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ORIGINAL ARTICLE

Effects of stream intermittency on minnow (Leuciscidae) and darter (Percidae) trophic dynamics in an agricultural watershed

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Abstract

Stream intermittency is predicted to increase where water withdrawals and climate warming are increasing. In regions coupled with high fish diversity, understanding how intermittency influences fish trophic ecology is critical for informing ecosystem function. This study compared fish diets across seasons in perennial and intermittent streams to estimate the immediate and cumulative effects of stream drying on fish foraging patterns. We used gut content analysis to compare the diets of small-bodied, secondary consumer fishes, including two minnow and three darter species found in the lower Flint River Basin of southwestern Georgia, during both the summer (before stream dry-down) and fall (post flow resumption) seasons. Fish communities in perennial streams had greater diet richness compared to fishes in intermittent streams for both seasons. Darter diets were characterised by rheophilic aquatic insects in perennial streams and by benthic crustaceans (copepods, cladocerans and isopods) and predatory aquatic insects in intermittent streams. Minnow diets were typified by freshwater sponges, eggs and organic detritus in intermittent streams and by terrestrial insects and diatoms in perennial streams. Fishes in intermittent streams consumed significantly more benthic crustaceans in the fall (37% increase in proportional volume) compared to preflow cessation conditions in the summer, suggesting these organisms play an important, yet relatively unrecognised role in supporting fish communities in southeastern streams. Our findings enhance our understanding of how stream intermittency influences the trophic dynamics of secondary consumer fishes in an agricultural watershed increasingly affected by water scarcity.

KEYWORDS

benthic crustaceans, diet richness, intermittency, invertebrate recolonisation, secondary consumer fishes, trophic dynamics

1 | INTRODUCTION

Intermittent rivers and ephemeral streams, or waterways that include periods of flow cessation, are arguably the most widespread lotic water bodies globally (Datry et al., 2018; Larned et al., 2010). Intermittent waterways are projected to become more common in parts of the world with increasing human population densities, warmer and drier climates and/or anthropogenic water abstraction (Datry et al., 2018). Naturally intermittent streams are characterised by predictable flooding and drying regimes that generate dynamic habitats that filter species assemblages through various flow-related adaptations (Poff et al., 1997). Fishes that have evolved in intermittent stream networks have adapted to natural changes in flow by migrating to perennial refugia (Kerezsy et al., 2017), timing spawning with predictable (seasonal) flooding events (Lytle & Poff, 2004) and persisting in isolated pools (Pires et al., 2010). However, the response by fishes to intermittency in human-altered flow regimes remains unknown for many species (Lytle & Poff, 2004). Furthermore, the cumulative effects of flow cessation on food resource availability and the resulting dynamics in fish trophic ecology are not well known (Mas-Martí et al., 2010; Matthews & Marsh-Matthews, 2003) and yet could potentially influence changes in the distributions, demographics and vital rates of species, and ultimately stream ecosystem function.

The southeastern United States (SE USA) is a global hotspot for fish biodiversity, supporting 79% of all freshwater fish species found in the United States and Canada (Elkins et al., 2019). Streams in this region often support multiple species of small-bodied fishes belonging to two highly diverse families; the minnows (Leuciscidae; Tan & Armbuster, 2018), and darters (Percidae) (Boschung & Mayden, 2004; GMNH, 2008). Often functioning as secondary consumers in stream food webs, many minnows opportunistically forage in the water-column on drifting aquatic and terrestrial invertebrates, whereas darters typically are more selective feeders with preference for benthic animal prey. However, minnows and darters are generally flexible consumers (e.g. algivores, insectivores, detritivores, opportunistic piscivores; Alexandre et al., 2015; Power & Dietrich, 2002), and thereby function as integral conduits of energy and nutrient transfer in stream food webs (Taylor et al., 2010). Therefore, alterations to the food resources of secondary consumer fishes could have important implications for ecosystem processes, and especially in the southeastern United States where minnows are the most abundant vertebrate consumers in streams (Wheeler et al., 2018). Thus, comparing the trophic ecology of minnows and darters in perennial and intermittent streams may guide predictions regarding the effects of increasing intermittency on food web dynamics, and allow us to understand how populations adapt to shifting food availability as a function of drying and rewetting cycles (McIntosh et al., 2017).

The lower Flint River Basin (LFRB; Apalachicola River basin) of southwestern Georgia, USA, provides a context to test predictions about how stream fishes may cope with increasing intermittency. Streams in this region have historically supported diverse assemblages of aquatic biota, including fishes, and species endemic to the Apalachicola basin (Albanese, 2020; Boschung & Mayden, 2004). However, land use changes and intensification of irrigated agriculture have altered streamflow regimes such that historically perennial streams now cease to flow during some growing seasons (Golladay et al., 2016; Rugel et al., 2012). Additionally, climate change models predict a warmer climate, shifts in precipitation regimes, increased evapotranspiration rates and more frequent and intense droughts in this region (Ingram, 2013; IPCC, 2007). Thus, climate change coupled with water extraction for irrigated agriculture has the potential to exacerbate the frequency of intermittency of those streams most affected by groundwater pumping (Gordon et al., 2012). However, we do not know whether or how reductions in stream flows will alter the availability of food resources and resulting trophic dynamics for small-bodied fishes.

Although our study did not quantify food resource availability for fishes, research has demonstrated that available food resources often become reduced in streams that experience chronic drying due to the loss of aquatic-obligate taxa and shrinking aquatic habitats resulting in more simplified food webs (McHugh et al., 2015; McIntosh et al., 2017). In our study area, decreased aquatic insect richness has been documented in intermittent streams compared to perennial streams due to the delayed recolonisation of taxa lacking the necessary adaptations to persist or complete their life cycles under drying conditions (e.g. Ephemeroptera, Plecoptera, Trichoptera; Smith et al., 2017). In contrast, LFRB intermittent streams are characterised by abundant noninsect invertebrate assemblages that can withstand desiccation and rapidly recolonise streams upon inundation (e.g. Isopoda, Copepoda, Ostracoda; Smith et al., 2017). Fish trophic responses to low flows in the LFRB can also vary significantly depending on the respective trophic guilds of fishes. For instance, Davis et al. (2020) showed that as streams dry to isolated pools, diet shifts were not observed in the redeye chub (*Pteronotropis harperi*) (aquatic invertivore) but were significant for the Apalachee shiner (*Pteronotropis grandipinnis*) (terrestrial invertivore); presumably due to the reduced availability of terrestrial prey under drying conditions (Davis et al., 2020). Therefore, previous research in our study area collectively provides valuable context regarding the availability of invertebrate prey taxa and corresponding responses by fishes to inform our predictions for this study.

This research sought to understand both the immediate and cumulative effects of stream intermittency on the trophic ecology of minnows and darters in southwestern Georgia. We tracked the diet shifts of five fish taxa between early summer (prior to stream drying) and fall (following flow resumption in intermittent streams) to separate seasonal and hydrological controls on trophic patterns. We assessed diets at the species-level to account for distinct trophic guilds (e.g. terrestrial invertivores, aquatic invertivores, opportunistic piscivores) being represented within minnow and darter groups, thus allowing more comprehensive coverage of trophic patterns observed at the family level. To evaluate the immediate effects of stream drying, we identified the food resources that were most common in fish gut contents soon after flow resumption in intermittent streams following a period of flow cessation. To evaluate the cumulative effects of stream intermittency, we compared diets of darters and minnows between perennial and intermittent streams and evaluated whether stream drying would cause fishes to converge on food resources.

Additionally, we made the following specific predictions for this study. First, we expected fish diets would have greater richness in perennial streams compared to intermittent streams during both summer and fall seasons because of more hydrologically stable streams supporting more rheophilic taxa year-round. Secondly, we predicted fishes would display significant seasonal shifts in diet in intermittent, but not in perennial systems due to summer-time flow cessation in intermittent streams altering available food resources in the fall. Specifically, we expected that upon flow resumption, fish diets in intermittent streams would be dominated by aquatic invertebrate taxa with desiccation-resistant adaptations. Lastly, we expected that species-specific diets would differ between perennial and intermittent streams even in early summer, that is, following a period of sustained flow, due to the potential delayed reassembly of benthic insect communities. We specifically predicted that diet differences would be more apparent in darters compared to minnows, because of their greater reliance on benthic aquatic insect prey as opposed to drifting, terrestrially derived or basal prey resources.

