

# Increasing fish taxonomic and functional richness affects ecosystem properties of small headwater prairie streams

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## SUMMARY

1. Stream fish can regulate their environment through direct and indirect pathways, and the relative influence of communities with different taxonomic and functional richness on ecosystem properties likely depends on habitat structure. Given this complexity, it is not surprising that observational studies of how stream fish communities influence ecosystems have shown mixed results.

2. In this study, we evaluated the effect of an observed gradient of taxonomic (zero, one, two or three species) and functional (zero, one or two groups) richness of fishes on several key ecosystem properties in experimental stream mesocosms. Our study simulated small (less than two metres wide) headwater prairie streams with a succession of three pool-riffle structures (upstream, middle and downstream) per mesocosm. Ecosystem responses included chlorophyll *a* from floating algal mats and benthic algae, benthic organic matter, macroinvertebrates (all as mass per unit area), algal filament length and stream metabolism (photosynthesis and respiration rate). Ecosystem responses were analysed individually using general linear mixed models.

3. Significant treatment (taxonomic and functional richness) by habitat (pools and riffles) interactions were found for all but one ecosystem response variable. After accounting for location (upstream, middle and downstream) effects, the presence of one or two grazers resulted in shorter mean algal filament lengths in pools compared to no-fish controls. These observations suggest grazers can maintain short algal filaments in pools, which may inhibit long filaments from reaching the surface. Accordingly, floating algal mats decreased in mid- and downstream locations in grazer treatment relative to no-fish controls.

4. At the scale of the entire reach, gross primary productivity and respiration were greater in treatments with two grazer species compared to mixed grazer/insectivore or control treatments.

5. The distribution of stream resources across habitat types and locations within a reach can therefore be influenced by the taxonomic and functional composition of fishes in small prairie streams. Thus, disturbances that alter diversity of these systems might have unexpected ecosystem-level consequences.

*Keywords:* floating algal mats, functional groups, habitat complexity, mesocosm experiment, prairie streams

## Introduction

Effects of fishes on stream ecosystems have increasingly been recognised since the classic work by Power, Matthews and colleagues identified the influence of the grazing fish functional group on the structure of stream

periphyton communities (Power & Matthews, 1983; Power, 1984; Power, Stewart & Matthews, 1988). Subsequent studies illustrated the potential for stream macroconsumers (fish and crustaceans) to affect other aspects of ecosystems including changes in benthic invertebrate community structure (Gilinsky, 1984; McIntosh &

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Townsend, 1996) and nutrient cycling (Vanni, 2002; McIntyre *et al.*, 2008; Schmitz, 2008; Flecker *et al.*, 2010). Nevertheless, our understanding of the location and timing of fish effects on ecosystem properties is still incomplete (Vanni, 2010).

Headwater streams make up over 70% of total stream length in catchments (Benda *et al.*, 2005; Lowe & Likens, 2005) and ecosystem processes in these systems affect larger downstream reaches through downstream transport of materials (Vannote *et al.*, 1980), including water, nutrients (Dodds & Oakes, 2008), organic matter, invertebrates and larger debris (Wipfli & Gregovich, 2002; Compton *et al.*, 2003). Thus, headwater systems contribute to the ecological integrity of larger stream networks (Freeman, Pringle & Jackson, 2007). Fish can affect numerous headwater stream properties including structure of periphyton and macroinvertebrate communities and stream function (most often measured as primary production and respiration). For instance, central stoneroller (*Campostoma anomalum*, Cyprinidae) from small headwater streams in Oklahoma influenced ecosystem attributes including algal height, multiple measures of benthic organic matter, bacterial abundance and several measures of primary productivity and nutrient concentrations (Gelwick & Matthews, 1992). These results agreed with earlier studies reporting ecosystem effects of central stoneroller (e.g. Power & Matthews, 1983; Power, 1984; Power *et al.*, 1988). However, differences in functional composition of headwater stream fish communities might further influence the ecosystem response. For example, Flecker (1992) compared small-bodied (2–4 cm) grazing fish (Loricoridae) to small-bodied insectivores (Crenuchidae) and found grazers reduced both the number of invertebrates and sediment dry mass more than insectivores. Freshwater animals are among the most endangered species on Earth (Dudgeon *et al.*, 2006) and understanding how changes in fish population or community structure in headwaters affect ecosystem properties is necessary to anticipate the catchment-scale consequences of human activities that result in community alteration.

Community composition has been proposed as a strong determinant of stream ecosystem structure and function through processes such as complementarity, resource use or trophic cascades (Hargrave, 2009). Presence of some dominant species, such as grazing fishes (Gelwick & Matthews, 1992, 1997; Bertrand & Gido, 2007) and detritivores (Flecker, 1996; Winemiller *et al.*, 2006), has been associated with changes in algal and invertebrate biomass, but those effects might be mediated by the presence of other species. For example,

Power, Matthews & Stewart (1985) showed grazing minnows reduced algal filament lengths in pools, but when a predatory bass was added, minnow grazing shifted to shallow pool margins and filament lengths increased in deep areas. In addition, Vanni (2002) found that nitrogen and phosphorus excretion rates varied approximately 10-fold across 26 species of fishes in Venezuelan streams. Hargrave (2009) found that fish species richness in stream mesocosms was positively associated with primary production, and that the relationship strengthened over the 42-day duration of the experiment. Combined, these studies suggest various mechanisms by which taxonomic and functional composition of fish communities may influence stream ecosystem properties.

