AQUATIC CONSERVATION: MARINE AND FRESHWATER ECOSYSTEMS

Aquatic Conserv: Mar. Freshw. Ecosyst. **25**: 307–321 (2015)

DOI: 10.1002/aqc.2559

Mesohabitat associations of the threatened San Marcos salamander (Eurycea nana) *across its geographic range*

PETER H. DIAZ[a], JOE N. FRIES[b], TIMOTHY H. BONNER[a], MARA L. ALEXANDER[b] and WESTON H. NOWLIN[a,*]

[a]*Department of Biology, Aquatic Station, Texas State University, San Marcos, Texas 78666, USA*

[b]*San Marcos Aquatic Resource Center, United States Fish and Wildlife Service, San Marcos Texas 78666, USA*

ABSTRACT

1. Habitat loss is one of the most critical factors affecting the loss of species. However, habitat conservation of many threatened species is performed with incomplete information on habitat requirements and trophic ecology, thus presenting a challenge to designing and implementing recovery plans.

2. The San Marcos salamander (*Eurycea nana*) is a federally threatened spring-associated organism whose geographic distribution is limited to the headwaters of the San Marcos River in Texas, USA. Although its designated critical habitat includes the headwaters and the first 50 m of the river, little is known of its habitat requirements or co-occurrence with benthic macroinvertebrates and macrophytes.

3. This study examined mesohabitat associations of the salamander and patterns of co-occurrence with macrophytes and benthic invertebrates within its critical habitat. Surveys of mesohabitat characteristics were conducted during a one-year period and data were analysed to assess mesohabitat associations of the San Marcos salamander and patterns of co-occurrence with invertebrates and macrophytes.

4. Salamanders were distributed throughout the critical habitat, but were almost exclusively found in mesohabitats containing cobble and gravel with coverage of *Amblystegium* and filamentous algae. The salamander did not exhibit consistent co-occurrence with specific invertebrates or macrophytes across the critical habitat, indicating that salamanders were probably selecting mesohabitats based on benthic substrate and not the biotic communities.

5. Protection of a specific mesohabitat type within the critical habitat of the San Marcos salamander is likely to be one of the most important conservation measures, given that it accounts for ~7% of the total area within the designated critical habitat. These results also emphasize that habitat conservation plans for species at risk should consider that contemporary spatial distribution of species within habitats may be influenced not only by their evolutionary history but also by past and current human pressures.

Received 21 August 2013; Revised 16 December 2014; Accepted 10 January 2015

KEY WORDS: spring; river; endangered species; habitat management; amphibians; invertebrates; impoundment; sedimentation

---

*Correspondence to: Weston H. Nowlin, Department of Biology, Aquatic Station, Texas State University, San Marcos, Texas, 78666, USA. E-mail: wn11@txstate.edu

INTRODUCTION

It is universally recognized that habitat loss and fragmentation are some of the most important factors leading to loss of species (Sih *et al*., 2000). Ecologists and conservation biologists have concentrated on the maintenance of diversity because losses in overall taxonomic diversity or the loss of individual and functionally important species from a community can have potentially far-reaching implications (Hooper *et al*., 2005). However, owing to the large number of attributes and scales of biological diversity, the practicality of monitoring multiple aspects of diversity to preserve or manage habitat presents an exceedingly difficult challenge. In order to overcome these challenges, monitoring programmes often use a single species or a limited number of species in the community (Lambeck, 1997; Carignan and Villard, 2002). In particular, many management efforts focus on habitat conservation for vulnerable or rare species. Although the utility of approaches in which a single or limited set of species within the community is used to preserve habitat, overall biodiversity, or ecosystem functioning has been questioned (Simberloff, 1998), conservation approaches that focus on the preservation or protection of habitat of a single or a limited set of species in the community are widespread (Carignan and Villard, 2002).

In North America, habitat preservation and management often is associated with taxa designated as vulnerable or under threat of extinction. The United States Endangered Species Act (ESA) was created to protect species under threat of extinction and to preserve their habitats. One of the provisions of the ESA requires that a Recovery Plan is developed that will lead to a reduction of extinction threats and an increase in the population of the endangered or threatened species (USFWS, 1995a). Listing an organism as threatened or endangered requires that critical habitat be designated. Under the ESA, critical habitat is defined as the specific areas within the geographic areas occupied by a species that contain biological and physical features essential for the conservation of the species in question (US ESA, 1988). Although designation of critical habitat is an important part of the recovery process, it should be done with sufficient information, such as detailed data on how variation in habitat quality within the protected critical habitat area can affect the abundance and distribution of listed taxa (Rosenfeld and Hatfield, 2006). Indeed, determining the role of habitat in the limitation and viability of imperilled populations represents one of the most difficult issues associated with endangered species management (Rosenfeld and Hatfield, 2006). Funding for recovery plans is limited, but it is imperative to determine the factors that lead to recovery of listed populations and to maximize efforts to remove species from the list (Lawler *et al*., 2002). Data on species–habitat interactions as habitat associations, spatial variation in habitat quality, and dietary requirements should be included in recovery plans to augment more efficient recovery (Harvey *et al*., 2002; Moore and Gillingham, 2006).

Throughout the arid and semi-arid south-western United States, many threatened or endangered aquatic species are associated with spring-fed ecosystems (Sada *et al*., 2005). The abundance and distribution of many spring-associated organisms are thought to be dependent upon the physical and biological stability of the spring environment (Holsinger and Longley, 1980; Humphreys, 2006). Spring-fed aquatic ecosystems generally display consistent hydrologic and physicochemical characteristics (Roca and Baltanas, 1993), and ecologists have hypothesized that this constancy has led to local adaptation of fauna in spring-influenced ecosystems (Hubbs, 1995). In addition, many spring ecosystems in the arid south west exhibit relatively high levels of endemism, with many taxa having ranges limited to a single spring (Seidel *et al*., 2009). At present, many spring-associated species experience habitat degradation from human activities, including decreased flows caused by groundwater extraction and introduction of non-native species (Bowles *et al*., 2006) and these stresses have led to extirpation of native spring-associated species (Strayer, 2006).

The upper San Marcos River is a spring-fed ecosystem located along the margin of the Edwards Plateau in Central Texas. Spring Lake is the headwaters of the San Marcos River and artesian spring water from the Edwards Aquifer emerges into the lake from ~200 openings. The San Marcos River and its headwaters are located along the margin of the Edwards Plateau, a region in central

Texas characterized by the presence of abundant spring openings, a large number of endemic spring and aquifer-associated biota, and rapidly expanding human populations (Bowles and Arsuffi, 1993). The upper San Marcos River contains a relatively large number of federally protected taxa, including the San Marcos salamander (*Eurycea nana*), Texas wild rice (*Zizania texana*), the fountain darter (*Etheostoma fonticola*), and the Comal Springs riffle beetle (*Heterelmis comalensis*). Owing to the large number of protected species with limited geographic ranges within this area and the threat of human development and groundwater extraction, the upper portion of the San Marcos River receives a great deal of conservation effort to protect and enhance habitats.