2 | METHODS

2.1 | Study area

The Ichawaynochaway Creek Basin (ICB) is a major sub-drainage of the lower Flint River Basin in the Coastal Plain of southwestern Georgia, USA (Figure 1; Golladay & Battle, 2002). The ICB geology is defined by two distinct physiographic districts including the rolling Fall Line Hills in the north and the karstic Dougherty Plain in the south (Figure 1; Golladay & Battle, 2002). ICB streams begin as springs and seeps in the headwaters of the Fall Line Hills that transition into streams with porous Ocala limestone reaches, high groundwater inputs and floodplain swamps in the Dougherty Plain (Golladay & Battle, 2002; Rugel et al., 2012). Nearly half of the land cover in the ICB is agriculture while the remaining land use is largely forested with pine uplands, riparian hardwood forest and forested wetlands (Golladay & Battle, 2002; Rugel et al., 2016).

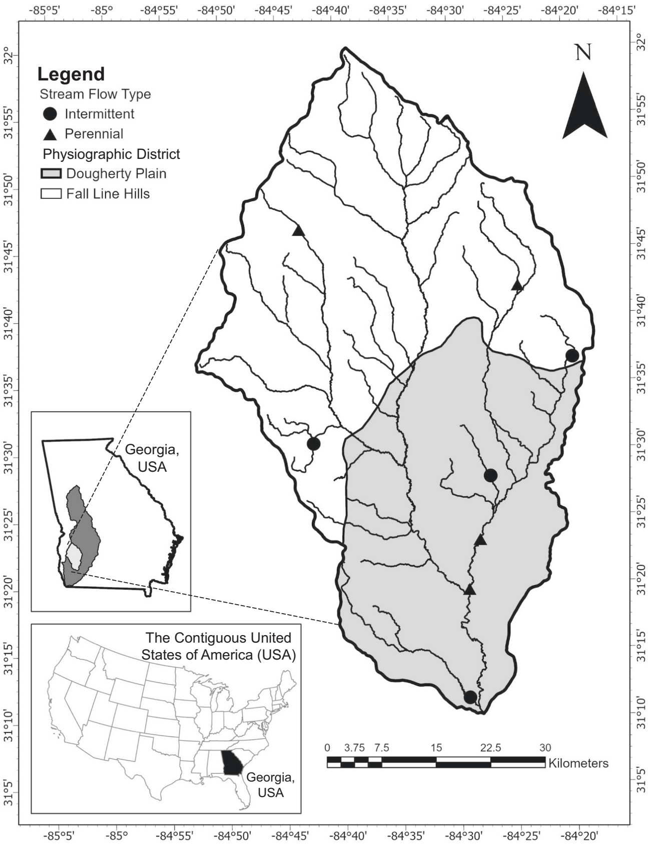

FIGURE 1 Map of eight stream study sites in the Ichawaynochaway Creek Basin (ICB) (284,835 ha), Georgia, USA

Much of the agriculture in the ICB is in large fields of row crops with centre-pivot irrigation that relies largely on pumping groundwater from the Upper Floridan Aquifer in the south, and from surface water withdrawals in the northern portions of the basin (Rugel et al., 2012, 2016). This agriculture was adopted in the region in the 1970s and has resulted in a shift in historical flow regimes, such that annual 7-day minimum stream flows in the Ichawaynochaway Creek have been reduced by 61% compared to preirrigation levels (Rugel et al., 2012). The typical hydrological regime for the ICB is characterised by high flows in the winter and spring and low flows in the summer and fall when evapotranspiration rates and air temperatures are high (Atkinson et al., 2009). Summer low flows frequently overlap with peak irrigation season, and often include the predictable drying of intermittent streams and below-average flows in perennial streams during drier years (Rugel et al., 2012). Additionally, during recent periods of climatological, multi-year drought, some historically perennial streams have ceased to flow (Golladay et al., 2003; Rugel et al., 2016), indicating that climate change and increased frequency and intensity of groundwater pumping in this region are pushing historically perennial streams towards increasing intermittency (Gordon et al., 2012; Rugel et al., 2012).

We sampled fishes in both perennial and intermittent streams before and after an annual stream drying event (August–September 2019) to isolate the effects of chronic and recent stream drying on fish trophic ecology in the ICB. Although the ICB endured a moderate to severe drought in September and October (Konrad et al., 2019), all of the perennial sites maintained flow throughout this study, whereas intermittent sites ceased flowing for 2–6 months. Throughout our sampling period, an intermittent groundwater-fed tributary of the Chickasawhatchee Swamp (Little Spring Creek) maintained standing water at low levels, while all the other intermittent streams transitioned between states of dry stream beds, isolated pools or standing water between the months of August and October. Sustained flow resumed in intermittent streams between November and December.

2.2 | Sampling design

We implemented a randomised complete block design by selecting eight sites across perennial and intermittent streams within two distinct physiographic districts of the ICB (Table 1). Sites were blocked by network position in the ICB, by being either within the limits of the Dougherty Plain or the Fall Line Hills (Figure 1). In this study, we were not specifically interested in the effect of physiographic district on fish diets. However, the quality and concentration of allochthonous and autochthonous basal food resources can vary significantly across the Fall Line Hills and Dougherty Plain due to factors including stream channel morphology, stream size, groundwater inputs and connectedness with surrounding floodplains (Atkinson et al., 2009; Golladay et al., 2000). Therefore, the blocked design allowed us to account for potentially distinct basal resources and respective primary consumer assemblages (e.g. shredder, scraper, collector, predator invertebrates) yielding variable prey resources for fishes. We identified streams as locations for prestudy sampling reconnaissance if records confirmed that they historically hosted a majority of our target fish species (Davis, 2017). We then randomly selected sites from the subset of stream locations that yielded positive detections of at least one or more of our target fish species. Our target fish species included two minnows: the redeye chub (*Pteronotropis harperi*) and Apalachee shiner (*Pteronotropis grandipinnis;* endemic to our study basin), and three darters: the brown darter (*Etheostoma edwini*), swamp darter (*Etheostoma fusiforme*) and blackbanded darter (*Percina nigrofasciata*). At each site we sampled the representative stream reaches twice, once before the growing season minimum flow/dry-down (June-July), and again in the dormant season once flows had resumed in intermittent streams (October-January). During each sampling event, we conducted fish community surveys (Appendix S1: Table S1), collected fish for gut content analysis (GCA), and recorded wetted width and mid-channel depth (every 12.5 m along the reach), and measured discharge.

2.3 | Fish sampling methods

We implemented protocols that met animal care and use standards to capture and euthanise fishes (e.g. minimised handling time, used appropriate electroshocking settings and euthanasia concentrations). We designated a 50-m representative stream reach in each of the eight selected sites based on (1) accessibility and feasibility for effective seining and (2) representation of stream microhabitats (e.g. runs, riffles, pools, shallow areas along channel margins) that host our target darter and minnow species. Upon flow resumption in intermittent streams, we waited between 7 and 30 days before resampling sites to allow time for fish to recolonise streams from perennial water bodies. We collected fish using a 1.2 m × 3.1 m seine net with 3.2 mm mesh and a Smith-Root LR-24 Backpack Electrofisher. Within each study reach, we conducted 20 independent kick-sets (in which two people held a seine perpendicular to stream flow with the lead line on the stream bottom, and a third person electrofished a 2 m^2 area immediately upstream). Fishes captured were identified to the species level, measured to the nearest millimetre (total length) and immediately released live into the habitat from which they were captured. A subset of up to 20 individuals that represented a broad range of size classes were kept for GCA per each target fish species. These fish were humanely euthanised by immersion in a 250 mg/L solution of MS-222 buffered with sodium bicarbonate (Leary et al., 2013). We preserved euthanised fishes on ice prior to transport to the laboratory. Each fish was assigned a unique numeric code and either 1) immediately dissected fresh or 2) stored in a −25°C freezer until we performed subsequent GCA analyses. All fishes were measured for length prior to freezing.

