

Distribution, habitat suitability, conservation state and natural history of endangered salamander *Bolitoglossa pandi*

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Background. Pandi's mushroom-tongue salamander *Bolitoglossa pandi* is one of the most threatened amphibians in South America, as well as a spotlight species on conservation Colombian agenda. Few studies have provided relevant information that helps Colombian government to formulate lines of action for its conservation. Given this dearth of information, its threat assessments have been based on very limited information.

Methods. We conducted surveys for salamanders in four municipalities of Cundinamarca-Colombia, using two approaches: visual encounter surveys (Guaduas and Villeta) and the basic sampling protocol for single-species occupancy modeling (Supatá and Venecia). A multivariate approach was employed to explore correlation between habitat structure and natural history traits, abundance, as well as detection / non-detection of *B. pandi*. We evaluated *B. pandi* activity pattern through kernel density curves for each sampling occasion and explored salamander abundance variability during their activity period by performing a nested ANOVA.

Results. We report the discovery of two new populations of *B. pandi* which represent the most northwestern records known for the distribution of this species. A significant correlation between body length, body mass and habitat structure were observed. Multivariate analyses indicated that leaf litter depth, environmental mean temperature, percent vegetation cover, and altitude were the habitat variables that explain 60.32% of *B. pandi* abundance variability, as well as being the main drivers of its optimal habitat. *B. pandi* exhibit an activity pattern characterized by two main activity peaks, in which niche partitioning was observed. We found a healthy, stable, highly dense, with high replacement rate between body size classes and constant recruitment rate population of *B. pandi* (>1300 individuals).

Discussion. Given the high habitat specificity of *B. pandi*, it is highly vulnerable to local changes on its habitat variables. Thus, we recommend that *B. pandi* is retained as Endangered (EN) on the IUCN Red List based on the IUCN Criterion B, given its restricted extent of occurrence (ca. 2,500 km²), as well as the ongoing habitat loss within its range due to agriculture, cattle ranching, logging, and urban development that constantly reduce its suitable habitat.

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endangered salamander *Bolitoglossa pandi***

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Abstract

Background. Pandi's mushroom-tongue salamander *Bolitoglossa pandi* is one of the most threatened amphibians in South America, as well as a spotlight species on the Colombian conservation agenda. Few studies have provided relevant information that helps the Colombian government to formulate lines of action for its conservation; so, given this dearth of information, its threat assessments have been based on very limited information.

Methods. We conducted surveys for salamanders in four municipalities of Cundinamarca–Colombia, using two approaches: visual encounter surveys (Guaduas and Villeta) and the basic sampling protocol for single-species occupancy modeling (Supatá and Venecia). A multivariate approach was employed to explore the correlation between habitat structure and natural history traits, abundance, as well as detection / non-detection of *B. pandi*. We evaluated the *B. pandi* activity pattern through kernel density curves for each sampling occasion and explored the salamander abundance variability during their activity period by performing a nested ANOVA.

Results. We report the discovery of two new populations of *B. pandi*, which represent the most northwestern records known for the distribution of this species. A significant correlation between body length, body mass, and habitat structure as was observed. Multivariate analyses indicated that leaf litter depth, environmental mean temperature, percent vegetation cover, and altitude were the habitat variables that explain the 60.32% of the *B. pandi* abundance variability, as well as the main drivers of its optimal habitat. *B. pandi* exhibits an activity pattern characterized by two main activity peaks, in which niche partitioning was observed. We found a healthy, stable, highly dense, with high replacement rate between body size classes and constant recruitment rate population of *B. pandi* (>1300 individuals).

Discussion. Given the high habitat specificity of *B. pandi*, the species is highly vulnerable to local changes on its habitat variables. Thus, we recommend that *B. pandi* is retained as Endangered (EN) on the IUCN Red List based on the IUCN Criterion B, given its restricted extent of occurrence (ca. 2,500 km²), as well as the ongoing habitat loss within its range due to agriculture, cattle ranching, logging, and urban development that constantly reduce its suitable habitat.

Introduction

Colombia is home to more than 850 species of amphibians, and much of this diversity is concentrated in the Andean Forest (Acosta-Galvis, 2019). Geographic distributions of fauna in this biome are often restricted, and also endemism is common (Lynch & Suárez-Mayorga, 2002). Andean forests are the most threatened habitats in the country due to the drastic transformation of native vegetation by urban growth, mining, agriculture, and cattle ranching (Etter, Andrade, Saavedra, & Cortés, 2018). As a

consequence, and given that the majority of threatened Colombian amphibian species are concentrated between 1800-3600 a.s.l., habitat loss is their shared major threat.

Among threatened Colombian amphibians, salamanders of the *Bolitoglossa* genus are one of the spotlight amphibian groups on Colombian conservation agenda. This genus is the most diverse and geographically widespread lineages of Plethodontidae salamanders inhabiting the Neotropics. Currently, *Bolitoglossa* comprises 131 species, 24 of which reside in Colombia across several types of tropical habitats (Wake, 2017; Acosta-Galvis, 2019). Many species of *Bolitoglossa* exhibit patterns of restricted geographic ranges [i.e. *Bolitoglossa capitata* (Brame & Wake, 1963b), *B. hypacra* (Brame Jr & Wake, 1962), *B. hiemalis* (Lynch, 2001), among others]. However, the high morphological crypsis and an incomplete understanding of the morphological variability among Andean *Bolitoglossa* species make suitable taxonomic decisions about their distributional ranges hard to achieve (Acosta-Galvis & Gutiérrez-Lamus, 2012).

The ecology and life history of *Bolitoglossa* salamanders are poorly understood compared to their Nearctic congeners; so less than 11.5% of *Bolitoglossa* species have been investigated with published research on their diet, reproduction, foraging activities, thermal ecology, ecological interactions, microhabitat use, or habitat preferences (Anderson & Mathis, 2006; Bruce, 1997; Cadenas, Pérez-Sánchez, Villa, & De Ascensão, 2009; Cruz, Galindo, & Bernal, 2016; del Río-Gracia, Serrano-Cardozo, & Ramírez-Pinilla, 2014; Houck, 1977; Jimenez, 1994; Ortega, Monares-Riaño, & Ramírez-Pinilla, 2009; Salgado-Aráuz, 2005; Sarmiento et al., 2011). Given this dearth of information, the conservation status of many of these species is Data Deficient (DD) or has been based on limited information.

According to the IUCN Red List of Threatened Species, *Bolitoglossa pandi* (Brame & Wake, 1963b) is Endangered B1ab (iii), based on its restricted geographical range; it was originally described based on a single specimen (holotype ZSZMH 2858, an adult female with SVL 50.4 mm, collected in 1913 by Wilhelm Frietsche) from the municipality of Pandi, Cundinamarca (exact locality unknown), in the cloud forests on the western slope of the Cordillera Oriental of Colombia. Subsequently, Hanken and Wake (1982) reported a second specimen 75km north (airline) from the type locality, in a bromeliad inside the primary cloud forest, near the municipality of Albán (Cundinamarca), at 2400 m a.s.l.

Acosta-Galvis and Rueda-Almonacid (2004) reported a third specimen (an adult female with snout-vent length (SVL) 44.7 mm, collected by Franz Kaston, ICN 45500) from nearby the type locality and described the associated habitat as relict wet areas covered by lush trees and shrubs. Acosta-Galvis and Gutiérrez-Lamus, (2012) included a new record for the Supatá region (adult male with SVL 37.63 mm, MUJ 7921) representing the northernmost record of the species. Hence, the known distribution of *B. pandi* includes four localities from the sub-Andean forests between 1300-2400 m a.s.l.,

throughout the western slopes of the Cordillera Oriental. Since its first extinction risk assessment was performed 14 years ago, few studies have provided relevant information for the formulation of strategies and an action plan for its conservation (del Río-García et al., 2014).

The aims in this study are: (1) to describe the geographic range extension of *B. pandi* along the western slopes of the Cordillera Oriental; (2) to explore the relationship between habitat structure and natural history traits of this species; (3) to describe its activity pattern and population structure; (4) to expand the knowledge of the variability of certain morphological characters and life-history traits of this poorly known species; (5) and to provide a conservation state reassessment of *B. pandi*.

Materials & Methods

Ethics statement

Sex was not determined on living salamanders due to the high risk of injury to the animal. Fieldwork was done under the scientific research non-commercial purpose permit of collection of wild specimens of biological diversity issued by the National University of Colombia (Research Project 38615), and the Colombian National Environmental Licensing Authority (ANLA) by resolution No. 0255 of 14 March 2014. This study was conducted following the Colombian animal welfare law and the collection of wild specimens of the biological diversity acts (Ley 1774, 2016; Decreto 1376, 2013), as well as considering the Universal Declaration on Animal Welfare (UDAW) endorsed by Colombia in 2007.

Study area

We searched for salamanders at nine localities in four municipalities of Cundinamarca located on the western slope of the Cordillera Oriental of Colombia: Guaduas, Supatá, Villeta, and Venecia (Fig. 1). Searches at each locality were carried out within an altitudinal gradient ranging from 1638 to 2315 m a.s.l (Table 1). The sampled area includes sub-Andean and Andean forests, as well as areas transformed by urban growth, agriculture, and cattle ranching. The sampling area is characterized by bimodal climate [high dry season (2nd half of Dec, Jan, Feb, 1st half of Mar); high rainy season (2nd half of Mar, Apr, May, Jun); low dry season (Jul, Aug, 1st half of Sept), low rainy season (2nd half of Sept, Oct, Nov, 1st half of Dec)]. We monitored the environmental temperature (ET) and relative humidity (RH) at sampling sites using thermo-hygrometers model EBI 20-TH1 Ebro®.

Sampling and data collection

We conducted surveys for salamanders using two approaches. First, during the rainy season in April–May 2013, we performed visual encounter surveys (VES; Crump & Scott, 1994) in three localities associated with cloud forests throughout an altitudinal gradient (1648–2002 m a.s.l.) at the municipalities of Guaduas and Villeta (Table 1). Two researchers surveyed day and night for five consecutive days investing a total of 100 h of sampling effort.

