

Eastern Illinois University
The Keep

Masters Theses

Student Theses & Publications

1-1-2005

Resilience of an amphibian community following the removal of introduced fish

Leroy J. Walston

Eastern Illinois University

This research is a product of the graduate program in [Biological Sciences](#) at Eastern Illinois University. [Find out more](#) about the program.

Recommended Citation

Walston, Leroy J., "Resilience of an amphibian community following the removal of introduced fish" (2005). *Masters Theses*. 928.
<http://thekeep.eiu.edu/theses/928>

This Thesis is brought to you for free and open access by the Student Theses & Publications at The Keep. It has been accepted for inclusion in Masters Theses by an authorized administrator of The Keep. For more information, please contact tabruns@eiu.edu.

*******US Copyright Notice*******

No further reproduction or distribution of this copy is permitted by electronic transmission or any other means.

The user should review the copyright notice on the following scanned image(s) contained in the original work from which this electronic copy was made.

Section 108: United States Copyright Law

The copyright law of the United States [Title 17, United States Code] governs the making of photocopies or other reproductions of copyrighted materials.

Under certain conditions specified in the law, libraries and archives are authorized to furnish a photocopy or other reproduction. One of these specified conditions is that the reproduction is not to be used for any purpose other than private study, scholarship, or research. If a user makes a request for, or later uses, a photocopy or reproduction for purposes in excess of "fair use," that use may be liable for copyright infringement.

This institution reserves the right to refuse to accept a copying order if, in its judgment, fulfillment of the order would involve violation of copyright law. No further reproduction and distribution of this copy is permitted by transmission or any other means.

THESIS REPRODUCTION CERTIFICATE

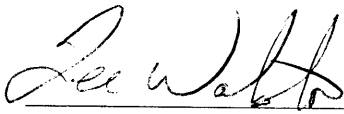
TO: Graduate Degree Candidates (who have written formal theses)

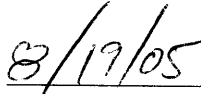
SUBJECT: Permission to Reproduce Theses

The University Library is receiving a number of request from other institutions asking permission to reproduce dissertations for inclusion in their library holdings. Although no copyright laws are involved, we feel that professional courtesy demands that permission be obtained from the author before we allow these to be copied.

PLEASE SIGN ONE OF THE FOLLOWING STATEMENTS:

Booth Library of Eastern Illinois University has my permission to lend my thesis to a reputable college or university for the purpose of copying it for inclusion in that institution's library or research holdings.

 _____

 _____

Author's Signature

Date

I respectfully request Booth Library of Eastern Illinois University **NOT** allow my thesis to be reproduced because:

Author's Signature

Date

This form must be submitted in duplicate.

RESILIENCE OF AN AMPHIBIAN COMMUNITY FOLLOWING
THE REMOVAL OF INTRODUCED FISH

by

Leroy J. Walston

THESIS

Submitted in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE in BIOLOGICAL SCIENCES

In the Graduate School, Eastern Illinois University

Charleston, Illinois

2005

I hereby recommend that this thesis be accepted as fulfilling this part of the graduate
degree cited above

Date

Thesis Director

Date

Department/School Head

Abstract

Introduced species have become one of the most important anthropogenic impacts in aquatic ecosystems and are implicated in the declines of many native amphibian populations. Despite the effects of introduced species on amphibian populations, there is little information concerning the responses of amphibians following the removal of introduced fish. I conducted a field experiment to assess the amphibian community-level responses to the removal of introduced fish, and determine the effects of fish removal on smallmouth salamander (*Ambystoma texanum*) larval dynamics. Species diversity of the amphibian community and smallmouth salamander juvenile recruitment improved following the removal of fish. My results indicate that introductions of predaceous fish can have adverse effects on amphibians at the community and population levels. Furthermore, my results indicate that amphibian communities might be capable of rapid recovery following the removal of introduced fish.

Table of Contents

Abstract	i
Dedication	iii
Acknowledgments	iv
List of Tables	v
List of Figures	vi
Introduction	1
Methods	6
Results	11
Discussion	14
Literature Cited	22
Appendix I	38

Dedication

To my wife, Meredith, for her love and support over the past two years while I was completing this thesis research.

Acknowledgments

Funding for this research was provided by the Illinois Department of Natural Resources (IDNR) Wildlife Preservation Fund, The Graduate School at Eastern Illinois University (EIU), and the EIU College of Sciences. Field work was conducted under the authorization of IDNR Scientific Collection Permit #NH04-0946 and an IDNR Nature Preserves Commission Permit “Mullin.” Laboratory procedures were in accordance with EIU’s Institutional Animal Care and Use Committee (protocol #04-008). I would like to thank my committee members, Profs. Stephen Mullin, Eric Bollinger, and Kipp Kruse for all of their advice and support in the development of my thesis. I would also like to thank Prof. Scott Meiners for his advice and answers to many of my statistical questions. The following people have assisted with field work and/or data collection: B. Towey, E. Casey, C. Foster, D. Cox, J. Florey, S. Klueh, M. Sikich, M. Fincel, and M. Walston. Special mention is warranted to L. B. Hunt and B. Szafoni for permission to conduct field work.

List of Tables

Table 1. Relative abundance (proportion) of amphibians captured at mitigated (n = 2) and reference (n = 2) ponds at Warbler Woods Nature Preserve, Coles County, Illinois, between the temporal periods of pre- and post-Rotenone™ application. Shannon-Weiner Diversity Index values (H') are displayed for both pond types during each temporal period.

Table 2. Results of the analysis of covariance (ANCOVA) that tested for the effects of temporal period (pre-application or post-application) and pond type (mitigated or reference) on growth (size at metamorphosis; SVL) of smallmouth salamanders (*Ambystoma texanum*) at WWNP between 2001 and 2004. Time to metamorphosis (Julian date of metamorphosis) was used as a covariate (df = 1,1297 in all cases).

Table 3. Results of the analysis of variance (ANOVA) that tested for the effects of temporal period (pre-application or post-application) and pond type (mitigated or reference) on the juvenile recruitment of smallmouth salamanders (*Ambystoma texanum*) at WWNP between 2001 and 2004 (df = 1,12 in all cases).

List of Figures

Figure 1. A portion of Warbler Woods Nature Preserve (WWNP), Coles County, Illinois, with the location of the four experimental ponds. Fish were introduced into mitigated ponds (Ponds B and C) prior to the commencement of this study, whereas reference ponds (Ponds A and D) never contained fish.

Figure 2. Size (mm) of metamorphosing juvenile smallmouth salamanders (*Ambystoma texanum*) as a function of time (month of emergence). Juveniles emigrating from mitigated ponds were larger than those from reference ponds throughout the study period (2001-2004), and size was positively associated with length of the larval period (Julian date of metamorphosis). All individuals metamorphosed between May and November for every year of the study period. Data are presented as means \pm 1 SE.

Figure 3. Changes in larval smallmouth salamander (*Ambystoma texanum*) growth (size at metamorphosis) between temporal periods (pre-application and post-application) and pond types (mitigated or reference) at Warbler Woods Nature Preserve, Coles County, Illinois. Juveniles emigrating from mitigated ponds were larger than those from reference ponds throughout the study period (2001-2004). All data are presented as means \pm 1 SE.

Figure 4. Changes in smallmouth salamander (*Ambystoma texanum*) juvenile recruitment (proportion of metamorphosing juveniles to number of breeding females) in mitigated and reference ponds between temporal periods (pre- and post-application of Rotenone™) at WWNP. All data are presented as means \pm 1 SE.

Introduction

Pond-breeding amphibians are important members of wetland ecosystems. As larvae, anurans typically feed on primary production such as phytoplankton and periphyton (Dickman 1968; Kupferberg et al. 1994) whereas caudates are consumers of secondary production such as aquatic insects and other amphibian larvae (Brophy 1980; Holomuzki et al. 1994). In many aquatic ecosystems, amphibians comprise the greatest amount of protein biomass available to organisms of higher trophic levels (Burton and Likens 1975; Blaustein et al. 1994), suggesting the importance of amphibian populations in the energy dynamics of these ecosystems (Holomuzki et al. 1994; Blaustein et al. 1996). Thus, fluctuations in amphibian populations undoubtedly impact other wetland taxa. Because of their indirect life cycles, consisting of an aquatic larval stage and a terrestrial adult stage, pond-breeding amphibians have complex habitat requirements and are susceptible to both aquatic and terrestrial stressors. Consequently, pond-breeding amphibians often serve as valuable indicators of environmental integrity (Blaustein 1994; Collins and Storfer 2003).

Pond-breeding amphibians exist as metapopulations (Sjogren 1991) that experience frequent local extinctions and high rates of population turnover (Hecnar and M'Closkey 1996; Skelly et al. 1999). Their populations are often regulated by a number of abiotic and biotic factors including hydroperiod (the length of time a pond continuously contains water; Pechmann et al. 1989; Phillips et al. 2002), larval density (Scott 1990), and precipitation (Jensen et al. 2003). Although natural populations are known to annually fluctuate by as many as 1-2 orders of magnitude (Pechmann and

Wilbur 1994), concern about population declines exceeding the limits of what are considered normal population fluctuations is growing (Alford and Richards 1999; Collins and Storfer 2003). Factors such as disease (Muths et al. 2003), chemical contaminants (Sparling et al. 2001), introduced species (Kupferberg 1997; Kiesecker and Blaustein 1997; Knapp and Matthews 2000), and habitat fragmentation (Means et al. 1996; Kolozsvary and Swihart 1999) have been suspected in amphibian declines, and there is evidence that many of these factors act synergistically (Lips 1998; Collins and Storfer 2003).

The impacts of anthropogenic environmental perturbations in amphibian population declines have received considerable attention, particularly the effects of invasive species. In many aquatic systems, invasive species constitute the most important anthropogenic impact (U.S. Congress 1993; Lodge et al. 2000). Invasive species negatively impact amphibian populations in a variety of ways, including interspecific competition and predation (Kiesecker and Blaustein 1997; Kupferberg 1997; Knapp and Matthews 2000), and may be responsible for the extirpation of local amphibian populations (Bradford 1989). For example, the introduction of the bullfrog (*Rana catesbeiana*) in the western U.S. is implicated in the decline of native frog species through predation and competition by adult and larval bullfrogs, respectively (Kiesecker and Blaustein 1997, 1998; Kupferberg 1997). Similarly, sport fish have been introduced into many historically fishless lakes throughout the U.S. for recreational purposes (Bahls 1992), altering amphibian populations primarily through predation upon larvae (Kiesecker and Blaustein 1998; Tyler et al. 1998; Knapp and Matthews 2000).

A number of hypotheses exist to explain the decline of amphibian populations due to fish introductions. Certainly, many introduced fish species are capable of consuming amphibian eggs and larvae (Gamradt and Kats 1996; Tyler et al. 1998; Goodsell and Kats 1999), directly influencing local populations of amphibians by decreasing reproductive success and subsequent juvenile recruitment. Furthermore, females of many amphibian species avoid breeding in bodies of water containing fish (Kats and Sih 1992; Hopey and Petranka 1994), indicating that introduced fish may indirectly effect amphibian populations by limiting the number of potential breeding sites. Lastly, introduced fish may exacerbate the effects of an already established exotic species. For instance, Kiesecker and Blaustein (1998) reported that the synergistic effects of introduced fish and bullfrogs in the western U.S. have drastically reduced the survival of native anuran tadpoles, although the individual effects of introduced fish and bullfrogs were not as pronounced.

Predation is considered to be a major selective force in amphibian community dynamics (Hecnar and M'Closkey 1997), restricting some species to ephemeral wetlands that lack predators, or resulting in the evolution of adaptations that allow amphibians to coexist with predators such as fish. For many amphibians, the lack of defensive adaptations is the primary reason for their allotopic distribution with fish (Kats et al. 1988; Semlitsch 1988; Hecnar and M'Closkey 1997). Amphibians that are capable of coexistence with fish have developed antipredator defenses such as the use of chemical repellents (Kats et al. 1988), shifts in activity patterns (Petranka et al. 1987; Figiel and Semlitsch 1990; Hoffman et al. 2004), or chemically-mediated avoidance of predators (Petranka et al. 1987). Much of the current research has focused on behavioral

adaptations of amphibian larvae in response to predation risk (Kiesecker and Blaustein 1997; Relyea 2002, 2004; Hoffman et al. 2004). Recent experiments have suggested that some amphibian larvae respond to predators with phenotypically-plastic behaviors such as reduced activity and increased use of specific microhabitat refugia (Kiesecker and Blaustein 1997; Relyea 2002, 2004). Additionally, some amphibians respond to predation risk by altering diel patterns, shifting from diurnal activity periods to nocturnal foraging periods (Hoffman et al. 2004).

Although plastic behaviors might permit some amphibians to coexist with fish, there are often fitness costs associated with the expression of these traits. Reduced activity of larval amphibians, a common response to predation risk, often results in a decline in larval growth (Relyea 2002, 2004), thereby affecting population parameters such as size and age at metamorphosis (Figiel and Semlitsch 1990; Sih et al. 1992). Size at metamorphosis is often considered an indication of fitness as larger juveniles might have increased survival (Werner 1986; Altwegg and Reyer 2003) and increased reproductive potential as adults (Semlitsch 1985a; Semlitsch et al. 1988; Berven 1990). Juvenile recruitment into the adult population is also critical for maintaining amphibian populations, and variation in recruitment might have pronounced effects on future amphibian population densities (Semlitsch 1983; Semlitsch et al. 1996). Fish have been observed to reduce larval growth and juvenile recruitment of amphibian populations (Figiel and Semlitsch 1990; Sih et al. 1992; Tyler et al. 1998). Therefore, the addition of predators to aquatic systems, including fish, may have negative consequences on larval amphibian dynamics.