2.4 | Gut content analysis

We performed GCA on 228 individual fish and excluded all fishes with empty or near empty (<10% full) guts from analyses. Minnows lack a true stomach; thus, to standardise methods across all taxa, we collected items found in the entire gastrointestinal (GI) tract for each fish we examined. Each GI tract was dissected, from the oesophagus to the anal vent (Davis et al., 2020), and preserved in a 70% ethanol solution (Rybczynski et al., 2008). We focused on individuals > 25 mm standard length (SL) to make species-level inferences based on the larger individuals of the population that can forage without gape limitations. Individuals that were < 25 mm SL were only included if sample sizes were limiting for a given species. We performed permutational analysis of variance (PERMANOVA) tests to determine if diet composition (represented by volumetric proportions for each diet item) varied significantly across SL (using categorical 5 mm bins) for each species. However, since we failed to find diet differences between the largest and smallest individuals within a species, we did not include SL as an effect in subsequent analyses.

We employed a modified approach (described later) of an indirect volumetric method, known as the “squash” method, to estimate the volumetric contribution of gut contents. In this method, all gut contents are “squashed” to a uniform depth and the total area of the gut contents are measured (Hellawell & Abel, 1971; Hyslop, 1980). We analysed each gut under a stereo microscope (Olympus SZX7) and used a transparent Petri dish with 1 mm^2 grid graph paper to measure the volume that each distinct food item category contributed to the total gut contents. We quantified food item volume by counting the total number of 1 mm^2 grids occupied to the nearest tenth of a millimetre. To determine the total gut content volume, we summed the volume measurements for each distinct food item present. For food items that could be represented as distinct units or individuals, we recorded the total number of each food item present. To estimate the volume of diatoms that were integrated into amorphous detrital components we haphazardly selected 10 grids to estimate the proportion of grids occupied; and extrapolated the proportion of grids occupied into total volumetric contributions (Rybczynski et al., 2008).

We identified macroinvertebrates to the family taxonomic level (with the exception of Chironomidae sub-families), while all other food items were identified to the lowest practical level (Davis et al., 2012). To calculate diet richness, we considered all individual diet items categorised at the family level or broader. The family-level resolution of gut content items was summarised as Frequency of Occurrence (%F) values (Appendix S1: Tables S2–S6). Because many insect families were sparsely represented, we pooled aquatic insects to Order (except for Diptera) for subsequent analyses. The fish taxa analysed for GCA were all small bodied, thus the size and volume of

TABLE 1 Physical characteristics of stream reaches sampled throughout the study period

Stream site	Stream-flow type	Physiographic district	Stream order	Mean stream cross-section area (m^2)	Mean discharge (ft^3/s)	Mean flow-state rank
Chk1-Elmodel	P	DP	4	7.67 ± 3.15	29.21 ± 16.38	4.14 ± 0.38
Chk2-Clear Lake	P	DP	4	6.87 ± 4.14	12.54 ± 14.12	3.83 ± 0.75
Pachitla	P	FLH	4	2.29 ± 0.37	10.25 ± 5.23	4.00 ± 0.00
Big Cypress	I	DP	3	2.99 ± 0.96	5.04 ± 2.95	1.91 ± 2.02
Brantley	P	FLH	3	2.30 ± 1.09	8.02 ± 4.66	4.33 ± 0.52
Kiokee	I	FLH	2	0.76 ± 0.11	1.95 ± 4.29	3.42 ± 1.56
Little Spring	I	DP	2	4.10 ± 1.15	9.99 ± 7.07	3.89 ± 0.78
Tennelle	I	FLH	1	1.50 ± 0.34	0.50 ± 0.73	3.00 ± 1.83

Note: Mean hydrology data for each site were monitored monthly (June 2019–January 2020), while mean stream cross-section area was collected at each sampling event. Mean discharge indicates approximate average monthly discharge values measured only when streams maintained a connected channel (including zero flow values). Flow-state ranks are derived from five fixed values that reflect the average state of flow the stream site was in throughout the study. Flow-state rank values are as follows: 0 = Dry, 1 = Isolated pools, 2 = Connected channel with zero flow (standing water), 3 = Connected channel with minimal flow, 4 = Average flow, 5 = Above average flow. Variation is represented as ± 1 standard deviation.

food items were similar across taxa and were consistently less than or equal to 1 mm in height. Therefore, we used the number of grids occupied (1 mm length × 1 mm width) and a 1 mm height assumption as a surrogate for true gut volume since this measurement was consistently comparable and standardised across all food items analysed. This modified approach of the "squash" method allowed us to rapidly assess a greater sample size of guts by permitting easier separation and identification of distinct food items. For rare food items that exceeded 1 mm in height, we measured the total volumetric displacement in a 10-mL graduated cylinder and converted millilitres to cubic millimetres for volumetric contribution. Volumetric contributions of each food item were then converted into a proportion value of the total gut content amorphous mass as such:

1. Total gut content volume (mm^3)* = Total area of grids occupied (mm^2) × 1 mm height
2. Food item volume (mm^3)** = Area of grids occupied by food item (mm^2) × 1 mm height
3. Food item proportion = Food item total volume (mm^3) / Total gut content volume (mm^3)

* = Add the volumetric displacement (mm^3) of food items >1 mm height to the total gut content volume (mm^3) for applicable gut samples.

** = Use the volumetric displacement (mm^3) of food items >1 mm height to determine the food item volume (mm^3) for food items >1 mm height.

2.5 | Statistical analyses

We calculated taxa richness of food items within each fish gut to characterise the diet richness for each stream-flow type, season and species combination. Furthermore, since we were interested in different life stages of aquatic insects as they related to fish foraging behaviour, we considered larval, pupal and adult stages of dipterans as distinct diet categories. We then used a three-way analysis of variance (ANOVA) followed by post hoc pairwise comparisons of true means with the Tukey's Honestly Different test to determine if fish diet richness was significantly different between three-way interactions of stream-flow type, season and species. When we were only interested in a subset of treatment combinations generated from a three-way ANOVA, we used the "emmeans" package to conduct pairwise comparisons of interest (Lenth, 2020). All statistical analyses were performed in R v.3.6.3 (R Core Team, 2019). We used Shapiro–Wilk's test and Bartlett's test to confirm that all variables in parametric analyses met the assumptions of normality and variance homogeneity.

To explore patterns in fish diets between perennial and intermittent streams we conducted multivariate analysis on gut content data (as volumetric proportions) with nonmetric multidimensional scaling (NMDS) and permutational multivariate analysis of variance (PERMANOVA). To meet the assumptions of normality, we performed arc-sine square root transformations on all gut content data represented as volumetric proportions in NMDS and PERMANOVA analyses (Rybczynski et al., 2008; McDonald, 2014). When using NMDS, we selected the models that allowed convergence with the fewest possible axes while maintaining stress below 20 (Clarke, 1993). When three axes were required for NMDS convergence, we selected the two axes that best distributed the samples along the x-axis for data visualisation purposes. NMDS analysis was conducted using Bray–Curtis dissimilarity and was performed on all gut content data (n = 228) simultaneously. We included gut content items that occurred in ≥ 5% of the entire fish population in the analyses (Bonato et al., 2018). Gut content items that were < 5% were grouped when applicable (e.g. terrestrial Coleoptera and spiders as "terrestrial arthropod other") or were excluded from analyses. We then performed similarity percentages analyses to determine which diet items most significantly contributed to divergent fish diets.

We performed PERMANOVAs to explore diet differences between fish family, stream-flow type and season interactions. We tested the assumption of homogeneity of multivariate dispersion and when significant, included the blocking variable of physiographic district in the analysis. We only included physiographic district as a block in multivariate analyses that included volumetric proportions of available diet items as the response variable (except for the indicator analysis which did not allow a blocking variable). This is because we were only interested in accounting for how a physiographic district may influence the types of diet items that are made available to fish, rather than the overall quantity or richness of diet items made available to fish. If fish family, stream-flow type, season or their interaction was found to be significant, we performed pairwise comparisons and constructed Bonferroni corrected p values for the entire model and each interaction term respectively (Arbizu, 2020).

We then implemented a modified version of a traditional indicator analysis to determine which diet items were most associated with certain fish families in perennial and intermittent streams. A traditional indicator analysis identifies species that exhibit fidelity and specificity towards habitat types based on abundance and frequency factors (Dufrêne & Legendre, 1997). Our modified indicator analysis used the volumetric proportions (instead of abundance) of all diet items identified to the family level or broader across all five fish species. Due to sample size limitations at the intraspecific level, we grouped fishes by family to identify indicator diet items for fish family and stream-flow type interactions (see Table 2 legend for the modified indicator analysis formula).