The San Marcos salamander is a small-bodied (<50 mm total length), plethodontid salamander that has an exclusively aquatic life cycle and exhibits a geographic range limited to the San Marcos River headwaters (Chippindale *et al*., 1998). The USFWS and the State of Texas currently lists the San Marcos salamander as threatened and the critical habitat is designated as the headwaters of the river (Spring Lake) and the first 50 m of the river below a dam at the end of Spring Lake (USDI, 1980). This area is thought to encompass virtually all of the geographic range of the San Marcos salamander (USFWS, 1995b). Although this species can be abundant in portions of the headwaters (Tupa and Davis, 1976), a limited geographic range and threats of declining groundwater levels led to its listing (USDI, 1980). Estimation of the population size of San Marcos salamanders in the San Marcos River headwaters has been performed many times (Tupa and Davis, 1976; Nelson, 1993; Lucas, 2006). However, habitat associations of the San Marcos salamander within the headwaters of the San Marcos River has only been semi-quantitatively examined once and the study concluded that salamanders were most commonly observed within the aquatic moss *Amblystegium* sp. along a 40 m long stretch on the north-western side of Spring Lake, typically in close proximity to spring openings (Tupa and Davis, 1976). Laboratory studies subsequently found that San Marcos salamanders exhibit a relatively low upper thermal tolerance, indicating adaptation to thermally stable spring environments (Berkhouse and Fries, 1995) and salamanders prefer current velocities around 1 cm s^{-1} (Fries, 2002). Cumulatively, these studies suggest that San Marcos salamanders are likely to exhibit some habitat preference within their larger designated critical habitat, but there has been no systematic and quantitative examination of mesohabitat associations of salamanders in their natural habitat.

The objective of this study was to assess whether the San Marcos salamander exhibits specific mesohabitat preferences within its greater critical habitat area. A further aim was to examine whether the San Marcos salamander co-occurred with specific groups of benthic invertebrate and macrophyte species inside their critical habitat area, potentially indicating species-specific interactions that may be critical for the San Marcos salamander. The study also explored the conservation consequences of managing specific mesohabitat conditions within the larger critical habitat area for this threatened species in particular and other species at risk that exhibit small geographic ranges. Given the probable spatial variation in physicochemical conditions and biological communities within the critical habitat of the San Marcos salamander, data on associations with specific mesohabitat types in their critical habitat would contribute to more effective management and recovery of this species. It was hypothesized that the abundance and distribution of San Marcos salamanders is associated with specific mesohabitat types within its critical habitat, thereby indicating that the salamander uses only a portion of its larger designated critical habitat.

MATERIALS AND METHODS

Study site

The upper San Marcos River, including Spring Lake, is located in Hays County, central Texas (Figure 1(A)). The lake and upper river are almost exclusively supported by Edwards Aquifer groundwater which arises from the upper portion of the Spring Arm of the lake. In contrast, the Slough Arm of the lake is largely fed by surface runoff from the ephemerally flowing Sink Creek. Water flows through the lake, over a dam, and into the upper San Marcos River. The dam was

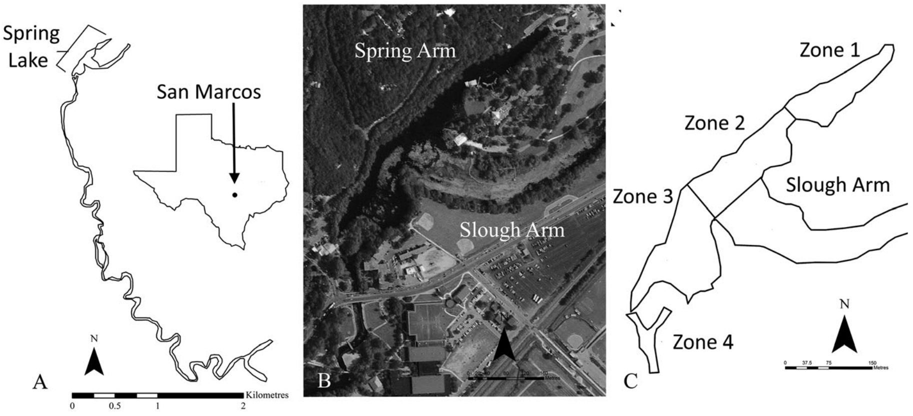

Figure 1. (A) Map of the location of Spring Lake and the upper San Marcos River in Texas, USA. (B) Aerial photograph of the designated critical habitat area of the San Marcos salamander in Spring Lake and the upper 50 m of the San Marcos River below Spring Lake dam. (C) Map of the critical habitat area showing the sampling zones in the Spring Arm of the lake (zones 1–3) and the zone in the upper river (zone 4).

constructed in 1849 to power a gristmill; the impoundment, Spring Lake, did not exist before this and early descriptions of the headwaters describe it as a stream which on average was approximately 20 m wide and 1 m deep (McClintock, 1930; Kimmel, 2006).

Study design

The San Marcos salamanders' designated critical habitat was divided into four zones, based upon hydrology and morphometric characteristics (Figure 1(B) and (C)). The first three zones were located within Spring Lake. Zone 1 encompassed the first 240 m of the spring arm of Spring Lake and contained the greatest number of spring openings. Zone 2 began immediately below zone 1 and extended 180 m down the lake to where the Slough Arm converges with the Spring Arm of the lake. Zone 2 contained some spring openings dispersed along the remnant river channel. At this point, zone 3 encompassed the remaining 150 m of the lake and contains relatively few spring openings. Zone 4 began below the dam at the outflow of the lake and ended 50 m downstream. Although all of Spring Lake is designated as critical habitat for the San Marcos salamander, the Slough Arm of the lake was excluded in the study sampling design because its hydrology, sediment composition, and water quality are substantially different from the rest of the lake and the salamander has not been observed in the Slough Arm (W.H. Nowlin, pers. obs.).

Assessment of mesohabitat associations

In this study, mesohabitats are defined as biologically and physicochemically distinct habitats (Pardo and Armitage, 1997). For example, relatively small areas (i.e. several square metres) with clearly differing vegetation types and benthic substrate composition would be classified as different mesohabitats. San Marcos salamander mesohabitat associations were assessed during five sampling events (3–4 days each event) over the course of the study. Sampling was carried out in 2008 in the summer (May, July, and August) and winter (November and December); the role of seasonality was expected to be minimal because of the consistent physicochemical (e.g. temperature and water chemistry) and hydrological characteristics of this groundwater-based system. On each sampling occasion sample collections were divided equally between day and night to determine whether San Marcos salamanders show diel variation in mesohabitat associations.

Spatial distributions in mesohabitats and the abundance and distribution of San Marcos salamanders were assessed using a modified random sampling design nested within each of the four zones within the critical habitat. For the first sampling event (May 2008), transects within each zone were spaced 15 m apart, perpendicular to the long axis of the spring arm of Spring Lake and the river channel in the first 50 m of the upper San Marcos River. After transects were established, points were selected randomly along each transect within each zone and a total of 47 sampling points were located across the entire sampling area. Of the initial 47 samples, the number of sampling points within each zone was not evenly distributed among zones; the number of sampling points within a zone increased with the total area of the zone. This was done to equalize sampling effort among zones so that approximately the same proportional area inside each zone was sampled regardless of the total area of the zone.

After sampling points were assigned, two SCUBA divers lowered a 1 m^2 PVC quadrat onto the substrate at each sampling point. The water column height (water depth) above the quadrat was measured and dissolved oxygen concentration (DO; mg L^{-1}), pH, water temperature (°C), conductivity (μS cm^{-1}), and salinity (ppt) were recorded in the middle of each quadrat approximately 0.5 m above the benthic surface using a water quality sonde (YSI 6600 V2-4 Multi-parameter sonde). Divers then identified macrophytes (if present) to genus, the percentage cover of each macrophyte species, and the dominant substrate type within each quadrat. Substrate types were classified using a modified Wentworth classification scheme (Cummins, 1962; Nielsen and Johnson, 1989), which included five categories: (1) a mix of fine-grained mud and silt; (2) sand; (3) gravel; (4) cobble; and (5) boulder. Obvious spring openings were noted if present in a quadrat.