Low taxonomic and, in turn, functional diversity of headwater streams (Schlosser, 1982; Martin *et al.*, 2013) and relatively simple food web dynamics allow for straightforward manipulations of community structure to investigate the influence of species and functional groups on stream ecosystem properties. As processes in headwater streams accumulate downstream (Peterson *et al.*, 2001; Lowe & Likens, 2005), measuring the influence of variable functional and taxonomic diversity on ecosystem process might help in understanding processes at the catchment scale (Freeman *et al.*, 2007). In this study, we evaluate how fish species richness affects properties of prairie stream ecosystems. We examined this question by stocking experimental stream mesocosms that included pool-riffle habitats across a simulated upstream-to-downstream gradient with three fish communities reflecting the nested structure of headwater prairie streams. These communities included three common prairie stream fish species; central stoneroller, southern redbelly dace (*Chrosomus erythrogaster*, Cyprinidae), and creek chub (*Semotilus atromaculatus*, Cyprinidae). Dietary analyses of stonerollers and dace in Kings Creek reveal they are primarily algivorous, with incidental ingestion of animal matter (Bertrand *et al.*, 2009; Martin *et al.*, 2013). Creek chub are opportunistic generalists that consume a diverse array of food items including some plant material, numerous aquatic and terrestrial insects and fishes when the chub are large enough (Goldstein & Simon, 1999; Quist, Bower & Hubery, 2006). Stoneroller are often the only species in small, ephemeral stream reaches, thus comprised the single species community in our study; stoneroller and dace often occur concurrently in small, perennial stream reaches; and all three species exist together in larger, perennial stream reaches (Martin *et al.*, 2013). It is unusual to find stream reaches where only dace or chub occur alone, and long-term monitoring of four sites of

variable size and water permanence indicated no instances where these two species consistently comprise a community (Franssen *et al.*, 2006).

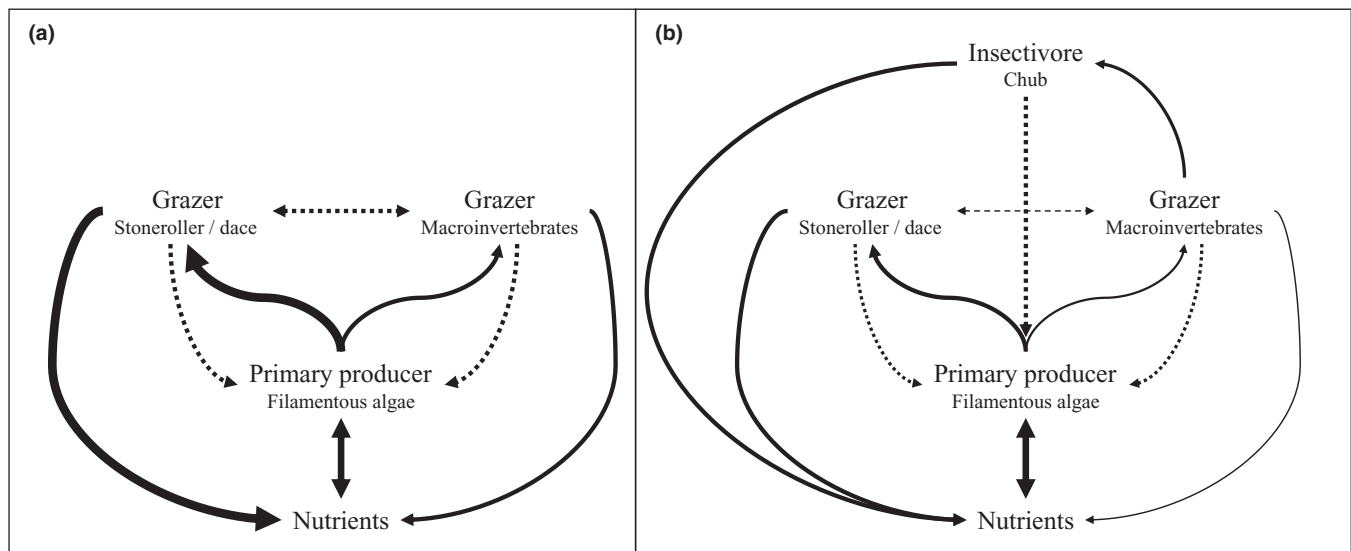
Our main objective was to quantify how natural variation in species richness and composition of small-bodied prairie stream fishes affect whole-stream metabolism and biomass distribution of benthic organic matter, algae and benthic macroinvertebrates in experimental mesocosms. On the basis of previous experiments by our group quantifying trophic interactions and nutrient dynamics in prairie streams (Gido, Dodds & Eberle, 2010; Kohler *et al.*, 2011), we hypothesised that effects of increased fish richness and corresponding compositional changes on measured ecosystem properties would depend on the relative importance of direct versus indirect effects (Fig. 1). In the experiment described below, overall fish biomass and size structure was similar across treatments, thus according to basic metabolic scaling (e.g. Vanni, 2002), we predicted that rates of nutrient remineralisation would be similar across different levels of species richness. Therefore, variation in ecosystem properties among different levels of species richness were predicted to be through interspecific differences in trophic ecology and behaviour. The addition of either algivorous species (i.e. stoneroller or dace) is expected to decrease algal biomass via consumption and

bioturbation, but the magnitude of this effect could be offset by nutrient remineralisation. Grazing fishes may also homogenise resources within pools where they graze, but might increase heterogeneity between habitats by stimulating algal growth in riffles indirectly through excreted nutrients. The addition of the third species (i.e. creek chub, an insectivore) is anticipated to stimulate algal growth directly through nutrient remineralisation of nutrients. Although not explicitly tested in this study, insectivores might indirectly increase primary producers through a trophic cascade (i.e. by consuming grazing insects).