We also performed independent species-level PERMANOVAs on three fish taxa (*E. edwini*, *P. harperi* and *P. nigrofasciata*) that yielded adequate sample size to assess intraspecific diet differences between perennial and intermittent streams during extended-flow conditions in the early summer. We included gut content items that occurred in $\geq$ 5% of each sub-population (including each species, stream-flow type and season combination of interest). Each species-level PERMANOVA assessed if volumetric proportions of diet items (arc sin square root transformed) were significantly different as a function of stream-flow type and physiographic province was included as a blocking variable when significant. All multivariate analysis was performed using the "vegan" and "labdsv" packages (Oksanen et al., 2019; Roberts, 2019).

To compare the proportion of benthic crustaceans consumed by fishes, we used the "glmmTMB" package to construct generalised linear models using a beta distribution with the "logit" link (Brooks et al., 2017). Fixed effects included stream-flow type, season and their interaction and site and fish species were included as random effects (herein referred to as the single original model). Wald tests were used to extract chi-square values and significant p values ($p \leq .05$) associated with generalised linear models (Fox & Weisberg, 2019). Organisms grouped into benthic crustaceans included ostracods, isopods, copepods, cladocerans and unidentified microcrustaceans. To allow model convergence, the numeric response variable of the proportion of benthic crustaceans (x) was scaled as follows: $(x * (\text{length}(x) - 1) + 0.5)/\text{length}(x)$, where length represents the number of the observational units (Douma & Weedon, 2019). We created a single original model and a separate covariate model containing a dispersion parameter (a season and stream-flow type interaction) to relax the assumption of beta models which specifies that the dispersion is the same across treatments (Douma & Weedon, 2019). For model selection, we then used Akaike's information criteria (AIC) and the package "AICcmodavg" to compare Akaike weights between the two models (Mazerolle, 2020). We then compared 95% confidence intervals to determine significant differences between season and stream-flow type interaction terms.

3 | RESULTS

3.1 | Comparing minnow and darter diets in perennial and intermittent streams

Fishes in perennial streams consumed a greater number of diet items in the summer ($n = 46$) and fall ($n = 43$) compared to fishes in intermittent streams in the summer ($n = 33$) and fall ($n = 17$). Diets had significantly greater richness in perennial streams compared to intermittent streams (ANOVA, $F_{1,212} = 24.89$, $p = 1.27\text{e-}06$). There was a significant interaction between season and stream-flow type (ANOVA, $F_{1,212} = 5.79$, $p = .017$) as well as species and stream-flow type (ANOVA, $F_{3,212} = 4.33$, $p = .006$). When compared to intermittent streams, diets in perennial streams had slightly more richness in the summer (Tukey's HSD, $p = .039$) and substantially more richness in the fall (Tukey's HSD, $p = < .001$) (Figure 2a). Within stream-flow types, richness was not significantly different across seasons for perennial ($p = .302$) or intermittent streams ($p = .315$).

We could compare diet richness between stream types for four of the five species in at least one season. These comparisons showed mixed results. *P. nigrofasciata* consumed more diet items in intermittent streams ($\bar{x} = 8.00 \pm 1.02$ SE) compared to perennial streams ($\bar{x} = 5.92 \pm 0.46$ SE) in summer, although this difference only approached statistical significance ($\text{Pairwise}_{\text{emmeans}}$, $p = .064$) (Figure 2b). In contrast, two minnow species consumed significantly more diet items in perennial streams compared to intermittent streams; *P. grandipinnis* in the summer ($\text{Pairwise}_{\text{emmeans}}$, $p = .0003$) and *P. harperi* in the fall ($\text{Pairwise}_{\text{emmeans}}$, $p = .0005$) (Figure 2b). Differences in diet richness were nonsignificant for *E. edwini* for both seasons and *P. harperi* in the summer. Comparisons could not be made for fishes that were not collected in perennial streams (*E. fusiforme*) or in intermittent streams in the fall (*P. grandipinnis* and *P. nigrofasciata*).

Fifteen food items had the greatest probability ($p \leq .001$) of driving divergent diets among individual fish in relation to NMDS axes (NMDS Stress = 16.9, $k = 3$; Figure 3; Appendix S1: Table S7). Minnows tended to separate from darter species along Axis 2, containing diets with more organic detritus, wood, sand and diatoms compared to chironomids, Ephemeroptera and Trichoptera (Figure 3). Samples from intermittent streams tended to score higher on Axis 3, positively correlated with Cladocera and Copepods and

TABLE 2 Indicator analysis output for diet items identified as significant indicators for fish family and stream-flow type interactions

Diet items	Fish family and stream-flow type	Indval	*p* Value	Frequency
Chironomidae (Tanypodinae)	Percidae, Intermittent	0.373	0.001	98
Copepoda	Percidae, Intermittent	0.225	0.002	24
Aquatic Coleoptera larvae	Percidae, Intermittent	0.178	0.001	10
Isopoda	Percidae, Intermittent	0.153	0.001	6
Trichoptera	Percidae, Perennial	0.588	0.001	82
Chironomidae (Non-Tanypodinae)	Percidae, Perennial	0.411	0.002	190
Ephemeroptera	Percidae, Perennial	0.306	0.005	72
Ancylidae	Percidae, Perennial	0.091	0.023	9
Detritus	Leuciscidae, Intermittent	0.309	0.026	218
Wood	Leuciscidae, Intermittent	0.225	0.012	71
Porifera	Leuciscidae, Intermittent	0.129	0.019	12
Unidentified microcrustacea	Leuciscidae, Intermittent	0.113	0.016	11
Aquatic egg	Leuciscidae, Intermittent	0.093	0.033	8
Diatoms	Leuciscidae, Perennial	0.525	0.001	54
Terrestrial Diptera	Leuciscidae, Perennial	0.385	0.001	54
Unidentified terrestrial Insects	Leuciscidae, Perennial	0.139	0.020	19
Formicidae	Leuciscidae, Perennial	0.133	0.015	15
Unidentified Insects	Leuciscidae, Perennial	0.089	0.045	9

Note: Diet items were identified in gut content analysis of darters ($n = 84$) and minnows ($n = 144$) in perennial and intermittent streams. Indval was modified from Dufrêne and Legendre (1997), where *N* (abundance) is substituted with the proportional volumetric contribution of diet items (*P*), individuals are substituted with diet items, and sites are substituted with individual fish as follows: ($\text{Indval}_{ij} = \text{Pdiet item}_{ij}/\text{Pdiet item}_{i} * \text{Nfish}_{ij}/\text{Nfish}_{j} * 100$). Frequency indicates how many times a particular diet item was present in a distinct individual in gut content analysis of all fishes ($n = 228$). *P* value indicates all diet items found to be significant indicators of fish family and stream-flow type interactions when $\alpha = 0.05$.

negatively with terrestrial insects (Figure 3). Samples from perennial streams ranked slightly higher along Axis 1 and were positively correlated with aquatic insects and basal resources and negatively with benthic crustaceans and most terrestrial insects. Similarity percentages analysis indicated that organic detritus, non-Tanypodinae chironomids and Ephemeroptera contributed to the greatest dissimilarity between diets classified by fish family, stream-flow type or season (explaining >30% of the total variation for each comparison; Appendix S1: Table S7). Additional diet items contributing to differences between families, stream types and seasons included Tanypodinae, Trichoptera, Cladocera, diatoms, unidentified aquatic insects and sand (Figure 3; Appendix S1: Table S7).

The blocking variable of physiographic district (Fall Line Hills vs. Dougherty Plain) had a significant effect on the volumetric proportions (arcsine square root transformed) of fish diet items ($\text{PERMANOVA}_{\text{adonis}}$, $F_{1,226} = 8.45$, $p = .001$) and was included in the subsequent community-level PERMANOVA. Fish diets were significantly different as a function of a three-way interaction between stream-flow type, season and fish family ($\text{PERMANOVA}_{\text{adonis}}$, $F_{7,220} = 12.47$, $p = .001$) and there was a significant interaction between stream-flow type and family ($\text{PERMANOVA}_{\text{adonis2}}$, $F_{1,220} = 4.30$, $p = .002$). Fish diets were distinct between minnows and darters in intermittent streams in the summer ($\text{Pairwise}_{\text{adonis}}$ $p = .028$) but not in the fall ($\text{Pairwise}_{\text{adonis}}$, $p = 1.00$). In perennial streams, fish diets were distinct between fish families in both summer ($\text{Pairwise}_{\text{adonis}}$, $p = .028$) and fall ($\text{Pairwise}_{\text{adonis}}$, $p = .028$). Across both families, fish diets were significantly different between seasons in intermittent streams ($\text{Pairwise}_{\text{adonis}}$, $p = .006$), but not significantly different in perennial streams ($\text{Pairwise}_{\text{adonis}}$, $p = .012$). Indicator analysis determined that 18 diet items (out of 52 total) were representative of minnow and darter diets in perennial and intermittent streams (Table 2). Terrestrial-derived prey items were strictly associated with minnows in perennial streams (Table 2). Specifically, terrestrial insects, including formicids, dipterans, hymenopterans and coleopterans were important prey in perennial streams for one minnow species, *P. grandipinnis* (Figure 3; Appendix S1: Table S5), with terrestrial arthropods accounting for 15% and 30% of summer and fall gut content volume in perennial sites respectively (Figure 4). Both minnows, *P. grandipinnis* and *P. harperi*, also consumed substantial quantities of diatoms in perennial streams (Figure 4, Table 2).