After recording data on habitat characteristics, divers searched for San Marcos salamanders inside each quadrat for a 5 min period by turning over rocks and debris. Salamanders were captured with aquarium nets or by hand, and counted. Immediately following the search for salamanders, a 500 μm aperture mesh 0.5 m wide drift net was placed in a downstream corner of each quadrat and the benthic surface disturbed within this 0.25 × 0.25 m area for a 2 min interval to collect invertebrates. Invertebrates were preserved in 70% ethanol and identified and counted in the laboratory. Identification was performed to the lowest practical taxonomic level using keys (Wiggins, 1996; Merritt *et al*., 2008).

The initial collection of 47 quadrat samples in May indicated that San Marcos salamanders were not widely distributed and rare in most portions of the sampled area; a total of seven salamanders were observed in 47 quadrats across all zones on the first sampling occasion. Because of these low numbers, the sampling design was modified for the subsequent sampling events (July, August, November, and December): this reduced the total potential area to be sampled within each zone. The area of the remnant river channel was determined within the Spring Arm of the lake. The remnant river channel begins at zone 1 and runs throughout zone 2, zone 3 in the lake, and encompasses the total area of zone 4 (i.e. the river below the lake). Sampling efforts were limited to the remnant river channel because: (1) it was historically the habitat of the San Marcos salamander before the building of the Spring Lake dam; (2) it is situated along the central portion of the Spring Arm of the lake; (3) it still represents a substantial portion of the total area within zones; and (4) it contains all of the major mesohabitat types that were observed in the first sampling event. The remnant river channel was delineated in the lake by diving and marking the channel with flags, and adding an additional 1 m buffer on each side. Subsequently and to be consistent with the initial sampling in May, transects were placed across the delineated remnant river channel every 15 m down the length of the Spring Arm of the lake and the upper 50 m of the river. A sampling design similar to that of the first sampling event was used by randomly selecting sampling points along each transect in each zone and ensuring that the number of sampling points within a zone increased with the total potential sampling area inside each zone. For the remaining sampling events, 145 quadrat samples were collected to assess mesohabitat characteristics, salamander densities, and invertebrate community composition using the same methods as before.

In order to determine the extent of specific mesohabitat types within the critical habitat area, an additional survey was performed. Transects were created 10 m apart longitudinally down the lake and observations of dominant substrate type were made every 5 m along that transect for the entire length and width of the lake. Areas inside these quadrats were classified according to dominant substrate type (using the same scheme as the initial mesohabitat survey) and whether non-rooted and low-profile primary producers (e.g. *Amblystegium* and filamentous algae) or rooted macrophytes were present. Data were collected from an additional 568 1 m^2 quadrats from the Spring Arm of the lake.

Data analysis

Principal component analysis (PCA) was carried out to assess spatial variation in mesohabitats within the study area and how these habitats were distributed among the zones of the sampled areas, using the software package R (R Development Core Team, 2008). Principal component analysis often is used to examine environmental gradients and variation in habitat types (Krebs, 1999). Continuous and percentage data (e.g. water temperature, water pH, DO, conductivity, water depth, percentage total vegetation cover, and percentage cover of the various macrophyte species) were *z*-score transformed before analyses. The presence or absence of spring openings (0 = present, 1 = absent) and the dominant substrate type were coded as dummy variables (dominant substrates were categorized as values from 1 to 5). PCA scores for each quadrat were identified by source (lake or river) to assess concordance among mesohabitats between Spring Lake and the San Marcos River.

In order to assess potential mesohabitat associations of San Marcos salamanders and benthic invertebrates, canonical correspondence analysis (CCA) with Canoco version 4.5 (ter Braak, 1986) was used. Canonical correspondence analysis is a multivariate analytical method used to examine the relationships between biological assemblages and environmental data. In particular, CCA is an appropriate method to examine habitat associations of organisms along environmental gradients (ter Braak and Verdonschot, 1995). Environmental data used for the CCA included the dominant substrate type in each quadrat and the percentage cover of the various macrophyte species. Percentage cover of macrophyte species was included in the environmental dataset because macrophyte communities often play a substantial habitat and structural role in many freshwater communities (Thorp *et al.*, 1997; Weaver *et al.*, 1997). Although DO, water temperature, conductivity and pH varied among critical habitat zones, the magnitude of differences in DO (5.65–7.87 mg L^{-1}), temperature (21.68–22.50 °C), conductivity (570–580 $\mu S\ cm^{-1}$), and pH (7.43–7.67) were probably not biologically relevant and were thus not included in the CCA. The biological data in the CCA included the density of San Marcos salamanders found inside each quadrat and the density of aquatic invertebrates captured during sampling. The CCA model significance was determined using Monte Carlo analysis with 999 permutations and $\alpha = 0.05$ (ter Braak and Verdonschot, 1995).

RESULTS

Assessment of the physicochemical environment in the four zones showed that there was substantial spatial variation in the distribution of mesohabitat types in critical habitat area. This general trend in the spatial distribution of mesohabitats between the lake and river samples was evident in the PCA (Figure 2, Table 1). Principal component analysis axes I and II cumulatively explained 25% of the variation in the measured mesohabitat characteristics among quadrats. The environmental gradient represented along PC I separated sites sampled in zones 1–3 from those in zone 4. This axis represented a gradient of mesohabitats in the river

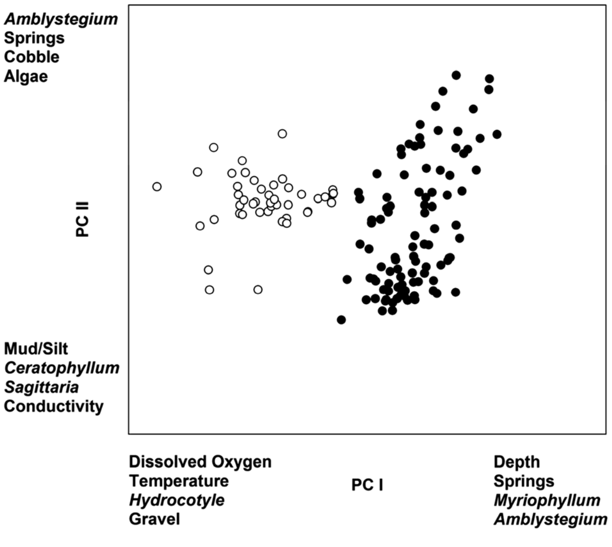

Figure 2. Principal component analysis (PCA) ordination of environmental data (water depth, dominant substrate type, percentage cover of different macrophyte species and filamentous algae, DO concentration, conductivity, water temperature, and the presence of springs) collected from quadrats within the critical habitat for the San Marcos salamander. Data for individual quadrats collected from Spring Lake (●) and in the San Marcos River below the dam (○) are presented.

