DISPERSAL VERSUS SITE TENACITY OF ADULT AND JUVENILE RED-BACKED SALAMANDERS (*PLETHODON CINEREUS*)

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ABSTRACT: In many species of amphibians that display territoriality, it is unclear at what life stage and to what extent dispersal occurs. We examined whether differences existed between life stages in dispersal and homing in red-backed salamanders (*Plethodon cinereus*) in continuous habitat. In a mark–recapture study, we found that juvenile and adult movements between years were not significantly different. The median distances moved by both adults (0.85–0.88 m) and juveniles (1.14–1.22 m) of *P. cinereus* between years were similar to the diameter of adult home ranges in this area (1.15 m). In a homing experiment, we found that, although the probability of recapture of translocated individuals under their original cover object increased with body size, both adults and juveniles successfully orientated toward their original cover objects when displaced over short distances (1.5 m and 6.25 m), but their orientation was random when displaced long distances (12.5 m and 25 m). Despite the current assumption that juveniles of *P. cinereus* lack territorial behaviors, our results suggest that regardless of life stage, site tenacity accounts for lack of movements.

Key words: Dispersal; Homing; Life stages; Mark–recapture; *Plethodon cinereus*

TERRITORIALITY can be defined as the defense and advertisement of an area by an animal with the goal of exclusive occupancy (Wilson, 1975). Maintenance of a territory and the site fidelity associated with such maintenance can be energetically expensive because they limit time available for other activities such as foraging, and may lead to injury or death during defense (Marler and Moore, 1988; Marler et al., 1995). However, territoriality can benefit individuals through exclusive access to resources, including prey, refugia, and mates (Brown and Orians, 1970; Stamps, 1994).

In cases where adult vertebrates defend territories, juveniles may be tolerated or excluded entirely due to current or future competition with adults for resources (Stamps, 1994). Juveniles may permanently disperse to establish their own territories or remain in their natal areas (reviewed in Koenig et al., 1992). In recent years, juvenile amphibians have become an important conservation consideration (Cushman, 2006), because juvenile movements are disproportionately affected by human activities (Patrick et al., 2008). Like other vertebrates, dispersal is well-documented in juveniles of pond-breeding amphibians, but little is known about movements of juvenile terrestrial amphibians (reviewed in Pough et al., 2003).

Homing behavior is the tendency for individuals to return to their home range or territory after displacement. Homing occurs in many amphibians (Diego-Rasilla and Phillips, 2007). However, prior to the establishment of territories, individuals may not home (Jaeger et al., 1993).

Plethodontid salamanders, in particular the red-backed salamander, *Plethodon cinereus*, are excellent model organisms for studies of behavioral ecology, especially territoriality (Mathis et al., 1995). Because a cover object (rock or log; CO) can retain moisture longer than leaf litter (Mathis et al., 1995), male and female adults of *P. cinereus* defend COs and surrounding territory as feeding sites and moist refugia (Mathis, 1990, 1991). Following displacement from distances far greater than the radius of their home range, most adults and some older juveniles of *P. cinereus* homed to their territories, specifically their COs (Kleeberger and Werner, 1982; Marsh et al., 2005). Despite their homing abilities, adults of *P. cinereus* are hypothesized to have limited movements, at least within the summer season (Mathis, 1991), and move only infrequently between cover objects over multiple years (Gillette, 2003). Cabe et al. (2007) found small, but significant genetic differentiation within continuous habitat (unfragmented hab-

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itat with no obvious geographic barriers) over as little as 200 m, suggesting a lack of gene flow and dispersal. However, some individuals moved between 30 m and 40 m across open fields to colonize new habitat (Marsh et al., 2004) and the northern range of *P. cinereus* was expanding at a rate of 80 m/yr (Cabe et al., 2007). It is unclear whether long-distance movements are normal for *P. cinereus* in continuous habitat or whether movement only occurs when new habitat is available.

Although the movements of adult *P. cinereus* have been well-studied, little is known about the movements of juveniles in any fully terrestrial species of amphibian, including their ability to home, disperse, and establish territories (reviewed in Pough et al., 2003). For *P. cinereus*, data on juvenile movements are very limited (reviewed in Petranka, 1998) because they are infrequently found under cover objects and are difficult to locate in the leaf litter (Jaeger et al., 1995).

We tested the hypothesis that dispersal and homing differ in juveniles and adult *P. cinereus* by comparing their behavior in a homing experiment and in mark–recapture studies over single and multiple years. Comparison of fine-scale movements in the two life stages may elucidate the discrepancy between studies that indicate limited dispersal (molecular: Cabe et al., 2007; field: Gillette, 2003; Mathis, 1991) and those that indicate range expansion through long-distance movements (Kleeberger and Werner, 1982; Marsh et al., 2004).

