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Food habits of imperiled Plains Topminnow and diet overlap with invasive Western Mosquitofish in the Central Great Plains

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Food habits of imperiled Plains Topminnow and diet overlap with invasive Western Mosquitofish in the Central Great Plains

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Abstract: Plains Topminnow (*Fundulus sciadicus)* populations have experienced large declines throughout the Central Great Plains, with Western Mosquitofish (*Gambusia affinis)* introductions suggested as a contributing factor. There are limited studies identifying the food habits of Plains Topminnow and the trophic interactions with Western Mosquitofish. This study sought to determine if a diet overlap exists between the Plains Topminnow and the introduced Western Mosquitofish by identifying the feeding habits of both species. We analyzed diets from lentic and lotic populations of Plains Topminnow captured in August and found lentic topminnows employed a generalist diet while lotic topminnow selected for gastropods. Additionally, Western Mosquitofish diets from regionally proximate lotic and lentic populations also displayed a generalist diet consisting of benthic, littoral and terrestrial macroinvertebrates. The two species did not show overlapping diets based on Schoener's Index. Therefore we suggest the introduced Western Mosquitofish do not likely impact Plains Topminnow populations through food resource competition.

Keywords: diet overlap, Great Plains, imperiled, invasive species, mosquitofish, topminnow.

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Introduction

Plains Topminnow (*Fundulus sciadicus)* is endemic to the Central Great Plains and has experienced a decrease in historic range and local abundance (Schumann et al. 2016). As a habitat specialist with the capability to move great distances the Plains Topminnow was once common throughout the Great Plains (Schumann 2015[a]; Schumann et al. 2016). Recent studies report a 72 percent decline in distribution throughout the Plains Topminnow's native range, which includes both a northern and a southern spatially separated disjunct populations (Pasbrig et al. 2012). Nebraska represents 67 percent of historical occurrence sites and is thought to be the central stronghold of Plains Topminnow distribution; however, a 66 percent decline in occurrence was reported (Pasbrig et al. 2012). Nebraska currently list Plains Topminnow as a Tier 1 species of special concern with populations considered at risk (NatureServe 2016).

The decline in Plains Topminnow presence is likely linked to a multitude of reasons. Factors hypothesized to limit Plains Topminnow persistence vary from species competition, predation, habitat loss, stream fragmentation, climate change, water quality, and stream dewatering (Fischer and Paukert 2008; Pasbrig et al. 2012). Over the last century, much of the Central Plains landscape has been converted from grasslands to agriculture (Samson and Knopf 1994), causing reductions in stream habitat and biologic diversity (Pringle 1988; Jenkins et al. 2003). Habitat alterations and fish stockings across the Plains have increased interactions with non-native competitors and predators. Additionally, large-scale landscape alterations have favored non-native generalist species, which decreased native fish populations, presumably from decreased habitat diversity (Smith et al. 2014). Western Mosquitofish (*Gambusia affinis*) are among the generalist species encountered by Plains Topminnow populations and are commonly found to dominate local fish assemblages in disturbed streams (Chapman and Warburton 2006).

Western Mosquitofish are the most widely distributed fish in the World and are the subject of increased research for their impacts on native fishes. These fish were introduced in Nebraska for mosquito control starting in

1972 (Kaufmann and Lynch 1991). Western Mosquitofish have been implicated in the reduction of other topminnow species due to direct predation of larval fish, with little supporting evidence for resource partitioning impacts (Minckley 1973; Meffe 1984; Goldsworthy and Bettoli 2006; Laha and Mattingly 2006; Sutton et al. 2012). In mesocosm experiments, Plains Topminnow experienced 70% mortality in co-existence with Western Mosquitofish, but other Fundulidae species survived (Haas 2005, Schumann et al. 2015[b]). However, distinguishing the impacts between cohabited wild Plains Topminnow and Western Mosquitofish populations is complicated by both the conservation status of Plains Topminnow and short temporal scale of co-habitation.

