Notes on the Distribution and Feeding Ecology of a Relict Population of the Cardinal Shiner, Luxilus cardinalis (Teleostei: Cyprinidae), in Kansas

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Notes on the distribution and feeding ecology of a relict population of the cardinal shiner, *Luxilus cardinalis* (Teleostei: Cyprinidae), in Kansas

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We reviewed historical patterns in distribution and investigated dietary habits of a relict population of cardinal shiner (*Luxilus cardinalis*) in southeastern Kansas. Historical (pre-1980) collections of cardinal shiner ranged throughout portions of the upper Neosho and Verdigris basins, and contemporary collections (post-1980) documented the species in many of the same locations. Reduction in cardinal shiner range has apparently occurred in the Neosho River basin just downstream of John Redmond Reservoir. Diets of 95 individuals collected from five sites in the Cottonwood River drainage of the upper Neosho River basin during summer of 2010 included primarily aquatic invertebrates (occurred in 93.7% of individuals, constituted 29.3% of diet), followed by terrestrial invertebrates (76.8%, 9.5%) and algae and plant material (70.5%, 4.8%). Diet diversity increased as cardinal shiner size increased, so that aquatic invertebrates constituted the majority of the diet among cardinal shiner >80 mm total length. Although previous studies conducted in Oklahoma suggested cardinal shiner consumes nearly equal amounts of invertebrates and plant material, our findings suggest the relict population in Kansas consumes primarily aquatic invertebrates and occasionally terrestrial invertebrates and plant material.

Introduction

Understanding the ecology of stream organisms is a necessary step for conservation and management of freshwater biodiversity. Less than one percent of the Earth's water exists in the form of freshwater streams, and disproportional losses in global biodiversity have occurred in these ecosystems (Dudgeon et al. 2006). Freshwater fish represent a group of organisms with well-documented global declines in abundance and distribution (Helfman 2007), including imperilment of nearly 40% of species in North America (Jelks et al. 2008) and 47% of species in Kansas (Haslouer et al. 2005). Among imperiled fishes, species with isolated or disjunct populations are among the most highly imperiled and represent potential priorities for conservation of ecological and molecular diversity (Lesica and Allendorf 1995; Perkin, Shattuck and Bonner 2012). In the Great Plains region of North America, isolated and disjunct populations of endemic freshwater fishes are common and many are in need of conservation (Hoagstrom, Brooks and Davenport 2011).

Distribution of some Great Plains fishes was historically widespread throughout interior highland rivers of North America. During the Pleistocene period, expansion and contraction of glaciers caused changes in the structure of drainage basins to the extent that many Great Plains streams were redirected into adjacent drainage basins (Gerking 1947; Hocutt and Wiley 1986). This process had consequences for the distribution of freshwater fishes, resulting in isolated populations with restricted ranges. Populations that persisted in regions where dramatic changes in the surrounding environment occurred are referred to as relict, and many relict populations evolve to form new species because long-term isolation results in genetic drift and natural selection (Mayden 1988a; Lesica and Allendorf 1995). In present-day southeastern Kansas and portions of

Figure 1. Male cardinal shiner (*Luxilus cardinalis*) in breeding coloration. Photo by Keith Gido. Used with permission.

Arkansas, Missouri, and Oklahoma, the striped shiner (*Luxilus chrysocephalus*) species group is an example of glacial relict populations that formed unique species from a widely distributed common ancestor (Mayden 1988a). One of these species, the cardinal shiner (*Luxilus cardinalis*; Fig. 1), inhabits portions of the Arkansas and Red river basins in Arkansas, Kansas, Missouri, and Oklahoma (Mayden 1988b). In southeastern Kansas, a relict population exists in the upper Neosho River basin and is now isolated completely because of construction of John Redmond Reservoir (completed in 1964; Fig. 2). Occasional collections have also been taken from the upper Verdigris River basin of Kansas (Cross 1967; Mayden 1988b). Whereas some aspects of cardinal shiner ecology have been studied since its description, especially in portions of Oklahoma (e.g., McNeely 1987), little is known about the life history and feeding ecology of the relict population in the upper Neosho River drainage of Kansas (Cross and Collins 1995).

