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Chapter 3

Breeding Amphibian Population Declines Following Loss of Upland Forest Habitat Around Vernal Pools in Massachusetts, USA

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Abstract — Rapid urbanization is known to imperil populations of pond-breeding amphibians, in part through loss of terrestrial non-breeding habitat. However, we are aware of no published case studies that document changes in pond-breeding amphibian population size or structure following the loss of significant terrestrial habitat to urbanization. Here, we describe changes in breeding population size of three species of vernal pool-breeding amphibians following destruction of forested upland habitat surrounding vernal pools at two sites in Massachusetts. At the first site, Shopper's World, a small population of *Rana sylvatica* collapsed by the first spring breeding season following loss of 90% of the upland forest surrounding a vernal pool; no evidence of breeding by this species could be found by the third year following habitat alteration. At a second site, Marlboro Road, relatively large breeding populations of *R. sylvatica*, *Ambystoma maculatum*, and *A. laterale-jeffersonianum* hybrids declined sharply after 41% of contiguous upland forest within 300 m of a vernal pool was cleared for residential houses. Breeding populations of the same three species showed lesser changes in size at another vernal pool, separated by <100 m from the altered Marlboro Road pool, but on the other side of a road crossed by relatively few amphibians. Additionally, recapture rates of individually marked *Ambystoma maculatum* at the altered pool were lower following construction than at the relatively unaltered pool. By the third year following construction, breeding population sizes of *A. laterale-jeffersonianum* hybrids, but not of the other two species, had returned to pre-construction levels. Our observations demonstrate that breeding populations of amphibian species at both sites declined as a consequence of anthropogenic habitat loss. We recommend that more such case studies be conducted so that meta-analyses of the resulting data will allow conservationists to better predict the consequences of urbanization–related alterations of terrestrial amphibian habitat.

Key words — *Ambystoma*, Amphibian, Case Study, Habitat Loss, Population Decline, *Rana sylvatica*, Vernal Pool

Several recent studies have described the dependence of pond-breeding amphibian species on adjacent forested terrestrial habitat (Raymond and Hardy 1991; Windmiller 1996; Vasconcelos and Calhoun 2004; Regosin et al. 2005) and the critical importance of protecting such habitat to amphibian conservation efforts (Windmiller 1990; Semlitsch 1998; Calhoun et al. 2005). Among pond-breeding amphibian species in eastern North America, correlational studies have demonstrated that both the abundance and probability of occurrence of several amphibian species are lower in ponds surrounded by relatively little forested area or, conversely, by relatively high proportions of urbanized land cover and roadways (Windmiller 1996; Gibbs 1998; Homan et al. 2004; Rubbo and Kiesecker 2005; Clark et al. 2008).

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However, we are not aware of any studies that have compared pond-breeding salamander population structure prior to and following urbanization-related habitat alteration despite the fact that terrestrial habitat loss to urbanization is known to be a substantial threat to amphibian populations in eastern North America (Klemens 1993). Here, we provide data from two case studies that document declines in breeding population sizes of pond-breeding amphibian species following the destruction of forested upland habitat adjacent to vernal pool breeding sites. Additionally, we provide recommendations for conducting similar studies in the future.

Materials and Methods

Species Studied — The species studied, Wood Frog (*Rana sylvatica*), Spotted Salamander (*Ambystoma maculatum*), and mixed-ploidy hybrids of Blue-spotted Salamanders (*A. laterale*) and Jefferson Salamanders (*A. jeffersonianum*), all typically breed in fishless, often temporary ponds and other wetlands (Petranka 1998). In eastern North America, the breeding habitats most commonly associated with these species are often referred to as vernal pools in regulatory and scientific literature (Preisser and Clark 2000). The salamanders that we henceforth refer to as "hybrid Blue-spotted Salamanders" or "*Ambystoma laterale* hybrids" were predominately triploid females of the "*Ambystoma jeffersonianum* complex" (Uzzell 1963), with morphological characters of most individuals resembling those of "LLJ" triploids (*sensu* Klemens 1993). Measurement of mean erythrocyte areas of a sample of 121 female salamanders from the Marlboro Road site indicated that 75% were polyploid (Homan 2003; R. Homan et al. 2007).

Study Areas and Site Histories — We monitored amphibians in three vernal pools before and after significant adjacent upland habitat loss at two of the pools. The Shopper's World Vernal Pool was located in an urbanized, commercial district in Framingham, Massachusetts (N42° 18.10', W71° 23.55'). The vernal pool was fishless, 0.13 ha in area, and had a seasonal inlet and outlet and dried completely in the summer of some years but not in others. Prior to June 1994, the Shopper's World Vernal Pool was ringed on three sides by heavily trafficked roadways, parking areas, and shopping centers. To the east, however, the vernal pool abutted 6.3 ha of contiguous forested uplands and a larger area of marshland and forested wetland.

In June 1994, 90% of the contiguous upland forest adjoining Shopper's World Vernal Pool was cleared for the construction of a large, multiplex movie cinema and parking area (Fig. 1). The remaining upland forested habitat, totaling 0.6 ha in area, included a 38 m wide buffer zone adjacent to the edge of the vernal pool and two smaller habitat fragments to the north. Pre-construction drainage into the vernal pool from a large adjacent parking area was temporarily interrupted during construction and the vernal pool dried completely at the abnormally early date of 25 June 1995.