Food selection and diet overlap between Plains Topminnow and cohabitate species have not been documented. Western Mosquitofish have a generalist diet (Mansfield and Mcardle 1998) preventing them from being an effective biological control agent of mosquito populations (Kumar and Hwang 2006). However, knowledge of Plains Topminnow food habits remains limited and represents a gap in ecological understanding (Bestgen 2014). Previous studies deduced from morphological characteristics, associated habitat use, and observed feeding behavior that Plains Topminnow are surface feeders that primarily feed on Ostracods, chironomidae, and occasionally feed on Gastropods from the Genus Physa (Stribley and Stasiak 1982; Rahel and Thel 2004; Haas 2005; Bestgen 2014). Historic evidence from actual diet analysis of 12 specimens supported this hypothesized feeding behavior of surface insects (Ellis 1914), while a recent effort from the southern range of distribution found a more diverse diet including greater utilization of benthic organisms (Thompson 2014). However, the diet contents from Missouri were from lentic specimens only and may be the result of dissimilar prey availability in lentic and lotic environments. Determining food habits of Plains Topminnow will help define the ecological role of this species, as well as provide additional information on the diet overlap between this species and the non-native Western Mosquitofish in Nebraska. Thus, the objectives of this study are to 1). Provide an additional assessment of Plains Topminnow diet; 2). Describe prey selectivity of Plains Topminnow from a lentic and lotic population; and 3). Assess diet overlap between regionally proximate populations of Plains Topminnow and Western Mosquitofish.

Methods

***Fish collection*:** Fish collections were made at 4 separate sample sites including 2 lotic and 2 lentic systems. Ideally, wild populations of Plains Topminnow would be used for this assessment, but the protected status of Plains Topminnow precludes the sacrifice of wild specimens and therefore Plains Topminnow specimens from two Nebraska Game and Parks Commission broodstock locations were used. Additionally, cohabitated populations of Plains Topminnow and Western Mosquitofish would best identify resource partitioning but mingled populations are rare as mosquitofish have been suggested to rapidly displace topminnow (Thiessen 2016). Introducing Western Mosquitofish to Plains Topminnow broodstock populations in an effort to create a cohabitated population were not considered in the interest of maintaining a viable broodstock population. Therefore, Western Mosquitofish were collected from similar aquatic systems within regional proximity of Plains Topminnow broodstock populations.

A total of 30 Western Mosquitofish were collected from Blue Hole WMA (lentic) and 30 from Sandy Channel State Recreation Area (lotic) using DC pulsed backpack electrofishing techniques from August to September 2015 (Schumann et al. 2015[b]). A total of 30 Plains Topminnow from Sac-Wilcox WMA (lentic) and 30 from Rock Creek Fish Hatchery (lotic) were collected using a 381mm tall, 3 paneled cloverleaf trap; with 6mm mesh galvanized wire and 12mm openings August to September 2015. Study specimens ranged in length from 58mm to 73mm. All study specimens were preserved in 4:1 ethanol:water solution immediately after capture. The entire digestive tract was removed from each fish and prey items were identified to order and quantified as frequency of occurrence.

***Prey collection*:** Prey availability at Plains Topminnow collection sites was determined using samples of both benthic and littoral macroinvertebrates. Preliminary studies found zooplankton to be nonexistent in Plains Topminnow diets (NGPC unpublished data); therefore zooplankton sampling was not conducted. Macroinvertebrate collections were completed at three sites for Rock Creek Fish Hatchery and Sac-Wilcox WMA pond, using a 1m x 1m 500µ mesh macroinvertebrate kick net. Each site sampled $2m^2$ using a floating PVC quadrat (Barbour et al. 1999); water depth never exceeded 1m, which allowed representation of the entire water column in the samples. All samples were immediately preserved in 4:1 ethanol:water solution. Prey items from each sample site were identified to order and quantified as frequency of occurrence.