Ecological and life history information for the cardinal shiner is limited. In Arkansas and Missouri, the species associates with clear, gravel-bottom streams with perennial flow and occupies deep riffles or pools (Robison and Buchanan 1988; Pflieger 1997). Reproductive ecology of cardinal shiner was studied in Oklahoma, where the species associated with mound-building fishes and spawned during May (Miller 1967). Spawning was also observed during April to June in Arkansas (Robison and Buchanan 1988), and April to May in Kansas and Missouri (Cross and Collins 1995; Pflieger 1997). In Kansas, cardinal shiner is known to spawn in riffles with notable current when mound-building species are not present (Cross and Collins 1995). Feeding ecology was documented during only a single study conducted in Oklahoma, where cardinal shiner consumed equal amounts of invertebrate and plant material (McNeely 1987). Interestingly, cardinal shiner have simple (as opposed to complex) gut morphology

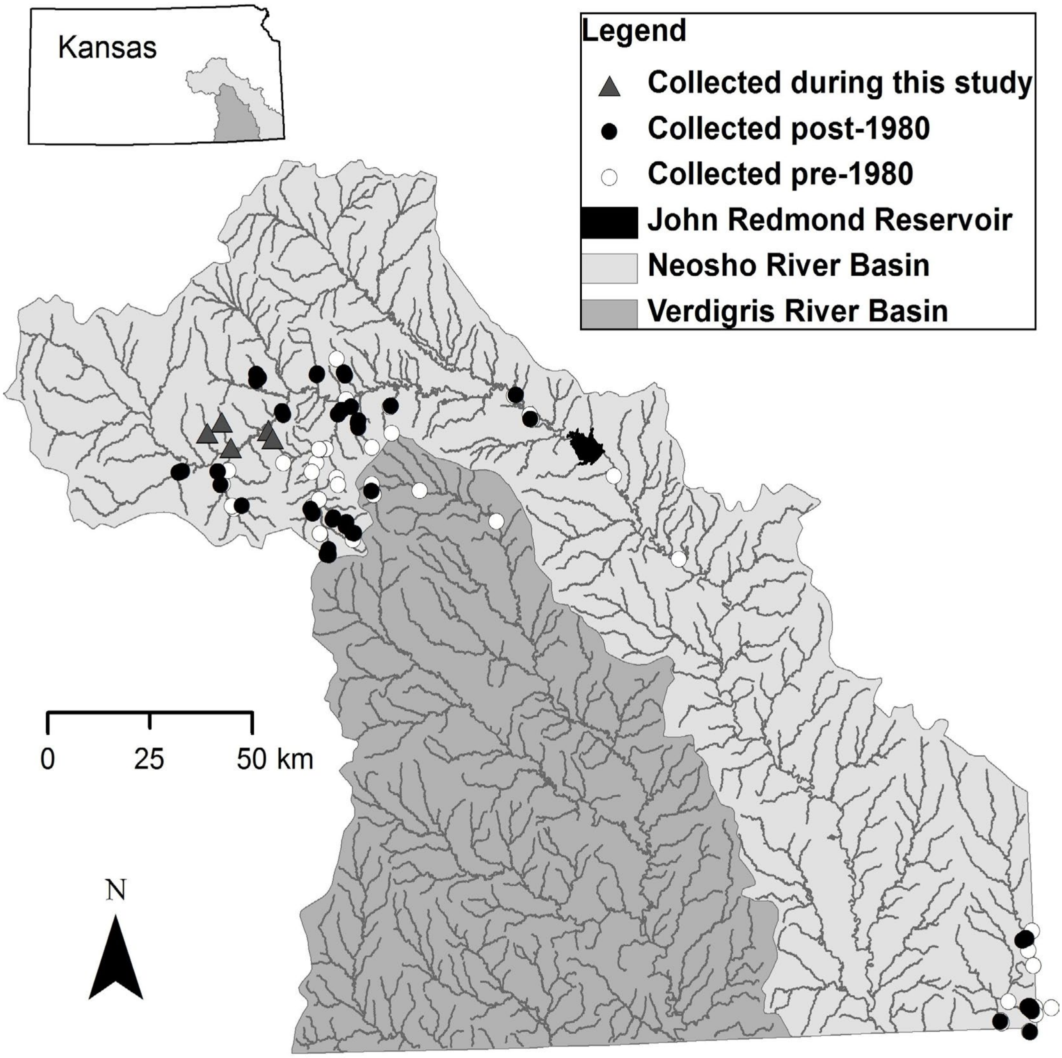

Figure 2. Distribution of cardinal shiner (*Notropis cardinalis*) in Kansas illustrating collections taken prior to 1980 (white circles), after 1980 (black circles), and during this study (gray triangles) from the Neosho and Verdigris river basins. The upstream population in the Neosho River basin is a glacial relict and is now separated from the adjacent Spring River and Shoal Creek populations by John Redmond Reservoir (black polygon; completed in 1964). Fish collection data are held in the Kansas Aquatic Gap Database (Keith Gido, Kansas State University); contemporary collections were made largely by the Kansas Department of Wildlife, Parks, and Tourism Stream Survey Program.

characterized by short overall length, only two bends, and a black colored peritoneal lining (Pflieger 1997). Short and simple gut morphology is ideal for consumption of insect prey items with greater energy availability relative to algae or plant material (Marshall 1947; Perkin, Shattuck and Bonner 2012); however, black peritoneal lining is characteristic of omnivorous or algivorous species that prefer algae or plant material (Goldstein and Simon 1999). Further study is needed to determine if cardinal shiner consume primarily animal or plant material in Kansas streams.

We present notes on the feeding ecology of a relict population of cardinal shiner in the upper Neosho River basin of Kansas. Although available evidence suggests cardinal shiner populations in Kansas are stable, our hope is that additional ecological information will be helpful given the species is considered in need of conservation (Haslouer et al. 2005). The objectives of this study were to: (1) describe historical (pre-1980) and contemporary (post-1980) distribution of the relict cardinal shiner population in the upper Neosho and Verdigris rivers, (2) describe the summer feeding ecology of the species to determine if equal parts of animal and plant material are consumed, and (3) determine if proportions of prey items change as a function of cardinal shiner size.