The second study site, Marlboro Road, was located in a predominately suburban area in Sudbury, Massachusetts, approximately 28 km west of Boston. The site contained two fishless vernal pools, separated by an undivided two-lane road with moderate vehicular traffic density, and a habitat mosaic of wetlands, forested uplands, and single-family housing surrounding the pools (Figs. 2, 3, see Regosin et al. 2005 for a more detailed description of the site). The two pools, referred to here as North Pool (N42° 24.24', W71° 25.35') and South Pool (N42° 24.20', W71° 25.27'), were, respectively, approximately 0.32 and 0.23 ha in spring-time surface area. During the six-year study period, the North Pool was entirely dry only once for a brief period of time. The South Pool, in contrast, dried completely by late September in most years.

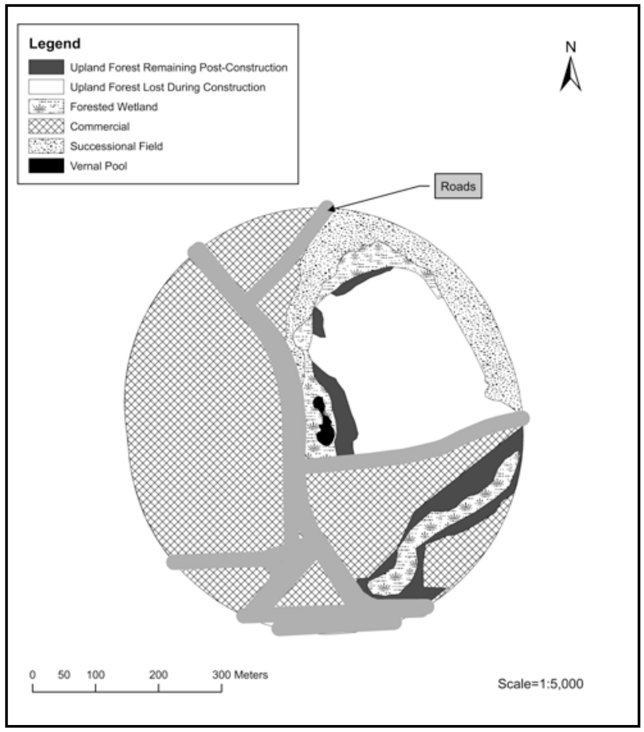

Fig. 1. Land cover changes in a 300 m buffer area drawn around Shopper's World Vernal Pool in Framingham, Massachusetts, that occurred during construction of a cinema complex in summer 1994. Land cover polygons interpreted from MassGIS orthophotos.

In March 1996, a new roadway was cleared to the west and south of the South Pool. During 1997 and 1998, 17 single-family houses were constructed in the vicinity of the South Pool. Land clearing was approximately 15% complete by March 1997 and more than 90% complete by March 1998. In all, residential construction removed 40.7% of the contiguous upland forest cover within 300 m of the South Pool (Fig. 2, Table 1). The North Pool, in contrast, suffered relatively little loss of adjacent contiguous forested habitat during the study period. During the summer of 1998, a single house was constructed northeast of the pool, causing a loss of 8.2% of contiguous upland forest within 300 m of the North Pool (Fig. 3).

Fig. 2. Land cover changes in a 300 m buffer area drawn around the more impacted South Pool (center point of figure), Marlboro Road site, Sudbury, Massachusetts. The relatively unaltered North Pool lies across Marlboro Road and a third vernal pool, not part of this study, is in the northeastern corner of the figure. Changes in land cover, as shown, occurred during construction of a residential housing subdivision, primarily in the summer of 1997. Land cover polygons were interpreted from MassGIS orthophotos. Location of 1997 drift fence array is shown (curved lines) in the "XYZ Area," an area that had been partially isolated from surrounding forest by road construction prior to the spring of 1997.

Fig. 3. Land cover changes in a 300 m buffer area drawn around the relatively unaltered North Pool (center point of figure), Marlboro Road site, Sudbury, Massachusetts. The more impacted South Pool lies across Marlboro Road and a third vernal pool, not part of this study, is in the northeastern corner of the figure. Changes in land cover, as shown, occurred during construction of a residential housing subdivision, primarily in the summer of 1997. Land cover polygons were interpreted from MassGIS orthophotos. Area A, as shown, was cleared of forest cover in the summer of 1998

Data Collection and Analysis at Shopper's World Site — In March 1991, we first observed chorusing *R. sylvatica* in the Shopper's World Vernal Pool and tadpoles of the species were observed in May of that year. In late winter 1992, we installed a drift fence — pitfall trap array around the margins of the Shopper's World Vernal Pool. Drift fences consisted of 0.9 m high plastic siltation fencing suspended from wooden stakes, with the bottom 0.2 m of plastic buried beneath the ground surface. We buried 25 pairs of 19-L plastic buckets on either side of the pond-encircling drift fence, at intervals of approximately 9 m. We checked traps daily from late March until late summer of the years 1992–1997. We closed the traps each summer after two weeks had elapsed since the last capture of a *R. sylvatica* metamorph leaving the pool;

Table 1. Changes in forested area within 300 m of Marlboro Road South and North Pools following construction in summer, 1997. Contiguous forested areas are those not bisected by Marlboro Road, which relatively few amphibians crossed.

