

FINAL PERFORMANCE REPORT



Federal Aid Grant No. F16AF01297 (T-92-R-1)

**Assessing the impacts of road culverts on stream fishes
in eastern Oklahoma**

Oklahoma Department of Wildlife Conservation

Grant Period: July 1, 2017 - June 30, 2019

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Project Leader: Thomas M. Neeson

I. Project Objective: The aims of this project were to: 1) Determine the current distribution and population status of fish species of greatest conservation need in the Ozarks and Ouachita Mountain regions of Oklahoma; 2) Determine the impact of stream fragmentation from road culverts on these fish communities; and 3) Determine the location and physical characteristics of road culverts that are impassable to fish species of greatest conservation need.

II. Summary of Progress

Executive Summary

Small rivers and streams in the Ozarks and Ouachita Mountains regions of Oklahoma are highly fragmented by road crossings that disrupt the movements of native fishes. This ecosystem fragmentation is a key stressor of stream fishes and the ecosystem services they support. Conservation practitioners lack data on the location and impacts of these barriers, as well as the distribution and population status of fish species of greatest conservation need. Our aims were to 1) determine the current status of fish communities in small rivers and streams in the Ozarks and Ouachita Mountains regions of Oklahoma, and 2) assess the impact of road culverts on these fish communities. During the summer of 2018, we surveyed 39 free-flowing sites and 29 sites that contained a physical structure that potentially blocked fish movements. At each site, we measured a suite of physical and hydrological attributes of the stream system and sampled the fish community; in sites with a physical structure, we also measured a suite of physical attributes of the structure itself. We observed large differences in species richness, abundance, and community structure when comparing sites with a physical structure to free-flowing sites. We also found that the degree to which a physical barrier impacted fish richness and abundance depended on culvert type (i.e., pipe culvert vs. other culvert designs) and the physical characteristics of that culvert (i.e., outlet drop height, culvert length, and plunge pool depth). We also report fish community composition throughout all 58 sites, including the abundances of six species of greatest conservation need: Wedgespot Shiner, Redspot Chub, Cardinal Shiner, Orangebelly Darter, Plains Minnow, and Black Buffalo. Management recommendations include 1) expanded surveys to create a complete inventory of the location and physical characteristics of stream barriers in the Ozarks and Ouachita Mountains regions, 2) further study that includes direct observation of fish movements to better understand how barrier passability varies

seasonally with stream discharge, and 3) exploration of opportunities for collaboration between fish managers and transportation agencies to jointly address problematic culverts.

A. Introduction

Habitat fragmentation is a leading cause of declining freshwater biodiversity and the ecosystem services it supports (Palmer et al., 2007, Perkin et al., 2015). In river ecosystems, fragmentation occurs when anthropogenic structures such as road-stream crossings, dams, weirs, or other infrastructure block the movements of aquatic organisms, disrupt ecological process, or modify the physical habitat of the stream (Lindenmayer and Fischer, 2006; Gido et al., 2010; Hoagstrom et al., 2011). Fragmentation often results in disconnected habitat patches and isolated subpopulations of organisms, which can affect the persistence of species and the ecosystem services they provide (Perkin and Gido, 2012). Over time, this can lead to increased endemism, extirpation, and loss of genetic diversity (Jager et al., 2001).

In the south-central United States, habitat fragmentation has had dramatic effects on stream fishes. There are an estimated 19,000 anthropogenic barriers in this region, with strong effects on both stream biota and physical habitat (Costigan and Daniels, 2012; Perkin et al., 2015). Many key fish species are pelagic spawners; these species spawn within the water column and require long, free-flowing sections of river to allow the fertilized eggs to develop (Perkin and Gido, 2011; Worthington et al. 2017). In many locations, the effects of habitat fragmentation are exacerbated by drought and declining water availability. Termed an ecological “ratchet effect” (Perkin et al., 2015), physical barriers such as dams and road-stream crossings block fish movements during drought periods, when fish would otherwise attempt to escape the impacts of drought by moving to locations of the stream network with higher flow and lower temperatures. Given that many future scenarios include projected declines in water availability and increased periods of drought (McPherson et al. 2016; Bertrand and McPherson 2018), the interactive effects of habitat fragmentation and declining water availability may grow stronger over the coming decades.

Fish managers focused on small rivers and streams in the Ozarks and Ouachita Mountains regions of Oklahoma have key information gaps concerning habitat fragmentation. Previous research has shown that the ability of aquatic organisms to pass through a road culvert or other physical structure is related to flow depth and velocity within the structure (Warren and Pardew, 1998; Schaefer et al., 2003), the longitudinal length of the culvert (Schaefer et al., 2003; Bouska and Paukert, 2010), and the depth of the plunge pool at the culvert outlet (MacPherson et al., 2012). Many native fishes are weak leapers, and are thus unable to navigate a vertical drop at the downstream outlet of a structure (Mueller et al., 2008; Ficke, Myrick, and Jud, 2011). Despite these previous studies, both stream characteristics and standard engineering practices for the design of road-stream structures often vary regionally (e.g., pipe culverts vs. box culverts; Sleight and Neeson 2018), meaning that fragmentation studies from other regions (e.g., the Great Plains) may not be transferable to small rivers and streams in the Ozarks and Ouachita Mountains regions of Oklahoma.

This project aimed to address the primary conservation issue in two of the highest priority ecosystems in Oklahoma. Small river ecosystems in the Ozarks and Ouachita Mountains regions of Oklahoma are designated “very high conservation priority” by the Oklahoma Comprehensive Wildlife Conservation Strategy (OCWCS; ODWC 2016). For these ecosystems, the highest priority conservation issue is “Incomplete data for species of greatest conservation need and

habitat, an impediment for effective conservation planning and implementation.” Thus, a key objective of this project was to conduct field surveys to update and expand existing data sets on the distribution and population status of fish species of greatest conservation need in these two regions. These updated distribution and population data are essential for prioritizing conservation efforts in small rivers and streams of eastern Oklahoma.

The second aim of our project was to assess the impacts of road culverts on stream fishes in the Ozarks and Ouachita Mountains biogeographic regions. Habitat fragmentation from road-stream crossings is a leading driver of declining stream fish populations and the ecosystem services they support in Oklahoma (Perkin and Gido 2011). There is growing enthusiasm for restoring ecosystem connectivity in the region by installing “fish-friendly” road culverts (Perkin et al. 2015). However, conservation practitioners lack comprehensive data on the location and ecosystem impacts of these barriers, which inhibits their efforts to systematically direct conservation investments to the most cost-effective projects (Januchowski-Hartley et al. 2013). Our assessment of the impacts of road culverts on Oklahoma will provide managers with a partial inventory of the most problematic road culverts in small rivers of the Ozarks and Ouachita Mountains, as well as an enhanced understanding of the characteristics of road culverts that block fish movements.

In this study, we conducted a field survey of fishes in streams and small river ecosystems in the Ozarks and Ouachita Mountains regions of Oklahoma. During the summer of 2018, we surveyed 39 free-flowing sites and 29 sites that contained a physical structure that potentially blocked fish movements. At each site, we measured a suite of physical and hydrological attributes of the stream system and sampled the fish community; in sites with a physical structure, we also measured a suite of physical attributes of the structure itself. We then analyzed the effects of physical structures on fish abundance, species richness, and community structure, and quantified how the magnitude of these effects depended on structure type, condition, size, and other physical characteristics. We also report on fish community composition across the 58 study sites, including the population abundances of species of greatest conservation need.

B. Methods

During the summer of 2018, we conducted field surveys at a total of 68 sites across the Ouachita and Ozark Mountain regions of eastern Oklahoma. We examined 29 sites that contained a physical structure (road culvert, low-water ford, etc.) that might potentially block fish movements (Fig. 1), and 39 free-flowing stream segments that contained no physical structure (Fig. 2). At each location, we recorded physical and hydrological measurements of the stream upstream and downstream of the physical structure, and measured a suite of physical characteristics of the structures themselves. After taking these measurements, we sampled the fish community upstream and downstream of the structures to assess the fish community structure on either side of the barrier.

We measured the physical and hydrological characteristics of the structures using a small barrier assessment data sheet from Bain and Stevenson (1999). Specifically, we measured the outlet drop height (i.e, distance from the bottom of the culvert outlet to the water surface below the culvert), structure length, width, type of structure, structure condition, road condition, average velocity of water going through the culvert (cm/s), pool depth (cm), and culvert height (Bouska and Paukert, 2010). We used a flow meter to determine average velocity of the water flowing through the culverts.

Following Bain and Stevenson (1999), we measured a suite of physical and hydrological variables associated with the stream itself. Using a flow meter, we took a cross section measurement of the stream by taking the bank full width of the stream and dividing the stream width into 20 equal intervals. For each interval we set the depth of our flow meter to be 60% of the stream depth. We then recorded a suite of physical characteristics of the stream environment: water temperature, percentage canopy cover, percentage of certain substrates (bedrock, gravel, mud, sand, cobble), stream width, stream depth at thalweg, flow velocity, and discharge (Bouska and Paukert, 2010; Zbinden and Matthews, 2017). We replicated these measurements for our control (i.e., free-flowing) sites, upstream segments of our fragmented sites, and downstream segments of our fragmented sites.

