

THE MANY FACES OF PREDATION: HOW INDUCTION, SELECTION, AND THINNING COMBINE TO ALTER PREY PHENOTYPES

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Abstract. Populations in nature face a number of factors that can alter their traits and subsequent performance. Predation is one factor that can have widespread effects on the mean trait value in a prey population because predators can impact prey traits through a number of processes. Predators can directly induce prey phenotypes through visual and chemical cues, predators can indirectly alter prey phenotypes by thinning the prey population (thereby reducing competition), and predators can cause selection on prey phenotypes through nonrandom killing. Ecologists are beginning to understand each of these three processes in isolation, but these processes act on prey phenotypes simultaneously, and we lack information on the relative importance of these processes in determining the final phenotype of prey.

I used a system of gray treefrog tadpoles (*Hyla versicolor*) and dragonfly predators (*Anax longipes*) to determine the separate and combined impacts of induction, thinning, and selection on the behavior, morphology, and growth of tadpoles reared in aquatic mesocosms. Using combinations of hand thinning, cues emitted from caged predators, and lethal predators, I demonstrated that the impact of lethal predators on growth was mediated primarily through thinning, the impact on morphology was primarily through induction, and the impact on behavior was affected similarly through thinning and induction. Surprisingly, while we know from numerous studies that the dragonflies kill tadpoles nonrandomly, selection did not have a significant impact on the final phenotypes of the tadpoles. This work appears to be the first study that identifies the relative magnitudes of the three predatory processes on a suite of prey phenotypes. The three processes can have opposing or supporting effects, the relative magnitudes of which likely differ among systems, illustrating why we often observe contradictory results when we examine the impact of lethal predators on prey phenotypes. Thus, it is important that we identify the mechanisms underlying the outcomes of predation, rather than simply observing the outcomes and then inferring which processes are responsible.

Key words: *Anax longipes*; *antipredator traits*; *Hyla versicolor*; *phenotype*; *phenotypic plasticity*; *predation*.

INTRODUCTION

Most organisms observed in nature possess phenotypes that are the product of two important processes: natural selection and environmental induction. Natural selection can alter the phenotypic range of populations in the current generation by serving as a filter that favors the survival of particular phenotypes (Weiss and Gorman 1990, Kingsolver 1995, Van Buskirk and Relyea 1998). Selection also can alter phenotypes over several generations by altering the genetic composition and, thus, the range of possible phenotypes within populations (Scheiner and Lyman 1991, Schluter 1994, Grant and Grant 1995). Concurrently, environmental conditions can have substantial impacts on phenotypes by inducing changes in behavior, morphology, physiology, or life history of most organisms, and these changes are often in adaptive directions (Bradshaw 1965, Schlichting 1986, Sultan 1987, West-Eberhard

1989, Travis 1994, Schlichting and Pigliucci 1998, Tollrian and Harvell 1999). Therefore, when one observes an organism in its natural environment, the phenotype observed is produced by a combination of both selection and induction, but it is difficult to determine the relative importance of these two processes.

Predation is a pervasive process in nature that causes selection on, and induction of, prey phenotypes. Predators differentially kill prey of different sizes and shapes, and thereby produce a selective filter that alters the mean phenotype of the survivors in the current generation (Hall et al. 1970, Havel and Dodson 1984, Brönmark and Miner 1992, Wellborn 1994, Van Buskirk et al. 1997). Predation also can induce prey to decrease foraging activity, decrease growth, and alter morphology (Havel 1987, Sih 1987, Lima and Dill 1990, Kats and Dill 1998, Tollrian and Harvell 1999). In addition to directly inducing phenotypic changes in their prey, predators can indirectly induce prey phenotypes by thinning prey populations and reducing competition. Reduced competition increases resources, causing many organisms to decrease foraging activity,

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increase growth, and alter morphology in directions similar to that induced by predators (Harvell 1990, Pfennig 1992a, b, Reilly et al. 1992, Anholt and Werner 1995, Van Buskirk and Yurewicz 1998, Relyea 2000, 2002a). Thus, predators can alter the phenotypes in a population of prey by selection (nonrandom killing), direct induction of traits, and indirect induction of traits via reduced competition. This trio of processes may explain why studies arrive at different conclusions when they examine the impact of lethal predators on traits such as prey growth (Morin 1983, Figiel and Semlitsch 1990, Wilbur and Fauth 1990, Gascon and Travis 1992, Semlitsch 1993, Diehl and Eklöv 1995, McIntosh and Townsend 1996, Moran et al. 1996). The relative importance of each process may vary among systems, preventing investigators from correctly inferring which process underlies their observed phenomenon. Thus, we need experiments that can examine these processes in isolation and in combination.

Larval anurans pose an ideal system for addressing this phenomenon because we have extensive studies documenting the separate impacts of predator selection, predator induction, and reduced competition on tadpole phenotypes. First, predators kill tadpoles nonrandomly; tadpoles with high activity, relatively shallow tail fins, and relatively large bodies are killed more often than tadpoles with the opposite suite of traits (Skelly 1994, Anholt and Werner 1995, Van Buskirk et al. 1997, Van Buskirk and Relyea 1998, Relyea 2001b). Second, chemical cues from many predators induce tadpoles to alter their phenotype. Typically, tadpoles become less active and develop a relatively small body and deep tail fin, particularly in the presence of the most risky predators (Lawler 1989, Smith and Van Buskirk 1995, Van Buskirk et al. 1997, Relyea and Werner 1999, 2000, Relyea 2001a, 2002c Relyea and Mills 2001.). Third, reduced competition induces tadpoles to become less active and develop a relatively small body and deep tail fin (Anholt and Werner 1995, Relyea 2002a). Therefore, when one examines a tadpole in a natural pond that contains predators, the observed phenotype is the combined outcome of (1) the predator selecting phenotypes from the morphological and behavioral variation present in the tadpole population, (2) the predator directly inducing the tadpoles via predatory cues, and (3) the predator indirectly inducing the tadpoles by thinning the prey population and reducing competition. Past experiments have considered each of these processes in isolation. My objective in the current study was to examine the relative impacts of each process on the final phenotype of gray treefrog tadpoles (*Hyla versicolor*) by controlling each process separately and in combination. My hypothesis was that selection, induction, and reduced competition would have similar absolute impacts on the growth, behavior, and morphology of treefrog tadpoles. While natural selection on prey phenotypes can certainly occur in any environment, in this study I restrict the use of the term "se-

lection" to the process of nonrandom killing of prey by predators.