Pool	Habitat Type	Pre-Construction Area (ha)	Post-Construction Area (ha)	% Change
South Pool	Total Forest	24.8	19.3	-22.2
	Contiguous Forest	16.2	11.4	-29.5
	Total Upland Forest	17.2	11.8	-31.4
	Contiguous Upland Forest	11.8	7.0	-40.7
North Pool	Total Forest	25.0	21.1	-15.7
	Contiguous Forest	15.1	14.4	-4.9
	Total Upland Forest	16.1	12.4	-23.0
	Contiguous Upland Forest	8.5	7.8	-8.2

gaps were left in the drift fencing to permit free movement of animals during periods when the traps were closed. Captured amphibians and other vertebrates were recorded by trap number, sexed when possible, and released opposite their points of capture. Captured amphibians were not marked during the course of the study. In addition to pitfall trap sampling, we counted *R. sylvatica* egg masses by walking transects through the vernal pool in April of 1993, 1997, and 2003. In 1997 and 2003, we also surveyed the vernal pool for newly hatched *R. sylvatica* tadpoles on the same day as the egg mass count, using numerous dip net sweeps throughout the pool.

To determine whether the number of *R. sylvatica* breeding at the Shopper's World Vernal Pool changed following the clearing of most of the adjacent forest, we divided the study into a "pre-construction" phase (March 1992–June 1994) and a "post-construction" phase (July 1994–August 1997). We then tested the null hypothesis that the total number of emigrating adult and newly metamorphosed *R. sylvatica* captured per year would be the same under pre- and post-construction periods. We compared pre- and post construction total captures of adult and newly metamorphosed *R. sylvatica* using contingency table analysis, with expected values in pre- and post-construction categories dependent upon the proportion of study years in each period. We used the number of adult *R. sylvatica* captured emigrating from the vernal pool each year as the basis of between-year comparisons since, in some years, the traps were not opened early enough to capture all immigrating breeders.

Additionally, we tested the hypothesis that the number of emigrating adult *R. sylvatica* captured in 1995, the spring after construction, was lower than would be expected given the previous years' capture data. To do so, we calculated the expected number of 1995 breeders using *R. sylvatica* demographic parameters provided by Berven (1990) as follows: annual survivorship for adult *R. sylvatica* = 13.4% (mid-point of values given in Berven 1990 for males and females), years to first breeding from metamorphosis = 1 for males and 2 for females (the modal values in Berven 1990), and survivorship from metamorphosis to first breeding = 7.8% for females (at age 2) and 37.9% for males (at age 1). We assumed that the sex ratio among newly metamorphosed *R. sylvatica* was 1:1 (Berven 1990).

Data Collection and Analysis at Marlboro Road Site — Our study commenced in March 1996 and continued until September 2001. For comparative purposes, we divided the study into "pre-construction" (March 1996–June 1997) and "post-construction" (July 1997–September 2001). We captured ambystomatid salamanders and *R. sylvatica* entering and leaving the South and North Pools with pond-encircling drift fence and pitfall arrays similar to those described above for the Shopper's World Vernal Pool. We checked pitfall traps daily during the spring breeding period and at least 3–4 times weekly throughout the summer and autumn months. We usually closed the pitfall traps during the winter months, leaving gaps in the fencing. We identified captured amphibians to species, sexed all adults, and released them on the opposite side of the fence. We weighed and measured samples of captured individuals. In 1996 and 1997, we individually marked all captured *Ambystoma* (990 hybrid *A. laterale* and 506 *A. maculatum*) with toe clips using the system described by Hero (1989). In the spring of 1998, we recorded the 1997 toe clip numbers for approximately the first 1/3 of *Ambystoma* captured that year (37.3% of hybrid *A. laterale* and 33.1% of *A. maculatum*). In 1998, 147 Spotted Salamanders and 262 hybrid Blue-spotted Salamanders were marked individually with subdermally inserted PIT-tags (Ott and Scott 1999). From spring 1998 onwards, most of the remaining captured *Ambystoma* were toe-clipped with batch-marks to indicate the year and nearest breeding pool at which they were first captured. Wood Frogs were generally not marked.

The first drift fence array, constructed in March 1996, enclosed 34% of the perimeter of the South Pool and included 10 pairs of pitfall traps. A partial drift fence also was constructed around 48% of the North Pool perimeter in 1996 but the pitfall traps were open for only a portion of the breeding migration. In early March 1997, the drift fence array was extended to include 82% of the South Pool perimeter using 24 pairs of pitfall traps; the remainder of the South Pool perimeter consisted of lawns and adjacent residential property to which we had no access. At the same time, the North Pool was completely encircled with a drift fence array consisting of 24 pairs of pitfall traps.

Additionally, in March 1997, we installed three parallel arcs of drift fencing across a 1.54 ha patch of upland forest, referred to henceforth as the "XYZ Area" (Fig. 2), that had been partially isolated from contiguous forested areas by newly constructed roadways. The XYZ Area drift fence array originally consisted of 19 pairs of pitfall traps. In the summer of 1997, approximately 67% of the XYZ Area was cleared for houses and lawns, leaving a reduced fragment of 0.50 ha of upland forest. The drift fence system in the XYZ Forest Fragment was reduced accordingly to 9 pairs of pitfall traps for the remainder of the study. In addition to the pond-encircling drift fence around the North Pool, a large array of drift fencing, mostly in arcs parallel to the Pool, was constructed in the upland and wetland forest surrounding the North Pool during 1998 and 1999; the entire array is described in Regosin (2003).