## Methods

### Mesocosm design and treatments

For this study, we used experimental stream mesocosms (as described in Matthews *et al.*, 2006) located on Konza Prairie Biological Station (KPBS) in the summer of 2011. Reaches consisted of a series of three 2.5 m<sup>2</sup> pool habitats connected by 0.8 m<sup>2</sup> riffle habitats (Fig. 2), for a total area of 10.1 m<sup>2</sup> per reach. A nearby low nutrient spring (mean ammonium-nitrogen concentration 35 µg L<sup>-1</sup> and soluble reactive phosphorus 10 µg L<sup>-1</sup>; Konza Prairie LTER unpubl. data) is used to fill streams and provide a



**Fig. 1** Conceptual diagram illustrating potential direct and indirect effects of headwater prairie stream fishes in experimental stream mesocosms. Solid lines represent direct effects (e.g. consumption, bioturbation, excretion) and dashed lines indicate potential competition. Line thickness indicates strength of a relationship. Panel a illustrates the potential effects of grazers, where the addition of either grazing algivorous species (i.e. stoneroller or dace) is expected to decrease algal growth via consumption and bioturbation. The addition of the third species (b; creek chub, an insectivore) is expected to stimulate algal growth through both a trophic cascade (i.e. consuming grazing insects) and nutrient remineralisation (relative to consumption/remineralisation of the two grazing species), thus diminishing the influence of algivorous fishes on both abundance and distribution of resources.

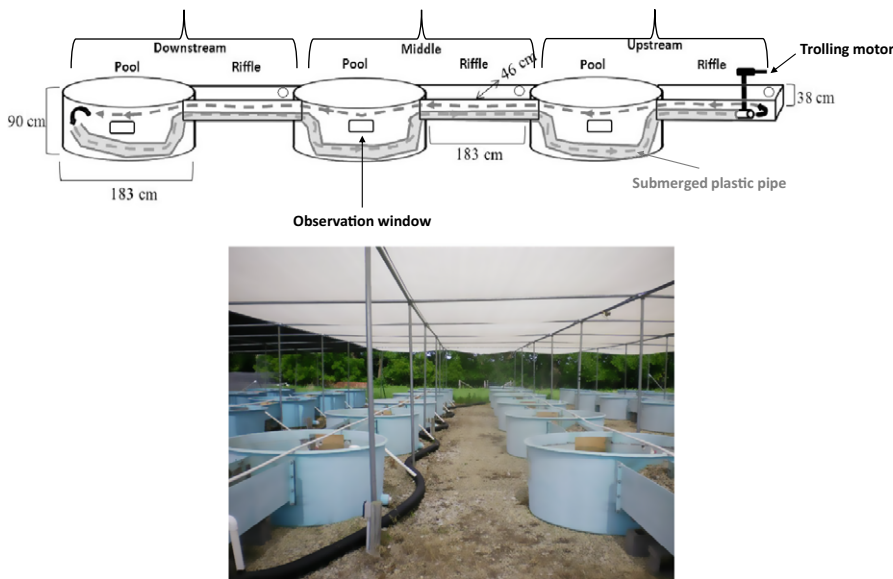


Fig. 2 Diagram (top, one reach) and photograph (bottom) of experimental stream mesocosms on the Konza Prairie Biological Station. Mesocosms are located outdoors under a canopy that provides 60% shading. Flow (indicated by dashed lines, direction follows arrows) is generated from a trolling motor in the upstream riffle that pulls water from the downstream pool through a 15 cm diameter plastic pipe buried under the substrate. Fresh spring water continuously flows into the stream from a pipe running across the top of all streams (not pictured) and water overflows through holes in each riffle. Observations windows are located on each pool.

continuous source of water to offset evaporative loss. Microbial communities colonise the experimental streams primarily by residual spores from previous experiments or those carried by wind. Macroinvertebrates colonise the experimental stream from a nearby (<300 m) stream when flying adults deposit eggs into the system. Prior to running the experiment, mesocosms were power washed, drained and refilled to remove the majority of organic matter and homogenise conditions among reaches (Bertrand *et al.*, 2009). Flow was generated by pulling water through a 15.2 cm diameter plastic pipe with a trolling motor with an average discharge of  $\sim 10 \text{ L s}^{-1}$  (Bertrand & Gido, 2006). Thus, a reach consisted of three locations (i.e. up-, middle- and downstream), each containing a pool and a riffle habitat (Fig. 2). Velocity in pools was  $<0.1 \text{ m s}^{-1}$  and in riffles  $\sim 0.2 \text{ m s}^{-1}$ , with little variation among up-, middle- and downstream locations (Martin *et al.*, 2013). We defined a reach as the series of three connected pool and riffle habitats. Three reaches were randomly assigned in a balanced arrangement to one of four experimental treatments consisting of a control (i.e. no fish) and three fish communities, namely stoneroller (single species), stoneroller with dace (two species), and stoneroller, dace and creek chub (three species).

Fish were caught by backpack electrofishing in Kings Creek, on the KPBS. Single species treatments and combined species treatments were stocked at a constant density of 90 individuals per reach ( $26.6 \text{ fish m}^{-2}$ ; approximately  $15 \text{ g m}^{-2}$ ). The rationale for a constant stocking density across richness treatments was based

on our long-term observations from Kings Creek (1995–2012) which showed that species richness at headwater sites was relatively stable (i.e. 2–4 species) and not associated with abundance, which varied over orders of magnitude. We selected fish of similar sizes (60 to 70 mm total length) for all species. Based on length-weight relationships, stoneroller weight ranged from 2.38 to 3.71 g, southern redbelly dace weight ranged from 2.12 to 3.38 g and creek chub weight ranged from 2.26 to 3.60 g. These fish were likely age-1 (spawned the previous year) and mature adults (Schmulbach, 1957; Schemske, 1974; Settles & Hoyt, 1978).

