replicated 6 times. For the salinity treatments, we created stock solutions of the low (500 mg NaCl/L) and high (1000 mg NaCl/L) salinity treatments using NaCl (Fisher Scientific Sodium Chloride Certified for Biological Work) and aged tapwater. Temperature treatments were created by placing the containers in either a thermostat controlled lab (25°C) or a thermostat controlled greenhouse (30.5°C). Each experimental unit consisted of a clear plastic container (21 cm x 14 cm x 5 cm) filled with 500 mL of the appropriate salinity solution. Each container had 5 tadpoles.

Every 3 days we refilled each container with water of the appropriate salt concentration that had been acclimated to the treatment temperature. We also removed feces and any remaining food. We fed the tadpoles 0.05g of crushed Purina Rabbit Chow per tadpole every 3 days. After 10 days, we recorded the number of tadpoles alive in each container and weighed the survivors to the nearest 0.001 g after blotting dry. We used two-way ANOVAs to analyze the effects of temperature and salinity on tadpole mass and survivorship separately.

Results

Survivorship to the end of the experiment was higher in the 25 °C treatments than in the 30.5°C treatments (25°C: 0.80 ± 0.063 [N = 18], 30.5°C: 0.267 ± 0.096 [N = 18]; $F_{1,30} = 22.26$, $P < 0.0001$). Salinity had no effect on tadpole survivorship (Control: 0.65 ± 0.14 [N = 12], Low: 0.47 ± 0.12 [N = 12], High: 0.48 ± 0.11 [N = 12]; $F_{2,30} = 1.49$, $P = 0.24$). The interaction between temperature and salinity was not significant ($F_{2,30} = 1.18$, $P = 0.32$).

Mean tadpole mass was not significantly affected by temperature treatment (25°C: 0.0530 ± 0.0037 g [N = 17], 30.5°C: 0.0614 ± 0.0085 g [N = 6]; $F_{1,17} = 1.05$, $P = 0.32$). Mean tadpole mass was also not affected by salinity treatment (Control: 0.052 ± 0.0009 g [N = 8], Low: 0.053 ± 0.006 g [N = 7], High: 0.060 ± 0.009 [N = 8]; $F_{2,17} = 0.98$, $P = 0.40$). The interaction term was not significant ($F_{2,17} = 0.50$, $P = 0.61$).

Discussion

Our results indicate that NaCl concentrations of 500 and 1000 mg L^{-1} did not affect the growth or survivorship of wood frog tadpoles. This is in contrast to other studies that have found salinity to negatively affect survivorship, growth, and size at metamorphosis in wood frogs at concentrations ranging up to 1400 mg L^{-1} (Sanzo and Hecnar, 2006; Karraker et al., 2008). However, Petranka and Doyle (2010) found increased mortality in wood frog tadpoles only at concentrations of 4500 mg L^{-1} and no effect at lower concentrations. Thus, there appears to be a range of susceptibility to salinity in wood frog tadpoles across their geographical range. What drives such variability in susceptibility is unknown but warrants further investigation.

The survivorship of wood frog tadpoles at 25°C was greater than their survivorship at 30.5°C, but mean tadpole mass did not differ between the temperatures. Castano et al. (2010) found that survivorship of wood frog tadpoles was better at 17°C than at 25°C; however, they found that tadpoles at 25°C were larger than tadpoles at 17°C. Our results for survivorship are generally consistent with Castano et al. (2010) in that survivorship is better at the cooler temperature. Our results are also consistent with the observation that 25°C is near the maximum tolerated temperature for wood frog tadpoles from Ohio (Manis and Claussen, 1985). Thus, higher temperatures appear to negatively affect survivorship in wood frog tadpoles. However, our results for mass and those from Castano et al. (2010) suggest that growth in wood frog tadpoles in this Ohio population is generally better at higher temperatures since growth at 25°C was greater than at 17°C (Castano et al., 2010) and similar to that at 30.5°C (this study). The results of these two experiments do suggest that increasing temperatures associated with a warming climate could have serious consequences for wood frogs, especially since the increases in growth performance appear to level off after 25°C. However, spring temperatures for this Ohio population would have to drastically increase to have substantial impacts on wood frogs (water temperatures for the source pond in this experiment averaged 13.7°C in early April; Dougherty et al., 2005).

In summary, we found that wood frogs are not strongly affected by NaCl concentrations of 500 and 1000 mg/l, but they do suffer a reduction in survivorship when exposed to a warmer temperature of 30.5°C in comparison to 25°C. There does not however appear to be a synergistic effect of these stressors on wood frogs in this population, as evidenced by the lack of a significant interaction between temperature and salinity.

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