Methodology

We reviewed historical and contemporary accounts of cardinal shiner distributions in the state of Kansas to evaluate status of the species, including the relict population in the upper Neosho River basin. We used fish collection samples in the Kansas Aquatic Gap Database (Keith Gido, Kansas State University) to plot historical (pre-1980) and contemporary (post-1980) distribution of cardinal shiner in Kansas. We used 1980 because this year marked a reduction in major environmental alterations including construction of large reservoirs, expansion of irrigated land, and expansion of farm land in Kansas (Gido, Dodds and Eberle 2010). We also reviewed contemporary literature regarding the status of cardinal shiner in Kansas and North America (Haslouer et al. 2005; Jelks et al. 2008).

We collected cardinal shiner from five privately-owned stream sites in Chase County, Kansas during July of 2010 as a part of a larger study (Perkin and Gido 2012). Sampling sites were positioned in close proximity to control for abiotic and biotic conditions such as regional species pools, climate variables (e.g., rainfall), as well as air and stream temperature regimes (Fig. 2, gray triangles). Streams ranged in size from second- to third-order and were characterized by low turbidity, gravel substrate, pool and riffle habitats, and intact riparian corridors. Collections were made using a combination of backpack electrofishing and seining so that all habitats within a representative reach of stream (i.e., 40x the mean stream width) were sampled at five sites (Fischer and Paukert 2009). Cardinal shiner individuals were opportunistically retained to achieve a diversity of size classes with a minimum of 20 individuals from each site. Remaining cardinal shiner and fish community members were identified and counted. We euthanized all retained cardinal shiner in a lethal dose of MS-222 (80 mg/l) followed by preservation in 10% formalin for laboratory analysis.

In the laboratory, we measured for total length (mm) and dissected individuals to obtain a quantitative diet description. Gut tracts between the sphincter of the esophagus and second bend in the intestine were removed and contents were classified into lowest practical taxonomic groups, usually family for aquatic insects and class or order for terrestrial or non-insect items. Insect and insect parts were identified based on taxonomic keys by Merritt et al. (2008) and Thorp and Covich (2001). Underneath a dissecting microscope, we used the two-dimensional area (mm) of each taxonomic group to quantify abundance by spreading items across the bottom of the Petri dish and then counting the number of 1x1 mm cells covered by each group (Franssen and Gido 2006). We then calculated the frequency of occurrence (percentage of fish that contained each prey item) and percent area (taxon area / total content area * 100) for each taxonomic group across all individuals to summarize dietary habits (Bowen 1996).

