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EUROPEAN PLETHODONTID SALAMANDERS ON THE FOREST FLOOR: LOCAL ABUNDANCE IS RELATED TO FINE-SCALE ENVIRONMENTAL FACTORS

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***Abstract.*—Understanding habitat selection by animal populations is one of the most relevant topics in ecology. In this study, we analyzed fine-scale habitat selection in a population of the salamander *Speleomantes strinatii* living in a Mediterranean forest environment. We used repeated surveys to estimate salamander abundance on 40 plots (12 m^2) by applying hierarchical models to repeated count data. In addition, we estimated seven habitat variables for each plot to infer the relationships between local factors and salamander abundance. The salamander population showed a patchy distribution; higher abundances were found in plots with a prevalent North aspect and with a high number of rocks lying on the forest floor. Conversely, there was a negative influence of superficial water runoff on salamander abundance. Our results demonstrate that fine-scale environmental factors, mainly related to the physiological constraints of the salamanders, shape local abundance even in apparently suitable environments. Finally, the overall estimate of 0.86 individuals/m^2 (95% CI 0.34–1.08) was similar to one obtained in a similar environment with removal sampling, suggesting efficiency of N-mixture modelling to estimate abundance of European plethodontids.**

Key Words.—abundance estimation; habitat selection; hierarchical modelling; N-mixture models; repeated count data; *Speleomantes strinatii*

INTRODUCTION

Terrestrial salamanders belonging to the family Plethodontidae are important components of temperate forest ecosystems where they contribute to the regulation of soil-dwelling invertebrates and to the cycling and storing of nutrients (reviewed by Davic and Welsh 2004; and by Wells 2007). Plethodontid salamanders are lungless, and respiration occurs mainly though their skin and oral mucosa (Wells 2007). Because their skin must be kept moist to respire, surface activity of terrestrial plethodontids is restricted by temperature, moisture, and wind speed (Peterman and Semlitsch 2014), which also influence their dispersal between suitable habitat patches (Heatwole 1962, Wells 2007; Peterman and Semlitsch 2013).

Although the family Plethodontidae is widely distributed in North America, Central America, and northern South America (Wake 2013), only one species occurs in Asia (Min et al. 2005), and only eight species are found in southern Europe (seven species of *Speleomantes* and one species of *Atylodes*, option 5 in Wake 2013). In Europe, these fully terrestrial salamanders are found in Mediterranean habitats in southern France, continental Italy, and on the island of Sardinia (Lanza et al. 2006; Lanza 2007). In particular, *Speleomantes* spp. are primarily epigean, living in interstitial subsurface habitats, on the forest floor, and in humid rock crevices (Lanza et al. 2006; Lanza 2007). They are, however, capable of establishing stable populations in natural and artificial underground habitats (i.e., caves) and, have been categorized by Lanza (2007) as eutroglophiles (as reviewed by Sket 2008). To date, studies on the ecology of European plethodontid species have been limited to environmental factors influencing the distribution and the selection of microhabitat inside underground habitats or dry stone walls (Salvidio et al. 1994; Ficetola et al. 2012; Lunghi et al. 2015; Manenti 2015). Few studies have analyzed ecological factors influencing the distribution and abundance of *Speleomantes* in forest environments where, under favorable conditions, they may attain relatively high population densities (Salvidio 1998, 2007).

The goals of this study were to evaluate habitat selection as a function of environmental factors, to obtain local estimates of abundance and density for the focal population, and to compare our results with those published in other studies. Specifically, we conducted repeated surveys at multiple sites, recording counts of *Speleomantes strinatii* (Aellen 1958) and analyzing repeated count data by hierarchical modeling (Royle 2004). Moreover, we included several microhabitat variables to allow a better estimation of micro-habitat selection, in particular to allow inference on these parameters on local abundance of salamanders.