#### Ecosystem response variables

Measured response variables included algal filament length (cm), benthic chlorophyll *a* ( $\mu\text{g cm}^{-2}$ ), floating chlorophyll *a* ( $\text{mg cm}^{-2}$ ), macroinvertebrate community structure and biomass ( $\text{g m}^{-2}$ ), benthic organic matter ( $\text{mg cm}^{-2}$ ) and whole-stream metabolism [gross primary productivity ( $\text{g O}_2 \text{ m}^{-2} \text{ day}^{-1}$ , GPP), net ecosystem production ( $\text{g O}_2 \text{ m}^{-2} \text{ day}^{-1}$ , NEP) and ecosystem respiration ( $\text{g O}_2 \text{ m}^{-2} \text{ day}^{-1}$ , ER)] based on diel changes in dissolved oxygen concentrations. These responses were measured on 30 July 2011, four weeks after initiation of the study.

Mesh baskets (6 cm deep,  $10 \times 10 \text{ cm}$  surface area) were filled with pebble substrate from the experimental streams and three baskets were placed in every pool and riffle habitat. One basket was removed from each habitat and one pebble from each basket collected for chlorophyll analysis. Benthic chlorophyll *a* was extracted



by submerging pebbles in a 95% ethanol solution that was heated for 5 min at 78 °C and the extract analysed using a spectrophotometer after 24 h. Concentrations of chlorophyll *a* were corrected for cross-sectional area of pebbles (see Sartory & Grobbelaar, 1984; Bertrand & Gido, 2007 for detailed methods).

Floating algal mats were quantified by first photographing the water surface of each habitat in a reach and calculating the proportion of the water surface covered with floating algae. A subsample from each floating algal mat was taken using a square mesh basket (10 × 10 cm) and chlorophyll *a* was extracted as described above. Three subsamples (one in each pool) were taken in each reach and the amount of chlorophyll *a* was averaged and multiplied by the estimated surface area of floating algal mats in the reach (mean chlorophyll *a* per m<sup>2</sup> × surface area covered by mat = total biomass) to estimate total biomass of floating chlorophyll *a*.

Algal filament length was sampled at three points along three different transects in each riffle ( $n = 9$  per reach) and at six points along six different transects in each pool ( $n = 36$  per reach) to quantify structural properties of periphyton. Filament length was defined as the length of the longest filament attached to a pebble that occurred on each transect point.

Macroinvertebrates and benthic organic matter (BOM) samples were collected from each of the three pebble baskets in each habitat that were used for chlorophyll *a*. Pebbles were emptied into 8 L of water in a 20 L bucket. The substrate was vigorously stirred and a 500 mL subsample of the slurry was collected for fine and coarse BOM. The remaining slurry was elutriated to separate inorganic substrate from organic matter and poured through a 250 µm mesh sieve to capture macroinvertebrates. Samples were preserved in 10% formalin, returned to the laboratory, and macroinvertebrates were counted and identified to order or family. Lengths of macroinvertebrates were measured for all individuals to calculate biomass using standard length-mass relationships (Benke, 1984). The 500 mL benthic organic matter samples were preserved using 10% formalin and were filtered (GF/F 0.7 µm microfiber filter), dried and ashed to obtain the amount of benthic organic matter (Wallace, Hutchens & Grubaugh, 2007). The ash-free dry mass (AFDM, g m<sup>-2</sup>) was standardised by surface area of the basket.

Whole stream metabolism (GPP, NEP and ER) was measured at the reach level based on fluctuations in dissolved oxygen content (g O<sub>2</sub> m<sup>-2</sup> day<sup>-1</sup>) of the water measured every 10 min over a 24 h period using YSI

ProODO optic dissolved oxygen sensor. One sensor was deployed in the downstream pool of each reach on the final 24 h of the experiment. These rates were corrected for variation in temperature, dissolved oxygen saturation, light, atmospheric pressure and stream mesocosm morphology based on the single-station modelling technique outlined in Riley & Dodds (2013) and Dodds *et al.* (2013). In short, this method uses a standard equation (Marzolf, Mulholland & Steinman, 1994) to predict dissolved oxygen concentration. Modelled dissolved oxygen is compared to observed dissolved oxygen using the Solver function in Microsoft Excel (version 2007; Microsoft Corporation, Redmond), which uses a Newton search method to minimise the sum of squares of error between modelled and observed values by changing the basic rates of GPP, ER and gas transfer coefficient (*k*). Full model results are presented in Appendix S1.

#### Data analysis

Each response variable was fitted with a general linear mixed model. For all responses, the statistical model included the fixed effects of treatment (i.e. 0, 1, 2, 3 species). For responses other than whole-stream metabolism, the model also included habitat (i.e. pool versus riffle), location (i.e. upstream, middle or downstream) and all two- and three-way interactions, as well as a random effect for reach nested within treatment to recognise the experimental unit for treatment. Degrees of freedom were estimated using Kenward–Roger's approach, with manual fine-tuning whenever needed. Heterogeneous residual variances were fitted to meet model assumptions, as was further supported by enhanced model fit assessed with the Bayesian Information Criterion. All variance components were estimated using residual maximum likelihood. Model assumptions were evaluated using externally studentised residuals and were considered to be appropriately met. Observations were screened for outliers based on a Bonferroni-corrected test on externally studentised residuals.

All statistical models were fitted using the GLIMMIX procedure of SAS (2000 Version 9.4, SAS Institute, Cary) implemented using Newton–Raphson with ridging as the optimisation technique. Estimated least square means and corresponding standard errors are presented. Relevant pairwise comparisons, focusing on treatment effects and their interactions, were conducted using a Tukey–Kramer or Bonferroni adjustment, as appropriate in each case, to avoid inflation of Type-I error rate due to multiple comparisons. Tailored contrasts were built to address comparisons between functional treatment

groups (e.g. the two grazer treatments versus grazers plus an insectivore and versus control treatments) at the proper level of inference.

