

Trophic plasticity, environmental gradients and food-web structure of tropical pond communities

CHRISTOPHER M. SCHALK* , CARMEN G. MONTAÑA*[†], KIRK O. WINEMILLER* AND LEE A. FITZGERALD*

*Department of Wildlife and Fisheries Sciences, and Biodiversity Research and Teaching Collections, Texas A&M University, College Station, TX, U.S.A.

[†]Department of Biological Sciences, Sam Houston State University, Huntsville, TX, U.S.A.

SUMMARY

1. Freshwater pond communities exhibit strong patterns in species composition in response to environmental gradients such as ecosystem size, disturbance and productivity, serving as excellent systems for studies of food-web structure.
2. We surveyed 13 ponds that varied along environmental gradients of canopy cover, pond size and hydroperiod at the beginning and end of the rainy season in the semi-arid thorn forests of the Gran Chaco ecoregion of Bolivia. We collected basal resources and consumers (tadpoles, macroinvertebrates and fishes) from these ponds and used stable isotope ratios of carbon ($^{13}\text{C}/^{12}\text{C}$) and nitrogen ($^{15}\text{N}/^{14}\text{N}$), to quantify the spatiotemporal dynamics of the food webs in these tropical ponds.
3. There were no relationships between vertical structure of the food webs and the environmental gradients associated with the ponds. Consumers within these ponds exhibited high trophic variability, with multiple taxa occupying more than one trophic position across space or time.
4. Consumers from larger ponds had a greater degree of trophic redundancy. More shaded ponds supported food webs that had lower trophic diversity. More permanent ponds had significantly greater trophic diversity as well as a greater range of basal resource diversity.
5. The breeding ponds utilised by tadpoles and macroinvertebrates are patchily distributed across space and time. In these dynamic habitats, a feeding strategy of trophic generalism and plasticity enables consumers to exploit a broad range of resources and promote species coexistence. These results suggest that high diversity in tropical ponds does not necessarily translate into specialisation of trophic function.

Keywords: canopy cover, hydroperiod, Neotropics, pond size, stable isotopes

Introduction

A fundamental goal in ecology is to determine the mechanisms responsible for community organisation and species coexistence (Chesson, 2000; Siepielski & McPeck, 2010). Studies of community structure can generate hypotheses about how and why attributes of communities allow their assembly and persistence within certain environments. Food webs are depictions of consumer–resource interactions and can provide insights as to trophic relationships and resource use among coexisting species.

Freshwater pond communities exhibit strong patterns in species composition in response to environmental gradients such as area, hydroperiod and productivity, serving as excellent systems for studies of food-web structure (Wellborn, Skelly & Werner, 1996; De Meester *et al.*, 2005; Williams, 2006; Werner *et al.*, 2007). Most studies of species inhabiting lentic waterbodies have attributed patterns of community structure as a response to pond permanence, and trade-offs that are associated with the constraints of pond drying and predation (Wellborn *et al.*, 1996). Werner *et al.* (2007) documented

Correspondence: Christopher M. Schalk, Department of Wildlife and Fisheries Sciences, and Biodiversity Research and Teaching Collections, Texas A&M University, 210 Nagle Hall, MS 2258 College Station, TX 77843, U.S.A. E-mail: schalkchris@gmail.com

a decline in amphibian richness in ponds with longer hydroperiods, which was clearly attributed to the presence of fish in more permanent ponds. In addition to pond hydroperiod, canopy cover has also been shown to affect structure of pond communities (Werner & Glennemeier, 1999; Pazin *et al.*, 2006; Binckley & Resetarits, 2007; Werner *et al.*, 2007). Patterns of amphibian distribution among ponds without fish were strongly linked with abiotic variables, specifically canopy cover and pond area (Werner *et al.*, 2007). Some species appeared to be intolerant to increased canopy cover, and changes in distribution of species among ponds were associated with increasing forest encroachment around breeding ponds (Skelly, Werner & Cortwright, 1999; Werner & Glennemeier, 1999; Binckley & Resetarits, 2007, 2009). Canopy cover affects the resource type and quality by reducing productivity and this in turn affects consumer developmental rates (Werner & Glennemeier, 1999; Grether *et al.*, 2001; Skelly, Freidenburg & Kiesecker, 2002; Halverson *et al.*, 2003).

Theoretical studies have converged on the pattern that less predictable environments (e.g. those with less predictable disturbance regimes or primary production) tend to support food webs with shorter food chains (Pimm & Lawton, 1978; Pimm, 1982). However, most empirical studies have been conducted in temperate regions (Post, 2002; McHugh, McIntosh & Jellyman, 2010; Schriever & Williams, 2013; Schriever, 2015). There is much to learn from examination of the structure of tropical pond communities and how local assemblage structure of aquatic and semi-aquatic species may or may not follow the patterns and trade-offs that are apparent in temperate systems. Tropical taxa tend to reveal greater specialisation and diversification in functional traits (Schemske *et al.*, 2009). Consumers in temporary tropical ponds, such as tadpoles, have high eco-morphological diversity compared to their temperate counterparts (Altig & Johnston, 1989; Altig & McDiarmaid, 1999). However, temporary ponds, by their very nature, eventually dry, and species occupying these habitats experience changing environmental conditions, such as reduction in habitat size and/or water quality. Harsh conditions such as these may act as a filter that restricts local assemblage composition to species that possess a similar suite of traits (Chase, 2007). In turn, these coexisting species may be functionally redundant, whereby distinct taxa fulfil similar ecological roles (Rosenfeld, 2002). Thus, different patterns may emerge when comparing food-web structure in the tropics.

Here, we analyse stable isotope ratios of major food-web components in tropical ponds to reveal variation in

food-web structure along gradients of habitat disturbance (pond drying), pond area and productivity (canopy cover). We predicted that larger and more stable ponds (i.e. more permanent) would support food webs with lower trophic redundancy and greater trophic diversity compared with more ephemeral ponds. We predicted that less productive (more shaded) ponds would have lower trophic redundancy and diversity because these have a lower availability and diversity of food resources for aquatic consumers. We further predicted that most tadpoles in these tropical ponds would be trophic specialists resulting in low trophic redundancy within local assemblages when compared to tadpoles from temperate ponds.

Methods

Study site

Our study was conducted in the semi-arid thorn forests of the Gran Chaco ecoregion of south-eastern Bolivia. The region has a warm, rainy season (November–March) and a cool, dry season (April–October). Vegetation in this region is predominately thorn forest; common tree species include *Schinopsis lorentzii* (Anacardiaceae) and *Aspidosperma quebracho-blanco* (Apocynaceae), with cacti [e.g. *Opuntia* spp. (Cactaceae), *Cleistocactus baumannii* (Cactaceae) and *Eriocereus guelichii* (Cactaceae)] and bromeliads common in the understory (Navarro & Maldonado, 2002). The study area is located in one of the most xeric regions of the Bolivian Chaco; annual rainfall and temperature average 513 mm and 24.6 °C respectively (Navarro & Maldonado, 2002).