Second, we employed the basic sampling protocol described by Mackenzie et al., (2003) for single-species occupancy modeling in six localities, three in the municipality of Supatá and three in Venecia. We randomly selected a total of 296 quadrats (5 m x 5 m), which were located throughout an altitudinal gradient at each sampling locality (1600–2300 m a.s.l.), grouping the following vegetation covers: relicts of the Andean forest, restored the Andean riparian forest, grove pastures, pastures and roadsides (Table 1). During three sampling occasions (September–October 2017, March 2018, and July 2018), each quadrat was surveyed day and night for five consecutive days by ten researchers, resulting in a total of 2100 h of sampling effort. During each survey, the detection / non-detection of *B. pandi* specimens were recorded. When a salamander was present, we measured its perch height using a measuring tape (± 0.1 cm). Once salamanders were caught, we measured their weight (Mass g) with a Pesola® dynamometer of 50g (± 0.1 g) and took photographs to measure their body size [SVL = snout-vent length (mm), TL = Tail length (mm)] using the software Image–J version 1.52 (Schneider, Rasband, & Eliceiri, 2012). The sex was not determined on living salamanders due to the high risk of injury to the animal. All specimens of *B. pandi* were subsequently released near the quadrat in which they were sighted.

Based on 17 specimens collected, euthanized using lidocaine 2%, and fixed in 10% formalin (Chen & Combs, 2001), we described the morphological variability of *B. pandi*. We made a small incision in the groin region to identify the sex and sexual maturity through macroscopic observation of the gonads. All the morphological terminology employed follows several contributions (Brame Jr & Wake 1962, 1963; Acosta-Galvis & Restrepo 2001; Lynch 2001; Acosta-Galvis & Hoyos 2006; Acosta-Galvis & Gutierrez-Lamus 2012; Brcko, Hoogmoed, & Neckel-Oliveira, 2013; Bingham, Papenfuss, Lindstrand, & Wake, 2018). All these specimens were deposited in the amphibian collection at Instituto de Investigación de Recursos Biológicos Alexander von Humboldt, (IAvH-Am), as well as in the amphibian collection at the Instituto de Ciencias Naturales de la Universidad Nacional de Colombia (ICN).

Habitat structure data collection

We used the Point Intercept Method described by Elzinga, Salzer, and Willoughby (1998) for estimation of the percent vegetation cover. We grouped the plants into eight life forms: graminoids, forbs, palm trees, mosses, lichens, vines, shrubs, and trees. We

divided vegetation into layers: 0–0.1 m, 0.1–1 m, 1–1.5 m, 1.5–3 m, 3–5 m, 5–12 m. We also considered the cover of other abiotic items such as leaf litter, bare soil and bare rocks. We estimated the vegetation cover of each of the quadrants where salamanders were surveyed, employing a set of 15 intercept points distributed in three parallel lines of five points separated by one meter of distance. At each point, we used a sampling bar of 1.5 m to register the contact of the life forms of each vegetation layer below 1.5 m. This provided us with a 6.67% cover resolution by layer. We assessed the vegetation percentage cover of the vegetation layers above 1.5 m (mostly trees) using five intercept points: the corners of the quadrant and the central point of the third line. In this way, we reached a 20% cover resolution for upper layers.

Statistical analysis

We evaluated the association between the habitat structure and the natural history traits of *B. pandi* by multiple correlation analysis, with $P < 0.05$ as the significance criterion. The following variables were considered: SVL (mm), weight (g), perch height (mm), leaf litter depth (mm), vegetation layers, vegetation life form, vegetation percentage cover, and altitude. Using the habitat variables, we performed a principal component analysis (PCA) to explore which of these variables presents greater variability between quadrats with the detection / non-detection of *B. pandi* and, therefore, which of these are responsible for the differences observed between them. The variable suitability for PCA analysis was tested performing a Kaiser-Meyer-Olkin test ($KMO > 0.5$, $P < 0.05$). Afterward, a quadratic discriminant analysis was performed to determine which of the habitat variables have the greatest discrimination capacity between quadrants where *B. pandi* was detected or non-detected.

We assessed the variability in the salamander abundance observed in the Supatá population through multiple regression analysis. First, we considered the following habitat structure variables at each sampling quadrant: leaf litter depth (mm), vegetation layers, vegetation life form, vegetation percentage cover, altitude, temperature, and relative humidity of the environment. All variables were Ln-transformed previously to perform the statistical analysis.

Second, we evaluated assumptions of normality, autocorrelation, and homoscedasticity using Kolmogorov–Smirnov’s test, Durbin–Watson test and Breusch–Pagan test, respectively. Given that the p-value of the Durbin–Watson test can easily be less than 0.05 when data size is very large, we used the Durbin–Watson statistic test (DW) as an autocorrelation criterion. According to Durbin and Waston (1950), a DW of less than 1 indicates a strong positive autocorrelation, a DW greater than 4 indicates a strong negative autocorrelation, values between 1 and 3 suggest a moderate autocorrelation, and a value close to 2 means that there is no autocorrelation.

Third, we tested for multicollinearity between the aforementioned variables using the variance inflation factor (VIF) with a threshold of 10. Fourth, we selected the “best” regression model employing the Akaike Information Criterion (AIC; Akaike, 1973), considering that models with ΔAIC values of less than two are equally plausible (White & Burnham, 1999). Finally, we used the hierarchical partitioning method to evaluate the contribution of all the independent variables of the regression model (Chevan & Sutherland, 1991).

Activity pattern and population structure

We only assessed the activity pattern and the population structure of the *B. pandi* population at the Cuzcungos locality given the remarkable abundance observed (Table 1). We estimated the activity pattern through Kernel Density Curves for each sampling occasion and explored the salamander abundance variability during their activity period by performing a nested ANOVA. Hence, the activity period of *B. pandi* was divided into nine-time intervals (from H1 = 18:30–19:30, H2 = 19:31–20:30, H3 = ... until H9 = 02:31–03:30), and each salamander sighting was allocated into its respective interval. The sampling occasion was employed as the primary factor, and the time intervals as the secondary factor nested in the primary factor. We evaluated assumptions of normality and homogeneity of variances using a Shapiro-Wilk test and Levene test, respectively. Additionally, we analyzed the variability in observed body size over the nine-time intervals through a non-parametric ANOVA using a Kruskal-Wallis test (KW) as a measure of the central tendency of the samples (Sokal & Rohlf, 1981).

We used SVL as a descriptive variable of the population structure of *B. pandi*. We compared the variability in population structure between sampling occasions through a Wilcoxon test, with the null hypothesis that the median of the population structure is the same across all sampling occasions. According to the categories proposed by Acosta-Galvis and Gutierrez-Lamus (2012) and Del Río-Gracia et al (2014), as well as the reproductive stage of the collected salamanders, the population was divided into body size classes as follows: neonates (≤ 23 mm), juveniles (24–30 mm), and adults (≥ 30 mm).

All statistical analyses were performed using the software Rwizard 4.3 (Guisande-González C, Vaamonde-Liste, A. Barreiro-Felpeto, 2014) and the following R packages: car (Fox & Weisberg, 2019), hier.part (Walsh & Macnally, 2015), lawstat (Hui, Gel, & Gastwirth, 2008), nortest (Gross & Ligges, 2015), overlap (Meredith & Ridout, 2017) stat (R Core Team, 2012) and usdm (Naimi, Hamm, Groen, Skidmore, & Toxopeus, 2014).

Results

Geographic distribution

We found a total of 34 individuals of *B. pandi* in three new localities, extending the geographical range of the species by 96.5 Km (airline) northwest from the type-locality, and 33.6 Km (airline) west from the northernmost locality in the municipality of Supatá (Fig. 1). These new localities belong to the municipalities of Guaduas and Villeta in the department of Cundinamarca. All the salamanders in the new localities were found at night, within the understory of oak groves dominated by ferns. The salamanders were sighted in different vertical strata ranging from leaf litter, where they remained hidden, to shrubby substrates up to 2.5 m. Also, two salamanders were found in ecotonal areas associated with sugar cane crops and rangeland areas for livestock.

Associations of habitat structure and natural history traits

We observed a significant association between habitat structure and natural history traits. Snout-vent length, tail length, and mass were significantly associated with all the habitat structure variables assessed, but less so with the vegetation layers. SVL, TL, and mass showed a negative correlation with leaf litter and altitude (Table 2). In contrast, SVL and mass showed a positive correlation with perch height and percent vegetation cover (Fig. 2). The square coefficients obtained indicate that the variability of SVL, TL, and mass correlated in 19.3%, 16.8%, and 18.6%, with habitat structure variables respectively.

Similarly, the habitat structure variables explain the detection or non-detection of *B. pandi* throughout the sampling quadrants. The first two components of the PCA analysis explained 62.41% of the variability observed. The habitat structure variability was clustered in two groups associated with the detection or non-detection of *B. pandi* (Fig. 3A). These groups were moderately overlapped in multivariate spaces, but they were strongly differentiated by altitude, leaf litter depth, and percent vegetation cover: the presence of *B. pandi* was positively correlated with quadrants highly structured and with a depth of leaf litter (Table 3). The quadratic discriminant analysis confirms that the detection or non-detection of *B. pandi* depends on habitat variables. The cross-validation percentage was 92.6%, indicating that the quadrants in which *B. pandi* was detected can be clearly distinguished by habitat variables such as vegetation layers, leaf litter and environmental mean temperature (Fig. 3B).

Results of the multiple regression analysis showed that leaf litter depth, environmental mean temperature, percent vegetation cover, and altitude were the variables that contributed the most to explaining the observed variability in the abundance of *B. pandi* ($R^2 = 60.32\%$, $P < 0.001$). These habitat variables also composed the “best” fitted regression model (Table 4). However, this model showed a moderate autocorrelation ($DW = 1.10$), signifying that the autocorrelation is very small and the variance explained by the habitat variables may be close to 60.32% (Fig. 4).

Activity pattern

Bolitoglossa pandi is completely nocturnal, its activity period extends throughout the night, beginning from 18:30 h until 05:00 h. The environmental temperature and relative humidity recorded during this activity period ranged from 12.6–25.6 °C (\bar{x} = 15.3), 73.8–98.8 RH (\bar{x} = 80.3). The activity peaks were all subject to environmental influences showing a significant association with the local weather conditions. The observed salamander abundance was strongly and positively correlated with the environmental temperature ($R_{ET} = 0.251$, $P < 0.001$), whereas the moderately and negatively were correlated with the relative humidity ($R_{RH} = -0.174$; $P = 0.017$). Despite the sampling occasion, we observed two main activity peaks, the first from 20:30 h to 21:30 h, and the second from 23:30 h to 00:30 h (Fig. 5). Results of the nested ANOVA indicated that salamander abundance between these peaks was significantly different ($F_{9-259} = 4.57$, $P < 0.001$; Table 5), being more abundant in the second activity peak during the second and third sampling occasions, and the opposite during the first sampling occasion (Fig. 6A).

