

U.S. Fish and Wildlife Service

Preliminary Testing of Flow-Ecology Hypotheses Developed for the GCP LCC Region

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Cooperator Science Series # 108-2014

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Contractual References:

This document fulfills reporting requirement for Wildlife Management Institute Grant Number GCP LCC 2012-003. Previously published documents that partially fulfilled any portion of this contract are referenced within, when applicable. (USGS IPDS #: IP-057262)

Recommended citation:

Brewer S. K. and M. Davis. 2014. Gulf Coast Prairie Landscape Conservation Cooperative Regional Hypotheses of Ecological Responses to Flow Alteration. U.S. Department of Interior, Fish and Wildlife Service, Cooperator Science Series FWS/CSS-108-2014, Washington, D.C.

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**PRELIMINARY TESTING OF FLOW-ECOLOGY HYPOTHESES
DEVELOPED FOR THE GCP LCC REGION**

Report to the U.S. Fish and Wildlife Service,
Gulf Coast Prairie Landscape Conservation Cooperative

Wildlife Management Institute Grant Number GCP LCC 2012-003



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July 1, 2014

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Acknowledgements

We would like to express our sincere appreciation to the many people and agencies that supported this work. In particular, the authors and other members of the Flow-Ecology Hypothesis Committee of the GCP LCC Instream Flow Project provided the basic hypotheses for this study. The hypothesis authors were: Shannon Brewer, Mary Davis, Jacquelyn Duke, Kimberly Elkin, Nicole Farless, Timothy Grabowski, Kevin Mayes, Robert Mollenhauer, Trevor Starks, Kevin Stubbs, Andrew Taylor, Caryn Vaughn, and Tom Worthington. Daniel Fenner, Thom Hardy, David Martinez, Robby Maxwell, Bryan Piazza, and Ryan Smith provided helpful reviews and improved the quality of the hypothesis compilation report. We thank Emily Tracy-Smith and Michele DePhilip for comments provided on an earlier draft. Funding for this work was provided by the Gulf Coastal Prairie Landscape Conservation Cooperative of the U.S. Fish and Wildlife Service. The contract was administered by the Wildlife Management Institute (Grant Number GCP LCC 2012-003). Any use of trade, firm, or product names is for descriptive purposes and does not imply endorsement by the U.S. Government.

Executive Summary

The Ecological Limits of Hydrological Alteration (ELOHA) framework calls for the development of flow-ecology hypotheses to support protection of the flow regime from ecologically harmful alteration due to human activities. As part of a larger instream flow project for the Gulf Coast Prairie Landscape Conservation Cooperative (GCP LCC), regional flow-ecology hypotheses were developed for fish, mussels, birds, and riparian vegetation (Davis and Brewer 2014¹). The objective of this study was to assess the usefulness of existing ecological and hydrological data to test these hypotheses or others that may be developed in the future. Several databases related to biological collections and hydrologic data from Oklahoma, Texas, and Louisiana were compiled. State fish-community data from Oklahoma and Louisiana were summarized and paired with existing USGS gage data having at least a 40-year period of record that could be separated into reference and current conditions for comparison. The objective of this study was not to conduct exhaustive analyses of these data, the hypotheses, or analyses interpretation, but rather to use these data to determine if existing data were adequate to statistically test the regional flow-ecology hypotheses. The regional flow-ecology hypotheses were developed for the GCP LCC by a committee chaired by Shannon Brewer and Mary Davis (Davis and Brewer 2014). Existing data were useful for informing the hypotheses and suggest support for some hypotheses, but also highlight the need for additional testing and development as some results contradicted hypotheses. Results presented here suggest existing data are adequate to support some flow-ecology hypotheses; however, lack of sampling effort reported with the fish collections and the need for ecoregion-specific analyses suggest more data would be beneficial to analyses in some ecoregions. Additional fish sampling data from Texas and Louisiana will be available for future analyses and may ameliorate some of the data concerns and improve hypothesis interpretation. If the regional hydrologic model currently under development by the U.S. Geological Survey for the South-Central Climate Science Center is improved to produce daily hydrographs, it will enable use of fish data at ungaged locations. In future efforts, exhaustive analyses using these data, in addition to the development of more complex multivariate hypotheses, would be beneficial to understanding data gaps, particularly as relevant to species of conservation concern.

¹ <http://southeastaquatics.net/resources/sifn-resources/documents/gcplcc-instream-flow-documents/flow-ecology-relationships/gcp-lcc-regional-hypotheses-of-ecological-responses-to-flow-alteration>

Background

The relationship between flow regime and aquatic biota is both fundamental and multifaceted, to the extent that flow regime is described as a ‘master variable’ controlling stream ecosystems and driving ecological processes (Power et al. 1995; Hart and Finelli 1999). Both directly, and indirectly through its impact on other regulatory factors (e.g., water quality and energy transfer), the flow regime (i.e., seasonal and inter-annual pattern of discharge timing and fluctuations), regulates the ecological integrity of aquatic ecosystems (Poff et al. 1997). Discharge is a key driver of physical habitat providing a template that determines both taxonomic and functional community structure (Poff and Allan 1995; Bunn and Arthington 2002; Bêche et al 2006). Sediment and solutes are transported through the system creating spatial and temporal gradients in physicochemical conditions. Discharge shapes the flow of energy through a river system (e.g., River Continuum Concept, Vannote et al. 1980) and between the channel and surrounding ecosystems (e.g., Flood Pulse Concept, Junk et al. 1989). Disturbance in the form of high and low-flow events promotes habitat complexity and patchiness through lateral and longitudinal connectivity, and natural fragmentation (Lake 2000). Discharge drives hydraulic conditions that stimulate biological processes like nutrient uptake and decomposition (Doyle et al. 2005) and predator-prey interactions (Hart and Finelli 1999). The ecological consequences of flow alteration are pervasive, although the degree and direction of flow alteration varies spatially and temporally (Poff et al. 1997; Naiman and Turner 2000).

The importance of natural flow regimes to the ecological integrity of rivers has been established for decades, but protective regulatory and management instream flow criteria or standards are lagging behind the science (Richter 2009). More specific relationships of ecological response to flow alteration are needed to develop and implement scientifically credible instream flow standards and management recommendations (Richter 2009). The Ecological Limits of Hydrologic Alteration (ELOHA) framework for developing instream flow standards calls for supporting flow-ecology hypotheses with relationships established in the scientific literature or testing them with existing hydrologic and ecological information (Poff et al. 2010). However, recent reviews of the literature to support the development of instream flow standards reveal there is little available information that helps quantify specific ecological responses to flow alteration (e.g., Poff and Zimmerman 2009; McManamay et al. 2012). Further work is needed to quantify flow-ecology relationships using existing hydrologic and ecological information.

