

## FINE-TUNED PHENOTYPES: TADPOLE PLASTICITY UNDER 16 COMBINATIONS OF PREDATORS AND COMPETITORS

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**Abstract.** It is now well appreciated that most organisms can alter their phenotypes when faced with environmental variation. Decades of empirical investigations have documented hundreds of examples of phenotypic plasticity, yet most studies have focused on the presence or absence of a single environmental factor. As a result, we know little about how organisms respond to gradients of environmental factors (i.e., threshold responses vs. continuous responses), nor do we understand how organisms respond to combinations of environmental variables. I examined how larval wood frogs (*Rana sylvatica*) altered their behavior, morphology, and growth in response to combined gradients of predation and competition. Increased predation risk induced lower activity, deeper tails, and shorter bodies, which collectively caused slower growth. Increased competition caused slower growth which induced higher activity, shallower tails, and longer bodies. For both environmental gradients, the responses were frequently continuous rather than threshold responses. Moreover, predation and competition had interactive effects. Responses to predators were always larger under low competition than under high competition. Responses to competition were larger under low predation risk when predation and competition induced traits in the same direction, but larger under high predation risk when predation and competition induced traits in opposite directions. The results demonstrate that responses to phenotypically plastic traits can be fine-tuned to a wide variety of environmental combinations.

**Key words:** *anuran; competition; dose response; larval wood frogs; phenotypic plasticity; predation; Rana sylvatica; tadpole.*

### INTRODUCTION

The phenotype that organisms express is the product of the genes that they carry and the environment that they experience (phenotypic plasticity; Schlichting 1986, Sultan 1987, Karban and Baldwin 1997, Tollrian and Harvell 1999). Theory predicts that phenotypic plasticity should evolve when alternative phenotypes are favored in different environments (Via and Lande 1985, 1987, Gomulkiewicz and Kirkpatrick 1992, Moran 1992). In most evolutionary theory, plasticity models are restricted to only two environments for ease of mathematical tractability. Empirical tests of plasticity theory have, in turn, also largely restricted themselves to examining only two environments (Baldwin et al. 1990, Kingsolver 1995, Dudley and Schmitt 1996, Agrawal 2000, Sell 2000, Trussell 2000, Van Buskirk 2000). Environments are rarely this simplistic, but working with a limited number of environmental variables is an important first step in determining whether a particular response is adaptive (not all responses are adaptive; Travis 1994). Now that we have hundreds of case studies that document examples of adaptive plasticity, it has become increasingly apparent that we need to examine more complex environmental scenarios that better approximate nature (Relyea 2003).

In nature, there are both discrete environments (e.g., water vs. air) and continuous environments (e.g., competitor density). When environments are discrete, selection may favor discrete phenotypes (“threshold responses” or “on-off switches”) to ensure a good match between the phenotype and the environment. This type of response is epitomized by the classic work on *Ranunculus* plants forming aquatic and aerial leaves (Cook and Johnson 1968). However, a great many environments are continuous. In this case, selection should favor continuous phenotypes, particularly when extreme phenotypes confer no additional fitness benefits but have associated fitness costs (due to phenotypic trade-offs; Pigliucci 2001). While it is likely that these criteria for continuous responses are commonly met, an empirical focus on presence/absence experiments precludes us from determining whether most environmentally induced responses are continuous or discrete responses (although this is by far a bigger problem in animal studies than in plant studies). We need to conduct experiments that determine whether organisms respond to continuous environments with continuous phenotypes.

A focus on only two environments also restricts our understanding of how organisms respond to different combinations of environments. For example, most prey in nature face combinations of different predators, yet most empirical studies have examined prey responses to one predator at a time (Sih et al. 1998, Relyea 2003).

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Similarly, most prey in nature face combinations of predators and competitors (Gurevitch et al. 2000, Chase et al. 2002), yet few studies have examined the combined effects of predators and competitors on a prey's phenotype. Studies that have examined combinations of predators and competitors typically have been limited to two-by-two factorial designs (the presence and absence of predators crossed with the presence and absence of competitors; Gilliam and Fraser 1987, Horat and Semlitsch 1994, Werner and Anholt 1996, Relyea 2002a). We need studies that test whether organisms can fine-tune their phenotypes to a wide range of environmental combinations. If organisms respond to combinations of environments with continuous responses, then it means we cannot condense phenotypic responses down to simple two-by-two factorial outcomes.