Surveys and sample preparation

We surveyed 13 ponds at two sites, Kuaridenda (19.17°S, 62.53°W; $n = 5$ ponds) and Yapiroa (19.61°S, 62.57°W; $n = 8$ ponds) in the indigenous territory of Isono, Cordillera Province, Santa Cruz Department, Bolivia. Both natural and constructed ponds were surveyed along environmental gradients of pond permanency (mean \pm SD = 85.8 \pm 7.9 days; range = 76–93 days), pond size (mean \pm SD = 724.1 \pm 780.8 m²; range = 74.4–2441 m²) and canopy cover (mean \pm SD = 26.9 \pm 23.6%; range = 1.5–68.7%) (Appendix S1). All ponds sampled dry annually and lacked aquatic vegetation. Among frog species that breed exclusively in ponds, reproduction is most intense during the rainy season (Schalk & Saenz, 2016). All frogs in the study area are pond breeders and have exotrophic tadpoles (Perotti, 1997).

We collected tadpoles, macroinvertebrates and fishes at the beginning (December 2010/January 2011) and at the end (April 2011) of the rainy season using an area-based sampler and dip net. At the beginning of the rainy season, all ponds were sampled within 2 weeks after filling with water. Only two ponds were in close proximity to an ephemeral stream (ponds 7 and 8 were 150 and 100 m from the stream respectively), which can connect to these ponds during heavy rain events. We used a 120-L plastic cylinder (trashcan) as a drop sampler (height = 0.43 m, sampling area = 1.3 m²). We dropped the sampler every 3 m along the pond's longest axis and cleared the sampler using a dip net (mesh size = 2 mm). To ensure that all organisms had been removed from the sampler, we conducted at least 10 sweeps with the dip net and 10 additional sweeps after the last animal was collected (Werner *et al.*, 2007). During each survey period, we retained samples of tadpoles, fishes and aquatic invertebrates per pond for isotopic analyses.

Tissue samples for stable isotope analysis were taken from three specimens of each species during each survey period. In addition, we collected potential basal sources (organic sediment, seston and C3 plants) from each pond by hand. We collected seston by filtering 100 mL of water through pre-combusted Whatman GF/F filter (GE Healthcare, Little Chalfont). Samples for stable isotope analysis were preserved in salt as described by Arrington & Winemiller (2002). In the laboratory, tissues were soaked in distilled water for 4 h, and then rinsed again to remove salt. Samples were then dried at 60 °C for 48 h in a drying oven. Dried samples were ground to a fine powder with a mortar and pestle, and then stored in clean glass vials. Subsamples of each ground samples were weighed (1.5–3 mg) and packaged into a Ultra-Pure tin capsules (Costech Analytical, Valencia) and sent to the Analytical Chemistry Laboratory, Institute of Ecology, University of Georgia, for analysis of stable isotope ratios of carbon (¹³C/¹²C) and nitrogen (¹⁵N/¹⁴N). During mass spectrometry, two different standards were processed between every 12 samples, and precision was ±0.22‰ for δ¹³C and ±0.20‰ for δ¹⁵N. Isotope ratios were reported in parts per thousand (‰) standardised in relation to reference material (Pee Dee Belemnite for C, atmospheric nitrogen for N) and reported as $\delta X = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 10^3$, where $R = {}^{13}\text{C}/{}^{12}\text{C}$ or ${}^{15}\text{N}/{}^{14}\text{N}$.

We quantified environmental gradients at each pond monthly from January to April 2011. Canopy cover was estimated using a spherical densitometer with a measurement taken at every 5 m along the pond's longest axis. At each sampling site, four readings were taken,

one facing each cardinal direction. Canopy cover was estimated as the average of four readings taken at each measurement site, then averaging those to get an overall mean canopy cover for a pond (Werner *et al.*, 2007). Pond area was determined using a measuring tape to measure the pond's longest and widest axis and using the formula of an ellipse to calculate surface area. Hydroperiod (the number of days the ponds held water within the study period) was determined by checking the ponds for water approximately every 2 weeks. When a pond was found to be dry, we assumed the pond dried in the midway point between surveys.

Data analyses

Trophic position from isotopic data (TP SIA) was estimated based on fractionation of ¹⁵N between the consumer and basal production sources collected from its locality (Vander Zanden & Rasmussen, 1999; Post, 2002) using the formula:

$$\text{Trophic position} = [(\delta^{15}\text{N}_{\text{consumer}} - \delta^{15}\text{N}_{\text{reference}})/2.54] + 1$$

where δ¹⁵N_{reference} was the mean δ¹⁵N of basal sources (C3 plants, seston, sediment) and 2.54‰ is the mean trophic fractionation value (Vanderklift & Ponsard, 2003).

To compare food-web structure between ponds and periods, we used community-wide metrics based on isotopic data (Layman *et al.*, 2007). We examined the degree of trophic redundancy [mean nearest neighbour distance (NND)], evenness of trophic niches (standard deviation of NND), average degree of isotopic diversity [centroid distance (CD)], magnitude of food-web trophic diversity [convex hull of the total area (TA)], maximum vertical structure [δ¹⁵N range (NR)] and basal resource diversity [δ¹³C range (CR)] (Layman *et al.*, 2007). These metrics may be biased when isotopic signatures of basal sources are not considered, because δ¹³C values might be influenced by physicochemical and other environmental characteristics across systems (Hoeinghaus & Zeug, 2008). In our study, however, these metrics are appropriate given that between-pond variation in ratios of basal sources was minimal within a given survey period (Fig. 1 and Appendix S2). The SIAR (Stable Isotope Analysis in R) package in the R software version 3.0.2 (R Core Team, 2013) was used to calculate food-web metrics. We used generalised linear models (GLMs; using a normal distribution and link function) to explore relationships between food-web structure and environmental gradients. Because there was no relationship between pond area, hydroperiod or canopy cover (Appendix S2), we analysed the

