7-17-2011

Survival, Breeding Frequency, and Migratory Orientation in the Jefferson Salamander, *Ambystoma Jeffersonianum*

Stephen P. De Lisle

Kristine L. Grayson
University of Richmond, kgrayson@richmond.edu

Recommended Citation

SURVIVAL, BREEDING FREQUENCY, AND MIGRATORY ORIENTATION IN THE JEFFERSON SALAMANDER, *AMBYSTOMA JEFFERSONIANUM*

STEPHEN P. DE LISLE[1,2] AND KRISTINE L. GRAYSON

Department of Biology, University of Virginia, Charlottesville, Virginia 22904, USA
[1]Corresponding author; e-mail: spd2f@virginia.edu
[2]Present address: Department of Ecology and Evolutionary Biology, University of Toronto, Toronto, Ontario Canada M5S 3B2

Abstract.—Accurate estimates of demographic parameters, such as survival and breeding frequency, are necessary for the conservation and management of animal populations. Additionally, life-history data are required for gaining an empirical understanding of the ecology of natural populations. We monitored a population of Jefferson Salamanders (*Ambystoma jeffersonianum*) breeding in a permanent mountain-top pond at the southern limit of this species' geographic range in Virginia over four years. We used closed multistate mark-recapture models with Pollock's robust design to estimate the demographic parameters of this population. Additionally, we used point-of-capture data to compare the orientation of migrations into and out of the pond within and among years. Our model selection results support consistent annual adult survival across years with higher estimates for males compared to females. Our estimates of the probability of breeding in sequential years were high for both sexes during the four years of our study. Our model rankings and capture probability estimates indicate that females had a higher probability of detection when entering the breeding pond, likely reflecting differences between the sexes in arrival time to the pond. We found directionality in some, but not all, annual migrations, despite indications of individual fidelity in orientation across years. Our study provides the first estimates of breeding probability and assessment of migratory orientation patterns for *A. jeffersonianum* and contributes to the understanding of the reproductive ecology and natural history of pond-breeding amphibians.

Key Words.—*Ambystoma jeffersonianum*; breeding probability; Jefferson Salamander; migratory orientation; multistate mark-recapture; survival

INTRODUCTION

Recent and dramatic global amphibian declines have increased the need for accurate knowledge of amphibian population demography and habitat use (Houlahan et al. 2000; Semlitsch 2000). For many pond-breeding amphibians, migration across distinct habitat boundaries makes these animals an important component of landscape biota and creates challenges for conservationists, land managers, and wildlife biologists (Semlitsch 2008). Salamanders of the family Ambystomatidae have been identified as having a significantly greater number of declining species than other amphibian families (Stuart et al. 2004). Accurate estimates of annual survival and breeding frequencies, as well as spatiotemporal movement patterns, are essential to inform management decisions and to provide biologists with the life-history knowledge necessary for understanding the population dynamics of these animals. Plasticity in reproductive behavior and complex life cycles common to many migratory pond-breeding amphibians adds to the difficulty associated with obtaining accurate demographic information (Wilbur 1980; Church et al. 2007).

Jefferson Salamanders (*Ambystoma jeffersonianum*) are a pond-breeding salamander ranging throughout the northeastern United States. *Ambystoma jeffersonianum*

tend to breed in permanent or ephemeral ponds in upland forests, usually arriving at a pond in late winter or early spring and migrating back to the terrestrial habitat after a short breeding period (Petranka 1998). For *A. jeffersonianum*, early arrival at breeding ponds across much of their range may have community wide effects and distinguishes them from sympatric Ambystomatid species that have been the focus of most recent demographic studies. To our knowledge, annual adult survival rates in a breeding population of Jefferson Salamanders have not been estimated since a study by Williams (1973). Additionally, due to anthropogenic habitat fragmentation, many amphibian populations now exist in isolation (reviewed by Cushman 2006). Estimates of adult survival probability are particularly important for isolated *A. jeffersonianum* populations (Mullin and Klueh 2009) and this life-history parameter is likely impacted by terrestrial habitat quality (Faccio 2003).

For iteroparous Ambystomatids, breeding frequency is a key factor in determining reproductive success in spatiotemporally variable pond habitats and can be limited by the costs of reproduction (e.g., Bull and Shine 1979). For pond-breeding amphibians, these costs can be byproducts of the requisite migration to and survival in a breeding pond or be directly related to the energetics of reproduction. Annual environmental variation

associated with often-ephemeral breeding ponds can influence breeding 'decisions' and result in individuals skipping breeding opportunities (Husting 1965), especially in females (Church et al. 2007; Gamble et al. 2009). Amphibian iteroparity may also be limited by the time required to recover energy costs associated with reproduction (e.g., Harris and Ludwig 2004). Variable reproductive patterns in amphibian populations have created challenges for conservationists attempting to interpret population fluctuations (Pechmann et al. 1991) as well as biologists attempting to identify breeding frequency in light of imperfect census data (the Husting dilemma, *sensu* Gill 1985). To our knowledge, no studies have estimated breeding frequency for *A. jeffersonianum*.

Finally, orientation of migrations to and from breeding ponds can provide an indication of adult terrestrial habitat use (Madison and Farrand 1998) as well as metamorph dispersal patterns. For pond-breeding amphibians, knowledge of migratory routes and upland habitat use is necessary for successful conservation efforts, which must focus on both aquatic and terrestrial habitat (Semlitsch 2000). Directional migrations (Dodd and Cade 1998; Malmgren 2002; Marty et al. 2005; Jenkins et al. 2006) as well as intra- and inter-annual fidelity of individual migratory orientation (Shoop 1965; Stenhouse 1985; Phillips and Sexton 1989) have been documented in several amphibian populations. Although terrestrial habitat quality is a key resource for *A. jeffersonianum* (Faccio 2003), no previous studies have examined migratory orientation patterns or fidelity in this species.

We used four years of mark-recapture data from a Jefferson Salamander population breeding in a permanent mountain-top pond at the southern limit of this species' range to estimate survival and breeding probability. Point of capture data was used to test for directionality in migratory orientation and to make comparisons of orientation across years. Conducting orientation analysis in conjunction with mark-recapture methods allowed us to track migratory orientation of marked individuals across years and measure the level of directional fidelity of individuals.

MATERIALS AND METHODS

Study site.—We monitored an *A. jeffersonianum* population breeding in Sylvatica Pond (area = 828 m², depth = 0.7 m) at Mountain Lake Biological Station (Giles County, Virginia, USA, 37º22'32"N, 80º31'20"W, elevation 1160 m). We studied the breeding migrations of adults from 2006–2009 and the emigration of juveniles from 2005–2008. Sylvatica Pond is a permanent, fishless, rain-fed pond constructed in the late 1960s and provides breeding habitat for a number of amphibian species (e.g., *Hemidactylium*

scutatum, Notophthalmus viridescens, Pseudacris crucifer, Rana sylvatica). We also monitored a second permanent pond, Horton (area = 630 m², depth = 1.1 m), located 100 m from Sylvatica Pond, during this period using the same methods.