Natural variation in predator and competitor densities presents an excellent contextual background for testing responses to gradients of environmental combinations. Predators can induce changes in prey behavior and morphology that make prey more resistant to predation, but at the cost of slower growth or reduced reproduction (Havel 1987, Lima and Dill 1990, Kats and Dill 1998, Tollrian and Harvell 1999). Competitors can also induce behavioral and morphological responses that allow the organism to grow better under low food conditions, but at the cost of increased vulnerability to predation (Anholt and Werner 1995, Relyea 2002a, d). Because predators and competitors typically induce prey behavior and morphology in opposite directions, one might expect prey to evolve an ability to fine-tune their phenotypes in a way that balances the challenges posed by predators and competitors. If so, the two types of environments should have interacting effects on prey phenotypes such that the response to predators depends on the intensity of competition and the response to competitors depends on the intensity of predation risk.

I tested for fine-tuned phenotypes using wood frog tadpoles reared under gradients of predator-competitor combinations. Wood frogs are well known for altering their behavior, morphology and growth in response to predators and competitors (Van Buskirk and Relyea 1998, Relyea and Werner 2000, Relyea 2000, 2001a, b, 2002a, b, c), yet we know nothing about the interactive effects of predation and competition on tadpole phenotypes across predator and competitor gradients. I tested three hypotheses: (1) predator and competitor-induced responses are continuous responses (i.e., not threshold responses); (2) predator-induced changes in behavior, morphology, and growth are strongest under low competitor density and weakest under high competitor density; and (3) competitor-induced changes in behavior, morphology, and growth are strongest under low predator density and weakest under high predator density.

## METHODS

I conducted the experiment at the University of Pittsburgh's Pymatuning Laboratory of Ecology. On 24–26 April 2001, I filled 1300-L cattle watering tanks with 1000 L of well water. To simulate natural ponds, I added 300 g of dry leaves (primarily *Quercus* spp.), 25 g of commercial rabbit chow, and an aliquot of plankton from a mixture of 10 nearby ponds. I covered all tanks with 60% shade cloth lids to prevent other organisms from colonizing and then allowed the tanks to sit for nine days to develop sufficient periphyton to serve as a tadpole food source.

The experiment used a randomized block design. There were four spatial blocks with each block containing a factorial combination of four levels of predator risk (0, 1, 2, or 4 caged dragonfly larvae; *Anax junius*) and four levels of intraspecific competition (20, 40, 80, or 160 wood frog tadpoles). Thus, there were a total of 16 treatment combinations replicated four times (64 experimental units). The competitor treatments translated to 11, 22, 44, and 89 wood frogs/m<sup>2</sup>, which is well within the natural range of densities observed for wood frogs (up to 450 wood frogs/m<sup>2</sup>; R. A. Relyea, E. E. Werner, D. K. Skelly, and K. L. Yurewicz, *unpublished data*). Wood frogs were a mixture of larvae collected as 10 egg masses from a nearby pond shortly after oviposition and hatched in outdoor wading pools. On 5 May, the tadpoles were added to the tanks (individual mean mass  $\pm$  1 SE = 108  $\pm$  8). A sample of 20 tadpoles was set aside to estimate survivorship due to handling; the 24 hr survival of this sample was 100%.

All tanks contained four predator cages constructed of 10  $\times$  10-cm drain pipe with window screen on each end. Using predator cages allows predators to emit chemical cues without killing the target tadpoles in the experiment (Petranka et al. 1987, Kats et al. 1988). For each level of predation risk, the appropriate number of predatory dragonfly larvae were added to the cages (one predator per cage) and the remaining cages were left empty. Approximately 300 g of wood frog tadpoles was fed to each predator three times per week. Empty cages were briefly lifted and put back into the tank to equalize disturbance among tanks.

Midway through the experiment (23 May), I observed tadpole activity four times in every tank. To quantify activity, I counted the number of tadpoles that I could observe and the number of observed tadpoles that were active (moving) using scan sampling (Altmann 1974). Dividing the latter by the former estimates the proportion of active tadpoles. This approach has been successfully used in several past studies (Peacor and Werner 1997, Relyea 2002a, b).