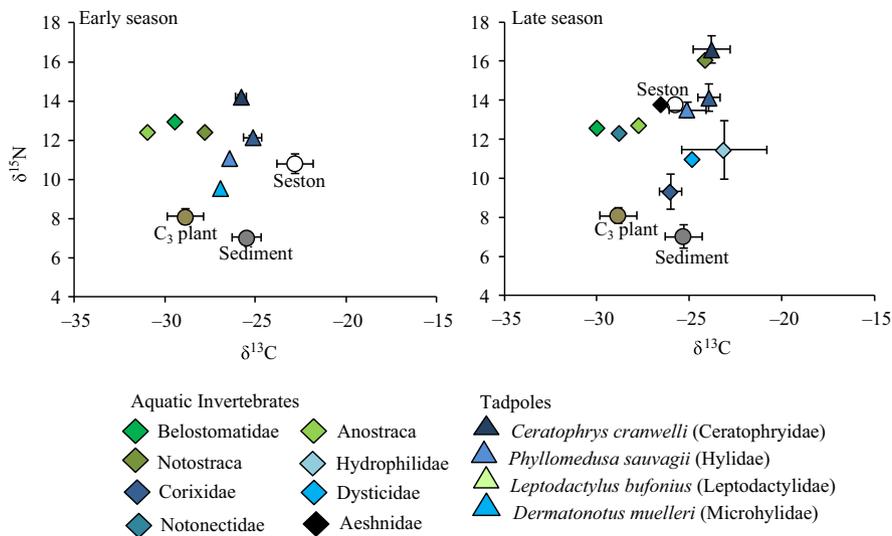


Fig. 1 Bi-plots of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ for tadpoles (triangles), aquatic invertebrates (diamonds) and three basal sources (circles) for one of the ponds (Pond 6) in the Gran Chaco ecoregion of Bolivia. Isotope ratios were compared for taxa that occurred near the early season and late season of the annual rainy season.

influence of these gradients on food-web structure separately. Data were log transformed prior to analyses that were conducted using the statistical software PAST (Hammer, Ryan & Harper, 2001).

Results

Samples from 318 consumers (early season = 152, late season = 165) and 98 basal sources (early season = 54, late season = 44) were analysed for stable isotope ratios. Consumer samples included 11 tadpole species, 5 fish species and 16 macroinvertebrate taxa. Consumers $\delta^{13}\text{C}$ ranged from -34.9 to -16.9‰ at the beginning of the rainy season, and from -32.7 to -19.7‰ at the end of rainy season. Consumer $\delta^{15}\text{N}$ ranged from 4.3 to 15.9‰ at the beginning of the rainy season, and from 5.8 to 19.2‰ at the end of rainy season. Producer $\delta^{13}\text{C}$ ranged from -31.3 to -19.4‰ at the beginning of the rainy season, and from -31.3 to -20.5‰ at the end of the rainy season. Producer $\delta^{15}\text{N}$ ranged from 4.5 to 13.4‰ at the beginning of the rainy season, and from 4.2 to 13.7‰ at the end of rainy season. Stable isotope bi-plots indicated consumers became more enriched in $\delta^{15}\text{N}$ near the end of the rainy season (Fig. 1). Most consumer $\delta^{15}\text{N}$ values were consistently higher than those of basal production sources, but there were some exceptions (e.g. in ponds 5 and 7 in the early rainy season; Appendix S2). Seston $\delta^{15}\text{N}$ was highly variable and mean values sometimes were greater than those of consumer taxa (Appendix S2).

Trophic positions

Macroinvertebrates and tadpoles revealed large isotopic variation across space and time, and the range for

estimates of trophic position for several taxa exceeded an entire level (Fig. 2). Consumers tended to occupy higher trophic positions near the end of the rainy season (Fig. 3). Near the beginning of the rainy season, mean trophic position of consumer taxa ranged from 1.3 [*Dermatoneotus muelleri* (Microhylidae) tadpoles] to 3.2 [*Neofundulus ornatipinnis* (Rivulidae) killifish]. Near the end of the rainy season, mean trophic position of consumer taxa ranged from 1.1 (Hydrophilidae beetles) to 5.7 [*Scinax nasicus* (Hylidae) tadpoles] (Fig. 2). During the early rainy season, tadpoles tended to have lower trophic positions compared to tadpoles sampled near the end of the rainy season (Fig. 3a, b) and mean trophic position of tadpoles was similar to mean values of other consumer groups during this period [tadpole mean (\pm SD) = 2.1 (± 0.4), macroinvertebrate = 2.1 (± 0.3), fish = 2.3 (± 0.6); Fig. 3a, b]. A few macroinvertebrate taxa had high trophic positions near the end of the dry season (Fig. 3a). Near the end of the rainy season, trophic positions of tadpoles tended to be higher with distributions more left skewed compared to other consumer groups (tadpole mean \pm SD = 3.2 ± 1.2 , macroinvertebrate mean \pm SD = 2.5 ± 1.0 , fish mean \pm SD = 2.3 ± 0.6 ; Fig. 3b).

Food-web structure

Food-web structure varied along environmental gradients across seasons. Near the beginning of the rainy season, consumers were significantly more packed within the isotopic space occupied by consumers with increasing pond size [GLM, $R^2 = 0.17$, $P = 0.02$; mean NND = $0.40285 - 0.16142\log_{10}(\text{area})$] (Fig. 4). There was a statistically non-significant trend showing that