MATERIALS AND METHODS

Study species and sampling site.—*Speleomantes strinatii* is a fully terrestrial plethodontid salamander occurring in southern France and northwestern Italy (Lanza 2007). This species, long-lived with a maximum snout-vent length of 70 mm (Lindström et al. 2010), is found in different habitats from sea level up to 2000 m above sea level (a.s.l.) in the Alps. We conducted our study at an elevation of about 900 m a.s.l.in a Supra-Mediterranean Deciduous Mixed Forest (Blondel and Aronson 1999) dominated by Chestnut trees (*Castanea sativa*). The study area is a small valley located in northwestern Italy, Province of Genova (44°34′00″N; 9°08′10″E) and bisected in north-facing and south-facing slopes by a first-order Apennine stream. In this area, the species shows a bimodal pattern of activity, which peaks in the spring and autumn during rainy periods when the soil moisture is high (Salvidio 1993). At the study site, *S. strinatii* is relatively abundant, and two other salamanders, *Salamandrina perspicillata* (Savi 1821) and *Salamandra salamandra* (Linnaeus 1758), are also present (Salvidio et al. 2012).

Field surveys.—On both sides of the stream, we randomly selected 40 plots and individually marked each with plastic flags. Each plot was 12 m^2 (rectangular plots 3 × 4 m, covering a total area of 480 m^2) and spaced at least 20 m apart. We used these plots as sites for our spatially and temporally replicated surveys. During autumn 2013 (from 5 October to 17 November 2013), we visited each site five times during daylight (0900–1100), in cloudy or rainy days, and at regular intervals (8–9 d). During each survey, we searched leaf litter, lifted cover objects (e.g., rocks and logs), checked rocky crevices with electric flashlights, and recorded the number of salamanders found at each plot. We did not attempt to capture the salamanders found. In addition, at each site we measured four environmental variables: (1) aspect of the plot (Aspect; e.g., north and south), (2) slope with an inclinometer (Slope), (3) distance from stream with a digital telemeter (Stream), and (4) distance from the nearest rock cliff with a digital telemeter (Cliff). We considered three other variables: (1) presence of dead-wood logs (Wood), (2) presence of water runoff on the soil surface during rainfalls (Water Runoff, presence or absence directly observed in the field during or after rainfalls), and (3) percentage coverage of rocks larger than 20 cm (Rock) in classes of high (> 20%), medium (6–20%), low (1–5%), or none. We estimated percentage cover using a comparison chart for visual percentage cover estimation.

Data analysis.—As a first step, we conducted a correlation analysis between site covariates to identify possible collinearity (MacNally 2002), and avoided including correlated variables in the same model. We analyzed repeated count data using Royle's (2004) N-mixture model. The major assumption of this model is that the population is closed to births, deaths, emigration, and immigration during the sampling period (i.e., the population abundance remains constant during surveys). Because our sampling lasted only one month, we are confident that this assumption could be met (Peterman and Semlitsch, 2013). N-mixture models provide estimates of two parameters: the state variable, i.e., mean abundance of salamanders per site (λ), and the probability of detection per individual (r; Royle 2004). Total abundance or density can be obtained as derived parameters. As a first step in modeling abundance we built several global models (i.e., the model with all the covariates and in which other candidate models are nested) using different distributions (e.g., NB: Negative Binomial; P: Poisson; ZIP: Zero-Inflated Poisson) and values for the upper limit of integration (K). We tested all candidate models for goodness of fit with a Pearson chi-square test (MacKenzie and Bailey 2004), using a parametric bootstrap procedure (1000 re-sampling). Among these models, P-value and over dispersion parameter (*c-hat*) have been used to select the best global model (Burnham and Anderson 2002; Balestrieri et al. 2015). From the selected global model we built all possible candidate models deriving from covariate combinations and detection probability structures: the abundance of salamanders (λ) was modeled as a function of site-specific covariates; whereas, detection probability (r) was considered to be constant over time or survey-dependent. We selected models according to second-order Akaike's Information Criterion (i.e., AICc, Akaike 1973; Burnham and Anderson, 2002), and we considered that models with a ΔAICc > 2 show substantial differences (Burnham and Anderson 2002). We conducted all statistical analyses in R with the package Unmarked (Fiske and Chandler 2011) and AICcmodavg (Mazerolle 2011).

RESULTS

During our surveys we counted 255 plethodontid salamanders. We did not find any other family of salamanders inside the plots. Spearman's rank correlation coefficient for site covariates indicated lack of significant correlation ($\rho_s < 0.5$; $P > 0.050$), allowing us to use all covariates in model building procedure. Among the global model candidate set, the model with NB distribution and $K = 45$ resulted had the best fit ($P = 0.225$) and lower over dispersion (*c-hat* = 1.01),

TABLE 1. List of best models laying within four points of AICc for fine-scale habitat selection in a population of the salamander *Speleomantes strinatii* living in a Mediterranean forest environment.