***Plains Topminnow prey selectivity*:** Stomach contents were quantified using percent composition by frequency of each diet item occurrence (number of each diet item group divided by the total number of diet items found in stomachs of each site group). Prey electivity by Plains Topminnow was evaluated using the Strauss's prey electivity index (1979):

$$L = r_i - p_i,$$

where r_i and p_i represent the relative abundance of prey in the diet and the environment, respectively. Relative prey abundance in Plains Topminnow diets (r_i) were determined by dividing the number of individual diet item groups found in the stomachs from each waterbody, by the total number of diet items consumed from the same group. Diet item proportions in the environment (p_i) were calculated by dividing the density of each prey item group by the total density of all prey items available in the environment. Strauss's index value (L) can range from total avoidance (-1) to absolute selectivity (1) for a given prey item. Similar to previous studies (Dettmers and Stein 1992; Sullivan et al. 2011), a value of ± 0.15 was chosen as the cutoff to determine selectivity or avoidance. Prey items with index values between 0.15 and -0.15 represent prey consumed proportionately to their availability (Sullivan et al. 2012). We defined opportunistic prey selection as electivity values between 0.15 and -0.15 and prey selection as values > 0.15 or < -0.15. Diet and available prey items from lotic and lentic sample locations were tested for significant differences using a paired t-test with α = 0.05, to determine if differences between aquatic systems exist (Childs 2006).

***Diet overlap*:** Diet overlap between Plains Topminnow and Western Mosquitofish was determined using Schoener's diet overlap index (Schoener 1970):

$$C_{xy} = 1 - 0.5 \left(\sum [p_{xi} - p_{yi}]\right),$$

where C_{xy} is the index value, p_{xi} is the relative percentage of prey type i used by species x, p_{yi} is the relative percentage of prey type i used by species y. Index values, (C_{xy}), range from 0 to 1, with a value of 0 indicating no overlap and a value of 1 indicating complete overlap. Diet overlap index values ≥ 0.6 were considered biologically significant (Wallace 1981).

Results

***Plains Topminnow diet and prey selectivity*:** The composition of prey available between the lentic and lotic environments (**Figure 1**) did not significantly differ (t-stat