## Results

### Algal filament lengths

After accounting for differences between up-, mid- or down-stream locations, effect of treatments on algal filament lengths differed by habitat (treatment  $\times$  habitat  $P = 0.027$ ) (Fig. 3). That is regardless of location, algal filaments were significantly shorter in pools subjected to grazer communities (single and two species combination) than in control pools ( $P_{\text{adj}} = 0.018$ ). Filament lengths in pools subjected to three species were intermediate to pools of grazer and control treatments, but not significantly different from either. Algal filaments were also shorter for pools relative to riffles in single species, three species and control treatments ( $P_{\text{adj}} < 0.020$ ), but this habitat difference was not significant in two species treatments. Across locations and treatments, estimated algal filament lengths were more than two orders of magnitude more variable in riffles than in pools.

### Benthic chlorophyll

Regardless of location, the effects of treatments on benthic chlorophyll differed by habitat (treatment  $\times$  habitat  $P = 0.052$ ) (Fig. 4). Across up-, mid- or down-stream loca-

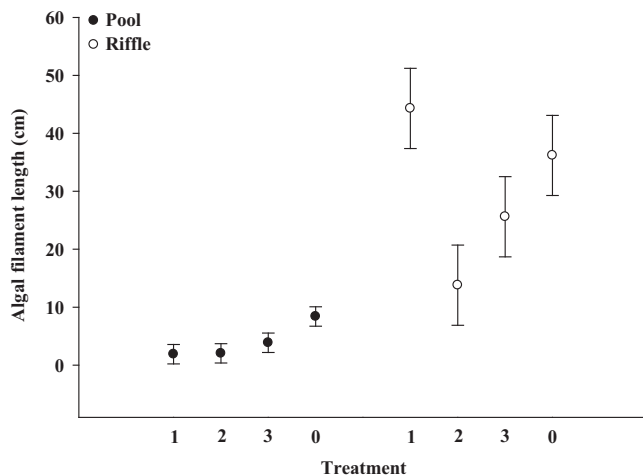


Fig. 3 Least square mean estimates and standard error (SE) for algal filament lengths in pool and riffle habitats of experimental stream mesocosms averaged across up-, mid- and down-stream locations. Treatments 1 – one species, 2 – two species, 3 – three species and 0 – no fish control.

tions, benthic chlorophyll in pools of three species treatments was significantly greater than in control pools ( $P_{\text{adj}} = 0.016$ ). Pools of grazing fish treatments showed intermediate amounts of benthic chlorophyll that was not significantly different from either control or 3-species pools. For all locations, riffles of single species reaches had higher benthic chlorophyll than control riffles ( $P_{\text{adj}} = 0.024$ ). No other evidence for treatment effects was apparent in riffles.

### Floating chlorophyll

The presence of floating algal mats was highly variable across habitats, locations and treatments, with a significant location-by-habitat interaction ( $P = 0.002$ ). In middle and downstream locations, floating chlorophyll biomass was significantly lower for grazer treatments than for controls ( $P_{\text{adj}} < 0.026$ ), whereas the three species treatment was intermediate and not significantly different from the other treatments (Fig. 5). In turn, there was no evidence of any treatment differences in floating chlorophyll in upstream locations. Across locations and habitats, variability in floating chlorophyll biomass was four times greater in control and three species treatments than in grazer treatments.

### Benthic organic matter

Treatments, locations and habitats interactively affected BOM ( $P = 0.045$ ) (Fig. 6). In upstream pools and riffles,

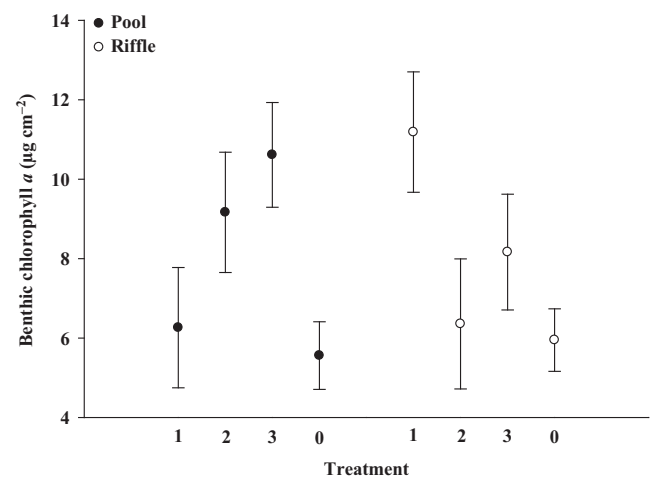


Fig. 4 Least square mean estimates and standard error (SE) for benthic chlorophyll *a* measured in experimental stream mesocosms that included pool and riffle habitats averaged across up-, mid- and down-stream locations. For treatment abbreviations see Fig. 3.

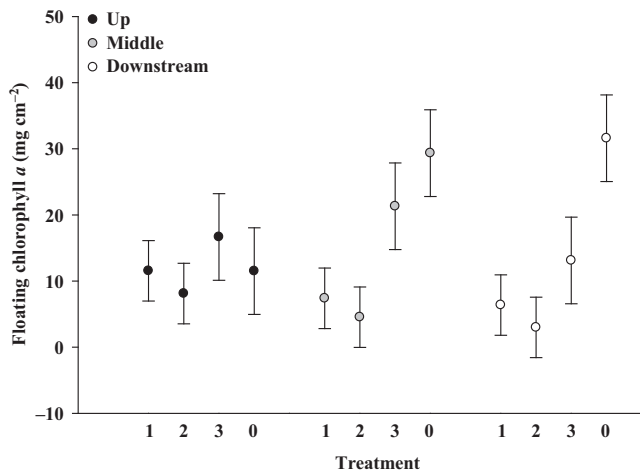


Fig. 5 Least square mean estimates and standard error (SE) for chlorophyll *a* measured from floating algal mats in experimental stream mesocosms that included upstream, middle and downstream locations averaged across riffle and pool habitats. For treatment abbreviations see Fig. 3.

grazer treatments had lower BOM than control treatments ( $P_{\text{adj}} < 0.01$ ). In downstream riffles, BOM of reaches with three species was greater than that of grazers or controls ( $P_{\text{adj}} < 0.02$ ).