Table 1. Results from the PCA for environmental variables in the mesohabitat survey conducted in the critical habitat area of the San Marcos salamander. Values represent loadings along PC1 and PC2 for variables used in the PCA. '—' indicates that a variable did not have a relatively high loading value on a principal component axis

Variable	PC1	PC2
Amblystegium	0.15	0.39
Ceratophyllum	—	–0.31
Cobble	—	0.25
Conductivity	—	–0.15
Dissolved oxygen	–0.43	—
Gravel	–0.21	—
Hydrocotyle	–0.21	—
Myriophyllum	0.17	—
Sagittaria	—	-0.2
Silt/mud	—	-0.52
Spring openings	0.21	0.31
Spring openings		
Water depth	0.36	—
Water temperature	–0.38	—

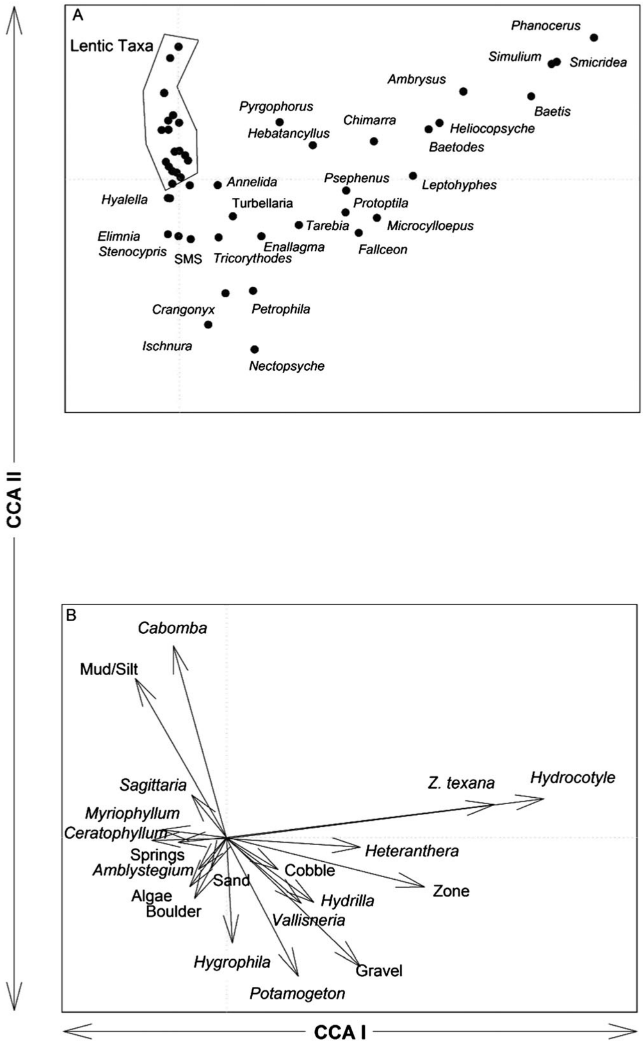

Figure 3. Canonical correspondence analysis (CCA) plot of (A) the densities of macroinvertebrates and San Marcos salamanders, and (B) the environmental conditions (dominant substrate type, percentage cover of different macrophyte species and filamentous algae, the presence of springs) and spatial location (zone number) for quadrats throughout the Spring Arm of Spring Lake and the upper 50 m of the San Marcos River. The irregular polygon in panel (A) encompasses a group of tightly clustered, primarily lentic, invertebrate taxa. The length of the arrows (vectors) in panel (B) indicates the relative importance of the environmental parameter in determining variation among quadrat sites.

with higher DO concentrations, greater temperature, higher percentage cover of the macrophyte *Hydrocotyle* sp. and substrate dominance of gravel to mesohabitats in the lake which had greater water depth, the presence of spring openings, and greater percentage cover of the macrophyte *Myriophyllum* sp. and the aquatic moss *Amblystegium* sp. In contrast to the gradient displayed along the first principal component, PC II described a gradient of mesohabitats within Spring Lake. This axis represented an environmental gradient of mesohabitats dominated by silt/mud, with increasing percentage cover of the macrophytes *Ceratophyllum* sp. and *Sagittaria* sp., and higher conductivity, to mesohabitats dominated by *Amblystegium* cover, the presence of spring openings, cobble as the dominant substrate type, and higher percentage cover of filamentous algae. The gradient along PC II is indicative of the spatial distribution of mesohabitats found largely throughout the upper portion of the lake, with a transition to the dominant mesohabitat types found in zones 2 and 3.

The entire dataset including both lake and river quadrats was examined using CCA, which showed a large influence of the longitudinal position of quadrats in the critical habitat (i.e. large influence of zone) (Figure 3(A) and (B), Table 2). The CCA accounted for 30% ($P = 0.001$) of the variation in abundance and distribution of San Marcos salamanders and benthic invertebrates across the critical habitat area. Of the 30% explained by the model, canonical axis (CA) I explained 44% ($P = 0.001$), and CA II explained an additional 15% of the total variance in community composition. Canonical axis I largely revealed differences in mesohabitats associated with the different zones, reflecting a gradient of mesohabitats composed of *Hydrocotyle*, *Zizania texana*, and *Heteranthia* (zone 4), to mesohabitats composed of mud/silt, *Ceratophylum*, *Myriophyllum*, and *Cabomba* (zones 1–3). Canonical axis II exhibited a

Table 2. Results from the CCA for environmental variables in the mesohabitat survey conducted across the entire critical habitat area of the San Marcos salamander. Values represent loadings along CA1 and CA2 for variables utilized in the CCA. '—' indicates that a variable did not have a relatively high loading value on a CA axis

Variable	CA1	CA2
Cabomba	–0.13	0.68
Ceratophyllum	–0.17	–
Gravel	–	–0.41
Habitat zone	0.48	–
Heteranthia	0.36	–
Hydrocotyle	0.76	0.12
Hygrophila	–	–0.35
Myriophyllum	–0.15	–
Potamageton	–	–0.43
Sagittaria	–	0.11
Silt/mud	–0.22	0.53
Valisneria	–	–0.21
Zizania texana	0.64	–

gradient of mesohabitats within Spring Lake comprised of *Cabomba*, mud/silt, *Hydrocotyle*, and *Sagitarria* to negative loadings of *Potamogeton*, gravel, *Hygrophila*, and *Vallisneria*.

Given the substantial differences among mesohabitat environmental variables and biological assemblages between Spring Lake and the San Marcos River (Figures 2 and 3), further examinations of salamander and invertebrate mesohabitat associations were performed separately for the sampling sites in Spring Lake and those in the San Marcos River. Thus, separate CCA models were constructed for sites in Spring Lake and one in the zone in the river below Spring Lake dam. In the Spring Lake portion of the critical habitat, the CCA accounted for 20% ($P = 0.002$) of the variation in the abundance and distribution of the Spring Lake community (Figure 4(A) and (B), Table 3). Of the 20% explained by the model, CA I explained 41% ($P = 0.022$), and CA II explained an additional 25% of the total variance in community composition. The environmental gradient on CA I described mesohabitats characterized by gravel and boulder substrates and a higher percentage cover of filamentous algae and *Amblystegium* to mesohabitats composed of silt/mud substrates and higher percentage cover of the macrophytes *Cabomba* sp., and *Sagittaria*. Of the 100 quadrats portrayed in the Spring Lake CCA model, ~17% were dominated by gravel/boulder substrates with higher percentage cover of filamentous algae and *Amblystegium*. Gravel and boulder mesohabitats were distributed throughout zone 1 but were restricted to the old river channel in zones 2 and 3. The environmental gradient along CA II portrayed lake mesohabitats composed of gravel substrates, higher percentage cover of *Myriophyllum* and *Cabomba*, and the presence of spring openings changing to mesohabitats composed of boulder and silt/mud substrates with higher cover of *Ceratophyllum* and *Sagittaria*.