The effects of introduced fish on amphibian populations have been well documented (Bradford 1989; Kiesecker and Blaustein 1998; Tyler et al. 1998; Knapp and Matthews 2000), but there is a growing body of evidence indicating that these impacts can be reversed. Current research suggests that some amphibian species are particularly resilient following the removal of introduced fish (Knapp et al. 2001; Hoffman et al. 2004; Vredenburg 2004). Ecological resilience, in this context, refers to the rate at which a community or a species returns to its previous state once a perturbation is removed (*sensu* Knapp et al. 2001). In these situations, the removal of introduced fish has resulted in improved rates of colonization to the restored wetland by a single species of pond-breeding amphibian (Knapp et al. 2001; Vredenburg 2004) as well as shifts in larval behavior that appear to favor larval growth and survival (Hoffman et al. 2004). To my knowledge, however, no study has examined the amphibian community-level response following the removal of introduced fish. Additionally, no studies have investigated the effects of fish removal on amphibian larval growth and juvenile recruitment, metrics that may improve adult fitness and population stability (Semlitsch et al. 1988; Berven 1990; Altwegg and Reyer 2003).

Herein, I present the results of a field experiment designed to elucidate the effects of introduced fish on a pond-breeding amphibian community in east-central Illinois. The objectives of this study were to 1) assess the amphibian community-level responses to removal of introduced fish, and 2) determine the effects of introduced fish on larval growth and juvenile recruitment of the smallmouth salamander (*Ambystoma texanum*), a locally abundant pond-breeding amphibian.

Methods

Study Site

I collected data from May 2003 to November 2004 at Warbler Woods Nature Preserve (WWNP), an 81.5 ha parcel of land in Coles County, Illinois, under joint-jurisdiction of private ownership and the Illinois Department of Natural Resources (Fig. 1). My data were combined with data collected from May 2000 through April 2003 at this same site to provide four and a half years of continuous amphibian monitoring data. A mixture of old field upland habitat and deciduous oak-hickory hardwood forests dominate WWNP. Within the forested areas, four man-made ponds were created prior to 1985 (K. Kruse, pers. com). I labeled these ponds east to west alphabetically: A, B, C, and D. Ponds A and B are closest to each other, separated by a 5-m forested ridge. Ponds B and C are separated by 80 m of old field habitat, and Ponds C and D are separated by 280 m of old-field upland habitat (Fig. 1). The ponds are between 0.04 and 0.09 ha in size and, although each pond is completely surrounded by deciduous forest (terrestrial buffer), each pond is in close proximity (≤ 20 m) to different landcover types including active agricultural fields, old field growth, roads, and residential property. Vegetation in and immediately peripheral to the ponds included green algae, common duckweed (*Lemna minor*), Dudley's rush (*Juncus dudleyi*), stinging nettle (*Urtica dioica*), as well as a few other aquatic macrophytes. During the years of this study, all ponds possessed a permanent hydroperiod except for Pond D, which dried before 2 August of every year.

Sampling Procedure

Drift fence / pitfall trap arrays were constructed in May 2000, encircling each pond to monitor the movement of amphibians (modified from Corn 1994). The drift fence consisted of plastic-weave silt fence material, approximately 45 cm high with 10 cm buried beneath the soil surface to prevent individuals from eluding the traps by burrowing under the fence. The fence was supported upright, attached to wooden stakes that were driven into the ground. Pitfalls (3-L plastic tubs, 20-cm deep) were placed on both sides of the fence at 7.5-m intervals. I labeled every pitfall trap with a unique code to distinguish among ponds, individual buckets, and orientation (side of the fence). Additionally, I made small (< 2 mm diameter) punctures in the bottom of the buckets to allow water to drain during rain events and prevent captured organisms from drowning. This design is efficient in capturing juveniles of all species of amphibians at WWNP, and adults of those species not capable of escaping the pitfall traps. Adults of treefrogs (e.g., *Hyla* and *Pseudacris*) and large ranids (*R. catesbeiana* and *R. utricularia*), however, might be capable of escaping the pitfall traps. Due to inter-pond size variation, there were unequal numbers of pitfall traps among ponds. In total, there were 19, 23, 14, and 12 pairs of pitfall traps for Ponds A, B, C, and D, respectively. I placed lids on the buckets during periods of inactivity (early December to early February) to prevent mortality of non-target organisms such as small mammals. Excepting these periods, pitfalls were checked at least once every 48 h, depending on the season, from February 2001 through November 2004.

I measured the snout-vent length (SVL; ± 1 mm) for all amphibians captured in the pitfalls. I determined the sex of adult pond-breeding amphibians during the breeding season by inspecting the swollen cloacae of male salamanders (*Ambystoma*) and by inspecting the swollen nuptial pads on the fore-feet of male anurans. Depending on size, amphibians captured outside of the breeding season were identified as either “juveniles” for individuals that had undergone metamorphosis in the same year as they were captured, or “non-breeding adults” for individuals that had metamorphosed at least one year previously. I marked all captured amphibians to a year-by-pond specific cohort, using a pre-determined pattern of toe clipping (following Donnelly et al. 1994). All individuals were examined at time of capture and I omitted recaptures from the analyses.

Mitigation of Fish Introductions

During 1985 and 1986, centrarchids (primarily *Lepomis cyanellus*) were introduced into Pond C and black bullhead catfish (*Ameiurus melas*) were introduced into Pond B. Previous studies have demonstrated the ability of centrarchids and black bullhead catfish to have negative impacts on amphibian populations by depredating amphibian larvae (Kruse and Francis 1977; Figiel and Semlitsch 1990; Adams 2000). Therefore, the fish introduced into Ponds B and C at WWNP could have adverse effects on amphibian abundance. Intermittent field visits documented the presence of fish in both ponds between the mid-1980’s and mid-1990’s (K. Kruse, pers. com), and fish were still present within both ponds in 2000 when this study commenced.

Beginning in May 2000, amphibian activity was monitored at all four ponds via the drift fence / pitfall trap arrays. However, data collected in 2000 did not include

breeding adult census data; therefore, data collected during this year were omitted from analyses. Hence, only data collected from February 2001 through November 2004 were included in this study. In December 2001, Rotenone™ was applied to Ponds B and C to eradicate all fish. Rotenone™ is a plant-based isoflavonoid pesticide most commonly used in fish management (McClay 2000). Although Rotenone™ might be toxic to non-target organisms including amphibians (Fontenot et al. 1994), a recent report by Mullin et al. (2004) suggested that amphibians are less sensitive to the pesticide than fish and are capable of quickly recovering from exposure. Rotenone™ has a greater half-life in colder waters (Gilderhus et al. 1988), and is therefore most potent in lentic habitats when applied during the winter while most pond-breeding amphibians are dormant. Applied during this season, the poison is effective for less than a month (Fontenot et al. 1994), becoming inert before amphibian reproductive activity resumes the following spring. Bullfrog (*Rana catesbeiana*) tadpoles were the only amphibian to overwinter in the ponds at WWNP, and were therefore the only species exposed to Rotenone™ during application. Bullfrogs were ubiquitous throughout WWNP; therefore, recolonization of the treated ponds was likely should the poison have adversely affected overwintering tadpoles. The application of Rotenone™ in December 2001 did not completely remove all black bullhead catfish from Pond B and a second dose was applied in January 2003.

I grouped the four ponds into two categories: mitigated ponds, ponds in which fish had been present and subsequently removed (Ponds B and C); and reference ponds, ponds in which fish had never been present (Ponds A and D). Using the census data obtained from the drift-fence / pitfall trap arrays, I determined amphibian species richness at both pond types. Additionally, I used the mean relative species abundance in both

pond types and temporal periods to calculate the Shannon-Weiner Diversity Index ($H' = -\sum p_i \log_{10} p_i$), where p_i is the proportion of the i^{th} species. These results provided temporal metrics of species diversity (H') between pond types. Temporal periods were determined with respect to Rotenone™ application; the post-application period commenced after the 2001 season for Pond C and after the 2002 season for Pond B. The period between the 2002 and 2003 field season was used to discriminate temporal periods in reference ponds, as this was the final period in which Rotenone™ was applied to any of the mitigated ponds.

Ambystoma texanum Larval Growth and Recruitment

The smallmouth salamander (*Ambystoma texanum*) is a medium-sized (< 19 cm total length) pond-breeding salamander native to the central U.S. (Petranka 1998). In Illinois, *A. texanum* is abundant in poorly drained woodland habitats where adults breed in forest pools (Phillips et al. 1999). Adults were commonly observed breeding in all ponds at WWNP. I examined the effects of introduced fish on the population dynamics of *A. texanum*, particularly larval growth and juvenile recruitment. I quantified larval growth as the SVL (± 1 mm) of all juveniles emigrating from each pond. For each pond, I also determined annual juvenile recruitment by calculating the proportion of emigrating juveniles to the number of breeding females. Julian dates of metamorphosis were recorded for each captured juvenile and were used as a metric of larval period.

Statistical Analyses

To evaluate the resilience of the amphibian community following the removal of fish, I tested for temporal differences in species diversity (H') within and between pond types by performing paired t-tests (Zar 1999). For these analyses, a Bonferroni adjustment was made prior to comparisons, correcting the α -level to 0.0125.

I tested for the effects of introduced fish on smallmouth salamander larval growth by performing a 2 x 2 analysis of covariance (ANCOVA), with time period (pre-mitigation or post-mitigation) and pond type (mitigated or reference) as fixed factors. Time to metamorphosis (Julian date of emergence) was used as the covariate. In this analysis, I log-transformed juvenile SVL and square-root transformed the Julian dates of metamorphosis to normalize the data. To test for the effects of introduced fish on *A. texanum* juvenile recruitment, I performed a 2 x 2 analysis of variance (ANOVA) on juvenile recruitment for both pond types between the two temporal periods. Measures of recruitment were rank-transformed prior to analyses to conform to the assumptions associated with nonparametric statistics (Zar 1999).

Results

Fish have not been observed in either of the two mitigated ponds since the final application of Rotenone™ to Pond B in January of 2003. A total of 10 species of amphibians were sampled among the four ponds at Warbler Woods Nature Preserve during this study (Table 1). Before fish were removed, the American toad (*Bufo americanus*) was the most abundant species at WWNP, accounting for 90.5% and 66.8% of the captures at mitigated and reference ponds, respectively. During this period,

amphibian species diversity in reference ponds (0.361) was greater than the species diversity in mitigated ponds (0.195; $t_{666} = 5.52$; $P < 0.001$). There were also more species present in reference ponds ($n = 9$) than in mitigated ponds ($n = 8$; Table 1), but this difference was not statistically distinguishable.

Following the removal of fish, relative abundance of American toads decreased by 86.7% and 96.6% in mitigated and reference ponds, respectively. Smallmouth salamanders became the most abundant amphibian, accounting for 40.6% of all amphibians in mitigated ponds and 53.9% in reference ponds (Table 1). Following fish removal, species richness and diversity of both pond types increased compared to their pre-application conditions. Amphibian species diversity in mitigated ponds increased, from 0.195 during the periods when fish were present to 0.738 during the periods after fish were removed ($t_{1498} = 25.56$; $P < 0.001$). Species diversity in reference ponds also increased in the years following fish removal, increasing from 0.361 during the periods when fish were present to 0.597 during the periods after fish were removed ($t_{733} = 7.40$; $P < 0.001$). Although species diversity within both pond types increased following the removal of fish, diversity within mitigated ponds showed a greater increase from pre-mitigation conditions than did reference ponds (Table 1). Additionally, amphibian species diversity was greater in mitigated ponds (0.738) than in reference ponds (0.597) during the temporal period after fish were removed ($t_{970} = 5.92$; $P < 0.001$).

A total of 1302 emerging juvenile *A. texanum* were captured between both pond types during the study period. Within mitigated ponds, 12 juveniles were captured during the time period prior to the removal of fish, whereas 874 juveniles were captured during the years following the removal of fish. Within reference ponds, a total of 29 juveniles

were captured prior to the removal of fish, whereas 400 juveniles were captured after fish removal. Size of juvenile *A. texanum* was influenced by time to metamorphosis, temporal period, and pond type (Table 2). There was a positive association between juvenile size and time to metamorphosis (Pearson's correlation $r = 0.194$; $P = 0.012$; Fig. 2).

Throughout the study period, juvenile *A. texanum* emerging from mitigated ponds were larger than juveniles emerging from reference ponds (Table 2; Fig. 3), with an average size at metamorphosis of 33.6 ± 3.4 mm and 30.5 ± 3.2 mm within mitigated and reference ponds, respectively. Following the removal of fish, *A. texanum* size at metamorphosis decreased within both pond types (Fig. 3). In the years following the removal of fish, juvenile size declined by 19.3 % and 18.7 % within mitigated and reference ponds, respectively. There was no interaction between pond type and temporal period on *A. texanum* size at metamorphosis (Table 2).