Because the diets of cyprinids are known to change with increasing body size, either because of gape limitation (Perkin, Williams and Bonner 2009) or increased energy demands at greater sizes (Frazer and Cerri 1982), we hypothesized that cardinal shiner would become increasingly opportunistic as total body length increased because of associated increases in gape. We first used linear regression to characterize the relationship between gape width (mm; dependent variable) and total length (mm; independent variable). We then calculated diet diversity for each individual using Simpson's Index of Diversity (1-D, where D = 1/sum [proportion of diet for each prey category]^2) and regressed diet diversity against total length to test for greater prey diversity at greater total lengths (and gapes). We then aggregated individuals into 10-mm length classes and tested for differences in the mean percent area of aquatic invertebrates, plant material, terrestrial invertebrates, and detritus (i.e., decaying organic matter) in cardinal shiner guts. Differences in percent area of each diet category were tested using analysis of variance (ANOVA) and Fisher's least significant differences (LSD). We used a Bonferroni-adjusted alpha to control for four tests of changes in diet among length classes (i.e., $\alpha = 0.05/4 = 0.012$) during ANOVA and LSD tests to account for experiment-wise error.

Results

Distribution of cardinal shiner in Kansas decreased little during the period of historical records (Fig. 2), and this pattern was consistent with literature reviews regarding the status of the species. Contemporary collection reports for southeastern Kansas occurred in most of the systems in which historical records existed, with the exception of a section of the Neosho River downstream of John Redmond Reservoir. Literature review revealed that cardinal shiner was proposed to be a species in need of conservation (SINC) in the state of Kansas because their range encompasses habitats that have undergone or are currently undergoing major changes and because cardinal shiner is part of a distinctive fauna that is rare in the state (Haslouer et al. 2005). Cardinal shiner was not included on the list of North American species considered imperiled by the American Fisheries Society's Endangered Species Committee (Jelks et al. 2008).

Collections from five sites in the upper Neosho River basin visited during July of 2010 yielded 95 cardinal shiner that were retained for laboratory analysis. Among sites, cardinal shiner ranged 3-59% relative abundance (i.e., 3%, 13%, 21%, 24%, and 59%) and tended to co-occur with redfin shiner (*Lythrurus umbratilis*), creek chub (*Semotilus atromaculatus*), largemouth bass (*Micropterus salmoides*), green sunfish (*Lepomis cyanellus*), fantail darter (*Etheostoma flabellare*), and orangethroat darter (*E. spectabile*). Total lengths of retained individuals ranged 39-108 mm, with the greatest number of individuals falling within the 81-90 mm size class. Sample sizes within classes included 39-50 mm (n = 7), 51-60 mm (n = 9), 61-70 mm (n = 16), 71-80 mm (n = 17), 81-90 mm (n = 28), and 91-108 mm (n = 17). Among these individuals, gape widths ranged 2.75-7.5 mm and indicated a significant increase as a function of total length ($n = 95$, $r^2 = 0.91$, slope = 0.07, $F_{1,93} = 969.05$, $P < 0.01$).

Cardinal shiner consumed a diversity of prey items ranging from aquatic to terrestrial invertebrates. Among the 95 individuals retained for analysis, only one had an empty gut tract. By percent frequency of occurrence, aquatic invertebrates occurred in 93.7% of guts, followed by terrestrial invertebrates in 76.8% and algae and plant material in 70.5% (Table 1). Unidentified prey items in the form of amorphous detritus occurred in 96.8% of individuals. By percent area, aquatic invertebrates were on average 29.3% of diet items, followed by detritus (16.1%), terrestrial invertebrates (9.5%), and algae and plant material (4.8%). Dominant aquatic invertebrates included Diptera (62.1% occurrence, 1.8% area), Ephemeroptera (57.9%, 5.7%), and Odonata (26.3%, 0.7%), and unidentified aquatic invertebrate parts occurred in 74.7% of individuals and constituted on average 9.5% of diet area. Dominant terrestrial invertebrates included

Table 1. Frequency of occurrence and mean percent by area of gut contents for 95 cardinal shiner (*Notropis cardinalis*) collected from the upper Neosho River basin of Kansas during July 2010.