#### Macroinvertebrate biomass

Across up-, mid- and down-stream locations, the effects of treatments on macroinvertebrate biomass differed by habitat (treatment  $\times$  habitat  $P = 0.023$ ) (Fig. 7). Regardless of location, macroinvertebrate biomass in riffles of two species reaches was greater than in control riffles ( $P_{\text{adj}} = 0.022$ ). Additionally, macroinvertebrate biomass in riffles for two-species reaches was higher than for the corresponding pools ( $P_{\text{adj}} = 0.001$ ). With the exception of pool habitats in the two-species treatment, macroinvertebrate biomass was generally more variable in fish treatments than in control treatments.

#### Whole stream metabolism

We found evidence for significant treatment differences in GPP ( $P = 0.01$ ) and ER ( $P < 0.01$ ), but not for NEP. Both GPP (oxygen production) and ER (oxygen consumption) increased in the two-species treatment relative to control and three-species treatments (Fig. 8a and b respectively). The single species treatment was not significantly different from the rest of the treatments for any of the whole-stream metabolism responses. Large uncertainty in the single species treatment was partially due to extreme values of whole-stream metabolism

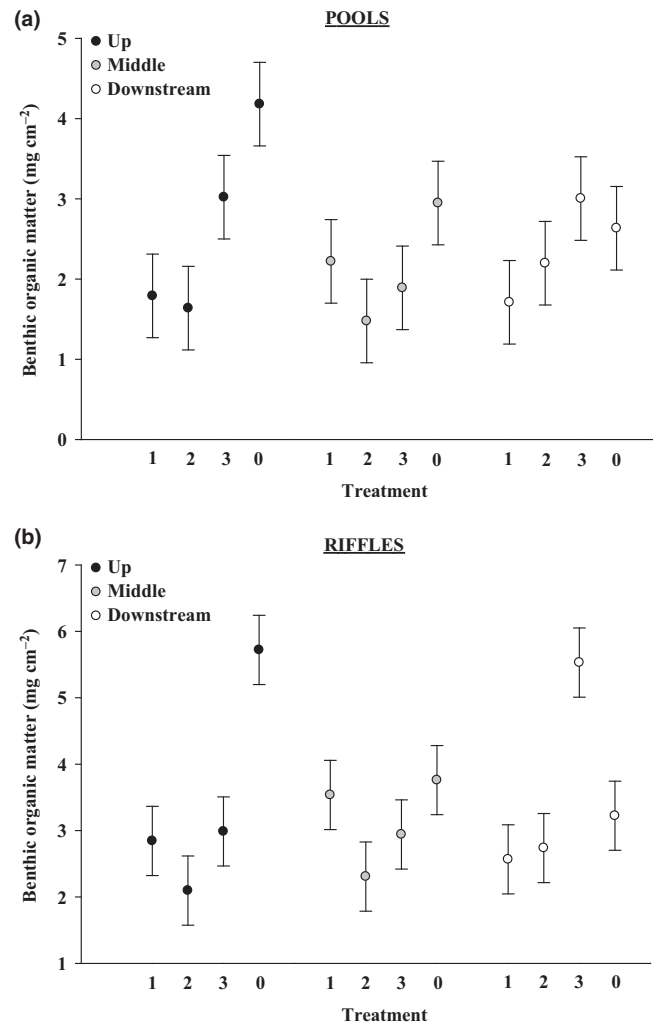


Fig. 6 Least square mean estimates and standard error (SE) for benthic organic matter from experimental stream mesocosms that included pool (a) and riffle (b) habitats as well as upstream, middle and downstream locations. For treatment abbreviations see Fig. 3.

observed on one of the stream reaches. These extreme observations did not show any evidence for technical faults neither were they extreme enough (based on a Bonferroni-adjusted test on externally studentised residual) to grant legitimate exclusion from analyses. As a result, these extreme observations were accounted for by fitting heterogeneous residual variances, which depict the increased estimation uncertainty in the single-species treatment.

#### Discussion

Our study provides evidence on how fish community structural and functional composition of small prairie streams might influence the distribution of primary pro-

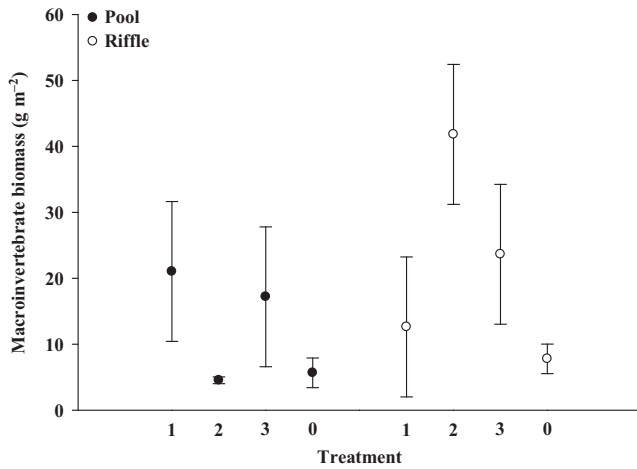


Fig. 7 Least square mean estimates and standard error (SE) for benthic macroinvertebrates collected from experimental stream mesocosms that included pool and riffle habitats averaged across up-, mid- and down-stream locations. For treatment abbreviations see Fig. 3.

