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Author(s): Tiffany D. Biagas, Alexander S. Hall, Alexis L. Ritzer, and Benjamin A. Pierce
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TIME OF DAY DOES NOT AFFECT DETECTION IN VISUAL-ENCOUNTER SURVEYS OF A SPRING-DWELLING SALAMANDER, *EURYCEA NAUFRAGIA*

Tiffany D. Biagas, Alexander S. Hall, Alexis L. Ritzer, and Benjamin A. Pierce*

Department of Biology, Southwestern University, Georgetown, TX 78626
**Correspondent: pierceb@southwestern.edu*

Abstract—During 10-weeks, visual-encounter surveys were conducted on the Georgetown salamander *Eurycea naufragia* at a spring pool in Williamson County, Texas. There was no significant difference in number of salamanders observed or percentage of objects used as cover between surveys in mornings and afternoons. Number of salamanders detected and percentage of objects used as cover were not correlated significantly with any environmental variable that was assessed. These results suggest that visual-encounter surveys conducted during daylight hours are not biased by time of day.

Resumen—Durante un periodo de 10 semanas se realizaron muestreos visuales de la salamandra de Georgetown, *Eurycea naufragia*, en un manantial en el condado de Williamson, Texas. No hubo diferencias significativas en el número de salamandras observadas o el porcentaje de objetos utilizados como escondite entre los muestreos realizados durante las mañanas o las tardes. No existió ninguna correlación entre el número de salamandras encontradas ni el porcentaje de objetos utilizados como escondite con las otras variables ambientales que fueron medidas. Estos resultados sugieren que los muestreos visuales realizados durante el día no están sesgados entre las diferentes horas del día.

Salamanders are important components of many spring and headwater-stream communities and they often are the most abundant vertebrates in these habitats (Burton and Likens, 1975; Peterman et al., 2008) where they play significant roles as predators and prey. Stream and spring salamanders are important indicators of health of ecosystems (Welsh and Ollivier, 1998) and numerous studies have demonstrated that these animals are sensitive to effects of urbanization (e.g., Orser and Shure, 1972; Bowles et al., 2006; Miller et al., 2007), construction of roads (Welsh and Ollivier, 1998; Ward et al., 2008), and harvesting of timber (Corn and Bury, 1989; Lowe and Bolger, 2002). Because many species of these salamanders are of conservation concern (Chippindale and Price, 2005), they often are included in watershed-assessment programs and are frequently the focus of research and monitoring (e.g., Jung et al., 2000).

Different techniques have been used to assess diversity and abundance of stream and spring salamanders, including dip netting (Nowakowski and Maerz, 2009), quadrat sampling (Jung et al., 2000), electroshocking (Sepulveda and Lowe, 2009), funnel trapping (Griffiths, 1985; Willson and Dorcas, 2004), sampling with leaf-litter bags (Chalmers and Droege, 2002; Waldron et al., 2003), and visual-encounter surveys (Crump and Scott, 1994; Jung et al., 2000). Visual-encounter surveys have become widely employed because they require minimal equipment, they have low impacts on target species, and they can be used for many species and in a variety of habitats. This technique allows researchers to visually detect presence of salamanders, often accompanied by actively searching potential objects in the water that are used as cover (Barr and Babbitt, 2001; Quinn et al., 2007; Marsh, 2009).

Previous studies have examined accuracy and precision of different visual-encounter surveys, compared visual-encounter surveys to other methods, or both (Barr and Babbitt, 2001; Jung et al., 2000; Quinn et al., 2007; Marsh, 2009; Mackey et al., 2010). A few studies investigated effects of environmental and sampling variables, such as season (e.g., Orser and Shure, 1975) and multiple observers (e.g., Marsh, 2009), on results of visual-encounter surveys. A criticism of traditional visual-encounter surveys is that they often fail to account for variation in probability of detection (Mackenzie et al., 2006; Mazerolle et al., 2007). One potential source of variation in probability of detection is time of day when the survey is conducted. Some stream salamanders are nocturnal (Orser and Shure, 1975; Petranka, 1984), but visual-encounter surveys often are conducted during daylight hours. If species are nocturnal, surveys conducted during morning might yield more observations than surveys in afternoon. We focused our research on the

Georgetown salamander *Eurycea naufragia,* a threatened species endemic to the San Gabriel River drainage in central Texas (Chippindale et al., 2000; Pierce et al., 2010). We investigated the effect of time of day and several environmental variables on number of salamanders observed during visual-encounter surveys.

