The Relative Abundance of the Juvenile Phase of the Eastern Red-Spotted Newt at Harvard Forest Prior to the Arrival of the Hemlock Woolly Adelgid

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Abstract - The invasive insect pest *Adelges tsugae* (Hemlock Woolly Adelgid) threatens the ecologically unique *Tsuga canadensis* (Eastern Hemlock)-dominated forest type throughout its range. Relatively little is known about how the loss of this forest type will affect the relative abundance of amphibians. This study assessed the relative abundance of the juvenile phase of *Notophthalmus viridescens viridescens* (Eastern Red-spotted Newt, Red Eft) in Eastern Hemlock-dominated stands (n = 5) and mixed deciduous stands (n = 5) at Harvard Forest in Petersham, MA, using both transect surveys of the forest floor surface (n = 368 Red Eft observations over four seasons), and intensive searches of quadrats (n = 27 Red Eft observations over two seasons). Using data from transect surveys, the average relative abundance of Red Efts was more than two times greater in Eastern Hemlock-dominated stands than in mixed deciduous stands, however the differences were not statistically significant (P = 0.146). Quadrat surveys yielded relative abundance estimates for Red Efts that were more than 5 times greater in Eastern Hemlock-dominated stands than in mixed deciduous stands, but again the differences were not statistically significant (P = 0.213).

Introduction

The long-lived, shade tolerant conifer species *Tsuga canadensis* Carrière (Eastern Hemlock) has been described as a foundation species that creates unique habitat and impacts core ecosystem processes (Ellison et al. 2005a). This ecologically important species is threatened throughout its range by the invasive insect, Adelges tsugae Annand (Hemlock Woolly Adelgid [HWA]) (Hemiptera: Adelgidae; Orwig 2002, Orwig and Foster 1998). Native to Japan, HWA was first discovered in Virginia in the 1950s (Souto et al. 1996) and has spread throughout a great percentage of Eastern Hemlock's range via a number of dispersal agents including wind, birds, deer, and humans (McClure 1990). As of 2004, when this study was conducted, HWA was present in 50% of Eastern Hemlock-dominated stands in Massachusetts, but was not yet present at Harvard Forest (Orwig and Povak 2004). Unfortunately, no natural predators of the aphid-like insect occur in the United States (McClure 1995). HWA can cause mortality in all age classes of Eastern Hemlock within 4–10 years of infestation (McClure 1991). In central Massachusetts, cold winter temperatures have slowed mortality of Eastern Hemlock in infested stands, though anticipated warming trends threaten to accelerate rates of mortality and dispersal (Orwig et al. 2012). At Harvard Forest, Eastern Hemlock will likely be replaced by mixed deciduous species such as Betula lenta L. (Black Birch), Quercus rubra L.

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(Red Oak), and *Acer rubrum* L. (Red Maple) (Orwig and Foster 1998, Sullivan and Ellison 2006).

Eastern Hemlock-dominated forests are structurally unique, providing important habitat to assemblages of invertebrates, amphibians, birds, and mammals (Ellison et al. 2005b, Ingwell et al. 2012, Mathewson 2009, Tingley et al. 2002, Yamasaki et al 2000). For example, the dense canopy of Eastern Hemlock-dominated forests provides breeding habitat preferred by several songbird species including *Dendroica virens* Gmelin (Black-throated Green Warbler), *Vireo solitarius* Wilson (Solitary Vireo), and *Dendroica fusca* Müller (Blackburnian Warbler) (Benzinger 1994, Tingley et al. 2002, Yamasaki et al. 2000). These dense canopies greatly reduce light penetration resulting in forest floors that are cooler, darker, and with moister soil than surrounding mixed deciduous stands (Benzinger 1994, Lustenhouwer et al. 2012, Rogers 1980). Many groups of invertebrates are more abundant in Eastern Hemlock litter than mixed deciduous litter including collembolans, mites and ticks, coleopterans, hymenopterans, and dipterans (8,5,4,2.5, and 2.5 times more abundant, respectively; Hartman 1977).