In 2012, the Gulf Coast Prairie Landscape Conservation Cooperative (GCP LCC) funded the Southeast Aquatic Resources Partnership (SARP) to work with partners (see Acknowledgements) and develop science-based resources to support instream-flow regulation and management in the GCP LCC region. Regional flow-ecology hypotheses were developed by a committee of aquatic experts for fish, mussels, birds, and woody riparian vegetation that were representative of rivers and streams in Louisiana, Oklahoma, and Texas (Davis and Brewer 2014). The hydrologic foundation was developed and existing ecological data were gathered for preliminary analyses. Additional resources were also developed, such as a regional river classification framework and flow-alteration assessment and can be found in Davis (2014). The principal focus of this project was to determine whether available data were sufficient to support quantification of the flow-ecology hypotheses. The regional hydrologic foundation and ecological information available in the GCP LCC, used to support this study, are discussed below.

Hydrologic Foundation

The ELOHA framework specifies a “hydrologic foundation” to quantify the alteration of ecologically significant components of the flow regime (Poff et al. 2010). The hydrologic foundation allows for the investigation of ecological responses to a holistic suite of hydrologic conditions that represent all components of the hydrograph (e.g., extreme low, high flow pulses, overbank flow; Poff et al. 2007, Tharme 2003). An ideal hydrologic foundation is based on daily hydrographs for a minimum of 15 years for reference and current conditions, respectively (See Poff et al. 2007 and Gao et al. 2009). This allows for calculation of the relative change due to human impacts on selected hydrologic conditions at locations where the ecological condition has been measured. This report investigated two aspects of the hydrologic foundation: the source of hydrologic data and specification of ecologically significant hydrologic metrics for the flow-ecology hypotheses.

There are several sources of hydrologic data with pros and cons for each that need to be carefully weighed for each flow-ecology project. Observed and modeled sources of hydrologic data differ in their availability, cost, accuracy, and management of error (Apse et al. 2008). For example, long-term gage data includes observed water levels that encompass the entire hydrograph (i.e., low to high water levels) and is readily available from the U.S. Geological Survey (USGS). These data are inexpensive, and relatively simple analysis tools (e.g., Index of Hydrologic Alteration; IHA) are readily available. However, long-term gage data have limited spatial and temporal availability and often are not located where ecological data have been collected. Further, determining human impacts on the hydrologic regime is confounded with climatic cycles when using comparisons of observations during historic reference and current altered time periods. In comparison, modeled hydrologic data are likely to have the greatest error at the extreme high and low ends of the hydrograph, which can be the most limiting ecological conditions (Lake 2000, Bunn and Arlington 2002). They are usually only available for limited geographies or on highly managed systems, have coarse spatial and temporal scales (e.g., Davis et al. 2013), and are expensive to develop (Apse et al. 2008). However, modeled hydrologic data can be developed for ungaged locations and greatly increase the use of existing ecological data. A fundamental part of the ELOHA framework is the development of hypotheses that predict ecological responses to flow alteration (Poff et al. 2010). The hypotheses are based on the premise that if the flow conditions change, those changes can be related to corresponding changes in the ecological condition of interest (e.g., reduced summer low flows results in increased water temperature and increased stress and mortality of mussels). Specifying the flow metric is challenging for several reasons. First of all, there are myriad hydrologic metrics, many of which are highly correlated (Poff et al. 2007). Rather than selecting metrics to minimize correlation among hydrologic metrics (e.g., ecological deficit; Gao et al. 2009), the hydrologic metrics should be selected to represent the proposed mechanism. Further, *a priori* selection among several closely related metrics is difficult without knowledge of their statistical behavior. To continue the example of altered summer low flows, several hydrologic metrics can be calculated to represent this condition. Whether the minimum 7-day flow during June through August is more meaningful than the maximum change in the three monthly minimum flows is difficult to determine at the hypothesis formation phase of ELOHA. Although these metrics are likely to be correlated, ecologists need to describe the limiting hydrologic condition as clearly as possible when formulating the hypothesis. The final selection of the metric for the flow-ecology

relationship is an iterative process during analysis. The hydrologic metric selected for the flow-ecology hypotheses should be related to human impacts and should be manageable.

The available hydrologic data for developing a hydrologic foundation was very limited in the GCP LCC region. Several hydrologic models exist in the region. However, no single model covered the three-state region of interest. Where models did exist, they were not able to be included in this effort because they were developed for different objectives (i.e., error could not be easily managed) and had monthly time steps. We chose to test the ability of existing gage data in the region to support analyses of the regional flow-ecology hypotheses.

Ecological Information

Part of the ELOHA framework is to use ecological data to test the hypothesized ecological responses to flow alterations (Poff et al. 2010). Ideally, the ecological data are representative of the whole aquatic ecosystem: the biological, chemical, and physical components. The ecological measurements would be taken under reference and altered hydrologic conditions. The measurements could be either taken in many places that differ in the degree of hydrologic alteration or taken repeatedly in places where hydrologic alteration changed over time. Under ideal conditions, the collection methods used would be standardized and efficient. Measurements need to be comparable over time and space so they can be used to calculate the relative change in ecological condition between reference and altered conditions. Finally, the ideal data have a sufficient number of measurements where hydrologic data exist to support statistical analyses. We investigated the regional sources of ecological data and the metrics that could be developed in this report.

Sources of ecological data were limited that could support testing the GCP LCC flow-ecology hypotheses for fish, mussels, birds, and woody riparian vegetation. In fact, fish-sampling data from the state game and fish agencies, in addition to other sources, were the only available aquatic ecological data in the region. These data provide counts by species for a sampling event at a specific location. Macroinvertebrates are the only other biota sampled on a regular basis across the region at adequate numbers of sampling sites to support analyses. However, no flow-ecology hypotheses for macroinvertebrates were developed as part of this instream-flow project (Davis and Brewer 2014). Further, fish data were the only data available in a *relatively* standardized format from Oklahoma and Louisiana (but see caveats throughout the report). The state fish data were previously compiled and placed within the Multistate Aquatic Resources Information System (MARIS) database, making them readily accessible. Texas fish data were not available at the time this study was initiated, but their data have been added to the Multistate Aquatic Resources Information System (MARIS) for future efforts.