Data collection.—We constructed a continuous drift fence of aluminum flashing in May 2005 around both ponds to monitor inbound and outbound amphibian movement. We buried pitfall traps (19 L plastic buckets) flush with the surface and spaced equally around the circumference of the pond (14 pairs at Sylvatica and 12 pairs at Horton). We opened and checked traps daily from 15 June – 10 October 2005, 15 March – 29 September 2006, 1 March – 2 October 2007, 4 March – 1 October 2008, and 8 March – 7 May 2009. During the late fall and winter (October-February), we placed lids on the pitfall traps and we installed and opened a sliding door in the fence between each pitfall trap pair to allow winter movements of all species. The target species for this drift fence was *Notophthalmus viridescens* (Grayson 2010) and we expected that some early arrivals of adult *A. jeffersonianum* entered the pond undetected before the trap opening date in early March. We structured our mark-recapture models to accommodate drift fence trespass and the possibility of differences between entry and exit capture probabilities (see *Mark-recapture analysis*).

Our mark-recapture models assume all individuals exited the pond at the end of the breeding period. The drift fence around each pond was within 2 m of the pond edge, before the start of the surrounding forest habitat, and it is unlikely that adults overwintered terrestrially inside the fence. Sylvatica and Horton Pond were also sampled extensively every fall by seine as part of a newt mark-recapture study (Grayson et al. 2011). Because we did not capture any *A. jeffersonianum* in the pond during these surveys, we conclude that none overwintered in the pond.

We weighed captured salamanders and determined their sex using the swollen appearance of the cloaca to distinguish males. Upon first capture, we marked all individuals on the ventral side with a unique color combination using visible implant elastomer (VIE, Northwest Marine Technology, Inc., Shaw Island, Washington, USA; Davis and Ovaska 2001; Bailey 2004). We returned all salamanders the same day to the opposite side of the fence at their point of capture. Capture and recapture records were used to construct a capture history for each individual.

Mark-recapture analysis.—We used multistate mark-recapture (MSMR) models to estimate annual survival and transitions between breeding and nonbreeding states while accounting for unequal capture probabilities. We applied Pollock's robust design to our sampling (Pollock

1982), where each primary sampling period (*i*) constituted a year with two secondary samples (*j*), entering (*j* = 1) and leaving the pond (*j* = 2). Traditionally, data from secondary samples within a primary period are analyzed using closed population models and intervals between primary periods are considered open to population additions and removals (Kendall and Nichols 1995). This model assumes demographic closure between secondary samples (i.e., while breeding individuals are in the pond) and allows for a single annual survival estimate. This model also accommodates imperfect and variable detection probabilities (i.e., fence trespass).

Advances in multistate modeling have provided methods to treat a population as 'open' between secondary sampling periods in order to estimate survival within a breeding pond separately from survival in the forest during the nonbreeding season (the gateway robust design; Bailey et al. 2004). While we acknowledge that our study population was likely in some violation of the demographic closure assumption during the interval between secondary samples, we proceeded with closed multistate models for two reasons. First, studying such a small population (total captures = 81 individuals) with an unobservable state limits the number of parameters that can be uniquely and precisely estimated (Bailey et al. 2010). The additional complexity associated with gateway robust design multistate models can greatly reduce their utility when applied to small amphibian populations (see Gamble et al. 2009; Muths et al. 2010). Second, given the short time period that breeding individuals spent in the pond (mean = 10 d ± 0.77 d SE) in our population and the short breeding period of this species across its range (Douglas 1979; Petranka 1998), we had no reason to expect a high levels of mortality during the breeding period. McCaffery and Maxwell (2010) provide a recent example of the same multistate closed robust design models applied to amphibians.

Four types of parameters were present in our models: (1) apparent survival probability, S_i^k the probability that a marked animal in state *k* survives between primary period *i* and *i*+1; (2) transition probability, ψ_i^{kj}, the probability that an animal in state *k* at primary period, *i*, is in state *j* at primary period *i*+1, given that it survives; (3) capture probability, p_{ij}^k, is the probability that an animal alive in state *k* at secondary sampling occasion *j* of primary period *i* is first captured; and (4) recapture probability, c_{i2}^k, the probability that an animal alive in state *k* is recaptured leaving the pond (*j* = 2) during primary period *i*. We sampled adult salamanders starting in spring 2006 (*i* = 1) until spring 2009 (*i* = 4). Our model included two states: breeders (B), individuals captured at the drift fence during spring breeding migrations, and unobservable non-breeders (U), individuals alive in the terrestrial habitat but skipping a breeding year (Husting 1965). Our designation of

migrants as 'breeders' refers to the attempt of an individual to reproduce based on its entrance into a pond, and does not make assumptions about the success of the breeding attempt.

We only captured seven individuals (six males and one female) at the Horton Pond drift fence; too small a number to include another state to separately estimate parameters for this pond. Excluding these data from the analysis would knowingly bias estimates of survival and breeding frequency, as we know that these individuals were alive and making a migration to breeding grounds. The only biologically reasonable and statistically feasible solution was to include the Horton Pond captures with the Sylvatica Pond captures in one mark-recapture analysis. We also used data from the 7 Horton Pond individuals in our body mass analyses, but not in our orientation analyses (see *Orientation analysis*).

A global model with full time-specificity in all parameters is statistically unidentifiable because it contains parameters that cannot be uniquely estimated (parameter redundancy; Gimenez et al. 2004). We applied constraints to our global model and candidate model set based on the biology of *A. jeffersonianum*. Nonbreeders were unobservable because individuals skipping breeding years were not sampled at the drift fence; hence, we set capture and recapture probability to zero for individuals in this state at all times. A necessary assumption in models with unobservable states is that survival probability for the unobservable state must be assumed equal to an observable state (Kendall 2004; Bailey et al. 2009). Therefore, we assumed $S_i^B = S_i^U$ for all primary periods. The infeasibility of sampling nonbreeding fossorial salamanders makes such assumptions inherent in MSMR studies of Ambystomatids (Church et al. 2007; Gamble et al. 2009). Additionally, in the robust design, the final capture probability is confounded with recapture probability (Armstrup et al. 2005). Thus, in our sampling design, p_{i2} and c_{i2} are not individually identifiable. Solutions include assuming capture probability is equal within a primary period ($p_{i1} = p_{i2}$) or assuming capture probability is equal to recapture probability ($p_{i2} = c_{i2}$). Given our knowledge that capture probability entering the pond was likely lower than capture probability exiting the pond due to the date the traps were opened, we chose the latter assumption. We have no reason to expect that detection in pitfall traps along the drift fence was impacted by previous capture. Survival and capture probability are also confounded in the final sampling period, as in all open live mark-recapture models (Lebreton et al. 1992; Kendall and Nichols 2002). To allow for full time-specify in survival, we assumed $p_3 = p_4$ for both secondary samples.

We also limited our modeling of transition probabilities (between breeders and nonbreeders) based

on the scope of our data set. With four years of data on breeders, we only had two opportunities to observe an individual skip a year of reproduction (ψ_i^{BU}) and then return to breed (ψ_i^{UB}). In other words, a salamander could breed in 2006 and breed again next in 2008 or breed in 2007 and breed again next in 2009. Given the low occurrence of these capture histories in our data set (see results), we modeled transition probability without variation between primary periods (*time*). Additionally, we only had one opportunity to observe individuals remaining in the unobservable state (ψ_i^{UU}) by skipping two years of breeding (i.e., breed in 2006, skip 2007 and 2008, breed again next in 2009). We observed no individuals in our study with this capture history and thus, $\psi_i^{UU} = 0$ and $\psi_i^{UB} = 1$.