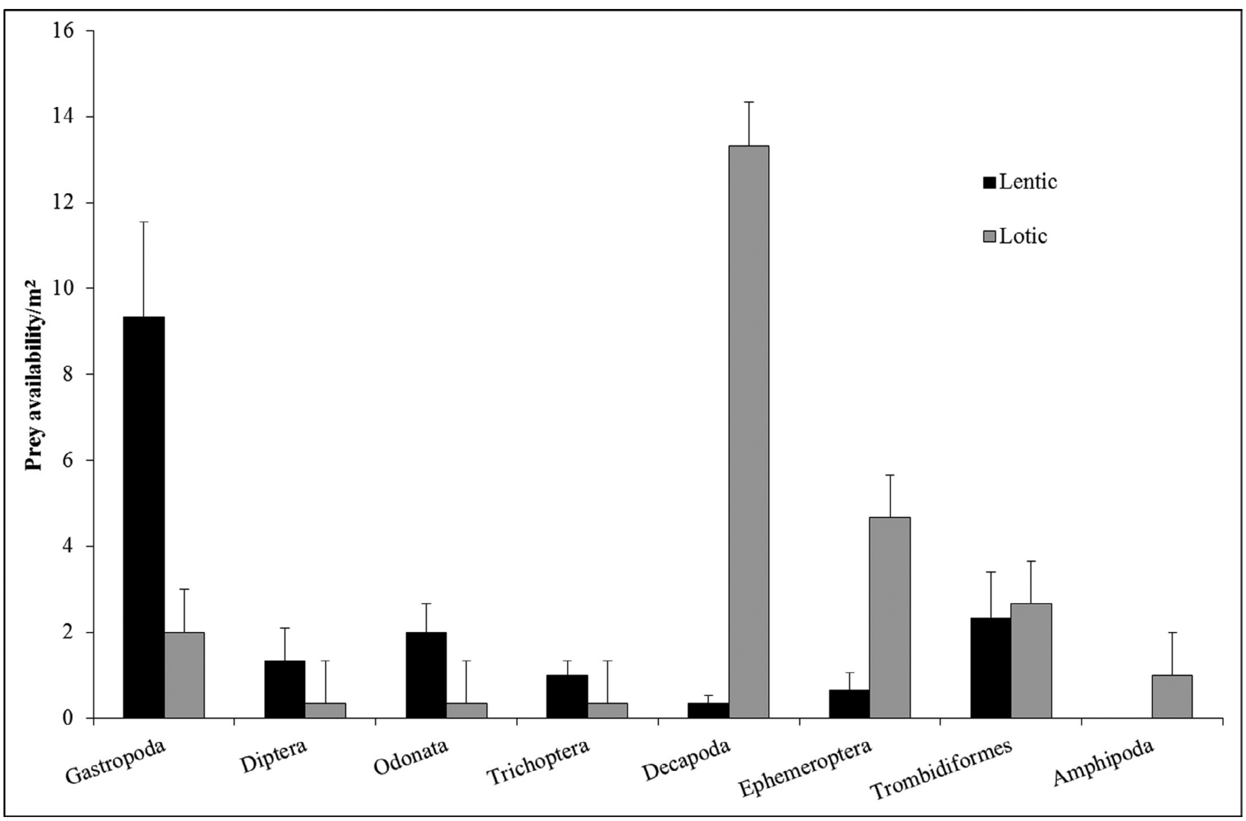

Figure 1. Total prey availability (prey count/1m^2) at three sample zones for each lentic and lotic Plains Topminnow study site.

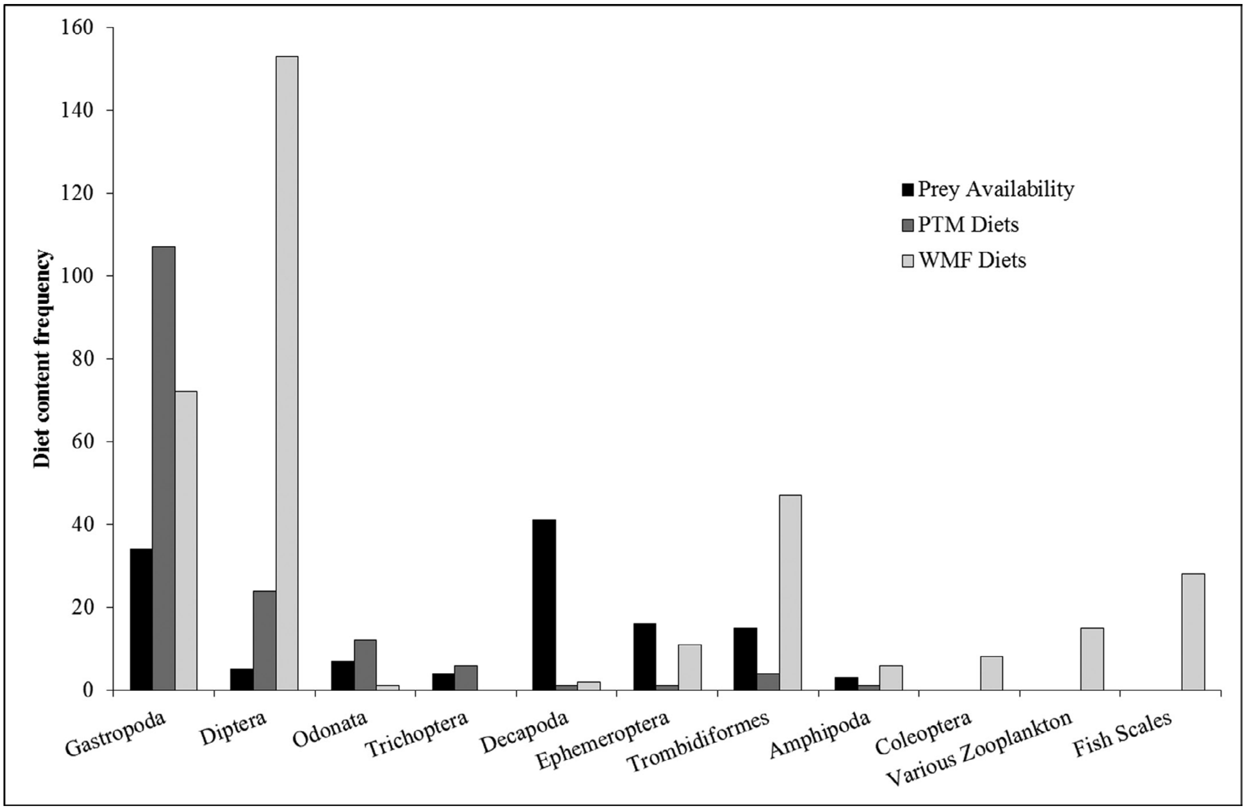

Figure 2. Total prey item frequency of occurrence at Plains Topminnow sample sites, and for Plains Topminnow and Western Mosquitofish diet content.