Model	Parameters	AICc	ΔAICc	AICc*wt*
r (Survey) λ (Aspect + Runoff + Rock)	11	572.27	0.00	0.20
r (Survey) λ (Aspect)	9	573.18	0.91	0.13
r (Survey) λ (Aspect + Runoff)	10	573.35	1.08	0.12
r (Survey) λ (Aspect + Runoff + Rock + Slope)	11	575.12	2.86	0.05
r (Survey) λ (Aspect + Runoff + Slope)	10	575.41	3.14	0.04
r (Survey) λ (Aspect + Slope)	9	575.43	3.16	0.04
r (Survey) λ (Aspect + Stream)	9	575.59	3.32	0.04
r (Survey) λ (Aspect + Runoff + Cliff)	10	575.88	3.61	0.03
r (Survey) λ (Aspect + Rock)	9	575.94	3.67	0.03
r (Survey) λ (Aspect + Runoff + Rock + Stream)	11	575.95	3.68	0.03

allowing us to employ this as the global model. From this global model we built 200 nested candidate models. From model selection, using AICc, we obtained a set of three models within a ΔAICc $\leq$ 2 range (Table 1).

Regarding the detection probability structure, all the best models accounted for survey-dependent detectability (mean = 0.34 ± [SD] 0.11; Table 2). Survey specific detection estimates were averaged by models laying within 2 points of AICc (Table 2). Ranking of variables based on their importance (*W*t), denoted as the sum of AICc weights (AICc*W*t) of models including the variable (Mazerolle et al. 2005), showed that Aspect was the most important variable in describing salamander abundance (W*t* = 0.45), followed by Water runoff (W*t* = 0.32) and Rock (W*t* = 0.20).

Averaged Beta estimates of abundance covariates show that Aspect, besides being the most important variable (Table 2), also had the strongest effect on describing salamander abundance (Beta estimate = 1.56; 95% CI, 1.02–2.09; Fig. 1), highlighting higher salamander estimates on north facing slopes. Also Rock had a fairly positive effect on salamander abundance (Beta estimate = 0.32; 95% CI, 0.04–0.6), while, conversely, Water Runoff (Fig. 2) denoted a strong negative effect on salamander density (Beta estimate = -0.8; 95% CI, -1.54 to -0.06). The averaged mean estimate abundance per site λ was 10.3 individuals (95% CI, 7.5–13.2), while the total abundance for the study area, estimated from the best models, was 414 (95% CI, 167–519), resulting in a mean density of 0.86 (95% CI, 0.34–1.08) salamanders/m^2.

DISCUSSION

Understanding habitat selection by animal populations is relevant to understanding ecology and in species conservation (Manly et al. 2002). The association of a species with specific environmental conditions within a habitat type influences population density and dynamics, dispersion of individuals, survival and reproductive rates, source and sink dynamics, and also evolutionary trajectories (for a review see Pulliam and Danielson 1991; Clobert et al. 2001, 2012). A main finding of our study is that, after correcting for detection probabilities,

TABLE 2. Model-averaged estimates (95% Confidence Intervals) of abundance and detection parameters, and estimates of covariate coefficients (Beta) for a population of the salamander *Speleomantes strinatii* living in a Mediterranean forest environment. The acronym Wt = weights of variables calculated as the sum of AICc*Wt* of models including the variable.

Detection probabilities (*r*)				
Survey 1	Survey 2	Survey 3	Survey 4	Survey 5
0.167 (0.111–0.244)	0.322 (0.236–0.422)	0.453 (0.362–0.422)	0.428 (0.337–0.524)	0.329 (0.242–0.429)

Covariate coefficient (Beta)		
Aspect	Water Runoff	Rock
1.56 (1.03–2.09) (Wt = 0.45)	-0.81 (-1.55– -0.07) (Wt = 0.32)	0.32 (0.04–0.60) (Wt = 0.20)

