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# Can translocated native fishes retain their trophic niche when confronted with a resident invasive?

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Abstract – Diet interactions between native and non-native fishes may influence the establishment of native species within their historical range (i.e., reintroduction). Therefore, we illustrated the food web structure of and followed the transition of the federally endangered humpback chub *Gila cypha* into a novel food web following translocation and determined the potential for a non-native species, rainbow trout *Oncorhynchus mykiss*, to influence translocation success. Humpback chub and rainbow trout used resources high in the food web and assimilated similar proportions of native fishes, suggesting non-native rainbow trout may occupy an ecological role similar to humpback chub. Subsequently, humpback chub may be well suited to colonise tributaries because of their ability to consume resources high in the food web. Additionally, diet partitioning may occur between all members of the fish community as indicated by separation in trophic niche space and little trophic overlap; although all species, particularly bluehead sucker *Catostomus discobolus*, used a broad range of food resources. Rainbow trout stomach content analysis corroborated stable isotope analysis and suggested rainbow trout diet consisted of aquatic and terrestrial macroinvertebrates, while larger rainbow trout (>120 mm total length) consumed a greater proportion of fish (incidence of piscivory = 5.3%). Trophic interactions may reveal an underutilized niche space or biotic resistance to the establishment of translocated native fishes. Continued translocation of humpback chub into tributaries appears to be one option for conservation. However, successful establishment of humpback chub may depend on continued removal of non-native trout, increasing availability of diet sources at higher trophic levels.

Key words: humpback chub; translocation; reintroduction; isotopes; food web

## Introduction

Conservation of endangered fishes may be facilitated by the establishment of species into portions of their historical ranges (i.e., reintroduction) (Minckley 1995; Olden et al. 2011). However, similar to invasion success of non-native fishes, trophic interactions with the fish community may influence where and whether establishment of native fishes is feasible (Gido & Franssen 2007; Jackson et al. 2012; Jackson & Britton 2013). Therefore, an increased understanding of the trophic structure and how endangered species settle into food webs of established freshwater fish communities where translocations are to occur

may aid translocation efforts (Vander Zanden et al. 2003; Armstrong & Seddon 2008) by providing insight into community interactions and population dynamics (Burress et al. 2013).

Trophic interactions with non-native fishes are implicated in the declines in native fishes globally (Gozlan et al. 2010). In the Southwest USA, predation is in part responsible for the decline or extinction of the region's highly endemic native fish fauna (Blinn et al. 1993; Bestgen et al. 2006; Propst et al. 2008). Non-native fishes may also consume food resources at similar trophic levels to many native species and force natives to consume resources at lower trophic levels and lead to reduced condition and

growth (Pilger et al. 2010; Walsworth et al. 2013), particularly in relatively simple foods webs typical in Southwest USA (Tyus & Saunders 2000; Cross et al. 2013). Therefore, effectively implementing management and conservation strategies to restore the range of native fish populations may depend on trophic interactions between native and non-native fishes.

Non-native fish removal has been used extensively in the Southwest USA (Coggins et al. 2011) and may facilitate translocation of native fishes back into their historical ranges (Marsh et al. 2005). However, complete eradication of non-native fishes is difficult and thus often results in incomplete removal (Mueller 2005; Meyer et al. 2006). As a result, managers may revert to suppressing non-native abundances potentially reducing threats to newly established native populations (Peterson et al. 2008; Propst et al. 2008) although direct predation or trophic overlap by non-native species is one cause of failed translocations to establish new populations (Harig et al. 2000; Vander Zanden et al. 2003). Therefore, when non-native fishes are present at release sites, managers must decide the severity of risks and whether or not to proceed with translocations involving potentially limited numbers of native fishes.

The federally endangered humpback chub is endemic to the Colorado River of the Southwest USA with the largest extant population near the confluence of the Colorado and Little Colorado rivers within Grand Canyon National Park, Arizona. Successful humpback chub reproduction and recruitment occurs in the Little Colorado River; however, recruitment is limited in the mainstem Colorado River downstream of Glen Canyon Dam, due to altered temperature and flow regimes and by predation and competition from non-native fishes (Paukert et al. 2006; Yard et al. 2011). Nevertheless, aggregations of juvenile and adult humpback chub are found in several reaches of the Colorado River outside the Little Colorado River (Valdez & Ryel 1995; Paukert et al. 2006).

Translocation of humpback chub into tributaries of the Colorado River may be an important management option in humpback chub recovery as these fish use these tributaries to carry out many life history stages (Hayden et al. 2013; Trammell et al. 2012). Tributaries may possess suitable habitat, water temperature and food resources to support humpback chub populations and may also serve as grow-out locations to augment mainstem Colorado River aggregations. Translocation efforts to either establish an independent humpback chub population or provide rearing opportunities in tributary systems will depend on humpback chub exploiting available food resources. However, trophic interactions with non-native fishes may reduce survival and increase emigration from the system which may prevent initial establishment (Pine et al. 2013).