Taxon	Frequency of occurrence (%)	Mean percent by area (%)
Aquatic Invertebrates	93.7	29.3
Coleoptera	14.7	0.9
Acari (sub-class)	1.1	<0.1
Elmidae	1.1	0.3
Georissidae	3.2	0.2
Psephenidae (larvae)	3.2	<0.1
Diptera	62.1	1.8
Chironomidae	50.5	0.6
Ephemeroptera	57.9	5.7
Baetidae	37.9	1.4
Heptageniidae	34.7	1.8
Hemiptera	17.9	0.7
Corixidae	4.2	0.1
Megaloptera	2.1	0.1
Sialidae	1.1	0.1
Odonata	26.3	0.7
Anisoptera (sub-order)	1.1	<0.1
Aeshnidae	1.1	0.2
Zygoptera (sub-order)	1.1	<0.1
Ostracoda	18.9	0.1
Plecoptera	1.1	0.1
Collembola	22.1	0.5
Trichoptera	15.8	0.3
Aquatic Insect Parts	74.7	9.5
Terrestrial Invertebrates	76.8	9.5
Attelabidae	1.1	<0.1
Araneae	10.5	0.3
Hymenoptera	17.9	0.3
Formicidae	11.6	0.2
Lepidoptera	5.3	0.3
Crambidae (larvae)	4.2	0.1
Orthoptera	9.5	0.9
Terrestrial Insect Parts	73.7	7
Algae and Plant Material	70.5	4.8
Filamentous Algae	55.8	1.8
Non-filamentous Algae	9.5	0.7
Vascular Plant	42.1	2.3
Detritus and Other	98.9	21.3
Detritus	96.8	16.1
Insect Parts	29.5	4.5
Fish Scales	20	0.7

Hymenoptera (17.9% occurrence, 0.3% area) and Araneae (10.5%, 0.3%), and unidentified terrestrial invertebrate parts (mainly wing parts) occurred in 73.7% of individuals and constituted on average 7.0% area. Filamentous algae occurred in 55.8% of individuals and constituted on average 4.8% of area, while vascular plants occurred in 42.1% of individuals and constituted on average 2.3% of area. Remaining taxonomic groups (e.g., ostracods) occurred in low abundances. Tests for differences in prey consumption among size classes suggested variability in feeding ecology with size. Aquatic invertebrates constituted between 34% and 55% of diet area among size classes, but proportions did not significantly differ (ANOVA, $F_{5,88} = 1.4$, $P = 0.23$; Fig. 3). Similarly, terrestrial invertebrates constituted between 5% and 19% of diet area, but did