During mesohabitat surveys, 126 San Marcos salamanders were observed in Spring Lake. Zone 1 contained 89 salamanders; in quadrats where salamanders occurred there was a mean of 1.9 salamanders m^{-2}. In zone 2, 34 salamanders were found ($\overline{X} = 0.94$ salamanders m^{-2} in quadrats that contained salamanders). The total number and density of salamanders declined along the longitudinal axis of the lake, with three salamanders captured throughout the entire study in the most downstream zone immediately above the spillway of the lake ($\overline{X} = 0.16$ salamanders m^{-2} in quadrats containing salamanders). In total, 68 336 aquatic invertebrates were collected and counted from quadrats in Spring Lake. The San Marcos salamander was associated with mesohabitats composed of gravel and cobble substrates, the presence of spring openings, and higher percentage cover of *Amblystegium* (Figure 4(A)). Four macroinvertebrate taxa (*Stenocypris* cf. *major*, Chydoridae, Turbellaria and *Elimnia* sp.) were associated also with this mesohabitat type. In contrast, the gastropods *Elimnia*, Hydrobidae, and *Gyraulus* sp. were associated with mesohabitats containing *Myriophyllum*, *Cabomba*, and gravel. The shrimp *Palaemonetes* sp. and the ephemeropteran *Tricorythodes* sp. were associated with mesohabitats containing boulder-sized substrates and *Ceratophyllum*. *Hyalella* sp., the most abundant invertebrate encountered in Spring Lake, had a cosmopolitan distribution across mesohabitats.

The CCA model generated for the riverine portion of the critical habitat explained 46% of the variation in community composition and distribution ($P = 0.003$) (Figure 4(C) and (D), Table 3). In this model, CA I explained 44% ($P = 0.010$) of the total variance, while CA II explained an additional 22%. Canonical correspondence axis

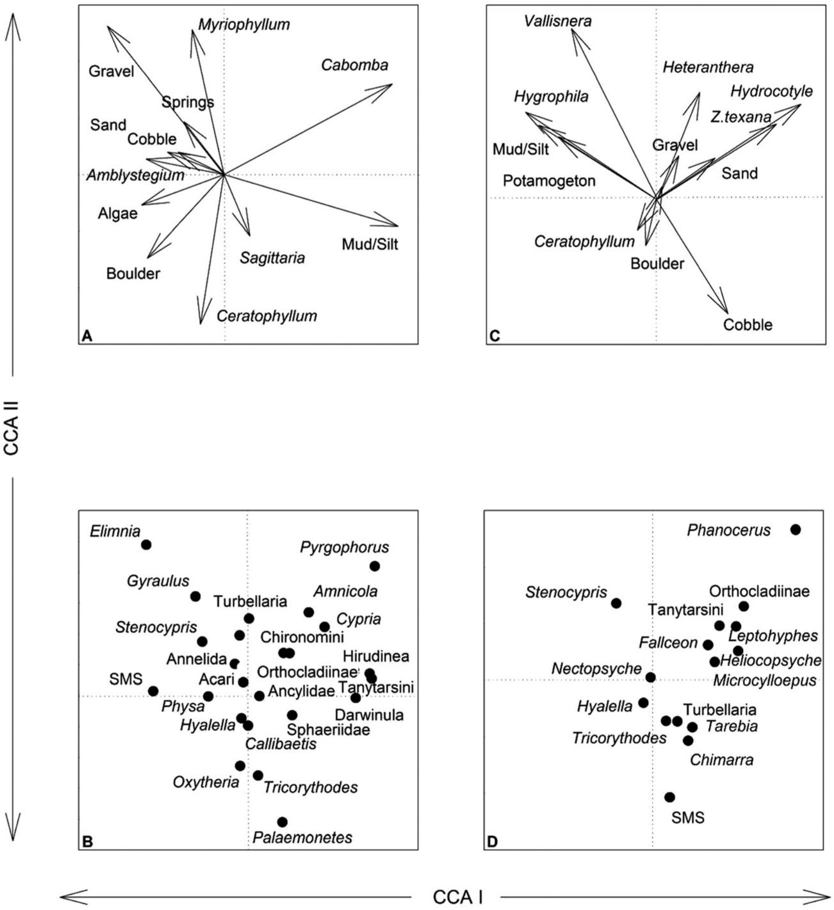

Figure 4. Canonical correspondence analysis (CCA) plot of (A) the densities of macroinvertebrates and San Marcos salamanders in quadrats found throughout the Spring Arm of Spring Lake (zones 1–3 of the critical habitat), (B) the environmental conditions for quadrats found throughout the Spring Arm of Spring Lake, (C) the densities of macroinvertebrates and San Marcos salamanders for quadrats found in the first 50 m of the San Marcos River (zone 4), and (D) the environmental conditions for quadrats found in the first 50 m of the San Marcos River. The length of the arrows (vectors) in panels (B) and (D) indicates the relative importance of the environmental parameter in determining variation among quadrat sites.

I described a gradient of mesohabitats dominated by cobble and sand substrates and containing higher cover of *Hydrocotyle* and *Zizania texana*, to mesohabitats composed of silt/mud substrates with higher cover of *Hygrophila* sp., *Potamogeton* sp., and *Vallisnera* sp. Canonical correspondence axis II characterized a mesohabitat gradient of mud/silt substrates with higher cover of *Vallisnera*, *Heteranthera* sp., *Hydrocotyle*, and *Z. texana* to mesohabitats containing cobble substrates with little to no macrophyte cover.

In the riverine portion of the critical habitat, 62 San Marcos salamanders were observed and these individuals were associated generally with mesohabitats consisting of cobble and boulder substrates with relatively low levels of macrophyte cover (Figure 4(C) and (D)). In total, 10 208 aquatic invertebrates were recorded from riverine quadrats; invertebrate taxa that co-occurred with San Marcos salamanders were turbellarian flatworms and the caddisfly *Chimarra* sp. Similar to Spring Lake, *Hyalella* was widely distributed across all mesohabitat types and was ordinated near the centre of the species biplot.

DISCUSSION

Results presented here indicate that the San Marcos salamander exhibited definite mesohabitat associations within its greater critical habitat area and were most frequently associated with

Table 3. Results from the CCA for environmental variables in the mesohabitat survey conducted in the lake and river portions of the critical habitat area of the San Marcos salamander. Values represent loadings along CA1 and CA2 for variables utilized in the CCA. '--' indicates that a variable did not have a relatively high loading value on a CA axis

	Lake		River	
Variable	**CA1**	**CA2**	**CA1**	**CA2**
Amblystegium	-0.31	--	–	–
Boulder	-0.31	-0.29	--	--
Cabomba	0.69	0.32	--	--
Ceratophyllum	--	-0.51	--	--
Filamentous algae	-0.33	--	--	--
Gravel	-0.47	0.51	--	--
Myriophyllum	--	0.51	--	--
Potomageton	--	--	--	--
Sagittaria	0.09	-0.21	--	--
Silt/mud	0.71	-0.18	-0.54	--
Spring openings	--	0.18	--	--
Zizania texana	--	--	0.55	0.30
Cobble	--	--	0.33	-0.46
Sand	--	--	0.27	--
Hydrocotyle	--	--	0.67	0.38
Hygrophila	--	--	-0.60	--
Potamageton	--	--	-0.45	--
Valisneria	--	--	-0.40	0.70
Heteranthia	--	--	--	0.42

mesohabitats dominated by cobble, boulder, and gravel substrates with higher percentage coverage of *Amblystegium* and filamentous algae. The findings also showed that there is substantial spatial variation in habitat quality within the larger critical habitat area of the San Marcos salamander, but that San Marcos salamanders were not closely associated with any specific invertebrate group or groups.