Our global model, $S(sex, time)$, $\psi(sex)$, $p(sex, time)$, included variation between the sexes and over time (primary periods/years) in survival and capture probability and variation between the sexes in transition probability. The global model and all reduced models included variation in capture probability between secondary samples ($p_{i1} \neq p_{i2}$) based on the opening date of the pitfall traps (i.e., we expected $p_{i1} < p_{i2}$). We followed a sequential modeling process where we first used the global model structure for S and ψ and tested models of capture probability structure (Lebreton et al. 1992). We tested four capture probability models with all combinations for *sex* x *time*. Next, we constructed a candidate model set of reduced models for S and ψ using the capture probability structure from the top ranked model. Our candidate model set included all combinations of *sex* x *time* for survival probability and *sex* for transition probability, for a total of eight models.

Akaike's Information Criterion corrected for small sample size (AIC$_c$) was used to select among competing models and Akaike weights (w) were used to identify the relative weight of evidence for each model in the candidate model set (Burnham and Anderson 2002). We used program MARK (White and Burnham 1999) to rank our candidate models and obtain maximum likelihood estimates of model parameters. A formal goodness-of-fit test currently does not exist for most robust design models. Therefore, we restructured our global model as an open multistate model with survival and transition probabilities constrained within primary periods and used the median $\hat{c}$ approach in Program MARK to estimate overdispersion.

Orientation analysis.—We used the pitfall trap location recorded for each individual capture to examine *A. jeffersonianum* movements into and out of Sylvatica Pond. The 14 trap pairs were located at approximately even arc distance from each other around the roughly-circular pond, so we treated each capture location as a point on a circular scale of 360°/14. We used the Rayleigh test to determine if samples were randomly distributed or directional, although our power was reduced due to the grouping of data by pitfall trap location, as opposed to being on a true continuous scale (Batschelet 1981; Zar 1996). For multi-sample comparisons of orientation distributions, we used Pearson's chi-square tests. In cases where zero individuals were captured at a pitfall trap in the samples being compared, the zero observation cells were removed from the analysis. In all tests at least 80% of the expected values were greater than five without pooling cells. The data did not meet the assumptions of other parametric (i.e., Watson-Williams *F*-test) or non-parametric (i.e., Watson's U^2 test) multi-sample circular tests due to the grouping by trap (approximately 26 degrees) and multi-modality in some samples (Batschelet 1981; Zar 1996).

The order of our circular analyses and comparisons were structured based on *a priori* hypotheses and limited based on sample sizes. We first pooled all observations (entering and exiting across all years and both sexes) and tested for directionality. We then repeated this procedure with data separated into entering and exiting migrations, and used chi-squared to compare the two samples. We then further separated observations with significant directionality by year and tested each year of our study for directionality. For observations of exiting metamorphs, we conducted a multi-sample test with data separated by year. We did this despite of a lack of statistically significant directionality in the pooled metamorph data because a clear bi-modality (a violation of assumptions of the Rayleigh test) was present. Because Pearson's chi-squared tests for any difference between two or more samples (not just mean direction; Batschelet 1981), comparisons of groups without a significant mean direction were appropriate.

We performed three circular correlations to assess individual orientation fidelity both within and across years using data from: (1) individuals captured both entering and exiting in a breeding year; (2) individuals captured exiting and then entering the pond in a sequential year; and (3) individuals captured exiting the pond in sequential years. The first correlation tested if individuals exited the pond in the same direction as their entrance within a year. The second tested if individuals entered the pond in a similar direction as their exit the year before and the third tested if individuals were consistent in their exit orientation between years. We determined the significance of the correlations using a jackknife procedure. We used Oriana 3.0 (Kovach Computing Services, Pentraeth, Wales, UK) for all circular statistical analysis and chi-squared tests.

We used a two-way ANOVA with year as a random effect and sex as a fixed effect to test for annual and sex-based differences in adult body mass for individuals entering Sylvatica Pond and Horton Pond (Proc GLM, SAS v. 9.2, Cary, North Carolina, USA). We used a

TABLE 1. Summary of *Ambystoma jeffersonianum* capture events at Sylvatica Pond from 2005–2009. The percentage of recaptures indicates the portion of individuals that were observed in a given year that had been observed in a previous year. Any adult captures at nearby Horton Pond that were included in the mark-recapture analysis are given; for example 7 (2H) males entering in 2008 means that seven males were included in the analysis, two of which were captured at Horton Pond.

		Males	Females	Metamorphs
2005	OUT	--	--	47
	Mean OUT Date	--	--	20 July
2006	IN Captures	8	8	--
	Mean IN Date	16 March	16 March	--
	OUT Captures	14	11 (1H)	15
	Mean OUT Date	21 March	25 March	20 July
	Number of Individuals	16	13 (1H)	--
	% Recaptures	--	--	--
2007	IN Captures	21 (2H)	10	--
	Mean IN Date	10 March	13 March	--
	OUT Captures	23 (2H)	7	8
	Mean OUT Date	19 March	20 March	17 July
	Number of Individuals	25 (3H)	11	--
	% Recaptures	40%	55%	--
2008	IN Captures	7 (2H)	5	--
	Mean IN Date	4 March	4 March	--
	OUT Captures	25 (2H)	6	23
	Mean OUT Date	9 March	13 March	14 July
	Number of Individuals	25 (2H)	6	--
	% Recaptures	36%	50%	--
2009	IN Captures	9	7	--
	Mean IN Date	17 March	17 March	--
	OUT Captures	25 (1H)	6	--
	Mean OUT Date	18 March	26 March	--
	Number of Individuals	25 (1H)	7	--
	% Recaptures	76%	14%	--
Total Individuals Captured		54 (6H)	27 (1H)	92

one-way ANOVA to test for annual differences in the body mass of metamorphs exiting Sylvatica pond from 2006–2008. Normality and homoscedasticity assumptions were met for both data sets without transformation. For all tests, $\alpha = 0.05$.

RESULTS

We observed 81 individuals (27 females, 54 males) over 180 capture events during the four years of our study (Table 1). Females that were captured and weighed both entering and exiting the pond in a year lost an average of 25.2% ± 1.9% SE of their incoming body mass while in the pond (n = 14, mean mass loss = 3.45 g ± 0.29 g SE). Males that were captured and weighed both entering and exiting the pond in a year lost an average of 8.5% ± 1.4% SE of their incoming body mass while in the pond (n = 20, mean mass loss = 0.94 g ± 0.18 g SE). Adult female body mass was significantly higher than that of males ($F_{1,3} = 53.8$, $P = 0.005$), while year had no significant effect ($F_{3,67} = 1.34$, $P = 0.27$) and

there was not a significant year*sex interaction ($F_{3,67} = 0.89$, $P = 0.45$). Metamorph mass was significantly different among years ($F_{2,43} = 25.1$, $P < 0.001$).

Model selection.—We found no evidence of overdispersion in our data based on median ĉ results (ĉ = 0.93) and therefore, made no adjustments in model selection criteria and variance estimates. Among our capture probability models, the model with full time and sex dependence in *p* carried 96% of the AIC_c weight ($AIC_c = 397.69$). All other capture probability structures had $\Delta AIC_c > 6.5$ and $w < 0.04$, indicating a majority of support for the top model. As a result, we used this capture probability structure in all of our candidate models.