Soils in Eastern Hemlock-dominated forests are more acidic than in mixed deciduous forests due to the species ability to thrive in acidic conditions and the acidity of hemlock needles themselves (Benzinger 1994). This association with high soil acidity led to the perception that amphibians are less abundant in Eastern Hemlock forests. Wyman and Jancola (1992) suggested the relative abundance of Plethodon cinereus Green (Eastern Red-backed Salamander) was found to be lower in Eastern Hemlock-dominated stands than in Fagus grandifolia Ehrh. (American Beech) stands in Albany County, NY due to higher soil acidity in the former. However, the relative abundance of Eastern Red-backed Salamanders was found to be greater in Hemlock-dominated stands than in mixed deciduous stands at Harvard Forest using surveys of artificial cover objects (ACOs; Mathewson 2009). No difference was found in the relative abundance of Eastern Red-backed Salamanders in the two forest types using intensive searches of quadrats (Mathewson 2009). At Harvard Forest, soil pH in Eastern Hemlock-dominated stands, while lower than in mixed deciduous stands, is above the level that negatively impacts the relative abundance of Eastern Red-backed Salamanders (Mathewson 2009, Wyman and Jancola 1992).

Notophthalmus viridescens viridescens Rafinesque (Eastern Red-spotted Newt) is the second most widely distributed salamander in North America (Petranka 1998). It is also perhaps the most familiar salamander, especially as a terrestrial juvenile, or Red Eft, due to its bright coloration and active diurnal behavior on the surface of the forest floor (Petranka 1998). This bright coloration serves as a warning to potential predators of the Red Eft's highly toxic skin (Hurlbert 1970). Although there are several variations, the most common life cycle involves 4 distinct stages—egg, aquatic larva, terrestrial Red Eft, and aquatic adult (Petranka 1998). The Red Eft stage usually lasts from 4–7 years (Petranka 1998). Despite their ubiquity, little is known regarding differences in the relative abundance of Red Efts in different forest types, and no study has ever assessed the relative abundance of Red Efts in

forests dominated by Eastern Hemlock. The only estimate of the relative abundance of Red Efts comes from an oak-pine woodland located 800 m from a breeding pond in central Massachusetts where the density of Red Efts was 0.03 individuals/m² (Healy 1975).

Red Efts do not appear to be affected by low soil pH likely because their skin is coarser than the lungless salamanders, making them less sensitive to acidic soils (Wyman and Jancola 1992). Therefore, low soil pH does not likely impact the relative abundance of Red Efts in Eastern Hemlock-dominated forests at Harvard Forest. However, food supply and moisture are important factors in habitat selection by Red Efts, and both factors may be favorable in Eastern Hemlock-dominated stands (Healy 1975). In addition, Eastern Hemlock-dominated stands have an abundance of mushrooms, and Red Efts are often observed feeding on invertebrates found around decaying mushrooms (MacNamara 1977). Finally, anecdotal observations of Red Efts in Eastern Hemlock-dominated stands at Harvard Forest suggests that the abundance of this species is greater in Eastern Hemlock stands than in mixed deciduous stands (B.G. Mathewson, pers. observ.).

If the relative abundance of Red Efts is higher in Eastern Hemlock-dominated stands than in mixed deciduous stands, a transition to mixed deciduous stands due to HWA could lead to a reduction in the relative abundance of Red Efts at these sites. Less desirable terrestrial habitat may also impact aquatic communities. Only 1–2% of Eastern Red-spotted Newts survive the larval stage to become Red Efts (Petranka 1998). Therefore, a change in survivorship of the juvenile phase could have an important impact on population densities of the Eastern Red-spotted Newt.

Salamanders inhabiting the forest floor are ecologically important due to their significant contribution to the overall biomass of vertebrates in the forest, and their position in the middle of the food web (Burton and Likens 1975a, Welsh and Droege 2001). At Hubbard Brook Experimental Forest in Coos Country, NH, salamander biomass equaled small-mammal biomass, and was twice the biomass of breeding birds (Burton and Likens 1975a). Red-backed Salamanders accounted for 93.5% of salamander biomass with a density of 0.25 individuals/m² at Hubbard Brook, while Red Efts were rare due to the lack of suitable aquatic breeding habitat within 1 km of study sites (Burton and Likens 1975a).