The association of the San Marcos salamander with cobble substrates across both of the major habitat areas within the critical habitat (i.e. the lake and the river sections) is similar to habitat associations reported for other species of *Eurycea* (Tumlison *et al*., 1990; Bowles *et al*., 2006). Rock and gravel substrates serve as spatial refuges for small-bodied salamanders from potential predators such as fish (Barr and Babbitt, 2002) and crayfish (Tumlison *et al*., 1990; Bowles *et al*., 2006). Stream-dwelling small-bodied salamanders are susceptible to crayfish and fish predation and the presence of these predators may lead to their local extirpation (Petranka, 1983; Barr and Babbitt, 2002; Davic and Welsh, 2004). Both crayfish and fish are present in the upper San Marcos River. Carnivorous centrarchids (*Lepomis* and *Micropterus* spp.) are abundant and dominant members of the fish community in the upper portion of the river (Perkin and Bonner, 2010) and these fish are likely to consume salamanders (Semlitsch, 1987).

Although the San Marcos salamanders exhibited similar general mesohabitat associations across all of the areas sampled, some habitat characteristics associated with San Marcos salamanders differed across habitat areas. In the lake portion of the habitat (zones 1–3), the San Marcos salamander was positively associated with gravel, cobble, and boulder substrates with relatively higher percentage cover of *Amblystegium*, and negatively associated with softer mud/silt habitats and rooted macrophytes. Much like lake-resident salamanders, riverine salamanders (zone 4) were primarily associated with cobble-sized substrates, but riverine salamanders were not strongly associated with *Amblystegium* and were more frequently found in mesohabitats composed of bare rock surfaces with no macrophyte cover (>85% of riverine San Marcos salamanders occurred in quadrats with no or very low macrophyte cover). The aquatic moss *Amblystegium* was much less abundant in the riverine zone; approximately 2% of quadrats sampled in the riverine zone of the critical habitat area contained *Amblystegium*, whereas 18% of quadrats in the upper headwaters region of the lake (zone 1) contained *Amblystegium*. Aquatic bryophytes are associated with larger rocky substrates (Steinman and Boston, 1993), presumably due to higher substrate stability and less opportunity for sloughing and abrasion losses from surfaces (Suren, 1991). For San Marcos salamanders, both the upper headwaters area (zone 1) and the riverine section (zone 4) contain substantial areas of habitat dominated by relatively large rocky substrates; however, shallower water depths (<0.5 m deep in the river and depths up to 6.7 m in the lake) and subsequent increased water velocities in the riverine portion of the habitat may limit the cover of *Amblystegium* owing to the higher probability of abrasion and scouring losses. Aquatic mosses such as *Amblystegium* can provide habitat and contain food resources for consumers

(Brusven *et al.*, 1990), but these findings indicate that it is likely that salamanders are primarily associated with mesohabitats consisting of benthic substrates of a specific size and composition (i.e. cobble) and that the observed association with *Amblystegium* in the lake portion is probably a consequence of the types of substrates they use. The findings of the present study further suggest that, from a critical habitat management perspective, cobble-dominated mesohabitat is a more important physical habitat feature than the presence of *Amblystegium*.

Mesohabitat patches composed of relatively large rocky substrate occurred throughout the critical habitat, but the proportion of this mesohabitat type, and correspondingly the density and total number of San Marcos salamanders, varied substantially. Within the lake portion of the habitat (zones 1–3), proportions of the various general mesohabitat types varied longitudinally down the lake. Within the upper headwaters of the Spring Arm of the lake (zone 1), gravel, cobble, and boulder-dominated mesohabitats comprised about 21% of the sampled area, whereas this mesohabitat type comprised only about 14% and 2% of the sampled area in zones 2 and 3, respectively. Correspondingly, densities (number of salamanders m^{-2}) and total number of salamanders differed across the lake zones. Within the lake, zone 1 had the highest mean density of San Marcos salamanders and the highest number of total observed salamanders within a zone. The riverine portion exhibited much less spatial variation of mesohabitat types. Approximately 50% of the sampled area in the riverine section was dominated by boulder or cobble substrates and had the second highest mean density of salamanders (1.4 m^{-2}) and the second highest total number of salamanders throughout the study period (n = 62). Thus, heterogeneity in the distribution and abundance of San Marcos salamanders within the managed critical habitat is driven by the distribution of specific mesohabitat types within the larger critical habitat area.

Results of this study indicate that the San Marcos salamander has a restricted distribution within its designated larger critical habitat. In particular, the limited distribution of San Marcos salamanders within the Spring Lake shows that the salamander occupies only a fraction of the designated and protected critical habitat. The mesohabitats occupied by salamanders (i.e. areas dominated by cobble, gravel or boulder substrates and low macrophyte cover) within the lake are located generally in the headwaters portion of the lake (zone 1) and along the remnant river channel along a narrow corridor (approximately 1–2 m wide) that runs longitudinally down the lake. These areas constitute only a small fraction of the entire critical habitat in Spring Lake. Based on data from the original mesohabitat assessment and the additional survey, an estimate that about 14% of the total area within the Spring Arm of the lake (approximately 5475 m^2 of 39 109 m^2) would be considered mesohabitats associated with San Marcos salamanders (i.e. habitats composed of cobble, gravel or boulder substrates). In addition, the Slough Arm of the lake (39 960 m^2) is almost completely dominated by silt/mud substrates, dense stands of rooted, submerged, and floating macrophytes (W.H. Nowlin, unpublished data), making this entire portion of the critical habitat unlikely to be used by the San Marcos salamander. When the entire designated critical habitat is considered (i.e. all of Spring Lake including the Slough and Spring Arms, and the upper 50 m of the river; 81 519 m^2 in total), ~7% of the total area (6086 m^2) contains substrate types associated with the presence of San Marcos salamanders. In summary, these findings indicate that although all of Spring Lake and the upper 50 m of the river are considered both the entire geographic range and critical habitat for the San Marcos salamander, their distribution is limited to a substantially smaller portion of the total area.

The San Marcos salamander co-occurred in mesohabitats with a diversity of invertebrate taxa. Across both the lake and riverine portions of the habitat, the San Marcos salamander co-occurred with *Stenocypris* (an ostracod), *Hyalella* (an amphipod), *Elimnia* (a gastropod), tubellarian flatworms, and the ephemeropteran *Tricorythodes*. However, when examination of co-occurrence was made separately in the lake and the river, there were slightly different invertebrate communities. Several of the invertebrate taxa are widespread and exhibit cosmopolitan distributions across a variety of mesohabitat types. In particular, *Hyalella* is widely distributed across mesohabitats and San Marcos salamanders frequently co-occurred with it. Indeed, *Hyalella* was the most abundant invertebrate, comprising 58% of all invertebrates collected. In the lake, San Marcos salamanders co-occurred with several taxa, including *Stenocypris* and the snails *Physa* and *Elimnia*. In contrast, salamanders in the riverine portion of the critical habitat co-occurred with the snail *Tarebia*, *Tricorythodes*, and the caddisfly *Chimarra*. This difference in associated invertebrates was due presumably to the affinity of these invertebrates with their typical habitats. Ostracods and the snails *Physa* and *Elimnia* are more commonly found in slower-flowing or stagnant habitats (Johnson and Brown, 1997; Brown, 2001), whereas the snail *Tarebia* (Liu and Resh, 1997), *Tricorythodes* (Edmunds and Waltz, 1996) and *Chimarra* (Wiggins, 1996) typically occur in more lotic environments. The San Marcos salamander does not appear to exhibit co-occurrence patterns with specific invertebrate taxa, suggesting that conservation of the salamander is not tightly linked to conserving specific invertebrate groups.