On 31 May–1 June, I terminated the experiment. The surviving tadpoles were counted and weighed to estimate individual growth rate ((final mass minus initial mass)/26 or 27 d). A sample of 10 tadpoles was eu-

thanized and preserved in 70% formalin (10 tadpoles have proven to well represent an experimental unit in past experiments). The preserved tadpoles were measured using an image analysis system in which I measured tail depth and body length (see Fig. 1 in Relyea 2000 for a photo of these dimensions). I selected these two dimensions because they are consistently induced by predators and competitors and appear to be under strong selection (Van Buskirk and Relyea 1998, Relyea 2002a, c).

#### Statistical analysis

A challenge in analyzing tadpole shape is to remove differences that are due to differences in tadpole size. To remove differences in size, I conducted a linear regression of each log-transformed linear dimension on log-transformed mass (the values were log-transformed to improve the linearity of the relationship). The residual values from this regression were saved because they represent mass-independent measures of shape. I then calculated the mean residuals for each tank. This approach has been used successfully in numerous previous studies (Relyea 2000, 2001b, 2002a, b, c, d).

I analyzed the data using a multivariate analysis of variance (MANOVA) because multiple responses from each individual were not independent. I analyzed the mean responses from each tank (survival, growth rate, activity, relative tail depth, and relative body length). None of the block interactions were significant, so I pooled the interaction sums of squares with the error term. To determine whether increased densities of competitors and predators caused continuous phenotypic changes, I conducted Fisher's tests for mean comparisons. I defined a continuous response as one in which changes along an environmental gradient caused phenotypic changes between more than one pair of treatments (e.g., a phenotypic difference between zero and one predators plus a change between one and two predators).

#### RESULTS

The multivariate analysis found significant effects of blocks, competitors, and predators as well as a significant competitor-by-predator interaction (Table 1). Tadpole survival was high (89–97%) and was not affected by any of the treatments.

Tadpole activity was affected by predators, competitors, and their interaction (Table 1, Fig. 1). Adding more predators (from zero to four) decreased tadpole activity by 33% under low competition ( $P = 0.021$ ) and by 13% under high competition (marginally non-significant,  $P = 0.078$ ). Adding more competitors (from 20 to 160) increased activity by 17% under low predation risk ( $P = 0.021$ ) and by 33% under high predation risk ( $P < 0.001$ ). The predator-induced changes were continuous with 20 competitors (0 vs. 1 predator,  $P = 0.002$ ; 1 vs. 2 predators,  $P = 0.014$ ) and 40 competitors (0 vs. 1 predator,  $P = 0.002$ ; 1 vs. 4

TABLE 1. Results of a multivariate analysis of variance that examined the impact of competitors and caged predators on the survival, growth, activity, and relative morphology of wood frog tadpoles.

A) Multivariate values			
Factor	df	F	P
Block	15, 113	5.8	<0.0001
Competitors	15, 113	25.1	<0.0001
Predators	15, 113	13.1	<0.0001
Competitors × Predators	45, 186	2.6	<0.0001
B) Univariate values			
Factor	Competitors (P)	Predators (P)	Competitors × Predators (P)
Survival	0.246	0.641	0.094
Activity	<0.00001	<0.00001	0.046
Tail depth	0.006	<0.00001	0.036
Body length	<0.00001	<0.00001	0.084
Growth	<0.00001	<0.00001	<0.00001

predators,  $P = 0.047$ ), but discontinuous with 80 competitors (0 vs. 1 predator,  $P = 0.055$ ; 1 vs. 2 or 4 predators,  $P > 0.2$ ) and 160 competitors (0 vs. 1 predator,  $P = 0.057$ ; 1 vs. 2 or 4 predators,  $P > 0.8$ ). The competitor-induced changes were discontinuous with zero predators (20 vs. 40 or 80 competitors,  $P > 0.1$ ; 20 vs. 160 competitors,  $P = 0.021$ ) and 1 predator (20 vs. 40 competitors,  $P = 0.365$ ; 20 vs. 80 competitors,  $P = 0.006$ ; 80 vs. 160 competitors,  $P = 0.390$ ), but continuous with two predators (20 vs. 40 competitors,  $P > 0.001$ ; 40 vs. 160 competitors,  $P = 0.039$ ) and four predators (20 vs. 80 competitors,  $P = 0.008$ ; 80 vs. 160 competitors,  $P = 0.041$ ).