Diet items associated with minnows in intermittent streams included freshwater sponges (Porifera), unidentified microcrustacea (likely degraded copepods and/or ostracods), organic detritus, wood and sand (Table 2). Darter diets were composed mainly of aquatic arthropods, with benthic crustaceans as a common dietary item in intermittent streams (Figure 4). Predatory aquatic insects including Tanypodinae and Coleoptera larvae (primarily Gyrinidae, Dytiscidae and unidentified larvae) were associated with darters in intermittent streams (Table 2). Trichoptera and Ephemeroptera were strongly associated with darter diets in perennial streams (Table 2). In intermittent streams, both minnows and darters consumed substantial

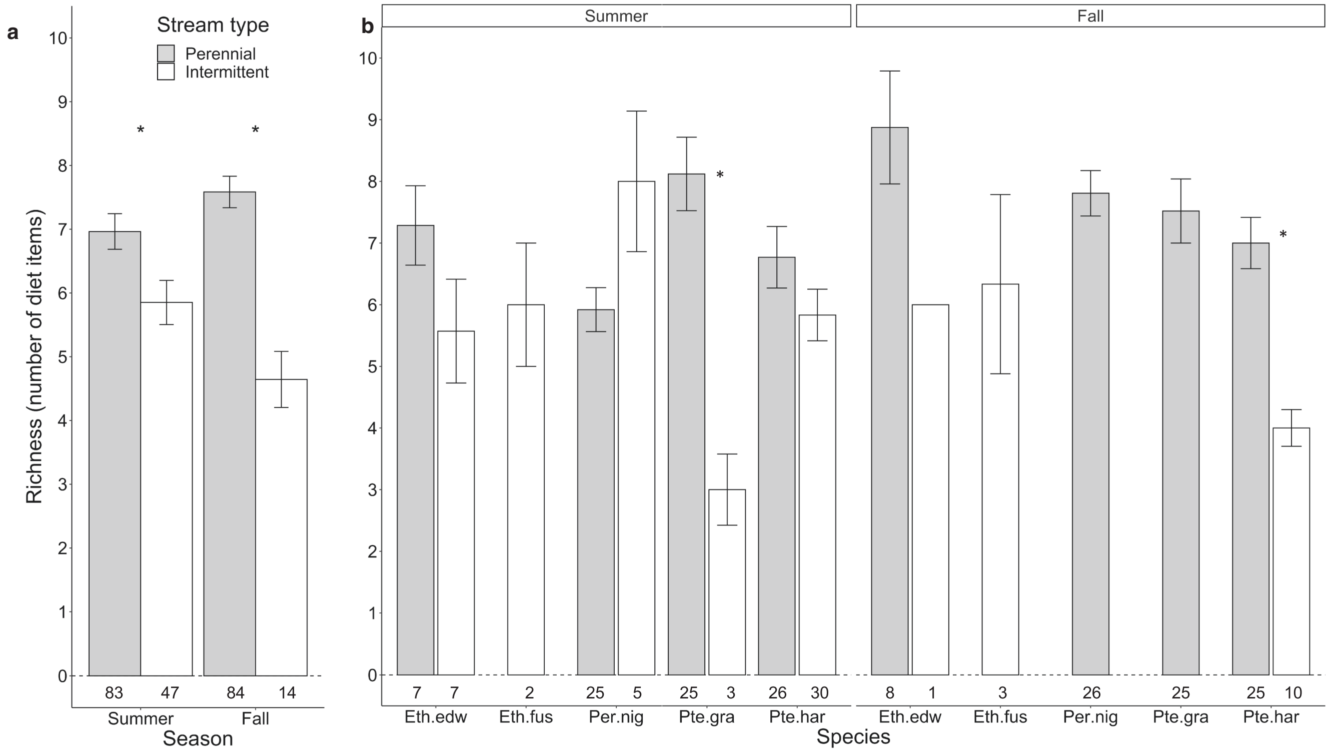

FIGURE 2 Comparison of mean richness of fish diets in perennial and intermittent streams at the community level (a) and species level (b) for summer and fall seasons. Richness is represented as the number of distinct diet items, identified at the sub-family level or broader, in all fishes ($n = 228$). Sample sizes of fish stomachs analysed per treatment are indicated below each bar in both panel (a) and (b). Variation is represented as ±1 standard error. Asterisks represent within season comparison of stream-flow types that had significantly different diet richness ($\alpha = 0.05$; (a) Tukey's Honestly Significant Difference and (b) $\text{Pairwise}_{\text{emmeans}}$). Vacant columns indicate negative species collections ($n = 0$) for respective stream-flow type and season combinations. Species are abbreviated by the first three letters of their scientific genus and species name respectively. The full scientific name for each species is defined as such: *Etheostoma edwini* (brown darter), *Etheostoma fusiforme* (swamp darter), *Percina nigrofasciata* (blackbanded darter), *Pteronotropis grandipinnis* (Apalachee shiner) and *Pteronotropis harperi* (redeye chub)

quantities of benthic crustaceans (Figure 3), which were abundant in fish diets in the fall (Figure 4). For fall intermittent stream samples across three fish taxa ($n = 14$), total volumetric contributions of benthic crustaceans were Cladocera (49%), Isopoda (29%), Copepoda (20%) and Ostracoda (2%). In the same subset of samples ($n = 14$), the percent frequency of cladocerans (71%) and copepods (36%) was greater than isopods (21%) and ostracods (14%) (Appendix S1: Table S2 and S3, and S6; Figure S1).

3.2 | Intraspecific diet differences between perennial and intermittent streams during extended-flow conditions

The blocking variable of physiographic district was significant for *P. harperi* ($p = .001$) and nonsignificant for *P. nigrofasciata* ($p = .212$) and *E. edwini* ($p = .31$). Volumetric proportions (arcsine square root transformed) of diet items were significantly different between intermittent and perennial streams in the summer following extended-flow conditions for one minnow, *P. harperi* ($\text{PERMANOVA}_{\text{adonis}}$, $F_{1,54} = 4.32$, $p = .001$), and both darter species that were collected in perennial sites (i.e. *P. nigrofasciata*, $\text{PERMANOVA}_{\text{adonis}}$, $F_{1,28} = 3.65$, $p = .001$, and *E. edwini*, $\text{PERMANOVA}_{\text{adonis}}$, $F_{1,12} = 3.14$, $p = .023$). *P. harperi* guts contained substantial volumes of organic detritus in both stream types, but greater amounts of diatoms and insects in perennial streams (Figure 4). Darter diets from perennial streams contained a greater volume of Trichoptera than those from intermittent streams (Figure 4) but otherwise were similar. Volumes of Ephemeroptera were greatest in perennial streams for *E. edwini* and in intermittent streams for *P. nigrofasciata* (Figure 4). Although overall volumetric contributions were minimal, juvenile minnows were only observed in the guts of *P. nigrofasciata* from perennial streams (Figure 4).