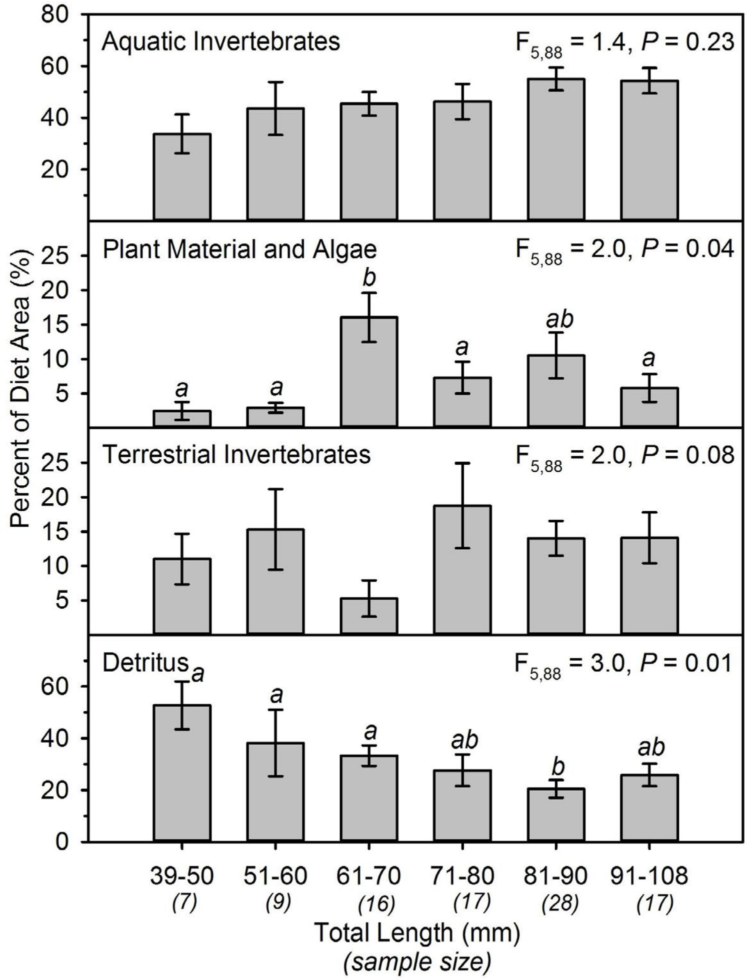

Figure 3. Mean (± 1 standard deviation) percent of diet comprised by aquatic invertebrates, algae and plant material, terrestrial invertebrates, and detritus among 95 cardinal shiner (*Notropis cardinalis*) of various total lengths (mm) collected from the upper Neosho River basin during July of 2010. Sample sizes in each length class are given parenthetically below lengths and results of single factor analysis of variance as well as Fisher's least significant differences (α = 0.05/4 = 0.012) are given.

not change significantly among size classes (ANOVA, $F_{5,88} = 2.0$, $P = 0.08$). The quantity of detritus generally declined as cardinal shiner length increased and some larger individuals (i.e., 81-90 mm) contained significantly less detritus relative to smaller size classes (ANOVA, $F_{5,88} = 3.0$, $P = 0.01$, Fishers LSD). Presence of plant material and algae was variable among size classes, ranging 2% to 16% and significantly differing among size classes (ANOVA, $F_{5,88} = 2.0$, $P = 0.04$, Fishers LSD). Intermediate sized cardinal shiner (61-70 mm) consumed the greatest majority of plant material, consistent with an associated decline in proportion of terrestrial invertebrates. Finally, taxonomic diversity among prey items increased as a function of total length among all cardinal shiner analyzed ($n = 95$, $r^2 = 0.08$, $F_{1,93} = 8.27$, $P = 0.005$).

Discussion

Dietary habits of 95 cardinal shiner ranging 39 to 108 mm in total length and collected during July of 2010 suggested aquatic and terrestrial invertebrates were dominant prey items in terms of diet area, but plant material occurred in a large portion of individuals analyzed. Invertebrate prey items were consumed by all cardinal shiner size classes and indicated no significant difference in percent of diet among size classes. Plant material and algae were also consumed by all size classes and occurred most in 61-70 and 81-90 mm cardinal shiner, but percent of diet composed of plant material and algae was always less than 25%. Furthermore, although total diversity of prey items generally increased as cardinal shiner size (and consequently gape width) increased, the portion of plant material and algae in diets remained low. These findings represent to our knowledge the first report of feeding ecology for the relict population of cardinal shiner in the upper Neosho River basin of Kansas.

Fishes in the genus *Luxilus* generally consume aquatic invertebrates as well as terrestrial invertebrates and plant material. For example, striped shiner (*L. chrysocephalus isolepis*) is known to consume nearly equal parts plant material and terrestrial arthropods during the day but greater consumption of plant matter at night (Hambrick and Hibbs 1976). Duskystripe shiner (*L. pilsbryi*) and bleeding shiner (*L. zonatus*) are classified as insectivores (Goldstein and Simon 1999), though duskystripe shiner consumes algae at times (Robison and Buchanan 1988; Pflieger 1997). Similarly, cardinal shiner (previously *Notropis pilsbryi*) in the Neosho River of Oklahoma is known to consume equal parts plant and animal material, with vascular plants and filamentous algae being the most important food items across all individuals collected over a 20-month period spanning October 1979 to May 1981 (McNeely 1987). Deviations from these base-line trophic patterns might be useful for identifying environmental perturbations, but such an approach is most useful when adequate information is available for all species within a community (Goldstein and Simon 1999). Given trophic classification information for cardinal shiner is currently lacking (Goldstein and Simon 1999), our results are useful for classifying the relict population of cardinal shiner in the upper Neosho River basin. Using the terminology of Goldstein and Simon (1999), the main food of cardinal shiner in our study system was aquatic and terrestrial invertebrates, primary trophic level was invertivore, and secondary trophic level included both benthic and drifting prey items. Some aspects of cardinal shiner were consistent with omnivorous fishes, including occurrence of a dark peritoneum and consumption of both plant and animal materials (Schlosser 1982). However, cardinal shiner lack the elongated gut characteristic of omnivores and plant material never exceeded 25% of their diet, which is consistent with generalized insectivores (Schlosser 1982). Consequently, we believe cardinal shiner to be insectivorous, feeding primarily on aquatic invertebrates and exhibiting opportunistic and generalist behavior by consuming plant material as well as terrestrial subsidies (Goldstein and Simon 1999).