Juvenile recruitment of *A. texanum* was also influenced by temporal period (Table 3). There was no difference in juvenile recruitment between pond types prior to the removal of fish (Fig. 4). After the removal of fish, however, juvenile recruitment increased 73-fold and 6-fold within mitigated and reference ponds, respectively (Fig. 4). Despite this observation, however, the only statistically distinguishable increase in juvenile recruitment occurred within mitigated ponds, indicated by a significant temporal period by pond type interaction (Table 3; Fig. 4).

Discussion

This study demonstrated that the presence of introduced fish in the experimental ponds at WWNP had adverse effects on amphibians at the community and population levels. Additionally, this study demonstrated that amphibians may be resilient following the removal of introduced fish, as indicated by improved species diversity of the entire amphibian community and increased *A. texanum* juvenile recruitment. The temporal shifts in amphibian species diversity varied between the experimental ponds at WWNP. Within-pond type changes in species diversity were likely due to changes in species relative abundance rather than changes in species richness (Table 1). Although species diversity improved within both pond types, the increase within mitigated ponds was greater than that within reference ponds. The increase in species diversity within mitigated ponds coincided with the removal of fish via the application of Rotenone™.

Many studies have documented the species-level response to the removal of introduced fish (Knapp et al. 2001; Hoffman et al. 2004; Vredenburg 2004). My results, however, are the first to describe the resilience of an amphibian community following such mitigation. The interaction between predation and interspecific competition is an important factor known to regulate many amphibian communities (Wilbur 1980, 1987) and is the most plausible explanation for the temporal shifts in amphibian relative abundance observed at WWNP. Metamorphosing juveniles accounted for 76.8% of all *B. americanus* captured during this study, suggesting that the shifts in toad relative abundance might have occurred during the larval stage. Previous studies have demonstrated that *B. americanus* tadpoles are unpalatable to fish, whereas tadpoles of competing species might be more susceptible to depredation by fish (Kruse and Stone

1984; Smith et al. 1999). Therefore, the presence of fish might have had positive indirect effects on *B. americanus* abundance by depredating and reducing the abundance of competing amphibians. Because *B. americanus* tadpoles are inferior competitors compared to other species (Wilbur 1987), the observed decline in toad relative abundance following the removal of fish was likely due to increased competition with other amphibians.

The trends that I observed in species' relative abundance at WWNP are possibly confounded for many species, as shifts in relative abundance were similar between mitigated ponds and reference ponds (Table 1). This correlation in amphibian community composition between pond types might be attributable to interpond dispersal by adults during or following reproduction, and juvenile emergence from each pond upon metamorphosis. Interpond distances among the 4 experimental ponds at WWNP are within the dispersal distances of juvenile and adult pond-breeding amphibians, which range from 100 m to over 1000 m (Berven and Grudzien 1990; Semlitsch and Bodie 1998). Due to the close proximity of the experimental ponds at WWNP, therefore, adult amphibians may have visited more than one pond during the breeding season. Although recaptures were omitted from analyses in this study, some adult amphibians might not have reproduced in the first pond where they were captured, traveling to a nearby pond and reproducing as a recaptured individual. Females of some amphibian species are known to deposit egg masses in multiple wetlands during one breeding season (Ritke et al. 1991). Therefore, reproduction by adult amphibians in other experimental ponds following their initial capture may also have contributed to the dynamics of the amphibian community observed at WWNP.

There were noticeable temporal shifts in relative abundance of some species, primarily occurring within mitigated ponds. For instance, the changes in relative abundance of hylid frogs (*Pseudacris crucifer* and *Hyla versicolor* x *chrysoscelis*) were different between pond types during the study period. Whereas both species were observed within reference ponds, they were either absent (*H. versicolor* x *chrysoscelis*) or rare (*P. crucifer*) in mitigated ponds during the years when fish were present. After the removal of fish, however, relative abundance of hylids improved in both pond types, but the degree of change was greater within mitigated ponds (Table 1). Previous studies have demonstrated the susceptibility of hylids to fish predation (Kruse and Stone 1984; Adams 2000). Therefore, the presence of fish within mitigated ponds might have reduced hylid relative abundance via predation upon larvae, and the increase in hylid relative abundance following the removal of fish indicates that these species might be particularly resilient following the removal of introduced fish.

My results indicate that the presence of fish did not influence *A. texanum* larval growth. Jackson and Semlitsch (1993) found similar results in a study of the effects of sunfish (*Lepomis* spp.) on the larval growth of mole salamanders (*A. talpoideum*). My observations, however, are in contrast to other experiments that have documented the negative effects of predation risk on amphibian larval growth. In manipulative field experiments, Figiel and Semlitsch (1990) reported that the presence of sunfish (*Lepomis* spp.) altered the larval growth of spotted salamanders (*A. maculatum*). Additionally, laboratory experiments have demonstrated the adverse effects of predation risk on growth of anuran tadpoles (Relyea 2002, 2004).

One plausible explanation for this observation is in the development of behavioral antipredator adaptations. Larvae of *Ambystoma* species that tolerate populations of introduced fish have developed behavioral adaptations that aid in survival (Figiel and Semlitsch 1990; Hoffman et al. 2004). For many larval amphibians, the expression of these behaviors compromise fitness traits such as age and size at metamorphosis (Sih et al. 1992; Kiesecker and Blaustein 1997; Relyea 2002). Because fish had no effect on *A. texanum* larval growth, and because fewer juveniles successfully metamorphosed in the presence of fish, it is likely that *A. texanum* did not exhibit any antipredatory behaviors that would promote coexistence with fish. The lack of these behaviors would make *A. texanum* larvae more susceptible to fish predation, although the growth of surviving larvae might not be compromised. I was unable to determine whether or not larval *A. texanum* at WWNP possess antipredator adaptations. I did not quantify *A. texanum* behavioral responses to fish; however, previous studies have found that *A. texanum* is capable of exhibiting behavioral plasticity in response to fish predators (Kats 1988).

During the years when fish were present, *A. texanum* size at metamorphosis was greater within mitigated ponds than within reference ponds. Furthermore, when fish were present within mitigated ponds, the size distribution of emigrating *A. texanum* juveniles was less variable within mitigated ponds than within reference ponds (Fig. 3). This observation may be explained by fish morphology. As gape-limited predators (Golub and Brown 2003), the fish introduced at WWNP might not have been capable of consuming *A. texanum* larvae that had surpassed a threshold size. Thus, the presence of fish might be responsible for a greater, less-variable size at metamorphosis among juvenile *A. texanum* through predation upon smaller *A. texanum* larvae.

Although fish presence did not affect *A. texanum* larval growth, fish did impact juvenile recruitment, a measure of reproductive success. The presence of fish nearly eliminated *A. texanum* juvenile recruitment within mitigated ponds (Fig. 4). Predation is the most plausible explanation for this observation, as larval ambystomatids have been shown to be palatable to fish (Figiel and Semlitsch 1990; Sih et al. 1992; Tyler et al. 1998). Despite this, other mechanisms such as disease transfer might be responsible. I am unaware, however, of any reports documenting introduced fish transmitting infectious agents to native amphibians. Furthermore, successfully metamorphosing juvenile *A. texanum* did not appear diseased during the years when fish were present at WWNP, suggesting that the transmission of pathogens was unlikely to suppress *A. texanum* reproductive success.

Juvenile recruitment into the adult population is an important factor determining population dynamics of many vertebrate taxa including fish (Lewis 1997), amphibians (Semlitsch 1983; Berven 1990; Semlitsch et al. 1996), and mammals (Gaillard et al. 1998). Field and laboratory experiments have demonstrated the adverse effects of introduced fish on the reproductive success of larval amphibians (Sih et al. 1992; Tyler et al. 1998; Goodsell and Kats 1999; Smith et al. 1999), reducing the number of juveniles successfully metamorphosing into the adult population. The fish species introduced to the ponds at WWNP were capable of consuming amphibian egg masses and larvae (Orchard 1992; Adams 2000). Therefore, it is likely that the introduced fish present within mitigated ponds at WWNP reduced *A. texanum* larval abundance and suppressed juvenile recruitment via predation.

Following the removal of fish, *A. texanum* juvenile recruitment improved within mitigated ponds at WWNP. My results support those of previous studies that have documented rapid (i.e., < 5 years) improvement in amphibian reproductive success following the removal of introduced fish (Hoffman et al. 2004; Vredenburg 2004). For instance, Hoffmann et al. (2004) reported that larval abundance of the northwest salamander (*A. gracile*) increased following the removal of introduced brook trout (*Salvelinus fontinalis*) in the western United States. Rapid improvement in amphibian reproductive success, therefore, indicates that these amphibian species might be particularly resilient following the mitigation of introduced fish.

My results also indicate that the application of Rotenone™ might be an effective method to remove fish for future amphibian conservation efforts. Previous studies have cautioned against the use of Rotenone™ due to the adverse effects on amphibians by interfering with oxygen uptake across gill membranes (i.e., Fontenot et al. 1994). When Rotenone™ is applied at the appropriate season, however, amphibian exposure to the poison can be minimized. Rotenone™ was applied to the mitigated ponds at WWNP during winter months when most amphibian activity had ceased. Additionally, the successful metamorphosis of overwintered bullfrog (*Rana catesbeiana*) tadpoles following the application of Rotenone™ in mitigated ponds suggests that at least some amphibian species might be more resistant to the poison than fish (Mullin et al. 2004). The future use of Rotenone™ to improve amphibian-breeding habitat in other aquatic systems relies on a thorough understanding of amphibian biology and life history. Care should be taken to minimize the impact of Rotenone™ on as many non-target organisms as possible, especially rare or endangered species. For this reason, Rotenone™ might not

be an appropriate option in wetlands where many fully aquatic (e.g., paedeomorphic) amphibian species exist.

Although my observations are most likely attributable to the removal of introduced fish, these results may be due to a number of abiotic phenomena. Amphibian communities are often regulated by factors such as precipitation, temperature, and pond hydroperiod (Semlitsch 1985b; Phillips et al. 2002; Jensen et al. 2003). Analyses of daily precipitation and temperature data obtained from Coles County Regional Airport, approximately 12 km from WWNP, revealed no differences in mean daily precipitation or temperature patterns between temporal periods (MANCOVA; $F_{2,1450} = 0.600$; $P = 0.549$). Thus, differences in amphibian relative abundances and *A. texanum* larval dynamics are unlikely due to these environmental changes. All ponds at WWNP possessed a permanent hydroperiod except for Pond D, which dried before 2 August of each year of this study. Variation in pond hydroperiod among ponds at WWNP may have contributed to these observations, however, as amphibian larval growth and survivorship are often influenced by hydroperiod length (Pechmann et al. 1989; Phillips et al. 2002).

Improvements in species diversity of the amphibian community and *A. texanum* larval dynamics at WWNP coincided with, and are apparently attributable to, the removal of introduced fish. Amphibian population sizes and reproductive output fluctuate naturally among years (Pechmann and Wilbur 1994) and could also explain my results. Therefore, long-term amphibian monitoring datasets are vital in field studies to accurately determine the effects of anthropogenic perturbations on amphibian populations.

Literature Cited

- Adams, M.J. 2000. Pond permanence and the effects of exotic vertebrates on anurans. *Ecological Applications* 10: 559-568.
- Alford, R.A., and S.J. Richards. 1999. Global amphibian declines: a problem in applied ecology. *Annual Review of Ecology and Systematics* 30: 133-165.
- Altwegg, R., and H.U. Reyer. 2003. Patterns of natural selection on size at metamorphosis in water frogs. *Evolution* 57: 872-882.
- Bahls, P.F. 1992. The status of fish populations and management of high mountain lakes in the western United States. *Northwest Science* 66: 183-193.
- Berven, K.A. 1990. Factors affecting population fluctuations in larval and adult stages of the wood frog (*Rana sylvatica*). *Ecology* 71: 1599-1608.
- Berven, K.A., and T.A. Grudzien. 1990. Dispersal in the wood frog (*Rana sylvatica*): implications for genetic population structure. *Evolution* 44: 2047-2056.
- Blaustein, A.R. 1994. Chicken Little or Nero's fiddle? A perspective on declining amphibian populations. *Herpetologica* 50: 85-97.
- Blaustein, A.R., D.B. Wake, and W.P. Sousa. 1994. Amphibian declines: judging stability, persistence, and susceptibility of populations to local and global extinctions. *Conservation Biology* 8: 60-71.
- Blaustein, L., J. Friedman, and T. Fahima. 1996. Larval salamandra drive temporary pool community dynamics: evidence from an artificial pool experiment. *Oikos* 76: 392-402.

- Bradford, D.F. 1989. Allotopic distribution of native frogs and introduced fishes in high Sierra Nevada lakes of California: implication of the negative effect of fish introductions. *Copeia* 1989: 775-778.
- Brophy, T.E. 1980. Food habits of sympatric larval *Ambystoma tigrinum* and *Notophthalmus viridescens*. *Journal of Herpetology* 14: 1-6.
- Burton, T.M., and G.E. Likens. 1975. Salamander populations and biomass in the Hubbard Brook Experimental Forest, New Hampshire. *Copeia* 1975: 541-546.
- Collins, J.P., and A. Storfer. 2003. Global amphibian declines: sorting the hypotheses. *Diversity and Distributions* 9: 89-98.
- Corn, P.S. 1994. Straight-line drift fences and pitfall traps. *In* W.A. Heyer, M.A. Donnelly, R.W. McDiarmid, L.C. Hayek, and M.S. Foster, (eds.), *Measuring and Monitoring Biological Diversity: Standard Methods for Amphibians*, pp. 109-118. Smithsonian Institution Press, Washington, D.C.
- Dickman, M. 1968. The effect of grazing by tadpoles on the structure of a periphyton community. *Ecology* 49: 1188-1190.
- Donnelly, M.A., C. Guyer, E. Juterbock, and R.A. Alford. 1994. Appendix 2: Techniques for marking amphibians. *In* W.A. Heyer, M.A. Donnelly, R.W. McDiarmid, L.C. Hayek, and M.S. Foster, (eds.), *Measuring and Monitoring Biological Diversity: Standard Methods for Amphibians*, pp. 277-284. Smithsonian Institution Press, Washington, D.C.
- Figiel, C.R., Jr., and R.D. Semlitsch. 1990. Population variation in survival and metamorphosis of larval salamanders (*Ambystoma maculatum*) in the presence and absence of fish predation. *Copeia* 1990: 818-826.