3.3 | Benthic crustaceans in fish diets in intermittent streams after flow resumption

The covariate model ($w = 0.74$) that included the dispersion parameter of a season and stream-flow type interaction term had greater support than the original model ($w = 0.26$). Hence, we selected the covariate model to predict the consumption of benthic crustaceans in perennial and intermittent streams. Season and stream-flow type interaction significantly affected the volumetric proportion (scaled) of benthic crustaceans in fish diets (df = 1, $\chi^2 = 23.39$, $p = < .001$;

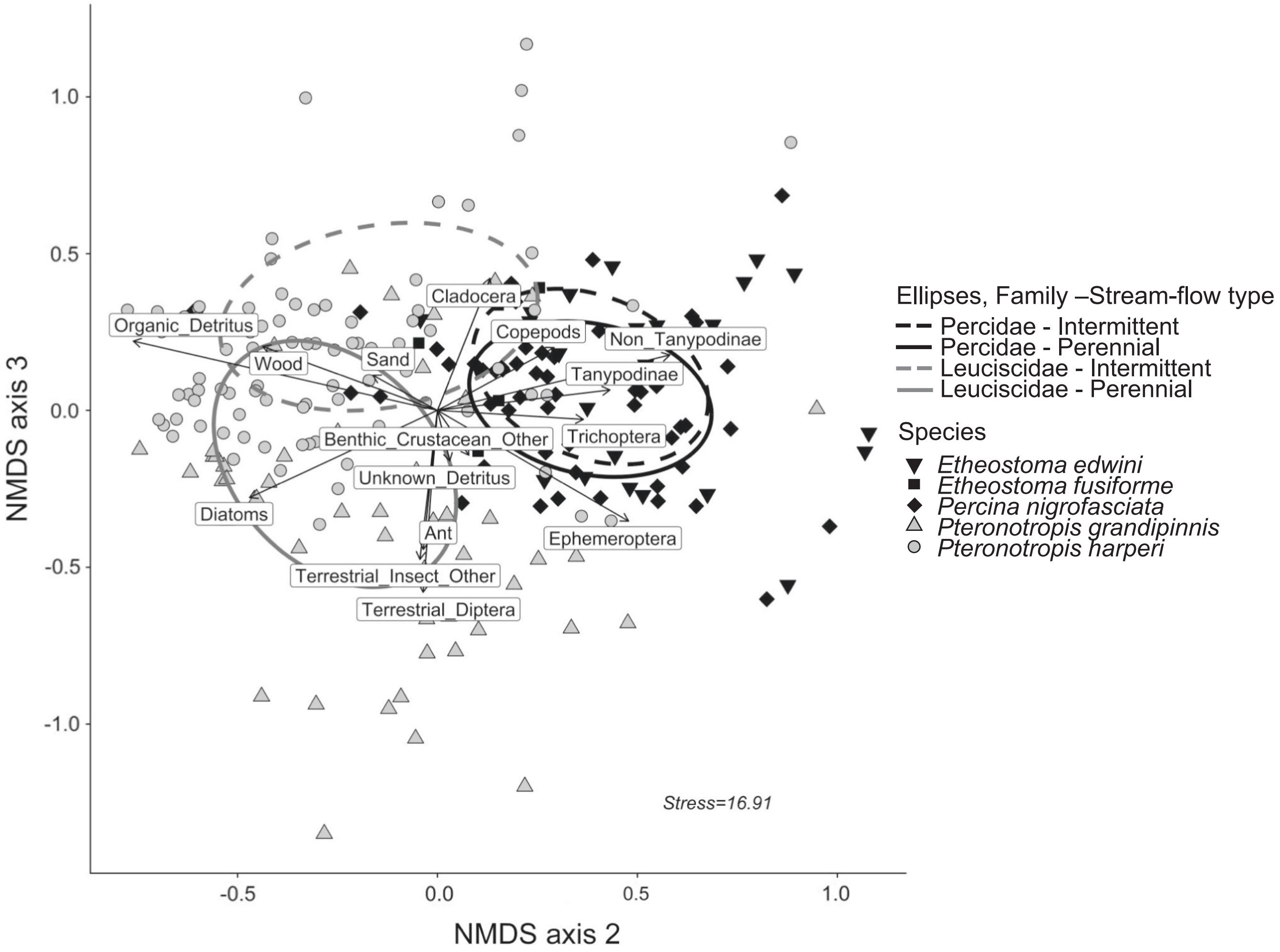

FIGURE 3 Nonmetric multidimensional scaling (NMDS) two-dimensional ordination on arcsine square-root transformed volumetric proportions of gut content samples for five fish taxa in perennial and intermittent streams ($n = 228$). Bray–Curtis dissimilarity distances were determined for all pooled fishes collected throughout the study. 95% confidence intervals for fish family and stream-flow type interactions are represented as ellipses. The significant cut-off value for diet item vectors is $p \leq .001$. A complete list of all significant vectors ($p < .05$) with respective similarity percentage values can be found in the Appendix S1: Table S7

Akaike weight = 0.74). Confidence intervals (95%) showed that the consumption of benthic crustaceans was significantly greater in intermittent streams in the fall when compared to intermittent streams in the summer, and when compared to perennial streams in either season (Figure 5). There was no significant effect of season on the consumption of benthic crustaceans in perennial streams (Figure 5). The predicted volumetric proportion (scaled) of benthic crustaceans was an order of magnitude larger in intermittent streams during the fall (0.44) when compared to all of the other stream-flow type-sampling period combinations (intermittent-summer: 0.07; perennial-summer: 0.06; and perennial-fall: 0.07; Figure 5).

4 | DISCUSSION

As climate change and water extraction increasingly exert pressures on aquatic biota (Datry et al., 2014), there is a pressing need to determine how more frequent and intense periods of water scarcity will alter fish trophic ecology and resulting ecosystem function. This study identified intraspecific and community-level differences in minnow and darter diets as a function of stream hydrology and seasonal interactions. From this work, we found that diets of fishes have significantly reduced richness in intermittent streams compared to perennial streams, especially immediately following flow resumption. Our results highlight that the trophic ecology of darters and minnows in this system reflect macroinvertebrate (Smith et al., 2017) and fish communities (Davis et al., 2020) shaped by intermittency. Our diet results suggest these intermittent communities are characterised by desiccation resistant invertebrates (Smith et al., 2017) and resilient fish taxa that exhibit trophic flexibility (Davis et al., 2020). Our research also illuminates the potential for minnows and darters to serve as sentinels for pelagic and benthic food resource availability in stream networks that experience dynamic shifts in hydrology. Here, we discuss the potential implications of our results regarding regional increases in intermittency and in other watersheds that are subjected to water withdrawals to support