In contrast to the many hydrologic metrics that could be calculated from the hydrologic data, selection of testable ecological hypotheses was very limited due to the limited ecological data available at this time. First, the hypotheses for biota other than fish could not be tested due to a lack of supporting ecological data. These flow-ecology relationships must be supported by the scientific literature and best professional judgment or ideally by future analyses using defensibly-collected data. Responses of fish abundance to flow alteration were supported by the available

fish data; however, relative abundance had to be used as a surrogate for true abundance (see section on analyses). All flow-ecology relationships involving fish reproduction, life stage, growth, mortality, condition, and habitat must be supported using other approaches or data collection.

Study Objectives

The objective of this study was to use the available hydrologic and ecological data in the GCP LCC Region to determine if existing data would be adequate to statistically test flow-ecology hypotheses developed as part of the overall project. The aim of the study was not to conduct exhaustive analyses of these data or data interpretation to recommend standards, but simply to explore the fish and hydrology data, and provide some recommendations on how these data might be improved and suggestions for data analyses.

Methods

Our general approach was to apply regression analyses to fish-sampling data paired with USGS gage data to test the GCP LCC flow-ecology hypotheses for fish relative abundance. Details of our methods to acquire and condition the data, calculate hydrologic and ecological metrics, and perform the analyses are provided below.

Fish-Community Data

Fish-community samples from Oklahoma and Louisiana were extracted from the MARIS database (<http://www.marisdata.org/>), and then paired with USGS stream gages that had been screened for environmental studies (GAGES II; Falcone et al. 2010, Falcone 2011). Fish samples that were collected within 10 km (upstream or downstream) of a USGS gage that had no second order or larger stream between the gage and the collection location were paired with that stream gage. All other fish samples were omitted from the dataset. We removed additional samples that were collected during a period when the gage was not operational. We also removed fish samples that were binary (presence/absence). Several samples were taken within close proximity to one another and often on the same day (perhaps by the same investigator). The catch from these samples, often at commonly accessed points (bridge locations), was averaged to provide one fish sample for that date. We identified dams and their maximum storage that occurred upstream of every gage using the National Inventory of Dams². Additionally, we identified the five largest tributary dams (all were small watershed dams and only 5 were recorded because there could be > 50 in some cases) and their maximum storage within 50 km upstream of each gage. Existing dam information was used to screen the hydrology data (*see* subsection below).

Hydrologic Data

Flow alteration was calculated for the selected USGS gages. The period of daily flow record was obtained from USGS for each of the selected gages and reviewed for completeness of record (i.e., no prolonged periods with no data) and at least 40 years of observations.

² <http://nid.usace.army.mil/>. Last accessed on July 23, 2013..

Reference and altered time periods were selected for each gage using criteria intended to minimize variation in flows due to differences in climate (Figure 1). The reference time period included 20 years early in the gage record that overlapped 1940-1960 as closely as possible. The altered time period included the 20 years of record prior to the most recent fish sampling date at that location, which usually included the years 1985-2005. A 20-year time period of record was selected for analyses to cover major climate cycles (Poff et al. 2007, Gao et al. 2009). On rivers with controlled flow regimes due to large dams, reference and altered periods reflected pre- and post-dam construction periods.

The Index of Hydrologic Alteration (IHA Version 7; The Nature Conservancy³) was used to calculate select hydrologic metrics for each gage for the reference and altered time periods. Hydrologic metrics were selected based on specific flow-ecology hypotheses that were developed by an expert committee (*see* Davis and Brewer 2014). Default and nonparametric IHA analysis settings were used to compare the reference and altered flow conditions (Appendix A). Analyses were run using the batch feature that allowed reference and altered time periods to be specified for each gage. Separate analyses were completed for hydrologic metrics with specific seasons (e.g., annual, April-June, and June-August). Percent of flow alteration was calculated manually to maintain direction of alteration from reference conditions (i.e., increase or decrease of an altered flow metric from reference conditions).

Calculation of Fish Metrics

Species in the data set were determined to be native or non-native at each sample location. First, the USGS website for nonindigenous aquatic species⁴ was used to determine the native distribution of each species; however, if a fish species was not available on the USGS website or if the location of capture was near the edge of the range, then field guides were used to determine the distribution (Page and Burr 1991; Pflieger 1997; Miller and Robison 2004). The status (native or nonnative) of each species was also verified by a professional in each state of record.

Fish collections had been obtained using a variety of sample gears and levels of effort. Because level of effort was not reported for most sampling events, we were limited to calculating fish metrics the data would support. We calculated relative abundance (abundance of the species or guild/total abundance of fish in that sample) to account for the lack of standardized approach. Relative abundance was calculated for the following: 1) four individual species (*Etheostoma spectabile*, *Noturus nocturnus*, *Notropis girardi*, and *Atractosteus spatula*); 2) all pelagic broadcast spawning cyprinids within Oklahoma and Louisiana (*Macrhybopsis aestivalis*, *Macrhybopsis hyostoma*, *Macrhybopsis storeriana*, *Notropis bairdi*, and *Notropis sabiniae*); 3) sucker species for the following genera: a) *Moxostoma* (*Moxostoma poecilurum*, *Moxostoma carinatum*, *Moxostoma erythrurum*, *Moxostoma duquesnei*) b) *Carpionodes* (*Carpionodes velifer*, *Carpionodes cyprinus*, *Carpionodes carpio*) and c) *Hypentelium* (*Hypentelium nigricans*); and 4) lithophilic species. The individual species were chosen based on the hypotheses generated via an

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<http://www.conservationgateway.org/ConservationPractices/Freshwater/EnvironmentalFlows/MethodsandTools/IndicatorsofHydrologicAlteration/Pages/IHA-Software-Download.aspx> Last accessed on June 5, 2013.

⁴ <http://nas.er.usgs.gov/>, Last accessed October 22, 2013.

expert committee (See Davis and Brewer 2014). Briefly, the pelagic broadcast spawning fishes, including Arkansas River Shiner (federally threatened), were chosen because of existing evidence suggesting flow and fragmentation are linked to their decline (see Worthington et al. 2014 and references therein). Species belonging to the lithophilic reproductive guild, including many darters and madtoms, were chosen because they are thought to require flowing water and silt-free substrates for successful reproduction (Balon 1975, see more detailed description below). Finally, several of the genera of sucker were chosen for hypothesis development because they are considered sensitive to altered flow regimes (Freeman and Marcinek 2006).