At each study site, we used seine nets to sample the fish community. Following the approach used in Perkin et al. (2015), a team of two people would seine the available habitat within the stream reach. Fish were collected and stored in a Frabill three-gallon minnow bucket with an aerator attached. Sampling time ranged from approximately 40-100 minutes depending on the size of the reach and how many fish were collected (Zhibden and Matthews, 2017). After seining, we identified individuals to the species level and recorded their length. All fish were released back into the stream segment they were sampled in as quickly as possibly once identified, counted, and measured; as a result, mortality was minimal and typically less than 10 individuals per site. This process was replicated in the upstream, downstream, and control segments.

As a direct test of whether physical structures hindered fish movement, we used Visible Implant Elastomer (VIE) tags to mark fish and observe their location over the course of the summer. For our first five sites with physical structures, we marked all fish captured on both sides of the structure. We injected the elastomer into the dorsal side of the fish near the dorsal fin, and used two different VIE tag colors to differentiate fish captured upstream vs. downstream of the barrier (Bouska and Paukert, 2015). We repeated the sampling and VIE marking three times over the course of the summer, at an approximately monthly interval, using VIE tags to mark both recaptured fish and non-tagged fish that had not been observed during previous sampling.

Data Analysis

Our field survey efforts resulted in 8,370 fish comprising 55 species. To assess whether and how stream barriers affect stream fishes in our study sites, we performed a series of statistical analyses on community structure and barrier attributes. All statistical tests were conducted using R.

To test whether mean species richness and abundance differed among upstream, downstream, and control (i.e., unfragmented) sites, we performed one-way ANOVAs on species richness and abundance between the positions (upstream, downstream, and control). We then ran paired t-tests to determine whether mean abundance and richness differed between upstream and downstream segments. We chose to use a paired t-test because we hypothesized that abundance and richness at paired sites would be subject to the same set of confounding factors.

We then performed several tests to determine how differences in species richness and abundance among fragmented vs. unfragmented sites might be related to physical and hydrological characteristics of the structures and stream sites. First, we hypothesized that differences in species richness and abundance upstream vs. downstream of a structure would be greater in locations where the structure contained a vertical drop that would block fish

movements (Mueller et al., 2008). To test this hypothesis, we first calculated the difference in species richness upstream vs. downstream of each structure. We then performed a paired t-test to compare mean difference in species richness between sites with a vertical outlet drop vs. sites without a vertical outlet drop. To test whether differences in species richness and abundance were related to the type of structure, we first separated our sites with structures into two groups: those with a pipe culvert (n = 11 sites) and those with any other type of structure (n = 18 sites; structures included box culverts, arch culverts, and low water dams). We then conducted separate paired t-tests for each of those groups (sites w/ pipe culverts, and sites without) to determine whether species richness and abundance differed between upstream and downstream stream segments.

We then fit two linear regression models to determine how the difference in species richness between upstream and downstream stream reaches, and the differences in abundance between upstream vs. downstream reaches, might be related to four physical dimensions of the structure: vertical drop height, structure length, plunge pool depth, and structure condition. We included vertical drop height in this model because it is known to hinder fish movement and therefore should drive differences in abundance and richness between upstream and downstream segments (Mueller et al., 2008). We included culvert length in this model because there is evidence to show that the length of the culvert can impede fish movement (Bouska and Paukert, 2010). Plunge pool depth was also included under the hypothesis that deeper plunge pools would have higher species richness and abundance (MacPherson et al., 2012). Lastly, structure condition was included due to a hypothesis that structures in poor condition (i.e., physically deteriorated structures) might hinder fish movement (Sleight and Neeson, 2018).

Finally, we conducted two analyses to determine how stream barriers might affect fish community structure. First, we calculated the Bray-Curtis Dissimilarity index for all possible pairs of upstream sites; all possible pairs of downstream sites; and for all possible pairs of control sites. The Bray-Curtis Dissimilarity Index quantifies how dissimilar a species community structure is between a pair of sites. It returns a value between zero and one; values closest to zero indicate that two sites have similar community structures, while values closer to one indicate that two sites have more dissimilar community structure (Brown and Milner, 2007). In this analysis, our objective was to determine how similar community structure was among all upstream sites; among all downstream sites; and among all control sites.

Our second community structure analysis was to assess community dissimilarity between upstream vs. downstream sites; and between upstream and control, and downstream and control sites. To do this, we first created pairs of fragmented and control sites by identifying pairs of sites that were as similar as possible to each other in terms of physical characteristics of the stream (e.g., flow, depth, and width). We then calculated the BCI between each pair of sites and used an ANOVA to compare mean community dissimilarity among upstream vs. downstream pairs; upstream vs. control pairs; and downstream vs. control pairs. All Bray-Curtis analyses were used with the vegan package in R.

C. Results/Discussion

In our surveys, we recorded 8,370 individual fish from a total of 55 species. We collected 1,570 fish in stream segments above physical structures, 2,731 fish in stream segments below physical structures, and 4,069 fish throughout our control sites. All sites were wadeable streams with water velocities ranging from nearly 0 to 33.13 cm/s.

We found that both mean species richness and mean abundance differed among control sites, upstream of the physical structure at fragmented sites, and downstream of the physical structure at fragmented sites. Specifically, we observed large differences in abundance between upstream segments (mean = 54.14 individuals, $n = 29$ sites), downstream segments (mean = 94.17, $n = 29$), and control segments (mean = 104.33, $n = 39$); these differences were statistically significant (ANOVA; $p < 0.05$; Fig. 3). We also saw differences in mean species richness among upstream segments (mean = 4.79 species), downstream segments (mean = 5.86 species), and control segments (mean = 6.36 species). Our ANOVA approached significance ($p < 0.1$; Fig. 4). We also found statistically significant differences between species richness upstream of physical structures (mean = 4.79 species) vs. downstream of physical structures (mean = 5.86 species; paired t-test; $p < 0.05$); and between abundance upstream of physical structures (mean = 54.14 individuals) vs. downstream of physical structures (mean = 94.17 individuals; paired t-test, $p < 0.05$).

We found that the differences in species richness and abundance between fragmented and unfragmented sites were related to a variety of physical attributes of barriers. Of the 29 sites with potential barriers, 20 sites had no vertical outlet drop and 9 sites had a vertical outlet drop. For sites with no vertical outlet drop, we observed a large difference in mean abundance between upstream segments (mean = 57.55 individuals) vs. downstream segments (mean = 78.7 individuals), but the difference only approached statistical significance (paired t-test; $p < 0.1$; Fig. 5). For sites with a vertical outlet drop, we saw much larger differences in abundance between upstream segments (mean: 46.56) vs. downstream segments (mean: 128.56); however, this difference only approached statistical significance, likely due to low sample size ($n = 9$) of sites with a perched culvert (paired t-test; $p < 0.1$; Fig. 6).

Of the 29 sites with potential barriers, 11 sites had pipe culverts and 18 had other types of barriers (box culvert, low water dam, or arch culvert). At sites with pipe culverts, we saw large differences in mean abundance between upstream segments (mean = 54.14 individuals) vs. downstream segments (mean = 94.17 individuals), but due to low statistical power ($n = 11$ sites) this difference only approached statistical significance (paired t-test, $p < 0.1$; Fig. 7). For other types of structures, we also saw large differences in mean abundance between upstream segments (mean = 64.17 individuals) vs. downstream segments (mean = 102.13 individuals), but a paired t-test only approached significance (paired t-test; $p < 0.1$; Fig. 8). At sites with pipe culverts we found a statistically significant difference in mean species richness between upstream segments (mean = 4.79 species) vs. downstream segments (mean = 5.86 species; paired t-test; $p < 0.05$; Fig. 9). When we repeated this test excluding cyprinids, we did not find a statistically significant difference between species richness at upstream sites (mean = 2.69 species) and downstream species richness (mean = 3.28 species; paired t-test; $p > 0.05$; Fig. 10). Similarly, we did not find a statistically significant difference between abundance at upstream sites (mean = 15.45 individuals) and downstream sites (mean = 20.55 individuals; paired t-test, $p < 0.1$; Fig. 11).

For culverts with a vertical outlet drop, we found that the drop height (i.e., the distance between the water surface and the culvert outlet) was negatively correlated with differences in abundance (Fig. 12A, B) and richness (Fig. 12C, D) between the upstream and downstream sides of the structure (linear regression; $p < 0.05$; Fig. 12). We did not find a statistically significant relationship between the longitudinal length of the culvert and the differences in abundance between the upstream and downstream sides of the culvert ($p > 0.05$; Fig. 13A, B). We did find a statistically significant difference between the longitudinal length of the culvert and the

difference in species richness between the upstream and downstream sides of the culvert, but only when excluding cyprinids from the analysis ($p < 0.05$; Fig. 13D). We did not find any statistically significant relationships between plunge pool depth and the differences in abundance (Fig. 14A, B) or richness (Fig. 14C, D) between the upstream and downstream sides of structures.