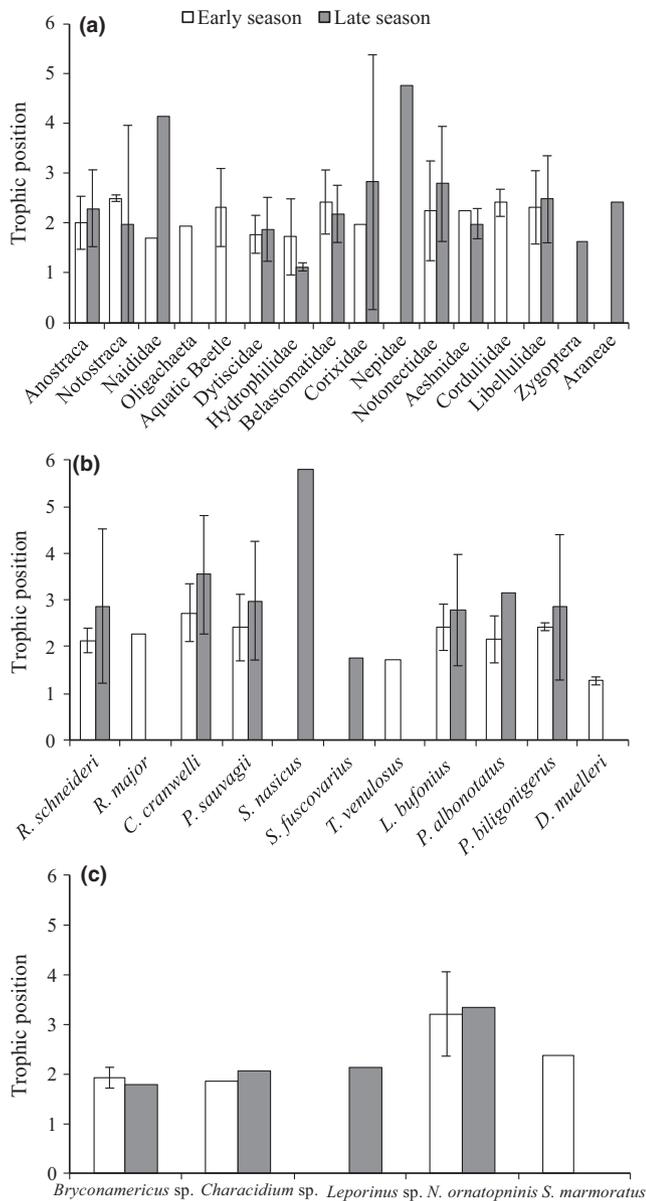


Fig. 2 Mean trophic position (± 1 standard deviation) of (a) macroinvertebrates, (b) tadpoles and (c) fish collected in 13 ponds at the early season (white bars) and late season (grey bars) of the annual rainy season. *R. schneideri* = *Rhinella schneideri*, *R. major* = *Rhinella major*, *C. cranwelli* = *Ceratophrys cranwelli*, *P. sauwagii* = *Phyllomedusa sauwagii*, *S. nasicus* = *Scinax nasicus*, *T. venulosus* = *Trachycephalus venulosus*, *L. bufonius* = *Leptodactylus bufonius*, *P. albonotatus* = *Physalaemus albonotatus*, *P. biligonigerus* = *Physalaemus biligonigerus*, *D. muelleri* = *Dermatonotus muelleri*, *N. ornatipinnis* = *Neofundulus ornatipinnis*, *S. marmoratus* = *Synbranchus marmoratus*.

consumers were more packed within isotopic space with increasing species richness (Appendix S3). Isotopic diversity was significantly lower with increasing canopy cover near the beginning of the rainy season [GLM, $R^2 = 0.47$, $P < 0.001$; $CD = 0.68649 - 0.16587 \log_{10}$

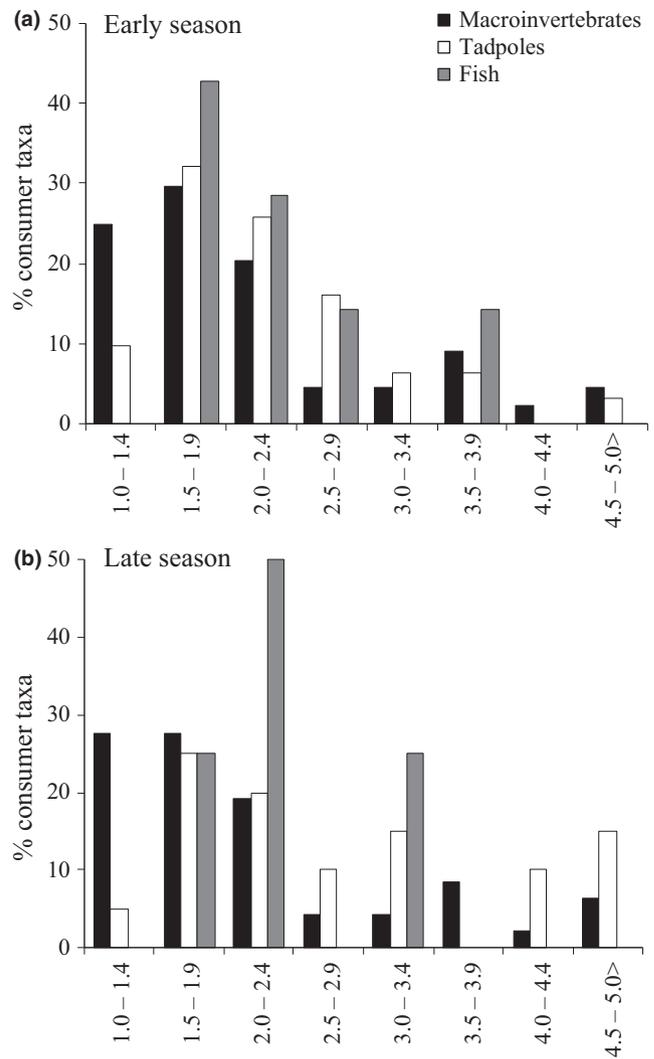


Fig. 3 Frequency histograms of trophic positions of macroinvertebrates (black bars), tadpoles (white bars) and fish (grey bars) collected across 13 ponds at the (a) early season and (b) late season of the annual rainy season.

(canopy cover)] (Fig. 4). Near the end of the rainy season, ponds with longer hydroperiods had significantly greater trophic isotopic diversity [GLM, $R^2 = 0.29$, $P = 0.047$; $CD = -1.532 + 1.088 \log_{10}$ (hydroperiod)] as well as a greater basal resource diversity as indicated by a greater range of $\delta^{13}\text{C}$ [GLM, $R^2 = 0.0073$, $P = 0.01$; range $\delta^{13}\text{C} = 1.741 - 2.3367 \log_{10}$ (hydroperiod)] (Fig. 4). There was no relationship between CR for basal sources or taxa richness and the three environmental gradients (Appendices S4 and S5 respectively). The distribution of taxa within assemblage isotopic space became less even with increasing canopy cover near the end of rainy season [GLM, $R^2 = 0.19$, $P = 0.036$; $CD = 0.13948 - 0.18091 \log_{10}$ (canopy cover)] (Fig. 4). Neither the range of isotopic diversity (range N) nor the

extent of isotopic diversity (TA) revealed significant patterns in relation to any of the three environmental gradients (Fig. 4).

Discussion

Trophic positions of macroinvertebrates and tadpoles varied considerably between ponds and within ponds between periods. Many consumer taxa would be classified as omnivores, and in some cases, spatiotemporal variation in species trophic level ranged from herbivorous to carnivorous. Tadpoles were traditionally considered to be herbivores or detritivores (Altig, Whiles & Taylor, 2007); however, our results indicate many tadpoles fed at higher trophic levels, which corroborates results from other studies of temperate tadpoles (Petranka & Kennedy, 1999; Schiesari, Werner & Kling, 2009). With the exception *N. ornatipinnis*, an annual killifish with a life cycle adapted for ephemeral aquatic habitats (Schalk, Montaña & Libson, 2014a), the fish species collected from these Chacoan ponds would have colonised them during intervals when they had surface water connections with the nearby stream. On average, *N. ornatipinnis* was a tertiary consumer, but like tadpoles and macroinvertebrates, had large within-taxon variation in trophic level during the early rainy season. While the other four fish species could be classified as herbivores or omnivores, our small sample sizes render this generalisation speculative. Although tropical tadpoles tend to reveal greater ecological specialisation in terms of functional traits (Altig & Johnston, 1989; Schemske *et al.*, 2009), our results suggest that greater eco-morphological specialisation does not necessarily translate into trophic specialisation for many tadpoles inhabiting Chacoan ponds. In these dynamic aquatic habitats, tadpoles display considerable trophic plasticity, consuming diverse resources opportunistically dependent upon availability. The flexible trophic strategy of tadpoles may explain the lack of a relationship between food-web vertical structure and environmental gradients in Chacoan ponds.