Our highest ranked candidate model, *S(sex) ψ(·) p(sex, time)* supported sex-, but not time-dependence in survival and did not support sex dependence in transition probability. This model accounted for 53% of the AIC_c weight (Table 2). The second ranked model, *S(sex) ψ(sex) p(sex, time)* accounted for 16% of the AIC_c

TABLE 2. Model selection results from a multistate mark-recapture analysis of breeding *Ambystoma jeffersonianum*. The top capture probability (p) structure was used for all candidate models (see results). A period ($\cdot$) denotes consistency for a parameter (i.e., not meaningfully variable over time or between sexes). Models were ranked according to Akaike's Information Criterion corrected for small sample size (AIC_c). Also shown are the relative differences in AIC_c (ΔAIC_c) as well as AIC_c weight (w) and the number of parameters in the model (K).

	Model Structure					
S	ψ^{BB}, ψ^{BU}	p	AIC_c	ΔAIC_c	w	K
sex	$\cdot$	sex, time	391.11	0.00	0.533	15
sex	sex	sex, time	393.50	2.39	0.162	16
$\cdot$	$\cdot$	sex, time	394.24	3.13	0.112	14
sex, time	$\cdot$	sex, time	395.20	4.09	0.069	19
$\cdot$	sex	sex, time	395.89	4.78	0.049	15
time	$\cdot$	sex, time	396.39	5.27	0.038	16
sex, time	sex	sex, time	397.69	6.58	0.020	20
time	sex	sex, time	397.99	6.88	0.017	17

weight. The most reduced model, $S(\cdot)$ $\psi(\cdot)$ $p(sex, time)$ was ranked third and accounted for 11% of the AIC_c weight.

Parameter estimates.—Capture probability estimates varied widely by sex and primary and secondary sampling period (Table 3). For males, capture probability entering the pond was lower than exiting the pond across all years. This is consistent with the observed sex ratios of captures, which when pooled across years indicate a higher male-bias for exiting than for entering captures (entering = 1.37 males : 1 female, exiting = 2.76 males : 1 female). Capture probabilities for females were generally higher and without a consistent pattern.

We obtained survival and transition probability estimates using averages from the entire model set weighted by AIC_c support. Consistent with our model rankings, survival estimates varied little across time for both males and females, with males having higher survival probabilities than females (Fig. 1). Model-averaged estimates of transition probability from breeding to the unobservable nonbreeding state (ψ^{BU}) were similar for males (0.24 ± 0.09 SE) and females (0.25 ± 0.13 SE), consistent with transition probability being constant in our top ranked model. In our capture history records, only six males and one female returned to breed after skipping a year of reproduction during the four years of our study. No individuals were observed returning to breed after skipping two breeding years.

TABLE 3. Maximum liklihood estimates of *Ambystoma jeffersonianum* capture probabilities at the Sylvatica Pond drift fence from 2006–2009. Parameter $p_{i,j,}$ represents the probability of capture at secondary sampling period j within primary sampling period i. Our study of a pond-breeding amphibian had two secondary sampling periods: individuals migrating into the pond (IN; j = 1) and individuals exiting (OUT; j = 2) within each of four primary sampling periods (years). We constrained $p_{3,1} = p_{4,1}$ and $p_{3,2} = p_{4,2}$ to allow for full time-specificity in survival (see methods).

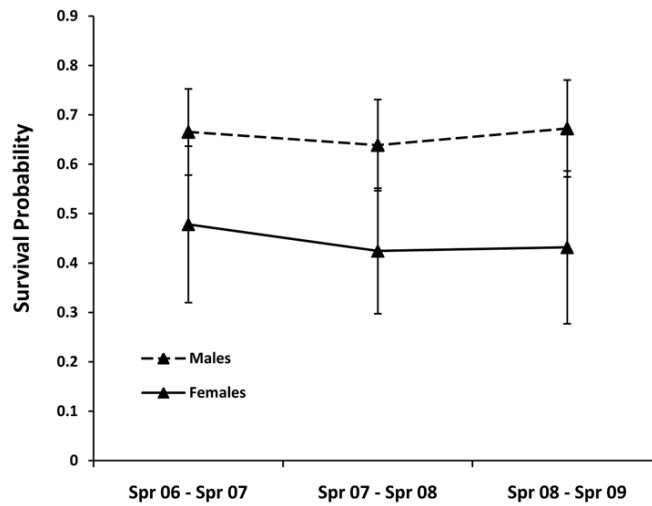| Parameter (p_{ij})	Sex	Year	Direction	Estimate	SE
$p_{1,1}$	M	2006	IN	0.43	0.13
$p_{1,2}$	M	2006	OUT	0.75	0.15
$p_{2,1}$	M	2007	IN	0.83	0.08
$p_{2,2}$	M	2007	OUT	0.87	0.07
$p_{3,1}/p_{4,1}$	M	2008/2009	IN	0.28	0.07
$p_{3,2}/p_{4,2}$	M	2008/2009	OUT	0.91	0.08
$p_{1,1}$	F	2006	IN	0.55	0.15
$p_{1,2}$	F	2006	OUT	0.75	0.15
$p_{2,1}$	F	2007	IN	0.87	0.12
$p_{2,2}$	F	2007	OUT	0.61	0.15
$p_{3,1}/p_{4,1}$	F	2008/2009	IN	0.92	0.08
$p_{3,2}/p_{4,2}$	F	2008/2009	OUT	0.92	0.08

FIGURE 1. Model-averaged annual survival probabilities (mean ± 1 SE) for male and female *Ambystoma jeffersonianum* breeding in Sylvatica Pond. Estimates represent annual survival between spring 2006 and spring 2007, spring 2007 and 2008, and spring 2008 and 2009. Data from seven individuals captured at nearby Horton Pond drift fence were included in these estimates (see methods).
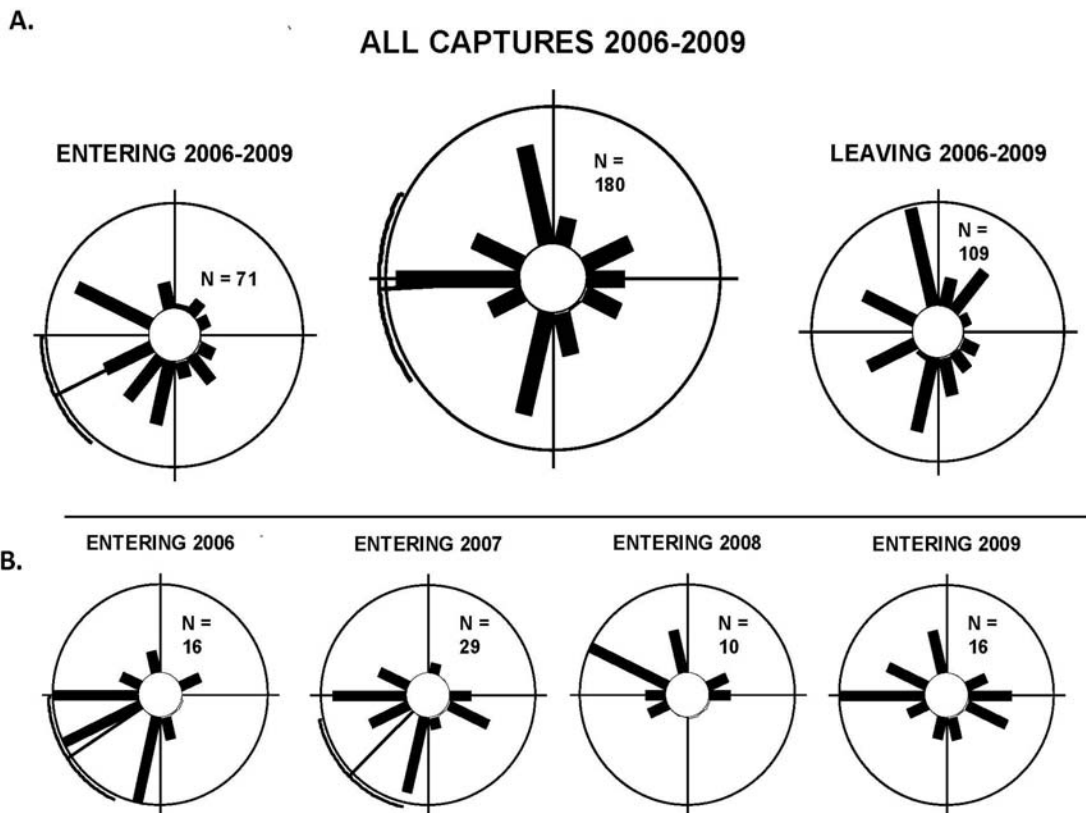