ducers, organic matter and macroinvertebrates across habitats. A surprising result of the experiment was the apparent ability of grazing fish to maintain short algal filaments, which in turn can inhibit long filaments from reaching the surface and forming floating algal mats, at least in pool habitats. In our experiment, after controlling for location, pools of grazer treatments (1 or 2 species) had shorter algal filaments than those of controls. This pattern coincided with lower biomass of floating algae in middle and downstream pools of grazer treatments relative to controls. Although grazers were present in three-species treatments, because we maintained a constant overall fish biomass, they were at lower densities, which might have minimised their influence. Although a reduction in algal filament lengths by grazing fishes is commonly observed in other studies (Power *et al.*, 1985; Gelwick & Matthews, 1992; Bertrand & Gido, 2007), to our knowledge the effects of grazing minnows on the development of floating algal mats has not been quantified. Gelwick & Matthews (1997), however, did observe in both field and mesocosms experiments that larger mats of floating algae were exported in treatments with stonerollers than in no fish controls.

A shift in the distribution of algal biomass from the benthos to floating mats can have important effects on ecosystem properties, such as shading benthic biota (Shigesada & Okubo, 1981), providing refugia from predators for macroinvertebrates or small fishes (Power *et al.*, 2008), aiding in the dispersal and emergence of macroinvertebrates (Highsmith, 1985; Power, 1990), and reducing the edibility of algal resources (Chick, Geddes

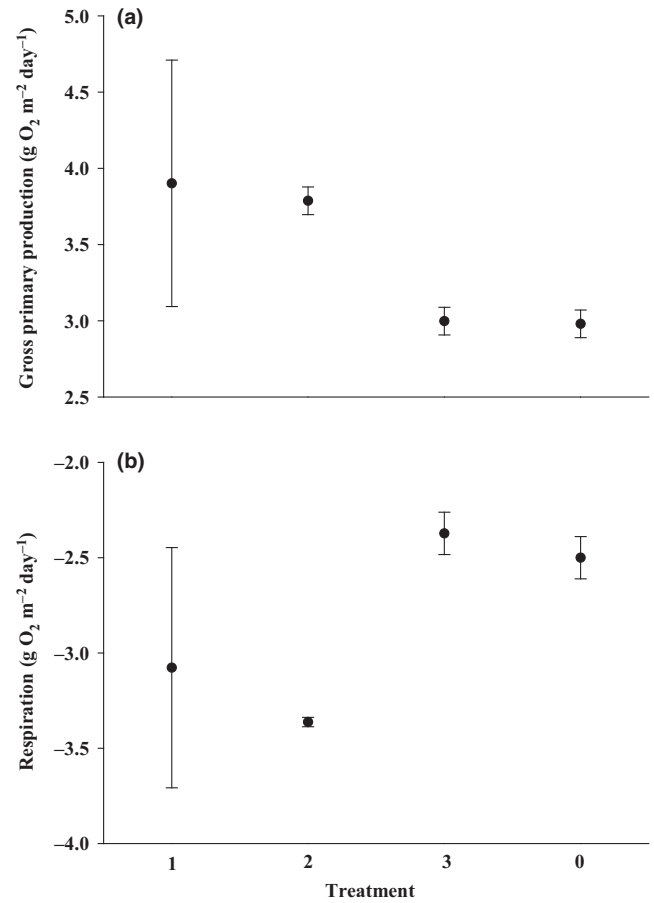


Fig. 8 Least square mean estimates and standard error (SE) for gross primary production (a; GPP) and ecosystem respiration (b; ER) measured in experimental stream mesocosms. For treatment abbreviations see Fig. 3.

& Trexler, 2008). Power (1990) showed that macroinvertebrate community structure was different in floating mats compared to benthic turf mats and that production and emergence of macroinvertebrates was two to six times higher in floating mats, largely due to reduced fish predation. Mature floating mats also might be less edible due to the calcareous matrix that deters fishes from foraging, as found by Chick *et al.* (2008) in the Florida Everglades. Our streams are fed from a limestone aquifer and we observed potential carbonate deposits on dry tops of the mats, though they were not analysed specifically for carbonate. A lack of grazing on these mats could allow rapid succession of algal populations, thus leading to algae senescence and accumulation of detritus (Lamberti, Ashkenas & Gregory, 1987). Unfortunately, we did not expect the development of algal mats, thus did not quantify macroinvertebrate abundances in those habitats. Therefore, a lack of concordance between treatment effects on benthic macroinvertebrates and floating



algal mats is not surprising as we likely underestimated the total biomass of invertebrates in mesocosms, particularly in those taxa that might have occurred in high abundance within floating algal mats.

There also was evidence that experimental manipulations of fish taxonomic and functional richness altered the distribution of resources between riffles and pools. Indeed, significant treatment  $\times$  habitat interactions were apparent for four of the five ecosystem response variables. For example after accounting for location effects, mean algal filament lengths were higher in riffles than in pools when a single grazer species was present. Macroinvertebrate density also was greater in riffles than pools with two grazers present. These results are consistent with the hypothesis that in addition to lower grazing in riffles, grazing fishes in pools excrete nutrients that flow out of the pools and stimulate algal growth in riffles. However, when the presence of two grazers was compared to a single grazer, the difference in algal filament lengths in riffles versus pools seemed to disappear (Fig. 3). Habitat use and grazing behaviour might explain the differences in the structure of algae in riffles and pools when a second grazer (dace) was added. As in other experiments in these mesocosms (Kohler *et al.*, 2011), we observed both species almost exclusively in pools, stoneroller spent the majority of their time in contact with the benthic surface, whereas dace occurred higher in the water column (Martin, 2014). Moreover, Kohler *et al.* (2011) showed that diet of dace in experimental stream mesocosms reflects the riffle algal community composition more than the pool community composition. Thus, it is possible dace more readily feed on riffle algae either with short forays into riffles during the day, foraging in those habitats during night, or eating material that sloughs from the riffles as it enters the pools (Kohler *et al.*, 2011).