- Fontenot, L.W., G.P. Noblet, and S.G. Platt. 1994. Rotenone hazards to amphibians and reptiles. *Herpetological Review* 25: 150-156.
- Gaillard, J.M., M. Festa-Bianchet, and N.G. Yoccoz. 1998. Population dynamics of large herbivores: variable recruitment with constant adult survival. *Trends in Ecology and Evolution* 13: 58-63.
- Gamradt, S.C., and L.B. Kats. 1996. Effect of introduced crayfish and mosquitofish on California newts. *Conservation Biology* 10: 1155-1162.
- Gilderhus, P.A., V.K. Dawson, and J.L. Allen. 1988. Deposition and persistence of rotenone in shallow ponds during cold and warm seasons. *Investigations in Fish Control*: 95. U.S. Fish and Wildlife Service, Washington DC. 7 pp.
- Golub, J.L., and G.E. Brown. 2003. Are all signals the same? Ontogenetic change in the response to conspecific and heterospecific chemical alarm signals by juvenile green sunfish (*Lepomis cyanellus*). *Behavioral Ecology and Sociobiology* 54: 113-118.
- Goodsell, J.A., and L.B. Kats. 1999. Effects of the introduced mosquitofish on pacific treefrogs and the role of alternative prey. *Conservation Biology* 13: 921-924.
- Hecnar, S.J., and R.T. M'Closkey. 1996. Regional dynamics and the status of amphibians. *Ecology* 77: 2091-2097.
- Hecnar, S.J., and R.T. M'Closkey. 1997. The effects of predatory fish on amphibian species richness and distribution. *Biological Conservation* 79: 123-131.
- Hoffman, R.L., G.L. Larson, and B. Samora. 2004. Responses of *Ambystoma gracile* to the removal of introduced nonnative fish from a mountain lake. *Journal of Herpetology* 38: 578-585.

- Holomuzki, J.R., J.P. Collins, and P.E. Brunkow. 1994. Trophic control of fishless ponds by tiger salamander larvae. *Oikos* 71: 55-64.
- Hopey, M.E., and J.W. Petranka. 1994. Restriction of wood frogs to fish-free habitats: how important is adult choice? *Copeia* 1994: 1023-1025.
- Jackson, M.E., and R.D. Semlitsch. 1993. Paedomorphosis in the salamander *Ambystoma talpoideum*: effects of a fish predator. *Ecology* 74: 342-350.
- Jensen, J.B., M.A. Bailey, E.L. Blankenship, and C.D. Camp. 2003. The relationship between breeding by the gopher frog, *Rana capito* (Amphibia: Ranidae) and rainfall. *American Midland Naturalist* 150: 185-190.
- Kats, L.B. 1988. The detection of certain predators via olfaction by small-mouthed salamander larvae (*Ambystoma texanum*). *Behavioral and Neural Biology* 50: 126-131.
- Kats, L.B., J.W. Petranka, and A. Sih. 1988. Antipredator defenses and the persistence of amphibian larvae with fishes. *Ecology* 69: 1865-1870.
- Kats, L.B., and A. Sih. 1992. Oviposition site selection and avoidance of fish by streamside salamanders (*Ambystoma barbouri*). *Copeia* 1992: 468-473.
- Kiesecker, J.M., and A.R. Blaustein. 1997. Population differences in responses of red-legged frogs (*Rana aurora*) to introduced bullfrogs. *Ecology* 78: 1752-1760.
- Kiesecker, J.M., and A.R. Blaustein. 1998. Effects of introduced bullfrogs and smallmouth bass on microhabitat use, growth, and survival of native red-legged frogs (*Rana aurora*). *Conservation Biology* 12: 776-787.

- Knapp, R.A., and K.R. Matthews. 2000. Nonnative fish introductions and the decline of the mountain yellow-legged frog from within protected areas. *Conservation Biology* 14: 428-438.
- Knapp, R.A., K.R. Matthews, and O. Sarnelle. 2001. Resistance and resilience of alpine lake fauna to fish introductions. *Ecological Monographs* 71: 401-421.
- Kolozsvary, M.B., and R.K. Swihart. 1999. Habitat fragmentation and the distribution of amphibians: patch and landscape correlates in farmland. *Canadian Journal of Zoology* 77: 1288-1299.
- Kruse, K.C., and M.G. Francis. 1977. A predation deterrent in larvae of the bullfrog, *Rana catesbeiana*. *Transactions of the American Fisheries Society* 106: 248-252.
- Kruse, K.C., and B.M. Stone. 1984. Largemouth bass (*Micropterus salmoides*) learn to avoid feeding on toad (*Bufo*) tadpoles. *Animal Behaviour* 32: 1035-1039.
- Kupferberg, S. 1997. Bullfrog (*Rana catesbeiana*) invasion of a California river: the role of larval competition. *Ecology* 78: 1736-1751.
- Kupferberg, S.J., J.C. Marks, and M.E. Power. 1994. Effects of variation in natural algal detrital diets on larval anuran (*Hyla regilla*) life-history traits. *Copeia* 1994: 446-457.
- Lewis, A.R. 1997. Recruitment and post-recruit immigration affect the local population size of coral fishes. *Coral Reefs* 16: 139-149.
- Lips, K.R. 1998. Decline of a tropical montane amphibian fauna. *Conservation Biology* 12: 106-117.

- Lodge, D.M., C.A. Taylor, D.M. Holdrich, and J. Skurdal. 2000. Nonindigenous crayfishes threaten North American freshwater biodiversity: lessons from Europe. *Fisheries* 25: 7-19.
- McClay, W. 2000. Rotenone use in North America (1988-1997). *Fisheries* 25: 15-21.
- Means, D.B., J.G. Palis, and M. Baggett. 1996. Effects of slash pine silviculture on a Florida population of flatwoods salamander. *Conservation Biology* 10: 426-437.
- Mullin, S.J., J.B. Towey, and R.E. Szafoni. 2004. Using Rotenone™ to enhance native amphibian breeding habitat in ponds (Illinois). *Ecological Restoration* 22: 305-306.
- Muths, E., P.S. Corn, A.P. Pessier, and D.E. Green. 2003. Evidence for disease-related amphibian decline in Colorado. *Biological Conservation* 110: 357-365.
- Pechmann, J.H.K., D.E. Scott, J.W. Gibbons, and R.D. Semlitsch. 1989. Influence of wetland hydroperiod on diversity and abundance of metamorphosing juvenile amphibians. *Wetlands Ecology and Management* 1: 3-11.
- Pechmann, J.H.K., and H.M. Wilbur. 1994. Putting declining amphibian populations in perspective: natural fluctuations and human impacts. *Herpetologica* 50: 65-84.
- Petranka, J.W., L.B. Kats, and A. Sih. 1987. Predator-prey interactions among fish and larval amphibians: use of chemical cues to detect predatory fish. *Animal Behaviour* 35: 420-425.
- Petranka, J.W. 1998. *Salamanders of the United States and Canada*. Washington and London: Smithsonian Institution Press. 587 p.
- Phillips, C.A., R.A. Brandon, and E.O. Moll. 1999. Field guide to amphibians and reptiles of Illinois. *Illinois Natural History Survey Manual* 8. 300 pp.

- Phillips, C.A., J.R. Johnson, M.J. Dreslik, and J.E. Petzing. 2002. Effects of hydroperiod on recruitment of mole salamanders (Genus *Ambystoma*) at a temporary pond in Vermillion County, Illinois. Transactions of the Illinois State Academy of Science 95: 131-139.
- Relyea, R.A. 2002. Costs of phenotypic plasticity. The American Naturalist 159: 242-282.
- Relyea, R.A. 2004. Fine-tuned phenotypes: tadpole plasticity under 16 combinations of predators and competitors. Ecology 85: 172-179.
- Ritkey, M.E., J.G. Babb, and M.K. Ritke. 1991. Breeding-site specificity in the gray treefrog (*Hyla crysoscelis*). Journal of Herpetology 25: 123-125.
- Scott, D.E. 1990. Effects of larval density in *Ambystoma opacum*: an experiment in large-scale field enclosures. Ecology 7: 296-306.
- Semlitsch, R.D. 1983. Structure and dynamics of two breeding populations of the eastern tiger salamander, *Ambystoma tigrinum*. Copeia 1983: 608-616.
- Semlitsch, R.D. 1985a. Reproductive strategy of a facultatively paedomorphic salamander, *Ambystoma talpoideum*. Oecologia 65: 305-313.
- Semlitsch, R.D. 1985b. Analysis of climatic factors influencing migrations of the salamander *Ambystoma talpoideum*. Copeia 1985: 477-489.
- Semlitsch, R.D. 1988. Allotopic distribution of two salamanders: effects of fish predation and competitive interactions. Copeia 1988: 290-298.
- Semlitsch, R.D., D.E. Scott, and J.H.K. Pechmann. 1988. Time and size at metamorphosis related to adult fitness in *Ambystoma talpoideum*. Copeia 1988: 978-983.

- Semlitsch, R.D., D.E. Scott, J.H.K. Pechmann, and J.W. Gibbons. 1996. Structure and dynamics of an amphibian community: evidence from a 16-year study of a natural pond, p. 217-247. *In*: M.L. Cody and J.A. Smallwood (eds.). Long-term studies of vertebrate communities. Academic Press Inc., San Diego, CA.
- Semlitsch, R.D. and J.R. Bodie. 1998. Are small, isolated wetlands expendable? *Conservation Biology* 12: 1129-1133.
- Sih, A., L.B. Kats, and R.D. Moore. 1992. Effects of predatory sunfish on the density, drift, and refuge use of stream salamander larvae. *Ecology* 73: 1418-1430.
- Sjogren, P. 1991. Extinction and isolation gradients in metapopulations: the case of the pool frog (*Rana lessonae*). *Biological Journal of the Linnean Society* 42: 135-147.
- Skelly, D.K., E.E. Werner, and S.A. Cortwright. 1999. Long-term distributional dynamics of a Michigan amphibian assemblage. *Ecology* 80: 2326-2337.
- Smith, G.R., J.E. Rettig, G.G. Mittelbach, J.L. Valiulis, and S.R. Schaack. 1999. The effects of fish on assemblages of amphibians in ponds: a field experiment. *Freshwater Biology* 41: 829-837.
- Sparling, D.S., G.M. Fellers, and L.L. McConnell. 2001. Pesticides and amphibian population declines in California, USA. *Environmental Toxicology and Chemistry* 20: 1591-1595.
- Tyler, T.J., W.J. Liss, R.L. Hoffman, and L.M. Ganio. 1998. Experimental analysis of trout effects on survival, growth, and habitat use of two species of Ambystomatid salamanders. *Journal of Herpetology* 32: 345-349.

- U.S. Congress. 1993. Harmful non-indigenous species in the United States. Office of Technology Assessment, OTA-F-565, U.S. Government Printing Office, Washington DC.
- Vredenburg, V.T. 2004. Reversing introduced species effects: experimental removal of introduced fish leads to rapid recovery of a declining frog. *Proceedings of the National Academy of Sciences* 101: 7646-7650.
- Werner, E.E. 1986. Amphibian metamorphosis: growth rate, predation risk, and the optimal size at transformation. *American Naturalist* 128: 319-341.
- Wilbur, H.M. 1980. Complex life cycles. *Annual Review of Ecology and Systematics* 11: 67-93.
- Wilbur, H.M. 1987. Regulation of structure in complex systems: experimental temporary pond communities. *Ecology* 68: 1437-1452.
- Zar, J.H. 1999. *Biostatistical analysis*. 4th ed. Prentice-Hall, Englewood Cliffs, New Jersey, USA.

TABLE 1. Relative abundance (proportion) of amphibians captured at mitigated (n = 2) and reference (n = 2) ponds at Warbler Woods Nature Preserve, Coles County, Illinois, between the temporal periods of pre- and post-Rotenone™ application. Shannon-Weiner Diversity Index values (H') are displayed for both pond types during each temporal period.