The physical condition of the structure also affected species richness and abundance. We found a statistically significant relationship between structure condition and the difference in abundance between the upstream and downstream sides of the structure when excluding cyprinids (linear regression; $p < 0.05$); however, this relationship was not significant when we retained cyprinids in the analysis ($p > 0.05$). We also found a statistically significant relationship between structure condition and the difference in species richness between the upstream and downstream sides of the structure (linear regression; $p < 0.05$); however, this relationship was not statistically significant when we excluded cyprinids from the analysis ($p > 0.05$).

Exploration of the Bray-Curtis Dissimilarity Index (BCI) values revealed differences in fish community structure among upstream, downstream, and free-flowing sites. Comparison of all pairwise combinations of sites upstream of a physical barrier showed that these sites tended to be dissimilar from each other (BCI min = 0.16, $\mu = 0.9$, max = 1). We saw similar results for all pairwise combinations of sites downstream of a physical structure (BCI min = 0.16, $\mu = 0.9$, max = 1) and for all pairwise combinations of control sites (BCI min = 0.11, $\mu = 0.85$, max = 1; Fig. 15). Upstream and downstream sites that were immediately adjacent to each other tended to have the lowest dissimilarity (mean BCI = 0.59); this was significantly lower (ANOVA; $p < 0.05$; Fig. 16) than the dissimilarity between pairs of upstream sites and physically similar control sites (mean BCI = 0.79) and the dissimilarity between pairs of downstream sites and physically similar control sites (mean BCI = 0.78).

Using the USFWS IPaC system prior to our field season, we had determined that three federally threatened or endangered species potentially occur in our study area: Arkansas River Shiner, Leopard Darter, and Neosho Madtom. However, we did not detect these species during our field survey efforts. Our field survey efforts detected six species listed as species of greatest conservation need in Oklahoma. We did not find any Tier I species. Tier II species identified were Wedgespot Shiner (45 individuals), Redspot Chub (2 individuals), Cardinal Shiner (2250 individuals), and Orangebelly Darter (2 individuals). Tier III identified were the Plains Minnow (262 individuals) and Black Buffalo (1 individual).

D. Conclusions/Recommendations

We found that physical structures such as road culverts have large negative impacts on stream fish abundance, richness, and community structure in streams in the Ozarks and Ouachita Mountains regions of Oklahoma. We also found that the magnitude of these effects is related to culvert type (i.e., pipe culvert vs. other culvert designs) and the physical characteristics of that culvert (i.e., outlet drop height, culvert, length, culvert condition and plunge pool depth). Overall, our study adds to the growing body of evidence on the effects of fragmentation on stream fish communities in the south-central United States (Bouska and Paukert, 2010; Perkin and Gido, 2012; Worthington et al., 2017) and other regions around the world (Nislow et al., 2011; Macpherson et al., 2012; Januchowski-Hartley et al., 2014; Maitland and et al., 2016).

This study also provided an update on the population status of stream fishes in the Ozarks and Ouachita Mountains regions of Oklahoma, including species of greatest conservation need.

Three federally endangered species are believed to occur in our study region: Arkansas River Shiner (*Notropis girardi*), Leopard Darter (*Percina pantherina*), and Neosho Madtom (*Noturus placidus*). However, we did not detect any of these three species during our field surveys. Our analysis also provides an update on the population status of species considered as Species of Greatest Conservation Need by the Oklahoma Department of Wildlife Conservation. We did not find any Tier 1 species. However, we did encounter four Tier II species: Wedgespot Shiner (*Notropis greenei*) (1 site; 45 individuals), Redspot Chub (*Nocomis asper*) (2 sites; 2 individuals), Cardinal Shiner (*Luxilus cardinalis*) (35 sites; 2,734 individuals), and Orangebelly Darter (*Etheostoma radiosum*) (1 site; 2 individuals). We also found two Tier III species: Plains Minnow (*Hybognathus placitus*) (13 sites; 262 individuals) and the Black Buffalo (*Ictiobus niger*) (1 site; 1 individual).

Our results are mostly in agreement with other studies on the characteristics of road culverts that influence fish passage. We found significant effects of culvert length on species richness (though not abundance; Fig. 15), which is similar to the results of Bouska and Paukert (2010) on the effects of culvert length on fish passage. Mechanistically, culvert length affects fish passage because longer culverts force fishes to swim against higher water velocities for longer lengths (Toepfer et al., 1999; Adams et al., 2000; Bouska and Paukert, 2010). We saw the biggest differences in species abundance and richness with structures that contained vertical outlet drops; thus, structures with a vertical outlet drop should be prioritized for restoration. Many native fishes are unable to traverse a vertical drop greater than 5cm (Ficke, Myrick, and Jud, 2011); thus, even a modest vertical drop at a culvert outlet would constitute an impassable barrier for many native fishes. Pipe culverts on average had a larger difference in species abundance from the upstream segment vs downstream segment than the other types of culverts we sampled; thus, pipe culverts in particular should be prioritized for replacement.

Management recommendations

Based on our results, we make three management recommendations to guide the restoration of ecosystem connectivity in small streams and rivers in the Ozarks and Ouachita Mountains regions of Oklahoma. The most common management strategy is to replace or retrofit existing culverts with “fish-friendly” designs that maximize fish passage, and to ensure that any new construction follows best engineering practices for aquatic organism passage (Perkin and Gido, 2011; Worthington et al., 2017). Culvert designs that maximize aquatic organism passage include those with natural stream bottoms (Bouska and Paukert, 2010), which also minimize geomorphic changes within the stream itself (Angermeier and Schlosser, 1995; Warren and Pardew, 1998; Bouska and Paukert, 2010). The replacement of problematic structures with fish-friendly designs has been shown to help reestablish or increase dispersal of native fishes (Catalano et al., 2007; Walters et al., 2014), and may also partially mitigate the ecological ratchet effect (Perkin et al., 2014, Perkin et al., 2015) that occurs from the interactive effects of drought and fragmentation. Thus, our three management recommendations are designed to support cost-effective barrier mitigation projects to restore ecosystem connectivity:

1) We recommend comprehensive field surveys of all road-stream crossings in high-priority streams and rivers.

Fish managers and conservation practitioners lack comprehensive data on the location and ecosystem impacts of physical barriers, which inhibits their efforts to systematically direct conservation investments to the most cost-effective barrier mitigation projects (Januchowski-

Hartley et al. 2013). Regional databases of dam and road crossing location, condition and ecosystem impact are patchy and incomplete. In some regions, existing road maps capture less than 15% of the true number of road crossings, largely due to poor data on rural private roads (Brooks et al. 2014). The lack of comprehensive barrier data sets for the region seriously hampers efforts at systematic conservation planning. In river networks, the costs and benefits of a proposed barrier removal project depend on whether neighboring upstream and downstream barriers will also be removed. Accordingly, conservation practitioners prefer to follow a “portfolio approach” of evaluating the return-on-investment of a suite of barrier removal projects (O’Hanley 2011). Although very efficient, portfolio approaches to project selection require information on the location, impact, and removal costs of every barrier in a river network; thus, the lack of a comprehensive barrier data set for the region precludes cost-effective portfolio approaches and forces conservation practitioners to adopt inefficient piecemeal strategies (Neeson et al. 2015).

For streams and rivers of high conservation priority, a field survey of all road-stream crossings in the study region would likely be relatively simple to perform and require only modest resources. For example, Sleight and Neeson (2018) surveyed more than 700 road culverts across Oklahoma during a three month field study using a team of three personnel (one graduate student, and two undergraduate student assistants) and basic surveying equipment (stadia rod, flow meter, etc.). For each culvert, they GPS-ed the culvert location and measured a suite of physical characteristics of the culvert and intersecting stream. Similarly, Januchowski-Hartley et al. (2014) surveyed more than 2,200 road culverts across Michigan and Wisconsin during two summer field seasons using a team of three personnel. Thus, a field survey of all physical barriers in streams in the Ozarks and Ouachita Mountains regions (or other priority regions) would be feasible, relatively inexpensive, and would lay the foundation for strategic planning for cost-effective barrier mitigation projects.

2) We recommend further study of how barrier passability varies seasonally with discharge; how the ecological ratchet effect may operate in these streams; and how future water availability may interact with habitat fragmentation.

Although we detected large differences in species richness and abundance between fragmented and free-flowing stream reaches, we did not directly quantify whether individual fish could pass through any barriers. Regrettably, our recapture rates from fish tagged with VIE markers were too low to allow us to make any inferences about barrier passability based on direct observation of fish movement through barriers. An expanded VIE tagging study would be important for quantifying how fish movement and barrier passability vary seasonally with stream discharge. The passability of barriers is known to vary seasonally with discharge (Januchowski-Hartley et al. 2014), and barriers that appear impassable during low flows (e.g., due to a vertical outlet drop) may be fully passable at higher flows. Conversely, some barriers may be fully passable at low flows, but higher flows within the culvert may present a velocity barrier to fishes.