= -0.36, df = 7, p = 0.36). Similarly, Plains Topminnow and Western Mosquitofish diets (**Figure 2**) did not significantly differ between lentic and lotic populations (t-stat = 1.24, df = 7, p = 0.13) and represented all available prey in both lentic and lotic aquatic environments (Figure 2). Gastropods were the second most abundant prey available (Figure 2) and were positively selected by Plains Topminnow at both lentic (L = 0.16) and lotic (L = 0.32) study sites (**Figure 3**). Decapods and ephemeroptera were the most and third most abundant prey available (Figure 2), respectively; however, lotic Plains Topminnow populations (Figure 3) selected against both decapods (L = -0.55) and ephemeroptera (L = -0.21). All other available prey items were consumed in proportion to their availability in both lotic and lentic environments (Figure 3).

***Diet overlap*:** Schoener's diet overlap index value between Plains Topminnow and Western Mosquitofish in Nebraska was C_{xy} = 0.43, which is below the C_{xy} = 0.6 threshold used to deem two species diet overlap as biologically significant (**Figure 4**). Despite having many similar prey items, Plains Topminnow and Western Mosquitofish do not have overlapping diets. Plains Topminnow most frequent prey item were juvenile gastropods which comprised 66 percent of their diets (Figure 4). Western Mosquitofish most frequent prey item were various diptera species, which represented 44 percent of their diet composition (Figure 4). Various diptera species represented 15 percent of topminnow diets and gastropods represented 22 percent of mosquitofish diets (Figure 4). Although both species diet content included similar macroinvertebrates, they consumed individual taxa at different frequencies of occurrence, with mosquitofish also consuming zooplankton (Figure 4). Additionally, fish scales were found in mosquitofish diets suggesting at least some level of piscivory (Figure 4). Zooplankton, fish scales, and coleoptera were absent from topminnow diets.

Discussion

***Plains Topminnow diet and prey selectivity*:** Lentic Plains Topminnow populations examined as part of this study can be considered generalist feeders as lentic study fish preyed mostly on organisms in proportion to their availability. In contrast, lotic populations were selective for gastropods and against decapods and ephemeroptera. However, decapods and ephemeroptera were absent and gastropods were of greater abundance at lentic