Likewise, the observed body length of the salamanders showed significant differences between the two activity peaks ($K_{8df} = 99.70$, $P < 0.001$, Fig. 6B), suggesting niche partitioning between body size classes. Most of the salamanders observed during the first activity peak had SVL > 30 mm (juveniles and adults), whereas the salamanders observed at the second activity peak had SVL < 27 mm (juveniles and neonates). However, in contrast with the differences in observed abundance between activity peaks, the variation between salamander SVL were consistent across the three sampling occasions.

Population structure

A total of 1391 individuals of *B. pandi* were observed throughout the study in the Supatá population, exhibiting a population density ranging from 0.04– 1.44 individuals/m². The population structure showed significant differences between sampling occasions, suggesting a high replacement rate between body size classes (Table 6). Regardless of the variability observed in the population structure of *B. pandi*, the population is mostly dominated by juveniles and neonates which represent between 34–64% of the individuals (Fig. 7). Neonates were observed across all sampling occasions, indicating constant recruitment. During the second sampling occasion, a notorious bias in the SVL median value toward small body length salamanders was observed, suggesting that the major recruitment peak occurs during March (Fig 7B). Adults were conspicuous throughout all sampling occasions, although predominantly in the first sampling occasion.

Morphological variability

Bolitoglossa pandi is a tiny species recognizable by SVL = 13.4–51 mm ($n = 1034$), 13.4–48mm adult males ($n = 7$), and SVL 14–51 mm adult females ($n = 8$). Extensively

webbed hands and feet with third toes and fingers triangular; ventral surfaces of digit tips without terminal flattened tubercles; snout short and rounded in the lateral profile; head length 4.9–10.1 mm; head width 5.1–7.9 (n =17); snout rounded in dorsal view irregular white spots, cream nasolabial grooves, and edges of the lips irregularly dark brown with irregular light spotting (Fig. 8A–B); protruding eyes on dorsal view, brown iris with black reticules (Fig. 8B); well-defined post-cephalic constriction; ventral surfaces (preserved); brown or dark grey with numerous tiny cream guanophores (Fig. 8C); inverted bracket shaped scapular spots (Fig. 9F); males have white testes.

Bolitoglossa pandi can be distinguished from its Colombian congeners by having an extensive webbing of hands and feet (versus absence of extensive webbing of hands and feet in *B. adspersa*, *B. hiemalis*, *B. hypacra*, *B. palmata*, *B. ramosi*, *B. savagei*, *B. tamaense*, *B. tatamae*, *B. walkeri* and *B. valleculea*); also, It can be distinguished from species with extensive webbing as *Bolitoglossa lozanoi* and *B. nicefori*, in having more protruding eyes, and longer and triangular third finger. *B. pandi* also differs from *B. biseriata* and *B. silverstonei* by having a dark brown or dark grey ventral surface with irregular white dots (versus cream ventral surface with brown suffusions and pits in *B. biseriata* and *B. silverstonei*), brown-reddish iris with black reticules (versus golden with brown pits in *B. biseriata* and *B. silverstonei*), and white testes in adult males (versus black mesorchium in testes of adult males of *B. biseriata*). It also differs from *B. medemi* by the absence of digital depressions in digital tips on the fingers and toes (present in *B. medemi*). *B. pandi* has an upper lip with irregular light spotting (versus uniform in *B. guaneae*). *B. pandi* can be differentiated from *B. altamazonica* and *B. leandrae* by having extensive interdigital webbing with a longer and triangular third finger (complete webbing and tips rounded in *B. altamazonica* and *B. leandrae*). *B. pandi* is morphologically very similar to *B. phalarosoma*, but it differs by having a dark brown or dark grey ventral surface with diffuse white pits and some white blotches (versus usually light brown ventral surfaces, with some irregular cream spots in *B. phalarosoma*), plantar and palmar regions in ventral dark brown or dark grey view (versus cream in *B. phalarosoma*). *B. pandi* also differs from *B. capitana* by having smaller adult size with a longer and triangular third finger (versus rounded third finger in *B. capitana*) and less head width.

Color variability

Bolitoglossa pandi exhibited a wide polychromatic variability ranging from a uniform dorsal color pattern of different shades of light to a dark brown one (Fig. 9A); one reddish-brown pattern that can have diffused grey or dark blotches scarcely distinguishable to the dorsolateral region (Fig. 9B), or an ochre pattern, with some diffuse irregular dark brown, yellow or cream spots (Fig. 9C–E). Some specimens exhibit an exceptionally diffuse band in the paravertebral region covering almost the entire dorsal surface or including a dark brown inverted triangle shape in the interorbital

region (Fig. 9 F–G). The caudal region is highly variable, ranging from uniform reddish-brown or completely ochre segments, with very small scattered white pits (Fig. 9 A,B, D) to irregular cream, yellow or orange patches (Fig. 9C, D), and very small or irregular longitudinal black spots (Fig. 9F–G). The distal end of the tail becomes uniform light brown and has cream blotches towards the proximal region in some individuals (Fig. 9B). The cephalic region is dark brown with some white spots with irregular pits up to the supralabial region in lateral view (Figure 8B); a loreal region with ochre patches and light brown iris with black reticles; nasolabial projections are creams; the ventrolateral surface is dark brown. Ventral surfaces are dark brown with some cream and white blotches with scattered pits; the mental and gular surfaces are uniformly dark brown or dark grey with some irregular white circular spots bordering the maxillary region. In adult males the mental gland is light brown; the palmar and plantar surfaces are always dark brown or dark grey.

Discussion

The new localities added to *Bolitoglossa pandi* distribution and the detailed examination of the Supatá population have allowed not only the expansion of knowledge about the distribution and natural history traits of this species but also the reassessment of its conservation status and the validation of its characters for the taxonomic identification among its congeners. The apparent local endemism of *B. pandi* is the result of a general misunderstanding due to the examination of restricted samples, insufficient sampling effort, lack of comparisons with the relevant type material, and the incomplete knowledge of the variability of certain morphological characters of *Bolitoglossa* salamanders (Wake, 2017).

Since Brame and Wake's (1963a) original description of *B. pandi*, several characters such as dorsal surfaces color (Figure 9) have had conflicting or ambiguous diagnostic characters (Acosta-Galvis and Gutiérrez-Lamus, 2012). However, our findings significantly increased the understanding of the morphological variability of *B. pandi*. The chromatic variations reported here contrast with the original description (Brame and Wake, 1963b) because these have not been described in life (and the holotype is poorly preserved), and evidence of a broad intrapopulational polychromatism was not available due to restricted sampling. Our findings allow clarification of taxonomical misidentifications in the literature. For example, Acosta-Galvis and Rueda-Almonacid (2004), during the first threat assessment of *B. pandi*, included erroneously a picture of *B. walker* as the species' portrait. The color and morphological description provided here comes from living specimens obtained in the municipalities of Guaduas, Supatá, and Villeta.

Neotropical salamanders have been considered inconspicuous amphibians of the Andean Forest (Brame and Wake, 1963; Gibbons, 1983; Barrio-Amoros and Fuentes, 1999; Acosta-Galvis and Gutiérrez-Lamus, 2012). Nevertheless, our results challenge

this general assumption and provide support to previous studies relating specific environmental conditions with high abundance and density of Andean salamander populations (Cadenas, Pérez-Sánchez, Villa, & De Ascensão, 2009; Ortega et al., 2009; Gutiérrez-Lamus, Lynch, & Martínez-Villate, 2011; del Río-García et al., 2014). Altitude (1700-2000 m. a.s.l), leaf litter depth (> 6 cm), vegetation layers (> 5 vegetation strata), and environmental mean temperature (16–19 °C) arise as the “best” predictor variables of presence, abundance and population density of *B. pandi*. Given the *B. pandi*’s arboreal habits, diet preferences, as well as thermoregulation behavior of Bolitoglossines (del Río-García et al., 2014; Cruz et al 2016; Mendieta Donaire et al., 2019);, the selection of these predictor variables by the regression models was expected.

Likewise, highly homogenous sampling quadrants in which pastures, graminoids, bare soil, and rocks were dominant, as a result of human activities, showed the lowest or no detection grade of *B. pandi*. Conversely, sampling quadrants in which *B. pandi* was present were positively correlated with highly structured habitats and deep leaf litter, characteristics associated with relicts of conserved Andean forest or highly restored Andean riparian forest (Rangel-Ch, Lowy, & Aguilar, 1997; Alvear, Betancur, & Franco-Rosselli, 2010; Quintero Vallejo, Benavides, Moreno, & Gonzalez-Caro, 2017). This result indicates that *B. pandi* requires Andean forests in a good conservation state and a narrow range of environmental conditions. This strong link between *B. pandi* and a narrow range of environmental conditions has been observed in other Andean salamanders such as *B. adspersa*, *B. altamazonica*, *B. nicefori*, and *B. orestes* (Valdivieso & Tamsitt, 1965; Cadenas et al., 2009; Gutiérrez-Lamus et al., 2011; Leenders & Watkins-Colwell, 2013). The detection and high local abundance of these species are strongly correlated with the same habitat variables that promoting the detection and abundance of *B. pandi*. Thus, the deep dependency of a narrow environmental range of conditions makes the Andean Bolitoglossines highly vulnerable to local changes in habitat variables.

Our study provided evidence for the first time of intraspecific niche partitioning on Bolitoglossine salamanders. Niche partitioning has been plenty reported on populations of Nearctic salamander species (Jaeger & Gergits, 1979; Wicknick, 1995; Arif, Adams, & Wicknick, 2007; Jaeger, Gollman, Anthony, Gabor, & Kohn, 2016). Evidence of Nearctic salamander’s niche intraspecific partitioning has been explained as a life history strategy to maximize foraging success, predator avoidance, and mating success (Jaeger & Gergits, 1979; Holomuzki, 1986; Cloyed & Eason, 2017). Nevertheless, our data are insufficient to infer any fundamental ecological trade-off between body size and microhabitat use, activity and vulnerability to predation, or activity and foraging success. Future experimental studies must be addressed to understand how niche intraspecific partitioning is acting through environmental heterogeneity in the ecology and evolution

of the South American Bolitoglossines. Also, future conservation efforts must consider this life history strategy into conservation interventions.