Relative tail depth was also affected by predators, competitors, and their interaction (Table 1, Fig. 1). Adding more predators caused a large increase in tail depth under low competition ( $P < 0.001$ ) but only a moderate increase in tail depth under high competition ( $P < 0.001$ ). Adding more competitors had no effect on tail depth under low predation risk ( $P = 0.446$ ) but induced shallower tails under high predation risk ( $P = 0.005$ ). The predator-induced changes were continuous with 20 competitors (0 vs. 1 predator,  $P < 0.001$ ; 1 vs. 2 predators,  $P = 0.049$ ), 40 competitors (0 vs. 1 predator,  $P < 0.001$ ; 1 vs. 2 predators,  $P = 0.013$ ), and 80 competitors (0 vs. 1 predator,  $P < 0.001$ ; 1 vs. 2 predators,  $P = 0.018$ ), but discontinuous with 160 competitors (0 vs. 1 predator,  $P < 0.001$ ; 1 vs. 2 or 4 predators,  $P \geq 0.09$ ). The competitor-induced changes in tail depth were not different with zero predators ( $P > 0.15$  for all comparison) and discontinuous for one, two, and four predators (20 vs. 40 competitors,  $P > 0.2$ ; 20 vs. 80 competitors,  $P > 0.6$ ; 80 vs. 160 competitors,  $P \leq 0.05$ ).

Relative body length was affected by competition and predation risk with a nearly significant interaction (Table 1, Fig. 1). Adding more predators induced a large decrease in body length under low competition

( $P < 0.001$ ) but only a moderate decrease under high competition ( $P < 0.001$ ). Adding more competitors induced a small increase in body length under low predator risk ( $P = 0.025$ ) but a large increase in body length under high predator risk ( $P < 0.001$ ). The predator-induced changes in body length were discontinuous with all competitor densities (0 vs. 1 predator,  $P \leq (0.05; 1$  vs. 2 or 4 predators,  $P > 0.1)$ ). The competitor-induced changes in body length were discontinuous with zero predators (20 vs. 40 or 80 competitors,  $P > 0.1$ ; 20 vs. 160 competitors,  $P = 0.036$ ) and two predators (20 vs. 40 competitors,  $P = 0.528$ ; 20 vs. 80 competitors,  $P < 0.001$ , 80 vs. 160 competitors,  $P = 0.168$ ), but continuous with one predator (20 vs. 40 competitors,  $P = 0.016$ ; 40 vs. 80 competitors,  $P = 0.009$ ) and marginally continuous with four predators (20 vs. 40 competitors,  $P = 0.072$ ; 40 vs. 80 competitors,  $P = 0.001$ ).

Tadpole growth was affected by competitors, predators, and their interaction (Table 1, Fig. 1). Adding more predators caused a small decrease in growth under low competition (9 mg/d;  $P < 0.001$ ) but had no effect on growth under high competition ( $P > 0.2$ ). Adding more competitors caused a large decrease in growth under low predation risk (27 mg/d;  $P < 0.001$ ) but a moderate decrease under high predation risk (16 mg/d;  $P < 0.001$ ). The predator-induced changes in growth were continuous with 20 competitors (0 vs. 1 predator,  $P < 0.001$ ; 1 vs. 2 predators,  $P < 0.001$ ) and 40 competitors (0 vs. 1 predator,  $P < 0.001$ ; 1 vs. 4 predators,  $P = 0.021$ ), but did not change with 80 or 160 competitors ( $P > 0.09$  for all comparisons). The competitor-induced reductions in growth were continuous with zero predators (20 vs. 40 competitors,  $P < 0.001$ ; 40 vs. 80 competitors,  $P < 0.001$ ; 80 vs. 160 competitors,  $P < 0.001$ ), 1 predator (20 vs. 40 competitors,  $P = 0.009$ ; 40 vs. 80 competitors,  $P = 0.002$ ; 80 vs. 160 competitors,  $P < 0.001$ ), two predators (20 vs. 40 competitors,  $P = 0.691$ , 20 vs. 80 competitors,  $P = 0.019$ ; 80 vs. 160 competitors,  $P < 0.001$ ), and four predators (20 vs. 40 competitors,  $P = 0.033$ , 40 vs. 80 competitors,  $P = 0.043$ , 80 vs. 160 competitors,  $P < 0.001$ ).