MATERIALS AND METHODS—Weekly visual-encounter surveys were conducted at a permanent spring on the North San Gabriel River fed by the Edwards Aquifer in Williamson County, Texas. The area sampled was the first 5 m of the spring run, a well-delineated rectangular area consisting of ca. 30 m^2 of wetted surface area. Previous research (Pierce et al., 2010) indicated that the majority of salamanders at this site occur within this segment of the spring run. The area sampled contained riffles and pools; substrate was varied, consisting mostly of silt, gravel, and large limestone rocks.

We conducted a total of 10 weeks of surveys during 16 September–20 November 2009. One morning and one afternoon survey were conducted during each of the 10 weeks. Weekly morning and afternoon surveys were separated by 48–52 h. We randomly chose morning or afternoon for the first survey of the week and surveyed during the alternative time for the second survey. Morning surveys were conducted at 0730–0930 h and afternoon surveys at 1600–1800 h. The survey team was the authors, who conducted surveys in a consistent manner. To avoid bias by observers, two members of the team were assigned randomly to each survey, with the constraint that no person conducted both morning and afternoon surveys in the same week.

During each survey, observers overturned all submerged and partially submerged objects that could provide cover for a salamander. Potential cover included rocks, leaves, and woody debris. Rocks that were heavy or embedded deeply within the substrate were not overturned. For each survey, we recorded number of salamanders observed, number of overturned objects, percentage cloud cover, temperature of water (°C), specific conductivity (μS), dissolved oxygen (mg O_2/L H_2O), and depth of water (cm). Temperature of water and specific conductivity were measured with a YSI Model 30-10 FT conductivity meter and dissolved oxygen was measured with a YSI Model 550A meter (YSI, Inc., Yellow Springs, Ohio). During each survey, depth of water was measured with a ruler at the same location within the pool. Percentage cloud cover was estimated visually. Each salamander observed was assigned to one of three size classes based on a visual estimate of total length from tip of snout to tip of tail: <2.5, 2.5–5.1, or >5.1 cm.

We used paired *t*-tests to compare number of salamanders observed and percentage of objects used as cover during morning and afternoon surveys. To test for order effects (e.g., reduced number of salamanders on the surface due to multiple surveys in a relatively short time), we also compared mean number of salamanders observed and percentage of objects used as cover in the first and second surveys of the week. We used Pearson correlations to assess influence of environmental variables (temperature of water, specific conductivity, dissolved oxygen, depth of water, and percentage cloud cover) on number of salamanders observed and on percentage of objects used as cover. Statistical analyses were performed using SPSS (SPSS 13.0 for Windows, release 13.0.1; SPSS, Inc., Chicago, Illinois).

RESULTS—Mean (± *SD*) number of salamanders observed during morning (6.40 ± 3.37 salamanders; $n =$ 10 surveys) was the same as mean number observed in afternoon (6.40 ± 3.17 salamanders; $n = 10$ surveys). In addition, mean percentage of objects used as cover ($n =$ 10 surveys) did not differ between morning (2.14 ± 1.06 objects) and afternoon (2.20 ± 1.16 objects; $t = -0.131$, $df = 9$, $P = 0.899$). Across all surveys, number of salamanders observed was correlated strongly with percentage of objects used as cover ($r = 0.92$, $P < 0.001$).

We detected no order effect between morning or afternoon surveys. There was no significant difference between mean number of salamanders observed in the first or second survey of the week ($t = -0.924$, $df = 9$, $P =$ 0.379), or between mean percentage of objects used as cover in the first and second surveys of the week ($t =$ -0.971, $df = 9$, $P = 0.357$).