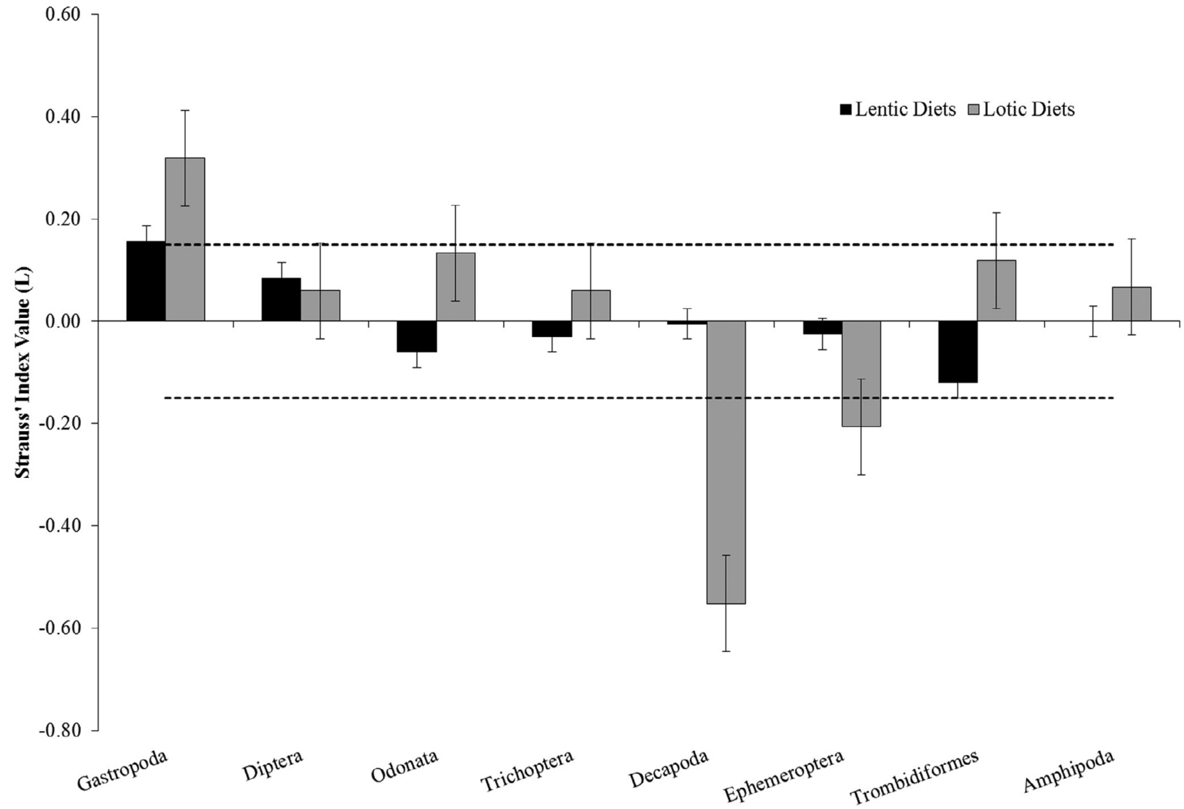

Figure 3. Strauss' prey selectivity index for lentic and lotic Plains Topminnow populations.