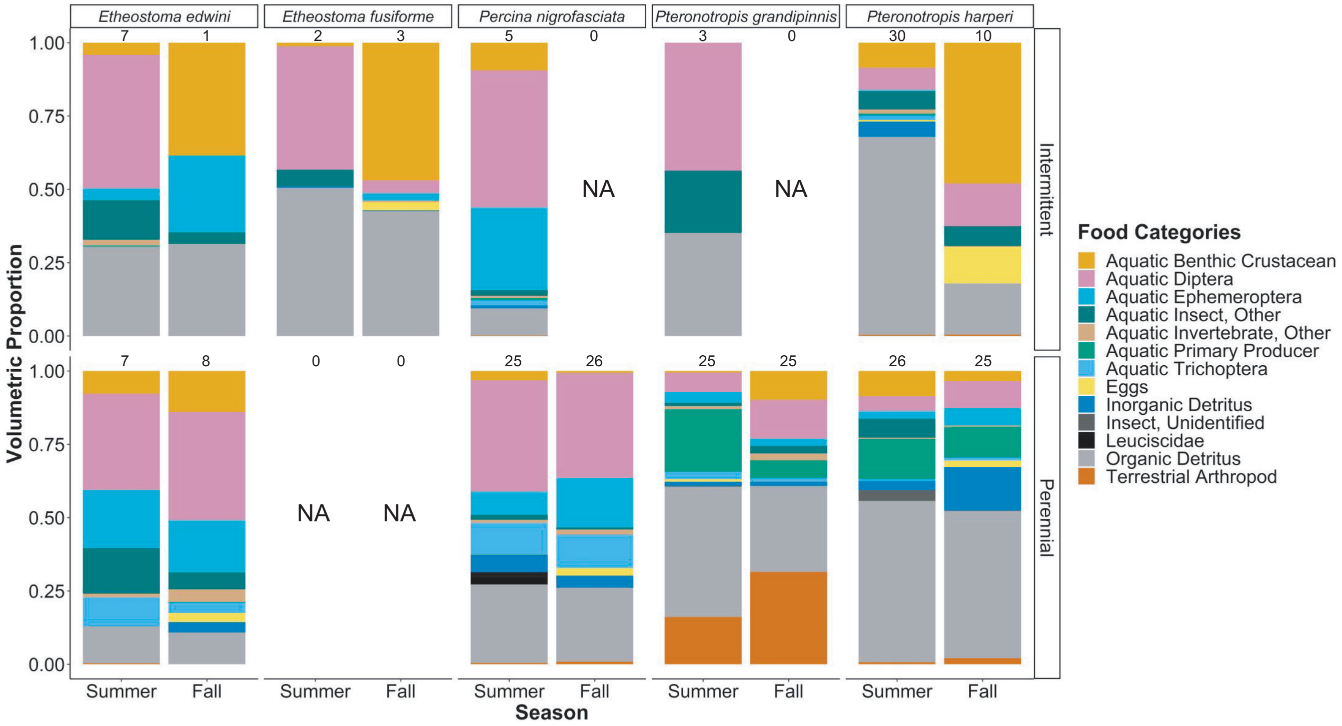

FIGURE 4 Gut content items present in five focal fish taxa collected in perennial and intermittent streams during summer and fall seasons. Sample sizes of fish stomachs analysed per treatment are indicated above each bar. Vacant columns with "NA" indicate where fish taxa were not collected in particular stream-flow type and season combinations. Food categories are represented at a broad resolution to effectively display significant diet shifts. Finer resolution of food categories identified to the family and order taxonomic level can be found in the Appendix S1: Tables S2–S6 and Figure S1 respectively

agricultural development. We acknowledge that our study is limited by small sample sizes due to low collections of fish specimens in intermittent streams. However, our confidence in the interpretation of our findings is upheld because our results mirror local patterns regarding macroinvertebrate (Smith et al., 2017) and fish community responses to reduced stream flows (Davis et al., 2020).

4.1 | Effects of stream hydrology on prey availability for minnows and darters

Our results indicate that food resource availability for secondary consumer fish communities may be more diverse and temporally stable in perennial streams. This likely explains why distinct diets were observed between minnows and darters in perennial streams, but not in intermittent streams. Similarly, Atlantic salmon (*Salmo salar*) and Brook charr (*Salvenius fontanalis*) in Newfoundland have been found to converge on food resources during low flows, due to reductions in available habitat and food resources resulting in increased interspecific interactions (e.g. competition and predation) (Brush et al., 2015). Furthermore, food webs in intermittent stream networks are often smaller and more simplified due to the shrinking of aquatic habitats and the loss of aquatic-obligate invertebrate taxa (McHugh et al., 2015; McIntosh et al., 2017). Akin to our study, Mas-Martí et al. (2010) assessed the cumulative effects of stream drying on Mediterranean barbel (*Barbus meridionalis*) and chub (*Squalius laietanus*) trophic ecology by comparing diets in perennial and intermittent reaches during flowing conditions (spring season). Likewise, they found that food resource availability and diet richness were significantly reduced in intermittent streams, due to the absence of rheophilic insect prey that were adversely affected by chronic stream drying.

Aquatic macroinvertebrate recolonisation is often delayed following flow resumption due to some taxa requiring time to aerially disperse from nearby perennial refugia or reliable flows to carry out their reproductive cycles (Bogan et al., 2015; Stubbington et al., 2017). Macroinvertebrate communities in SE USA streams have been found to require approximately 5–10 months to recover from stream drying and may remain significantly different from perennial communities more than a year after flow resumption, due to the lack of dispersal limited, desiccation intolerant and longer lived taxa (e.g. several Plecoptera, Diptera and Ephemeroptera genera) (Churchel & Batzer, 2006; Smith et al., 2017). In contrast, shorter lived invertebrate taxa are often the first to recolonise intermittent streams upon rewetting (Smith et al., 2017). In fact, due to the presence of early colonising and desiccation-tolerant taxa (including some benthic crustaceans), Smith et al. (2017) demonstrated that aquatic invertebrate assemblages in our study system can be equally abundant and diverse in both perennial and intermittent streams. However, although invertebrate assemblages may be diverse in intermittent streams, these taxa may not all function as prey resources for fish.

Small-bodied stream fishes may be limited in their ability to consume all invertebrate prey types in intermittent streams. For instance, although we collected amphipods in all intermittent sites during this study (pers. Obs.), they were not found in any fish guts. Furthermore, additional indicator invertebrate taxa for intermittent streams in our system (per Smith et al., 2017) were either rare (e.g. leeches, dragonfly larvae, limpets) or absent (e.g. snails, crayfish) in community-level fish diets. This suggests fish either selectively reject prey items that are available in intermittent streams (due to gape limitations or foraging tactics), or that the ability to successfully forage upon such taxa declines in these habitats (Mas-Martí et al., 2010). Therefore, the combined effects of losses in preferred insect prey and foraging limitations likely explains why fish diets at the community-level were significantly less diverse in intermittent streams, and why this trend was especially pronounced following flow resumption in the fall.

Some fish species may be more affected by shifts in food resources prompted by variable stream flows. Water-column foragers, such as *P. grandipinnis*, that consume drifting terrestrial prey may be disproportionately affected by increasing intermittency. Previous research in this system found that during periods of intermittence, *P. grandipinnis* trapped in isolated pools contained almost no terrestrial prey and 29% of individuals had consumed fewer than five prey items (Davis, 2017). Furthermore, we were only able to collect three *P. grandipinnis* specimens from a single, flowing intermittent stream in this study, and none of these individuals had terrestrially derived prey in their guts. We conclude that periodic elimination of drifting prey is a major consequence of intermittency for some fishes.

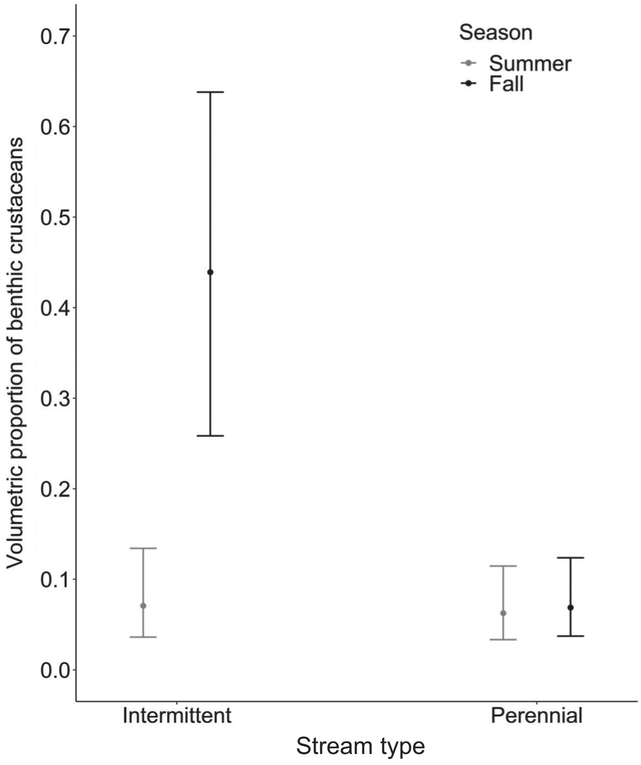

FIGURE 5 Beta-regression predicted proportions of benthic crustacean consumption in fishes as a function of stream-flow type and season. Variation is represented as prediction intervals. Prediction intervals are 95% confidence intervals that include the residual variance terms associated with random effects (site and fish species)

In contrast, aquatic invertivore fishes may be better suited to adapt to shifts in available food resources in intermittent streams (Davis et al., 2020). We saw some evidence of greater taxonomic richness for *P. nigrofasciata* diets in intermittent reaches compared to perennial reaches. This difference appeared to be a result of smaller bodied juveniles consuming larval beetles, benthic crustaceans, terrestrial insects and several worm taxa in intermittent streams in the summer (Appendix S1: Table S4). These results indicate that juvenile *P. nigrofasciata* may be able to exploit available prey in intermittent streams due to their adaptive foraging across a broad range of stream habitats. Similarly, *P. harperi* appears to use diverse foraging strategies since drifting/pelagic prey (e.g. terrestrial insects, copepods) and benthic prey (e.g. freshwater sponges, midges) were both components of their diets. Hence, fish taxa that exhibit trophic flexibility and greater diet richness may be better suited to tolerate habitat degradation (Lisi et al., 2018), and can serve as effective indicators of food resource availability under varying hydrological conditions in streams.