Estimating the separate and combined contributions of predator induction, reduced competition, and selection can be achieved through a number of environmental manipulations. In larval anurans, the process of predator induction can be isolated from the other two processes because aquatic predators emit chemical cues that induce antipredator responses in tadpoles (Petranka et al. 1987, Kats et al. 1988, McCollum and Leimberger 1997). Thus, caged predators can be used to simulate the predation threat without thinning or selecting on the prey population. The effect of predators thinning the prey population can be isolated using hand thinning that is conducted at the same rate as a lethal predator (but without the inductive and selective effects of a predator). Isolating the effect of selection without thinning is more problematic, but it may be estimable by comparing the results of randomly thinned vs. selectively thinned populations. By using these techniques in isolation and in combination, we can build up from environments with no selection, no induction, and no reduced competition (i.e., no-predator environments) to the full combination of all three processes (i.e., lethal-predator environments).

METHODS

I conducted the experiment in 51 100-L wading pools that served as pond mesocosms. I filled each pool with well water, and then added 100 g of deciduous leaves (primarily oak leaves, *Quercus* spp.), 5 g of Purina rabbit chow, and a sample of plankton from a nearby pond. The pools were randomly assigned one of eight treatments in a completely randomized design. The first six treatments were a factorial cross between the presence and absence of caged predators and one of three thinning treatments: no thinning, random thinning, or selective thinning. Each of the six treatments was replicated six times. I defined random thinning as thinning with a large aquarium net (20 × 15 cm) from which few tadpoles were able to escape, and selective thinning as thinning with a small aquarium net (6 × 8 cm) from which ~50% of the pursued tadpoles could escape (*personal observation*). Because deep-tailed tadpoles are better at escaping predators (Van Buskirk et al. 1997, Van Buskirk and Relyea 1998, Relyea 2001b), the smaller net should preferentially catch tadpoles with relatively shallow tails; however, I did not explicitly test this assumption. The remaining two treatments were two types of lethal (uncaged) predator treatments. The first lethal predator treatment was replicated six times, and remained undisturbed throughout the duration of the experiment. The second lethal predator treatment (hereafter termed the "test-predator treatment") was replicated nine times, and was destructively sampled in groups of three every four days over the course of the experiment to quantify the rate of predation on treefrog tadpoles. Using these treatments,

TABLE 1. Mean comparisons used to estimate the three impacts of predators on the phenotype of their prey (induction, thinning, and selection), both in isolation and in combination.

Process	Appropriate mean comparisons
Induction	Caged-predator vs. No-predator Caged-predator, random thin vs. No-predator, random thin Caged-predator, selective thin vs. No-predator, selective thin Lethal-predator vs. No-predator, selective thin
Thinning	No-predator, random thin vs. No-predator Caged-predator, random thin vs. Caged-predator
Selection	No-predator, selective thin vs. No-predator, random thin Caged-predator, selective thin vs. Caged-predator, random thin Lethal-predator vs. Caged-predator, random thin
Induction + Thinning	Caged-predator, random thin vs. No-predator
Induction + Selection	Caged-predator, selective thin vs. No-predator, random thin Lethal-predator vs. No-predator, random thin
Thinning + Selection	No-predator, selective thin vs. No-predator Caged-predator, selective thin vs. Caged-predator Lethal-predator vs. Caged-predator
Induction + Thinning + Selection	Lethal-predator vs. No-predator Caged-predator, selective thin vs. No-predator

I had multiple ways of estimating the separate and combined effects of induction, thinning, and selection (Table 1).

I equipped each pool with a single predator cage (Relyea 2002c). In pools assigned the no-predator treatment, cages remained empty. In pools assigned caged-predator treatments, each cage contained a single, late-instar aeshnid dragonfly larvae (*Anax* spp.). Larval aeshnids are voracious predators of tadpoles and naturally coexist with gray treefrog tadpoles. The density used ($1/m^2$) is well within natural aeshnid densities (up to $5/m^2$; R. A. Relyea, E. E. Werner, D. K. Skelly, and K. L. Yurewicz, unpublished data). In pools assigned the lethal-predator treatment, the single predator in each pool was initially caged for two days and then released; the empty cage remained in each pool. In pools assigned the test-predator treatment, the predators were caged for only a few hours and then released to allow predation to begin.

Each pool initially contained 60 treefrog tadpoles drawn haphazardly from a mixture of 12 sibships collected at Cornfield Pond, located in Livingston County, Michigan, USA. This density ($\sim 60/m^2$) is on the high end of what we observe in natural ponds (up to $53/m^2$; R. A. Relyea, E. E. Werner, D. K. Skelly, and K. L. Yurewicz, unpublished data), but initial densities of tadpoles are likely higher at the time of hatching in natural ponds and decline over time due to predation and competition. This decline from a dense population is precisely what I wanted to observe. I first added tadpoles to the nine test-predator pools (added on 17 June 1998, mean mass = 117 mg). Two days later, I added tadpoles to the remaining 42 pools (mean mass = 63 mg), and kept the lethal predators in their cages for two days to allow the tadpoles to acclimate to the predator's presence, and to allow the tadpoles to grow to a more similar size as those placed in the test-predator pools. The lethal predators were released on 21 June 1998, providing a four-day time difference be-

tween the test predators and the lethal predators. The test-predator pools were destructively sampled to provide me with an advanced estimate of the rate of thinning that was occurring in the undisturbed lethal-predator pools. Every four days, I removed all leaves and tadpoles from three randomly selected test-predator pools and counted the survivors. Based on this mortality rate, I reduced pools assigned to the thinning treatments by the same amount; I also moved a net through the non-thinned pools to equalize disturbance among all pools. Thus, the reduction in tadpole density by hand thinning was similar to the reduction in tadpole density due to predation. Once thinned, I weighed the tadpoles that were removed.