Ranges of environmental variables during the study were: temperature of water, 21.1–21.3°C; specific conductivity, 550–662 μS; dissolved oxygen, 6.44–7.56 mg O_2/L H_2O; depth of water, 12.9–24.10 cm; percentage cloud cover, 0–100%). No correlation between number of salamanders observed and environmental variables was significant.

DISCUSSION—Visual-encounter surveys commonly are used for assessing and monitoring stream and spring salamanders (Crump and Scott, 1994; Jung et al., 2000). Because these surveys are based on visual detection of salamanders, environmental factors that alter behavior of salamanders, visual acuity of observer, or both of these, may influence probability of detection. Studies have compared results of surveys conducted at night with those conducted during daylight hours. For example, Orser and Shure (1975) reported that mark-recapture surveys of the dusky salamander *Desmognathus fuscus* in a spring-fed stream in Georgia yielded higher estimates of size of populations when conducted at night than when surveys were conducted during daylight hours. They determined that the different estimates were largely the result of capturing more adults at night; numbers of juveniles were similar during night and day. Similarly, Petranka (1984) observed that larvae of the northern two-lined salamander *Eurycea bislineata* were under rocks and leaf litter during day, but emerged from cover at night and moved about in the open. Petranka (1984) observed that larvae fed continuously during night and day, suggesting that lack of movement during daylight was a predator-avoidance response. These studies suggest that, at least for some species, visual-encounter surveys conducted during nighttime may yield higher counts than those conducted during daylight. We know of no study that examined whether visual-encounter surveys of stream and spring salamanders conducted during daylight are affected by time of day. Most surveys are conducted during daylight and monitoring and research programs frequently combine and compare surveys conducted at different times of day.

Because some stream and spring salamanders are more active at night (Orser and Shure, 1975; Petranka, 1984), we hypothesized that surveys conducted during morning might yield higher counts than those in afternoon. However, our results demonstrated that time of day had no effect on number of salamanders observed. Average number of salamanders observed in morning and afternoon surveys was the same. Percentage of objects used as cover by salamanders also was similar in morning and afternoon surveys. Although our sample was not large, the degree of similarity of results for morning and afternoon surveys suggests that increasing the sample would have a negligible effect. Our results suggest that visual-encounter surveys of *E. naufragia* are not biased by time of day, allowing researchers to combine data from visual-encounter surveys taken during various daylight hours.

We detected no association between number of salamanders observed or percentage of objects used as cover with temperature of water, dissolved oxygen, specific conductivity, depth of water, or percentage cloud cover. The site where our study was conducted is a spring-fed pool with relatively constant temperature and chemistry of water. For other sites and species, where more environmental variation occurs, these parameters might play a larger role in abundance of salamanders. A limitation of our study was the relatively short time of sampling (10 weeks). Absence of significant correlations between environmental variables and number of salamanders may be related to brevity of our study.

Traditional visual-encounter surveys have been criticized because often they do not account for probability of detection, which may affect inferences about size and presence of populations (Mazerolle et al., 2007). Although we surveyed all available habitats within the spring pool, the extent to which salamanders may move into or out of the study area and access the subterranean aquifer is not known. Our objective, however, was not to draw inferences about size of population, but rather to determine whether time of day and environmental factors affect detection of salamanders in visual-encounter surveys.

Eurycea naufragia is a permanently neotenic species, and most salamanders we observed were adults (we rarely encountered juveniles in our surveys). Whether juveniles exhibit a similar lack of diurnal variation in detection is unknown. Orser and Shure (1975) detected no difference in numbers of juvenile *D. fuscus* between day and night. However, in his study of *E. bislineata*, Petranka (1984) discovered that smaller larvae were more active during predawn and afternoon hours than adults, but the effect was not strong and diurnal activity was independent of size. Additional research about variation in diurnal activity of larval salamanders would be informative.

Our study was conducted near the end of a severe drought in central Texas. However, a large rainfall occurred near the beginning of our study and spring flow was high throughout our sampling. We focused on a single season, but diurnal activity of salamanders might vary seasonally, although Petranka (1984) noted no strong seasonal effect in his study of *E. bislineata*. Further studies of activity patterns of additional species of stream and spring salamanders are warranted.

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