#### DISCUSSION

This study demonstrated that larval wood frogs possess an amazing ability to alter their phenotypes in response to a wide range of predator and competitor densities. Predators induced tadpoles to exhibit lower activity and develop relatively deep tails and short bodies. Reduced activity is a common response in a variety of taxa because it lowers the risk of being detected by predators, although this benefit comes at the cost of slower growth (Lima and Dill 1990, Werner and Anholt 1993, Kats and Dill 1998). Predator induction of relatively deep tails and short bodies has been observed repeatedly in several species of larval anurans using lab experiments, outdoor mesocosm experiments, and pond pen experiments (Van Buskirk 2000, Relyea

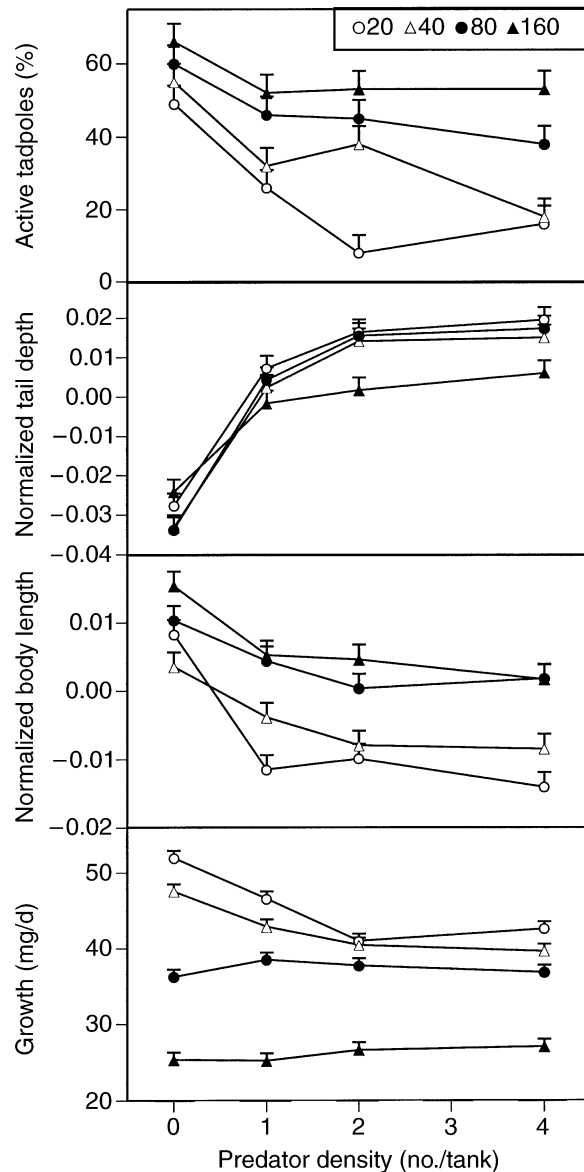


FIG. 1. The change in growth, activity, and relative morphology of wood frog tadpoles exposed to four densities of caged predators (0, 1, 2, and 4 aeshnid dragonfly larvae/1300-L tank) crossed with four levels of intraspecific competition (20, 40, 80, and 160 tadpoles). Each symbol represents a different level of intraspecific competition. Morphological measurements were made independent of size by regressing the dimensions of all individuals against mass and then saving the residuals. Data are least-squares means + 1 SE.

2001a, 2002c). Selection experiments have shown that these morphological changes are adaptive; tadpoles with deep tails and short bodies survive *Anax* predation better than tadpoles with shallow tails and long bodies (Van Buskirk et al. 1997, Van Buskirk and Relyea 1998). However, tadpoles with deep tails and short bodies experience slower growth (Van Buskirk and Relyea 1998, Relyea 2002a). Thus, as predicted from these



PLATE. 1. Comparison between these two individuals demonstrates the morphological plasticity of wood frog tadpoles reared in high-competition/low-predation vs. low-competition/high-predation environments. Photo credit: Rick Reylea.

past studies, large changes in predator-induced behavior and morphology in the current study (e.g., under low competition) caused slower growth, but small changes in predator-induced behavior and morphology (e.g., under low competition) had no effect on growth.