Because behavioral and morphological responses of prey likely depend upon the amount of chemical cue that is produced when predators consume tadpoles (Petranka et al. 1987, Kats et al. 1988, McCollum and Van Buskirk 1996), it was important that the tadpole mass consumed by the caged predators was similar to the mass of tadpoles being consumed by the lethal predators. One can estimate the mass of tadpoles consumed by the lethal predators by multiplying the number of tadpoles consumed during a time interval with the mean mass of tadpoles (during the same time interval) in pools that experienced both thinning and caged predators. Based on these estimates, I fed the caged predators the appropriate mass of prey every two days using the tadpoles that were removed during thinning.

During the experiment, I conducted behavioral observations to quantify the activity level of the tadpoles. On day 11 of the experiment, I conducted 20 observations on each pool, counting the number of tadpoles that could be observed and the proportion of observed tadpoles that were active (moving). The mean of all 20 proportions was used as the response variable for each pool (see Relyea 2002a, c, for prior use of this protocol).

I ended the experiment when I estimated that the

lethal-predator treatment was approaching 20% survival (day 13, based on the test-predator pools), leaving ~12 tadpoles to measure. All leaves were removed from the pools, and the surviving tadpoles were counted and weighed to determine survivorship and growth (final – initial mass). One of the lethal-predator pools contained a dead dragonfly predator; thus, data from this pool was not collected. I preserved a sample of up to 10 tadpoles from each pool and later measured their morphology using Optimas image-analysis software (Optimas Bioscan, Bothell, Washington, USA). Preserved tadpoles were placed under a video camera that projected the image onto a video screen, and seven linear dimensions were calculated: tail length and maximum tail depth, tail muscle depth, body depth, and body length (all from the lateral view); and body width and tail muscle width (both from the dorsal view). Tadpole tails were elevated on a piece of glass to allow them to lie in a natural, relaxed position (see Fig. 1 in Relyea 2000).

When examining morphological changes, it is important to account for differences in morphology due to differences in overall size, as well as differences in shape. In the current experiment, I was interested in shape differences, so I removed differences due to overall size by conducting a single regression on the seven linear dimensions of all individuals in the experiment against mass (all variables were log-transformed to improve linearity), and saving the residuals (Bookstein 1991). These residuals represent size-adjusted morphology. I then calculated the mean residual values for each pool, which represent the relative morphology for that pool. This is a commonly used protocol for quantifying the relative morphology of tadpoles (Van Buskirk and Relyea 1998, Relyea 2000).

Statistical analyses

I analyzed the 10 responses (survivorship, growth, activity, and seven morphological traits) using three multivariate analyses of variance (MANOVA, SYSTAT version 5). For all three analyses, the data met the assumptions of the analysis and, thus, were not transformed. After determining that a given multivariate test was significant, I examined whether the univariate responses were significant. For significant univariate responses in any of the three analyses, I conducted mean comparisons using Fisher's exact test. In the first analysis, I determined if there were differences in the responses among any of the seven treatments (the test-predator treatment was not included).

In the first analysis, several of the mean comparisons examined the same predatory process (e.g., there were four different comparisons that could estimate the impact of induction; Table 1). To determine the overall (average) impact of each process across different environmental contexts (e.g., the impact of thinning with and without a caged predator present), I conducted a second analysis. I defined each of the seven treatments

as containing either: (1) the presence or absence of predator induction, (2) the presence or absence of thinning (either by hand or via lethal predator), and (3) the presence or absence of selection. These assignments created an incomplete factorial design (some combinations were not possible; e.g., not-thinned, but selected). I then used a MANOVA to determine the impacts of induction, thinning, and selection on prey phenotypes across all environmental contexts.

Whereas the first two analyses determined if the seven treatments affected prey phenotypes and if the three predatory processes affected prey phenotypes across different environmental contexts, the analyses could not compare the relative magnitude of the processes in isolation and in combination. Therefore, my objective in the third analysis was to quantify the relative magnitude of response for a particular process (e.g., induction plus selection). To accomplish this objective, I focused on an appropriate treatment pair (e.g., caged-predator/selective thinning vs. no-predator/random thinning), calculated the difference between the first replicates of the two treatments, and divided the difference by the phenotypic value expressed in the first replicate of the no-predator/no-thinning treatment. This tells us how a particular predatory process alters the tadpole phenotypes relative to a no-predator environment. I then continued to calculate relative magnitudes for the second, third, fourth, fifth, and sixth replicates. If there was another treatment pair that also could be used to estimate the same process combination (e.g., lethal-predator vs. no-predator, random thinning), I repeated the above algorithm. Because each process combination was estimable from one to four original treatment pairs (using five to six replicates per treatment pair; Table 1), the seven combinations of induction, thinning, and selection were represented by 6–23 treatment differences. Analyses of variance are robust to such unbalanced designs providing that the other assumptions are not violated, and this proved to be the case in my data.

RESULTS

Analysis of the pool treatments

The pool treatments caused a significant multivariate effect on tadpole phenotype, and univariate tests indicated that survival, growth, behavior, and morphology were all affected (Table 2; all P values below refer to mean comparisons). The rate of hand thinning (both random and selective thinning in no-predator and caged-predator treatments) closely tracked the reduced survival observed in the lethal predator pools that were destructively sampled over time (Fig. 1). Survivorship due to thinning by hand or by lethal predators was one-fourth the survivorship of non-thinned pools ($P < 0.00001$; Fig. 2). Survival in the hand-thinned treatments and lethal-predator treatment did not differ ($P > 0.1$).