Conclusions

Our result agrees with the general pattern observed in other amphibian taxa in which habitat disturbance driven by human activities has deleterious effects on presence or population densities given their high habitat specificity (Lips, 1998; Collins & Storfer, 2003). Habitat loss continues protruding as the central threat for *B. pandi*. This fact has significant importance in conservation issues because the Andean forest is one of the most threatened ecosystems by human activities in Colombia (Etter et al., 2018). Therefore, despite that we directly observed a healthy and stable salamander population that showed high population density, high replacement rate between body size classes and constant recruitment in the municipalities of Supatá, Guaduas, and Villeta, we recommend that *B. pandi* is retained as Endangered (EN) on the IUCN Red List based on the IUCN Criterion B, given its restricted extent of occurrence (ca. 2,500 km²), as well as the ongoing habitat loss within its range due to agriculture, cattle ranching, logging, and urban development that constantly reduce its suitable habitat.

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References

- Acosta-Galvis, A.R., & Rueda-Almonacid, J. V. (2004). Salamandra de Pandi *Bolitoglossa pandi*. In J. V. Rueda-Almonacid, J. D. Lynch, & A. Amezcuita (Eds.), *Libro rojo de anfibios de Colombia* (pp. 206–209). Bogotá D.C.

- 501 Acosta-Galvis, A.R., & Gutiérrez-Lamus, D. L. (2012). A new species of salamander
502 (Bolitoglossa: Plethodontidae) from the Cordillera Oriental of the Colombian Andes.
503 *Papéis Avulsos de Zoologia (São Paulo)*, 52(18), 201–218.
504 <https://doi.org/10.1590/s0031-10492012001800001>
- 505 Acosta-Galvis, Andrés Rymel. (2019). Lista de los Anfibios de Colombia: Referencia en
506 línea. Retrieved February 12, 2019, from <https://www.batrachia.com>
- 507 Akaike, H. (1973). Information theory and an extension of the maximum likelihood
508 principle. In B. Petrovand & F. Caski (Eds.), *Proceeding of the isecnd international*
509 *Symposium on Information Theory* (pp. 1–451). Budapest: Akademiai Kiado.
- 510 Alvear, M., Betancur, J., & Franco-Rosselli, P. (2010). Floristic diversity and structure of
511 andean forests remnants near to los nevados national park, central colombian
512 andes. *Caldasia*, 32(1), 39–63.
- 513 Anderson, M. T., & Mathis, A. (2006). Diets of Two Sympatric Neotropical Salamanders,
514 *Bolitoglossa mexicana* and *B. rufescens*, with Notes on Reproduction for *B.*
515 *rufescens*. *Journal of Herpetology*, 33(4), 601. <https://doi.org/10.2307/1565576>
- 516 Arif, S., Adams, D. C., & Wicknick, J. A. (2007). Bioclimatic modeling, morphology, and
517 behavior reveal alternative mechanisms regulating the distributions of two
518 parapatric salamander species. *Evolutionary Ecology Research*, 9(5), 843–854.
- 519 Bingham, R. E., Papenfuss, T. J., Lindstrand, L., & Wake, D. B. (2018).
520 Phylogeography and Species Boundaries In the *Hydromantes shastae* Complex,
521 With Description of Two New Species (Amphibia; Caudata; Plethodontidae).
522 *Bulletin of the Museum of Comparative Zoology*, 161(10), 403–427.
523 <https://doi.org/10.3099/mcz42.1>
- 524 Brame, A. H., & Wake, D. B. (1963a). Contributions to the Salamanders of South
525 America. *Contributions In Science. Natural History Museum of Los Angeles County*,
526 69, 1–72.
- 527 Brame, A. H., & Wake, D. B. (1963b). The Salamanders of South America, (69), 1–71.
- 528 Brame Jr, A. H., & Wake, D. B. (1962). A new plethodontid salamander (genus
529 *Bolitoglossa*) from Venezuela with redescription of the Ecuadorian *B. palmata*
530 (Werner). *Copeia*, 1962(1), 170–177.
- 531 Brcko, I. C., Hoogmoed, M. S., & Neckel-Oliveira, S. (2013). Taxonomy and distribution
532 of the salamander genus *Bolitoglossa* Duméril, Bibron & Duméril, 1854 (Amphibia,
533 Caudata, Plethodontidae) in Brazilian Amazonia. *Zootaxa*, 3686(4), 401–431.
534 <https://doi.org/10.11646/zootaxa.3686.4.1>
- 535 Bruce, R. C. (1997). Society for the Study of Amphibians and Reptiles Life History
536 Attributes of the Salamander *Bolitoglossa colonnea*, 31(4), 592–594.
- 537 Cadenas, D. A., Pérez-Sánchez, A. J., Villa, P. ., & De Ascensão, A. A. (2009). Relative
538 Abundance, Habitat Use and Diet of *Bolitoglossa orestes* (Urodela: Plethodontidae)
539 in a Venezuelan Andean Cloud Forest. *ECOTRÓPICOS*, 22(2), 99–109.
- 540 Chen, M. ., & Combs, C. (2001). An alternative killing agent for amphibians.
541 *Herpetological Review*, 32(2), 93–94.
- 542 Chevan, A., & Sutherland, M. (1991). Hierarchical partitioning. *The American*
543 *Statistician*, 45, 90–96. <https://doi.org/1080/00031305.1991.10475776>
- 544 Cloyed, C. S., & Eason, P. K. (2017). Niche partitioning and the role of intraspecific
545 niche variation in structuring a guild of generalist anurans. *Royal Society Open*
546 *Science*, 4(3). <https://doi.org/10.1098/rsos.170060>

- Collins, J., & Storfer, A. (2003). Global amphibian declines: sorting the hypotheses. *Diversity and Distributions*, 9(2003), 89–98.
- Crump, M. L., & Scott, N. J. (1994). Visual encounters surveys. In W. Heyer, M. Donnelly, R. MacDiarmid, L. Hayeck, & M. Foster (Eds.), *Measuring and monitoring biological diversity, standard methods for amphibians* (pp. 354–352). Washington: Smithsonian Institution Press.
- Cruz, E. X., Galindo, C. A., & Bernal, M. H. (2016). Dependencia térmica de la salamandra endémica de Colombia *Bolitoglossa ramosi* (Caudata, Plethodontidae). *Iheringia. Série Zoologia*, 106(0). <https://doi.org/10.1590/1678-4766e2016018>
- del Río-Gracia, J. S., Serrano-Cardozo, V. H., & Ramírez-Pinilla, M. P. (2014). Diet and Microhabitat Use of *Bolitoglossa cf. pandi* (Caudata: Plethodontidae) from the Cordillera Oriental of Colombia. *South American Journal of Herpetology*, 9(1), 52–61. <https://doi.org/10.2994/sajh-d-13-00031.1>
- Elzinga, C. L., Salzer, D. W., & Willoughby, J. W. (1998). *Measuring and monitoring plant populations*. Denver: U.S. Dept. of the Interior, Bureau of Land Management.
- Etter, A., Andrade, Á., Saavedra, K., & Cortés, J. (2018). Actualización de la Lista Roja de los Ecosistemas Terrestres de Colombia. In G. I. Moreno, L. A., Rueda, C. y Andrade (Ed.), *Biodiversidad 2017. Estado y tendencias de la biodiversidad continental de Colombia* (p. 19). Investigación de Recursos Biológicos Alexander von Humboldt, Instituto de Ciencias Naturales de la Universidad Nacional de Colombia, WWF Colombia y Universidad de Manizales.
- Fox, J., & Weisberg, S. (2019). *An R Companion to Applied Regression* (Third edit). Thousand Oaks: Sage.
- Gross, J., & Ligges, U. (2015). nortest. Viena. Retrieved from <https://cran.r-project.org/web/packages/nortest/index.html>
- Guisande-González C, Vaamonde-Liste, A. Barreiro-Felpeto, A. (2014). Rwizard. Vigo: Universidad de Vigo. Retrieved from <http://www.ipez.es/RWizard/>
- Gutiérrez-Lamus, D. L., Lynch, J. D., & Martínez-Villate, G. C. (2011). Population estimators and adult sex ratio for a population of *Bolitoglossa altamazonica* (Caudata: Plethodontidae). *Basic and Applied Herpetology*, 25(2011), 43–54. <https://doi.org/10.11160/bah.11008>
- Hanken, J., & Wake, D. B. (1982). Genetic differentiation among plethodontid salamanders (genus *Bolitoglossa*) in Central and South America: implications for the South American invasion. *Herpetologica*, 32(2), 272–287.
- Holomuzki, J. R. (1986). Predator Avoidance and Diel Patterns of Microhabitat Use by Larval Tiger Salamanders. *Ecology*, 67(3), 737–748.
- Houck, L (1977). Reproductive biology of a Neotropical Salamander, *Bolitoglossa rostrata*. *Copeia*, 1977(1), 70–83.
- Hui, W., Gel, Y. R., & Gastwirth, J. L. (2008). Lawstat: An R package for law, public policy, and biostatistics. *Journal of Statistical Software*, 28(3). <https://doi.org/10.18637/jss.v028.i03>
- Jaeger, R.G., Gollman, B., Anthony, D. D., Gabor, C. R., & Kohn, N. (2016). *Behavioral Ecology of the Eastern Red-backed Salamander: 50 Years of Research*. New York: Oxford University Press.
- Jaeger, Robert G., & Gergits, W. F. (1979). Intra- and interspecific communication in salamanders through chemical signals on the substrate. *Animal Behaviour*,