As for many animals, tadpoles have diets that are context-dependent and often mediated by the density of conspecifics, heterospecific competitors or non-consumptive effects of predators (Caut *et al.*, 2013; Greig, Wissinger & McIntosh, 2013; Arribas *et al.*, 2015). In a temperate amphibian community, the presence of a crayfish competitor caused tadpoles to shift to a more detritus-based diet and consume resources with lower $\delta^{13}\text{C}$ (Arribas *et al.*, 2015). Trade-offs between foraging activity and predation risk are well established in tadpoles inhabiting ponds in temperate regions (Werner & Anholt, 1993;

Dayton & Fitzgerald, 2001). Whether this trade-off drives variation in trophic positions of macroinvertebrates and tadpoles in tropical ponds needs to be tested experimentally (*sensu* Caut *et al.*, 2013; Arribas *et al.*, 2015). Another challenge is to understand the functional roles of diverse tadpoles in tropical assemblages (Altig *et al.*, 2007). For example, it is not known whether Chacoan tadpoles with high trophic positions function as predators (Schalk *et al.*, 2014b) or scavengers, or both depending on context.

Certain aspects of food-web structure were non-random in relation to environmental gradients associated with pond habitats. Overall, the range of $\delta^{13}\text{C}$ was positively correlated with pond hydroperiod. In temperate regions, ponds with long hydroperiods have seasonally variable seston production and inputs from leaf fall (Schriever, 2015). We did not find a relationship between the range of $\delta^{13}\text{C}$ of the basal sources and hydroperiod of Chacoan ponds, however, it cannot be ruled out that samples of one or more important sources were not included in our analysis.

Food chain length was shown to be positively correlated with hydroperiod in studies of temperate-zone ponds (Schriever & Williams, 2013; Schriever, 2015). In contrast, we found no relationship between vertical structure (range of $\delta^{15}\text{N}$) and any of the three environmental gradients tested. There are important distinctions for the way hydrology affects ecological dynamics of ponds. Temporary ponds in temperate regions dry in a more predictable seasonal manner compared to ponds in many tropical and subtropical regions (Perotti, Jara & Úbeda, 2011; Schriever & Williams, 2013; Schriever, 2015). In the Chaco, ponds are very dynamic and may dry and refill multiple times within a single rainy season (Schalk, 2016a,b; Schalk & Saenz, 2016). Selection imposed by this unpredictable variation in pond drying is reflected in the diverse oviposition strategies of the regional frog fauna. Several species lay their eggs in foam nests or terrestrial nest chambers (Crump, 1974; Duellman & Trueb, 1994; Perotti, 1997; Schalk & Saenz, 2016), which likely protects developing larvae from desiccation associated with unpredictable pond drying (Crump, 1974, 2015; Duellman & Trueb, 1994). Temperate ponds with intermediate hydroperiods that dry in a predictable fashion tend to harbour the greatest diversity of amphibians and macroinvertebrates (Werner *et al.*, 2007; Schriever & Williams, 2013; Semlitsch *et al.*, 2015). Colonisation of these habitats by species with broad trophic niches naturally leads to an increase in overall trophic diversity by the end of the rainy season.

Among-pond variation in food-web structure in the Chaco apparently is influenced by pond size as well as