Tadpole growth also differed among treatments (Ta-

TABLE 2. MANOVA and ANOVA results of examining the effect of the seven pool treatments on the survival, growth, activity, and relative morphology of larval treefrogs.

Variable	df	F	P
MANOVA			
Treatment	60, 136	5.13	<0.00001
ANOVAs			
Survival	6, 34	57.46	<0.00001
Growth	6, 34	14.63	<0.00001
Activity	6, 34	11.53	<0.00001
Tail length	6, 34	1.62	0.171
Tail depth	6, 34	24.72	<0.00001
Muscle depth	6, 34	2.69	0.030
Muscle width	6, 34	2.07	0.083
Body length	6, 34	24.47	<0.00001
Body depth	6, 34	0.72	0.634
Body width	6, 34	2.28	0.059

ble 2, Fig. 2). In both the absence and presence of caged predators, growth increased when the population was thinned ($P < 0.0005$). On average, the five thinning treatments increased tadpole growth by 74% over the two non-thinned treatments. The addition of caged predators had no effect on growth in non-thinned pools, or in selectively thinned pools ($P > 0.1$), but there was a small increase in growth when caged predators were added to randomly thinned pools ($P = 0.049$). In the presence of the lethal predator, growth was intermediate between the caged predator treatments with no thinning ($P = 0.010$), and those with random or selective thinning ($P < 0.007$).

Tadpole activity also was affected (Table 2, Fig. 2). Within either the no-predator or caged-predator treatments, tadpoles in thinned pools had lower activity than tadpoles in non-thinned pools ($P < 0.04$). Within each of the three thinning treatments, the presence of caged predators reduced activity ($P \leq 0.03$). The activity of tadpoles experiencing lethal predators did not differ from the activity of tadpoles experiencing caged predators with either type of hand thinning ($P > 0.18$).

The relative morphology of larval treefrogs was altered by the pool treatments (Table 2, Fig. 3). Caged predators caused an increase in tail depth ($P < 0.0008$) and a decrease in body length ($P \leq 0.0001$) within each

hand-thinning treatment, but caged predators had no effect on the remaining morphological traits ($P > 0.05$). Hand thinning in the absence of caged predators had no effect on tail depth ($P > 0.4$), muscle depth ($P > 0.05$), or body length ($P > 0.35$). However, hand thinning in the presence of predators caused deeper tails ($P \leq 0.01$) and shorter bodies (selective thinning, $P = 0.014$; but for random thinning, $P = 0.121$). Body width experienced a nearly significant effect of pool treatments ($P = 0.059$); in the absence of predators, random and selective thinning caused narrower bodies ($P = 0.011$ and $P = 0.059$, respectively). However in the presence of predators, tadpoles exhibited narrower bodies with random thinning ($P = 0.047$), but not with selective thinning ($P = 0.374$). Hand-thinning had no effect on the remaining dimensions ($P > 0.05$). In comparisons of caged-predator/hand-thinned treatments and the lethal-predator treatments, there were never any differences in relative morphology ($P > 0.3$).

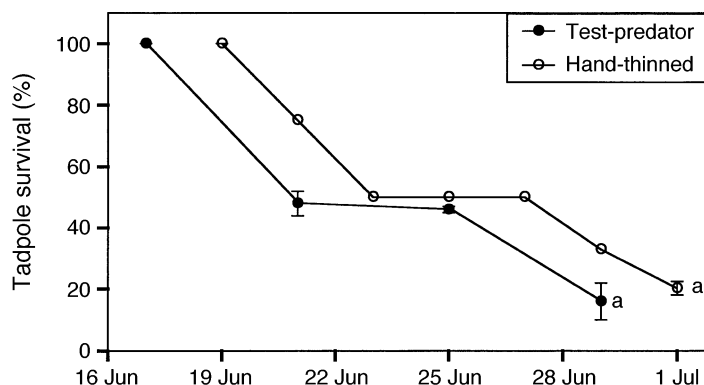
The overall effects of induction, thinning, and selection

In the second analysis, I examined the overall (average) impacts of induction, thinning, and selection on the prey phenotypes across the different environmental contexts (Tables 3 and 4). Both thinning and induction caused significant multivariate impacts on prey phenotypes, but selection did not. Thinning reduced activity and increased growth, while induction reduced activity but had no effect on growth. Induction caused longer and deeper tail fins, deeper tail muscles, and shorter bodies. Thinning also caused deeper tail fins and tail muscles, as well as shorter and narrower bodies. The remaining morphological dimensions were unaffected.

The relative magnitudes of induction, thinning, and selection alone and in combination

In the third analysis, I compared the relative magnitudes of induction, thinning, and selection both separately and in combination with each other (Fig. 4, Table 5). The different combinations had a significant multivariate effect on the prey phenotypes. Growth was impacted primarily by thinning; adding induction, selection, or both processes to thinning did not signifi-

FIG. 1. Survivorship over time for treefrog tadpoles in test-predator pools and hand-thinned pools. The test-predator treatments had lethal predators that were released two days prior to initiating hand-thinned treatments (thus the time lag). The density estimates for the hand-thinned treatments are based on the difference between the original density (60 tadpoles/pool) and the number of tadpoles removed with a dipnet over time. Thus, with the exception of the final value, there are no estimates of variation around the means. The final hand-thinned value was obtained by destructively sampling the pool and counting all survivors. Data are means \pm 1 SE. Means with common letters are not significantly different ($P > 0.05$).