Competitors induced wood frogs to be more active and develop relatively shallow tails and long bodies. Competitor induction of higher activity permits better detection and acquisition of resources at the cost of higher predation risk (Werner and Anholt 1993) and has been observed in a variety of taxa (Anholt and Davies 1987, Anholt and Werner 1995, Macchiusi and Baker 1992). Competitor induction of animal morphology is much less common. Some marine invertebrates produce spines, tentacles, and stolons in response to interference competitors (Stemberger and Gilbert 1987, Harvell 1990) and a few species of larval amphibians (tiger salamanders, *Ambystoma tigrinum*, and spadefoot toads, *Spea bombifrons* and *S. multiplicata*) produce carnivorous phenotypes when raised at high densities (Bragg 1957, Reilly et al. 1992, Pfennig 1992a, b). However, the competitor-induced morphology of the wood frog tadpoles (and gray tree frog tadpoles) is fundamentally different; they do not produce carnivorous phenotypes (Relyea 2002a, d, Relyea and Hoverman 2003; see Plate 1). These changes in larval wood frog behavior and morphology associated with competition are induced via changes in food concentration and the changes appear to be adaptive; competitor-induced tadpoles have a superior growth ability compared to non-induced tadpoles of similar size (Relyea 2002a). Thus, in the current experiment, increased competitor density directly reduced food levels, causing slower tadpole growth and inducing high activity, long bodies, and shallow tails.

When wood frogs were exposed to 16 combinations of predators and competitors, all of the traits had significant or nearly significant ( $P \leq 0.08$ ) interactive effects. This suggests that wood frogs possess a sophisticated plastic ability that is capable of four functions: (1) sensing the intensity of predation risk; (2) sensing the intensity of competition; (3) making decisions to balance these opposing challenges; and (4) enacting these decisions to produce a finely tuned phe-

notype. Past studies of predator-induced reductions in prey growth (using primarily two-by-two factorial designs) have found that the reductions are more extreme under lower competition than high competition (Werner and Anholt 1996, Van Buskirk and Yurewicz 1998, Relyea 2002a, d). Similarly, predator-induced changes in behavior and life history are more extreme under lower competition (Petranka 1989, Anholt et al. 1996, Van Buskirk and Yurewicz 1998, Weetman and Atkinson 2002). Few studies have examined the impact of competition on predator-induced morphology, but the small amount of existing data (using two-by-two factorial designs) suggest that morphological defenses are more extreme under lower competition (Relyea 2002a, d, Relyea and Hoverman 2003), although this is not always the case (Kuhlmann and Heckmann 1994). Thus, in two-by-two factorial experiments, predators induce large prey responses when competition is low and small prey responses when competition is high. Given that predator-induced traits typically come at the cost of slower growth, it appears that these interactive effects occur due to prey balancing the fitness benefits of surviving predators against the fitness costs of slower growth. However, because these studies are primarily two-by-two factorial experiments, one cannot determine whether prey can fine-tune their phenotypes to the many combinations of predators and competitors that exist in most natural systems. The current study demonstrates that wood frog tadpoles have the ability to fine-tune their phenotypes to all combinations of predators and competitors. Moreover, because these responses are frequently continuous responses (see *Discussion*), this means that prey responses to a large number of predator-competitor combinations cannot be simply condensed into a two-by-two set of prey decisions.

Because predator-induced traits were more extreme under low competition, I hypothesized that competitor-induced traits would be more extreme under low predation risk. Under low predation risk, competitor induction was more extreme for growth but it was less extreme for activity, tail depth, and body length. Thus, I rejected the original hypothesis. The two different outcomes occurred because predators and competitors

induced growth in similar directions but they induced activity, tail depth, and body length in opposite directions. When competitors and predators induce traits in opposite directions, predator-free environments allow the phenotype to already be quite competitive (high activity, shallow tail, long body); increased competition can only induce the phenotype to change a small additional amount (presumably due to a phenotypic limit). In contrast, high predation environments induce the phenotype to be much less competitive (low activity, deep tail, short body), allowing a large range of potential phenotypic change in response to competition. As a result, competition is able to induce larger phenotypic changes under high predation risk.