## Species trophic interactions following translocation

Conservation measures such as translocation and non-native fish control to restore native fish communities in tributaries of the Colorado River may benefit from an increased understanding of the trophic structure and interactions among species in these environments. Therefore, our primary objectives were to (i) determine where humpback chub transfer to a novel food web with respect to native and non-native members of the fish community and their potential food sources and (ii) determine how non-native rainbow trout may impede translocation efforts to establish an additional humpback chub population (e.g., predator–prey interactions; diet overlap with native fishes).

## Methods

### Study site

Shinumo Creek is a 20-km-long tributary of the Colorado River within Grand Canyon National Park, Arizona, USA, with a drainage area of 220 km$^2$ (Fig. 1). The mean stream width is 4 m, and the riparian zone consists of a densely vegetated understory (horsetail – *Equisetum*), and overhanging trees (willow – *Salix*; cottonwood – *Populus*). The creek maintains a perennial flow regime from numerous spring inflows, snowmelt and late summer (i.e., July–September) monsoons. Seasonal water temperatures range from 2° to 25° C (National Park Service; unpublished data). A 4-m waterfall near the mouth of Shinumo Creek isolates the fish community from further colonisation by mainstem Colorado River fishes.

Prior to humpback chub translocation, the fish community in Shinumo Creek consisted of native bluehead sucker, speckled dace *Rhinichthys osculus* and established non-native rainbow trout, which were intentionally stocked beginning in 1925 (Williamson & Tyler 1932). A total of 902 juvenile humpback chub (60–200 mm total length) were translocated into Shinumo Creek in June 2009 (302 fish), June 2010 (300 fish) and June 2011 (300 fish). Valdez et al. (2000) estimated a carrying capacity of approximately 200 adult humpback chub in Shinumo Creek based on the relative size of Shinumo Creek compared to the much larger Little Colorado River. A carrying capacity for small tributary streams within Grand Canyon National Park based on food availability has not been estimated; however, tributary streams may support high invertebrate densities (Whiting et al. 2014). Before translocation, humpback chub were not recorded in surveys above the waterfall separating Shinumo Creek from the mainstem Colorado River. During our study, a total of 21,521 fish were sampled consisting of two resident native species (speckled dace 87% and bluehead sucker 5%) and both translo-
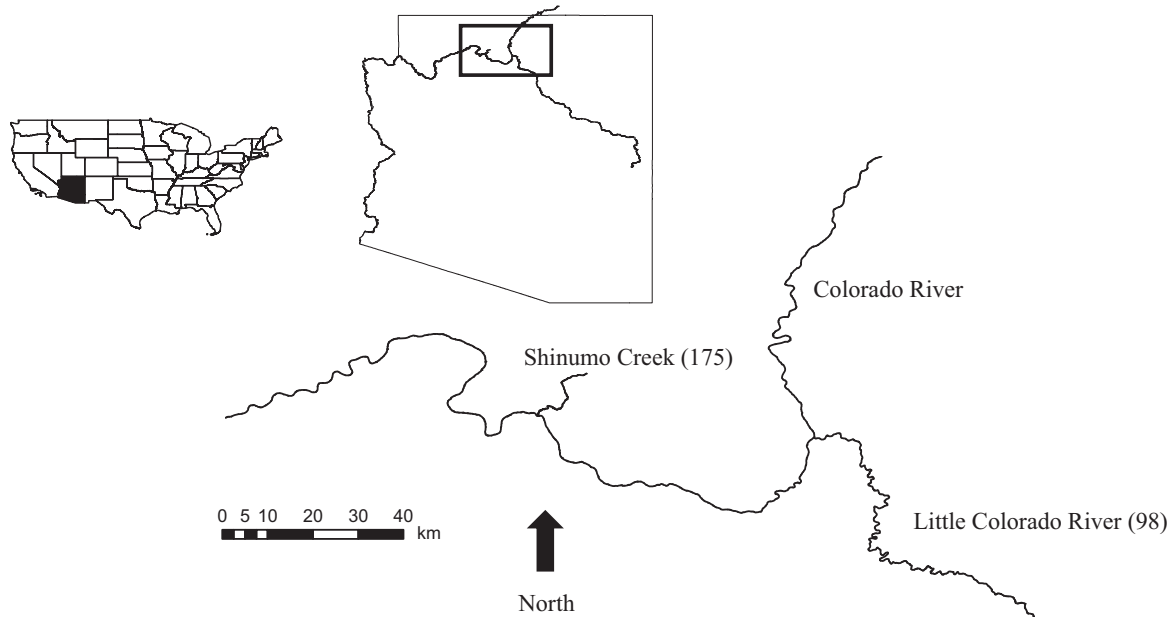
*Fig. 1.* Location of Shinumo Creek in comparison to the Little Colorado River, the source population for Humpback Chub translocated during this study. Parentheses indicate the approximate river kilometer along the Lower Colorado River of both Shinumo Creek and the Little Colorado River.

cated humpback chub (2%) and non-native rainbow trout (6%) (Healy et al. 2011).