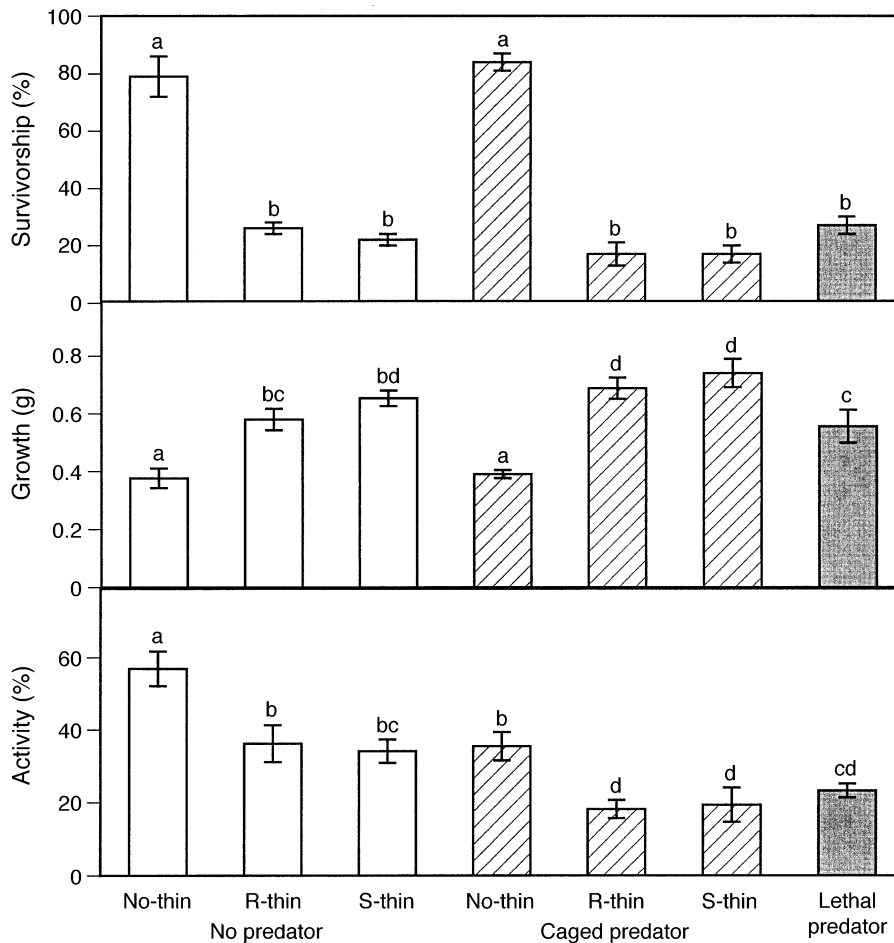


FIG. 2. Survivorship, growth, and activity of gray treefrog tadpoles when reared in either the absence (open bars) or presence (hatched bars) of caged predators combined with either no thinning (No-thin), random thinning (R-thin), or selective thinning (S-thin). A seventh treatment was the presence of a lethal, uncaged predator (shaded bar). Data are means \pm 1 SE. Means with common letters are not significantly different ($P > 0.05$).

cantly alter growth ($P > 0.3$). Activity was impacted by induction and thinning equally ($P = 0.300$), and each had greater impacts than selection alone ($P < 0.0001$). Adding selection to either process had no effect ($P > 0.7$). However, combining induction and thinning caused a larger activity reduction than either process alone ($P < 0.006$), and was similar to all three process combined ($P = 0.713$).

The relative impacts on morphology also differed among the three processes. Tail depth changes were dominated by induction as compared to thinning and selection ($P < 0.0001$). The impact of induction alone was similar to induction combined with thinning, selection, or both ($P > 0.08$). Muscle depth changes were similarly affected by the three processes ($P > 0.06$), and they were all similar to the two-way combinations ($P > 0.05$). Body length changes were dominated by induction rather than thinning or selection ($P < 0.00001$). In fact, induction combined with either thinning, selection, or both did not alter body length any

more than induction alone ($P > 0.06$). Body width changes were dominated by thinning more than induction or selection ($P < 0.02$); the addition of induction, selection, or both had no further impact on body width ($P > 0.23$). There were no differences in the relative impacts of the process combinations for tail length, muscle width, and body depth.

DISCUSSION

The data soundly reject the hypothesis that each of the three processes cause similar absolute impacts on the final phenotypes of the prey. This experiment critically relied upon a removal rate by hand that matched the removal rate by the lethal predator. By having an advanced estimate of the lethal predator's removal rate (based on the test-predator pools), I was able to remove tadpoles at a similar rate, and terminate the experiment with survival in the hand-thinning treatments that was similar to survival with lethal predators. Thus, any differences in tadpole phenotypes between the hand-