At the Marlboro Road Site, we estimated the breeding population sizes of *R. sylvatica* and *Ambystoma* spp. using pitfall trap captures for each study year at each of the two vernal pools. Breeding population estimates for *Ambystoma* spp., all of which were marked upon first capture, were calculated as: # captures of immigrants + # unmarked individuals among emigrants. For *R. sylvatica*, which were generally not marked, we used total captures of emigrating adults as our estimate of yearly breeding population size since some immigrants reached the breeding pools prior to our opening pitfall traps in at least one of the study years. Since the drift fence array around the South Pool in 1996 was smaller than in subsequent years, we calculated 1996 estimates of breeding population sizes by multiplying the number of individual amphibians of each species captured in 1996 by the ratio: # pitfall traps in 1997–2001 South Pool array / # pitfall traps in 1996 South Pool array (ratio = 2.4).

To determine whether breeding amphibian population sizes changed following destruction of upland forest habitat adjacent to the South Pool, we compared data from the 2-yr pre-construction and the 4-yr post-construction periods. Additionally, we used breeding population sizes at the relatively unaffected but adjacent North Pool (<100 m from the South Pool) as another point of comparison, asking whether the ratio of South Pool / North Pool captures for each species changed from the pre- to the post-construction period and whether the North Pool breeding populations showed changes similar to those observed in the South Pool following construction. Although forested habitat north and south of Marlboro were connected by amphibian migration and some individual *Ambystoma* metamorphs that emerged from each pool eventually bred in the other pool (J. Regosin unpubl. data), Marlboro Road served as a partial barrier to amphibian movement. In 1997, for example, only 3.5% of all *Ambystoma* individuals leaving the North Pool and 6.4% of all *Ambystoma* leaving the South Pool were captured in pitfall traps facing Marlboro Road and subsequently released on the opposite side of the road. Thus, we suggest that the partial barrier of Marlboro Road likely insulated the amphibian populations breeding in the North Pool from the full consequences of habitat alteration adjacent to the South Pool.

Results

Shopper's World Site — During the three pre-construction years of our study, we captured an average of 16.3 (range = 10–24) adult *R. sylvatica* emigrating from the Shopper's World Vernal Pool per year (Table 2). In sharp contrast, only one *R. sylvatica* was captured leaving the vernal pool in each of the three post-construction study years, a decline of 94% from the pre-construction mean. The 52 total *R. sylvatica* adults captured leaving the Shopper's World Vernal Pool were unequally distributed among the pre- and post-construction periods (chi-square = 41, df = 1, P <0.0001). The sharpest decline in the number of adult *R. sylvatica* captured occurred between 1994 and 1995, the period during which construction occurred. Consistent with this, the number of adults that would have been expected to breed in 1995, given capture data from 1992–1994 and using *R. sylvatica* survivorship parameters presented in Berven (1990), was 10.7; the actual number of emigrating adults captured was one.

As with adults, the number of juvenile *R. sylvatica* captured emigrating from the Shopper's World Vernal Pool was much lower in the four post-construction summer seasons (mean = 13.8, SE = 6.6; Table 2) than in the two pre-construction years (mean = 274, SE = 140.0), a decline of 95%. The 603 total *R. sylvatica* juveniles captured leaving the Shopper's World Vernal Pool were unequally distributed among the pre- and post-construction periods (chi-square = 899, df = 1, P <0.0001). In 1994, the year that construction activities resulted in abnormally early drying of the vernal pool, juvenile recruitment (1.3 juveniles per adult) was approximately 10% of the mean for the five years during which juveniles were captured (mean = 13.1). Searches for *R. sylvatica* egg masses and tadpoles in 1997 failed to detect any evidence of successful breeding and no juveniles were captured leaving the vernal pool in that year. Similarly, in 2003 we could not find a single egg mass or tadpole in the pool.

Marlboro Road Site: Overall Trends in Breeding Population Sizes — All three amphibian species had lower post-construction period (1998–2001) mean breeding population sizes at both vernal pools compared to pre-construction (1996–1997) (Table 3). This reflected the fact that, for all three species at both vernal pools, breeding population size values during the 1997–2001 period were highest in 1997. However, relative declines from 1997 pre-construction values to the post-construction mean values were greater at the South Pool, where more forested habitat was lost, than at the North Pool for all three amphibian species.

Upland forest destruction adjacent to South Pool occurred largely between the 1997 and 1998 breeding period. Notably,

Table 2. Annual totals of *Rana sylvatica* captured in pitfall traps leaving the Shopper's World Vernal Pool. Clearing of adjacent upland forest occurred in June 1994, prior to 1994 metamorph dispersal. Thus, for metamorphs, the pre-construction mean was for the years 1992-1993, and for adults it was 1992-1994. Standard errors are given in parentheses.

Year	# Adults	# Metamorphs	Metamorphs/ Emigrating Adult	Drying Date
1992	15	414	27.6	None
1993	10	134	13.4	July 6
1994	24	32	1.3	June 25
1995	1	11	11.0	July 16
1996	1	12	12.0	None
1997	1	0	0.0	July 2
Pre-Construction	16.3 (± 4.1)	274.0 (± 140.0)	20.5 (± 7.1)	
Post-Construction	1.0 (± 0.0)	13.8 (± 6.6)	8.1 (± 3.2)	

for all three species, the single greatest between-year decline at either vernal pool occurred at the heavily impacted South Pool between 1997 and 1998, with *R. sylvatica* numbers declining at the South Pool by 87%, hybrid *A. laterale* by 51%, and *A. maculatum* by 47% (Table 3). At South Pool, during the four study years following upland forest clearing, numbers of breeding *R. sylvatica* and *A. maculatum* never exceeded 60% of pre-construction values, regardless of whether pre-construction values are defined by the mean of 1996 and 1997 values or by 1997 values alone (Fig. 4, Table 3). Similarly, in the post-construction years, the ratio of South Pool / North Pool breeding population size estimates for *R. sylvatica* and *A. maculatum* never attained their 1997 pre-construction levels (Fig. 5). Hybrid *A. laterale* breeding population size at the South Pool, in contrast, exceeded 60% of the 1996–1997 mean pre-construction values in all post-construction years and exceeded the pre-construction mean in one year (2000). Furthermore, the ratio of hybrid *A. laterale* captured exiting the South Pool vs. the North Pool exceeded the 1997 level during the last two years of the study.