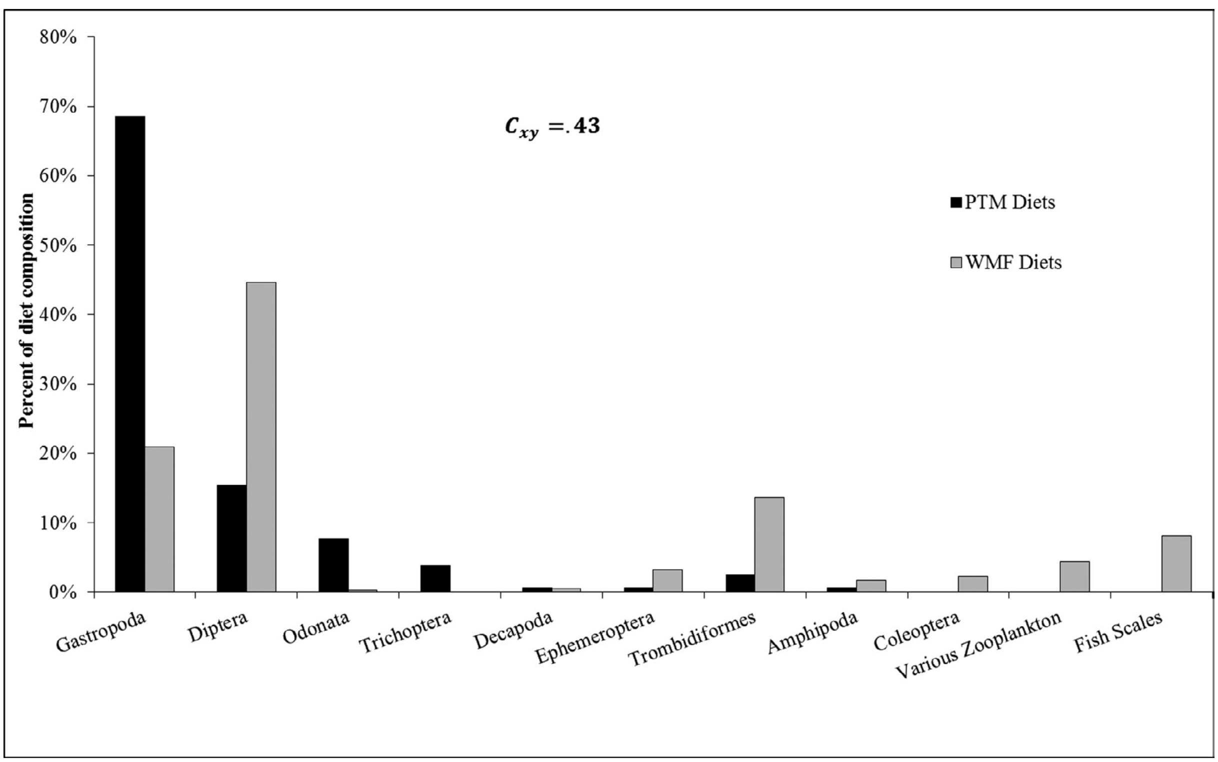

Figure 4. Percent of diet composition for each prey order and Schoener's diet overlap index for Plains Topminnow and Western Mosquitofish at 4 regionally proximate study locations.

study sites; which may be responsible for such a strong preference and avoidance of prey items. Prey selectivity favoring gastropods, of the genus *Physa*, supports spatial distribution and lotic habitat preferences associated with Plains Topminnow, as both taxa are commonly found in aquatic vegetated backwaters of shallow streams (Ross and Ultsch 1980; Fischer and Paukert 2008). Gastropods have a caloric equivalent of 2.6 calories/individual (Johnson et al. 2006) and represent a higher percentage of caloric biomass than littoral macroinvertebrates (Richardson et al. 1998); potentially providing Plains Topminnow with an energetic benefit. Prey selectivity towards gastropods may also result from differing energy outputs required for grazing or hunting, and the inability of a prey item to hide or ward off predators (Sullivan et al. 2012). Despite the preferred lotic prey selectivity observed in this study, both lentic and lotic Plains Topminnow populations consumed a variety of available prey taxa which make topminnow adaptable to changing environmental conditions and food sources. Therefore they may be less likely to encounter negative population impacts due to competition for food resources from other species.

Lentic and lotic Plains Topminnow diets and prey availability did not significantly differ in this study. Therefore, contrasting feeding habits for Plains Topminnow described in previous studies (Stribley and Stasiak 1982; Rahel and Thel 2004; Bestgen 2014; Thompson 2014) likely do not result from differing prey availability between lotic and lentic systems. Similarities in prey availability may be a result of Plains Topminnow being backwater specialists and seeking out slower moving water in lotic systems (Rahel and Thel 2004); which share similar characteristics with, and often function like, littoral zones in ponds and lakes (Barnes and Mann 1980). Previous studies suggest heavily vegetated backwater habitat is preferred for Plains Topminnow egg deposition, rearing cover (Rahel and Thel 2004), and now food preference. Furthermore, Schumann (2012) suggested fine sediment type, high abundance of submerged vegetation, and cooler water temperatures may be factors indicating quality Plains Topminnow habitat; which are also habitat characteristics associated with gilled snail preferred habitat (Ross and Ultsch 1980).

The selective avoidance of decapods and ephemerotera is consistent with the previous quantified diet studies of Plains Topminnow. Decapod avoidance is most likely a result of Plains Topminnow gape limitations, as only juvenile decapods were found in Plains Topminnow diets; though adult decapods were highly abundant in the environment. Gape limitations were identified as restrictive to dietary selection of Western Mosquitofish (Mansfield and Mcardle 1998), which share similar morphologic characteristics and prey items with Plains Topminnow. Decapods were not identified as prey in southern Plains Topminnow populations, which could mean they exhibit a similar avoidance (Thompson 2014). However, decapods were potentially underrepresented in previous Plains Topminnow diet studies as extraction efficiency was not assessed for the gastro-lavage technique employed (Thompson 2014), and decapods are reported to be under-represented as prey items in similar lavage techniques (Lundgren et al. 2014). Ephemeroptera prey avoidance was likely the result of spatial differences as the energetic benefit to Plains Topminnow would be twice that of gastropods (Ciancio et al. 2007). While ephemeroptera occurred in greater abundance in our lotic study sites, they are more prominent in channels and drifts of lotic systems which are less suitable for topminnow (Pastuchova et al. 2008).