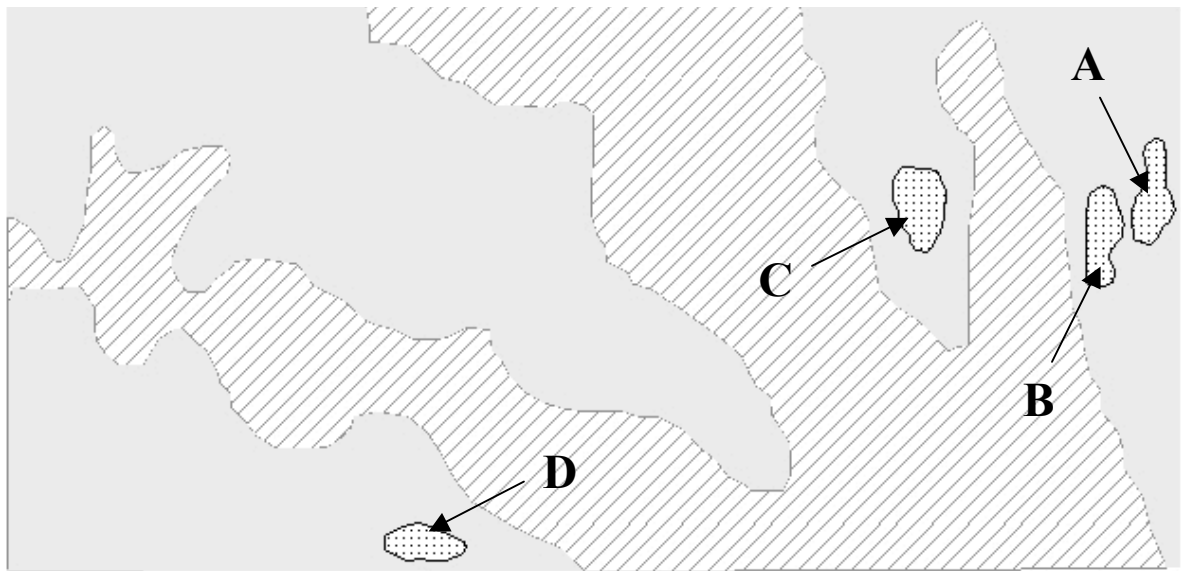
Species	<u>Pre-Application</u>		<u>Post-Application</u>	
	Mitigated Ponds	Reference Ponds	Mitigated Ponds	Reference Ponds
<i>Ambystoma texanum</i>	0.026	0.168	0.406	0.539
<i>Bufo americanus</i>	0.905	0.668	0.120	0.023
<i>Bufo fowleri</i>	< 0.001	< 0.001	–	0.002
<i>Acris crepitans</i>	0.002	< 0.001	0.003	< 0.001
<i>Hyla versicolor</i> x <i>chrysoscelis</i>	–	0.004	0.088	0.011
<i>Pseudacris crucifer</i>	0.003	0.012	0.141	0.047
<i>Pseudacris triseriata</i>	–	–	0.002	< 0.001
<i>Rana catesbeiana</i>	0.048	0.041	0.068	0.074
<i>Rana sylvatica</i>	0.010	0.077	0.022	0.162
<i>Rana utricularia</i>	0.007	0.029	0.149	0.141
(H')	0.195	0.361	0.738	0.597

TABLE 2. Results of an analysis of covariance (ANCOVA) that tested for the effects of temporal period (pre-application or post-application) and pond type (mitigated or reference) on growth (size at metamorphosis; SVL) of smallmouth salamanders (*Ambystoma texanum*) at WWNP between 2001 and 2004. Time to metamorphosis (Julian date of metamorphosis) was used as a covariate (df = 1,1297 in all cases).

Source	F	P
Temporal Period	131.91	< 0.0001
Pond Type	54.85	< 0.0001
Temporal Period x Pond Type	1.57	0.210
Time to Metamorphosis	51.85	< 0.0001

TABLE 3. Results of the analysis of variance (ANOVA) that tested for the effects of temporal period (pre-application or post-application) and pond type (mitigated or reference) on the juvenile recruitment of smallmouth salamanders (*Ambystoma texanum*) at WWNP between 2001 and 2004 (df = 1,12 in all cases).

Source	F	P
Pond Type	2.21	0.163
Temporal Period	13.52	0.003
Pond Type x Temporal Period	5.73	0.034



Habitat Types

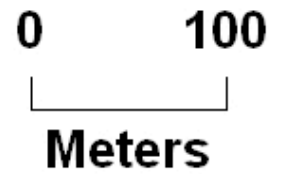
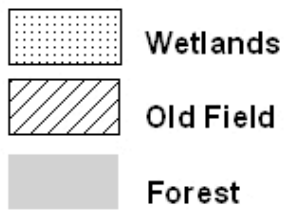


FIGURE 1. A portion of Warbler Woods Nature Preserve (WWNP), Coles County, Illinois, with the location of the four experimental ponds. Fish were introduced into mitigated ponds (Ponds B and C) prior to the commencement of this study, whereas reference ponds (Ponds A and D) never contained fish.

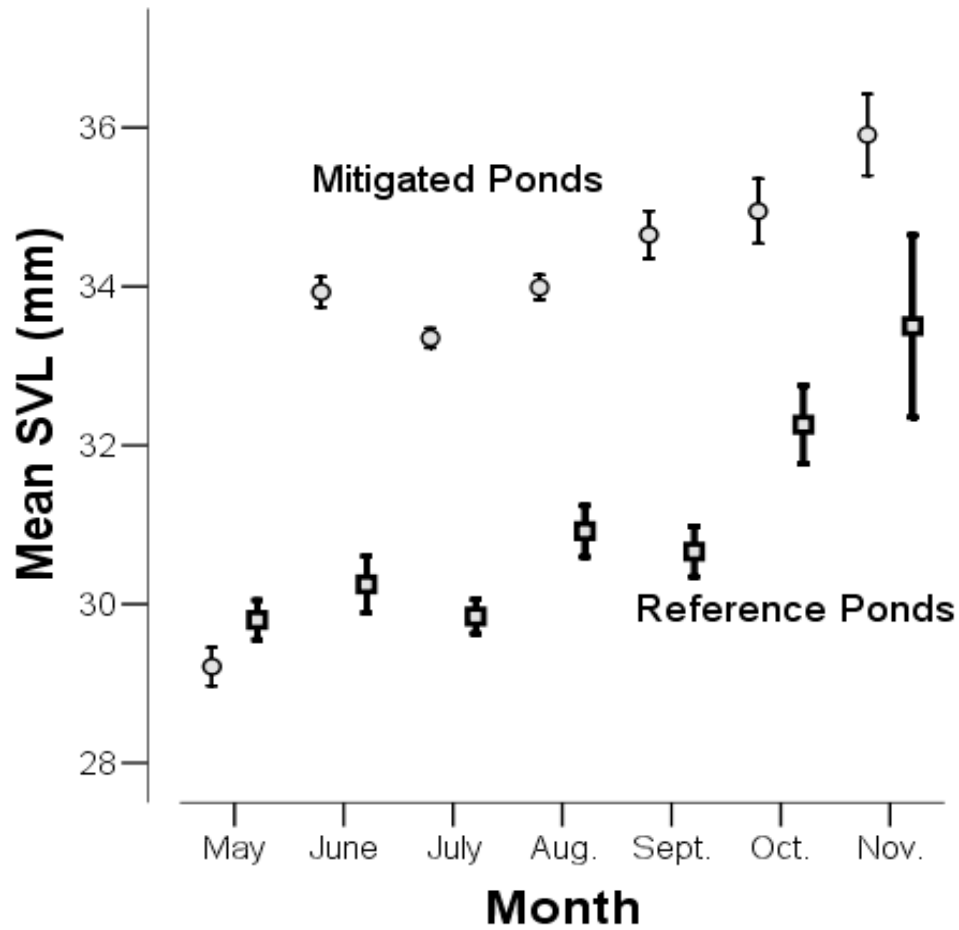


FIGURE 2. Size (mm) of metamorphosing juvenile smallmouth salamanders (*Ambystoma texanum*) as a function of time (month of emergence). Juveniles emigrating from mitigated ponds were larger than those from reference ponds throughout the study period (2001-2004), and size was positively associated with length of the larval period (Julian date of metamorphosis). All individuals metamorphosed between May and November for every year of the study period. Data are presented as means \pm 1 SE.

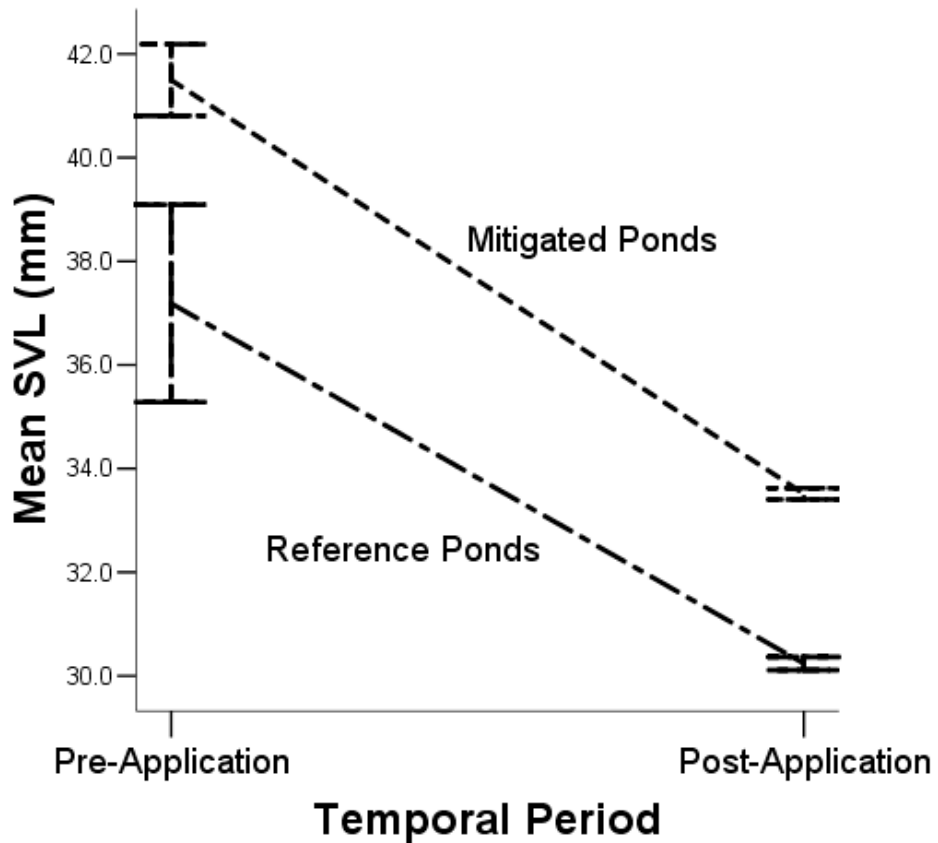


FIGURE 3. Changes in larval smallmouth salamander (*Ambystoma texanum*) growth (size at metamorphosis) between temporal periods (pre-application and post-application) and pond types (mitigated or reference) at Warbler Woods Nature Preserve, Coles County, Illinois. Juveniles emigrating from mitigated ponds were larger than those from reference ponds throughout the study period (2001-2004). All data are presented as means \pm 1 SE.

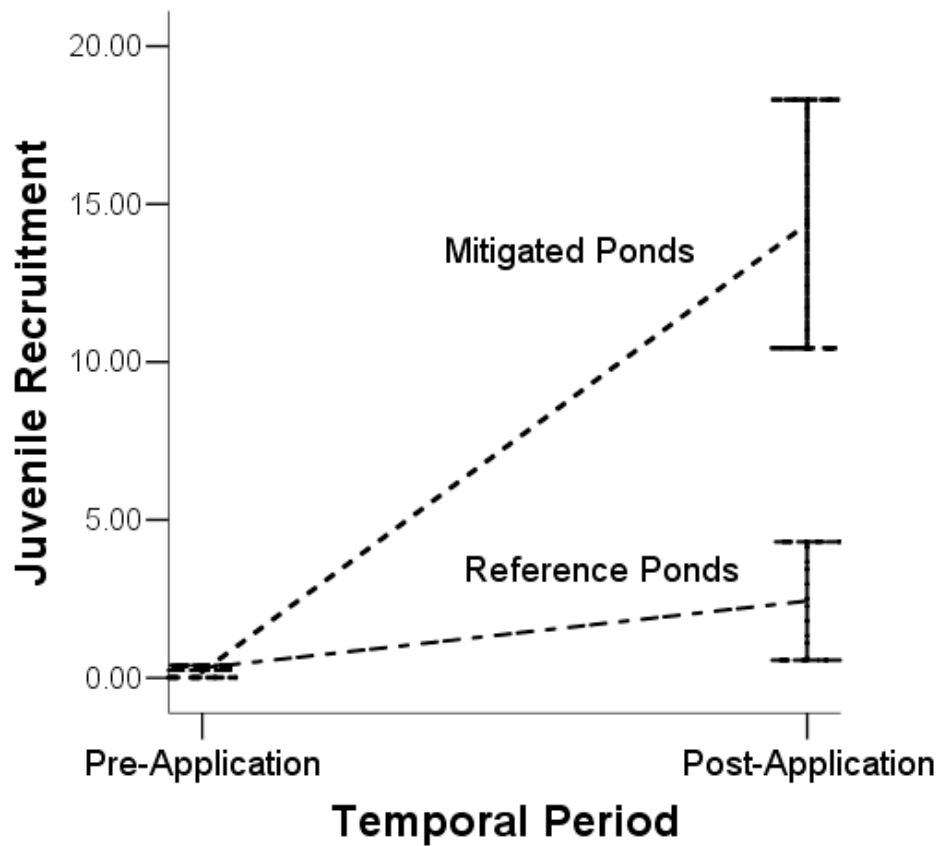


FIGURE 4. Changes in smallmouth salamander (*Ambystoma texanum*) juvenile recruitment (proportion of metamorphosing juveniles to number of breeding females) in mitigated and reference ponds between temporal periods (pre- and post-application of Rotenone™) at WWNP. All data are presented as means ± 1 SE.