One result of this difference in population trajectories of the two *Ambystoma* species is that the ratio of breeding adult *A. maculatum* to hybrid *A. laterale* at the relatively impacted South Pool decreased sharply after the construction period, from a 1996–1997 mean of 2.46 to near parity (1.18) in the post-construction years. At the relatively undisturbed North Pool, in contrast, hybrid *A. laterale* captures exceeded *A. maculatum* captures in all study years and the mean annual ratio of spotted to hybrid *A. laterale* in post-construction years (0.45) was similar to that observed in 1997 (0.37) and 1996 (0.64 among a sample of 95 total *Ambystoma* captured).

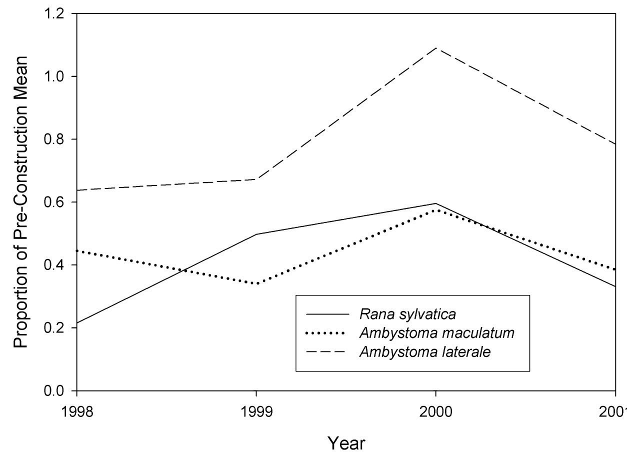

Fig. 4. Number of individual breeding amphibians of three species captured at South Pool in post-construction years, expressed as proportion of the mean number of breeding individuals captured in 1996 and 1997, the pre-construction period.

Marlboro Road Site: Trends among Sub-Population Groups — In spring 1998, immediately following the bulk of forest clearing near the South Pool, we observed that a smaller proportion of *A. maculatum* marked at the South Pool in spring 1997 were

Table 3. Breeding population sizes at Marlboro Road vernal pools. Construction – related habitat loss occurred primarily between 1997 and 1998 adjacent to the South Pool; 1996-1997 is the pre-construction period. Data for 1996 at the North Pool were insufficient to derive accurate breeding population size estimates. Standard errors are given in parentheses.

Pond	Year	# *R. sylvatica*	# *A. maculatum*	# *A. laterale* hybrids
South Pool	1996	701	288	81
South Pool	1997	3038	206	151
South Pool	1998	403	110	74
South Pool	1999	932	84	78
South Pool	2000	1115	142	126
South Pool	2001	618	95	91
South Pool Mean Pre-Construction		1869.5 (±1168.5)	247.0 (±41.0)	116.0 (±35.0)
South Pool Mean Post-Construction		767.0 (±158.9)	107.8 (±12.6)	92.2 (±11.8)
South Pool % Change Pre- vs. Post -Construction		-59.0	-56.4	-20.5
South Pool % Change 1997 vs. Post-Construction		-74.8	-47.7	-38.9
North Pool	1997	844	300	839
North Pool	1998	413	295	763
North Pool	1999	442	271	622
North Pool	2000	695	251	582
North Pool	2001	332	200	376
North Pool Mean Post-Construction		470.5 (±78.4)	254.2 (±20.2)	585.8 (±80.0)
North Pool % Change 1997 vs. Post-Construction		-44.3	-15.2	-30.2

recaptured that year (4.9% of a total of 206 marked) than was the case for *A. maculatum* captured at the relatively unaffected North Pool in 1997 (12.7% of a total of 300, Fisher's exact test, two-tailed P = 0.01). There was no significant difference among recapture rates for hybrid *A. laterale* marked at the two vernal pools in 1997 (South and North Pool respective recapture rates of 9.9% of a total of 151 and 12.4% of a total of 839). As noted previously, only approximately the first 1/3 of salamanders captured in 1998 were checked for 1997 toe-clips.

In the XYZ Area, south of the South Pool, which suffered a 67.5% reduction in forest cover between the springs of 1997 and 1998 (Fig. 2), annual per-trap captures of all three study species in the XYZ drift fence array fell sharply after 1997 (Table 4). Compared to 1997, mean annual per-trap captures for the remaining 4 yrs of the study declined by 77% for hybrid *A. laterale*, 84% for *A. maculatum*, and 96% for *R. sylvatica*. Additionally, the proportion of 1997 breeding *A. maculatum* marked in the XYZ area in 1997 that were recaptured in 1998 was 79% lower than the recapture rate of *A. maculatum* captured in all other traps in 1997, though the difference was not statistically significant (Fisher's exact test, two-tailed P = 0.11, Table 4). In contrast, the proportion of hybrid *A. laterale* captured in the XYZ array in 1997 that were observed among 1998 captures (12.2%) was almost identical to the recapture proportion of hybrid hybrid *A. laterale* first captured in all other traps in 1997 (12.0%, Fisher's exact test, two-tailed P = 1.0).