MATERIALS AND METHODS

Quantifying Dispersal

In 2005 we established a 12×12-m (144-m^2) plot at the University of Virginia's Mountain Lake Biological Station in Giles County, Virginia, United States. For survey purposes, we subdivided the plot into six 2×12-m transects. This plot was surveyed 39 times from 18 May to 14 October 2005 using diurnal and nocturnal surveys. Nocturnal surveys were conducted by walking each transect (in random order) and shining a flashlight ahead as the observer walked slowly down the transect. Nocturnal surveys were performed from 45 minutes after sunset up to three hours after

sunset only on nights when it had rained during the previous 24 hours. During diurnal surveys, which were conducted at least five days apart, we gently lifted up every natural CO on the entire plot. After processing (see below), salamanders were released at their point of capture (to the nearest cm; nocturnal surveys) or adjacent to the CO that they were located under (diurnal surveys).

At each capture we recorded the location (± 1.0 cm) and marked individuals uniquely using combinations of five fluorescent elastomers (Northwest Marine Technologies Inc., Shaw Island, WA). Marking using elastomers does not appear to influence behavior or survivorship of *P. cinereus* (Davis and Ovaska, 2001). We measured snout–vent length (SVL; ± 0.01 mm) using digital calipers (Mitutoyo America Corporation, Aurora, IL), and categorized individuals as juveniles (<36 mm) or adults (>36 mm; Sayler, 1966).

In the 2006 season, we surveyed the plot 31 times from 20 April to 13 October for salamanders that had been marked in 2005 and also marked salamanders that we had not previously captured. In 2007 we expanded the plot to 36×36 m (1296 m^2), subdividing it into 54 2×12-m survey transects. From 6 June to 2 August 2007, we conducted nocturnal surveys, searching six randomly selected transects on the plot per night, in order to recapture previously marked salamanders from the original 12×12-m plot. We conducted two nocturnal surveys per transect and five daytime CO searches per transect in random order in 2007.

We calculated the Euclidean distance from the grid coordinate location (± 1.0 cm) where marked salamanders were captured in the previous year(s) to determine dispersal distance from their previous home range(s). For individuals captured multiple times in a single year, we used the center of their home range for that year.

Dispersal Data Analysis

The original interyear distance data were not normal and no transformations helped the data meet the assumptions of normality. We used separate nonparametric Wilcoxon two-sample tests (SAS Institute Inc., 2004) to compare dispersal distances when we

searched plots of different areas (2005–2006 [144 m² searched] versus 2006–2007 [1296 m²]), over periods of one year (2006–2007) versus two years (2005–2007), and between juvenile versus adult life stages in each study period. One-sided tests, ranked tests that are the nonparametric equivalents of one-tailed t-tests, were used based on our predictions that dispersal distance would be farther during the year when a larger area was searched (1296 m² in 2006–2007), when more time had elapsed (two years: 2005–2007), and for juveniles because they are hypothesized to disperse (Mathis, 1991). Most salamanders were captured in only two of the three years, providing a single dispersal measure (2005–2006, 2006–2007, or 2005–2007). When salamanders were captured in all three years, we had three measures of annual dispersal for that individual. In order to meet the assumption of independence of data for our tests (e.g., 2005–2006 versus 2006–2007), we randomly chose one dispersal distance from those individuals (2005–2006, 2006–2007 or 2005–2007) for the between-year analyses and discarded the other two. Because we used the same dispersal data to make several comparisons, we applied the Bonferronni correction for conducting multiple tests on related data to our α values (Rice, 1989).

In order to look at differences in dispersal distances based on SVL, we used one-sided Spearman's rank correlations for each of the three time periods. One-sided tests were used based on the prediction that larger (older) salamanders are less likely to disperse (Mathis, 1991).

Homing Experiment

We conducted CO searches to capture red-backed salamanders between 1630 h and 1945 h from 17 June to 23 July 2007. Captured individuals were measured and categorized as adults or juveniles (Sayler, 1966) and marked with a single injection of fluorescent elastomer. We dipped individuals in powdered fluorescent pigments (Magruder Color Company, Fall River, MA) from their tail up to their fore-shoulders. These powders have been successfully used in the tracking of amphibians, including *P. cinereus* (Roberts and Liebgold, 2008), with no apparent effects on survival, growth, or integumentary gas

exchange (Orlofske et al., 2009). We powdered 67 individuals and translocated them to a different natural CO (with no salamander present) located 1.5, 6.25, 12.5, or 25 m away from their original CO in a random compass direction. If the random direction crossed a stream or a road, or there was no CO in that direction, we selected a new random direction. Translocated individuals were captured at least 50 m apart.