APPENDIX I

Population Responses of Wood Frogs (*Rana sylvatica*) to Overwintered Bullfrog (*Rana catesbeiana*) Tadpoles

Abstract

I examined the population responses of sympatric wood frogs to native overwintered bullfrog tadpoles in an effort to elucidate the mechanisms underlying the effects of bullfrog establishment within its native range. The presence of an overwintered bullfrog tadpole had a negative effect on the growth of wood frog tadpoles allotopic (naïve) to bullfrogs, whereas the presence of bullfrogs had no effect on growth of syntopic (experienced) wood frog tadpoles. There were also differential population responses of wood frogs to overwintered bullfrog tadpole visual and chemical cues, as allotopic wood frog tadpoles decreased activity levels and increased use of refugia in the presence of overwintered bullfrog tadpoles. These observations indicate that overwintered bullfrog tadpoles might exert a selective pressure on other amphibians, and that bullfrog establishment within its native range might have negative consequences on larval dynamics of other amphibian species.

Introduction

Biological invasions pose one of the greatest threats to global biodiversity (Soule 1990; Wilcove et al. 1998; Mack et al. 2000). Invasive species possess a number of attributes enabling them to become established in novel environments, and identifying these characteristics is critical in mediating their impacts on native species (Parker et al. 1999). Bullfrogs (*Rana catesbeiana*), native east of the Great Plains, are the largest

anuran native to North America. They are dietary generalists that consume a variety of invertebrate and vertebrate taxa including other amphibians (Corse and Metter 1980; Carpenter et al. 2002). Bullfrogs have been introduced throughout much of western North America and have also become established in other continents as well (Nussbaum et al. 1983; Stumpel 1992; Batista 2002). As a consequence, bullfrog invasion is implicated in the declines of native amphibian populations, primarily through interspecific competition and predation (Kiesecker and Blaustein 1997; Kupferberg 1997; Lawler et al. 1999).

Throughout their native range, bullfrogs are competitively superior to other amphibian species (Werner and Anholt 1996), and are considered important agents of amphibian community structure (Hecnar and M'Closkey 1997; Boone et al. 2004). The effects of bullfrogs on other amphibians, however, are often mediated by pond hydroperiod. In Illinois, bullfrogs typically require two years for tadpoles to complete metamorphosis (Phillips et al. 1999), and are thus restricted to permanent wetlands where fish may also occur. Native fish species are predators of many amphibians (Semlitsch 1988; Werner and McPeck 1994), capable of eliminating many amphibian populations from permanent ponds. Bullfrog tadpoles, however, are unpalatable to many fish (Kruse and Francis 1977; Werner and McPeck 1994; Smith et al. 1999), and are capable of persisting in these environments.

Although ephemeral ponds do not typically support bullfrog tadpole populations, long hydroperiod events may allow bullfrog tadpoles to overwinter in ponds that fail to dry between years, creating a situation in which bullfrog tadpoles coexist with other sympatric amphibians (Boone et al. 2004). Thus, the establishment of new bullfrog

populations within its native range is a potentially realistic scenario. In these instances, overwintering bullfrog larvae may adversely impact other native amphibian species. Additionally, permanent woodland ponds lacking fish predators provide an opportunity for other amphibian tadpoles to exist with bullfrog tadpoles. Boone et al. (2004) found that the presence of overwintered bullfrog tadpoles negatively effects the growth and survival of sympatric amphibian species naïve to bullfrog tadpoles. Similar studies of introduced bullfrog populations outside their native range have suggested some potential mechanisms underlying these effects on native amphibians. Kiesecker and Blaustein (1997) demonstrated that amphibians might respond to exotic bullfrog invasion by exhibiting predator-induced behavioral plasticity by reducing foraging activity and increasing use of microhabitat refugia.

Wood frog (*Rana sylvatica*) tadpoles commonly exhibit behavioral and morphological plasticity in response to predators and competitors (Petranka and Hayes 1998; Relyea 2002, 2004). Wood frogs and bullfrogs are sympatric throughout much of the eastern United States, though their distributions are usually allotopic with respect to pond hydroperiod (Phillips et al. 1999; Paton and Crouch 2002). Overwintered bullfrog tadpoles possess a size advantage over wood frog tadpoles and have been observed to depredate tadpoles of other ranids (Kiesecker and Blaustein 1997; Boone et al. 2004). In situations permitting overwintered bullfrog tadpoles to coexist in the same environments as wood frog tadpoles, therefore, bullfrog tadpoles might exert adverse effects on wood frog populations. Wood frog tadpoles may perceive bullfrog larvae as a predation threat, responding to their presence through predator-induced plastic traits. The predator avoidance hypothesis (*sensu* Lima and Dill 1990) suggests that, in response to predators,

organisms alter their behavior in efforts to avoid detection by predators. For wood frog tadpoles, these responses include reduced activity, increased use of microhabitat refugia, and morphological changes; previous studies have suggested that these responses are adaptive (Petranka and Hayes 1998; Van Buskirk and Relyea 1998; Relyea 2002).

Experience may play an important role in dictating how organisms respond to predators (Kats et al. 1988). For instance, Kiesecker and Blaustein (1997) reported that populations of red-legged frogs (*Rana aurora*) allotopic to introduced bullfrogs do not exhibit antipredator behaviors in the presence of bullfrogs, whereas populations syntopic to introduced bullfrogs responded by reducing activity and altering microhabitat use. In the presence of overwintered bullfrog tadpoles, therefore, wood frog tadpoles from populations allotopic to bullfrogs may respond differently than syntopic wood frog tadpoles. These differential population responses may further affect wood frog larval growth and survival, metrics that often confer adult fitness and population viability (Werner 1986; Berven 1990). Consequently, bullfrogs may exert a selective pressure on sympatric wood frogs, favoring behavioral adaptations that permit coexistence with bullfrogs.

Herein, I present results of laboratory experiments addressing the responses of sympatric wood frog populations to native overwintered bullfrog tadpoles in an effort to elucidate the mechanisms underlying the effects of bullfrog establishment within its native range. I used natural populations of wood frogs and bullfrogs to test two hypotheses: 1) overwintered bullfrog tadpoles induce changes in behavior, growth, and survival of sympatric wood frog tadpoles; and, 2) wood frog tadpoles from populations

syntopic (experienced) to bullfrogs respond differently than allotopic (naïve) populations when in the presence of overwintered bullfrog tadpoles.

Methods

Study Organisms

I collected 8 wood frog egg masses on 7 March 2005 from four ponds in Coles County, Illinois. Four egg masses each were collected from populations syntopic and allotopic to bullfrogs. Syntopic wood frog egg masses were collected from two permanent fishless ponds that also contained a breeding population of bullfrogs. Successful recruitment of bullfrog and wood frog juveniles has been observed at both ponds for each of the previous 5 years (S. Mullin, pers. comm.). Syntopic wood frog tadpoles, therefore, were considered experienced to overwintered bullfrog tadpoles. Allotopic wood frog egg masses were collected from two ephemeral ponds in which bullfrog tadpoles have been excluded (S. Mullin, pers. comm.). Allotopic wood frog tadpoles, therefore, were considered naïve to overwintered bullfrog tadpoles. Nearly 400 m separates the allotopic and syntopic populations. Although this is within the range of known dispersal distances of juvenile wood frogs (Berven and Grudzien 1990), the two sites are separated by a combination of roads, residential property, and old-field upland habitats that impede amphibian dispersal abilities. Therefore, gene flow between the two sites is unlikely and I assumed egg masses collected from the two sites to be from separate populations.

Upon collection, the egg masses were brought back to the laboratory where they were incubated at 20 °C on a 12:12 light:dark (L:D) photoperiod. While incubating, egg

masses were separated by population and kept in 25 L of aged tap water. Tadpoles that hatched prior to the commencement of this study were raised in cohort-specific aquaria filled with 25 L of natal pond water. Water was changed every 5-7 d and tadpoles were fed powdered rodent chow *ad libitum*. On 22 March 2005, I returned to the three permanent ponds where the syntopic wood frogs were originally collected and seined each pond for bullfrog tadpoles. These tadpoles were 7.32 ± 1.84 g (mean \pm 1 SE), too large to have hatched in 2005 (Gosner stage 30 – 35; Gosner 1960), and were considered to have overwintered from the previous year. Bullfrog tadpoles were brought back to the laboratory to be added to experimental enclosures. Those bullfrog tadpoles not immediately added to experimental enclosures were kept in a separate enclosure filled with aged tap water. Bullfrog tadpoles were fed and their water was changed in the same manner as the wood frog tadpoles.

Tadpole Growth and Survival

I used a randomized block design to test for the effects of overwintered bullfrog tadpoles on the growth and survival of wood frog tadpoles. The two independent variables each had two levels – the wood frog population source was allotopic or syntopic, and bullfrog tadpoles were present or absent. I randomly assigned enclosures to the four combinations of these variables, each of which was replicated 5 times; thus, there were a total of 20 experimental enclosures. I placed brown construction paper between each enclosure to prevent the potentially confounding effects of bullfrog visual stimuli on wood frogs in adjacent enclosures (38-L aquaria). All enclosures were maintained on a 12:12 L:D photoperiod in a temperature-controlled laboratory (20 °C).

On 22 March 2005, I filled 38-L aquaria with 25 L of aged tap water. I then added 2 g of leaf litter and 1 g of ground rodent chow to each enclosure to serve as a refuge and food resource, respectively. For the bullfrog treatments, I added a single bullfrog tadpole to an enclosure containing either 20 allotopic or syntopic wood frog tadpoles. In treatments in which bullfrogs were absent, I added 20 wood frog tadpoles from either population to experimental enclosures. I measured the mass (± 1.0 mg) of all tadpoles before adding them. All wood frog tadpoles were of the same developmental stage (Gosner stage 26 – 30) and all bullfrog tadpoles used in these treatments were within the same size distribution (7.27 ± 2.05 g; Kolmogorov-Smirnov test; $P = 0.20$).

On 5 April, I terminated the experiment and measured wood frog tadpole growth and survival. To quantify growth, I measured the mass (± 1.0 mg) of each wood frog tadpole and calculated the mean for each enclosure. I then subtracted the mean initial mass from the mean final mass to determine wood frog growth within each enclosure. Bullfrog tadpole growth was calculated in a similar manner. I quantified survival in each enclosure as the proportion of wood frog tadpoles that were alive to the total number initially stocked ($n = 20$).

Activity and Refuge Use

Organisms often respond to an array of sensory cues to identify predation risk, including chemical, tactile, and visual cues. Previous studies have documented the ability of many tadpoles to recognize and respond to predator chemical cues (Kiesecker and Blaustein 1997; Petranka and Hayes 1998). Due to the large size of overwintered bullfrog tadpoles, wood frogs might recognize and respond to visual and chemical cues

emitted by overwintered bullfrog tadpoles. To prevent the confounding effects of physical interaction between bullfrog and wood frog tadpoles, I tested for chemically- and visually-mediated avoidance of bullfrogs by assessing wood frog larvae activity levels and use of refugia. Additionally, I tested for differential responses between the two wood frog populations. For this experiment, I used a 2x2 factorial design using 38-L enclosures. I partitioned each enclosure into equal lengthwise sections by attaching a 1-cm high aluminum screen to the bottom of the aquarium with silicone. I then placed a 1-cm layer of leaf litter on one side of each aquarium. The leaf litter served as refugia, and the aluminum screening served as a divider between the two microhabitats (with refugia or without). Additionally, the 1-cm high screen partitioning was low enough to allow tadpoles to swim freely between both sides.

To test for the effects of bullfrog chemical and visual cues on wood frog activity and refuge use, I conducted trials once per day, beginning on 24 March 2005. Prior to each trial I added 20 L of aged tap water to all aquaria. Bullfrog enclosures, made of perforated clear 20-cm tall plastic cups, were submerged 12 cm beneath the water surface such that the opening of the cup was above the water surface, preventing bullfrog tadpoles from escaping. I attached one bullfrog enclosure to the wall of the each enclosure perpendicular to the 1-cm high screen partitioning. Individual bullfrog tadpoles placed into these enclosures were visible to wood frog tadpoles and chemical cues could disperse into the water column occupied by the wood frog tadpoles. For the bullfrog treatments, I randomly selected one bullfrog tadpole from a stock population and placed it in the plastic cup. To mimic the same amount of disturbance that the bullfrog enclosures create, empty bullfrog enclosures were attached to all control aquaria. For all

treatments, I added 20 randomly selected wood frog tadpoles from either an allotopic or syntopic stock population. All enclosures were maintained in a temperature-controlled laboratory (20 °C) and maintained at a constant 12:12 L:D photoperiod.