Discussion

Impacts of Upland Forest Destruction — Results of our studies at both the Shopper's World and Marlboro Road sites do not constitute an experimental study as they lack replication and true control. These data therefore cannot confirm a causal link between the large-scale habitat alterations that occurred adjacent to vernal pools and the subsequent changes that we observed in vernal pool-breeding amphibian populations. Nevertheless, we find that the data show sharp declines in breeding population sizes of amphibian species occurring immediately following destruction of upland forest surrounding two vernal pools, and comparatively little change at a relatively unaffected third pool. At the Shopper's World site, destruction of 90% of the upland forest patch adjacent to a vernal pool was followed by a 94% decline in the numbers of adult *R. sylvatica* captured relative to the three-year pre-construction study period. Indeed, in each of the three post-construction study years, only a single *R. sylvatica* adult was captured leaving the vernal pool. By 1997, three years after the upland forest clearing, we were unable to locate either *R. sylvatica* eggs or tadpoles in the pool. No metamorphs were captured leaving the pool that summer. Six years later, in 2003, we were still unable to find any *R. sylvatica* egg masses or tadpoles in the Shopper's World Vernal Pool.

Rana sylvatica populations are characterized by high between-year variability in numbers of breeding adults (Berven and Grudzien 1990; Berven 1995), and we cannot exclude the possibility that the dramatic change we observed in *R. sylvatica* captures at the Shopper's World Vernal Pool was the result of natural fluctuations. However, in reviewing available multi-year data on *R. sylvatica* breeding population sizes (Berven and Grudzien 1990; Berven 1995; B. Windmiller unpubl. data), we have yet to observe an instance of a sustained decline in a *R. sylvatica* population of similar magnitude to the 94% decline that we observed for three years at the Shopper's World Vernal Pool relative to the three pre-construction study years. Berven and Grudzien (1990), for example, provide 7 yrs of data on *R. sylvatica* breeding population sizes at six different ponds in a national forest in Virginia. Among 24 possible within-pond comparisons of two-year periods to preceding

Table 4. Total annual captures of adult amphibians per pitfall trap and between-year recapture rates for the XYZ Area, a 1.54 ha forested fragment of which 67.5% was cleared in summer 1997. Recapture rates are percentages of the cohort of 1997 — breeding individual *Ambystoma* that were recaptured in a sample of 1998 — breeders; the 1997 cohort is divided here between those individuals captured in 1997 in the XYZ Area pitfall traps and those 1997 — breeders captured elsewhere on the site.

	Year	*A. laterale* Hybrids	*A. maculatum*	*R. sylvatica*
	1997	3.00	3.53	35.37
	1998	0.67	0.33	2.00
XYZ Area	1999	0.67	0.44	2.29
per-Trap Captures	2000	1.00	0.55	1.14
	2001	0.44	0.89	0.14
Mean Post-Construction		0.69	0.56	1.39
% Change 1997 vs. Post-Construction		-77.0%	-84.1%	-96.1%
XYZ Recapture Rate	1997 - 1998	12.2%	2.1%	n.a.
	(N)	(41)	(47)	
All Other Trap Recaptures	1997 - 1998	12.0%	10.2%	n.a.
	(N)	(949)	(459)	

two-year periods, the largest observed decline for a Wood Frog breeding population was 83%. Among the 5 yrs of *R. sylvatica* breeding population data reported here for the relatively undisturbed North Pool in Sudbury, Massachusetts, the greatest between-year decline in *R. sylvatica* breeding numbers was 49%. Additionally, at the Marlboro Road Site, less than 12 km from the Shopper's World Site and thus sharing similar annual precipitation patterns, *R. sylvatica* populations were high in 1996 and 1997, two years during which the Shopper's World Vernal Pool *R. sylvatica* population was near extirpation.

Our data suggest that clearing of most of the adjacent Shopper's World upland forest in June 1994 caused exceedingly high mortality rates among *R. sylvatica* adults and juveniles in the period between forest clearing and breeding in March 1995, a conclusion supported by our calculation of predicted 1995 *R. sylvatica* captures using stage-specific demographic data from Berven and Grudzien (1990). Since, in northeastern North America, few *R. sylvatica* occur in dry upland forest during June (Regosin et al. 2003), it is unlikely that many *R. sylvatica* were directly killed during the forest clearing operations. Instead, the observations of Regosin et al. (2003) suggest that upland forest, particularly areas within 100 m of vernal pool breeding sites, is critical to *R. sylvatica* populations during the winter months. Perhaps, deprived of adequate overwintering sites, the *R. sylvatica* of the Shopper's World population experienced unusually high winter mortality rates during the winter of 1994–1995.

The effects of apparently low survivorship among *R. sylvatica* in the terrestrial environment during the year between March 1994 and March 1995 were compounded by the very low rate of juvenile recruitment during the summer of 1994, an event probably caused by the unusually early drying of the vernal pool as a consequence of construction activities. With little suitable upland forest remaining, the relatively few newly metamorphosed *R. sylvatica* that emerged from the vernal pool in 1995 and 1996 were apparently unable to survive to maturity in numbers sufficient to revive the population, which was small at the outset of the study. Surrounded by heavily trafficked roadways and extensive urbanization, the Shoppers' World *R. sylvatica* population was cut off from rescue through recolonization (cf. deMaynadier and Hunter 2000) and declined to extirpation.