The wood frog responses to predators were frequently continuous. Predator-induced changes in activity were continuous at the two lowest competitor densities, but not at the two highest competitor densities. Similarly, predator-induced changes in tail depth were continuous at the three lowest competitor densities, but not at the highest competitor density. Predator-induced changes in body length were discontinuous at all competitor densities. Predator-induced changes in growth were continuous at the two lowest competitor densities, but not at the two highest competitor densities. These results suggest that, for many of the traits, the largest changes occur with the addition of the first predator, but further changes are possible, depending on the intensity of competition. The continuity of predator-induced behavior and morphology is in agreement with the wide range of unique behavioral and morphological phenotypes produced by several tadpole species in response to different species of predators (Relyea 2001a, 2003). Moreover, increased predation risk (higher numbers of predators, higher numbers of killed prey, or higher concentrations of predator cues) causes continuous changes in anti-predator behavior, including increased vertical migration in *Daphnia* (Loose and Dawidowicz 1994, von Elert and Pohnert 2000), increased spatial avoidance of the predator in tadpoles (Anholt et al. 1996), reduced swimming speed in tadpoles (Anholt et al. 2000) and greater activity reductions in tadpoles (Petranka 1989, Anholt et al. 1996, 2000, Van Buskirk and Arioli 2002). Much less is known about the continuous nature of predator-induced morphology. The few existing studies suggest that morphological traits are continuously plastic for predator-induced protists (Kusch 1993, 1995), marine bryozoans (Harvell 1998), cladocerans (Barry and Bayly 1985), and European water frogs (*Rana lessonae*, Van Buskirk and Arioli 2002).

The wood frog responses to competitors also were frequently continuous. The competitor-induced changes in activity were discontinuous under the two lowest densities of predators but continuous under the two highest densities of predators. The competitor-induced changes in body length were discontinuous with zero and two predators, but continuous with one and four

predators. The competitor-induced changes in tail depth were discontinuous at all predator densities, but changes in growth were continuous at all predator densities. This again suggests that competitor-induced traits are frequently continuous but this continuity is highly dependent on the intensity of predation that is present. The continuity of competitor-induced behavior and morphology in tadpoles is in agreement with a recent study of wood frogs (Relyea 2002a).

Continuous responses might be expected to evolve when two conditions hold true: (1) organisms experience continuous gradients of environmental factors; and (2) when exhibiting a more extreme phenotype confers no additional fitness benefit but does carry an additional fitness cost. Based on long-term surveys of natural ponds, it is clear that wood frogs (and many other prey) do experience a continuous gradient of predation risk and competition over space and time (E. E. Werner, R. A. Relyea, D. K. Skelly, and K. L. Yurewicz, *unpublished data*). Further, extreme behavioral and morphological phenotypes do carry a substantial cost of either slower growth or lower survival (Relyea 2001c, 2002a, b). Thus, wood frogs meet the above criteria and it is clear from the current study that wood frogs have evolved continuous behavioral and morphological responses to predators and competitors. Although many models of plasticity evolution focused on two environments for ease of tractability (Via and Lande 1985, Lively 1986, Moran 1992, Van Tienderen 1997), models that incorporate continuous changes in environments appear to better represent animal responses to predators and competitors (Gomulkiweicz and Kirkpatrick 1992, Van Tienderen and Koelewijn 1994).

### Conclusions

The results of this study suggest that predator- and competitor-induced traits vary continuously in tadpoles and that the responses to combinations of predators and competitors are fine-tuned to balance the opposing phenotypic optima. Thus, while examining the presence and absence of environmental factors is an important first step in determining whether a given plastic response is adaptive, it does not allow us to determine whether the organism is sensitive to a wide range of environmental variation. Further, few organisms (if any) experience variation in one environmental factor in isolation of other environmental factors. It is critical that we appreciate a wider range of natural complexity in which organisms have evolved and how phenotypically plastic responses to one environmental gradient may be highly contingent on other environmental gradients. When we examine these more complex scenarios, we will better understand how plasticity has evolved and how it affects the ecology of the organisms that possess it.

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