Conservation implications

Globally a large number of species face increased risk of extinction, especially through the degradation or loss of habitat (Adams *et al*., 2013; IUCN, 2013). The conservation and recovery of threatened and endangered species has been a central focus of conservation research and management efforts in the USA since the ESA was enacted in 1973. Understanding linkages between local or small-scale processes and larger-scale patterns in the abundance and distribution of species (e.g. human effects across the entire geographic range of a species) is critical for setting and meeting recovery and restoration targets (Bond and Lake, 2003). Indeed, understanding how available suitable habitats are spatially or temporally distributed in larger landscapes is critical for conservation efforts for a number of species (Moore and Gillingham, 2006; Gómez-Rodríguez *et al*., 2010). Critical habitats and the biological and physical requirements of species associated with these areas are often not well understood because operational definitions, conservation approaches, and information needed to identify critical habitat are poorly elucidated. Rosenfeld and Hatfield (2006) described requisites for the identification and maintenance of critical habitats, including information on life histories, available habitat, specific recovery targets, habitat–abundance relationships, and the amount of habitat required to meet recovery goals.

In the present study, it was estimated that across the entire geographic range of the San Marcos salamander (i.e. the critical habitat area), individuals are likely to occupy <10% of the total area. Thus, preservation and management efforts should be focused not only on the larger critical habitat area, but also preserving specific mesohabitat types contained within the overall critical habitat. In addition, the extensive portion of the critical habitat not occupied by salamanders might also serve as a 'buffer' for the preferred mesohabitats. This study reinforces the need for information on the utilization of sub-areas within the larger critical habitat to inform conservation decisions and strategies. Small-bodied amphibians with spatially restricted ranges, such as the San Marcos salamander, are highly threatened and may require intense conservation efforts to maintain populations (Neckel-Oliveira *et al*., 2013).

In the present study, mesohabitat associations are probably a product of the environmental conditions in which the San Marcos salamander first evolved as a species. In particular, this is likely to be true for the observed mesohabitat associations within the lake portion of the critical habitat. Before the construction of Spring Lake dam in 1849, the San Marcos River headwaters (San Marcos Springs) were largely lotic in nature (McClintock, 1930; Kimmel, 2006), currently evidenced as the 'remnant' river channel (composed mostly of gravel, cobble, and boulders) that runs much of the length of the Spring Arm of the lake. This study found that the San Marcos salamander primarily occurs in mesohabitats that reflect these past lotic conditions and are thus constrained to this portion of the critical habitat area because of their evolutionary history. Such evolutionary and phylogenetic relationships are increasingly being used for the conservation designation of species and determining priorities for management and restoration (Sechrest *et al.*, 2002; Redding and Mooers, 2006).

The current study suggests that the distribution of the San Marcos salamander in Spring Lake shows the ‘ghost of disturbance past’ and that its present spatial distribution is probably influenced not only by its evolutionary history but also by past and existing human pressures (Knight and Arthington, 2008). Impoundment of the headwaters undoubtedly increased the overall wetted area, but this may have caused loss of some habitat (e.g. spring runs) for the San Marcos salamander. Impounding rivers decreases water velocity, increases water residence time in the impoundment, and increases the rate of siltation (Thornton, 1990). We hypothesize that decreases in spring flow resulting from groundwater extraction would lead to longer water residence times, greater siltation rates, and further decrease the abundance and distribution of San Marcos salamanders in the critical habitat.

ACKNOWLEDGEMENTS

We would like to thank Francis (Chekka) Lash, Patricia Caccavale, Valentin (Val) Cantu, and Randy Gibson for their invaluable help with the field work and David Baumgardner for several invertebrate identification verifications. This project was funded through a grant from the United States Geological Survey (USGS). The views expressed in this paper are the authors’ and do not necessarily reflect the view of the US Fish and Wildlife Service.

REFERENCES

Adams MJ, Miller DA, Muths E, Corn PE, Campbell Grant EH, Bailey LL, Fellers GM, Fisher RN, Sadinski WJ *et al.* 2013. Trends in amphibian occupancy in the United States. *PLOS One* **8**: e64347. DOI:10.137/journal.pone.0064347

Barr GE, Babbitt KJ. 2002. Effects of biotic and abiotic factors on the distribution and abundance of larval two-lined salamanders (*Eurycea bislineata*) across spatial scales. *Oecologia* **133**: 176–185.

Berkhouse CS, Fries JN. 1995. Critical thermal maxima of juvenile and adult *E. nana* (*Eurycea nana*). *Southwestern Naturalist* **40**: 430–434.

Bond NR, Lake PS. 2003. Characterizing fish-habitat associations in streams as the first step in ecological restoration. *Austral Ecology* **28**: 611–621.

Bowles BD, Sanders MS, Hansen RS. 2006. Ecology of the Jollyville Plateau salamander (*Eurycea tonkawae*: Plethodontidae) with an assessment of the potential effects of urbanization. *Hydrobiologia* **553**: 111–120.

Bowles DE, Arsuffi TL. 1993. Karst aquatic ecosystems of the Edwards Plateau of central Texas, USA: a consideration of their importance, threats to their existence, and efforts for their conservation. *Aquatic Conservation: Marine and Freshwater Ecosystems* **3**: 317–329.

Brown KM. 2001. Mollusca: Gastropoda. In *Ecology and Classification of North American Freshwater Invertebrates*, 2nd edn, Thorp JH, Covich AP (eds). Academic Press: San Diego, CA; 297–329.

Brusven MA, Meehan WR, Biggam RC. 1990. The role of aquatic moss on community composition and drift of fish-food organisms. *Hydrobiologia* **196**: 39–50.

Carignan V, Villard M-A. 2002. Selecting indicator species to monitor ecological integrity: a review. *Ecological Monitoring and Assessment* **78**: 45–61.

Chippindale PT, Price AH, Hillis DM. 1998. Systematic status of the San Marcos salamander, *Eurycea nana* (Caudata: Plethodontidae). *Copeia* **1998**: 1046–1049.

Cummins KW. 1962. An evaluation of some techniques for the collection and analysis of benthic samples with special emphasis on lotic waters. *American Midland Naturalist* **67**: 477–504.

Davic RD, Welsh HH. 2004. On the ecological role of salamanders. *Annual Review of Ecology and Systematics* **35**: 405–434.

Edmunds GF, Waltz RD. 1996. Ephemeroptera. In *Aquatic Insects of North America*, 3rd edn, Merritt RW, Cummins KW (eds). Kendall/Hunt Publishing Company: Dubuque, IA; 126–163.

Fries JN. 2002. Upwelling flow velocity preferences of captive adult San Marcos salamanders. *North American Journal of Aquaculture* **64**: 113–116.

Gómez-Rodríguez C, Díaz-Paniagua C, Bustamante J, Portheault A, Florencio M. 2010. Inter-annual variability in amphibian assemblages: implications for diversity assessment and conservation. *Aquatic Conservation: Marine and Freshwater Ecosystems* **20**: 668–677.

Harvey E, Hoekstra JM, O’Connor RJ, Fagan WF. 2002. Recovery plan revisions: progress or due process? *Ecological Applications* **12**: 682–689.

Holsinger JR, Longley G. 1980. The subterranean amphipod crustacean fauna of an artesian well in Texas. *Smithsonian Contributions to Zoology* **308**: 1–62.

Hooper DU, Chapin FS, Ewel JJ, Hector A, Inchausti P, Lavorel S, Lawton JH, Lodge DM, Loreau M, Naeem S, *et al.* 2005. Effects of biodiversity on ecosystem functioning: a consensus of current knowledge and needs for future research. *Ecological Monographs* **75**: 3–35.

Hubbs C. 1995. Springs and spring runs as unique aquatic systems. *Copeia* **4**: 989–991.