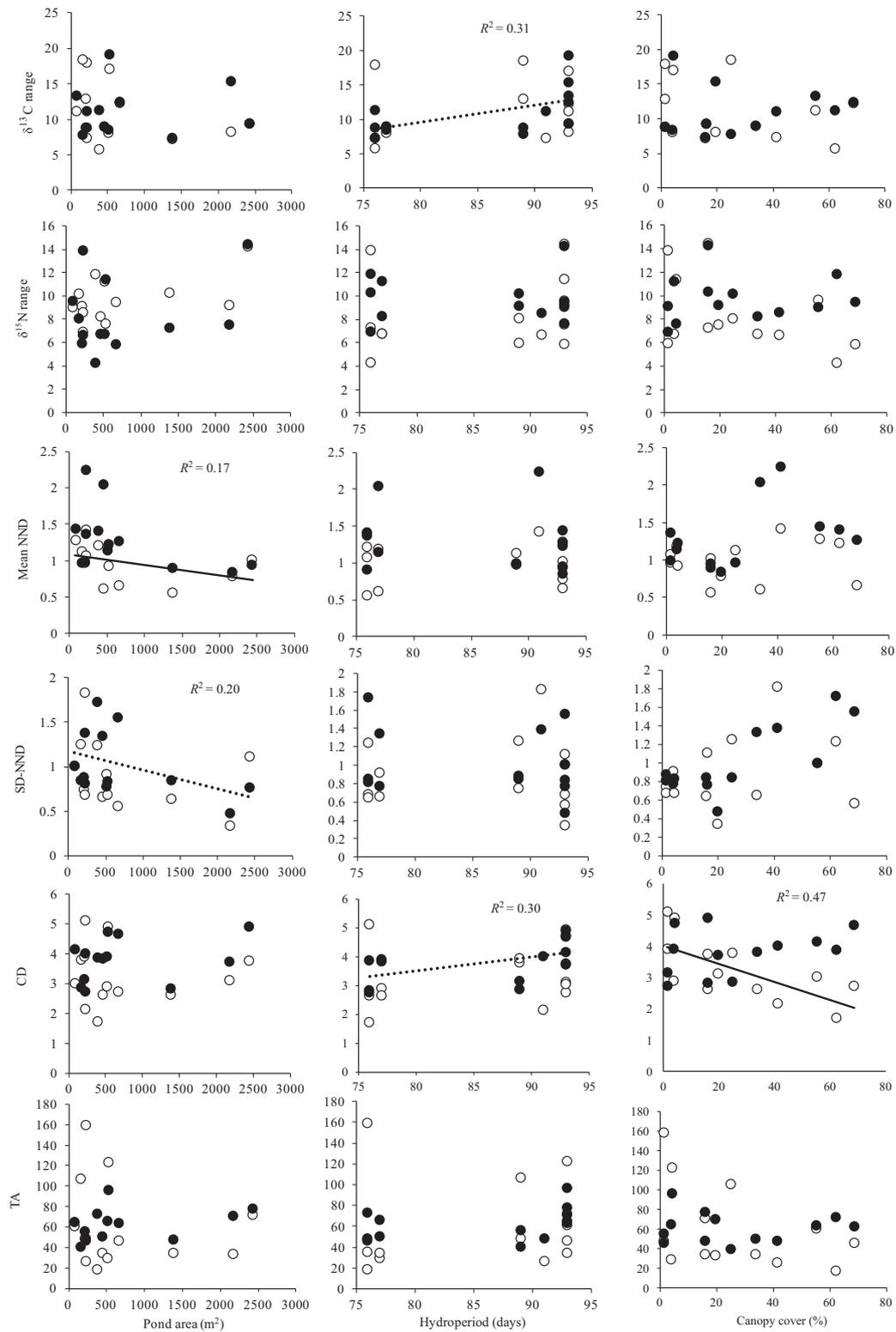


Fig. 4 Metrics of food-web structure in relation to the environmental gradients defined by pond area (first column), hydroperiod (second column) and canopy cover (third column) in the early season (open circles) and late season (filled circles) of the annual rainy season. Significant relationships between food-web structure and an environmental gradient are depicted by the presence of a solid line (early season) and dashed line (late season). NND, nearest neighbour distance; SD-NND, standard deviation of nearest neighbour distance; CD, distance to centroid; TA, total area.

trophic guild structure of local assemblages. During the early rainy season, there was evidence of higher trophic redundancy (i.e. consumers were more packed within isotopic space) with increasing pond size. Aquatic habitat area is recognised as a major factor affecting population abundance, assemblage composition and, consequently, food-web structure (McHugh *et al.*, 2015; Montaña, Layman & Winemiller, 2015; Semlitsch *et al.*, 2015). Larger habitats have higher rates of species colonisation, further contributing to a positive species–area relationship (Rosenzweig, 1995). Greater species diversity often is associated with greater functional diversity that facilitates species coexistence. We did not observe a relationship between pond area and taxon richness in Chacoan ponds. In these systems, stochastic factors may play a strong role in community assembly during the early rainy season (Chase, 2007; Chase & Myers, 2011), especially for smaller ponds that are likely to be colonised less frequently or predictably. Also, many basal resources are probably rare during initial stages of pond filling during the early wet season. Based on samples taken throughout the rainy season, there was a non-significant trend of increasing trophic redundancy in relation to taxa richness. If many consumers exploited similar resources due to low resource diversity, this also could explain high trophic redundancy. We also observed that trophic niches were more even with increasing pond size at the end of the rainy season. Since ponds were drying during this period, food resources may have been increasingly limited, thus promoting resource partitioning among consumers.

Trophic diversity was inversely correlated with canopy cover during the early rainy season. Canopy cover can be an important environmental gradient affecting aquatic ecosystems (Grether *et al.*, 2001; Mosisch, Bunn & Davies, 2001; Schiesari, 2006; Earl *et al.*, 2011). In temperate regions, pond canopy cover affects the distribution and abundance of aquatic species (Werner & Glennemeier, 1999; Skelly *et al.*, 2002; Binckley & Resetarits, 2007, 2009; Skelly, Bolden & Freidenburg, 2014). Field experiments that increased the canopy cover of tropical stream reaches reduced the abundance of primary producers, which in turn reduced the diversity and abundance of aquatic consumers (Ceneviva-Bastos & Casatti, 2014). Reduction in primary producer abundance can reduce growth rates and developmental times of consumers (Grether *et al.*, 2001). Given that most aquatic organisms in ephemeral ponds are larval stages of species with terrestrial adults, resource availability affecting growth also influences body size and timing of metamorphosis as well as mortality due to pond drying. Although we did not measure

standing crops of primary producers in these ponds or compare developmental/growth rates of taxa in shaded versus unshaded ponds, our results nonetheless are consistent with previous studies that found an inverse relationship between trophic diversity and canopy cover.

Isotopic analysis revealed that trophic plasticity and redundancy are high among tadpoles and macroinvertebrate taxa inhabiting Chacoan ponds. Some tadpole species apparently occupy multiple trophic positions, depending on time and location. Similar patterns have been observed for invertebrate communities in desert streams wherein coexisting species revealed high overlap among functional traits (Boersma *et al.*, 2013). In regions with variable and often harsh environmental conditions, such as the Gran Chaco, the regional species pool may contain taxa with high functional similarity owing to environmental filtering at a relatively large spatial scale (Chase, 2007; Schalk, Montaña & Springer, 2015; Schalk, 2016b). Because tadpoles and other aquatic consumers can affect food resource availability and quality, they can have a strong influence on aquatic food-web dynamics (Wallace & Webster, 1996; Whiles *et al.*, 2006, 2010). Moreover, pond-dwelling frogs and aquatic insects create trophic links between aquatic and terrestrial habitats (Whiles *et al.*, 2006; Earl *et al.*, 2011). Functional redundancy may provide a buffer that protects ecosystem function against species loss, however, this inference depends on the traits selected for analysis (Petchey & Gaston, 2006; Boersma *et al.*, 2013). Although we conclude that tadpoles and aquatic macroinvertebrates of Chacoan ponds have high trophic plasticity and redundancy, one also must consider that species could be dissimilar in other important functional aspects, a topic that merits further study.

Acknowledgments

We thank R.L. Cuellar and K. Rivero for providing logistical and permit support while in Bolivia. T.E. Lacher, A. Stronza and two anonymous reviewers provided constructive comments that greatly improved the manuscript. We also thank N. Angeli, D.E. Dittmer, T.J. Hibbitts, D.J. Leavitt, W.A. Ryberg, N.L. Smolensky, M.L. Treglia and M. Young for their support and insightful discussions during the course of this project. Support was provided by the National Science Foundation's Graduate Research Fellowship Program, the Applied Biodiversity Science NSF-IGERT Program at Texas A&M University (NSF-IGERT Award #0654377) and Carolyn Wierichs Kelso and George Kelso via the International Sportfish Fund. This is publication number

1533 of the Biodiversity Research and Teaching Collections at Texas A&M University.