4.2 | Effects of stream hydrology on prey availability for minnows and darters

Fishes in systems with high flow variation have been found to consume low-quality plant and detrital material to take advantage of available food resources (Pusey et al., 2010) and to persist in harsh abiotic conditions (Alexandre et al., 2015; Balcombe et al., 2005) when preferred animal prey may be limiting (Persson, 1983). Aquatic and terrestrial insect prey can be scarce in intermittent streams even months after flow resumption, due to the delayed recolonisation of sensitive macroinvertebrates after drought (Mas-Martí et al., 2010) or reductions in downstream transport of prey due to low flows. Our data shared similarities with these observations, as diet indicator items for minnows included organic detritus and wood in intermittent streams (especially for *P. harperi* in the summer), and terrestrial and aquatic rheophilic insect prey were more strongly associated with minnows and darters in perennial streams respectively.

Similar trophic patterns have been observed in fishes of the Mediterranean climate region; a region with similar intermittent hydrologic regimes. For instance, Alexandre et al. (2015) found that the diets of riverine Iberian barbel (*Luciobarbus comizo*) were largely composed of plant and detrital matter in intermittent streams, especially when flows were minimal in the summer. They also found fishes in perennial systems had greater diet richness, largely comprising aquatic insects, especially during peak flows in the winter. A difference observed in our study is that diatoms were found to be an indicator diet item for minnows in perennial streams. Since our target minnow species are not known to be algivorous, we expect that benthic diatoms were incidentally consumed alongside aquatic arthropod prey in larger open-canopy reaches where epilithon densities have been shown to be greater compared to smaller streams (Weigel et al., 2020).

Another study in the Iberian Peninsula documented that proportions of macroinvertebrates and terrestrial prey in fish diets were significantly greater, and macroinvertebrate taxa present were larger bodied, in perennial systems (Mas-Martí et al., 2010). Freshwater fishes have been found to prey on larger individuals when prey are abundant but become less selective as quantities of prey diminish (Werner & Hall, 1974). This may explain why benthic crustaceans such as copepods are an important prey resource for fish in intermittent streams, as rheophilic insect taxa (such as Ephemeroptera and Trichoptera) may become less available with drying (Stubbington et al., 2017). Additionally, larger predatory invertebrate taxa can become concentrated in intermittent streams during flow recession due to contracting habitats resulting in increased predation on primary consumer invertebrates (Stubbington et al., 2017). Following rewetting however, predatory taxa can be slow to recolonise, due to challenges of dispersal, reduced prey and the lack of desiccation-resistant adaptations (Bogan et al., 2013; Stubbington et al., 2017). This may explain why in our study, the larvae of Gyrinidae, Dytiscidae and Tanypodinae (all predatory taxa) were only found in darter guts in intermittent streams in the summer, but not in the fall (following rewetting).

Collectively, these patterns suggest that in streams where flows are dynamic and often unpredictable, omnivory, generalist (Pusey et al., 2010) and/or risk-prone (Pyke, 1984) foraging may be favoured for survival on available food resources. Nonetheless, switches to detritivorous/herbivorous diets have been shown to result in significant reductions in the individual growth rates of fish even though the consumption of algae and detritus can ensure energetic maintenance in fish when animal prey is limiting (Persson, 1983). Hence, future scenarios that project more frequent and/or extended drying may cause reductions of nutritious animal prey, ultimately limiting fish growth and risking population extirpation.

4.3 | The importance of noninsect invertebrate prey for fishes in intermittent streams

Our analysis shows evidence that fish diets may differ between perennial and intermittent streams even at the beginning of summer when all streams have been flowing for an extended period. In particular, rheophilic Trichoptera were more common in darter diets from perennial streams. However, overall differences were subtle. In contrast, our results show that benthic crustaceans, including copepods, cladocerans and isopods, are an important prey item for minnows and darters upon flow resumption in intermittent streams in the fall following summer drying. As streams experience increased intermittency, aquatic invertebrate communities often become simplified, nested subsets of species from perennial communities, that are dominated by taxa adapted to stream drying (Stubbington et al., 2017). Benthic crustaceans such as cladocerans and isopods are strong swimmers, strong crawlers and are ovoviviparous (Stubbington et al., 2017). These traits promote crustaceans' ability to relocate into hyporheic refugia during flow cessation and to recolonise streams rapidly once flows have resumed (Stubbington et al., 2017). Meiofauna such as ostracods and copepods are also able to tolerate low oxygen concentrations (Storey & Quinn, 2008) and desiccation via diapause (Stubbington et al., 2017; Thorp & Covich, 2009). Furthermore, research has demonstrated that benthic crustaceans are often the first invertebrate taxa to recolonise streams upon rewetting, when they can capitalise on abundant resources, low competition and low predation (Schneider & Frost, 1996; Smith et al., 2017; Wiggins et al., 1980).

In ICB intermittent streams, Smith et al. (2017) found the richness and abundance of noninsect invertebrates to be significantly greater in intermittent streams compared to perennial reaches. In fact, they found *Gammarus* amphipods, Isopoda, Copepoda and Ostracoda to be indicator taxa for intermittent streams that regularly dry during the summer months. Similarly, we found Copepoda, Isopoda and unidentified microcrustacea were indicator taxa for prey items of fish in intermittent streams, being especially important following rewetting. Research in dryland rivers of Australia found that during seasonal dry-down, benthic crustaceans such as calanoid copepods and cladocerans become increasingly important prey items for carnivorous, omnivorous and detritivorous fishes as insect prey became limiting (Balcombe et al., 2005; Medeiros & Arthington, 2008). Collectively, these findings indicate benthic crustaceans may provide an abundant and reliable food resource for secondary consumer fishes until macroinvertebrate taxa are able to recolonise intermittent streams.

In southwestern Georgia (Ingram, 2013) and many other regions of the world (IPCC, 2007) climate change coupled with water abstraction is predicted to cause more intense and frequent water scarcity, resulting in reduced stream flows and greater frequency and duration of stream intermittency on the landscape (Datry et al., 2014). Increasing water scarcity will continue to reduce the availability of perennial refugia for freshwater fish (Kerezsy et al., 2017); aquatic ectotherms that are threatened by climate warming (especially larger bodied taxa) due to their high sensitivity to water temperature changes and by stream drying resulting in degraded habitat (McIntosh et al., 2017). Moreover, simplified aquatic food webs (Sabo et al., 2010) and lower body condition in fishes (Mas-Martí et al., 2010) have been more correlated with intermittent streams compared to perennial streams. Therefore, increases in the duration and frequency of stream drying may also have consequences for fish fitness and subsequent food web structure, due to increases in lower quality or unsuitable prey and changes in the number and strength of food chain interactions respectively (McMeans et al., 2019).

4.4 | Conclusion

Our research contributes to our knowledge of how fish trophic pathways may change in an uncertain future where perennial streams are becoming more intermittent. Fishes that exhibit flexible foraging strategies under varying seasonal conditions can provide resilience to hydrological perturbations and can enhance food web stability (McMeans et al., 2019). Results from our study system suggest that the ability of minnows and darters to effectively utilise abundant benthic crustacean taxa as prey upon flow resumption may serve to buffer some fishes from the effects of increasing intermittency. However, since benthic crustaceans have been found to be more strongly associated with intermittent systems with longer hydroperiods compared to shorter hydroperiods (Bruno et al., 2001; Smith et al., 2017), the long-term reliability of this prey resource may depend on the degree to which streams experience drying. Hence, understanding how the timing and duration of stream drying influences available prey resources for fishes is important for informing food web structure and function, especially in systems with high fish diversity. Furthermore, ongoing research to promote irrigation efficiency and water conservation in agricultural watersheds has great potential to improve the security of perennial refugia for aquatic biota (Qi et al., 2020), which has also important implications for sustaining fish populations and promoting regional biodiversity resilience during drought (Robson et al., 2013).

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CONFLICT OF INTEREST

The authors have no conflict of interest to declare.

AUTHOR CONTRIBUTION

Christine Fallon was involved in fish sampling and collections, fish gut content analyses, statistical analyses, writing and study design. Krista Capps was involved with the supervision and conceptualisation of the research project and editing. Mary Freeman was involved with the supervision and conceptualisation of the research project, statistical analyses and editing. Stephen Golladay was involved with the supervision and conceptualisation of the research project, funding the research and editing. Chelsea Smith was involved with study design, statistical analyses and editing.

DATA AVAILABILITY STATEMENT

Additional information to support the findings in this study can be found in the Supporting Information files.

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