Replacing one-third of the herbivorous fish biomass with insectivorous creek chub (three species treatment) increased benthic algal biomass in pools relative to controls and increased BOM in downstream riffles relative to all other treatments. This result may be explained by a decrease in the amount of herbivore grazing pressure while maintaining nutrient remineralisation by all species present. Although we did not measure interspecific differences in excretion rates, because all individuals were similar size and weight, and that all fish assemblage treatments were stocked with equivalent biomass, we assumed community excretion rates were comparable across treatments. Thus, the major variable altered was the trophic level at which members of the community foraged.

The overall response of BOM to our experimental manipulations resulted in a complex three-way interaction between treatment, habitat and location, implying that the differential effects of fish richness treatments on BOM were dependent on the combination of riffle and pool habitats at up-, mid- or downstream locations. In a meta-analysis on the general effects of dace on stream ecosystem properties, Gido *et al.* (2010) found that presence of dace generally reduced fine particulate organic matter (FPOM). If one interprets both BOM and FPOM as representative of the amount of filamentous algae and detritus in the system, our observations are somewhat consistent in that BOM was lower in grazer treatments relative to controls at upstream locations; however, no evidence for this effect was present at middle or downstream locations. A possible explanation for reduced BOM in upstream pools with grazers is that foraging in the benthos likely stirs up sediments into the water column and those sediments are transported downstream. However, if this were the case, we would expect the greatest BOM accumulation in the downstream pools, which we did not observe. Other studies have found that high biomass of grazing fishes can have strong negative effects reducing sediment in streams (e.g. Flecker, 1992) or transport of BOM (Gelwick & Matthews, 1997). Our experimental mesocosms represent an early successional stage following filling, thus there were minimal sediments and presumably a large fraction of BOM was living algae. Thus, manipulation effects on BOM were likely combined effects of bioturbation of sediments, grazing of algae and nutrient dynamics.

Measurements of whole stream metabolism taken in reaches exposed to the different fish richness treatments suggested that different stream fish communities also can influence ecosystem rates. Specifically, GPP and ER increased in the grazer communities relative to the three species community and controls. This coincided with smaller algal filaments in grazer pools and less floating algae for grazer treatments in middle and downstream locations relative to controls. Previous research has suggested that presence or absence of predatory species of fish can influence the amount of carbon dioxide in aquatic systems, typically coinciding with a decrease in prey species and increase in primary producers (Atwood *et al.*, 2013). Although not measured, reduced filament and floating mats likely increased light penetration to benthic surfaces, which might have stimulated primary production. Given that ER mirrored rates of GPP, it appeared that both of these rates were driven by primary producers. Increased production in grazer treatments is in contrast with a previous study that found a

strong effect of a single grazer on algal filament lengths but not on GPP (Bertrand & Gido, 2007); however, floating algal mats did not develop in that study. Algal filaments have been described to induce strong light attenuation (Dodds, Biggs & Lowe, 1999), thus we speculate that grazers might indirectly influence light availability and provide an additional mechanism by which fishes could alter prairie stream ecosystems.

The ability of fishes to influence abundance and structural properties of primary producer communities is likely dependent on abiotic conditions (Gido *et al.*, 2010). In natural prairie streams, development of long algal filaments is often interrupted by flooding that scours stream substrates (Bertrand *et al.*, 2009), and long algal filaments leading to floating mats are more likely to occur during periods of low or no flow (K. Gido, personal observation). Therefore, we hypothesise that floating mats would most likely occur in natural stream locations during these low flow periods when no fish are present or in habitats dominated by insectivorous fishes.

Headwater streams (<3rd order) have relatively low taxonomic diversity (Schlosser, 1982), but the combined influence of these streams affect the entire river network's nutrient cycling and organic matter processing via longitudinal transport (Freeman *et al.*, 2007). Fish densities also can be quite high (>15 individuals per m<sup>2</sup>) in these habitats (Franssen *et al.*, 2006). Measuring the influence that variable functional and taxonomic fish diversity has on ecosystem processes in headwater streams might help to understand processes at the catchment scale, since headwater streams are common and results from more permanent reaches may not apply to intermittent upper reaches. Since streams worldwide are experiencing catastrophic losses of biodiversity due to direct and indirect anthropogenic land- and river-scape alterations (Allan & Flecker, 1993; Master, Flack & Stein, 1997; Dudgeon *et al.*, 2006; Jelks *et al.*, 2008), it is becoming increasingly urgent that we gather as much knowledge on protected systems as possible. Our study illustrates that functional composition of fish communities can alter the spatial distribution of primary producers and potentially consumers associated with those resources.

### Acknowledgments

James Whitney, Matt Troia, Dustin Shaw, Jeff Rogosch, Jason Fischer, Josiah Maine and Josh Perkin assisted in the field and laboratory. Funding was provided by the National Science Foundation through the Konza Prairie Long-Term Ecological Research Program (LTER). This is

publication #16-209-J from the Kansas Agricultural Experiment Station.

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### Supporting Information

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

**Appendix S1** Whole stream, single-station metabolism parameters used to model ER, GPP and NPP across treatment in the 12 replicate stream mesocosm reaches.

(Manuscript accepted 18 February 2016)