FIGURE 2. Capture location frequencies for adult *Ambystoma jeffersonianum* entering and leaving Sylvatica Pond from 2006–2009 (A) and entering for each year of the study (B). Circular axes indicate the location of pitfall traps around the pond from a fixed point. Radial axes show numbers of individuals captured at each pitfall trap. Sample sizes for each group are indicated. Mean direction ± 95% CI is indicated when there was significant directionality (Rayleigh's test, *P* < 0.05).
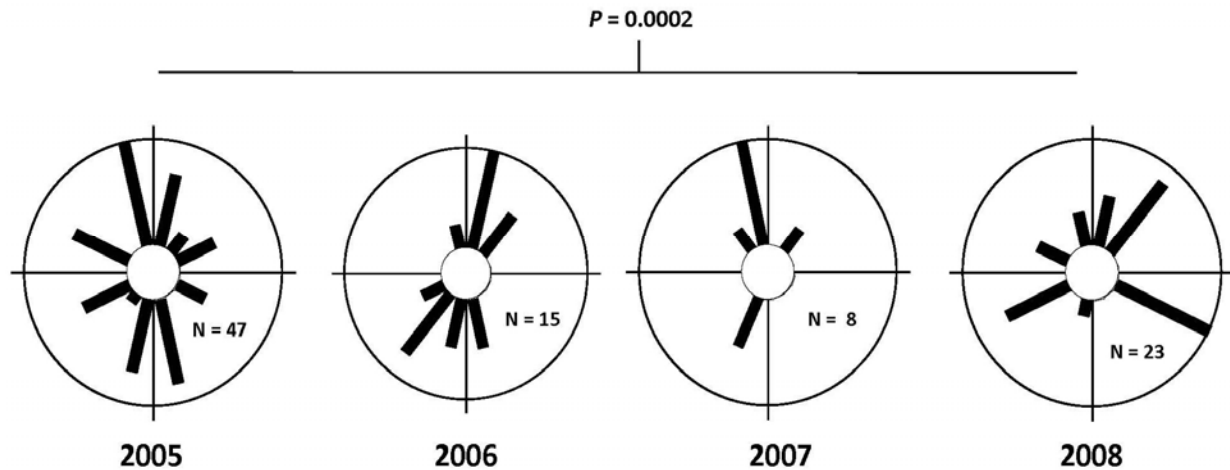
FIGURE 3. Capture location frequencies of metamorph *Ambystoma jeffersonianum* emigrating from Sylvatica Pond from 2005–2008. Circular axes indicate the location of pitfall traps around the pond from a fixed point. Radial axes show numbers of individuals captured at each pitfall trap. Sample sizes for each year are indicated. The bar and *P* value indicate a significant among-year difference in the orientation distributions.

Orientation analysis.—Pooled captures for all adults showed significant directionality (Rayleigh's $Z = 6.1$, $P = 0.002$, Fig. 2A). When observations were separated by direction of travel (entering or exiting), entering captures showed significant directionality (Rayleigh's $Z = 9.1$, $P < 0.001$, Fig. 2A), while exiting captures did not (Rayleigh's $Z = 1.7$, $P = 0.18$, Fig. 2A). A direct comparison showed the distributions of entering and exiting orientation to be significantly different ($\chi^2 = 33$, df = 13, $P = 0.002$). When entering observations were sub-grouped by year, 2006 and 2007 both showed significant directionality (Rayleigh's $Z = 5.0$, $P = 0.005$ and $Z = 5.5$, $P = 0.004$, respectively, Fig. 2B) while 2008 and 2009 observations did not (Rayleigh's $Z = 2.8$, $P = 0.055$ and $Z = 0.6$, $P = 0.55$ respectively, Fig. 2B). However, both multi-sample and pairwise chi-squared tests did not show the difference in the distribution of orientation across years to be significant. Metamorph captures pooled across all years did not show significant directionality (Rayleigh's $Z = 2.6$, $P = 0.07$). When grouped by year, all years showed no significant directionality ($P > 0.10$, Fig. 3). However, the orientation distributions for each year were significantly different ($\chi^2 = 74$, df = 36, $P < 0.001$, Fig. 3).

Entering and exiting orientations of individuals within a breeding year were significantly correlated, although the association was weak ($r = 0.059$, $P < 0.05$). Exiting orientations were significantly correlated with entering orientations in the next breeding year ($r = 0.103$, $P < 0.05$), and exiting orientations were significantly correlated across breeding years ($r = 0.254$, $P < 0.05$).

We used the absolute difference between the orientation of an individual in number of pitfall traps to graphically represent these associations (Fig. 4A, B and C, respectively).

DISCUSSION

We used four years of mark-recapture data from a small, isolated population of Jefferson Salamanders to obtain maximum likelihood estimates of adult terrestrial survival probability and breeding frequency. We also assessed migratory orientation during this period, examining population- and individual-level differences in entering and exiting orientation as well as differences across years. Our study provides the first estimates of breeding probability and migratory orientation patterns in this species.

Body mass in adult salamanders can be an indicator of habitat quality, as well as fecundity and individual condition (Salthe 1969). We found sex-based differences in body mass but no evidence for annual variation in mass of breeding adults. This could indicate little temporal variability in the terrestrial habitat during the four years of study, a hypothesis supported by the high rankings of our MSMR models without temporal variation in survival. In contrast to adults, emigrating metamorphs showed significant differences in mass across years, likely an indication of annual variation in pond habitat quality for aquatic larvae.

Our mark-recapture models indicated lower capture probabilities for *A. jeffersonianum* immigrants and emigrants compared to capture probabilities for
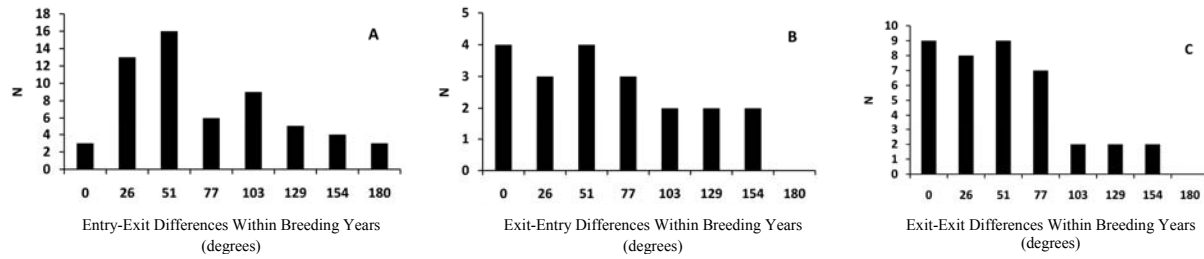
FIGURE 4. Frequencies of the differences between migratory orientations for individual *A. jeffersonianum* captured at Sylvatica Pond both entering and exiting within a year (A), exiting and entering in sequential breeding years (B), and exiting in sequential breeding years (C). A difference of zero degrees indicates migration along the same route and a difference of 180 degrees indicates a migration on the opposite side of the pond from the first observation. The associated circular correlations were all significant ($P < 0.05$).