After a 12 h habituation period, I quantified tadpole activity and microhabitat use. I used a method of scan sampling (Altmann 1974) to quantify activity, cautiously approaching each aquarium and counting the number of tadpoles that could be observed actively moving. I divided this number by the number of total tadpoles present ($n = 20$) to provide an estimate of tadpole activity (modified from Relyea 2002). I quantified refuge use in a similar fashion by counting the number of tadpoles present in the side of the enclosure that lacked the leaf-litter refuge. Subtracting the number observed in this side from the number of tadpoles present ($n = 20$) provided the number of tadpoles utilizing the leaf-litter refuge for that observation. I divided this number by the total number of tadpoles present ($n = 20$) to provide an estimate of refuge use. I recorded activity and refuge use three times for each replicate, each measurement separated by at least 1 h, and I calculated the mean as the metric of tadpole activity and refuge use for each replicate. After the final measurement was recorded for each replicate, all enclosures were drained and I measured the mass (± 1.0 mg) of each tadpole. Tadpoles were placed in separate containers and were never used in more than one treatment. I terminated all experiments on 6 April, after 5 replicates had been completed for each treatment.

Statistical Analyses

I tested for the effects of population source, bullfrog presence, and their interaction on the growth and survival of wood frogs using multivariate analyses of variance (MANOVA), followed by univariate analyses of variance (ANOVA). The block term was not significant, and pooled with the error term to increase the power of the test. To test for the effects of population source, bullfrog chemical cues, and their interaction on the activity and refuge use of wood frogs, I performed a multivariate analysis of covariance (MANCOVA), followed by a univariate analysis of covariance (ANCOVA). Mean wood frog mass was used as the covariate. In all analyses, wood frog mass was log-transformed and proportional data (survival, activity, and refuge use) were arcsine-square root transformed to normalize the data. All analyses were performed on SPSS 13.0 (2003) at $\alpha = 0.05$.

Results

Tadpole Growth and Survival

Bullfrog tadpoles affected the growth and survival of tadpoles from both wood frog populations (Table 1; Fig. 1). ANOVA results revealed that bullfrogs reduced survival for both allotopic and syntopic populations. In the presence of bullfrogs, survival of allotopic and syntopic larvae declined by 13.5 % and 6.1 %, respectively (Fig. 1B). There was no interaction between population source and the presence of the bullfrog tadpole on survival of wood frog larvae (Table 1), however, indicating that between-population survival rates were not different in the presence of bullfrogs. For all treatments, bullfrog survival was 100 %.

The growth of only allotopic wood frog larvae was reduced in the presence of overwintered bullfrog tadpoles (Table 1; Fig. 1A). In the presence of the bullfrog tadpole, growth of allotopic wood frog larvae declined by 61.9 %, whereas the growth of syntopic wood frog larvae declined by 16.1 %. Additionally, an interaction between wood frog population source and the presence of bullfrog indicates that, in the presence of the bullfrog tadpole, the growth rate of allotopic wood frog larvae is less than the growth rate of syntopic larvae (Table 1). The mean growth of all bullfrog tadpoles was 17.6 ± 0.32 mg/day and did not differ between allotopic and syntopic wood frog populations (ANOVA; $F_{1,8} = 2.124$; $P = 0.183$).

Activity and Refuge Use

There was an effect of population source, presence of bullfrog tadpole visual and chemical cues, and their interaction on the behavior (activity and refuge use) of wood frog larvae (Table 2; Fig. 2). There was no effect of the covariate (mean wood frog tadpole mass) on wood frog behavior (Table 2). Only allotopic wood frog tadpoles reduced their activity in the presence of bullfrog chemical cues (Fig. 2A). In the presence of bullfrog visual and chemical cues, activity of allotopic wood frog tadpoles declined by 66.2 %, whereas the activity of syntopic wood frog tadpoles declined by 18.8 %. Additionally, a population-by-treatment interaction indicates that, in the presence of bullfrog chemical visual and chemical cues, the activity of allotopic wood frogs was less than the activity of syntopic wood frogs (Fig. 2A).

Only allotopic wood frog tadpoles altered their microhabitat use in the presence of bullfrog tadpole visual and chemical cues, increasing their use of leaf-litter refugia by

16.0 % (Fig. 2B). There was no effect of bullfrog tadpole visual and chemical cues on syntopic wood frog microhabitat use, as use of leaf-litter refugia by syntopic tadpoles decreased by 1.2 % in the presence of bullfrog visual and chemical cues (Fig. 2B). There was also a population-by-treatment interaction indicating that, in the presence of bullfrog chemical cues, allotopic wood frog tadpoles increased refuge use more than syntopic wood frog tadpoles.

Discussion

The presence of overwintered bullfrogs had negative effects on fitness traits of both wood frog populations, reducing the survival of allotopic and syntopic wood frog tadpoles. Survival of larval amphibians has important population-level implications because population viability is often dependent upon the number of juveniles recruited into a population (Berven 1990). Predation is a plausible mechanism responsible for the decline in survival, as overwintered bullfrog tadpoles are capable of consuming congeneric tadpoles (Kiesecker and Blaustein 1997; Boone et al. 2004). Other mechanisms such as resource depletion resulting from exploitative competition could also have resulted in reduced tadpole survivorship (i.e., Kupferberg 1997; Lawler et al. 1999). Although I did not observe bullfrog tadpoles depredating any wood frog tadpoles, all dead wood frog larvae were missing upon the termination of the experiment, presumably having been consumed by the bullfrog tadpoles. Therefore, the decline in survivorship might be attributable to the interaction of predation and exploitative competition.

The results of this study emphasize the differential population responses of wood frog tadpoles to sympatric overwintered bullfrog tadpoles. Although bullfrogs negatively

affected the survival of tadpoles from both wood frog populations, bullfrogs only influenced the growth and behavior (activity and microhabitat use) of allotopic wood frogs. Other studies have cited similar effects of predators on wood frog behavior and development (Van Buskirk and Relyea 1998; Relyea 2002, 2004). My observations indicate that overwintered bullfrog tadpoles can induce physiological and behavioral shifts in wood frog larvae from allotopic populations.

Amphibian larval development also carries important population-level implications, as larger size at metamorphosis confers greater adult fitness in terms of survival and fecundity (Semlitsch 1985; Werner 1986). Overwintered bullfrog tadpoles may have reduced the growth of allotopic wood frog tadpoles via two mechanisms. Bullfrog tadpoles are superior competitors (Werner and Anholt 1996), and previous experiments have attributed declines in food resources to the presence of bullfrog tadpoles. Reduced food availability has negative consequences on amphibian growth (Lawler et al. 1999; Adams 2000). The reduced growth of allotopic wood frog tadpoles might also be a result of the predator-induced behaviors, as larval growth is often associated with activity (Relyea 2002, 2004) and increased use of refugia might also limit foraging ability (Petranka and Hayes 1998). Reduced activity and increased refuge use are common responses of many organisms to predation risk because they decrease the likelihood of being detected by predators (Lima and Dill 1990; Werner and Anholt 1996). However, this benefit often comes at a cost of slower growth (Relyea 2002, 2004). Allotopic wood frog tadpoles in my study responded to the presence of overwintered bullfrog tadpoles by decreasing activity levels and increasing use of refugia. Therefore, the reduced growth of allotopic wood frog tadpoles observed in my study might be

attributable to the effects of exploitative competition and/or reduced activity in response to the presence of bullfrog tadpoles.

Organisms exhibiting phenotypic plasticity in variable environments (e.g., varying levels of predation risk), are often at a selective advantage compared to organisms that do not exhibit plastic responses (Van Buskirk and Relyea 1998; Relyea 2002, 2004). For instance, Kiesecker and Blaustein (1997) reported that, in response to non-native overwintered bullfrog tadpoles, syntopic red-legged frogs (*R. aurora*) received a greater fitness benefit by exhibiting behavioral plasticity, as syntopic red-legged frog survivorship in the presence of bullfrog tadpoles was greater than that of allotopic wood frogs. My results contrast with these findings, however, indicating that sympatric wood frog populations received a greater fitness benefit if individuals exhibited a less plastic phenotype when in the presence of native overwintered bullfrog tadpoles.

Evolutionary experience may play an important role in the adaptation of such antipredator responses, as organisms that have evolved within the bullfrog's native range (i.e., wood frogs) might respond differently to the presence of overwintered bullfrog tadpoles than those organisms that share no evolutionary history with bullfrogs. In the presence of overwintered bullfrog tadpoles, syntopic wood frog tadpoles grew at a greater rate without exhibiting shifts in activity or microhabitat use. Other aquatic predators (e.g., *Anax* dragonfly larvae) are prevalent in fishless habitats, and previous studies have reported the ability of wood frog tadpoles to respond to such predators with antipredator behaviors (Petranka and Hayes 1998; Relyea 2002, 2004). Thus, it is likely that syntopic wood frog tadpoles still possess antipredator adaptations, but have been conditioned to

perceive overwintered bullfrog tadpoles as a lesser predation threat. Other behavioral or morphological adaptations might allow syntopic wood frogs to coexist with overwintered bullfrog tadpoles. For this reason, future studies should address other mechanisms responsible for the effects of native bullfrogs on sympatric amphibians, as well as those allowing coexistence with overwintered bullfrog tadpoles.

My results indicate that wood frog populations allotopic (naïve) to overwintered bullfrog tadpoles might not be able to maximize their fitness when developing in the same environments as overwintered bullfrog tadpoles. Although allotopic wood frog tadpoles perceived bullfrog tadpoles as a predation risk, responding with antipredator behaviors (reduced activity, increased refuge use), these responses may not be unique to bullfrog tadpoles. Numerous aquatic invertebrate predators (e.g., *Anax* dragonfly larvae) also exist in ephemeral aquatic habitats and may have created a selective pressure for allotopic wood frog tadpoles to develop similar antipredator behaviors despite lacking experience with overwintered bullfrog tadpoles. The responses of syntopic wood frog tadpoles to overwintered bullfrog tadpoles indicate that prolonged exposure to bullfrog tadpoles might permit other amphibian species amphibians to develop strategies that promote coexistence with bullfrogs.

Biological invasions have numerous adverse effects on native community dynamics (Soule 1990; Mack et al. 2000). Although the distributions of native bullfrogs and wood frogs are usually allotopic, fishless aquatic habitats that fail to dry between years may allow overwintered bullfrog tadpoles to become established. Therefore, bullfrogs have the potential to disperse to these habitats within their native range where they can have adverse effects on native amphibian community structure and composition.

This study demonstrates some of these effects, and provides evidence of some possible mechanisms that elicit these responses. Experience may play a role in the response of native amphibian species to the presence of bullfrogs, as those species that have evolved with bullfrogs (sympatric) respond differently than species that have not (allopatric). Despite these differences, bullfrog invasion within or outside of its native range might impact amphibian community dynamics. The effects of bullfrogs on native amphibians, however, are often associated with pond hydroperiod (Adams 2000). Therefore, future management strategies that focus on maintaining natural wetland hydroperiods and limit fish introductions should not only impede the spread of bullfrogs, but also provide quality breeding habitat for other amphibian species.

Acknowledgments. – This study was conducted in accordance with Institutional Animal Care and Use Committee guidelines (protocol #04-008). I would like to thank D. Mott and L. B. Hunt for permission to collect egg masses from their property. I am grateful to my committee members, Profs. S. Mullin, E. Bollinger, and K. Kruse for their constructive comments and advice on this manuscript.

Literature Cited

- Adams, M.J. 2000. Pond permanence and the effects of exotic vertebrates on anurans. *Ecological Applications* 10: 559-568.
- Altmann, J. 1974. Observational studies of behavior: sampling methods. *Behaviour* 49: 227-267.
- Batista, C.G. 2002. Natural history notes: *Rana catesbeiana* (effects on native anuran community). *Herpetological Review* 33: 131.
- Berven, K.A. 1990. Factors affecting population fluctuations in larval and adult stages of the wood frog (*Rana sylvatica*). *Ecology* 71: 1599-1608.
- Berven, K.A., and T.A. Grudzien. 1990. Dispersal in the wood frog (*Rana sylvatica*): implications for genetic population structure. *Evolution* 44: 2047-2056.
- Boone, M.D., E.E. Little, and R.D. Semlitsch. 2004. Overwintered bullfrog tadpoles negatively affect salamanders and anurans in native amphibian communities. *Copeia* 2004: 683-690.
- Carpenter, N.M., M.L. Casazza, and G.D. Wylie. 2002. Natural history notes: *Rana catesbeiana* (diet). *Herpetological Review* 33: 130.
- Corse W.A., and D.E. Metter. 1980. Economics, adult feeding, and larval growth of *Rana catesbeiana* on a fish hatchery. *Journal of Herpetology* 14: 231-238.
- Gosner, N. 1960. A simplified table for staging anuran embryos and larvae with notes on identification. *Herpetologica* 16: 183-190.
- Hecnar, S.J., and R.T. M'Closkey. 1997. Changes in the composition of a ranid frog community following bullfrog extinction. *American Midland Naturalist* 137: 145-150.