In many locations, the effects of habitat fragmentation are exacerbated by drought and declining water availability. Termed an ecological “ratchet effect” (Perkin et al., 2015), physical barriers such as dams and road-stream crossings block fish movements during drought periods, when fish would otherwise attempt to escape the impacts of drought by moving to locations of the stream network with higher flow and lower temperatures. Given that many future scenarios include projected declines in water availability and increased periods of drought (McPherson et al. 2016; Bertrand and McPherson 2018), the interactive effects of habitat fragmentation and

declining water availability may grow stronger over the coming decades. We recommend further study of the potential synergistic effects of habitat fragmentation and drought conditions on stream fish communities.

3) We recommend that fish managers and conservation practitioners seek partnerships with infrastructure groups (e.g., state and county departments of transportation) to explore cost-sharing strategies for replacing problematic road culverts.

Road culvert surveys (recommendation #1) and conservation projects to replace or retrofit existing road-stream crossings would be of broad interest to other government agencies concerned with infrastructure management. Aging infrastructure is a critical problem for many regions. In North America, many road crossings were constructed in the middle half of the 20th century and are nearing the end of their expected lifespan (Doyle et al. 2008). As these structures continue to age, up-to-date inventories of their condition will be critical for understanding both routine maintenance costs and the risk of catastrophic failures during extreme flow events. By providing basic information on road crossing condition, barrier surveys would enable cognizant agencies to better map the risks and potential impacts of catastrophic failures of road crossings.

Given that transportation agencies often replace or repair existing road culverts to meet their own management priorities, there could be widespread opportunities to enhance the cost-effectiveness of conservation investments by coordinating with infrastructure renewal efforts. For example, Neeson et al. (2018) tested whether accounting for road repair needs could boost conservation efficiency by piggybacking connectivity restoration projects on infrastructure maintenance. Using optimization models to align fish passage restoration sites with likely road repair priorities, they found potential increases in conservation return-on-investment of up to 25%. Importantly, these gains occur without compromising infrastructure or conservation priorities; simply communicating openly about objectives and candidate sites enabled greater accomplishment at current funding levels.

Sleight and Neeson (2018) applied this rationale to road culverts in Oklahoma to identify barriers that were likely to be a priority for both fish managers and transportation agencies. These high priority structures are road culverts that are both in poor condition (i.e., a priority for transportation agencies) and fully impassable to stream fishes (i.e., a priority for conservation groups). Given that, in our study, the culverts in the poorest condition had the greatest impact on stream fishes, there could be many opportunities for aligning fish passage projects with ongoing infrastructure maintenance.

Overall, this study provided a first assessment of the effects of road culverts on stream fish communities in the Ozarks and Ouachita Mountains of Oklahoma, and an update on the population status of stream fishes of conservation need in this region. These findings can be used to enhance on-the-ground efforts to restore aquatic ecosystem connectivity in the region by retrofitting impassable road culverts. Conservation practitioners could use our results to create a cost-benefit analysis for identifying the road culvert mitigation projects that might reconnect the most habitat for the stream system (O’Hanley and Tomberlin, 2005; Neeson et al., 2018). Going forward, efforts to restore aquatic ecosystem connectivity will need to occur alongside a broader suite of conservation actions: understanding flow variations, small barrier removal, and large-scale coordinated research between conservation agencies, road managers, and NGOs (Worthington et al. 2018).

III. Significant Deviations: None

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Tables

Table 1: Summary counts of species observed during our field study across all sites.

Lepisostidae	Column1	Column2	Total Caught	Total Fish Caught
	Common Name	Scientific Name		
	Spotted Gar	<i>Lepisosteus oculates</i>	11	
Clupeidae				8370
	Gizzard Shad	<i>Dorosoma cepedianum</i>	29	
Cyprinidae				
	Ozark Minnow	<i>Notropis nubilus</i>	113	
	Wedgespot Shiner	<i>Notropis greenei</i>	45	
	Golden Shiner	<i>Notemigonus crysoleucas</i>	8	
	Carp	<i>Cyprinus carpio</i>	3	
	Redspot Chub	<i>Nocomis asper</i>	2	
	Sand Shiner	<i>Notropis stramineus</i>	4	
	Redfin Shiner	<i>Lythrurus umbratilis</i>	9	
	Emerald Shiner	<i>Notropis atherinoides</i>	800	
	Bigeye Shiner	<i>Notropis boops</i>	303	
	Steelcolor Shiner	<i>Cyprinella whipplei</i>	226	
	Red Shiner	<i>Cyprinella lutrensis</i>	116	
	Central Stoneroller	<i>Camptostoma anomalum</i>	496	
	Cardinal Shiner	<i>Luxilus cardinalis</i>	2734	
	Plains Minnow	<i>Hybognathus placitus</i>	262	
	Suckermouth Minnow	<i>Phenacobius mirabilis</i>	61	
	Southern Redbelly Dace	<i>Chrosomus erythrogaster</i>	629	
	Bluntose Minnow	<i>Pimephales notatus</i>	69	
	Largescale Stoneroller	<i>Camptostoma oligolepis</i>	61	
	Bullhead Minnow	<i>Pimephales vigilax</i>	5	
	Creek Chub	<i>Semotilus atromaculatus</i>	52	
Catastomidae				
	Northern Hog Sucker	<i>Hypentelium nigricans</i>	23	
	River Redhorse	<i>Moxostoma carinatum</i>	13	
	Golden Redhorse	<i>Moxostoma erythrurum</i>	1	
	Black Buffalo	<i>Ictiobus niger</i>	1	
Ictaluridae				
	Slender Madtom	<i>Noturus exilis</i>	2	
	Yellow Bullhead	<i>Ameiurus natalis</i>	7	
	Freckled Madtom	<i>Noturus nocturnus</i>	3	
Esocidae				
	Redfin Pickerel	<i>Esox americanus</i>	1	
Fundulidae				
	Blackspot Topminnow	<i>Fundulus olivaceus</i>	1	
	Northern Studfish	<i>Fundulus catenatus</i>	11	
	Blackstripe Topminnow	<i>Fundulus notatus</i>	195	
Atherinopsidae				
	Brook Silverside	<i>Labidesthes sicculus</i>	738	
Poeciliidae				
	Western Mosquitofish	<i>Gambusia affinis</i>	376	
Cottidae				
	Banded Sculpin	<i>Cottus carolinae</i>	17	
Centrarchidae				
	Longear Sunfish	<i>Lepomis megalotis</i>	141	
	Bluegill Sunfish	<i>Lepomis macrochirus</i>	341	
	Redear Sunfish	<i>Lepomis microlophus</i>	72	
	Warmouth	<i>Lepomis gulosus</i>	3	
	Green Sunfish	<i>Lepomis cyanellus</i>	9	
	Rock Bass	<i>Ambloplites rupestris</i>	28	
	Largemouth Bass	<i>Micropterus salmoides</i>	235	
	Smallmouth Bass	<i>Micropterus dolomieu</i>	48	
	Redbreast Sunfish	<i>Lepomis auritus</i>	1	
	White Crappie	<i>Pomoxis annularis</i>	8	
Percidae				
	Channel Darter	<i>Percina copelandi</i>	1	
	Stippled Darter	<i>Etheostoma punctulatum</i>	6	
	Log Perch	<i>Percina caprodes</i>	1	
	Fantail Darter	<i>Etheostoma flabellare</i>	3	
	Banded Darter	<i>Etheostoma zonale</i>	3	
	Redfin Darter	<i>Etheostoma whipplei</i>	2	
	Orangebelly Darter	<i>Etheostoma radiosum</i>	2	
	Greenside Darter	<i>Etheostoma blennioides</i>	1	
	Orangethroat Darter	<i>Etheostoma spectabile</i>	38	

Figures



Figure 1: Example of a multi-culvert road crossing that likely blocks fish movement during most flow conditions. At the flow level in the image, all culverts have a vertical outlet drop and flow depths within the culverts are likely too shallow to allow fish movement.

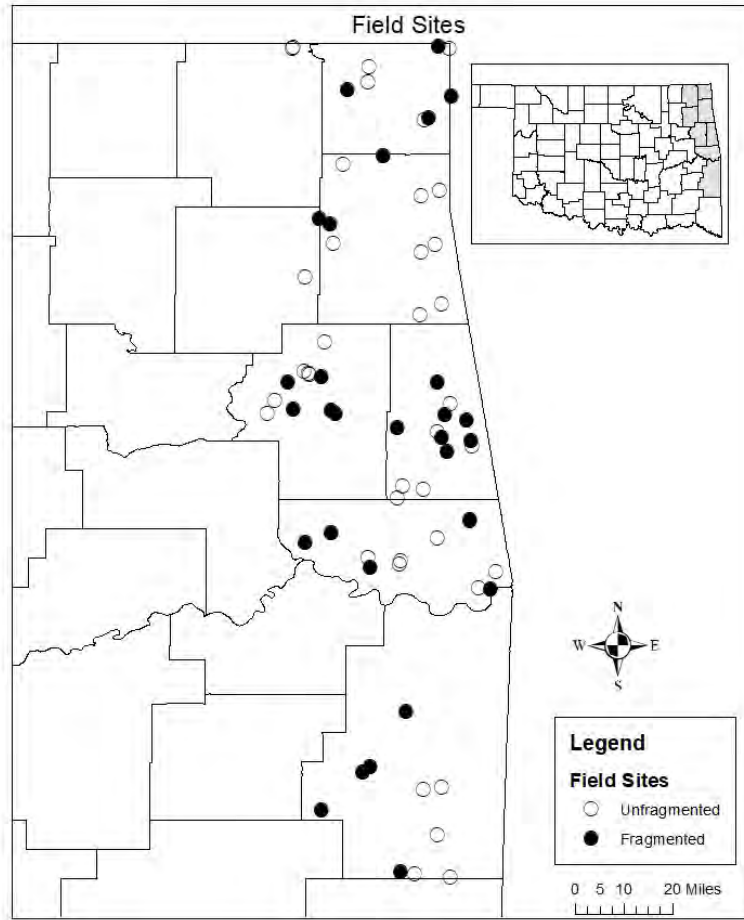


Figure 2: Locations of 58 study sites within eastern Oklahoma. Symbols denote whether the site was unfragmented ($n = 39$; open circles) or contained a physical barrier that potentially blocked fish movements (i.e., fragmented; $n = 29$; closed circles).