Broad diets including terrestrial prey items are common among stream fishes, especially small-bodied cyprinids such as the cardinal shiner. In reviewing terrestrial food subsidies in the diets of North American stream fishes, Sullivan, Zhang and Bonner (2012) found terrestrial subsidies were most associated with small-bodied fishes characterized by terminal mouth positions and inhabiting stream segments with dense vegetative cover. Cardinal shiner fits the description of a terminal mouth (lower jaw extending to the tip of the snout) and is small-bodied (maximum size 120 mm; Pflieger 1997). Furthermore, the streams sampled during this study were characterized by intact riparian corridors with dense vegetative stands that shaded streams (Perkin and Gido 2012). Temporal variability in terrestrial subsidies is driven by seasonal changes (Sullivan, Zhang and Bonner 2012), and the same is likely true over the diel period for consumption of plant material based on the findings of Hambrick and Hibbs (1976). Our sampling was limited to the summer period and did not allow for assessing seasonal changes in cardinal shiner diet, but we expect seasonal changes do occur as with the diets of other relict populations of cyprinids (e.g., Perkin, Shattuck and Bonner 2012). Although we did find that prey diversity increased as a function of cardinal shiner size, we did not find an associated increase in the portion of diet composed of terrestrial invertebrates. This suggests that while prey diversity is greater among larger cardinal shiner, there remains a bias toward aquatic invertebrates in terms of portion of their diet. However, increase in prey diversity with increased overall size might be related to numerous factors other than gape limitation, including behavioral or habitat changes, increased energetic demands, and improved ability to pursue and capture prey (Fraser and Cerri 1982). Given terrestrial subsidies are typically greater in stream segments with riparian vegetation (Sullivan, Zhang and Bonner 2012) and cardinal shiner consumed terrestrial prey items during all size classes, our findings support conclusions that intact riparian corridors are important for maintaining the trophic integrity of stream fish populations and communities (Fischer et al. 2010).

Although range of cardinal shiner has not declined in Kansas and the species is not imperiled throughout its North American range, there is still cause for concern regarding cardinal shiner persistence. The population of cardinal shiner in the upper Neosho River basin is confined to a small geographic range in which a single abiotic or biotic disturbance could affect the viability of a large proportion of individuals (Haslouer et al. 2005). Furthermore, Perkin and Gido (2012) found cardinal shiner responded negatively to stream fragmentation caused by road-stream crossings characterized by perched outflows, which is a growing concern for numerous fish species inhabiting streams in and outside of the Great Plains (Bouska and Paukert 2009; Fullerton et al. 2010). Construction of impoundments is known to disrupt the distribution of cyprinid species in Kansas streams (Falke and Gido 2006), and we found the only detectable change in cardinal shiner range was associated with construction of a reservoir on the mainstem Neosho River that potentially limited downstream dispersal. Introduction of predator species to streams or increases in the abundance of native nest-building predators (e.g., largemouth bass, green sunfish) have the potential to deplete local populations of stream-dwelling cyprinids (Knight and Gido 2005), but documented decreases in cardinal shiner abundance caused by predators do not exist to our knowledge. Other threats to cardinal shiner might include water quality degradation such as agricultural chemical spills capable of killing entire streams (e.g., Olmsted and Cloutman 1974) and habitat modification in the form of riparian buffer destruction (e.g., Fischer et al. 2010).

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