Notophthalmus at the same drift fence during the same period (Grayson et al. 2011) and *Ambystoma* species captured in other studies using similar methods (e.g., Church et al. 2007). We expected low capture probabilities for individuals entering the pond because the drift fence was opened relatively late in the breeding season for this species. Sex-dependence in capture probability in our models and estimates of capture probabilities that were lower for males entering the pond compared to exiting the pond indicate that males arrived earlier to the breeding pond than females in this population. In general, early arrival to breeding sites can provide males with a mating advantage (Morbey and Ydenberg 2001). Early male arrival has been found in many studies of Ambystomatids (e.g. Hillis 1977; Hardy and Raymond 1980; Semlitsch 1983, 1985; Briggler et al. 2004) but patterns of migration timing can vary across populations (Williams et al. 2009). Our results are consistent with a previous study of *A. jeffersonianum* (Douglas 1979) indicating that males often arrive earlier to the breeding pond than females in this species.

Our estimates of model parameters assume breeding fidelity of adults to the Sylvatica and Horton pond complex. Movement rates of adult Ambystomatids between breeding ponds have been found to be low in some mark-recapture studies (Church et al. 2007; Gamble et al. 2009), although recent genetic evidence indicates that there could be much inter-pond breeding in some species (Tennessen and Zamudio 2003; Williams and DeWoody 2009). We believe that unobserved movements to outside ponds were rare in our study based on the low rates of movement we observed between adjacent ponds and sampling of the only other permanent pond within a kilometer. Thus, we expect the impact of movement outside the study area to be minimal on our parameter estimates.

The closed mark-recapture models that we used provide survival estimates that represent the annual probability of survival in both the terrestrial and aquatic habitat. Our annual estimates of survival probability are comparable to estimates from many other studies of

Ambystomatids (reviewed by Gamble et al. 2009). Survival estimates for both sexes are much higher than the 25% survivorship rates in an Indiana Jefferson Salamander population reported by Williams (1973), which to our knowledge represents the only previous estimates of adult survival in this species. Much of this difference in survival estimates may be due to our multistate statistical approach that allowed us to account for unobservable but living individuals and imperfect capture probabilities. However, it is certainly possible that habitat or environmental differences between the populations also account for differences in the survival probability estimates.

The ranking of our MSMR models provides strong support for sex-based differences in survival probability. The higher survival estimates in males compared to females could indicate sex-based discrepancies in costs of reproduction for individuals in this population. Higher reproductive costs could have direct effects on annual survival. Alternatively, females in this population may be skipping more breeding seasons than we had the ability to observe. Both Church et al. (2007) and Gamble et al. (2009) found that female Ambystomatids may exhibit facultative breeding, where they can skip breeding in years that are not favorable for reproductive success, while males were less likely to forgo breeding opportunities. It is possible that differences in breeding strategy between males and females contributed to the sex-based survival differences that we observed in *A. jeffersonianum,* if a greater proportion of females than males remained in the unobservable state for longer than the time frame of our study allowed us to measure. The high ranking of our MSMR model with sex dependence in breeding probability supports this hypothesis. Our estimates of breeding probability over the four years of our study indicated that the frequency of breeding in consecutive years is high for both sexes. In other words, the probability of transitioning to the non-breeder state after breeding the previous year was low. This may indicate that *A. jeffersonianum* in this population are able to

sequester appropriate resources required to reproduce multiple seasons in a row.

However, four years of data provides limited opportunities to observe an individual skip a year of reproduction and return to breed again. We only had one opportunity to observe individuals skip two years and return to breed again. Long absences from breeding have been documented in Ambystomatids, especially for females. For example, California Tiger Salamanders (*Ambystoma californiense*) were recaptured six years after being initially marked at a breeding site (Trenham et al. 2000). It is likely that many individuals that we recorded as breeding only once in fact survived to breed again after a hiatus longer than we could observe in four years of study. As a result of limited data and short study period, our estimates likely underestimate the true survival rates of the population and probability of transitioning to the unobservable state, particularly for females. These results highlight the necessity of long-term studies to better understand the breeding and survival schedules of long-lived vertebrates.

For pond-breeding amphibians, directional migratory orientation patterns could be attributed to common migration routes and/or common terrestrial habitat use. The results of our orientation analysis provide some indication that adult *A. jeffersonianum* migrations to the pond are directional, however, not all years showed significant directionality in entering orientations. There was no indication of directionality in migratory orientation out of the pond. These results could be a result of high variability in orientations among individuals, small population size, or both. Other studies have shown directionality in adult amphibian migration orientation (e.g., Dodd and Cade 1998; Marty et al. 2005; Malmgren 2002; Jenkins et al. 2006) often associated with specific terrestrial habitat types, although there is a large range in the degree of orientation variation across these studies. Because Sylvatica Pond is immediately surrounded by a generally homogenous mixed deciduous forest with little topographic variability, there may be few environmental restrictions on the spatial distribution of terrestrial habitat use in this population.

An additional component of migratory orientation pattern is the consistency of individuals' orientations through time. Within-year and among-year orientation fidelity has been documented in pond-breeding amphibians and *Ambystoma* species (Shoop 1965; Stenhouse 1985; Phillips and Sexton 1989; Marty et al. 2005; Trenham and Cook 2008). Our data are consistent with these previous studies and indicates that individuals can exhibit a degree of fidelity in migratory orientation, not only within years, but also among years, despite no significant directionality in exiting orientation at the population level. Given the documented correlation between orientation at the pond edge and terrestrial habitat use in Ambystomatids (Madison and Farrand 1998), it is possible that such patterns may be due be to individual fidelity in terrestrial habitat use.

The clear lack of unimodality in metamorph migration out of the pond is a pattern observed in the juvenile stage of other amphibian populations (this study; Malmgren 2002; Jenkins et al. 2006; Patrick et al. 2007). The general variability we observed in both metamorph mass and orientation is consistent with other observations of temporal variability in metamorph output in amphibian breeding ponds (e.g., Pechmann et al. 1991; Church et al. 2007). However, although we only observed this population over a brief period, metamorph production in our permanent pond appears more consistent than in other populations breeding in ephemeral pond systems. For example, Church et al. (2007) observed virtually no metamorphs emigrating from one study pond in all but the final year of study, when over 100 metamorphs emigrated. Church et al. (2007) also observed extreme annual fluctuations in metamorph frequency at two other ponds during the same period.