In contrast to Shopper's World, the magnitude of post-construction declines observed in three vernal pool-breeding amphibian species studied at the Marlboro Road South Pool are within the range of reported natural variation in yearly breeding population sizes reported for *R. sylvatica* (Berven and Grudzien 1990; Berven 1995) and Spotted Salamanders (Husting 1965; Sexton et al. 1986). Moreover, all three species also showed declines at the relatively undisturbed North Pool relative to pre-construction conditions in 1997. The fact that mean annual breeding population sizes of all three study species were lower at the South Pool in the four year post-construction period than in the two year pre-construction period is not, in of itself, convincing evidence that the observed

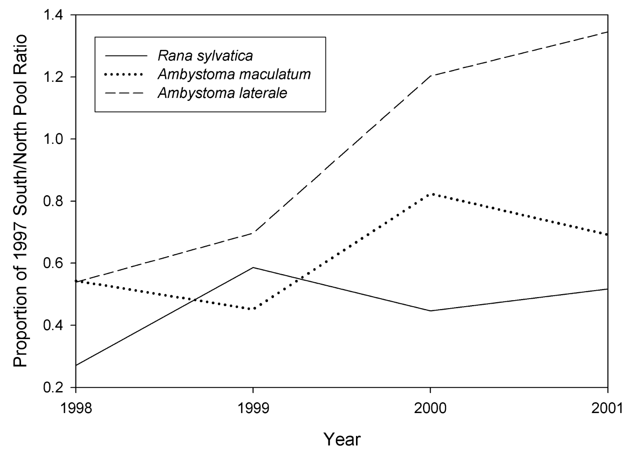

Fig. 5. Ratio of the number of individual breeding amphibians of three species captured at South Pool / North Pool post-construction years, expressed as proportion of the 1997 pre-construction ratio of South / North Pool captures of the same species.

declines were the result of construction-related changes in land cover.

However, taken as a whole, data comparing pre- and post-construction amphibian population structure at the South and North Pools strongly suggest that the loss of forested upland habitat during construction near the South Pool in 1997 caused increased mortality of resident *R. sylvatica* and *Ambystoma* populations and subsequent declines in the numbers of all three species. First, the 6 yrs of available breeding population data for the South Pool and 5 yrs of data for the North Pool provide nine between-year comparisons for each of the three species. For all three species, the single greatest between-year decline was recorded at the South Pool between 1997 and 1998, the primary period of habitat alteration. Second, although breeding population size data for the North Pool does not serve as a true experimental control for data recorded at the South Pool, these two adjacent vernal pools shared nearly identical climatic conditions. Additionally, *A. maculatum* egg mass count data from successive years suggest that variability in breeding effort for this species may be correlated among ponds within a region extending over tens of kilometers (Ruth et al. 1993; Windmiller 1996; B. Windmiller unpubl.data). Thus, it is notable that in the North Pool, <100 m from the South Pool, breeding population numbers for both species of *Ambystoma* showed little change between 1997 and 1998, and the greatest between-year decline for all three species at the North Pool was between 2000 and 2001, a period during which population size data declined sharply for all three species at both pools. Moreover, the ratio of breeding individuals at the South vs. North Pools declined sharply from 1997 to 1998 for all three species (> 45% decline for each species, Fig. 5) and, compared to 1997, remained depressed by >30% for all three species during the 1999 breeding season. In the 4-yr post-construction period, only hybrid *A. laterale* ever regained the 1997 ratio of North vs. South Pool captures.

Unlike the fate of the small *R. sylvatica* population at the Shopper's World Vernal Pool, the relatively large pre-construction populations of *R. sylvatica*, hybrid *A. laterale*, and *A. maculatum* at the South Pool persisted after alteration of much of the surrounding upland forest habitat. The breeding population sizes of all three species, however, declined sharply over the two years following construction and, for *A. maculatum* and *R. sylvatica*, remained at less than 60% of their pre-construction levels throughout our 4 yr post-construction monitoring period. Only the hybrid *A. laterale* population recovered to pre-construction breeding values. Recapture data for individually-marked *A. maculatum* suggest that mortality rates were higher for individuals that had resided in the altered South Pool vicinity during the construction period compared to those that had been last captured near the relatively unaffected North Pool. Although we cannot surmise the cause of increased mortality among South Pool-breeding amphibians during and following clearing of 41% of upland forest around their breeding pool, our frequent checks of the relatively sparsely trafficked subdivision roadways suggest that road-kill was not a primary factor.