We tracked individuals the same night as their capture (processing: 1630 h to 1945 h; track: 2130 h to 0000 h), using an ultraviolet flashlight to locate the powder trail. We measured the initial orientation, which was the angle formed between the straight-line path to the CO (0°) and the trail from the translocation CO, at approximately 1 m from the translocation CO.

Each original CO and translocation CO, as well as all potential COs within 2 m of those two sites, was examined three times (on days 1, 5, and 9 after displacement) for the marked salamander. If the marked animal was found under the original CO (or a CO <50 cm from its original CO), then the individual was considered to have homed successfully.

Homing Data Analysis

We used a logistic regression to identify factors associated with successful homing, including life stage (adult or juvenile), distance translocated (1.5, 6.25, 12.5, or 25 m) and the covariate SVL. We tested whether initial orientation was associated with successful homing in a separate logistic regression because the initial orientation data from 14 of 67 translocated salamanders were not available (individuals did not move in that first evening, left ambiguous trails resulting in unclear orientation, or it rained before trails could be measured).

We also tested what factors influenced the initial orientation of salamanders. The orientation data (but not the successful homing data) were combined into two displacement distance categories (short distances: 1.5 and 6.25 m and long distances: 12.5 and 25 m) per life stage instead of four, in order to meet the minimum sample sizes needed to conduct Rayleigh tests for nonrandom circular movements ($n = 10$; Batschelet, 1981) using the

TABLE 1.—Tests for the influences of life stage, dispersal time, and area searched on dispersal distances of *Plethodon cinereus* using one-sided Wilcoxon two-sample tests (SAS Institute Inc., 2004). Significant results are bolded.

		n	Median (m)	IQ range	Z	p	α
2005–2006	Life stage				1.39	0.0826	0.025
	Adult	68	0.85	0.43–1.85			
	Juvenile	58	1.14	0.52–3.58			
2006–2007	Life stage				−1.69	0.0454	0.017
	Adult	17	0.68	0.37–1.30			
	Juvenile	25	1.22	0.71–3.35			
2005–2007	Life stage				**−1.96**	**0.0249**	**0.025**
	Adult	10	1.06	0.74–1.38			
	Juvenile	14	4.85	1.22–8.45			
Years moved					1.44	0.0751	0.017
1	(2006–2007)	32	1.20	0.62–3.27			
2	(2005–2007)	17	1.66	0.77–5.54			
Area searched					0.93	0.1757	0.017
144 m²	(2005–2007)	109	1.04	0.43–2.09			
1296 m²	(2006–2007)	32	1.20	0.62–3.27			

program Oriana (Kovach Computing Services, Angelesy, Wales). In the Rayleigh test, when the lengths (r) of the mean vectors of movement (μ) are large, the mean vectors are significantly different than random movements (Batschelet, 1981). When $P < 0.05$, we used the 99% CI to determine whether salamanders moved toward their original COs (Rittenhouse et al., 2007).

RESULTS
Dispersal Study

There was no difference in dispersal distance during different years sampled using different sized study plots (Table 1). Dispersal distance over a two-year period was significantly greater in juveniles than in adults (Table 1). Although there was a tendency for juveniles to disperse farther during individual years, these were not statistically significant differences ($P > 0.05$; Table 1).

SVL was not significantly correlated with dispersal distance in single years (2005–2006:

$r = −0.144$, $P = 0.071$; 2006–2007: $r = −0.225$, $P = 0.068$). SVL was significantly negatively correlated with dispersal distance over the two-year period (2005–2007: $r = −0.360$, $P = 0.012$).

Homing Experiment

Of the total 63 salamanders that homed, all but one returned to their original COs. Logistic regressions revealed no effects of displacement distance, life stage, or initial orientation on the likelihood of homing (Table 2; distance: $\chi^2_2 = 1.31$, $P = 0.726$; life stage: $\chi^2_2 = 2.23$, $P = 0.136$; initial orientation: $\chi^2_2 = 0.12$, $P = 0.733$). There was a significant effect of SVL on homing ($\chi^2_2 = 5.10$, $P = 0.024$). From the odds ratio, which is used for estimating the size of the effect of variables within a logistic regression, a 1-mm increase in SVL increased the probability of homing by 22% (1.22; 95% CI: 1.03–1.44).

The initial orientation of adults and juveniles displaced short distances (1.5 or 6.25 m) was nonrandom (Table 2; adults: $n = 10$, $r =$

TABLE 2.—Homing success rates and initial orientation angles of juveniles and adults of *Plethodon cinereus* for various displacement distances. Sample sizes (n) are in parentheses.