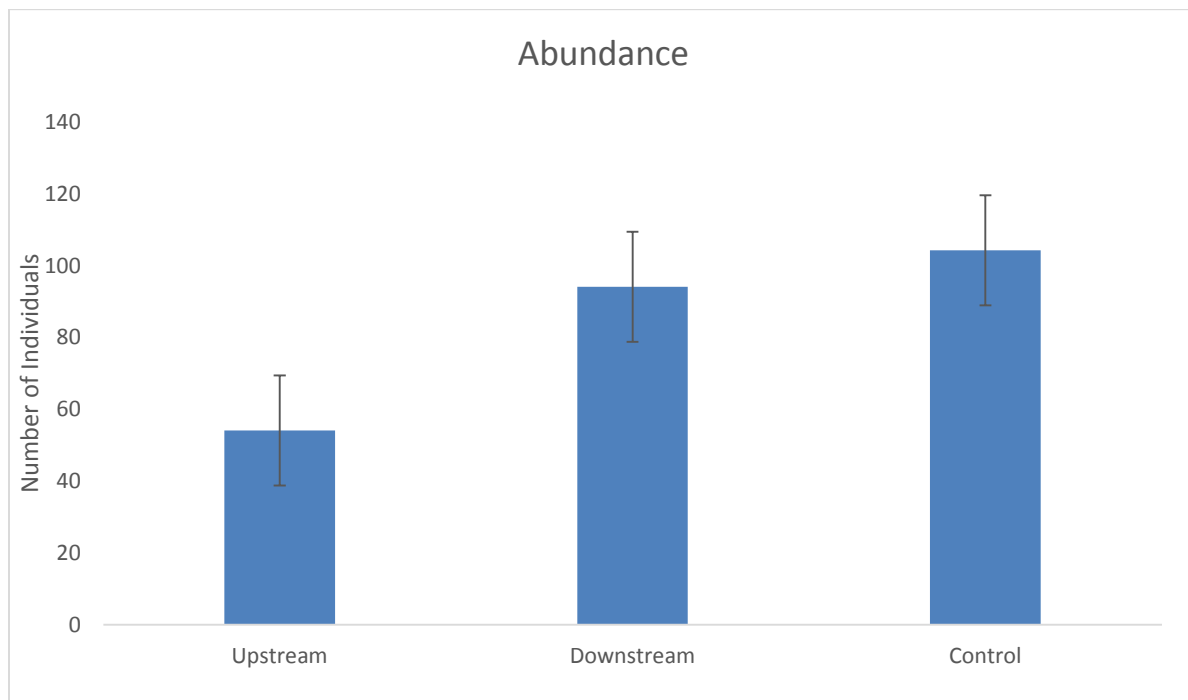


Figure 3: Mean abundance at each of three types of stream survey sites: upstream of structures at fragmented sites (mean = 54.14 individuals); downstream of structures at fragmented sites (mean = 94.17); and at free-flowing, unfragmented control sites (mean = 104.33). Error bars give the standard error. Differences in means are statistically significant (ANOVA; $p < 0.05$).

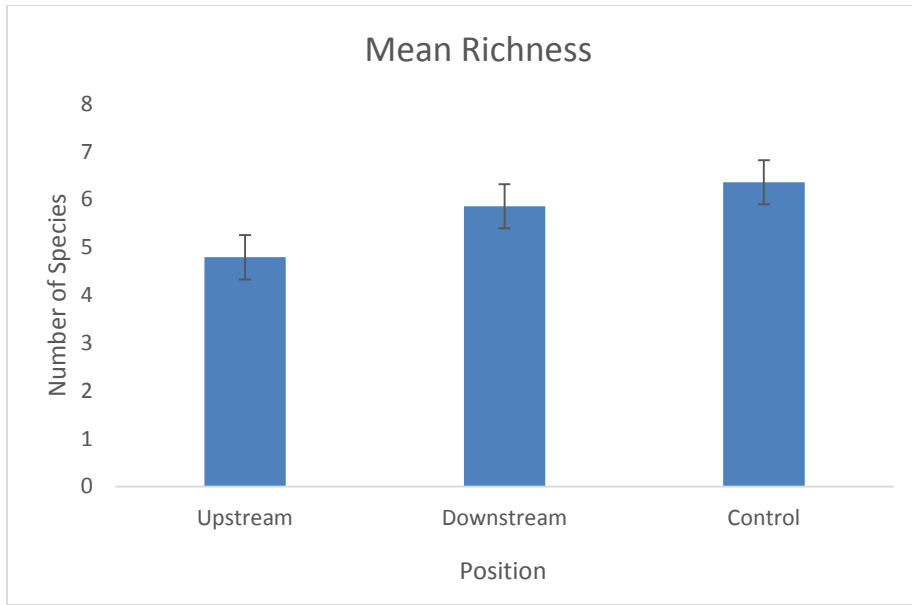


Figure 4: Mean richness at each of three types of stream survey sites: upstream of structures at fragmented sites (mean = 4.79 species); downstream of structures at fragmented sites (mean = 5.86); and at free-flowing, unfragmented control sites (mean = 6.36). Error bars give the standard error. Differences in means approached statistical significance (ANOVA; $p < 0.1$).

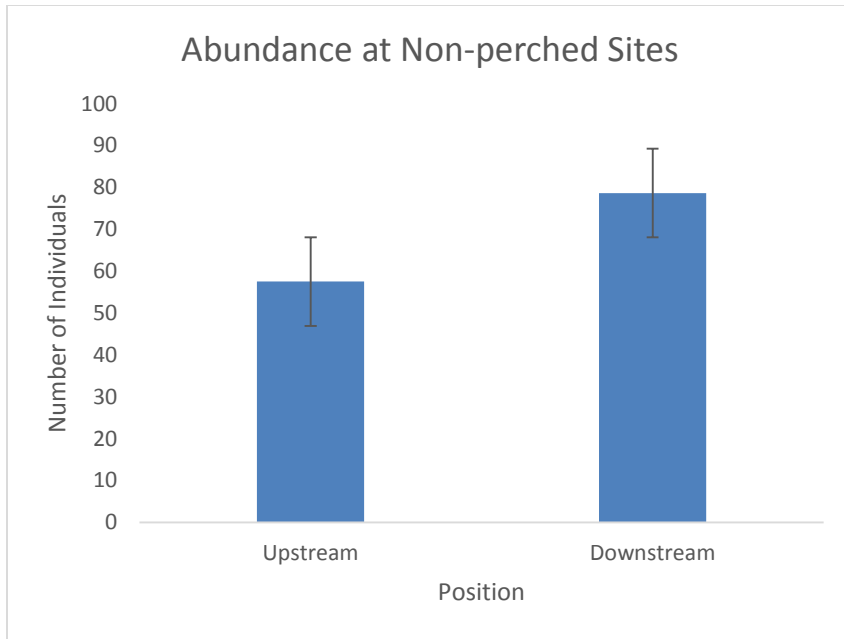


Figure 5: For sites with a physical barrier without a vertical outlet drop, abundance downstream of structure (mean = 78.70 individuals) vs. upstream of structure (mean = 57.55 individuals). Error bars give the standard error. The difference in means approached statistical significance (paired t-test; $p < 0.1$).