Humphreys W. 2006. Groundwater fauna. Paper prepared for the 2006 Australian State of the Environment Committee, Department of the Environment and Heritage, Canberra.

IUCN. 2013. IUCN Red List of Threatened Species. Version 2013.2. http://www.iucnredlist.org. Accessed on 17 January 2014.

Johnson PD, Brown KM. 1997. The role of current and light in explaining the habitat distribution of the lotic snail *Elimia semicarinata*. *Journal of the North American Benthological Society* **15**: 344–369.

Kimmel J. 2006. *The San Marcos: a Rivers's Story*. Texas A and M University Press: College Station, TX.

Knight JT, Arthington AH. 2008. Distribution and habitat associations of the endangered Oxleyan pygmy perch, *Nannoperca oxleyana* Whitley, in eastern Australia. *Aquatic Conservation: Marine and Freshwater Ecosystems* **18**: 1240–1254.

Krebs CJ. 1999. *Ecological Methodology*, 2nd edn. Addison-Welsey Educational Publishers: Menlo Park, CA.

Lambeck RJ. 1997. Focal species: a multi-species umbrella for nature conservation. *Conservation Biology* **11**: 849–856.

Lawler JJ, Campbell SP, Guerry AD, Kolozsvary MB. 2002. The scope and treatment of threats in endangered species recovery plans. *Ecological Applications* **12**: 663–667.

Liu HT, Resh VH. 1997. Abundance and micro distribution of freshwater gastropods in three streams of Moorea, French Polynesia. *Annals de Limnologie* **33**: 235–244.

Lucas LK. 2006. Population and conservation of Texas Hill Country Eurycea. Master's thesis, Texas State University, San Marcos.

McClintock WA. 1930. Journal of a trip through Texas and northern Mexico in 1846-1847. *Southwestern Historical Quarterly* **34**: 32–33.

Merritt RW, Cummins KW, Berg MB. 2008. *An Introduction to the Aquatic Insects of North America*, 4th edn. Kendall Hunt Publishing Company: Dubuque, IA.

Moore JA, Gillingham JC. 2006. Spatial ecology and multi-scale habitat selection by a threatened rattlesnake: the eastern Massasuaga (*Sistrurus catenatus catenatus*). *Copeia* **2006**: 742–751.

Neckel-Oliveira S., Galatti U, Faveri SB, Albareli LP, Nascimento HEM. 2013. Ecological correlates in Brazilian anurans: implications for conservation. *Amphibia-Reptilia* **34**: 217–232.

Nelson JM. 1993. Population size, distribution, and life history of *Eurycea nana* in the San Marcos River. Master's thesis, Southwest Texas State University, San Marcos.

Nielsen LA, Johnson DL. 1989. *Fisheries Techniques*. American Fisheries Society: Bethesda, MD.

Pardo I, Armitage PD. 1997. Species assemblages as descriptors of mesohabitats. *Hydrobiologia* **344**: 111–128.

Perkin JS, Bonner TH. 2010. Long-term changes in flow regime and fish assemblage composition in the Guadalupe and San Marcos Rivers in Texas. *River Research and Applications* **27**: 566–579.

Petranka JW. 1983. Fish predation: a factor affecting the spatial distribution of a stream breeding salamander. *Copeia* **1983**: 624–628.

R Development Core Team (2008). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051 -07-0, URL http://www.R-project.org.

Redding DW, Mooers AØ. 2006. Incorporating evolutionary measures into conservation prioritization. *Conservation Biology* **20**: 1670–1678.

Roca JR, Baltanas A. 1993. Ecology and distribution of Ostracoda in Pyrenean springs. *Journal of Crustacean Biology* **13**: 165–174.

Rosenfeld JS, Hatfield T. 2006. Information needs for assessing critical habitat of freshwater fish. *Canadian Journal of Fisheries and Aquatic Sciences* **63**: 638–698.

Sada DW, Fleishmen E, Murphy DD. 2005. Associations among spring-dependent aquatic assemblages and environmental land use gradients in a Mojave Desert mountain range. *Diversity and Distributions* **11**: 91–99.

Sechrest W, Brooks TM, da Fonseca GAB, Konstant WR, Mittermeier RA, Purvis A, Rylands AB, Gittleman JL. 2002. Hotspots and the conservation of evolutionary history. *Proceedings of the National Academy of Science* **99**: 2067–2071.

Semlitsch RD. 1987. Interactions between fish and salamander larvae. *Oecologica* **72**: 481–486.

Seidel RA, Lang BK, Berg DJ. 2009. Phylogeographic analysis reveals multiple cryptic species of amphipods (Crustacea: Amphipoda) in Chihuahuan Desert springs. *Biological Conservation* **142**: 2303–2311.

Sih A, Jonsson BG, Luikart G. 2000. Habitat loss: ecological, evolutionary and genetic consequences. *TREE* **15**: 132–134.

Simberloff D. 1998. Flagships, umbrellas, and keystones: is single species management passé in the landscape era? *Conservation Biology* **83**: 247–257.

Steinman AD, Boston HL. 1993. The ecological role of bryophytes in a woodland stream. *Journal of the North American Benthological Society* **12**: 17–26.

Strayer DL. 2006. Challenges for freshwater invertebrate conservation. *Journal of the North American Benthological Society* **25**: 271–287.

Suren AM. 1991. Bryophytes as invertebrate habitat in two New Zealand alpine streams. *Freshwater Biology* **26**: 399–418.

ter Braak CJF. 1986. Canonical correspondence analysis: a new eigenvector technique for multivariate direct gradient analysis in ecology. *Ecology* **67**: 1167–1179.

ter Braak CJF, Verdonschot PFM. 1995. Canonical correspondence analysis and related multivariate methods in aquatic ecology. *Aquatic Ecology* **57**: 255–289.

Thornton KW. 1990. Sedimentary processes. In *Reservoir Limnology: Ecological Perspectives*. John Wiley: New York; 43–70.

Thorp AG, Jones RG, Kelso DP. 1997. A comparison of water-column macroinvertebrate communities in beds of differing submersed aquatic vegetation in the tidal freshwater Potomac River. *Estuaries* **20**: 86–95.

Tumlison R, Cline GE, Zank P. 1990. Surface habitat associations of the Oklahoma salamander (*Eurycea tynerensis*). *Herpetologica* **46**: 169–175.

Tupa DD, Davis WK. 1976. Population dynamics of the San Marcos Salamander, *Eurycea nana* Bishop. *The Texas Journal of Science* **27**: 179–195.

USDI (United States Department of the Interior). 1980. Endangered and threatened wildlife and plants; listing of *E.*

nana as threatened, the San Marcos gambusia as endangered, and the listing of critical habitat for Texas wild rice, *E. nana*, San Marcos gambusia, and fountain darter. Federal Register 45(14 July 1980):47355–47364.

US ESA (US Endangered Species Act of 1973 as amended through the 100th Congress). 1988. US Congress, Washington, DC.

USFWS (US Fish and Wildlife Service). 1995a. Draft recovery planning guidance. US Fish and Wildlife Service, Washington, DC.

USFWS (US Fish and Wildlife Service). 1995b. San Marcos/Comal (revised) recovery plan. Albuquerque, New Mexico.

Weaver MJ, Magnuson JJ, Clayton MK. 1997. Distribution of littoral fishes in structurally complex macrophytes. *Canadian Journal of Fisheries and Aquatic Sciences* **54**: 2277–2289.

Wiggins GB. 1996. *Larvae of the North America Caddisfly Genera*, 2nd edn. University of Toronto: Toronto.