- 27(PART 1), 150–156. [https://doi.org/10.1016/0003-3472\(79\)90134-9](https://doi.org/10.1016/0003-3472(79)90134-9)
- Jimenez, C. E. (1994). Utilization of Puya dasylirioides (Bromeliaceae: Pitcairnoideae) as foraging site by Bolitoglossa subpalmata (Plethodontidae: Bolitoglossini). *Revista De Biología Tropical*, 42(3), 703–710. Retrieved from <http://search.ebscohost.com/login.aspx?direct=true&db=boh&AN=BACD199598404277&site=ehost-live>
- Leenders, T. A. A. M., & Watkins-Colwell, G. J. (2013). Morphological and behavioral adaptations in Bolitoglossa colonnea (Caudata, Plethodontidae) in relation to habitat use and daily activity cycle. *Phyllomedusa: Journal of Herpetology*, 2(2), 101. <https://doi.org/10.11606/issn.2316-9079.v2i2p101-104>
- Lips, K. (1998). Decline of a tropical amphibian fauna. *Conservation Biology*, 12(1), 106–117.
- Lynch, J. D. (2001). A small amphibian fauna from a previously unexplored páramo of the Cordillera Occidental in western Colombia. *Journal of Herpetology*, 35(2), 226–231.
- Lynch, J. D., & Suárez-Mayorga, A. M. (2002). Análisis Biogeográfico de los Anfibios Paramunos. *Caldasia*, 24(2), 471–480. <https://doi.org/10.1007/s11051-017-4041-1>
- Mendieta Donaire, R. A., Zolotoff Pallais, J. M., Chung, J. S., Cobos Rizo, M. A., De los Santos Rosales, M., Jun Lee, J., ... Casco-Robles, M. M. (2019). Constricted spatiotemporal foraging of the regenerating salamander, Bolitoglossa mombachoensis. *Ecosphere*, 10(10). <https://doi.org/10.1002/ecs2.2897>
- Meredith, M., & Ridout, M. (2017). Overview of the overlap package. *R Project*. <https://doi.org/10.1016/j.contraception.2006.09.005>
- Naimi, B., Hamm, N. A. S., Groen, T. A., Skidmore, A. K., & Toxopeus, A. G. (2014). Where is positional uncertainty a problem for species distribution modelling? *Ecography*, 37(2), 191–203. <https://doi.org/10.1111/j.1600-0587.2013.00205.x>
- Ortega, J. E., Monares-Riaño, J. M., & Ramírez-Pinilla, M. P. (2009). Reproductive Activity, Diet, and Microhabitat Use in Bolitoglossa nicefori (Caudata: Plethodontidae). *Journal of Herpetology*, 43(1), 1–10. <https://doi.org/10.1670/07-250r2.1>
- Quintero Vallejo, E., Benavides, A. M., Moreno, N., & Gonzalez-Caro, S. (2017). *Bosques Andinos, estado actual y retos para su conservación en Antioquia*. (E. Quintero Vallejo, A. M. Benavides, N. Moreno, & S. Gonzalez-Caro, Eds.). Medellín: Fundación Jardín Botánico de Medellín Joaquín Antonio Uribe.
- R Core Team. (2012). R: A language and environment for statistical computing. Viena: R Foundation for Statistical Computing. Retrieved from <http://www.r-project.org/>
- Rangel-Ch, O. J., Lowy, P. ., & Aguilar, M. (1997). Distribución de los tipos de vegetación en las regiones naturales de Colombia. In *Colombia diversidad biótica II: tipos de vegetación en Colombia* (pp. 383–402). Bogotá D.C: Instituto de Ciencias Naturales-Universidad Nacional de Colombia, Ministerio de Medio Ambiente.
- Salgado-Aráuz, H. . (2005). Caracterización de la población y del hábitat de la salamandra endémica (Bolitoglossa mombachoensis) en la Reserva Natural Volcán Mombacho. *Encuentro*, 37(73), 77–86.
- Sarmiento, J. F. M., Neckel-Oliveira, S., Fáveri, S. B., Lima, A. A., Galatti, U., Lima, C., & Suárez, P. (2011). Reproductive Traits of the Brazilian Salamander Bolitoglossa

- 639 paraensis (Urodela: Plethodontidae). *Copeia*, 2011(3), 457–462.
- 640 <https://doi.org/10.1643/ce-08-219>
- 641 Schneider, C. ., Rasband, W. ., & Eliceiri, K. (2012). ImageJ. *Nature Methods*, 9(7),
- 642 671–675. https://doi.org/10.1007/978-1-84882-087-6_9
- 643 Sokal, R. ., & Rohlf, R. (1981). *Biometry: The principles and practice of statistics in*
- 644 *biological research* (First edition). San Francisco: W.H. Freeman.
- 645 Valdivieso, D., & Tamsitt, J. R. (1965). Reproduction in a Neotropical Salamander,
- 646 *Bolitoglossa adspersa* (Peters). *Herpetologica*, 21(3), 228–236.
- 647 Wake, D. B. (2017). Persistent Plethodontid Themes: Species, Phylogenies, and
- 648 Biogeography. *Herpetologica*, (July). [https://doi.org/10.1655/herpetologica-d-16-](https://doi.org/10.1655/herpetologica-d-16-00065)
- 649 [00065](https://doi.org/10.1655/herpetologica-d-16-00065)
- 650 Walsh, C., & Macnally, R. (2015). Hierarchical Partitioning. Viena. Retrieved from
- 651 <https://cran.r-project.org/web/packages/hier.part/index.html>
- 652 White, G. ., & Burnham, K. (1999). Program MARK: survival estimation from 594
- 653 populations of marked animals. *Bird Study*, 4(sup1), S120–S139.
- 654 <https://doi.org/10.1080/00063659909477239>
- 655 Wicknick, J. A. (1995). *Interspecific competition and territoriality between a widespread*
- 656 *species of salamander and a species with a limited range*. University of
- 657 Southwestern Louisiana.

Figure 1

Distribution range of the poorly-known salamander *Bolitoglossa pandi*

New Northwestern records for the known distribution of *Bolitoglossa pandi* (IAvH-Am 10303-4, IAvH-Am 10305-8). Pentagon with a white square inside: La Esmeralda (municipality of Villeta), black star: Granada (municipality of Villeta), broad diamond: El Trigo (municipality of Guaduas). Sampled localities in the municipality of Supatá. Black square: San Marcos, asterisk: Cuzcungos Natural Reserve, solid pentagon: “El Alto” Vereda Monterey. Sampled localities in the municipality of Venecia. Narrow diamond: Buenos Aires, invert triangle: El Alto de la Chorrera, triangle: El Mirador.

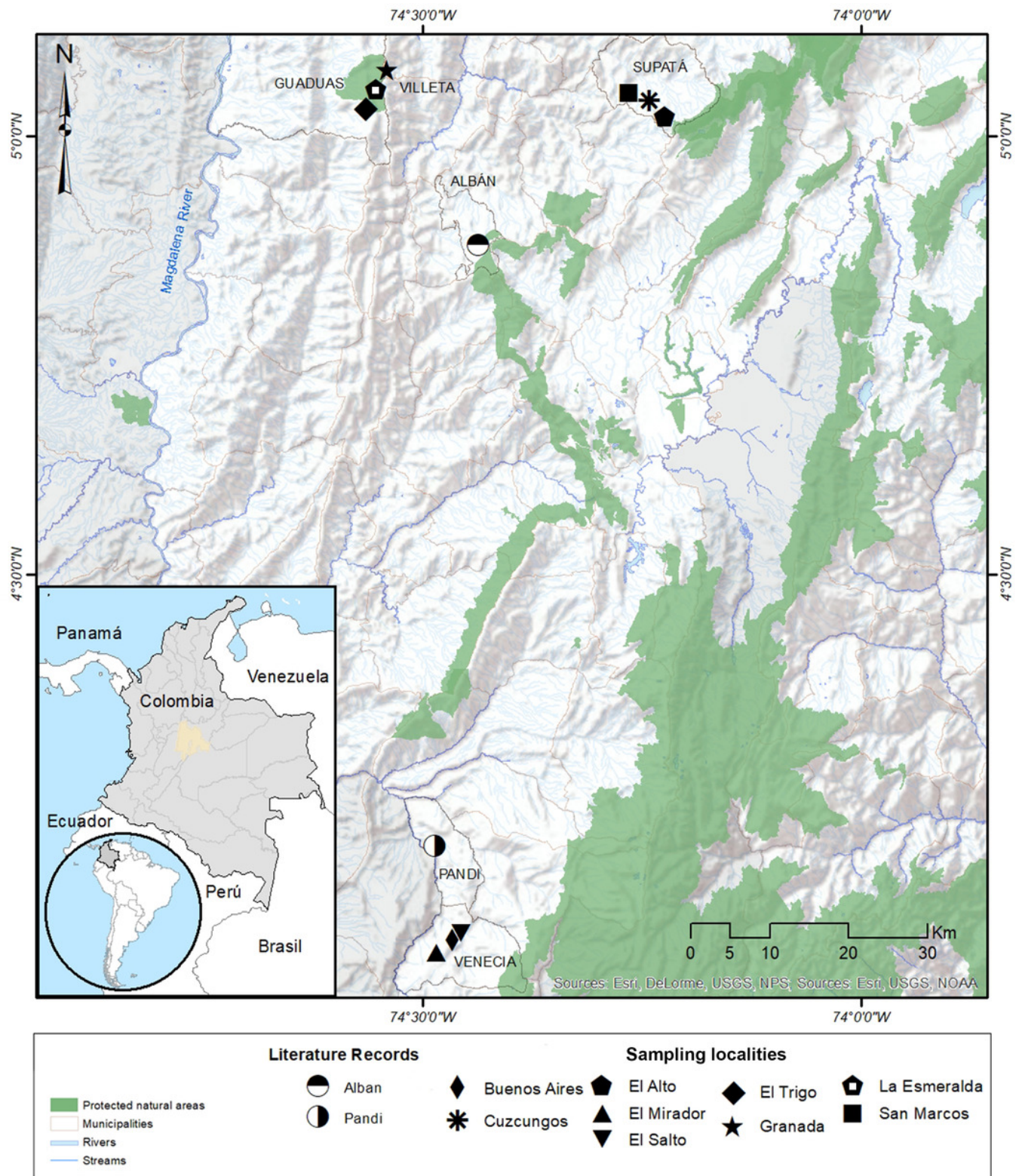


Figure 2

Scatterplot depicting the positive correlation between body length, weight and perch high