## Sample design

We sampled 6 days each in June and September 2010 and 2011 with a combination of gear types to ensure a complete representation of the Shinumo Creek fish community. Hoop nets ($N = 27$, $50 \times 100$ cm, 6-mm mesh, single 10-cm throat) and minnow traps ($N = 54$, $25 \times 25 \times 43$ cm, 3.18-mm mesh) baited with Purina AquaMax© trout food were placed in pool and run habitats overnight during each sampling period. We also used backpack pulsed-DC electrofishing (LR-20B Smith-Root Inc., 250–350 Volts; 35% for duty cycle; and 30 Hz frequency) and angling (artificial lures and flies).

## Community stable isotope data

We used stable isotope analysis to reconstruct the trophic structure and determine potential diet sources of the Shinumo Creek fish community. We collected pelvic fin tissue from humpback chub, rainbow trout, speckled dace and bluehead sucker (Walsworth et al. 2013). Fin tissue is a suitable substitute for muscle tissue (Shannon et al. 2001; Hanisch et al. 2010) and is less invasive and thus more suitable for endangered species (Andvik et al. 2010; Jardine et al. 2011). We collected >20 fin clips (Vinson & Budy 2011) from a range of sizes of each species to maximise variability in $\delta^{15}N$ and $\delta^{13}C$ values and provide a more repre-

sentative trophic structure that included seasonal and ontogenetic differences. Tissue samples were preserved in the field using salt, which does not affect carbon or nitrogen isotopic values (Arrington & Winemiller 2002).

We also collected aquatic and terrestrial macroinvertebrates and organic matter to determine potential diet sources among the fish community. We sampled dominant aquatic invertebrate groups with a Surber sampler and kick net in riffle, run and pool habitats each; we used sweep nets and beating sheets to collect terrestrial invertebrates along the riparian and upland vegetation zones. We separated aquatic macroinvertebrate predators from nonpredators, placed each in jars containing freshwater and allowed evacuation of gut contents for several hours to ensure stomach contents did not contaminate invertebrate tissue samples (Jardine et al. 2005; Hershey et al. 2007). Periodically changing the water in the jar prevented coprophagy. We identified invertebrates to order and preserved them in the field using salt (Arrington & Winemiller 2002). We collected organic matter, which included willow and cottonwood leaves, in-stream detritus, and algae (e.g., filamentous and Nostoc), dried samples in the field using aluminium foil and sunlight, and stored samples until further processing in the laboratory.

In the laboratory, we dried fin tissue, invertebrates and organic matter at 60 °C for 48 h and homogenised material using a mortar and pestle. We did not acidify samples due to the increased variability imposed on $\delta^{15}N$ values (Bunn et al. 1995; Pinnegar

& Polunin 1999). We packed samples (2 mg fin tissue and invertebrates; 4 mg organic matter) into pressed tin capsules and determined carbon and nitrogen stable isotopic composition using a Carlo Erba NA 1500 elemental analyser coupled to a Finnigan Delta Plus XL mass spectrometer via the ConFlo III interface. Replicate analysis of an acetanilide standard yielded standard deviation estimates of 0.1‰ for both $\delta^{13}C$ and $\delta^{15}N$ values. We analysed a total of 457 isotope samples from 347 fish, 74 invertebrate and 36 organic matter samples.

We constructed $\delta^{13}C$ and $\delta^{15}N$ bi-plots with standard ellipses and convex hull polygons for each member of the Shinumo Creek fish community. In addition, we adopted metrics from Layman et al. (2007), to quantify trophic structure and highlight resource overlap among members of the fish community (Jackson et al. 2012). These metrics were bootstrapped ($N = 10,000$ indicated with a subscript '$_b$') to allow comparison among populations in the fish community as sample sizes varied between the years (Feiner et al. 2013; Jackson & Britton 2013). Metrics included trophic niche width, the ranges in $\delta^{13}C$ ($CR_b$) and $\delta^{15}N$ ($NR_b$), population niche size, the standard ellipse area of isotope values corrected for small sample size (SEAc), trophic diversity, the mean distance to centroid ($CD_b$), trophic evenness and the standard deviation of the mean nearest neighbour distance ($SDNND_b$). We also calculated the per cent of niche overlap between bluehead sucker, speckled dace and rainbow trout with respect to humpback chub using the overlap of SEAc values (Jackson & Britton 2013). We estimated standard ellipse area, convex hull polygons and population metrics using Stable Isotope Analysis in R.

We examined differences among rainbow trout, humpback chub, speckled dace, and bluehead sucker $\delta^{13}C$ and $\delta^{15}N$ values using multivariate analysis of variance (R, Version 3.0; MANOVA) and used pairwise comparisons (R, Version 3.0; MANOVA, test = Wilks) to identify where differences in mean $\delta^{13}C$ and $\delta^{15}N$ values among species existed. We used a Bonferronni correction factor to account for possible type I error associated with multiple tests ($\alpha = 0.05$, number of test = 6; $\alpha_{corrected} = 0.05/6 = 0.008$). In addition, we used Spearman rank correlation (R, Version 3.0; cor.test, method = Spearman) comparing $\delta^{15}N$ and $\delta^{13}C$ values and rainbow trout length to evaluate the presence of ontogenetic shifts in diet with increasing lengths.