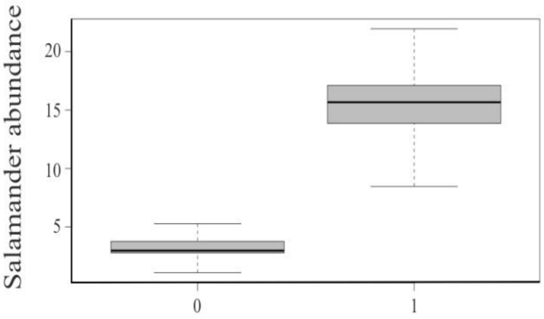

FIGURE 1. The estimated abundance of salamanders as a function of the prevalent aspect of the plot. 0 = South; 1 = North for a population of the salamander *Speleomantes strinatii* living in a Mediterranean forest environment.

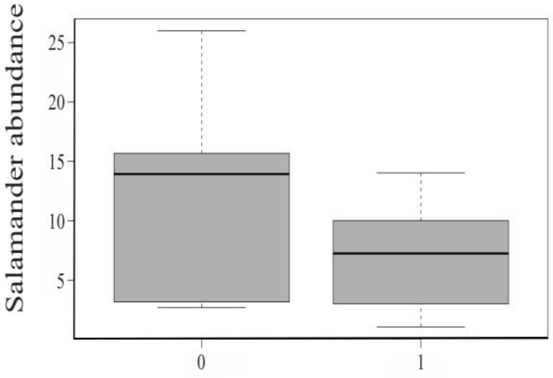

FIGURE 2. The estimated abundance of salamanders as a function of superficial Water Runoff for a population of the salamander *Speleomantes strinatii* living in a Mediterranean forest environment. 0 = No Runoff; 1 = Runoff.

the salamander *Speleomantes strinatii* has a patchy distribution on the forest floor and the distributio and relative abundance are influenced by three of the seven fine-scale habitat parameters selected *a priori* for ecological modelling. Two of these variables, north aspect and presence of rocks, positively affected salamander abundance, suggesting an influence of factors that provide high humidity on the forest floor, favorable shelters, and high prey availability (Jaeger 1980; Mathis 1990; Grover 1998). Conversely, an increase of water runoff on the forest floor negatively affected the density of salamanders.

Previous studies demonstrated the negative effects of water runoff on salamander populations living in freshwater habitats such as streams and ditches, because water flow increases stream bank instability and water turbidity (e.g., Orser and Shure 1972). However, the influence of this factor on salamander ecology has never been analyzed in detail in terrestrial ecosystems. Possibly, large volumes of rain water running off the soil surface removes leaf litter, displaces logs, and creates areas of bare soil that are unsuitable habitats for salamanders. In any case, all these parameters seem related to the physiological constraints of plethodontid salamanders and in particular to the retention of water, as reported in the North American species *Plethodon albagula* (Peterman and Semlitsch 2013).

To date, European plethodontids, habitat selection has been evaluated by modelling presence/absence data only (Ficetola et al. 2012; Manenti 2015 Lunghi et al. 2015), without estimating and accounting for their variation in abundance over the landscape. However, information on variation in abundance may be required to obtain more precise estimates on the use of available resources. This is especially true when the species is sampled in optimal or sub-optimal habitats, where its detection is relatively high and shows only a limited range of variation in abundance, as was the case of our study.

Finally, our density estimate of the forest population of *S. strinatii* (0.86 salamanders/m^2) is similar that reported for this species by Salvidio (1998; 0.8 salamanders/m^2, range 0.6–1.0). Salvidio (1998) estimated salamander density by temporary removal sampling, a technique that may be considered a generalization of a capture-mark-recapture method, where the probability of recapture is zero and the population closed (White et al. 1982). This result suggests efficiency of repeated count data analyzed with N-mixture models to estimate abundance of European plethodontids in forest environments and should also be experimented in other environments as for example underground habitats accessible to humans.

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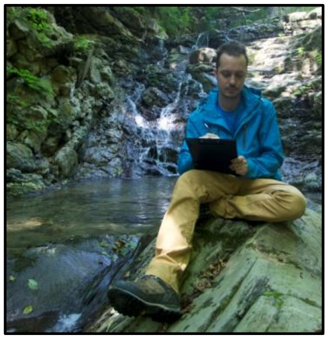

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