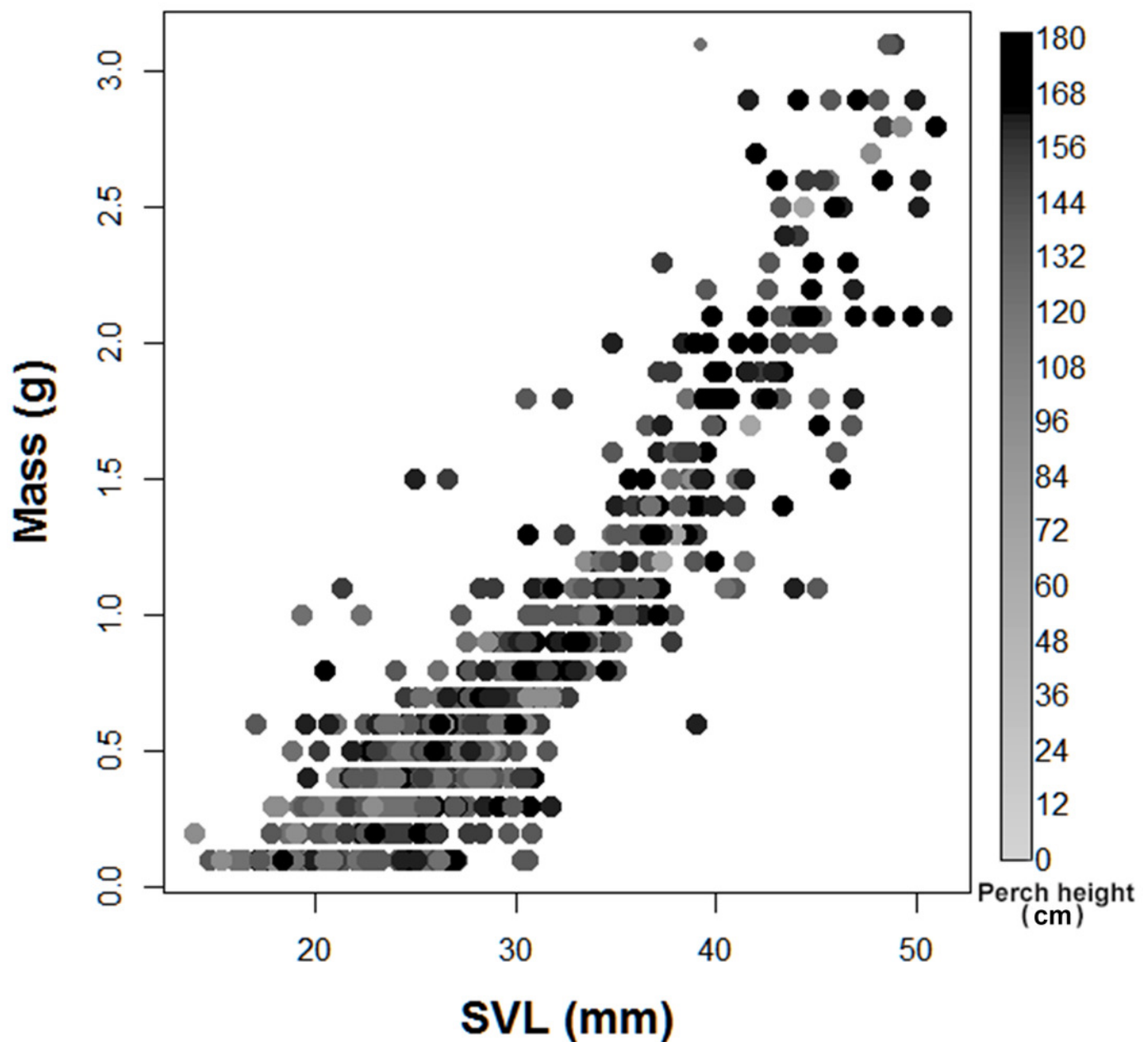


Figure 3

Main drivers of the *Bolitoglossa pandi* optimal habitat

(A) Principal component analysis showing the observed variability in habitat structure attributes between quadrats with detection /non-detection of *Bolitoglossa pandi*. PC1 = First principal component (38.8%). PC2 = Second principal component (23.6%). Veg.layers= Vegetation layers. Veg.per.cover = Vegetation percentage cover. Mean Temp = Environmental mean temperature. Black squares = detection of *Bolitoglossa pandi*. Grey dots = Non-detection of *Bolitoglossa pandi*. The inner ellipse represents 0.5 of significance; the outer ellipse represents 0.95 of significance. (B) Quadratic discriminant analysis depicting the habitat variables that have the greatest discrimination capacity between quadrants where *Bolitoglossa pandi* was detected or non-detected. The length of the vector denotes the discrimination capacity of each habitat variable.

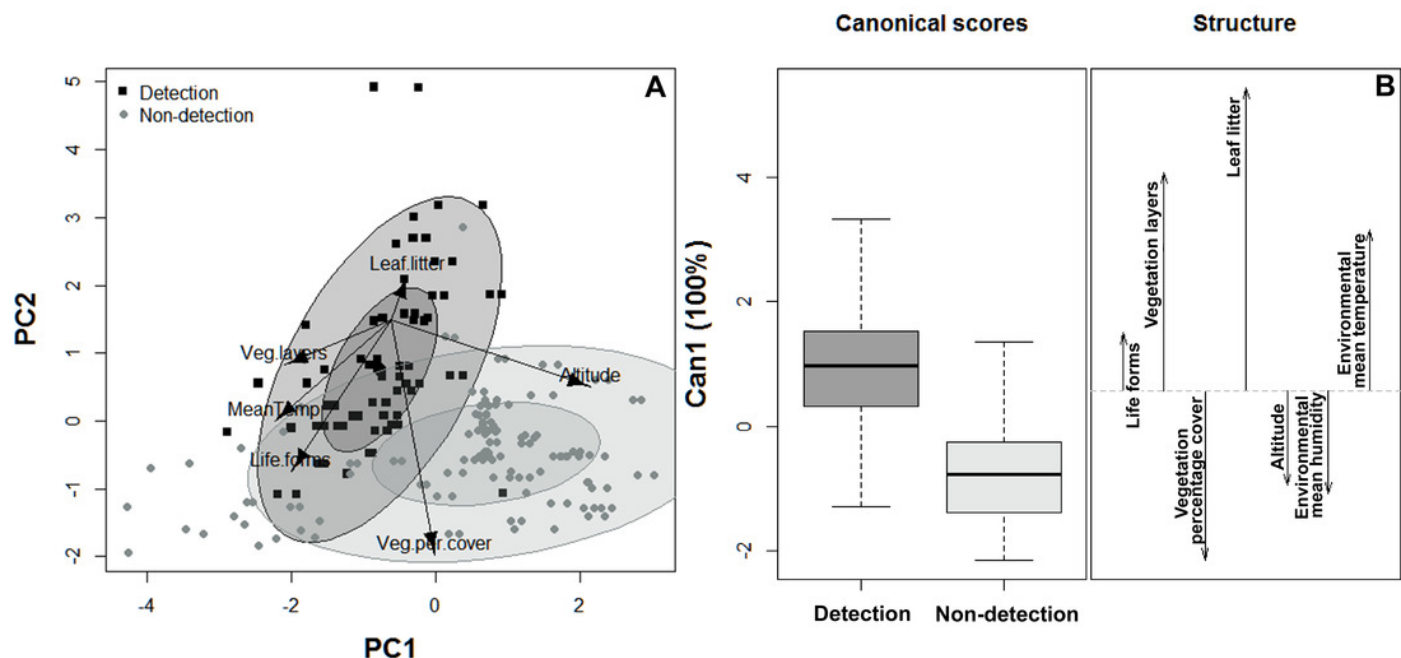


Figure 4

The “best” multiple regression model that relates *Bolitoglossa pandi* abundance and habitat structure variables

(A) Plots depicting linear regressions between *B. pandi* and habitat structure variables assessed. (B) Relative contributions of the variables that compose the “best” regression model.

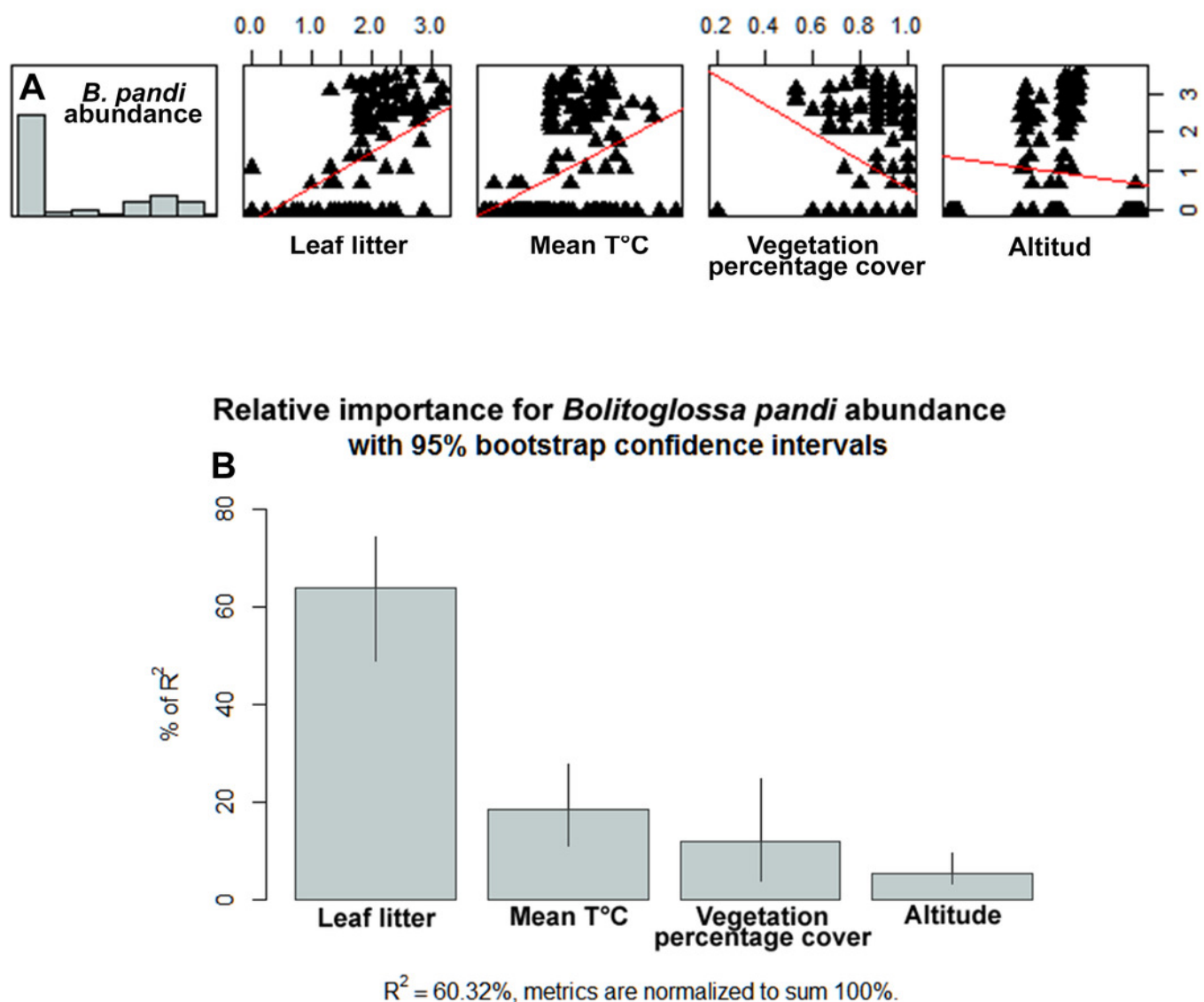


Figure 5

Circular kernel density models showing overall daily activity patterns of *Bolitoglossa pandi* during each sampling occasion.