Lithophilic species were defined using guilds by Balon (1975) and traits by Frimpong and Angermeier (2009). Balon acknowledged three reproductive guilds of lithophilic fishes defined as species that require oxygenated water for spawning, achieving this by spawning on clean substrate or spawning in a location that has clean flowing water: nonguarders (open substrate spawners), nonguarders (broodhidiers) and guarders (nest spawners). Frimpong and Angermeier (2009) defined a fish trait called lithophils but unlike Balon, divided the groups further to include selection for substrate type. Because selection of substrate varies by river and availability, we chose to use the broader definition of the group and combined all of the fish into one group of 'lithophilic fishes' regardless of selection for substrate type. The lithophilic guild included the following species (FishTraits database⁵): *Fundulus olivaceus*, *Hypentelium nigricans*, *Minytrema melanops*, *Moxostoma carinatum*, *Moxostoma duquesnei*, *Notropis greenei*, *Percina aurora*, *Pimephales tenellus*, *Alosa alabamae*, *Catostomus commersonii*, *Semotilus atromaculatus*, *Etheostoma lynceum*, *Hybopsis winchelli*, *Moxostoma erythrurum*, *Moxostoma poecilurum*, *Notropis girardi*, *Notropis boops*, *Notropis longirostris*, *Notropis shumardi*, *Phenacobius mirabilis*, *Campostoma anomalum*, *Campostoma oligolepis*, *Etheostoma spectabile*, *Ichthyomyzon castaneus*, *Luxilus cardinalis*, *Luxilus chrysocephalus*, *Percina carbonaria*, *Percina pantherina*, *Etheostoma stigmaeum*, *Etheostoma swaini*, *Ichthyomyzon gagei*, *Percina caprodes*, *Percina sciera*, *Percina shumardi*, *Semotilus atromaculatus*, *Nocomis asper* and *Nocomis leptocephalus*. Some of these species may also belong to other guilds (e.g., Arkansas River Shiner, pelagic broadcast spawning guild, see Taylor et al. 2014) but for simplicity, we have kept the designation provided in the FishTraits database for the lithophilic metric. A separate hypothesis was developed for Arkansas River Shiner and was applied to the pelagic broadcast spawning guild (see Worthington and Brewer 2014 for a list of associated species).

Analyses of Flow-Ecology Hypotheses

Exploratory plots of the data were completed to make assessments of how to best condition and analyze these data. Scatterplots were made of all data prior to analyses to identify outliers or overly influential data points and assess whether data caps or omissions would be needed to accommodate the use of relative abundance data. We also determined the relative shape of the patterns (e.g., linear, curvilinear) to see if thresholds in the data were apparent and whether data should be split to reflect increases or decreases in a flow metric (different forms of alteration that might be captured with different line directions).

⁵ <http://fishtraits.info/>. Last accessed October 12, 2013.

We were interested examining hypotheses at the ecoregion level to determine if hypotheses might differ depending on the influence of geology and climate (major influences of ecoregion designations). We found that using the Level-III Ecoregions created a situation where data were limiting (Figure 4B, Great Plains Ecoregion). To account for these shortcomings, we grouped ecoregions based on similarities between climatic conditions and fish-community structure and capped data to remove major outliers (Figure 4). Ecoregions were grouped as 1: Great Plains, 2: Southern Plains, and 3: Ouachita Mountain, Ozark Highlands, and Cross Timbers for the remaining analyses.

A series of simple and quantile regression analyses were conducted on these data to see if specific hypotheses could be supported given existing data in the GCP LCC Region. Simple least squares regression models the relationship of a response variable to X whereas quantile regression extends that basic concept to quantiles. Quantile regression is used extensively in ecological studies to explore the relationship between conditional quantiles (i.e., they can be defined by the user) of Y given one or more covariates. Quantile regression is considered to be especially useful when the rate of change in the relationship depends on certain percentiles of interest (e.g., median values or 90%), particularly if extremes are important. Quantile regression also makes no distributional assumption about the model error term (Koenker and Hallock 2001). The Simplex Algorithm was used to fit the flow-ecology relationship using quantile regression. We estimated a range of quantiles (50%, 75%, and 90%) because sampling variation can change rapidly near the extremes of the data. All statistical analyses were performed using SAS (SAS 2000).

Results

Substantial amounts of fish-community data were available in MARIS from Louisiana and Oklahoma. However, after data were truncated to gage locations and useable data (see section above on fish-community data), we only used 12% (405 of 3370 samples) and 4% (371 of > 10,000 samples) of data from OK and LA, respectively. There was a large amount of data collected in LA that was associated with bay and estuary locations so not applicable to this effort. After accounting for issues associated with multiple investigators, samples taken in close proximity to one another, gage pairing, etc., these fish sampling data were condensed from 62 to 39 locations in OK and 25 to 15 locations in LA.

Hydrologic Metrics

The selection of hydrologic metrics was a fundamental portion of the development the flow-ecology hypotheses as they represent the mechanisms driving the ecological response. Further, the statistical nature of the metric influences the outcomes of statistical tests. Although not all of the selected hydrologic metrics were used for analyses, an examination of the range of hydrologic conditions represented by the metrics and the statistical nature of the metrics is instructive for future flow-ecology efforts.

Hydrologic metrics specified in the flow-ecology hypotheses

Twenty six hydrologic metrics were calculated to reflect the metrics used in the 27 flow-ecology hypotheses developed for the GCP LCC region (Table 2). The metrics represent all of the ecological-flow components (EFC, i.e., extreme low flow, base flow, high flow pulse, and overbank flow). Base-flow metrics were the most commonly used in the flow-ecology hypotheses. EFCs were further described by flow attributes (i.e., magnitude, duration, frequency, timing, and rate of change) and magnitude and duration of the EFCs were the most common. Although the rate of change was not identified for these hypotheses, variability of daily base flows was included.