Plains Topminnow diets described in this study may change due to fish communities, trophic interactions, and seasonal prey availability. Study locations represented Plains Topminnow populations that coexist only with fathead minnows (*Pimephales promelas*) and terrestrial predators, such as birds, amphibians, and reptiles. The range of feeding habits exhibited in this study suggests Plains Topminnow are capable of adapting their diets to consume a variety of prey items. Additionally, the regional prey availability and food habits exhibited in this study represent the northern portion of a disjunct Plains Topminnow population and therefore a similar evaluation should be carried out for the southern Plains Topminnow populations.

***Diet overlap*:** Plains Topminnow and Western Mosquitofish did not have overlapping diets between the proximate populations sampled for this study, suggesting these two species could partition food resources if they cohabitated. Plains, Blackstripe *Fundulus notatus*, Gila *Poeciliopsis occidentalis*, Barrens *Fundulud julisia*, and Starhead *Fundulus dispar* Topminnows all share similar prey items with Western Mosquitofish (Rakes 1989; Childs 2006; Sutton et al. 2012). Additionally, a similar level of diet overlap was reported between Western Mosquitofish and Gila topminnow (Schoener's Index = 0.46) resulting in conclusions that food resources were not limiting cohabitation of these species, but rather other types of interaction (Mills et al. 2004; Childs 2006; Alcaraz et al. 2008). Plains Topminnow populations appear to be experiencing similar impacts other topminnow species have demonstrated when cohabiting with introduced Western Mosquitofish

(Schumann et al. 2016). Western Mosquitofish have been suggested as a main contributor in the demise of Barrens topminnow (Johnson and Bettoli 2003; Goldsworthy and Bettoli 2006; Laha and Mattingly 2007) and Sonoran topminnow *Poeciliopis occidentalis sonorensis* populations (Minckley 1969; Minckley 1973; Meffe et al. 1983; Meffe 1984). Studies have noted Western Mosquitofish predation on juvenile topminnow and harassing adults as the primary negative factor in reducing populations (Meffe et al. 1983; Laha and Mattingly 2007).

Management Implications: This study observed and supports Plains Topminnow populations consuming diptera in proportion to their availability in the environment. Western Mosquitofish also consume diptera species (i.e. mosquitos), but are not an effective biological control of mosquito populations (Kumar and Hwang 2006). Thus, stocking Plains Topminnow throughout its historic range as an alternative mosquito control mechanism may not provide additional or replicable mosquito abatement as previous studies have suggested (Bestgen 2014) as they consumed less diptera than mosquitofish. However, this hypothesis should be investigated further due to the potential avoidable impacts on native fish communities by limiting the continued introduction of a non-native species. Because Plains Topminnow consume diptera at a rate of which they are available, they may assist with mosquito abatement, while simultaneously preserving the integrity of native fish communities.

Gastropods are an important diet item for both lentic and lotic populations of Plains Topminnow and are associated with heavily vegetated backwater areas, the preferred habitat of Plains Topminnow (Schumann et al. 2015[a]). This affinity for prey in preferred habitat indicates the importance of habitat availability when developing management strategies and recovery plans. Conservation of Plains Topminnow moving forward should potentially include gastropods and their associated habitat as measurement for available preferred topminnow habitat.

Most topminnow species have not been able to cohabitate with Western Mosquitofish, and the Plains Topminnow is no exception (Meffe 1984; Laha and Mattingly 2007; Schumann et al. 2015[b]). Diet overlap of other topminnow species and Western Mosquitofish is limited primarily because mosquitofish cohabitate with topminnow for a short timeframe before displacement occurs, making cohabited populations difficult to encounter. Thus, in lieu of having co-mingled wild populations to examine we offered an alternative for examining diet overlap from regionally proximate sites for these two species. Our findings of a non-overlapping diet between Plains Topminnow and Western Mosquitofish were consistent with those found between Gila topminnow and Western Mosquitofish. The results of this study further support that future investigation of mosquitofish impacts on topminnow species should focus on other forms of competitive interactions, such as physical intimidation (Haas 2005) or direct predation of larval Plains Topminnow (Schumann et al. 2015[b]). Until these relationships are better understood repatriation events may have limited success (Schumann et al. 2017).

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