The small vertical bars on the bottom depicts the independent detections observed during day and night for five consecutive days by ten researchers. (A) First sampling occasion, September–October 2017. (B) Second sampling occasion, March 2018. (C) Third sampling occasion, July 2018.

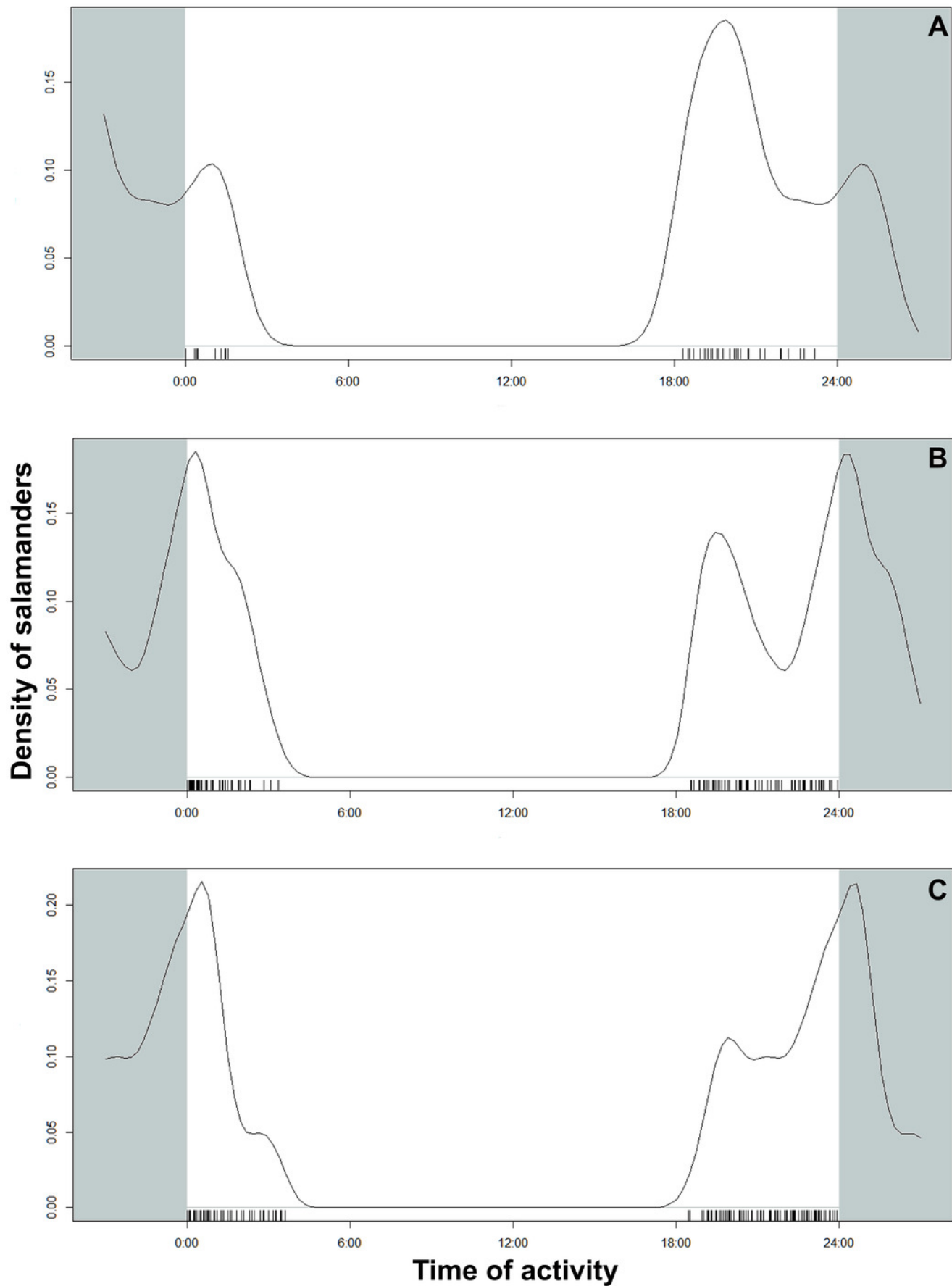


Figure 6

Abundance and body size variability during the activity of *Bolitoglossa pandi*.

(A) Nested ANOVA depicting the abundance variability among the activity peaks observed.

(B) Kruskal-Wallis test depicting niche partitioning between the body size classes

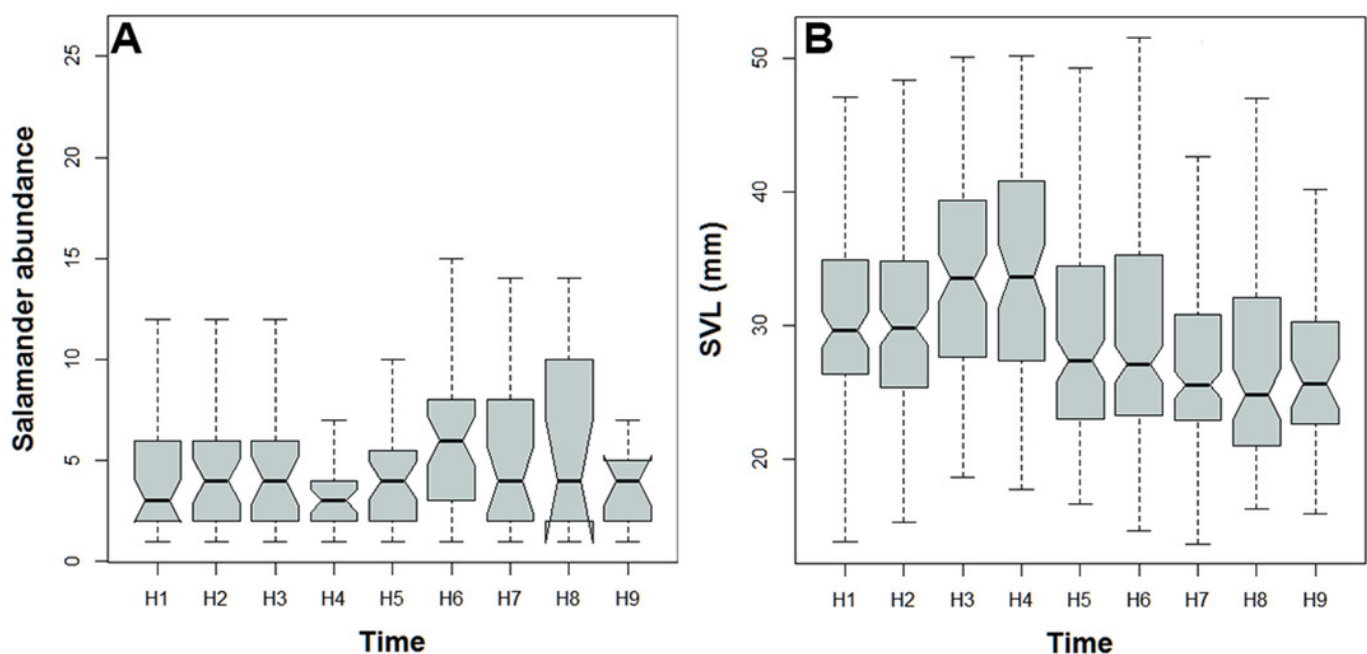


Figure 7

Population structure of the *Bolitoglossa pandi* population at the Cuzcungos locality across the sampling period.

(A) First sampling occasion, September–October 2017. (B) Second sampling occasion, March 2018. (C) Third sampling occasion, July 2018.

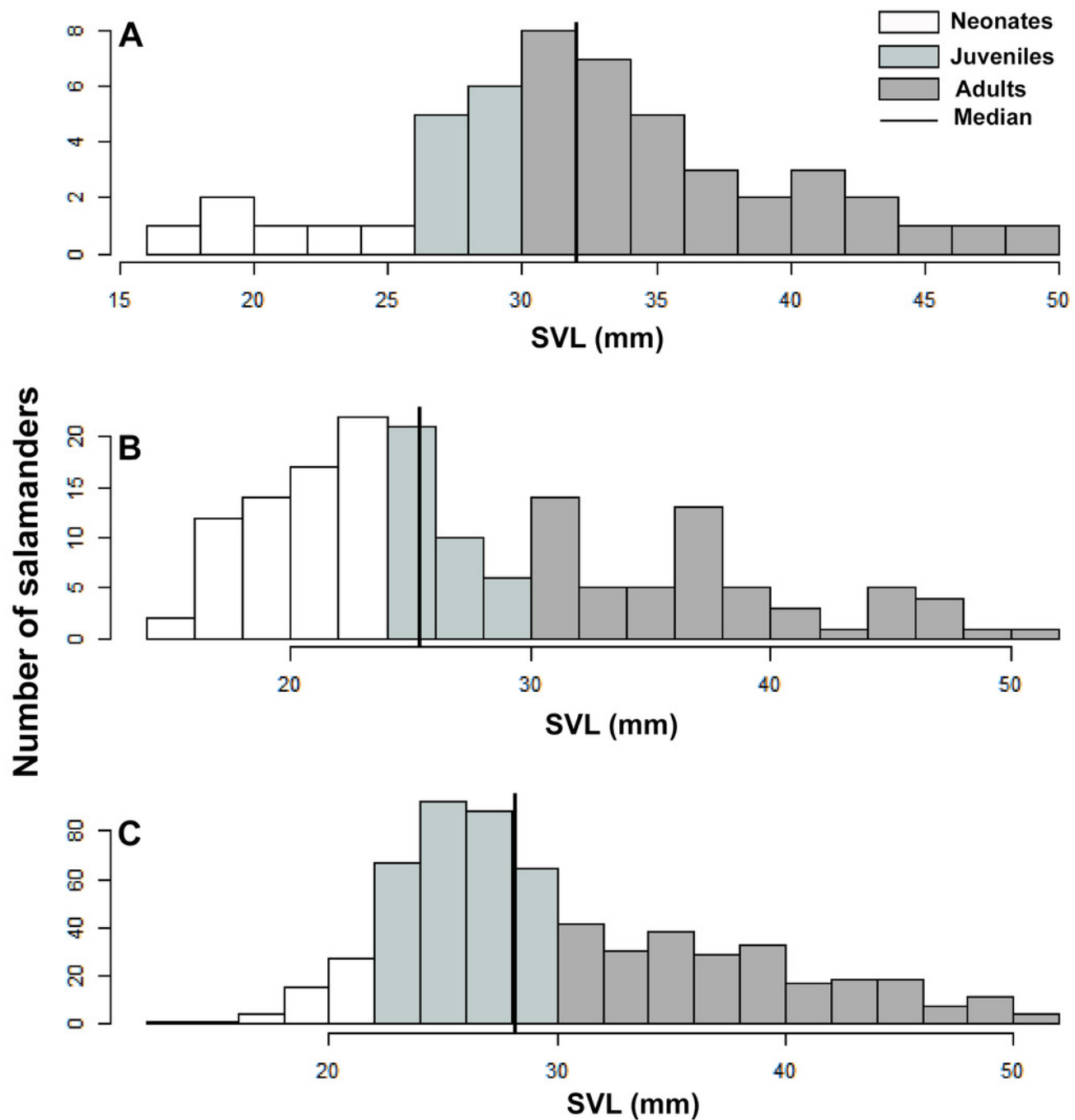


Figure 8

Color variability on ventral surfaces in preserved specimens of *Bolitoglossa pandi*.