Most of the hydrologic metrics specified a season when a flow condition was critical for completion of the life cycle (Table 2). For example, fish species differ in reproductive season so magnitude and duration metrics reflected the hydrologic conditions for the appropriate time of year for each species. Alligator gar, for example, requires access to inundated floodplains to spawn in the late winter and early spring (Etnier and Starnes 1993, Ferrara 2001), whereas, Arkansas River Shiner spawns in the summer months (generally June and July but sometimes in August⁶). Several flow-ecology hypotheses identified low summer flows as critical for buffering from extreme water temperatures. Hypotheses for riparian forests specified the growing season. Annual metrics were generally specified for hypotheses about physical habitat conditions. For example, high-flow pulses are important for maintaining habitat quantity, quality and availability regardless of when the habitat is actually used.

Another distinction among the hydrologic metrics was statistical in nature (Table 2). Use of medians was the most common. Medians are favored over means as being more representative of hydrologic conditions (C. Apse, TNC, and C. Konrad, USGS, personal communications, 2010). Mean values are more influenced by extreme events, such as very large floods. In the GCP LCC Region, many rivers are very flashy as there is minimal groundwater inflow and rainfall occurs during distinct periods of the year, so median values were preferred over means.

For several of the flow-ecology hypotheses, there were alternative hydrologic metrics that could be applied to a particular relationship. In these cases, it was not clear which metric best described the hydrologic condition driving the ecological response. In the case of ecological responses to reduced summer flows, for example, quantification of alteration of summer flow magnitude could be described several ways (Table 2). Alternative summer flow metrics included:

- 1) a change in the average daily flow for the time period (e.g., median cfs from June 1 to August 31) or
- 2) the maximum change in monthly mean flow for June, July, and August.

Alternative metrics were also identified for the base flow attributes and overbank flow magnitude.

⁶ http://www.fws.gov/ecos/ajax/docs/life_histories/E05X.html

Selected hydrologic metrics for analyses

The flow-ecology hypotheses tested for this exercise were selected based on the availability of biological data, particularly as related to specific ecoregions. Eleven hydrologic metrics for the hypotheses that predicted a change in fish abundance with flow alteration were calculated for analyses at the 54 study locations (Table 2). We visually examined the frequency distributions of the flow alteration for these metrics to ensure they represent a wide range of altered flow conditions and help inform the interpretations of the flow-ecology hypotheses test results.

A review of the frequency distributions of the flow alteration metrics is instructive for identifying a general pattern of flow alterations in the region between the reference and altered time periods (Figure 2). The direction and degree of flow alteration of all of the hydrologic metrics spanned a wide range from -1 (i.e., the maximum decrease in any hydrologic condition) to values much greater than 1 (i.e., values could greatly exceed 100% increase over reference conditions). Most sites had an increase in extreme low flow (Figure 2A) and annual base flow (Figure 2C) magnitudes. Summer base flow magnitudes remained relatively stable (Figure 2B). Both daily (Figure 2D) and annual (Figure 2E) flow variability decreased. Overbank flow magnitudes also decreased (Figure 2F).

The alternative hydrologic metrics for each flow condition revealed similar patterns in flow alteration (Figure 2). However, metrics based on median values for a time period (i.e., seasonal or annual) generally had a more narrow range of flow alterations than metrics based on maximum changes in monthly values. Maximum change in monthly values is calculated by selecting the greatest change from the individual months in the time period of interest (i.e., all 12 months for maximum annual change or 3 months for the summer months June-August). In contrast to change in median values for the time period of interest, the maximum change in monthly values focuses on the most extreme change for the ecological response. Maximum change in monthly values tends to include very large degrees of flow alteration and their distribution is skewed to larger values (e.g., Figure 2B and 2C). Statistically the median values are preferable, but more investigation is needed to identify which form of metric is more ecologically meaningful.

The range of flow alteration for each attribute would ideally span large decreases to large increases in flow alteration (i.e., $-1 \leq (\text{reference-altered})/\text{reference condition} \leq 100$) to best support tests of the flow-ecology hypotheses. The frequency distributions of the hydrologic metrics used in this effort suggest that the sites used for these analyses did cover a relatively broad range of flow alteration (Figure 2). There was a wide range of altered hydrologic attributes as well.

In addition, the frequency distributions of the hydrologic metrics (Figure 2) illustrate differences among the alternative metrics that would influence the quantification of the ecological responses to flow alteration and use of the results. Most of the sites fall within a continuum of levels of alteration. However, outliers with extreme degrees of alteration occurred for the maximum change of median monthly base flow magnitude (Figure 2C) and annual high flow pulse frequency (Figure 2E). *A priori* criteria to remove outliers were not used, but would improve the analyses results by removing isolated points that heavily influence the regression.

Ecological Response Metrics

A wide range of ecological metrics were specified for the GCP LCC flow-ecology hypotheses (Table 3). The Flow-Ecology Hypothesis Committee chose to focus on biological response metrics for two reasons. First, the use of these hypotheses in a stakeholder process to determine instream flow criteria or water management practices would be more effective if they related to ecological responses of interest to the public. This was also part of the rationale for focusing on fish. Secondly, the mechanisms involved with the biological responses often related to a holistic suite of hydraulic, physical, and water-quality processes of streams and rivers. It was not necessary for this effort to specify responses of these processes to flow alteration as they are inferred in the biologically-based flow-ecology hypotheses.

The committee also determined that flow-ecology hypotheses would not be limited by the availability of ecological data. Many ecological response metrics were specified that cannot be tested at this time. However, the importance of these responses to flow alteration has been documented and identifies additional data needs in the region (see Davis and Brewer 2014).

In fact, due to the ecological data limitations outlined above, none of the ecological metrics specified for the GCP LCC flow-ecology hypotheses could actually be tested. Flow-ecology hypotheses for fish abundance were tested using *relative* abundance as the best approximation of the original metric.

Analyses of Flow-Ecology Hypotheses

Several of the 27 flow-ecology hypotheses developed for the GCP LCC region (Davis and Brewer 2014) were tested. Analyses of species-level hypotheses were limited by using the fish-community data as many individual species had sparse data across the region. For example, Alligator Gar had only two data points across the state of Oklahoma so no analyses were possible. The regional flow-ecology hypotheses were proposed at the species level, but were anticipated to be broadly applicable to the guilds associated with the species (Davis and Brewer 2014). Therefore, we focused our preliminary analyses on guilds or other meaningful groups of fishes (e.g., redhorse suckers *Moxostoma* spp.). Many of the hypotheses were tested multiple times to determine data availability and ecoregion influences.