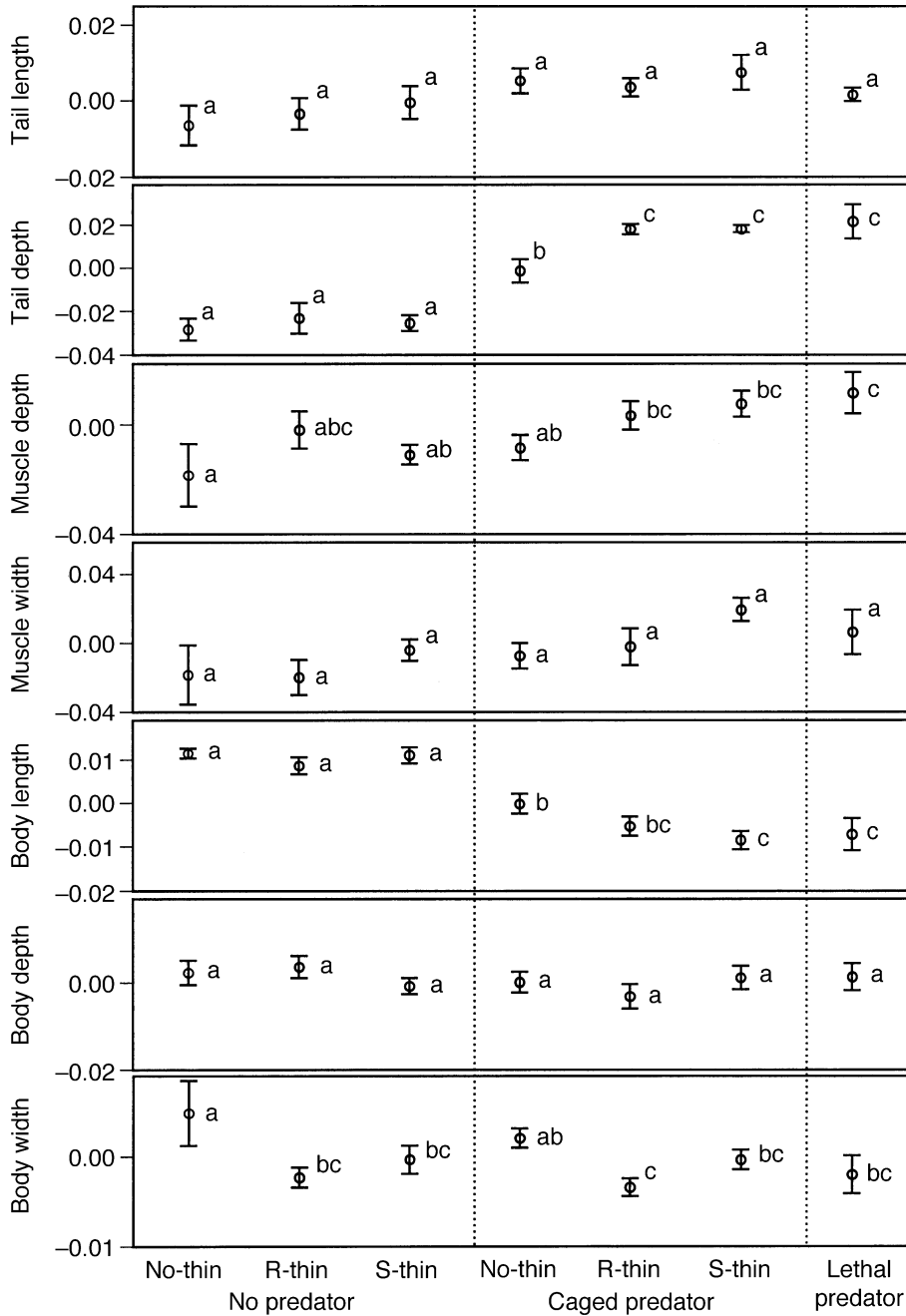


FIG. 3. Relative morphology of treefrog tadpoles when reared in either the presence or absence of caged predators, combined with no thinning (No-thin), random thinning (R-thin), or selective thinning (S-thin). A seventh treatment was the presence of a lethal, uncaged predator. Differences in morphology due to differences in overall size were removed prior to analysis by regressing all dimensions against tadpole mass and saving the residuals (mass-independent values on the y-axis). Data are means \pm 1 SE. Means with common letters are not significantly different ($P > 0.05$).

thinned and predator-thinned treatments were not due to differences in density over time. Below I discuss the effect of each process alone and then in combination.

Induction

Induction by predatory chemical cues caused the tadpoles to alter their phenotype in directions that were

consistent with past work. The tadpoles became less active in the presence of predators, a common response by many taxa (Sih 1987, Lima and Dill 1990, Kats and Dill 1998). Reduced activity makes prey less likely to encounter predators, and improves their probability of survival (Gerritsen and Strickler 1977, Houston et al. 1993, Werner and Anholt 1993, Skelly 1994, Relyea

TABLE 3. MANOVA results of examining the effect of predator induction, thinning, and selection on the survival, growth, activity, and relative morphology of larval treefrogs.

Source	df	Wilks' <i>F</i>	<i>P</i>
Induction	10, 26	13.2	<0.00001
Thinning	10, 26	24.2	<0.00001
Selection	10, 26	0.9	0.575
Induction × Thinning	10, 26	1.0	0.501
Induction × Selection	10, 26	1.6	0.162

2001a). The predator-induced increase in tail depth and decrease in body length has been documented in a number of larval anurans, including gray treefrogs (both *H. versicolor* and *H. chrysocelis*), chorus frogs (*Pseudacris triseriata*), wood frogs (*Rana sylvatica*), and leopard frogs (*R. pipiens*; Smith and Van Buskirk 1995, McCollum and Van Buskirk 1996, Relyea 2000, Relyea and Werner 2000). These morphological responses are adaptive; tadpoles with deeper tails and shorter bodies escape dragonfly predators better than tadpoles with the opposite morphology (Van Buskirk et al. 1997, Van Buskirk and Relyea 1998).

There was no overall effect of induction on tadpole growth. When antipredator responses reduce foraging ability, the costs of the responses must depend upon the rate of resource replacement and on the density of competitors. For example, in larval anurans, the addition of predatory cues causes relatively large growth reductions at low competitive densities, but small growth reductions at high competitive densities (Werner and Anholt 1996, Van Buskirk and Yurewicz 1998, Peacor and Werner 2000). Further, predator-induced growth reductions are common early in ontogeny, but disappear and sometimes even reverse later in ontogeny (Van Buskirk and Yurewicz 1998, Relyea and Werner 2000, Relyea 2001a). These changes may reflect either changes in resource recruitment rate or changes in antipredator responses as prey become less vulnerable with increased size. In the current experiment, we terminated growth relatively late in larval ontogeny, and thus found no effect of induction on growth.

Thinning

Thinning the tadpole population had a dramatic effect on tadpole traits. The associated increase in tadpole growth suggested that the tadpoles were released from competition, a result that has been commonly observed in larval anurans as well as many other taxa (Wilbur 1977, Connell 1983, Schoener 1983, Werner and Anholt 1996, Relyea 2000). The accompanying decrease in activity (over and above decreases caused by predator induction) further suggests that the treefrog larvae were competing. Decreased activity is an adaptive response to increased resources, allowing prey to spend a lower proportion of their time searching for food to meet their energy needs (Abrams 1991, Werner and Anholt 1993).