While no research has been conducted on the role of Red Efts in nutrient cycling or decomposition rates, it has been hypothesized that predation by Red-backed Salamanders on soil invertebrates that break down leaf litter reduces the rate of soil decomposition by decreasing the amount of surface area available to bacteria and fungi (Wyman 1998). Slowing down decomposition of organic matter on the forest floor slows down the rates of CO_2 emitted into the atmosphere (Wyman 1998). Thus, reducing predation on invertebrate decomposers may have major implications on the global carbon budget as it is estimated that six times as much CO_2 is emitted into the atmosphere by the breakdown of leaf litter as by any anthropogenic source (Wyman 1998). As predators of soil invertebrates, it is also possible that Red Efts have an impact on decomposition rates. Red Efts prey on a great diversity of invertebrates, including representatives from 25 orders and 58 families (MacNamara 1977). MacNamara (1977) and Burton (1976) reported that Red Efts' preferred prey (by percentage of overall diet by weight) are land snails (23.8% and 59.7%), mites and ticks (13.8% and 3.4%), springtails (10.6% and 9.1%), dipteran adults (9.7% and 8.8%), and lepidopteran larvae (7.9% and 2.3%).

Salamanders of the forest floor are also important as prey to larger vertebrates such as snakes, birds, and small mammals (Welsh and Droege 2001). Due to low metabolic rates, salamanders are extremely efficient at converting prey into protein, which is then passed up the food chain (Burton and Likens 1975b). However, Red Efts may not be as important prey to larger vertebrates as other salamanders because of toxins in their skin (Brodie 1968, Hurlbert 1970, Uhler 1939). Many potential diurnal predators including *Charadrius vociferus* L. (Killdeer), *Buteo jamaicensis* Gmelin (Red-tailed Hawk), *Falco sparverius* L. (American Kestrel), and *Thamnophis sirtalis* L. (Common Garter Snake) find Red Efts to be unpalatable (Hurlbert 1970, Uhler 1939). Other predators such as *Rana catesbeiana* Shaw (American Bullfrog), *Procyon lotor* L. (Raccoon), and *Bufo americanus* Holbrook (American Toad) appear to be less sensitive to the toxins in Red Efts' skin (Brodie 1968a, Hurlbert 1970).

I hypothesized that the relative abundance of Red Efts would be higher in Eastern Hemlock-dominated stands than in mixed deciduous stands based on preliminary field observations as well as the presence of biotic and abiotic habitat features preferred by Red Efts (Benzinger 1994, Hartmann 1977, Healy 1975, Lustenhouwer et al. 2012, Rogers 1980). In addition to testing this hypothesis, a secondary goal of this study was to look for relationships between the relative abundance of Red Efts and the average daily minimum and maximum temperatures in the spring and fall, soil pH, and estimated distances to potential breeding habitat. The third goal of this study was to establish baseline data on the relative abundance of Red Efts at Harvard Forest.

Field Site Description

This study was conducted in 10 second-growth stands at Harvard Forest in Petersham, MA (42.533°N, 72.190°W; 338 m elev.). I chose 1 mixed deciduous stand and 1 Eastern Hemlock-dominated stand at the Prospect Hill, Tom Swamp, and Slab City tracts, and 2 mixed deciduous and 2 Eastern Hemlock-dominated stands on the Simes tract. One of the stands, the mixed deciduous stand in the Tom Swamp tract (hereafter referred to as TS-MD), was selectively logged in 1998. The average distance from potential breeding habitats to the center of stands, estimated using maps in the lab, was 545 m in Eastern Hemlock-dominated stands and 430 m in mixed deciduous stands (Table 1). When this study was conducted, HWA was not known to be present in any of the stands studied.

I used tree species composition data from the Harvard Forest Archives to select stands and then qualitatively verified stand type in the field (Foster 1992). Eastern Hemlock contributed 63% of the total basal area in the Eastern Hemlock-dominated stand at Simes 1, and 60% in Simes 2 (Ellison et al. 2010). The dominant

overstory tree species in the mixed deciduous stand at Simes 1 were Red Oak (36%), Black Birch (24%), Red Maple (13%), and *Acer saccharum* Marsh. (Sugar Maple) (11%) (Ellison et al. 2010). In the mixed deciduous stand at Simes 2, the dominant overstory tree species were *Pinus strobus* L. (Eastern White Pine) (35%), Red Maple (17%), Black Birch (15%), and Red Oak (15%) (Ellison et al. 2010). Quantitative data for overstory tree species composition data was not available in the Prospect Hill, Slab City, or Tom Swamp sites. I qualitatively assessed Eastern Hemlock-dominated stands at these sites to be greater than 50% Eastern Hemlock. The primary species in mixed deciduous stands at these sites were Red Oak, Black Birch, Eastern White Pine, and Red Maple (Table 1).