Data were initially plotted to examine how best to complete the analyses and learn about the structure of these data. If the flow-ecology hypotheses were non-linear and directional (i.e., on each side of the baseline), we planned to develop more than one regression per hypothesis to examine the different relationships that may occur with decreases and increases in particular flow metrics. However, after plotting the data, it was clear that some flow metrics had very few data points related to the positive or negative change in flow metrics (Figure 3, hydrologic variable c). Further, it was evident that outliers of the ecological conditions existed in these data and were probably the result of using relative abundance as our ecological metric (e.g., Figure 3, lit > 80). Relative abundance is highly influenced by the presence of other fishes and methods used for sampling (unknown for these data). For example, if you encounter a school of fish during a sampling event, then the relative abundance of the particular species of interest could be substantially higher or lower. Further, certain gears are better able to capture some fishes over others (i.e., highly influenced by body morphology, behavior, etc) and can bias the overall catch.

We intended to test the hypothesis that abundance of Arkansas River Shiner would decrease with a decline in baseline flow variability during the spawning months (June-August) (Figure 5, Worthington and Brewer 2014). However, because data on individual species were often limited within an individual ecoregion, we instead tested the relationship by using the relative abundance of the pelagic broadcast spawning cyprinids (PBSC), the reproductive guild associated with Arkansas River Shiner⁷.

In the Southern Plains Ecoregion, there were only 8 capture locations of PBSC where the daily coefficient of variation (CV) of discharge was negative but 75 values where values were positive (Figure 6). Although substantial scatter in these data are apparent, there appeared to be a relationship of increasing relative abundance of these guild members with increasing alteration of CV. Simple linear regression indicated this relationship was statistically significant and explained 56% of the observed variation (Figure 7, Table 4 and Table 5). Unfortunately, the relationship is being undoubtedly influenced by a single point of $CV > 0.5$. Influential outliers can be ecologically meaningful but interpretation should be made with caution due to their undue influence on the statistical relationship. Given more data, this relationship might change or may only represent the linear portion of the hypotheses presented where increases in CV is positively and linearly related to PBSC (Figure 5).

Quantile regression of the PBSC hypothesis also indicated significant statistical relationships. The quantile regression coefficient of CV was significantly different from zero for the 75th ($y_{75} = -4.69 + 55.07x$) and 90th ($y_{90} = -3.89 + 61.52x$) percentiles but not the median (Table 6). This suggests that the different quantile regressions would provide different results for flow-management recommendations. However, interpretation of these data would vary on a case-by-case basis due to rare observations at the extremes that are pulling the direction of the relationship (Figure 8). Nonetheless, the sharp increase in the slope parameter shows where the parameter begins to differ from zero (i.e., 0.6), even though the confidence intervals around those estimates are relatively large (Figure 9). The relationships shown with both analyses (simple and quantile regression) confirm the linear portion of the hypothesis. The predicted threshold (where the line would become flat) is not apparent in these data but may still exist outside the range of the data used for the analyses.

Data were examined for testing the hypothesis that the abundance of lithophilic fishes would be reduced with changes in magnitude of monthly low-flow during summer. This hypothesis was based on Orangethroat Darter (Figure 10) but believed to represent the associated reproductive guild (Taylor et al. 2014). Again, noticeable outliers existed in these data (Figure 11, top panel). Decreases in baseflow showed no obvious pattern of ecological response (Figure 11, middle panel). However, data were truncated to examine the relationship between increases in median daily flows and increases in relative abundance of lithophils ($n=36$, Figure 11, bottom panel).

⁷ Note: Arkansas River Shiner was assigned to lithophils in the FishTraits database but has been assigned to the pelagic broadcast spawning guild by various authors. Guild designation is an important consideration in analyses of this type.

Whereas confidence intervals were wide, the relationship was statistically significant and explained approximately 42% of the observed variation in these data (Figure 12). When examining quantiles of these data, median values ($y_{50} = 3.20 + 78.78x$) and the 75% ($y_{75} = 19.85 + 64.62x$) were significant at $\alpha \leq 0.10$ (Figure 13, Table 7). Both approaches show significant results that contradict our hypothesis. It appears that the relative abundance of lithophilic fishes increases with increases in baseflow magnitude. A threshold response may become apparent outside of our tested data range. However, the relationship between decreases in baseflow magnitude and relative abundance is unclear and would benefit from further analyses via targeted data collection (i.e., from streams with this type of flow alteration).

Data were examined in this manner for other groups of fishes and most relationships were statistically significant when using guilds or some other combined ecological metric (e.g., groups of sucker species). However, depending on the hypothesis tested, adequate data were only available in some ecoregions. For example, relationships between flow metrics and *Moxostoma* spp. could only be examined in the 1) Southern Plains and 2) Ouachita Mountain, Ozark Highlands, and Cross Timbers. There were not enough data in the Great Plains to test the relationship. This suggests several options are available to test other regional flow-ecology hypotheses: 1) additional data collection, 2) developing other, ecologically meaningful ways to calculate ecological response metrics that are more broadly defined across regions, and 3) limit analyses to select locations.

Discussion

The results of this exercise using available hydrologic and ecological data to test the regional GCP LCC flow-ecology hypotheses were generally positive, despite limitations of these data for this application. The limitations included: only fish sampling data were available, few fish samples taken in some ecoregions, fish samples were collected for other purposes so were not ideal for testing our hypotheses (no effort or gear reported), and not enough data to represent some species (e.g., Alligator Gar). This indicates that the ecological responses to flow alteration we tested are robust enough to not be overwhelmed by the various sources of error. Although we were not able to test the entire suite of hypotheses, there were a number of lessons learned. These are discussed below with suggestions for further use of these data to support the development of flow-ecology relationships for regulatory or management purposes.

Use of Observed vs Modeled Hydrologic Data

Gage data provided an adequate hydrologic foundation for this study to statistically verify the flow-ecology hypotheses tested. While more data would improve our results, there was an adequate number of gages found near fish sampling locations with sufficiently long period of record to support quantification of flow alteration and statistical analyses. The daily hydrograph enabled the calculation of all the hydrologic metrics specified for the hypotheses. The use of gage data benefited from the large number of fish sampling locations in Oklahoma so that many of the available GAGES II gages could be paired with fish data and used in the analyses. The

major limitations of using gage data for the hydrologic foundation in this study are in the interpretation of the analyses. One reason for caution before using these results is that it is not clear whether flow alterations were due to human or climate influences. For example, increases in 7-day minimum flow and decreases in daily coefficients of variation at many locations could be explained by differences in weather or water management. This could be due to climate differences between the reference period (early in the period of record) and altered period (late in the period of record). Without separating the influences of human impacts from precipitation and temperature in the hydrograph, which is a significant modeling effort with its own set of data limitations, there is no way to distinguish the cause of the flow alteration. If the purpose of the exercise was to determine how humans cause ecological changes in rivers due to flow alteration, further analyses of the gage data would be needed. Nonetheless, it is clear that ecological changes have occurred in these stream systems.