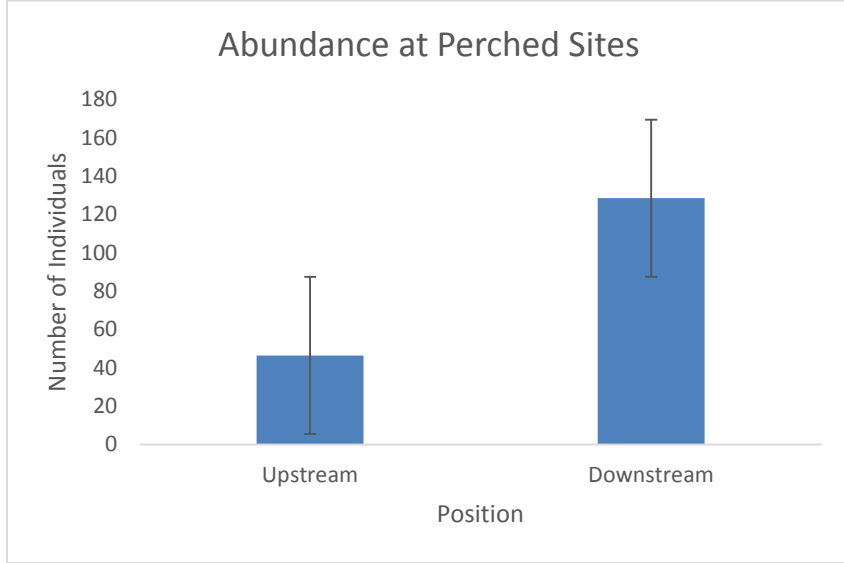


Figure 6: For sites with a physical barrier with a vertical outlet drop (i.e., perched sites), abundance downstream of structure (mean = 128.56 individuals) vs. upstream of structure (mean = 46.56 individuals). Error bars give the standard error. The difference in means approached statistical significance (paired t-test; $p < 0.1$).

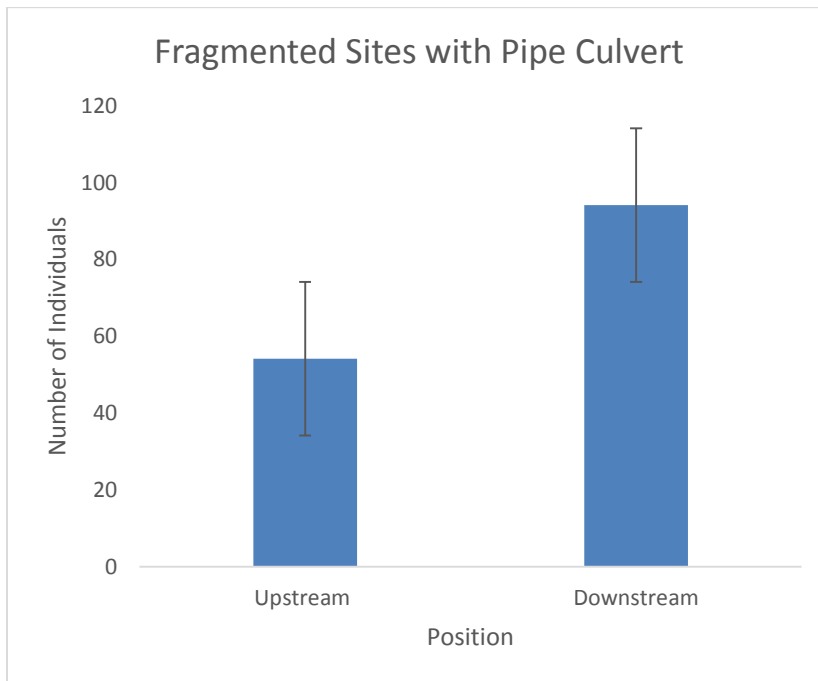


Figure 7: For sites with a physical barrier classified as a pipe culvert, abundance downstream of structure (mean = 94.17 individuals) vs. upstream of structure (mean = 54.15 individuals). Error bars give the standard error. The difference in means approached statistical significance (paired t-test; $p < 0.1$).

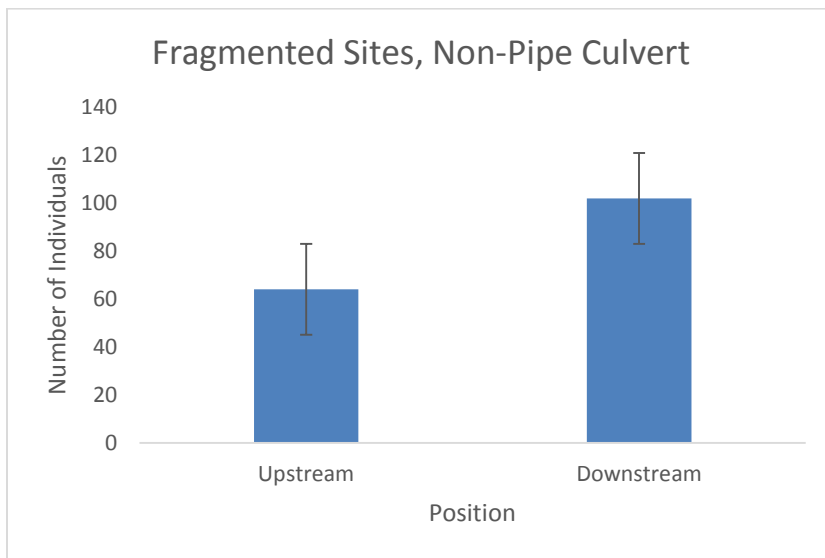


Figure 8: For sites with a physical barrier classified as a type other than pipe culvert (i.e., box culvert, arch culvert, or low-head dam), abundance downstream of structure (mean = 102.13 individuals) vs. upstream of structure (mean = 64.16 individuals). Error bars give the standard error. The difference in means approached statistical significance (paired t-test; $p < 0.1$).

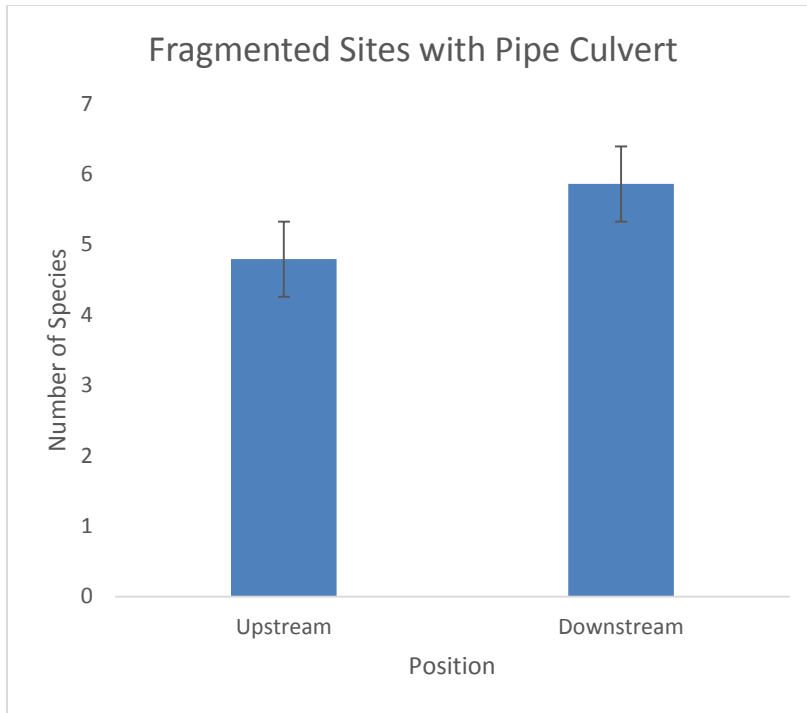


Figure 9: For sites with a physical barrier classified as a pipe culvert, species richness downstream of structure (mean = 5.86 species) vs. upstream of structure (mean = 4.79 species). Error bars give the standard error. The difference in means is statistically significant (paired t-test; $p < 0.05$).

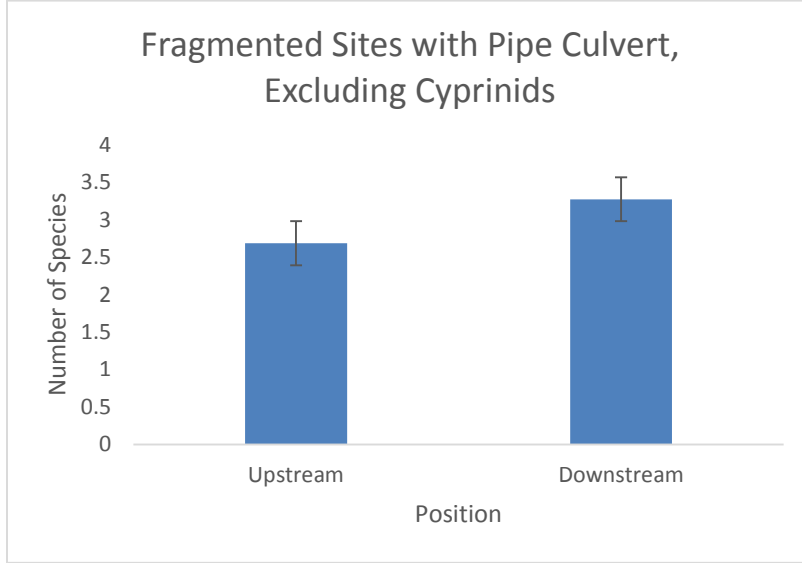


Figure 10: For sites with a physical barrier classified as a pipe culvert, species richness excluding Cyprinids downstream of structure (mean = 3.28 species) vs. upstream of structure (mean = 2.69 species). Error bars give the standard error. The difference in means is not statistically significant (paired t-test; $p > 0.05$).