Table 1. Description and measurements of environmental variables in 10 forest stands at 5 sites at Harvard Forest in Petersham, MA. Site codes are as follows: PH = Prospect Hill, S1 = Simes 1, S2 = Simes 2, SC = Slab City, TS = Tom Swamp. FT indicates forest type (EH = Eastern Hemlock-dominated; MD = mixed deciduous). Tree species codes are as follows: TSCA = *Tsuga canadensis* (Eastern Hemlock), PIST = *Pinus strobus* (Eastern White Pine), QUVE = *Quercus velutina* Lam. (Black Oak), QURU = *Quercus rubra* (Northern Red Oak), QUAL = *Quercus alba* (White Oak), BEPO = *Betula populifolia* Marshall (Gray Birch), ACRU = *Acer rubrum* (Red Maple), BELE = *Betula lenta* (Black Birch). Area = area covered by transects; dist = estimated distance to potential breeding habitat; low temp = average daily low temperature; high temp = average daily high temperature. Spring = 22 April 2004–7 June 2004; Fall = 22 September 2004–12 November 2004. SD = standard deviation.

								Spring		Fall	
				Stand				Low	High	Low	High
		Tree	Latittude	size	Area	Dist	Soil	temp	temp	temp	temp
Site	FT	species comp	longitude	(ha)	(m^2)	(m)	pН	(°C)	(°C)	(°C)	(°C)
PH	MD	QUVE-QURU	42°32.441'	1.0	180	700	4.2	7.4	22.9	7.6	13.1
		-BEPO	72°10.819'								
PH	EH	TSCA-PIST	42°32.372'	1.0	180	500	4.1	7.2	18.9	5.4	11.4
			72°10.750'								
S 1	MD	BELE-QURU	42°27.956'	1.0	180	500	4.4	7.6	25.6	5.2	13.6
		-ACRU	72°13.075'								
S1	EH	TSCA-QURU	42°28.313'	3.0	540	50	4.0	7.2	21.0	4.9	11.9
			72°13.025'								
S2	MD	PIST-BELE	42°28.758'	1.0	180	500	4.5	7.6	20.4	5.6	13.1
		-QURU	72°12.688'								
S2	EH	TSCA-BELE	42°28.511'	3.0	540	500	4.2	7.6	18.4	5.6	11.7
			72°12.782'								
SC	MD	QURU-ACRU	42°27.076'	0.4	185	1000	4.3	7.2	24.1	4.8	11.4
		-BELE-TSCA	72°10.098'							_	
SC	EH	TSCA-QURU	42°27.192'	0.5	248	850	4.1	7.4	18.7	5.2	11.4
TO		-PIST-ACRU	72°10.197'	1.0	212	0.5		- (24.0		10.0
18	MD	QUAL-QURU	42°30.232'	1.0	312	25	4.4	7.6	24.8	5.7	12.8
TO		- ACKU	/2°12.683	1.0	240	250	1.0	7 (10.5	C 1	11.0
18	ΕH	I SCA-PIS I	42°30.400'	1.0	248	250	4.0	/.6	18.5	5.1	11.2
MD	A	-ACKU	/2°12.886	0.0	207	E 1 E	4.4	75	22 (5 0	12.0
MD AVg		(0.9)	207	243 (255)	4.4	(0, 2)	(2.0)	3.8	12.8		
EILA	50			(0.3)	(39)	(355)	(0.1)	(0.2)	(2.0)	(1.1)	(0.8)
EH AVg			(1.2)	331	430	4.1	(0, 2)	19.1	3.2	(0.2)	
2	U U			(1.2)	(1/3)	(301)	(0.1)	(0.2)	(1.1)	(0.3)	(0.3)