- Kats, L.B., J.W. Petranka, and A. Sih. 1988. Antipredatory defenses and the persistence of amphibian larvae with fishes. *Ecology* 69: 1865-1870.
- Kiesecker, J.M., and A.R. Blaustein. 1997. Population differences in responses of red-legged frogs (*Rana aurora*) to introduced bullfrogs. *Ecology* 78: 1752-1760.
- Kruse, K.C., and M.G. Francis. 1977. A predation deterrent in larvae of the bullfrog, *Rana catesbeiana*. *Transactions of the American Fisheries Society* 106: 248-252.
- Kupferberg, S.J. 1997. Bullfrog (*Rana catesbeiana*) invasion of a California river: the role of larval competition. *Ecology* 78: 1736-1751.
- Lawler, S.P., D. Dritz, T. Strange, and M. Holyoak. 1999. Effects of introduced mosquitofish and bullfrogs on the threatened California red-legged frog. *Conservation Biology* 13: 613-622.
- Lima, S.L., and L.M. Dill. 1990. Behavioral decisions made under the risk of predation: a review and prospectus. *Canadian Journal of Zoology* 68: 619-640.
- Mack, R.N., D. Simberloff, W.M. Lonsdale, H. Evans, M. Clout, and F.A. Bazzaz. 2000. Biotic invasions: causes, epidemiology, global consequences, and control. *Ecological Applications* 10: 689-710.
- Nussbaum, R.A., E.D. Brodie, Jr., and R.M. Storm. 1983. *Amphibians and reptiles of the Pacific Northwest*. University of Idaho Press, Moscow.
- Parker, I.M., D. Simberloff, W.M. Lonsdale, K. Goodell, M. Wonham, P.M. Kareiva, M.H. Williamson, B. Von Holle, P.B. Moyle, J.E. Byers, and L. Goldwasser. 1999. Impact: toward a framework for understanding the ecological effects of invaders. *Biological Invasions* 1: 3-19.

- Paton, P.W.C., and W.B. Crouch. 2002. Using the phenology of pond-breeding amphibians to develop conservation strategies. *Conservation Biology* 16: 194-204.
- Petranka, J., and L. Hayes. 1998. Chemically mediated avoidance of a predatory odonate (*Anax junius*) by American toad (*Bufo americanus*) and wood frog (*Rana sylvatica*) tadpoles. *Behavioral Ecology and Sociobiology* 42: 263-271.
- Phillips, C.A., R.A. Brandon, and E.O. Moll. 1999. Field guide to amphibians and reptiles of Illinois. Illinois Natural History Survey Manual 8. 300 pp.
- Relyea, R.A. 2002. Competitor-induced plasticity in tadpoles: consequences, cues, and connections to predator-induced plasticity. *Ecological Monographs* 72: 523-540.
- Relyea, R.A. 2004. Fine-tuned phenotypes: tadpole plasticity under 16 combinations of predators and competitors. *Ecology* 85: 172-179.
- SPSS. 2003. Statistical Package for the Social Sciences. Version 13.0. Chicago, Illinois.
- Semlitsch, R.D. 1985. Reproductive strategy of a facultatively paedomorphic salamander, *Ambystoma talpoideum*. *Oecologia* 65: 305-313.
- Semlitsch, R.D. 1988. Allotopic distribution of two salamanders: effects of fish predation and competitive interactions. *Copeia* 1988: 290-298.
- Smith, G.R., J.E. Rettig, G.G. Mittelbach, J.L. Valiulis, and S.R. Schaack. 1999. The effects of fish on assemblages of amphibians in ponds: a field experiment. *Freshwater Biology* 41: 829-837.
- Soule, M.E. 1990. The onslaught of alien species, and other challenges in the coming decades. *Conservation Biology* 4: 233-239.

- Stumpel, A.H.P. 1992. Successful reproduction of introduced bullfrogs, *Rana catesbeiana*, in northwestern Europe: a potential threat to indigenous amphibians. *Biological Conservation* 60: 61-62.
- Van Buskirk, J., and R.A. Relyea. 1998. Selection for phenotypic plasticity in *Rana sylvatica* tadpoles. *Biological Journal of the Linnean Society* 65: 301-328.
- Werner, E.E. 1986. Amphibian metamorphosis: growth rate, predation risk, and the optimal size at transformation. *American Naturalist* 128: 319-341.
- Werner, E.E., and B.R. Anholt. 1996. Predator-induced behavioral indirect effects: consequences to competitive interactions in anuran larvae. *Ecology* 77: 157-169.
- Werner, E.E., and M.A. McPeck. 1994. Direct and indirect effects of predators on two anuran species along an environmental gradient. *Ecology* 75: 1368-1382.
- Wilcove, D.S., D. Rothstein, J. Dubow, A. Phillips, and E. Losos. 1998. Quantifying threats to imperiled species in the United States. *BioScience* 48: 607-615.

Table 1. (A) MANOVA results for the effects of bullfrog (*Rana catesbeiana*) tadpoles (present / absent), wood frog (*R. sylvatica*) population source (allotopic / syntopic), and their interaction on wood frog growth and survival (df = 2,15 in all cases). (B) ANOVA results for wood frog growth and survival as a function of bullfrog tadpole presence and wood frog population source. Experiment was conducted between 22 March and 5 April 2005 (df = 1,16 in all cases).

A) MANOVA			
Source		F	P
Population		16.67	<0.0001
Bullfrog		36.40	<0.0001
Population x Bullfrog		13.55	<0.0001
B) ANOVA			
Source	Response	F	P
Population	Growth	30.29	<0.0001
	Survival	4.59	0.048
Bullfrog	Growth	53.31	<0.0001
	Survival	22.33	<0.0001
Population x Bullfrog	Growth	27.97	<0.0001
	Survival	0.663	0.427

Table 2. (A) MANCOVA results for the effects of bullfrog (*Rana catesbeiana*) tadpole visual and chemical cues (present / absent), wood frog (*Rana sylvatica*) population source (allotopic / syntopic), and their interaction on wood frog activity and microhabitat use (df = 2,14 in all cases). (B) ANCOVA results for wood frog activity and microhabitat use as a function of bullfrog tadpole presence and wood frog population source. In both analyses, wood frog mass was used as a covariate (df = 1,15 in all cases).

A) MANCOVA			
Source		F	P
Mass		1.90	0.186
Population		11.01	0.001
Bullfrog		22.18	<0.0001
Population x Bullfrog		11.11	0.001
B) ANCOVA			
Source	Response	F	P
Mass	Activity	3.41	0.085
	Microhabitat	0.975	0.339
Population	Activity	12.51	0.003
	Microhabitat	8.88	0.009
Bullfrog	Activity	40.06	<0.0001
	Microhabitat	4.50	0.051
Population x Bullfrog	Activity	16.70	0.001
	Microhabitat	5.14	0.039

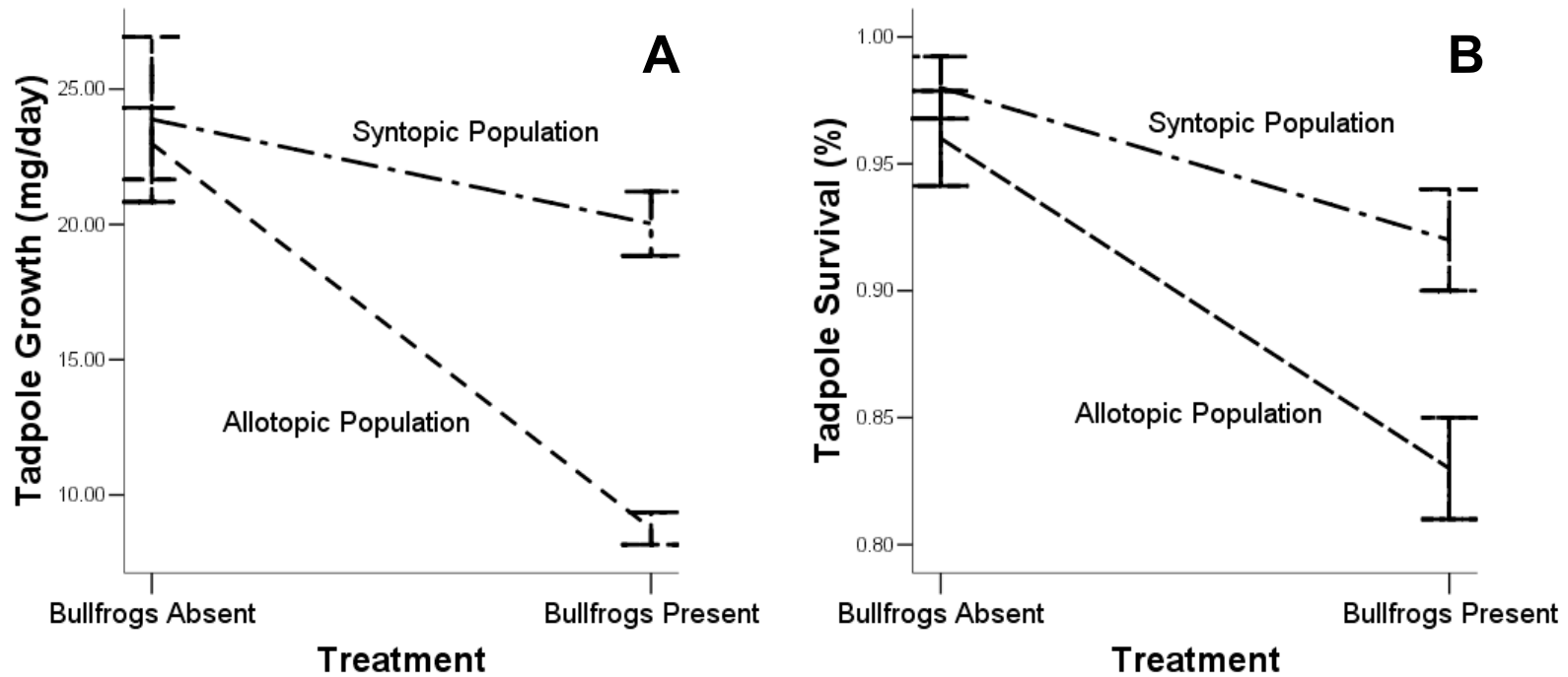


Figure 1. Growth (A) and percent survivorship (B) of wood frog (*Rana sylvatica*) larvae from syntopic or allotopic populations in response to the presence of a bullfrog (*R. catesbeiana*) tadpole. Experiments were conducted between 22 March and 5 April 2005. Data are represented as means ± 1 standard error.

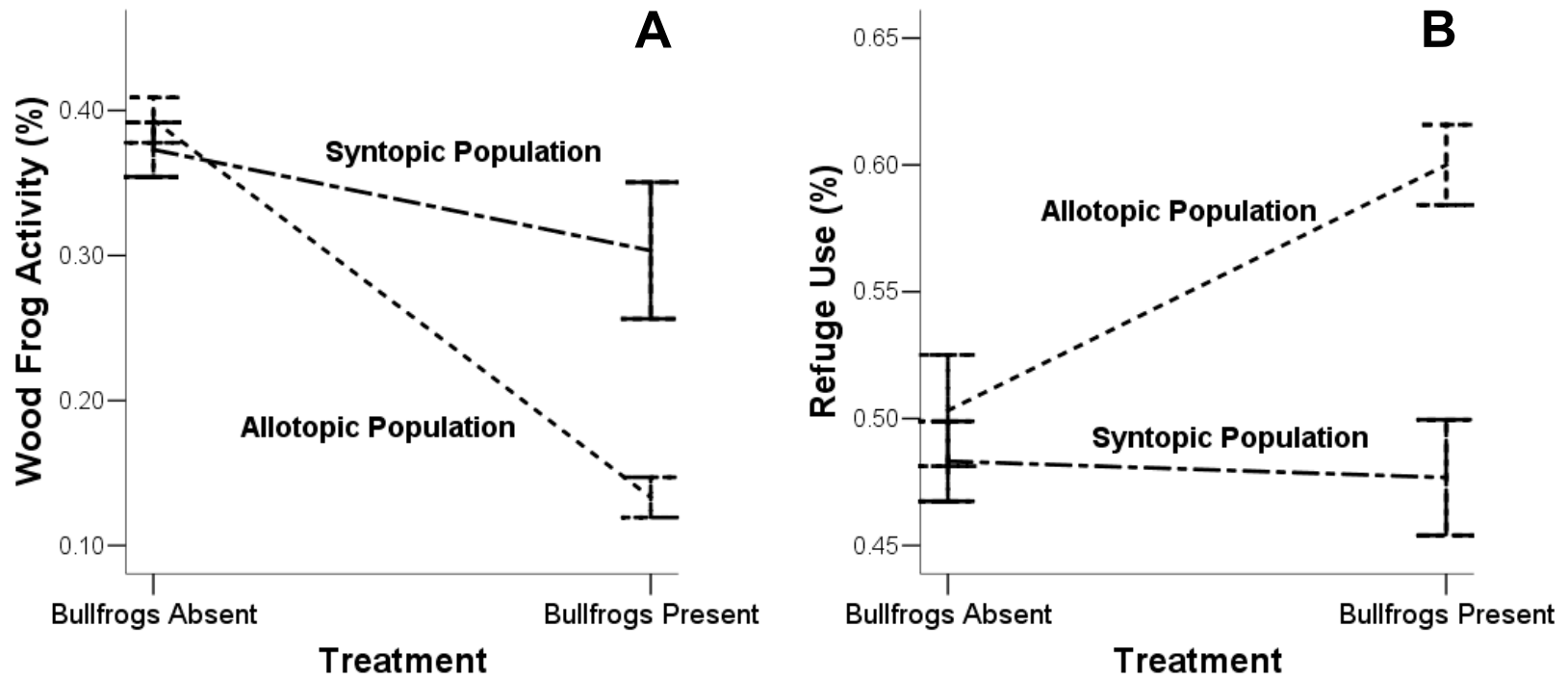


Figure 2. Activity (A) and refuge use (B) of wood frog (*Rana sylvatica*) larvae from syntopic or allotopic populations in response to the presence of bullfrog (*R. catesbeiana*) tadpole visual and chemical cues. Experiments were conducted between 22 March and 5 April 2005. Data are represented as means \pm 1 standard error.