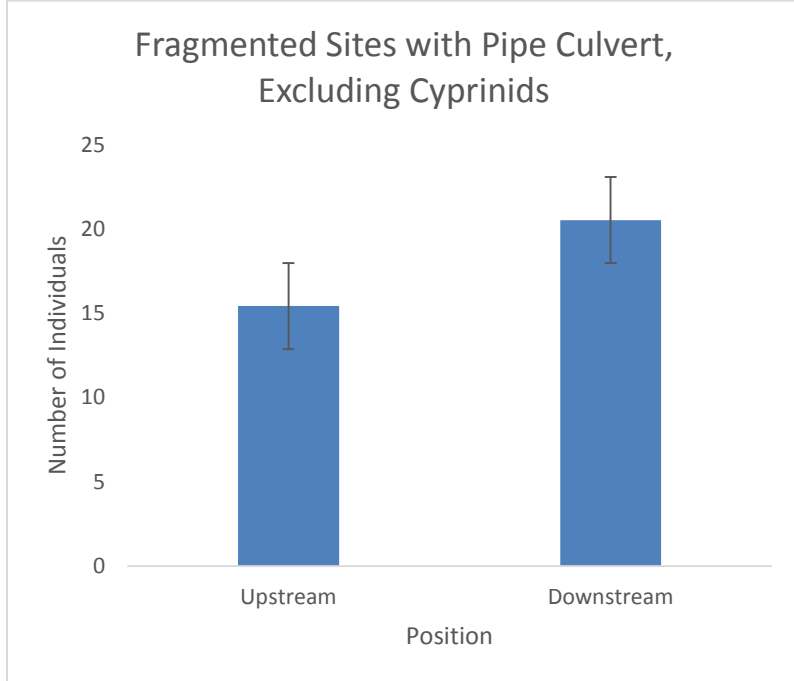


Figure 11: For sites with a physical barrier classified as a pipe culvert, abundance excluding Cyprinids downstream of structure (mean = 20.55 individuals) vs. upstream of structure (mean = 15.45 individuals). Error bars give the standard error. The difference in means approached statistical significance (paired t-test; $p < 0.1$).

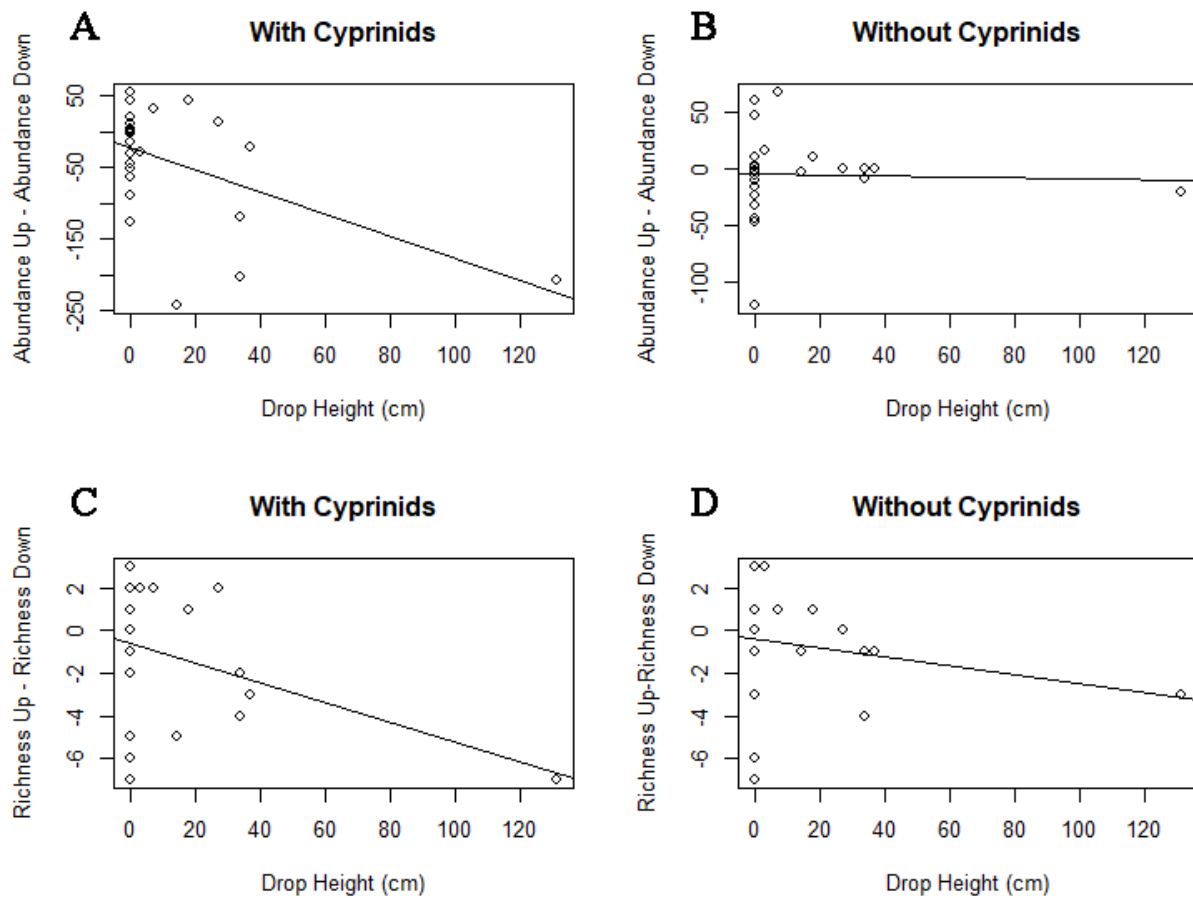


Figure 12: Relationships between the vertical distance between the water surface and the culvert outlet (drop height; x-axis) and fish abundance and species richness. Panels A and B give the difference in abundance between sites upstream and downstream of the structures for the entire fish community (A), and the same relationships without Cyprinids (B). Panels C and D give the difference in species richness between sites upstream and downstream of structures as determined for the entire fish community (C) and without Cyprinids (D). Lines give the best-fit linear regression to each set of points, and all regressions are statistically significant ($p < 0.05$).

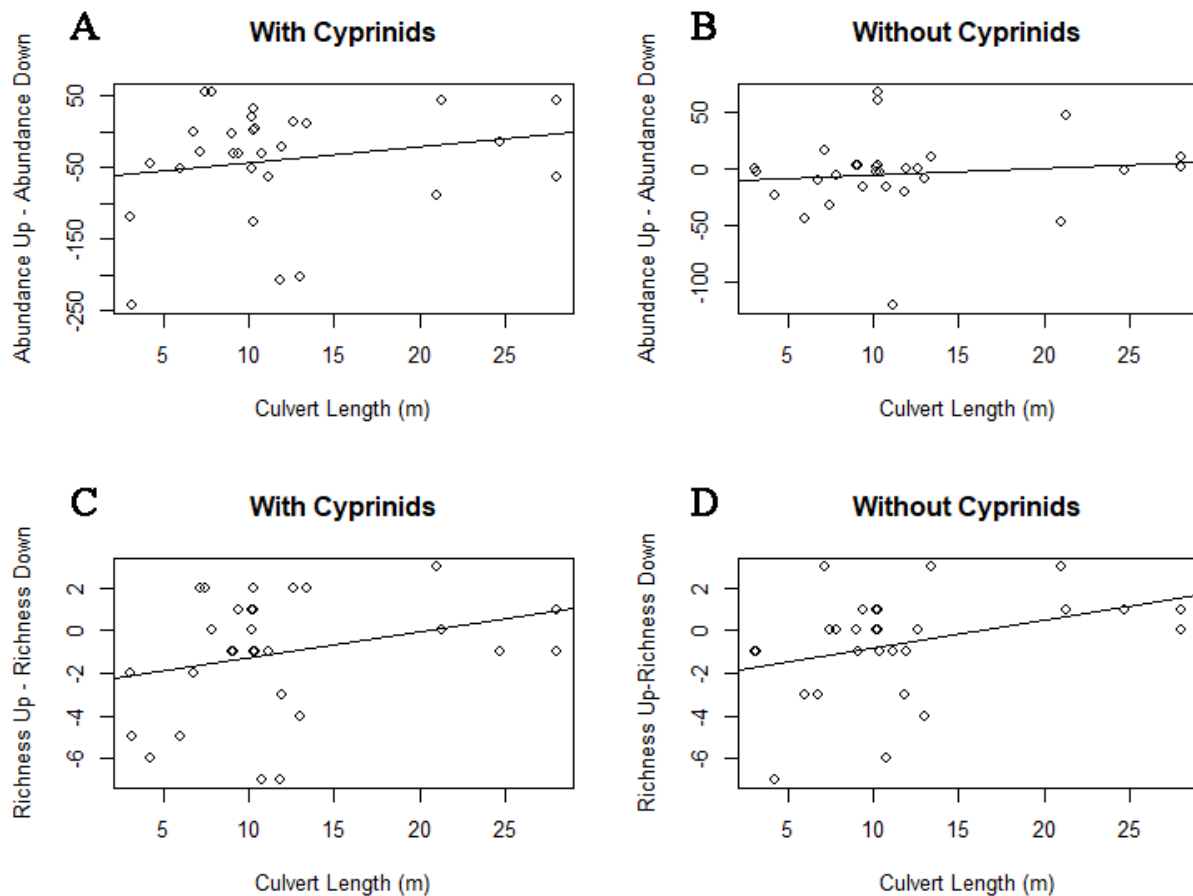


Figure 13: Relationships between the length of the culvert (culvert length; x-axis) and fish abundance and species richness. Panels A and B give the difference in abundance between sites upstream and downstream of the structures for the entire fish community (A), and the same relationships without Cyprinids (B). Panels C and D give the difference in species richness between sites upstream and downstream of structures as determined for the entire fish community (C) and without Cyprinids (D). Lines give the best-fit linear regression to each set of points. Regression lines in (A) and (B) are not statistically significant ($p > 0.05$), but lines in (C) and (D) are both significant ($p < 0.05$).