Methods

Red Eft sampling

My first method of assessing the relative abundance of Red Efts was daytime visual surveys of the surface of the forest floor (hereafter referred to as transect surveys). I conducted transect surveys 13–16 times in each stand on a total of 51 sampling days from 14 August 2003–29 October 2004 by walking transects and counting all Red Efts that were visible on the surface of the forest floor entirely within 0.5 m to the left and right of the transect. Natural cover objects such as rocks and stones were not turned over during these searches. Transects varied in length from 76–108 m because I established lengths so as to not extend the transects beyond the edge of the stand, and the distance to the edge of the stand was not constant. Each transect origin was chosen randomly. The area sampled in each stand was between 180–540 m², depending on the size of the stand (Table 1). I also randomly chose the order in which I sampled the stands; not all sites were sampled on the same day.

The second method of sampling for Red Efts was 2-minute time-constrained searches of natural cover objects (NCO) such as coarse woody debris, stones, and leaves in 1-m² quadrats on the surface of the forest floor (hereafter referred to as quadrat surveys). In each stand, I searched 20 quadrats during fall 2003 and 20 quadrats during spring 2004. I placed quadrats at randomly selected points along the same transects used in transect surveys, used a random number generator to determine sampling points along transects, and flipped a coin to determine whether to place the quadrat to the left or right of the transect. The order in which stands were sampled was random, and the same quadrat was never searched twice. Following searches, I returned all NCOs to their original position. I sampled both forest types on the same day at all sites except at Simes 2 when I sampled the two stands on consecutive days during the fall.

Measurements of habitat variables

I measured average daily high and low temperatures for each stand in the spring (22 April 2004–7 June 2004) and fall (22 September 2004–12 November 2004) using remote temperature sensors (1-Wire Thermochron iButtons \pm 1 °C, Maxim Integrated, San Jose, CA) that I placed on the soil surface in the center of each transect. These sensors recorded temperature every half hour in spring 2004 and every hour in fall 2004. To determine soil pH, I took 10–30 random samples from the organic layer of the soil just below the leaf litter in each stand, and used a Thermo Orion model 290 pH meter (\pm 0.005) to measure the pH of a slurry of 2.0 g of soil from each sample in 20 ml deionized water (Hendershot et al. 1993).

Measurements of precipitation, relative humidity, and hourly temperature from the Fisher Meteorological Station on the Prospect Hill Tract at Harvard Forest were used to report the weather conditions on all sampling days for both transect and quadrat methods. Weather conditions during transect surveys were reported for sites as opposed to individual stands because transect surveys of both forest types took only a few hours to complete and were completed in succession. Weather conditions during quadrat surveys were reported for individual stands as these surveys took more time and stands within a given site were often conducted at different times of the day, or in one case on different days. On several occasions the exact time of transect surveys was not recorded, and on one occasion exact time of quadrat surveys was not recorded. In these instances, weather conditions at noon on sampling day were used since this was the most frequent time searches were conducted.

Statistical analysis

All observations of Red Efts during transect and quadrat surveys were pooled to calculate the average relative abundance for each stand expressed as salamanders/m². In addition, I calculated the average relative abundance of Red Efts for surveys that were conducted when a rain event had and had not occurred in the prior 24 hours. I then used *t*-tests to test for differences in the average relative abundance of Red Efts in the two forest types using the two methods of sampling for all samples and for samples conducted within 24 hours of a rain event. All of these tests were run with and without TS-MD. Analyses without TS-MD were run because previous studies have found that selective harvesting can reduce the relative abundance of Plethodontids, and the same may be true for Red Efts (Harpole and Haas 1999). Further, piles of slash and dense stands of young vegetation may have reduced the probability of detection in this stand. I also conducted *t*-tests to evaluate differences in the estimated distance to potential breeding habitat, soil pH, and average daily high and low temperatures for each stand in the spring (22 April 2004–7 June 2004) and fall (22 September 2004–12 November 2004) in the two forest types. I used standard least squares regression analyses to test for individual relationships between each of the above variables and the relative abundance of Red Efts derived from the average of all transect surveys in a stand, both with and without TS-MD. All statistical tests were run using the statistical software program JMP IN version 5.1 (SAS Institute).