We used Bayesian isotopic mixing models to estimate the proportional contribution of different prey groups to humpback chub and rainbow trout diet (Parnell et al. 2010). Diet sources included allochthonous (e.g., cottonwood leaves) and autochthonous (e.g., filamentous algae) material, primary consumers (e.g.,

Ephemeroptera, Lepidoptera and Trichoptera), large predatory insects (Megaloptera and Odonata) and native fishes (e.g., speckled dace and bluehead sucker). The mixing model uses Markov Chain Monte Carlo simulation to produce values of dietary proportions of sources based on consumer and potential prey isotopic data. A primary benefit of these models is the inclusion of variability in model parameters (Parnell et al. 2010). We considered variation in $\delta^{13}C$ and $\delta^{15}N$ values for consumers and diet sources, the elemental concentration dependence for diet sources and the trophic enrichment factor (TEF). We used TEF values of $1.0 \pm 1.0‰$ for $\delta^{13}C$ and $3.3 \pm 1.0‰$ for $\delta^{15}N$, as these values are similar to commonly reported factors (Post 2002), those used in food web studies (Eloranta et al. 2011), and also include the variation of fractionation values observed by McCutchan et al. (2003) in a meta-analysis. The resulting probability density distributions were used to compare contributions of different source groups to humpback chub and rainbow trout diet. We estimated Bayesian mixing models using SIAR (Parnell et al. 2010).

### Non-native fish diet

We assessed rainbow trout diet to corroborate stable isotope analysis and obtain a finer resolution for prey type and predation levels on native fishes that cannot be provided by stable isotope analysis alone (Davis et al. 2012). We quantified rainbow trout diet from stomach contents collected from the anterior portion of the gut to the first bend of the digestive tract (Pilger et al. 2010). Stomach contents were sorted, identified to order and enumerated. We excluded rainbow trout captured in hoop nets and minnow traps from stomach content analysis because piscivory rates may be biased for fish collected with trap nets (Stone & Gorman 2006).

### Results

#### Fish community trophic structure

Humpback chub and rainbow trout consumed resources at higher trophic levels than speckled dace and bluehead sucker (Fig. 2). In 2010, rainbow trout trophic niche size, measured by SEAc, overlapped with humpback chub; however, little overall overlap in food resource use occurred among species (Table 1). Bluehead sucker used the greatest range in carbon sources, which contributed to their overall large isotopic niche width (Table 1). Overall, diversity of diets was similar among species ($CD_b$ and $SDNND_b$, Table 1). Humpback chub had the lowest overall isotopic niche width compared to other members of the fish community (SEAc, Table 1). All

members of the Shinumo Creek fish community differed with respect to their positions in multivariate isotope space in both 2010 (d.f. = 3, Wilks = 0.35, $F = 22.26$, d.f. = 6,200, $P < 0.0001$) and 2011 (d.f. = 3, Wilks = 0.27, $F = 44.35$, d.f. = 6,296, $P < 0.0001$). Comparisons among all species indicated $\delta^{15}N$ and $\delta^{13}C$ values differed (MANOVA; $F = 16.4$, $P < 0.0001$) except between speckled dace and bluehead sucker (MANOVA; $F = 2.4$, $P = 0.1079$). Rainbow trout $\delta^{13}C$ values increased with fish length in 2010 ($r = 0.52$, $P < 0.0010$; length range: 73 to 344 mm TL), but not in 2011 ($r = 0.16$, $P = 0.1717$; length range: 79–324 mm TL). Rainbow trout $\delta^{15}N$ values increased with fish length in 2011 ($r = 0.68$, $P < 0.0001$), but not in 2010 ($r = 0.02$, $P = 0.9067$).

Therefore, small rainbow trout may be consuming similar diet items as larger individuals. The SIAR isotope mixing models suggested humpback chub and rainbow trout may be assimilating similar proportions of native fishes at higher levels compared to other diet sources; all other diet sources contributed relatively similar proportions to both species (Fig. 3, Table 2).