Our estimates of survival and breeding frequency and assessment of migratory orientation patterns in an *A. jeffersonianum* population are important for understanding the demography of these animals and inform the conservation of pond-breeding amphibians. Mensurative studies of amphibians in natural wetlands have generally been biased towards complex assemblages breeding in multiple ephemeral ponds, in an effort to understand the evolution of complex life histories and the ecology of metapopulations. Semlitsch and Bodie (1998) have pointed out the dangers of ignoring the conservation of small and isolated wetlands for the maintenance of local biodiversity. Our results from four years of monitoring a small permanent pond indicate that such wetlands can provide important breeding habitat for amphibian populations.

LITERATURE CITED

Armstrup, C.J., T.L. McDonald, and B.F.J. Manly. 2005. Handbook of Capture-recapture Analysis. Princeton University Press, Princeton, New Jersey, USA.

Bailey, L.L. 2004. Evaluating elastomer marking and photo identification methods for terrestrial salamanders: marking effects and observer bias. Herpetological Review 35:38–40.

Bailey, L.L., S.J. Converse, and W.L. Kendall. 2010. Bias, precision, and parameter redundancy in complex multistate models with unobservable states. Ecology 91:1598–1604.

Bailey, L.L., W.L. Kendall, and D.R. Church. 2009. Exploring extensions to multi-state models with multiple unobservable states. Pp. 693–710 *In* Modeling Demographic Processes in Marked Populations. Thompson, D.L., E.G. Cooch, and M.J. Conroy (Eds.). Springer Science and Business Media, New York, New York, USA.

Bailey, L.L., W.L. Kendall, D.R. Church, and H.M. Wilbur. 2004. Estimating survival and breeding probability for pond-breeding amphibians: a modified robust design. Ecology 85:2456–2466.

Batschelet, E. 1981. Circular Statistics in Biology. Academic Press, London, England.

Briggler, J.T., J.E. Johnson, and D.D. Rambo. 2004. Demographics of a Ringed Salamander (*Ambystoma annulatum*) breeding migration. Southwestern Naturalist 49:209–217.

Bull, J.J., and R. Shine. 1979. Iteroparous animals that skip opportunities for reproduction. American Naturalist 114:296–303.

Burnham, K.P., and D.R. Anderson. 2002. Model Selection and Multimodel Inference: A Practical Information-theoretic Approach. Springer Verlag, New York, New York, USA.

Church, D.R., L.L. Bailey, H.M. Wilbur, W.L. Kendall, and J.E. Hines. 2007. Iteroparity in the variable environment of the salamander *Ambystoma tigrinum*. Ecology 88:891–903.

Cushman, S.A. 2006. Effects of habitat loss and fragmentation on amphibians: a review and prospectus. Biological Conservation 128:231–340.

Davis, T.M., and K. Ovaska. 2001. Individual recognition of amphibians: effects of toe clipping and fluorescent tagging on the salamander *Plethodon vehiculum*. Journal of Herpetology 35:217–225.

Dodd, C.K., Jr., and B.S. Cade. 1998. Movement patterns and the conservation of amphibians breeding in small, temporary wetlands. Conservation Biology 12:331–339.

Douglas, M.E. 1979. Migration and sexual selection in *Ambystoma jeffersonianum*. Canadian Journal of Zoology 57:2303–2310.

Faccio, S.D. 2003. Postbreeding emigration and habitat use by Jefferson and Spotted Salamanders in Vermont. Journal of Herpetology 37:479–489.

Gamble, L.R., K. McGarigal, D.B. Sigourney, and B.C. Timm. 2009. Survival and breeding frequency in Marbled Salamanders (*Ambystoma opacum*): implications for spatio-temporal population dynamics. Copeia 2009:394–407.

Gill, D.E. 1985. Interpreting breeding patterns from census data: a solution to the Husting dilemma. Ecology 66:344–354.

Gimenez, O., A. Viallefont, E.A. Catchpole, R. Choquet, and B.J.T. Morgan. 2004. Methods for investigating parameter redundancy. Animal Biodiversity and Conservation 27:561–572.

Grayson, K.L. 2010. Life history trade-offs of migration in a pond-breeding amphibian. Ph.D. Dissertation, University of Virginia, Charlottesville, Virginia, USA. 163 p.

Grayson, K.L., L.L. Bailey, and H.M. Wilbur. 2011. Life history benefits of residency in a partially migrating pond-breeding amphibian. Ecology 92:1236–1246.

Hardy, L.M., and L.R. Raymond. 1980. The breeding migration of the Mole Salamander, *Ambystoma talpoideum*, in Louisiana. Journal of Herpetology 14:327–335.

Harris, R.N., and P.M. Ludwig. 2004. Resource level and reproductive frequency in female Four-toed Salamanders, *Hemidactylium scutatum*. Ecology 85:1585–1590.

Hillis, D.M. 1977. Sex ratio, mortality rate, and breeding stimuli in a Maryland population of *Ambystoma maculatum*. Bulletin of the Maryland Herpetological Society 13:84–91.

Houlahan, J.E., C.S. Findlay, B.R. Schmidt, A.H. Meyer, and S.L. Kuzmin. 2000. Quantitative evidence for global amphibian population declines. Nature 404:752–755.

Husting, E.L. 1965. Survival and breeding structure in a population of *Ambystoma maculatum*. Copeia 1965:352–362.

Jenkins, C.L., K. McGarigal, and B.C. Timm. 2006. Orientation of movements and habitat selection in a spatially structured population of Marbled Salamanders (*Ambystoma opacum*). Journal of Herpetology 40:240–248.

Kendall, W.L. 2004. Coping with unobservable and mis-classified states in capture-recapture studies. Animal Biodiversity and Conservation 27:97–101.

Kendall, W.L., and J.D. Nichols. 1995. On the use of secondary capture-recapture samples to estimate temporary emigration and breeding proportions. Journal of Applied Statistics 22:751–76.

Kendall, W.L., and J.D. Nichols. 2002. Estimating state transition probabilities for unobservable states using

capture-recapture/resighting data. Ecology 83:3276–3284.

Lebreton, J.D., K.P. Burnham, J. Clobert, and D.R. Anderson. 1992. Modeling survival and testing biological hypotheses using marked animals: a unified approach with case studies. Ecological Monographs 62:67–118.

Madison, D.M., and L.I. Farrand. 1998. Habitat use during breeding and emigrating in radio-implanted Tiger Salamanders, *Ambystoma tigrinum*. Copeia 1998:402–410.

Malmgren, J.C. 2002. How does a newt find its way from a pond? Migration patterns after breeding and metamorphosis in Great Crested Newts and Smooth Newts. Herpetological Journal 12:29–35.

Marty, P., S. Angelibert, N. Giani, and P. Joly. 2005. Directionality of pre- and post-breeding migrations of a Marbled Newt population (*Triturus marmoratus*): implications for buffer zone management. Aquatic Conservation: Marine and Freshwater Ecosystems 15:215– 225.

McCaffery, R.M., and B.A. Maxwell. 2010. Decreased winter severity increases viability of a montane frog. Proceedings of the National Academy of Sciences 107:8644–8649.

Morbey, Y.E., and R.C. Ydenberg. 2001. Protandrous arrival timing to breeding areas: a review. Ecology Letters 4:663–673.

Mullin, S.J., and S. Klueh. 2009. Demographics of a geographically isolated population of a threatened salamander (Caudata: Ambystomatidae) in central Illinois. Herpetological Conservation and Biology 4:261–269.

Muths, E., R.D. Scherer, and B.A. Lambert. 2010. Unbiased survival estimates and evidence for skipped breeding opportunities in females. Methods in Ecology and Evolution 2010:123–130.