Results

The average relative abundance of Red Efts derived from transect surveys (n = 368 observations) was higher in Eastern Hemlock-dominated stands than in mixed deciduous stands, but the difference was not statistically significant (with TS-MD: 0.020 individuals/m² vs. 0.009 individuals/m², P = 0.146; without TS-MD: 0.020 individuals/m² vs. 0.011 individuals/m², P = 0.230). The same was true for the average relative abundance of Red Efts derived from quadrat surveys (n = 27 observations) (with TS-MD: 0.115 individuals/m² vs. 0.020 individuals/m², P = 0.234). The average relative abundance of Red Efts derived from transect surveys conducted within 24 hrs of a rain event (n = 307 observations) was also higher in Eastern Hemlock-dominated stands (with TS-MD: 0.036 individuals/m² vs. 0.019 individuals/m², P = 0.209), as was the average relative abundance of Red Efts derived from from transect from quadrat surveys (n = 20.209).

within 24 hrs of a rain event (n = 16 observations) (with TS-MD: 0.146 individuals/m² vs. 0.025 individuals/m², P = 0.153; without TS-MD: 0.146 individuals/m² vs. 0.033 individuals/m², P = 0.175) (Table 2), though, again, the differences were not statistically significant.

Soil pH was significantly lower in Eastern Hemlock-dominated stands than in mixed deciduous stands (4.1 vs. 4.4; P < 0.01) as was the average high temperature in the spring (19.1 °C vs. 23.6 °C; P < 0.01) and the average high temperature in the fall (11.5 °C vs. 12.8 °C; P < 0.05) (Table 1). The difference in estimated distance to potential breeding habitat in Eastern Hemlock-dominated stands versus mixed deciduous stands was not significant (430 m vs. 545 m; P = 0.600) (Table 1). Neither was the difference in average low temperature in spring (7.4 °C vs. 7.5 °C; P = 0.524) (Table 1) and the average low temperature in fall (5.2 °C vs. 5.8 °C; P = 0.332) (Table 1).

Regression analyses did not reveal a statistically significant relationship between any of the variables and the average relative abundance of Red Efts derived from transect surveys. However, when removing TS-MD, a statistically significant relationship was found between distance to potential breeding habitat and the average relative abundance of Red Efts derived from transect surveys (n = 9, r^2 adj = 0.76, P < 0.01) (Fig. 1). When TS-MD was included the results were not significant (n = 10, r^2 adj = 0.20, P < 0.19).

Table 2. Measurements of the average relative abundance (given in individuals/ m^2) of Red Efts in10 forest stands at Harvard Forest. Transect surveys of the forest floor surface conducted from fall 2003 to fall 2004 (excluding winter). Quadrat surveys of 1-m² quadrats conducted in fall 2003 and spring 2004. Site codes are as follows PH = Prospect Hill, S1 = Simes 1, S2 = Simes 2, SC = Slab City, TS = Tom Swamp. FT indicates forest type (EH = Eastern Hemlock dominated; MD = mixed deciduous). NA indicates that no sampling was conducted under these conditions.

		Average relative abundance of Red Ens							
			Transect surv	eys	Quadrat surveys				
Site FT		All	Within 24 hrs of rain event	Without rain event in prior 24hrs	All	Within 24 hrs of rain event	Without rain event in prior 24hrs		
PH	MD	0.012	0.017	0.002	0.050	0.050	NA		
PH	EH	0.020	0.029	0.000	0.050	0.050	NA		
S1	MD	0.016	0.033	0.001	0.025	0.050	0.000		
S1	EH	0.031	0.061	0.007	0.350	0.200	0.500		
S2	MD	0.015	0.021	0.006	0.025	NA	0.025		
S2	EH	0.028	0.041	0.000	0.150	0.300	0.000		
SC	MD	0.002	0.006	0.000	0.000	0.000	0.000		
SC	EH	0.000	0.000	0.000	0.000	NA	0.000		
TS	MD	0.001	0.001	0.001	0.000	0.000	0.000		
TS	EH	0.019	0.048	0.009	0.025	0.033	0.000		
MD Avg.		0.009	0.016	0.002	0.020	0.025	0.006		
5	SD	(0.007)	(0.013)	(0.002)	(0.021)	(0.029)	(0.013)		
EH Avg.		0.020	0.036	0.003	0.115	0.146	0.125		
SD		(0.012)	(0.023)	(0.004)	(0.143)	(0.127)	(0.250)		