### Non-native fish feeding strategy

We examined a total of 151 rainbow trout stomachs (June 2010, $N = 15$; September 2010, $N = 49$; June 2011, $N = 43$; and September 2011, $N = 44$). Rainbow trout consumed a combination of fish and eleven
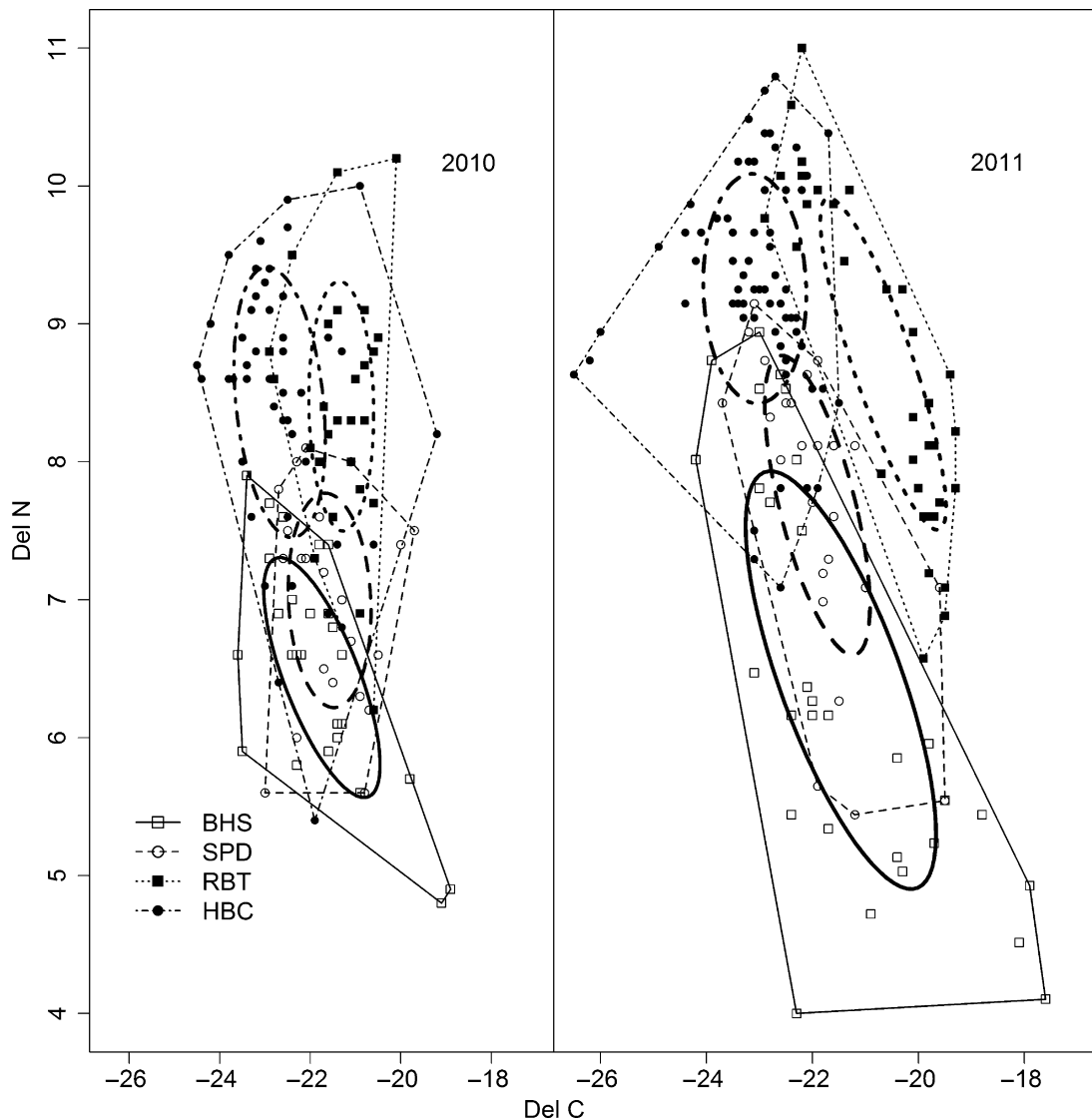