Another reason for caution before using results based on gage data is the relatively large degree of flow alteration (e.g., >50% alteration from reference condition) when compared with modeled data. As discussed above, gage data include at least two sources of flow alteration when comparing hydrologic records from historic and current time periods: climate cycles and human influences. Similar exercises testing flow-ecology hypotheses with modeled hydrologic data in other regions show a much more narrow range in the degree of flow alteration. These exercises use modeled data to compare reference (i.e., sources of alteration are removed from the hydrograph) and altered conditions for the same time period, which eliminates the influence of climate, and show most sites have hydrologic alteration within +/- 20% of reference conditions (e.g., NC EFSAB 2013). This difference in the degree of flow alteration between gaged data used in this study and modeled data used in other regions could be due to: 1) actual greater degrees of flow alteration in the GCP LCC region when compared to other regions (i.e., extreme groundwater pumping) or 2) influences of climate between different time periods used to calculate the flow alteration. Future efforts would benefit from addressing this uncertainty.

Interpreting the causes of this difference in quantifying the degree of flow alteration is beyond the scope of this project. However, it points to the problem of consistency in how the results are applied. Instream-flow standards could look very different depending on the source of the hydrologic foundation. For example, say an instream flow standard to maintain good ecological condition (*in sensu* Richter et al. 2011) was developed using gage data such that departures of impacted daily flows from reference conditions should be <30%. Standards based on modeled data with lower degrees of flow alteration could be lower for the same locations, possibly <10% allowable departure. Unless the user of these standards uses the same source of hydrologic data in their water resource regulatory or management system, the incorrect application of the standard would not have the desired outcome to manage ecological impacts due to flow alteration.

The answer to the question of whether gage or modeled data provide the best hydrologic foundation may be moot in many regions. The availability of data is likely to be the primary factor. As for the GCP LCC region, if no modeled data are available, the gage data may suffice. However, USGS intends to develop hydrologic models across the U.S. at fine temporal and

spatial scales (i.e., daily time step at all selected locations) under their Water Smart program (Lauren Hay, USGS, personal communication, 2014). The availability of these data, as well as hydrologic data from other sources, will greatly expand the use of existing fish-sampling data at ungaged locations. Errors in flow estimates at the extreme ends of the hydrograph can be managed if the models are calibrated for ecological applications. However, the question remains about keeping consistency in the application of results from one hydrologic source to another platform (e.g., water management planning models). Additional inquiry would help identify how the standards developed from different sources of hydrologic data can be applied before erroneous or irreversible water-management decisions are made.

Ecologically-Relevant Hydrologic Metrics

There has been a proliferation of hydrologic metrics in recent years to describe components of flow regimes for various purposes. Literally, hundreds of metrics can be found for management of water quality (e.g., 7Q10), hydraulics (e.g., bank-full discharge), habitat quality (e.g., velocity and depth), and other ecologically-significant components of the flow regime (e.g., monthly low discharge). Although we were not able to test differences in flow-ecology relationships using alternative hydrologic metrics, this exercise demonstrated the need for a full suite of metrics and flexibility in the *a priori* selection of hydrologic metrics. We found a productive approach was for ecologists to describe the cause-effect mechanism for the hypothesis as closely as possible and then select one or more applicable hydrologic metrics from a list (e.g., output variables from IHA). More than one metric was selected if there was question about how the metric would describe flow alteration over time. This allowed us to view the distribution of flow alteration for each metric. We could use metrics with well distributed degrees of flow alteration that best represented the mechanism for the ecological response and better understand the influence of outliers on the analysis results.

Many hydrologic metrics have been found to be correlated and down-selected to produce a more manageable size list for analyses (e.g., Poff et al. 2007, Gao et al. 2009). Reduction of correlated variables is important in river classification efforts that use multivariate statistical approaches to define hydrologic classes (e.g., Poff and Ward 1989, McManamay et al. 2011). However, the results of this study demonstrate that it is important to have a complete selection of hydrologic metrics that relate to the cause-effect mechanism of the flow-ecology hypotheses. Use of a down-selected set of hydrologic metrics may limit the ability of the hypotheses in describing the ecological response to specific flow alterations.

Fish-Assemblage Data

The fish assemblage data that were available for the GCP LCC region proved adequate to support statistical analyses of flow-ecology hypotheses. However, due to a number of limitations with how the data were collected and reported, it was useful for testing only few of the regional hypotheses that were based on responses in relative abundance to flow alterations.

The lack of a known effort and/or gear use was problematic as relative abundance was the only ecological metric that could be used for comparisons. Further, the use of relative abundance may increase the number of outlier observations because of gear bias and the possibility that large schools of fish may or may not be sampled in each event. Although it is important to make good use of these data so we can improve our understanding of flow-ecology relationships, the development of efficient sampling methods would improve our ability to make inferences about the effects of flow alteration on fish populations. It is clear that in some cases, the relationship between flow alteration and fishes are unknown (e.g., lithophilic fishes and increases in baseflow magnitude) and preliminary analyses of this type can help in identifying flow alterations that are problematic for biota.

The results of these analyses suggest the collection of quality fish-assemblage data would be beneficial to the development of instream-flow standards. An efficient and systematic sampling approach to sample aquatic communities or particular traits across a range of sites with varying levels of flow alteration would improve our ability to detect changes in biota with alteration. It is difficult to use the same monitoring approach across several ecoregions because of changes in environmental parameters (e.g., use of electroshocking across the range of conductivity); however, either developing protocols that can be compared across regions or at least within the same ecoregion are important to limit substantial bias in these data. Further, standardized-reporting methods would greatly increase the power and use of these data. Fewer of the data would have to be discarded due to unclear sample locations and effort. With comparable sampling approaches and data-reporting standards, many more flow hypotheses could have been tested including species occupancy, abundances, densities, demographics, etc.