Average relative abundance of Red Efts

Discussion

While these results do not confirm the hypothesis that the relative abundance of Red Efts is significantly greater in Eastern Hemlock-dominated stands than in mixed deciduous stands, they are suggestive of this hypothesis. Indeed, using data from transect surveys, 4 of 5 stands with the highest relative abundance of Red Efts were Eastern Hemlock-dominated stands. Further study with a larger sample size may yield statistically significant results. The extent of the differences in the relative abundance of Red Efts in the two forest types may be large given that transect surveys yielded estimates in Hemlock-dominated stands that were more than two times greater and quadrat surveys yielded estimates which were almost six times greater than those for mixed deciduous stands. If the relative abundance of Red Efts is greater in Eastern Hemlock-dominated forests, a shift from this forest type to the mixed deciduous forest type due to HWA would likely negatively impact populations of Eastern Red-spotted Newt at Harvard Forest.

The Red Eft, of course, is just one phase in the life cycle of the Eastern Redspotted Newt. The loss of Eastern Hemlock along wetland borders may impact



Figure 1. Relationship between the average relative abundance of Red Efts as measured by transect surveys and the estimated distance to potential breeding habitat at Harvard Forest (42.533°N, 72.190°W; 338 m elev.)—including the Tom Swamp mixed deciduous stand (n = 10, r^2 adj = 0.20, P < 0.19)—excluding the Tom Swamp mixed deciduous stand (n = 9, r^2 adj = 0.76, P < 0.01).

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aquatic adult and larval phases as well. For example, a shift from Eastern Hemlock to mixed deciduous species along wetland borders could cause increases in solar radiation in the late winter and early spring before mixed deciduous trees

Table 3. Sampling effort and average weather conditions at the time of transect surveys. All weather data were measured at the Fisher Meteorological Station on the Prospect Hill Tract at Harvard Forest ($42.533^{\circ}N$, $72.190^{\circ}W$; 338 m elev.). Site codes are as follows PH = Prospect Hill, S1 = Simes 1, S2 = Simes 2, SC = Slab City, TS = Tom Swamp. RH = relative humidity. The first number in each cell is the average for all sampling dates. The second number in each cell is the average for all sampling dates conducted within 24 hours of a rain event. The third number in each cell is the average for all sampling dates conducted when a rain event had not occurred within the previous 24 hours. SD = standard deviation.

0:4-		Turne (9C)		Avg total precipitation	Percentage of days sampled within 24 hrs
Site	n	Temp (°C)	KH (%)	prior 24 nrs (mm)	of precipitation
PH	13 (9, 4)	15.0 (15.1, 14.7)	76 (89, 50)	8.3 (11.9, 0.0)	71 (100, 0)
S1	18 (11, 9)	17.5 (15.3, 20.2)	64 (72, 56)	2.8 (5.0, 0.0)	55 (100, 0)
S2	17 (10, 7)	16.5 (15.6, 20.6)	66 (77, 52)	6.0 (10.3, 0.0)	59 (100, 0)
SC	18 (8, 10)	17.4 (17.3, 18.4)	63 (84, 51)	2.3 (5.8, 0.0)	43 (100, 0)
TS	18 (7, 11)	17.6 (14.2, 19.8)	65 (77, 58)	4.9 (12.7, 0.0)	39 (100, 0)
Avg	17 (9, 8)	16.8 (15.5, 18.7)	67 (80, 53)	4.9 (9.1, 0.0)	53 (100, 0)
SD	2 (2, 3)	1.1 (1.1, 2.4)	5 (7, 3)	2.4 (3.5, 0.0)	13 (0, 0)

Table 4. Average weather conditions at the time of quadrat surveys in 10 forest stands at Harvard Forest. All weather data were measured at the Fisher Meteorological Station on the Prospect Hill Tract at Harvard Forest ($42.533^{\circ}N$, $72.190^{\circ}W$; 338 m elev.). Site codes are as follows PH = Prospect Hill, S1 = Simes 1, S2 = Simes 2, SC = Slab City, TS = Tom Swamp. FT indicates forest type (EH = Eastern Hemlock-dominated; MD = mixed deciduous). The first number in each cell is the average for all sampling dates. The second number in each cell is the average for all sampling dates conducted within 24 hours of a rain event. The third number in each cell is the average for all sampling dates conducted within the previous 24 hours. SD = standard deviation.