(A) ventral surface of the head (ICN 45000; Pandi, Cundinamarca). (B) Lateral view of the lip of the edge showing the color pattern irregularly dark brown with irregular light spotting (ICN 58501, in life. Supatá, Cundinamarca). (C) ventral surfaces uniformly dark brown or dark grey with some irregular white circular spots (ICN 45000; Pandi, Cundinamarca. ICN 58502, Supatá, Cundinamarca).

A



B



C



5 mm

Figure 9

Polychromatic variability observed in living specimens of *Bolitoglossa pandi* obtained in the municipalities of Guaduas, Supatá, and Villeta.

(A) uniform dorsal color pattern. (B) reddish-brown dorsal color that can have diffused grey or dark blotches scarcely distinguishable to the dorsolateral. (C-E) dorsal surfaces exhibiting ochre pattern, with some diffuse irregular dark brown, yellow or cream spots. (F-G) Diffuse dorsolateral band in the paravertebral surfaces covering almost the entire dorsal surface or including a dark brown inverted triangle shape in the interorbital region.



Table 1 (on next page)

Sampling localities on the western slope of the Cordillera Oriental of Colombia.

VES: Visual encounter surveys (Crump and Scott 1994). QRS: Quadrats. N: Number of quadrants randomly selected.

Municipalities Vereda/locality	Latitude North	Longitude West	Altitude (m) a.s.l	Sampling protocol	<i>B. pandi</i> specimens observed	<i>B. pandi</i> specimens collected
Supatá						
Vereda Las Lajas/Monterey	From 5° 2' 18.5"	74° 14' 6.6"	2119–2315	QRS (N = 45)	1	0
	To 5° 1' 59.9"	74° 14' 3.5"				
Vereda Las Lajas/ Cuzcungos Natural Reserve	From 5° 2' 27.7"	74° 14' 32.2"	1931–2016	QRS (N = 43)	1390	12
	To 5° 2' 27.2"	74° 14' 27.9"				
Vereda San Marcos	From 5° 2' 58.9"	74° 15' 53.7"	1743–1773	QRS (N = 28)	0	0
	To 5° 2' 58.3"	74° 15' 54.9"				
Villeta						
Vereda La Esmeralda	5° 3' 17.9"	74° 32' 49.1"	1996	VES	15	6
El Tigre site	5° 2' 9.2"	74° 33' 48.8"	1650	VES	12	0
Guaduas						
Vereda Granada	5° 4' 1.49"	74° 32' 59.6	1816	VES	7	0
Venecia						
Vereda Buenos Aires	From 4° 4' 57.9"	74° 27' 57.7"	1809–2128	QRS (N = 90)	0	0
	To 4° 4' 44.1"	74° 27' 33.3"				
Vereda El Diamante/El salto de la Chorrera	From 4° 5' 24.6"	74° 27' 25.5"	1637–1735	QRS (N = 36)	0	0
	To 4° 5' 21.0"	74° 27' 33.2"				
Vereda El Alto/ Road Venecia- Cabrera	From 4° 4' 5.2"	74° 29' 6.9"	2034–2114	QRS (N = 54)	0	0
	To 4° 4' 11.1"	74° 28' 54.8"				

Table 2(on next page)

Multiple correlation analysis.

Bold values denote statistical significance at the $p < 0.05$ level.

Habitat structure variable	SVL (mm)	TL (mm)	Mass (g)	Perch height (mm)	Life forms	Percentage of vegetation cover	Vegetation layers	Leaf litter	Altitude (m) a.s.l
SVL (mm)	*****	0.890	0.901	0.316	0.216	0.287	0.006	-0.306	-0.155
TL (mm)	<0.001	*****	0.820	0.281	0.241	0.262	-0.037	-0.306	-0.126
Mass (g)	<0.001	<0.001	*****	0.288	0.194	0.271	0.018	-0.266	-0.134
Perch height (mm)	<0.001	<0.001	<0.001	*****	0.084	0.141	0.029	-0.058	0.004
Life forms	<0.001	<0.001	<0.001	0.026	*****	0.490	-0.039	-0.448	-0.043
Percentage of vegetation cover	<0.001	<0.001	<0.001	<0.001	<0.001	*****	0.063	-0.396	-0.152
Vegetation layers	0.883	0.322	0.627	0.445	0.300	0.096	*****	0.203	0.133
Leaf litter	<0.001	<0.001	<0.001	0.125	<0.001	<0.001	<0.001	*****	0.092
Altitude (m) a.s.l	<0.001	0.001	<0.001	0.906	0.260	<0.001	<0.001	0.015	*****

Table 3(on next page)

Principal component correlation matrix.

KMO: Kaiser-Meyer-Olkin test. BST: Kaiser-Meyer-Olkin test

Habitat structure variables	Life forms	Vegetation percentage cover	Vegetation layers	Leaf litter	Altitude (m) a.s.l	Environmental mean temp °C	KMO	BST
Life forms	*****	0.481	0.263	0.002	-0.384	0.368		
Vegetation layers	<0.001	*****	-0.078	0.340	-0.364	0.410		
Percentage of vegetation cover	<0.001	0.285	*****	-0.274	0.045	-0.061		
Leaf litter	0.979	<0.001	<0.001	*****	0.149	0.060	0.6	<0.0001
Altitude (m) a.s.l	<0.001	<0.001	0.541	0.040	*****	-0.629		
Environmental mean temp °C	<0.001	<0.001	0.401	0.409	<0.001	*****		

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Table 4(on next page)

Multiple regression models.

Akaike Information Criterion (AIC) employed to select the ‘best model’ that relates the fluctuation in habitat structure variables and the abundance of *Bolitoglossa pandi*.

Dependent variable: Ln-transformed abundance of *B. pandi* (Lnn). Independent variables: Veg.per.cover = Vegetation percentage cover. Ln-transformedMean Temp = Environmental mean temperature. LnMeanH = Ln-transformed relative humidity. Lnlife forms = Ln-transformed number of vegetation life form Veg.layers = Vegetation layers. Lnleaf litter= leaf litter depth. Nor.test = Kolmogorov-Smirnov’s test for normality, Hom.test = Breusch-Pagan test for homoscedasticity, and Aut.test = Durbin-Watson test for autocorrelation. Values shown are standard error (SE) and *t* test-value. Bold values denote statistical significance at the $p < 0.05$ level.

Multiple regressions models	AIC	Δ AIC	Nor.t est	Hom.test	DW
Lnn~Lnleaf litter+LnMean temp+Veg.per.cover+Altitude	-73.64	0.0			
Lnn~Lnleaf litter+LnMean temp+Veg.per.cover+Altitude+Lnlife forms	-73.08	-0.56			
Lnn~Lnleaf litter+LnMean temp+Veg.per.cover+Altitude+LnMeanHr	-71.92	-1.72	0.45	0.06	1.10
LnAbundance~Lnleaf litter+LnMeantemp+Veg.leyers+Veg.per.cover+Altitude+LnMeanH+Lnlife forms	-70.04	-3.60			
The “best” multiple regression model	Estimate	SE	t-value	P(> t)	
Intercept	3.33	1.93	2.79	0.006	
Ln leaf litter	0.88	0.071	12.34	<	0.0001
Ln Mean temp	0.69	0.167	4.12	<	0.0001
Vegetation percentage cover	-1.87	0.50	-3.73	< 0.001	
Altitude	-0.001	0.0004	-2.46	0.014	
F= 69.18, df= 4–182, P < 0.0001					

Table 5 (on next page)

Nested ANOVA results.

Values shown are mean standard error (SE) and *t* test-value. Bold values denote statistical significance at the $p < 0.05$ level.

Time interval	Salamander observed	Estimate	SE	t value	P(> t)	Normality test	Levene test
H1 (18:30 a 19:30)	Max= 16, Min= 1 □ = 4.16, N= 154	-0.0257	0.0234	-1.085	0.279		
H2 (19:31 a 20:30)	Max= 20, Min= 1 □ = 4.91, N= 172	0.010	0.022	0.461	0.645		
H3 (20:31 a 21:30)	Max= 12, Min= 1 □ = 4.73, N= 123	0.013	0.023	0.525	0.599		
H4 (21:31 a 22:30)	Max= 8, Min= 1 □ = 3.46, N= 97	-0.047	0.023	-2.037	0.042		
H5 (22:31 a 23:30)	Max= 16, Min= 1 □ = 4.393, N= 158	-0.006	0.020	-0.309	0.757	P = 0.08	P = 0.2
H6 (23:31 a 00:30)	Max= 20, Min= 1 □ = 6.05, N= 260	0.049	0.020	2.493	0.013		
H7 (00:31 a 01:30)	Max= 18, Min= 1 □ = 4.73, N= 196	0.040	0.022	1.834	0.067		
H8 (01:31 a 02:30)	Max= 26, Min= 1 □ = 6.47, N= 116	0.003	0.030	0.100	0.920		
H9 (02:31 a 03:30)	Max= 26, Min= 1 □ = 4.43, N= 62	0.003	0.030	0.100	0.920		

Table 6(on next page)

Wilcoxon test results.

N= number of salamanders observed. Bold values denote statistical significance at the $p < 0.05$ level.

Sampling occasion	Occasion 1	Occasion 2	Median	Neonates	Juveniles	Adults
Occasion 1	—	—	32.05	N = 4 (8%)	N= 13 (26%)	N = 33 (66%)
Occasion 2	W= 11825, P < 0.001	—	25.40	N = 55 (34%)	N= 49 (30%)	N = 57 (36%)
Occasion 3	W= 22590, P = 0.006	W = 84198, P < 0.001	28.20	N = 76 (13%)	N= 277 (46%)	N = 252 (42%)

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