Patrick, D.A., A.J.K. Calhoun, and M.L.J. Hunter. 2007. Orientation of juvenile Wood Frogs, *Rana sylvatica*, leaving experimental ponds. Journal of Herpetology 41:158–163.

Pechmann, J.H.K., D.E. Scott, R.D. Semlitsch, J.P. Caldwell, L.J. Vitt, and J.W. Gibbons. 1991. Declining amphibian populations: the problem of separating human impacts from natural fluctuations. Science 253:892–895.

Petranka, J.W. 1998. Salamanders of the United States and Canada. Smithsonian Institution Press, Washington, D.C., USA.

Phillips, C.A., and O.J. Sexton. 1989. Orientation and sexual differences during breeding migrations of the Spotted Salamander, *Ambystoma maculatum*. Copeia 1989:17–22.

Pollock, K.H. 1982. A capture-recapture design robust to unequal probability of capture. Journal of Wildlife Management 46:752–757.

Salthe, S.N. 1969. Reproductive modes and the number and sizes of ova in the Urodeles. American Midland Naturalist 81:467–490.

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. 1985. Analysis of climatic factors influencing migrations of the salamander *Ambystoma talpoideum*. Copeia 1985:477–489.

Semlitsch, R.D. 2000. Principles for management of aquatic-breeding amphibians. Journal of Wildlife Management 64:615–631.

Semlitsch, R.D. 2008. Differentiating migration and dispersal processes for pond-breeding amphibians. Journal of Wildlife Management 72:260–267.

Semlitsch, R.D., and J.R. Bodie. 1998. Are small, isolated wetlands expendable? Conservation Biology 12:1129–1133.

Shoop, R.C. 1965. Orientation of *Ambystoma maculatum*: movements to and from breeding ponds. Science 149:558–559.

Stenhouse, S.L. 1985. Migratory orientation and homing in *Ambystoma maculatum* and *Ambystoma opacum*. Copeia 1985:631–637.

Stuart, S.N., J.S. Chanson, N.A. Cox, B.E. Young, A.L. Rodrigues, D.L. Fischman, and R.W. Waller. 2004. Status and trends of amphibian declines and extinctions worldwide. Science 306:1783–1786.

Tennessen, J.A., and K.R. Zamudio. 2003. Early male reproductive advantage, multiple paternity and sperm storage in an amphibian aggregate breeder. Molecular Ecology 12:1567–1576.

Trenham, P.C., and D.G. Cook. 2008. Distribution of migrating adults related to the location of remnant grassland around an urban California Tiger Salamander breeding pool. Pp. 9–16 *In* Urban Herpetology. Mitchell, J.C., R.E. Jung Brown, and B. Bartholomew (Eds.). Society for the Study of Amphibians and Reptiles, Salt Lake City, Utah, USA.

Trenham, P.C., H.B. Shaffer, W.D. Koenig, and M.R. Stromberg. 2000. Life history and demographic variation in the California Tiger Salamander (*Ambystoma californiense*). Copeia 2000:365–377.

White, G.C., and K.P. Burnham. 1999. Program Mark: survival estimation from populations of marked animals. Bird Study 46 Supplement:120–138.

Wilbur, H.M. 1980. Complex life cycles. Annual Review of Ecology and Systematics 11:67 –93.

Williams, P.K. 1973. Seasonal movements and population dynamics of four sympatric mole salamanders, genus *Ambystoma*. Ph.D. Dissertation, Indiana University, Bloomington, Indiana, USA.

Williams, R.N., and J.A. DeWoody. 2009. Reproductive success and sexual selection in wild Eastern Tiger Salamanders (*Ambystoma t. tigrinum*). Evolutionary Biology 36:201–213.

Williams, R.N., D. Gopurenko, K.R. Kemp, B. Williams, and J.A. DeWoody. 2009. Breeding chronology, sexual dimorphism, and genetic diversity of congeneric Ambystomatid salamanders. Journal of Herpetology 43:438–229.

Zar, J.H. 1996. Biostatistical Analysis. 3rd Edition. Prentice-Hall, Englewood Cliffs, New Jersey, USA.
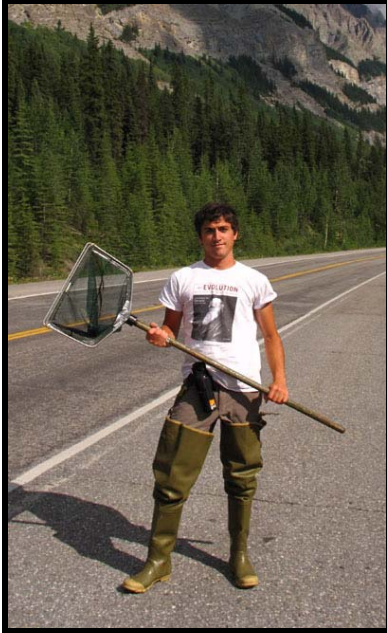
STEPHEN P. DE LISLE graduated from the University of Virginia with a B.S. in Biology in May 2010. He is currently a Ph.D. student in the Department of Ecology and Evolutionary Biology at the University of Toronto, and plans to study sexual selection and life history evolution in an amphibian species for his dissertation research. (Photographed by Matthew Robinson)
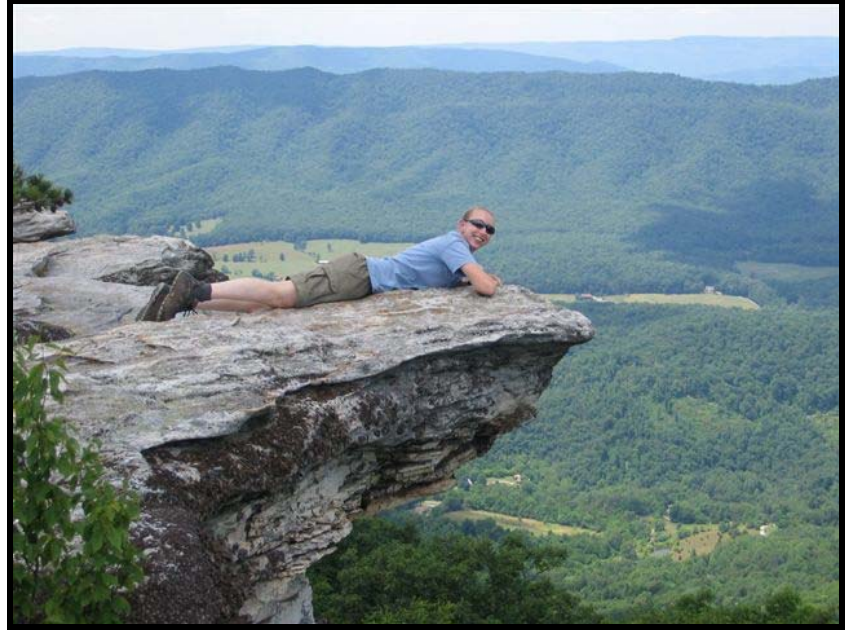
KRISTINE L. GRAYSON received a B.S. from Davidson College in North Carolina and completed her Ph.D. at the University of Virginia in May 2010. Her research examined the life-history trade-offs of migration in Red-spotted Newts (*Notopthalmus v. viridescens*) and was conducted at Mountain Lake Biological Station. She is currently studying population dynamics of Tuatara (*Sphenodon punctatus*) at Victoria University of Wellington in New Zealand. (Photographed by Kelly Hansen)