*Fig. 2.* Stable isotope bi-plot comparing $\delta^{13}C$ and $\delta^{15}N$ values in 2010 and 2011 for each species of the Shinumo Creek fish community (BHS, bluehead sucker; SPD, speckled dace; RBT, rainbow trout; HBC, humpback chub). Standard ellipses are corrected for small sample size (SEAc) and denote the main niche area of each species while the convex hulls surround all isotopic observations of each species and are indicative of overall niche diversity. Both SEAc and the convex hull area were estimated using methods in stable isotope analysis in R (SIAR).
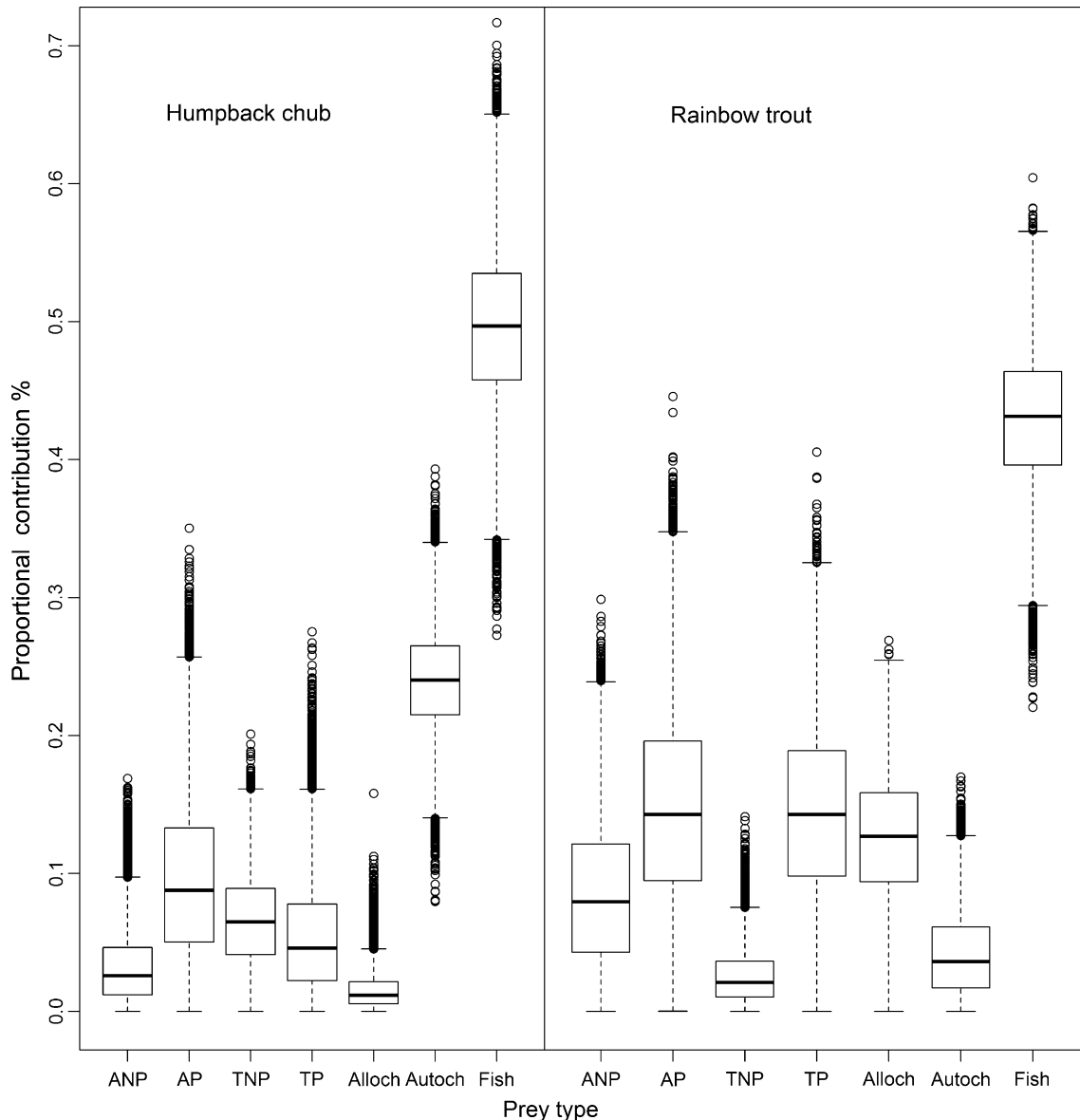
*Fig. 3.* Proportional contribution of prey types to rainbow trout and humpback chub diet from Shinumo Creek, 2010 and 2011. Bayesian isotopic mixing models were used to determine the proportional contribution of each prey type (Parnell et al. 2011). Prey codes are: ANP, aquatic non-predators; AP, aquatic predators; TNP, terrestrial non-predators; TP, terrestrial predators; Alloch, allochthonous material including cottonwood and willow leaves, grasses, and detritus; Autoch, autochthonous material including filamentous algae and nostoc; Fish, native speckled dace and bluehead sucker.

different orders of aquatic and terrestrial macroinvertebrates. Aquatic invertebrates (e.g., Trichoptera, Megaloptera, Ephemeroptera and Diptera) accounted for 51% and 61% of diet composition by mass in 2010 and 2011; fish accounted for 16% and 11% of diet composition by mass in 2010 and 2011. The occurrence of piscivory varied among sampling periods (June 2010 = 13%; September 2010 = 4%; June 2011 = 6.9%; and September 2011 = 2.2%) with a total of eight fish found in 151 rainbow trout stomachs (8/151 = 5.3%), six of those eight fish were identified as native resident speckled dace and

two were unidentified. Larger rainbow trout consumed more fish with 75% of rainbow trout over 200 mm TL containing fish in their stomach. However, stomachs from smaller rainbow trout (120 mm TL) also contained fish.

## Discussion

Consistent differences in isotopic values between humpback chub, rainbow trout, speckled dace and bluehead sucker suggest these species may partition food resources in Shinumo Creek, a system with

Table 1. Summary of number of isotope samples collected, mean size (mm total length) and range of fishes used and of adopted metrics from Layman et al. (2007), to quantify trophic structure and resource overlap among the fish community (Jackson et al. 2012).