Site	FT	Number of quadrats surveyed	Temperature (°C)	Relative humidity (%)	Average total precipitation prior 24 hrs (mm)
PH	MD	40 (40, 0)	20.1 (20.1, na)	60 (60, na)	10.3 (10.3, na)
PH	EH	40 (40, 0)	19.8 (19.8, na)	63 (63, na)	10.3 (10.3, na)
SI1	MD	40 (20, 20)	19.8 (14.9, 24.8)	60 (74, 47)	9.3 (18.5, 0.0)
SI1	EH	40 (20, 20)	19.2 (14.4, 24.0)	60 (71, 49)	5.3 (10.5, 0.0)
SI2	MD	40 (0, 40)	19.7 (na, 19.7)	60 (na, 60)	0.0 (na, 0.0)
SI2	EH	40 (20, 20)	18.9 (18.9, 18.8)	65 (96, 34)	7.2 (14.3, 0.0)
SC	MD	40 (20, 20)	13.5 (16.0, 11.0)	47 (39, 55)	0.2 (0.3, 0.0)
SC	EH	40 (0,40)	15.0 (na, 15.0)	43 (na, 43)	0.0 (na, 0.0)
TS	MD	40 (30, 10)	10.4 (7.9, 16.7)	56 (61, 45)	1.9 (2.5, 0.0)
TS	EH	40 (30, 10)	12.3 (10.9, 16.7)	45 (45, 45)	1.9 (2.5, 0.0)
MD avg $(n = 5)$	MD	40 (22, 18)	16.7 (14.7, 18.1)	57 (59, 52)	4.3 (7.9, 0.0)
SD	MD	0 (15, 15)	4.5 (5.1, 5.8)	6 (14, 7)	5.1 (8.3, 0.0)
EH avg $(n = 5)$	EH	40 (22, 18)	17.0 (16.0, 18.6)	55 (69, 43)	4.9 (9.4, 0.0)
SD	EH	0 (15, 15)	3.3 (4.1, 3.9)	10 (21, 6)	4.1 (5.0, 0.0)

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leaf out. This increased radiation could result in higher water temperatures and a reduction in size or even a complete disappearance of these wetlands. A comparison of populations of Eastern Red-spotted Newts in aquatic habitat within Eastern Hemlock-dominated and mixed deciduous forests would provide a more detailed understanding of how the relative abundance of this species may change with the loss of Eastern Hemlock.

Results from this study suggest that distance to breeding habitat may be the most important factor in driving differences in the relative abundance of Red Efts. Caution should be exercised when interpreting these findings, however, because the distances to breeding habitat were estimations and confirmation of actual breeding populations of Eastern Red-spotted Newts were not made. Additional research investigating the relationship between the relative abundance of Red Efts and distance to breeding habitat is warranted.

Data from this study, collected prior to the arrival of HWA at Harvard Forest, can be used in before-after analyses to directly monitor potential changes in the relative abundance of Red Efts in Eastern Hemlock-dominated stands throughout their decline and transformation into mixed deciduous stands. HWA was first discovered at Harvard Forest in 2006 near the Eastern Hemlock-dominated stand at Simes 1 (Ellison et al. 2010). As of 2009, it was present in 44% of the Eastern Hemlock trees in the two Eastern Hemlock-dominated stands at Simes 1 and Simes 2 (Ellison et al. 2010). Therefore, it makes sense to repeat sampling of Red Efts as soon as possible.

A long-term study of the relative abundance of Red Efts would be an important contribution to our understanding of populations of Eastern Red-spotted Newts because no similar study has ever been conducted. As a long-term ecological research (LTER) site, Harvard Forest is a perfect place for future studies to build on the baseline data presented here. The most efficient use of sampling time would be to conduct sampling within 24 hours of a rain event, if possible (Table 2). When comparing future data with data from this study, it is important to take into account average temperature and relative humidity at time of sampling along with total precipitation prior to sampling (Tables 3, 4).

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