Whereas we have a good understanding of how tadpoles and many other taxa alter their morphology in response to predator cues, we are only beginning to understand how prey alter their morphology in response to reduced competition (Harvell 1990, Pfennig 1992a, b, Reilly et al. 1992, Tollrian and Harvell 1999, Pfennig and Murphy 2000, Relyea 2000). The changes in morphology associated with thinning (increased tail depth and decreased body volume) were similar to responses observed in wood frog tadpoles (Relyea 2002a). When wood frogs experience high interspecific or intraspecific competition, they develop relatively shallower tails and larger bodies. Competitor-induced tadpoles are more competitive at the cost of reduced predator resistance. These morphological responses are fundamentally different than the cannibalistic morphs exhibited by a few amphibian species (e.g., Pfennig 1992a); wood frogs and gray treefrogs do not develop carnivorous mouthparts. In both species, the tadpoles use a combination of resource level cues and density cues to assess their intensity of competition, and they exhibit competitor-specific responses (Relyea 2002a; R. A. Relyea, unpublished data).

Curiously, thinning affected tadpole morphology only when predators were present. Tadpoles that increased their tail depth and decreased their body length in the presence of predators developed even more extreme phenotypes when competition was reduced (Fig. 3). Because predation resistance is conferred by deeper tails and shorter bodies, whereas competitive ability is conferred by shallower tails and longer bodies, this result suggests that tadpoles can simultaneously evaluate predation risk and competition intensity, and trade off these phenotypic demands. In contrast, when predators were absent, tadpoles should maximize their competitive ability. The lack of a more extreme morphological response under no thinning/no predators (Fig. 3) suggests that the tadpoles were already at the limit of their morphological dimensions and could not make further changes. Such limits are hypothesized to arise from a number of physiological and genetic constraints

TABLE 4. Univariate results from ANOVA tests for the effects of predator induction and thinning, the two main effects that caused significant multivariate impacts on prey traits (see Table 3).

Variable	Induction		Thinning	
	<i>F</i> _{1,35}	<i>P</i>	<i>F</i> _{1,35}	<i>P</i>
Survival	0.5	0.503	226.9	<0.00001
Growth	0.1	0.821	35.4	<0.00001
Activity	16.9	0.0001	26.6	0.00001
Tail length	5.9	0.021	0.0	0.867
Tail depth	70.9	<0.00001	6.6	0.014
Muscle depth	6.3	0.017	4.4	0.042
Muscle width	2.9	0.099	0.0	0.855
Body length	84.4	<0.00001	4.6	0.039
Body depth	0.0	0.989	0.1	0.707
Body width	0.7	0.396	12.4	0.001

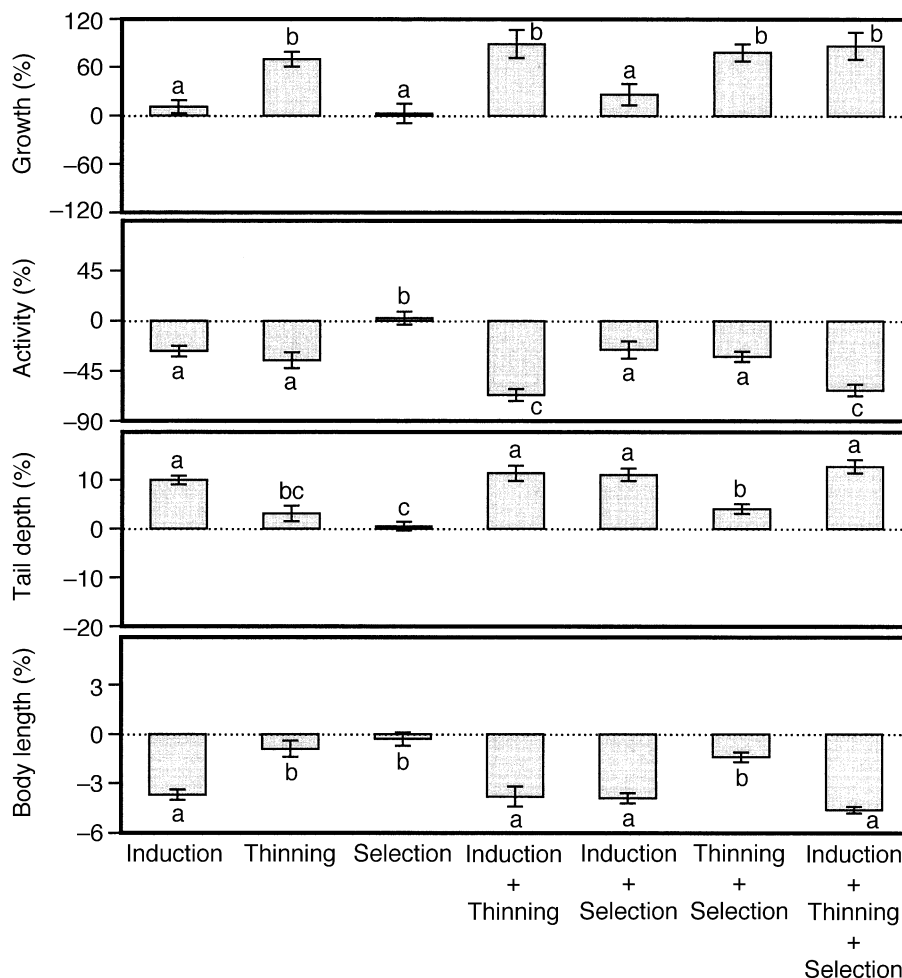


FIG. 4. The separate and combined relative contributions of induction, thinning, and selection on prey growth, activity, tail depth, and body length. The contributions were made relative to the phenotypes exhibited in the absence of all three processes. Data are means \pm 1 SE. Means with common letters are not significantly different ($P > 0.05$).

TABLE 5. MANOVA and ANOVA results of examining the relative contributions of seven combinations of predator processes on prey phenotypes (induction, thinning, selection, induction plus thinning, induction plus selection, thinning plus selection, and induction plus thinning plus selection).