| Year | Taxa | $N$ | Length | $CR_b$ | $NR_b$ | $CD_b$ | $MNND_b$ | $SDNND_b$ | $SEA_c$ | %Overlap |
|------|------|-----|--------|--------|--------|--------|----------|-----------|---------|----------|
| 2010 | BHS | 28 | 212 (88–336) | 4.6 (2.3–4.7) | 3.0 (2.0–3.1) | 1.2 (0.7–1.9) | 0.1 (0.0–0.2) | 0.2 (0.0–0.5) | 2.42 | 0 |
|      | SPD | 22 | 88 (40–170) | 3.1 (2.0–3.3) | 2.4 ( 1.7–2.5) | 1.0 (0.7–0.3) | 0.1 (0.0–0.2) | 0.2 (0.0–0.4) | 2.22 | 4 |
|      | RBT | 47 | 186 (73–344) | 4.5 (2.1–5.3) | 4.0 (2.1–4.6) | 1.2 (0.7–1.6) | 0.2 (0.0–0.3) | 0.3 (0.1–0.6) | 2.99 | 13 |
|      | HBC | 26 | 155 (110–209) | 2.7 (1.4–2.8) | 3.7 (1.9–4.4) | 0.9 (0.6–1.3) | 0.1 (0.0–0.2) | 0.2 (0.0–0.4) | 2.03 | |
| 2011 | BHS | 31 | 206 (76–298) | 6.3 (3.5–6.6) | 4.7 (3.6–4.8) | 2.0 (1.4–2.6) | 0.1 (0.0–0.3) | 0.3 (0.1–0.5) | 5.61 | 0 |
|      | SPD | 25 | 87 (23–144) | 4.0 (1.8–4.2) | 3.5 (1.9–3.6) | 1.2 (0.7–1.6) | 0.1 (0.0–0.2) | 0.2 (0.0–0.4) | 2.43 | 0 |
|      | RBT | 66 | 192 (79–324) | 4.4 (1.4–5.0) | 3.3 (1.8–3.6) | 1.0 (0.5–1.5) | 0.2 (0.1–0.3) | 0.2 (0.1–0.5) | 2.46 | 0 |
|      | HBC | 33 | 168 (124–256) | 3.5 (2.7–3.6) | 4.0 (2.5–4.3) | 1.5 (0.9–1.9) | 0.1 (0.0–0.2) | 0.2 (0.0–0.3) | 2.21 | |

These metrics were bootstrapped ($N$ = 10,000 indicated with a subscript '$_b$'). Parentheses indicate the range of the bootstrapped results. In addition, the per cent overlap (%Overlap) of niche space ($SEA_c$) shared with humpback chub is given. Taxa codes are as follows: BHS, bluehead sucker; SPD, speckled dace; RBT, rainbow trout; HBC, humpback chub.

Table 2. Summary of aquatic and terrestrial invertebrate feeding guilds and basal food resources from Shinumo Creek.

| Diet Source | $N$ | $\delta^{13}C$ | $\delta^{13}C$ Range | $N$ | $\delta^{15}N$ | $\delta^{15}N$ Range |
|-------------|-----|----------------|----------------------|-----|----------------|----------------------|
| Fish | 110 | −21.7 (1.3) | −24.2 to −17.6 | 106 | 6.7 (1.2) | 3.6–9.0 |
| ANP | 28 | −25.8 (2.5) | −29.2 to −19.8 | 28 | 3.5 (1.6) | 0.2–6.3 |
| AP | 26 | −24.2 (1.5) | −26.0 to −21.0 | 26 | 4.9 (1.7) | 2.4–7.7 |
| TNP | 16 | −24.9 (2.5) | −29.0 to −20.3 | 16 | 2.2 (2.8) | −2.6 to 5.8 |
| TP | 14 | −24.6 (1.7) | −26.3 to −21.4 | 14 | 5.5 (2.2) | 1.0–9.1 |
| Auto | 21 | −28.3 (2.1) | −27.3 to −13.9 | 15 | −0.9 (2.4) | −1.2 to −3.3 |
| Alloch | 6 | −20.0 (5.4) | −31.5 to −24.3 | 5 | 1.6 (1.9) | −6.2 to 3.5 |

The number of samples collected ($N$), mean and standard deviation in parentheses for carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$), and ranges are given. Mean values were also the raw data used in the Bayesian stable isotopic mixing models. Codes are as follows: Fish, native fish, (speckled dace and bluehead sucker), ANP, aquatic nonpredator (Trichoptera, Lepidoptera, Ephemenoptera, Diptera and Coleoptera); AP, aquatic predator (Megaloptera, Diptera and Hemiptera); TNP, terrestrial nonpredator (Coleoptera, Hymenoptera, Orthoptera and Hemiptera); TP, terrestrial predator (Arachnida, Hemiptera, Odonata and Coleoptera); Auto, autochthonous material (Filamentous algae and Nostoc); and Alloch, allochthonous material (Willow and Cottonwood leaves, Detritus).

relatively few fish species (Nakano et al. 1992; Meeuwig et al. 2011). For instance, humpback chub and rainbow trout are consuming resources at similar trophic levels, but currently appear to be partitioning those resources. However, diet partitioning may be related to competition for food resources and predation (Nilsson 1967) and thus may change as humpback chub population abundance increases. Humpback chub had the smallest trophic niche space among all members of the fish community and were the last to be established. Rainbow trout can exclude native species from food resources (Baxter et al. 2007), presumably through behavioural dominance (Baxter et al. 2004) and could also limit niche space of newly established species. Therefore, the presence of rainbow trout or other non-native species may have direct as well as indirect cascading effects on the native resident fish community by limiting niche space (Heermann et al. 2013; Walsworth et al. 2013).