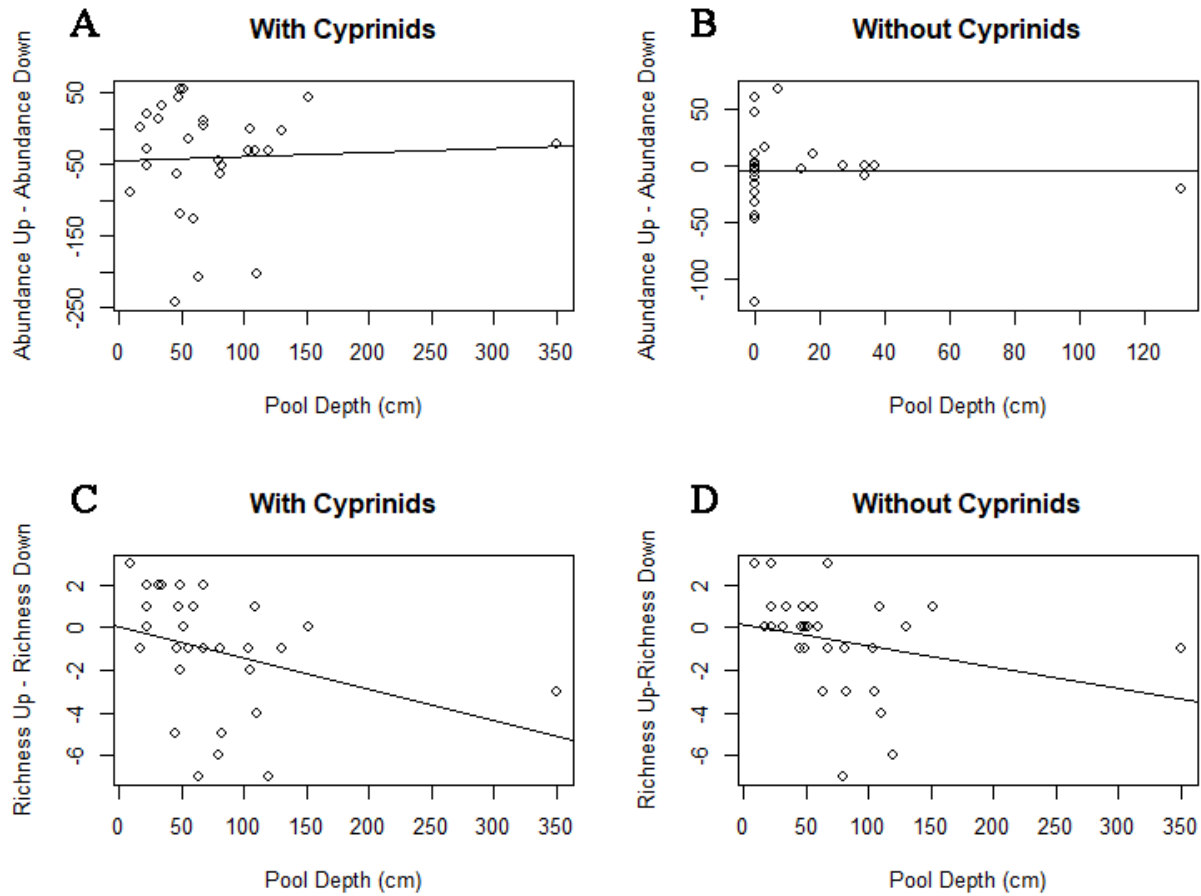


Figure 14: Relationships between the depth of the plunge pool (plunge pool; x-axis) and fish abundance and species richness. Panels A and B give the difference in abundance between sites upstream and downstream of the structures for the entire fish community (A), and the same relationships without Cyprinids (B). Panels C and D give the difference in species richness between sites upstream and downstream of structures as determined for the entire fish community (C) and without Cyprinids (D). Lines give the best-fit linear regression to each set of points; all regressions are not statistically significant ($p > 0.05$).

A Histogram of Upstream BC Pairs **B** Histogram of Downstream BC Pairs **C** Histogram of Control BC Pairs

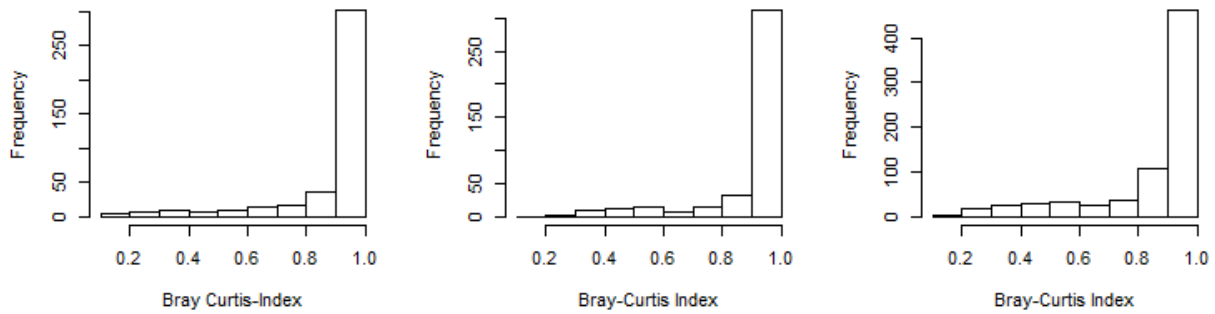


Figure 15: Histograms of fish community dissimilarity (as measured by the Bray-Curtis Index) for all pairwise combinations of all upstream sites (A), all downstream sites (B), and all non-fragmented control sites (C). BCI values of 1 indicate maximum dissimilarity in species composition.

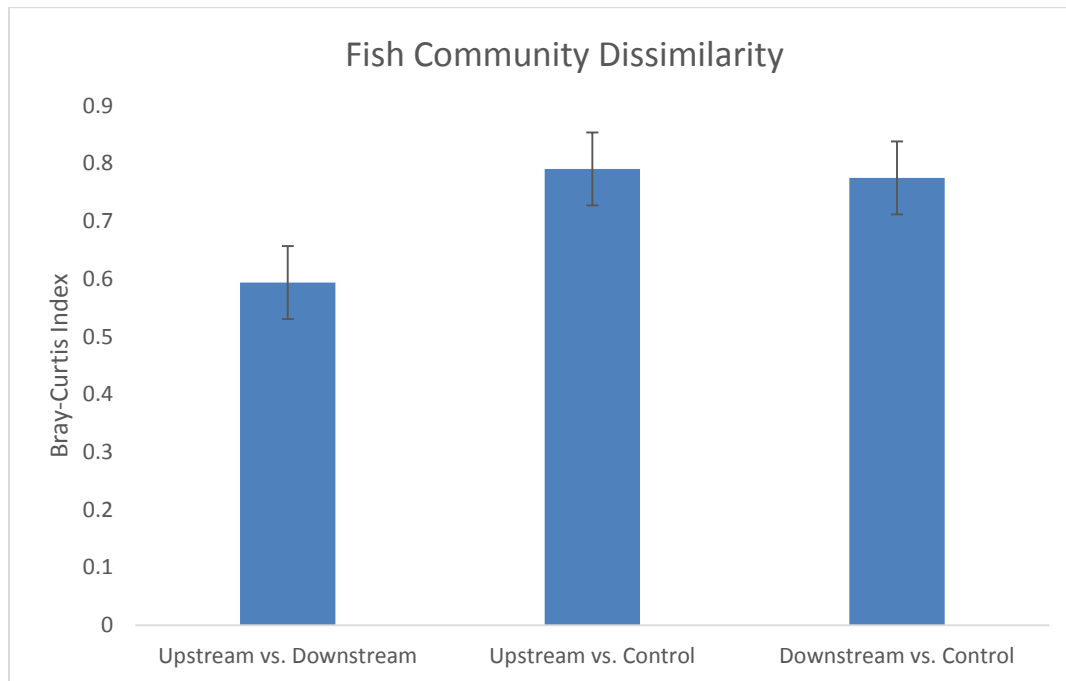


Figure 16: Fish community dissimilarity (as measured by the Bray-Curtis Index) between all pairs of upstream and downstream sites (mean BCI = 0.59), upstream and control sites (mean = 0.79), and downstream and control sites (mean = 0.78). BCI values of 1 indicate maximum dissimilarity in species composition. Error bars give the standard error. Differences in means are statistically significant (ANOVA; $p < 0.05$).