Variable	df	F	P
MANOVA			
Treatment	54, 422	0.1	<0.00001
ANOVAs			
Growth	6, 90	10.4	<0.00001
Activity	6, 90	13.4	<0.00001
Tail length	6, 90	2.0	0.071
Tail depth	6, 90	16.8	<0.00001
Muscle depth	6, 90	2.3	0.041
Muscle width	6, 90	1.3	0.256
Body length	6, 90	22.2	<0.00001
Body depth	6, 90	0.5	0.839
Body width	6, 90	5.8	0.00004

(DeWitt et al. 1998, Relyea 2002b). An intriguing question for future studies would be to investigate how well prey can finely tune their morphology to trade off their predation risk and competitive ability.

Selection

Nearly all of the analyses concluded that selection played no significant role in affecting the final phenotypes of the tadpoles. In the first analysis of all seven pool treatments, there were three individual comparisons with which I could assess the importance of selection. In the first two comparisons, I compared randomly thinned vs. selectively thinned tadpoles (in either no-predator or caged-predator environments) and found no phenotypic differences. This lack of phenotypic change might be because the small net used for thinning simply did not select on tadpole phenotypes even though personal observations indicated that the small net missed many of the tadpoles during capture attempts. Alternatively, it might be that the small net

was selective, but there was no signal of the selection by the end of the experiment. In the third selection comparison (caged predators plus random thinning vs. lethal predators), there also were no phenotypic differences (with the exception of a small growth increase). However, we know from numerous previous experiments (using overnight predation trials) that larval dragonflies preferentially kill tadpoles with relatively shallow tails (chorus frogs, Van Buskirk et al. 1997; wood frogs, Van Buskirk and Relyea 1998; gray treefrogs, R. A. Relyea, *unpublished data*). Thus, selection was likely taking place in the third comparison (and possibly the first and second comparisons), but there was no signal of the selection by the end of the experiment.

At first thought, it seems paradoxical that predators could cause selection on tadpole phenotypes, yet there is no signal of selection by the end of the experiment. Selection preferentially removes tadpoles with the shallowest tail fins, thus causing a truncation of the phenotypic distribution. However, for there to be no impact of this truncation at the end of the experiment, there must be a shift in phenotype by the survivors in the population toward the shallow-tailed end of the distribution. That is, selection that causes an increase in mean relative tail depth must have been opposed by the surviving tadpoles decreasing their relative tail depth. Whereas tadpoles that adopt this shallow-tailed phenotype would be at a higher risk of predation, this phenotype would make them more competitive, providing them with a higher growth rate and more rapid development (Relyea 2002*b*). This hypothesis requires that tadpoles are able to sense changes in their competitive environment, alter their morphology at a relatively rapid rate, and be able to reverse morphological inductions to at least a small degree. Recent data supports all three of these requirements (Relyea 2002*a*; R. A. Relyea, *unpublished data*). While this appears to be the most likely mechanism to explain the phenomenon, it clearly requires more extensive investigation.

Combining induction, thinning, and selection

The primary objective of this study was to determine the relative impacts of the three predatory processes (induction, thinning, and selection) on a prey population's phenotype within a given generation. The relative importance of each process depended upon the trait being examined. Lethal predators affected growth strictly through thinning, and tail depth and body length mostly through induction, and to a small degree through thinning. Behavior was equally affected by thinning and induction. As noted above, selection never played a significant role in determining the traits. These results are probably not limited to aeshnid predators; several other predator species have similar effects on tadpole phenotypes via induction, thinning, and selective thinning (Relyea 2001*a*; R. A. Relyea, *unpublished data*).

Few studies have attempted to separate the relative impacts of these three processes. Van Buskirk and Yurewicz (1998) separated the impacts of induction and thinning on prey growth and activity; they found that predators primarily affected activity through induction and secondarily through thinning, while predators affected growth through both induction and thinning. The current study builds on this foundation and appears to be the first to examine the role of all three processes on prey growth, behavior, and morphology. With the very limited amount of current data on this issue, it appears that the relative contributions of the three predatory processes may depend upon prey density and predation rate, but it remains much too early in our investigations to propose any reliable generalities. It is possible that the roles of induction and thinning are amplified in aquatic mesocosms to be stronger than those experienced in natural ponds (Skelly and Kiesecker 2001).

Induction, thinning, and selection probably impact the final phenotypes of many prey populations. Aquatic communities, in particular, have numerous examples of prey that experience these processes, including protists, rotifers, cladocerans, gastropods, bivalves, decapods, larval amphibians, and fish. However, this phenomenon is not necessarily restricted to aquatic systems; many terrestrial communities contain prey populations that are simultaneously thinned by predators and alter their behavioral phenotypes in the presence of predators (Lima and Dill 1990, Kats and Dill 1998, Tollrian and Harvell 1999). Presumably, these prey also undergo selection by their predators. Examining these processes is important because they can have opposing or supporting effects, making it difficult to determine the importance of each process. For example, predator-induced behavior and morphology typically decrease prey growth, but predator thinning of the prey population typically increases prey growth. Thus, when we examine the impact of lethal predators on prey growth, the net effect can be highly variable among studies because the relative magnitudes of the two processes likely vary among systems (Morin 1983, Figiel and Semlitsch 1990, Wilbur and Fauth 1990, Gascon and Travis 1992, Semlitsch 1993, Diehl and Eklöv 1995, McIntosh and Townsend 1996, Moran et al. 1996). Yet in many cases, the authors assume that one of the processes dominates the others without any direct evidence. Only when we document the magnitudes of each process can we begin to understand the underlying mechanisms that produce different outcomes in different systems.

CONCLUSIONS

In this study, I have demonstrated the importance of understanding the relative impacts of predators on numerous prey traits. It is clear that this information is critical for ecologists to understand the underlying mechanisms that cause changes in the traits and per-

formance of prey populations in nature. As we gain further insights from other systems in the coming years, we will begin to understand the conditions under which induction, thinning, and selection differentially impact a prey population's behavior, morphology, and growth, and how these impacts affect the evolution and ecology of both predators and prey in the larger community.

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