Humpback chub occupied relatively high trophic levels compared to speckled dace and bluehead sucker following translocation into Shinumo Creek. Previous studies within Grand Canyon have suggested that humpback chub feed at higher trophic levels as they get larger (>150 mm TL; Shannon et al. 2001; Bene-

nati et al. 2002) and documented piscivory in tributaries of the Colorado River (Stone 1999; Stone & Gorman 2006). Our mixing model results also suggest piscivory may be an important attribute in humpback chub life history. Additionally, our study suggests humpback chub may be well suited to colonise novel habitats because of their potential to use a broad resource pool high in the aquatic food web (Moyle & Light 1996; Olsson et al. 2009). However, similar to an invasion by a non-native species, the establishment of humpback chub, a native fish extirpated from portions of its historical range, may result in additive predation pressure upon native fish communities, particularly if non-native rainbow trout are present (Eby et al. 2006; Ricciardi & Simberloff 2009).

Humpback chub and rainbow trout appear to assimilate similar proportions of diet types and have expanded the food web of Shinumo Creek by occurring at higher trophic levels than speckled dace and bluehead sucker (Walsworth et al. 2013). Similarity in diet between the two species is consistent with other studies on the mainstem Colorado River, particularly where rainbow trout are abundant (Shannon et al. 2001; Donner 2011). Thus, rainbow trout, an abundant and widespread species within Grand Canyon (reviewed in Kennedy & Ralston 2011), may

occupy an ecological role formerly occupied by humpback chub, or other native *Gila* species (*G. robusta* or *G. elegans*), in the Colorado River or its tributaries. Therefore, if food resources become limited in areas where humpback chub and rainbow trout co-occur, reduced growth may occur and the establishment of humpback chub into Colorado River tributaries may be affected. Established non-native predators have hindered efforts to re-establish native species into their historical ranges when occupying similar trophic states (Vander Zanden et al. 2003). However, in the Southwest USA, native fishes may maintain similar growth rates in the presences of non-native fishes, but occur at lower abundance than in areas without non-native fishes (Walsworth et al. 2013). Sufficient growth and condition of humpback chub (Spurgeon 2012) suggests that food resources may not be limiting at current population abundances, diet partitioning may occur between humpback chub and rainbow trout, or humpback chub growth and condition were maintained partly as a result of mechanical (angling, electro-fishing and netting) suppression of rainbow trout that occurred concurrently with this study.

Conservation of native fishes and establishment of young-of-year or juvenile humpback chub may depend on reducing the potential of predation by non-native predators. Our results suggest larger rainbow trout may exert greater predation pressure on native fishes and corroborate those of Shannon et al. (2001) from the mainstem Colorado River and suggest rainbow trout undergo similar shifts in diet with increasing size in smaller tributary systems. Our stomach content analysis suggested rainbow trout are feeding generalists, and an ontogenetic shift to piscivory occurred sooner than previously estimated (150 mm TL, Shannon et al. 2001; Benenati et al. 2002), which is consistent with recent mainstem Colorado River work where trout as small as 100 mm TL consumed fish (Yard et al. 2011). In Shinumo Creek, seasonal and ontogenetic shifts in food sources and trophic position may occur in both native and introduced species. Therefore, temporal changes in food availability and diet overlap among fishes may occur in Shinumo Creek similar to macroinvertebrate communities in the mainstem Colorado River (Wellard Kelly et al. 2013). Cross et al. (2013) suggests large tributaries contribute a large diversity of energy sources to the mainstem Colorado River. Likewise, smaller tributary streams contain high diversity in food resources (Oberlin et al. 1999; Whiting et al. 2014) and may explain the high presence of omnivory by rainbow trout found in Shinumo Creek. The coexistence of rainbow trout and native bluehead sucker and speckled dace in Shinumo Creek over several decades may have resulted from the prevalence of omnivory and environmental stochasticity periodically reducing non-native abundance (Meffe 1984; Meffe & Minckley 1987).

Although other studies have determined how non-native fishes alter food webs (Pilger et al. 2010; Walsworth et al. 2013), few have assessed how a native, endangered fish will fit into a receiving food web following translocation into a presumably modified food web occupied by a non-native species. Our study has provided evidence that humpback chub and rainbow trout occupy similar trophic levels within Shinumo Creek, which suggests that rainbow trout are consuming food resources that could also be used by translocated humpback chub. Continued rainbow trout removal efforts may open up a food source potentially not available to humpback chub while also restoring ecological integrity. The ecological impacts of introduced species (or, in our case, an established non-native species, rainbow trout and a repatriated native species, humpback chub) may depend on the biotic and abiotic conditions of the system (Ruesink 2003). However, these initial results will be useful in understanding future trophic interactions as humpback chub abundances may increase over time through a combination of translocation and reproduction, and as rainbow trout densities are reduced through continual removal efforts.

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