References

- Altig R. & Johnston G.F. (1989) Guilds of anuran larvae: relationships among developmental modes, morphologies, and habitats. *Herpetological Monographs*, **3**, 81–109.
- Altig R. & McDiarmid R.W. (1999) Diversity: familial and generic characterizations. In: *Tadpoles: The Biology of Anuran Larvae* (Eds R. Altig & R.W. McDiarmid), pp. 295–337. The University of Chicago Press, Chicago.
- Altig R., Whiles M.R. & Taylor C.L. (2007) What do tadpoles really eat? Assessing the trophic status of an understudied and imperiled group of consumers in freshwater habitats. *Freshwater Biology*, **52**, 386–395.
- Arribas R., Díaz-Paniagua C., Caut S. & Gomez-Mestre I. (2015) Stable isotopes reveal trophic partitioning and trophic plasticity of a larval amphibian guild. *PLoS ONE*, **10**, e0130897.
- Arrington D.A. & Winemiller K.O. (2002) Preservation effects on stable isotope analysis of fish muscle. *Transactions of the American Fisheries Society*, **131**, 337–342.
- Binckley C.A. & Resetarits W.J. Jr (2007) Effects of forest canopy on habitat selection in treefrogs and aquatic insects: implications for communities and metacommunities. *Oecologia*, **153**, 951–958.
- Binckley C.A. & Resetarits W.J. Jr (2009) Spatial and temporal dynamics of habitat selection across canopy gradients generates patterns of species richness and composition in aquatic beetles. *Ecological Entomology*, **34**, 457–465.
- Boersma K.S., Bogan M.T., Henrichs B.A. & Lytle D.A. (2013) Invertebrate assemblages of pools in arid - land streams have high functional redundancy and are resistant to severe drying. *Freshwater Biology*, **59**, 491–501.
- Caut S., Angulo E., Díaz-Paniagua C. & Gomez-Mestre I. (2013) Plastic changes in tadpole trophic ecology revealed by stable isotope analysis. *Oecologia*, **173**, 95–105.
- Ceneviva-Bastos M. & Casatti L. (2014) Shading effects on community composition and food web structure of a deforested pasture stream: evidences from a field experiment in Brazil. *Limnologica*, **46**, 9–21.
- Chase J.M. (2007) Drought mediates the importance of stochastic community assembly. *Proceedings of the National Academy of Sciences of the United States of America*, **104**, 17430–17434.
- Chase J.M. & Myers J.A. (2011) Disentangling the importance of ecological niches from stochastic processes across scales. *Philosophical Transactions of the Royal Society of London B: Biological Sciences*, **366**, 2351–2363.
- Chesson P.L. (2000) Mechanisms of maintenance of species diversity. *Annual Review of Ecology and Systematics*, **31**, 343–366.
- Crump M.L. (1974) Reproductive strategies in a tropical anuran community. *Miscellaneous Publication – University of Kansas, Museum of Natural History*, **61**, 1–68.
- Crump M.L. (2015) Anuran reproductive modes: evolving perspectives. *Journal of Herpetology*, **49**, 1–16.
- Dayton G.H. & Fitzgerald L.A. (2001) Competition, predation, and the distributions of four desert anurans. *Oecologia*, **129**, 430–435.
- De Meester L., Declerck S., Stoks R., Louette G., Van de Meutter F., De Bie T. et al. (2005) Ponds and pools as model systems in conservation biology, ecology and evolutionary biology. *Aquatic Conservation: Marine and Freshwater Ecosystems*, **15**, 715–725.
- Duellman W.E. & Trueb L. (1994) *Biology of Amphibians*. Johns Hopkins University Press, Baltimore.
- Earl J.E., Luhring T.M., Williams B.K. & Semlitsch R.D. (2011) Biomass export of salamanders and anurans from ponds is affected differentially by changes in canopy cover. *Freshwater Biology*, **56**, 2473–2482.
- Greig H.S., Wissinger S.A. & McIntosh A.R. (2013) Top-down control of prey increases with drying disturbance in ponds: a consequence of non-consumptive interactions? *Journal of Animal Ecology*, **82**, 598–607.
- Grether G.F., Millie D.F., Bryant M.J., Reznick D.N. & Mayea W. (2001) Rain forest canopy cover, resource availability, and life history evolution in guppies. *Ecology*, **82**, 1546–1559.
- Halverson M.A., Skelly D.K., Kiesecker J.M. & Freidenburg L.K. (2003) Forest mediated light regime linked to amphibian distribution and performance. *Oecologia*, **134**, 360–364.
- Hammer Ø., Ryan P. & Harper D. (2001) PAST: paleontological statistics software package for education and data analysis. *Palaeontologia Electronica*, **4**, 9. Available at: http://palaeo-electronica.org/2001_1/past/issue1_01.htm.
- Hoeinghaus D.J. & Zeug S.C. (2008) Can stable isotope ratios provide for community-wide measures of trophic structure? Comment. *Ecology*, **89**, 2353–2357.
- Layman C.A., Arrington D.A., Montaña C.G. & Post D.M. (2007) Can stable isotope ratios provide for community-wide measures of trophic structure? *Ecology*, **88**, 42–48.
- McHugh P.A., McIntosh A.R. & Jellyman P.G. (2010) Dual influences of ecosystem size and disturbance on food chain length in streams. *Ecology Letters*, **13**, 881–890.
- McHugh P.A., Thompson R.M., Greig H.S., Warburton H.J. & McIntosh A.R. (2015) Habitat size influences food web structure in drying streams. *Ecography*, **38**, 700–712.
- Montaña C.G., Layman C.A. & Winemiller K.O. (2015) Species–area relationship within benthic habitat patches of a tropical floodplain river: an experimental test. *Austral Ecology*, **40**, 331–336.
- Mosisch T.D., Bunn S.E. & Davies P.M. (2001) The relative importance of shading and nutrients on algal production in subtropical streams. *Freshwater Biology*, **46**, 1269–1278.