Conservation Implications — Extirpation of the Shopper's World *R. sylvatica* population occurred even though all Federal, state, and local wetlands protection measures were applied to their fullest extent. Not only were all wetland areas untouched by construction, the Town of Framingham Conservation Commission required the developer of the site to leave a 38 m-wide wooded buffer area around the wetland boundary intact. Similarly, the Town of Sudbury Conservation Commission required the developers of the housing subdivision adjacent to the South Pool to leave intact all wetland areas on the sites, as well as a 30.5 m-wide forested buffer zone around the vernal pool. Additionally, because of the presence of a state-protected species, *Ambystoma laterale* complex (Species of Special Concern, as listed by the Massachusetts Division of Fisheries and Wildlife), the developers were further required to spare several patches of upland forest as far as 220 m from the South Pool. In our experience in Massachusetts, few construction projects near vernal pools in Massachusetts are as tightly regulated as those described here; most such projects are permitted to clear upland forest areas to within 10 m or less of the wetland boundaries. Our case studies at Shopper's World and Marlboro Road therefore support the conclusion, reached by others as well, that wetland protection regulations are inadequate tools to protect populations of amphibian species that depend upon both wetland and upland habitats from anthropogenic habitat alteration (Windmiller 1990; Semlitsch 1998; Calhoun and Klemens 2002; Homan et al. 2004).

The fate of the Shopper's World Wood Frog population represents a fairly extreme case of urbanization-related destruction of critical terrestrial habitat for vernal pool-breeding amphibians. Even in the densely settled northeastern United States, relatively few vernal pools remain within a setting as heavily urbanized as the Shopper's World Vernal Pool (Gibbs 1998). Additionally, relatively few construction projects adjacent to vernal pool breeding sites result in the conversion of as much as 90% of all surrounding upland forest habitat into buildings and paved surfaces, as was the case at Shopper's World. However, levels of upland forest destruction around vernal pools similar to or greater than those observed at the Marlboro Road site are common in the rapidly urbanizing northeastern United States (B.Windmiller pers. obs.), suggesting that anthropogenic amphibian population declines comparable to those that we observed at the South Pool are widespread. Furthermore, data from the XYZ Area, where per-trap amphibian capture rates declined much more than the proportionate reduction in forested area, indicate that leaving small patches of forest surrounded by houses, lawns, and roads is of dubious value to the conservation of vernal pool-breeding amphibians.

Finally, our data suggest the seemingly paradoxical conclusion that hybrid *A. laterale*, the only one of the three taxa studied that is protected as a rare species in Massachusetts, may be the most resistant of the species to the scale of upland forest destruction that occurred at the Marlboro Road study site. If this is generally true, possible explanations may include a greater ability by *A. laterale*-complex salamanders to use forest edge or wetland habitats (B. Windmiller unpubl. data) and the release of hybrid *A. laterale* from interspecific competition with *A. maculatum* if the latter species is reduced in number by habitat loss. However, urbanization may pose a particular threat to populations of hybrid *A. laterale-A. jeffersonianum*. Rebecca Homan et al. (2007) observed that the ratio of female to male hybrid *A. laterale* at the North Pool averaged 28.3:1 and increased over the course of the study. If the relatively few males present in such populations are particularly vulnerable to habitat destruction, urbanization may imperil populations of hybrid *Ambystoma* through removing opportunities for females to breed successfully, whether through gynogenesis or sexual reproduction (Klemens 1993). Future studies comparing the responses of these two species to habitat alteration should note the ploidy and genotypic make-up of *A. laterale-A. jeffersonianum* complex salamanders that are studied (Lowcock et al. 1987).

Recommendation for Future Studies — Regulators and conservationists would benefit greatly from an increased understanding of the specific responses of vernal pool-breeding amphibian species to urbanization. Correlational studies are able to provide considerable insight into the regional-scale trends in relationships between urbanization and amphibian abundance and distribution. Yet regulators are frequently called upon to make decisions of considerable economic importance on a much smaller site-specific scale. Studies such as the ones described here may allow regulators to optimize conservation outcomes by distinguishing between projects that are likely to cause only temporary increases in amphibian mortality versus those that carry a significant risk of causing permanent declines or extirpations. Additionally, data on the abilities of different amphibian species to survive in and move through an urbanized habitat matrix would allow regulators to more confidently choose between various site development alternatives.

Because it is inherently difficult to conduct controlled and replicated studies that involve large-scale conversion of natural habitat into urban land cover, we believe that it is important to maximize the knowledge gained from case studies such as ours. Meta-analyses of many well-conceived case studies that compare pre- and post-construction patterns in amphibian demography and habitat use might offer an excellent means of elucidating the responses of amphibian populations to urbanization. This would be particularly true if unpublished studies, perhaps from local consulting projects, could be compiled to reduce problems associated with publication biases (e.g., Sterne et al. 2000). Based on our experience with the studies reported here, we offer the following recommendations for the design of pre- vs. post-construction case studies of amphibian populations: (1) Maximize pre-construction data collection by beginning the study as soon as possible after the opportunity arises and initiating all study components at the outset; (2) Establish similar study protocols at nearby research areas that will be unaffected by the proposed construction; (3) Individually mark as many amphibians as possible during the pre-construction period, using marking systems such as PIT-tags (Ott and Scott 1999) or visible implanted elastomers (Bailey 2004) that allow identification over the lifetime of the amphibian; and (4) Track the movements and fates of individual amphibians through the pre- and post-construction landscape using radiotelemetry (Madison 1997), concentric drift fence arrays (Windmiller 1996; Regosin 2003), or direct observation. We recognize, however, that logistical considerations, timing of the permitting and construction process, and land ownership patterns will greatly affect the eventual study design.

The costs for pre- vs. post-construction case studies are most appropriately borne by developers seeking permits to alter wetlands, vernal pools, or other regulated habitat areas, with maximum study costs capped at a fixed percentage of estimated project costs. To reduce the possibility for conflicts of interest, we recommend that funding for the study be transferred from the developers to conservation agencies, which would, in turn, contract suitable researchers.

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