	% Homing success (n)		Mean initial orientation angle (n)	
Displacement distance (m)	Juveniles	Adults	Juveniles	Adults
1.5	0 (4)	17 (6)	83 ± 36° (3)	26 ± 14° (5)
6.25	0 (8)	57 (7)	10 ± 18° (7)	0 ± 24° (5)
12.5	36 (14)	11 (9)	53 ± 40° (11)	194 ± 23° (7)
25	0 (7)	0 (8)	36 ± 21° (7)	359 ± 35° (8)

0.74, Z = 5.40, P = 0.002; juveniles: n = 10, r = 0.56, Z = 3.13, P = 0.039) and directed toward their original COs (μ_{Adults} = 14.9°, 99% CI: 341.6°–82.1°; $\mu_{Juveniles}$ = 26.6°, 99% CI: 329.4°–83.1°). The initial orientation of adults and juveniles displaced long distances (12.5 or 25 m) was no different than random movements (Table 2; adults: n = 18, r = 0.15, Z = 0.35, P = 0.713; juveniles: n = 15, r = 0.28, Z = 1.41, P = 0.247).

DISCUSSION

Regardless of life stage, individuals of *P. cinereus* were found close to where they were captured in the previous year(s). The median distance moved by *P. cinereus* between single years (Table 1) was similar to the diameter of the mean adult home range (1.15 m: Liebgold and Jaeger, 2007). These data support molecular studies (Cabe et al., 2007; E. Liebgold, unpublished data) finding significant genetic differences over small distances, but not the argument that juveniles are not territorial (Mathis, 1991). However, juveniles did move farther than adults over a period of two years (Table 1) and size was negatively correlated with salamander movements over the two-year period, leading to the possibility that juveniles disperse farther distances than adults or shift their home ranges slightly more than adults over multiple years.

Koenig et al. (1996) argued that plot-based population dispersal studies of marked individuals can have a bias, favoring individuals that disperse shorter distances. To minimize this bias, we surveyed an area 900% larger than the area in which marking occurred (sensu Serrano et al., 2003). While we cannot rule out other factors that may have affected dispersal, we infer that the concordance of recapture distances between years despite the large increase in survey area provides some evidence that salamanders do not move far between years.

Adults and juveniles were also similar in that they immediately oriented toward their original CO when displaced short distances. However, the orientations of both adult and juvenile salamanders displaced larger distances were no different than random directions. The lack of immediate movements toward their original COs when displaced large

distances could be explained by a lack of familiarity with terrain far from their home range. Adults and juveniles of *P. cinereus* are known to have small home ranges within years (Liebgold and Jaeger, 2007; Mathis, 1991) and move only small distances between years (this study).

Contrary to our findings for initial orientation, homing success was not affected by the displacement distance. We note this lack of significance cautiously; logistic regressions are not very powerful tests because they use categorical, not continuous dependent data, often requiring large sample sizes. However, because logistic regressions parse out variance much like ANOVAs, any potential patterns based on distance (Table 2) may to be due to random distributions of salamander size between distance groups (but within age classes). Salamander size significantly affected homing success, with larger individuals much more likely to be found under their original COs than smaller ones. Similar return patterns based on size had been found in a previous homing experiment (Marsh et al., 2005).

The increase in homing success by larger individuals has three possible nonmutually exclusive explanations. These data may provide support for the hypothesis of a cognitive map, with individuals with more movement experience (e.g., older individuals) having a greater probability of homing (Jaeger et al., 1993). Secondly, our findings may support the hypothesis that juveniles are not territorial (Mathis, 1991). One last competing hypothesis is that leaf litter or underground habitat may be the preferred habitat of juveniles compared with adults (Cáceres-Charneco and Ransom, 2010), which are more frequently found under COs (Jaeger et al., 1995). If so, our initial homing orientation results lead to the inference that juveniles show site fidelity and differences in success may be a byproduct of increased detectability of larger individuals under COs, rather than a decreased desire or ability to return. Based on the similar and apparently successful initial homing orientations of juveniles and adults and the high site fidelity of juveniles, we conclude that this last hypothesis is the most likely.

Despite the widely held assumption that juveniles of *P. cinereus* are not territorial (e.g., Mathis, 1991), they rarely moved large distances between years and were consistently as able as adults to orient toward home ranges following displacement over small distances. These findings provide evidence that juveniles remain in a relatively small area between years and demonstrate site fidelity to that area. The limited movements and site tenacity of *P. cinereus* over all life stages supports previous mark–recapture estimates from smaller plots (Mathis, 1991) and accounts for significant genetic differences between nearby populations (Cabe et al., 2007).

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