- Navarro G. & Maldonado M. (2002) *Geografía ecológica de Bolivia: Vegetación y ambientes acuáticos*. Centro de Ecología Simón I. Patiño, Departamento de Difusión, Santa Cruz.
- Pazin V.F.V., Magnusson W.E., Zuanon J. & Mendonça F.P. (2006) Fish assemblages in temporary ponds adjacent to 'terra-firme' streams in Central Amazonia. *Freshwater Biology*, **51**, 1025–1037.
- Perotti M.G. (1997) Modos reproductivos y variables reproductivas cuantitativas de un ensamble de anuros del Chaco semiárido, Salta, Argentina. *Revista Chilena de Historia Natural*, **70**, 277–288.
- Perotti M.G., Jara F.G. & Úbeda C.A. (2011) Adaptive plasticity of life-history traits to pond drying in three species of Patagonian anurans. *Evolutionary Ecology Research*, **13**, 415–429.
- Petchey O.L. & Gaston K.J. (2006) Functional diversity: back to basics and looking forward. *Ecology Letters*, **9**, 741–758.
- Petranka J.W. & Kennedy C.A. (1999) Pond tadpoles with generalized morphology: is it time to reconsider their functional roles in aquatic communities? *Oecologia*, **120**, 621–631.
- Pimm S.L. (1982) *Food Webs*. Chapman and Hall, London.
- Pimm S.L. & Lawton J.H. (1978) On feeding on more than one trophic level. *Nature*, **275**, 542–544.
- Post D.M. (2002) Using stable isotopes to estimate trophic position: models, methods, and assumptions. *Ecology*, **83**, 703–718.
- R Core Team (2013) *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna. Available at: <http://www.R-project.org>.
- Rosenfeld J.S. (2002) Functional redundancy in ecology and conservation. *Oikos*, **98**, 156–162.
- Rosenzweig M.L. (1995) *Species Diversity in Space and Time*. Cambridge University Press, Cambridge.
- Schalk C.M. (2016a) Predator-induced phenotypic plasticity in an arid-adapted tropical tadpole. *Austral Ecology*, **41**, 415–422.
- Schalk C.M. (2016b) *Community Assembly of Neotropical Frogs Across Ecological Scales*. PhD Thesis, Texas A&M University, College Station.
- Schalk C.M., Montaña C.G. & Libson M. (2014a) Reproductive strategies of two Neotropical killifish, *Austrolebias vandenbergi* and *Neofundulus ornatipinnis* (Cyprinodontiformes: Rivulidae) in the Bolivian Gran Chaco. *Revista de Biología Tropical*, **62**, 109–117.
- Schalk C.M., Montaña C.G., Klemish J.L. & Wild E.R. (2014b) On the diet of the frogs of the Ceratophryidae: synopsis and new contributions. *South American Journal of Herpetology*, **9**, 90–105.
- Schalk C.M., Montaña C.G. & Springer L. (2015) Morphological diversity and community organization of desert anurans. *Journal of Arid Environments*, **122**, 132–140.
- Schalk C.M. & Saenz D. (2016) Environmental drivers of anuran calling phenology in a seasonal Neotropical ecosystem. *Austral Ecology*, **41**, 16–27.
- Schemske D.W., Mittelbach G.G., Cornell H.V., Sobel J.M. & Roy K. (2009) Is there a latitudinal gradient in the importance of biotic interactions? *Annual Review of Ecology, Evolution, and Systematics*, **40**, 245–269.
- Schiesari L. (2006) Pond canopy cover: a resource gradient for anuran larvae. *Freshwater Biology*, **51**, 412–423.
- Schiesari L., Werner E.E. & Kling G.W. (2009) Carnivory and resource-based niche differentiation in anuran larvae: implications for food web and experimental ecology. *Freshwater Biology*, **54**, 572–586.
- Schriever T.A. (2015) Food webs in relation to variation in the environment and species assemblage: a multivariate approach. *PLoS ONE*, **10**, e0122719.
- Schriever T.A. & Williams D.D. (2013) Influence of pond hydroperiod, size, and community richness on food-chain length. *Freshwater Science*, **32**, 964–975.
- Semlitsch R.D., Peterman W.E., Anderson T.L., Drake D.L. & Ousterhout B.H. (2015) Intermediate pond sizes contain the highest density, richness, and diversity of pond-breeding amphibians. *PLoS ONE*, **10**, e0123055.
- Siepielski A.M. & McPeck M.A. (2010) On the evidence for species coexistence: a critique of the coexistence program. *Ecology*, **91**, 3153–3164.
- Skelly D.K., Bolden S.R. & Freidenburg L.K. (2014) Experimental canopy removal enhances diversity of vernal pond amphibians. *Ecological Applications*, **24**, 340–345.
- Skelly D.K., Freidenburg L.K. & Kiesecker J.M. (2002) Forest canopy and the performance of larval amphibians. *Ecology*, **83**, 983–992.
- Skelly D.K., Werner E.E. & Cortwright S.A. (1999) Long-term distributional dynamics of a Michigan amphibian assemblage. *Ecology*, **80**, 2326–2337.
- Vander Zanden M.J. & Rasmussen J.B. (1999) Primary consumer $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ and the trophic position of aquatic consumers. *Ecology*, **80**, 1395–1404.
- Vanderklift M.A. & Ponsard S. (2003) Sources of variation in consumer-diet $\delta^{15}\text{N}$ enrichment: a meta-analysis. *Oecologia*, **136**, 169–182.
- Wallace J.B. & Webster J.R. (1996) The role of macroinvertebrates in stream ecosystem function. *Annual Review of Entomology*, **41**, 115–139.
- Wellborn G.A., Skelly D.K. & Werner E.E. (1996) Mechanisms creating community structure across a freshwater habitat gradient. *Annual Review of Ecology and Systematics*, **27**, 337–363.
- Werner E.E. & Anholt B.R. (1993) Ecological consequences of the trade-off between growth and mortality rates mediated by foraging activity. *The American Naturalist*, **142**, 242–272.
- Werner E.E. & Glennemeier K.S. (1999) Influence of forest canopy cover on the breeding pond distributions of several amphibian species. *Copeia*, **1999**, 1–12.
- Werner E.E., Skelly D.K., Relyea R.A. & Yurewicz K.L. (2007) Amphibian species richness across environmental gradients. *Oikos*, **116**, 1697–1712.

- Whiles M.R., Gladyshev M.I., Sushchik N.N., Makhutova O.N., Kalachova G.S., Peterson S.D. *et al.* (2010) Fatty acid analyses reveal high degrees of omnivory and dietary plasticity in pond dwelling tadpoles. *Freshwater Biology*, **55**, 1533–1547.
- Whiles M.R., Lips K.R., Pringle C.M., Kilham S.S., Bixby R.J., Brenes R. *et al.* (2006) The effects of amphibian population declines on the structure and function of Neotropical stream ecosystems. *Frontiers in Ecology and the Environment*, **4**, 27–34.
- Williams D.D. (2006) *The Biology of Temporary Waters*. Oxford University Press, New York.

Supporting Information

Additional Supporting Information may be found in the online version of this article:

Appendix S1. Mean values of the environmental gradients (canopy cover, pond area and pond hydroperiod) of 13 ponds sampled in the Gran Chaco ecoregion, Bolivia. Variables were collected monthly from December 2010 to April 2011.

Appendix S2. Bi-plots of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ for consumers and basal sources for the remaining ponds surveyed in the study area. Isotope ratios were compared for certain

taxa that occurred in the early season and late season of the annual rainy season. The bi-plots for pond 6 are depicted in Fig. 1 of the main text.

Appendix S3. Bi-plots of the relationship between taxa richness and mean nearest neighbour distance (NND) a proxy for trophic redundancy for consumers collected during the early season (open circles) and late season (filled circles) of the annual rainy season.

Appendix S4. Bi-plots of the relationship between $\delta^{13}\text{C}$ range of the basal sources and the environmental gradients of (A) pond area, (B) pond hydroperiod and (C) canopy cover for consumers collected during the early season (open circles; solid trendline) and late season (filled circles; dashed trendline) of the annual rainy season.

Appendix S5. Bi-plots of the relationship between taxa richness and the environmental gradients of (A) pond area, (B) pond hydroperiod and (C) canopy cover for consumers collected during the early season (open circles; solid trendline) and late season (filled circles; dashed trendline) of the annual rainy season.

(Manuscript accepted 16 November 2016)