The importance of ecoregion suggests some form of stream classification or block on these data are needed to better define the flow-ecology relationships. Our experience with the existing data suggests there is better predictive power in the eastern ecoregions. It is very likely that other landscape factors interact to influence the changes that may be observed. Further classification of the sampling locations or including landscape covariates (e.g., land use) may help alleviate these concerns. Additionally, it would be helpful to spend more time looking at these data in ways that may negate the effect of ecoregion (e.g., some combination of fish traits rather than just guilds). For example, there may be combinations of life-history strategies and morphologies that lead to success or failure in biotic responses to alteration. Examination of traits could be used to make predictions without consideration of specific taxonomy thereby making the conclusions applicable across regions.

Data to examine individual species trends may be supplemented by other sources. In many cases, there were not enough data to complete analyses of species (e.g., Alligator Gar). However, these data may be supplemented by digitizing records held in museums (e.g., Sam Noble Museum at the University of Oklahoma) and adding state-agency data if available in the region (e.g., Texas and Louisiana). Additionally, our examination of these data emphasizes the importance of well-developed long-term monitoring programs in determining responses to perturbations (e.g., flow alteration) over time and space.

Statistical Approaches

The simple and quantile regression approaches used in this exercise were useful in demonstrating the direction and significance of ecological response to flow alteration. Given the scoping level of these analyses and quality of the data, surprisingly robust relationships of ecological responses to flow alteration were derived. However, analysis protocols would be helpful in guiding decisions for data management and application of the results back to the flow-ecology hypothesis being tested. For example, analyses of these types would benefit from the creation of decision points (e.g., data caps) to deal with outlier observations without omitting points that may be ecologically meaningful.

There are a variety of other statistical approaches that may be beneficial when examining these data. A preliminary look at use of multiple regression analyses (not reported here) indicates there are statistically significant quadratic forms suggesting thresholds exist in the relationships. This is important because it indicates that there is likely an identifiable breakpoint where ecological condition begins to decline rapidly that may be helpful in identifying important management actions. Breakpoint analyses would be a helpful approach that could identify when abrupt changes occur (the trend changes from linear) and may provide a different type of result to base management options. General additive models are an additional analysis that could prove helpful when enough data exist that multiple lines may be fit to these data. This approach would identify different directions in these data (e.g., positive, negative, or flat lines) without the need for separate analyses.

Our preliminary analyses represent ecological relationships to single hydrologic variables when, in reality, fishes are more likely to respond to multiple metrics. The process in ELOHA suggests that multiple factors of the natural flow regime are important but univariate analyses are typically easier to interpret. Fish respond to a variety of hydrologic metrics because they are influenced by multiple factors of the natural flow regime. For example, one would expect fish that need clean gravel in relatively shallow-water habitats to spawn might respond to high-flow events (responsible for the overall conditioning of spawning sites) but also to normal low-flow periods where the areas where they spawn have sufficient water quality and quantity for egg survival. Because of the expected biological response to multiple aspects of the flow regime, we suggest a series of more complex, multi-variable analyses would better represent the importance of different aspects of the flow regime. For example, multiple regressions including quadratic terms where appropriate to identify possible thresholds in these data would be helpful for making management decisions. Feedback from the stakeholder group associated with the GCP LCC project indicated univariate approaches, including simple regression and quantile regression were of interest in this study due to their common use for examining flow responses and relatively simple interpretation as related to management actions. However, there are approaches that can be used to easily interpret, at a minimum, two variable responses, where the interaction between two metrics could be visualized (e.g., surface or biplots, Figure 14).

Development of Regional Flow-Ecology Hypotheses

The suite of flow-ecology hypotheses has proven to be an important resource for the region even though the majority of the hypotheses could not be tested statistically. They are supported by scientific literature and best professional judgment (Davis and Brewer 2014). Since these flow-ecology relationships are qualitative, their use in setting quantitative instream-flow standards or guiding specific water-management practices is limited until quantitatively tested. However, all of the regional flow-ecology hypotheses were useful in identifying flow-sensitive species and guilds. Distributions of these species can be identified, which indicates where flow alterations could have negative impacts. Alterations of ecologically significant components of the flow regime due to human influences (e.g., summer low flow duration, frequency of high flow pulses, and magnitude of overbank flows) can be assessed and areas identified where flow-sensitive species are threatened. The source of the flow alteration (e.g., impervious surfaces, water use, and reservoirs) can also be identified and managed in the appropriate program (e.g., regulatory, dam management, or conservation). This information is useful for many purposes. For example, it is useful for evaluating potential ecological impacts of a proposed water withdrawal, assessing the status of instream flows in a state water management plan, or in regional conservation planning efforts (*See the Instream Flow Resources on the SARP website*⁸). In addition, the hypotheses serve as an educational tool for understanding the dependence of riverine biota on all aspects of the flow regime.

Other efforts that have followed the ELOHA framework usually develop flow-ecology hypotheses at a watershed (Apse and Kendy 2012) or state (NC EFSAB 2013) scale. The results of this study help define the usefulness of regional flow-ecology hypotheses. First, these relationships apply to broad ecologically-relevant areas beyond political or watershed boundaries. This makes development of the flow-ecology hypotheses applicable to large areas but also benefits from using a broader pool of collected data (i.e., data from multiple state agencies). The region to which each hypothesis applies can be investigated during the statistical analyses. We demonstrated that classifying the data by regions improved data availability and also the derived relationships.

The development and statistical testing of additional flow-ecology hypotheses would be beneficial to water resource management recommendations made in this region. We suggest focusing on hypotheses that can be tested and can support science-based decisions. The ecological and hydrologic metrics need to be relevant to management decisions. For example, generating hypotheses that use departure from mean annual discharge parameters, while showing significant relationships to biota, may not be the optimal metrics if the results are anticipated to be used to develop management strategies. For example, how would an agency set limits on the degree of alteration of mean annual discharge value that would be ecologically meaningful to a species? It may also be more beneficial to link fitness (e.g., survival, growth or reproduction) of stream fishes, or other biota, to flow alteration. Although we did develop some hypotheses that focused on fitness, effort in this region has not been directed toward collecting data based on

⁸ <http://southeastaquatics.net/sarps-programs/sifn/instream-flow-resources>

fitness responses. Additionally, developing population-level responses (e.g., population models) that could be linked to flow alteration would be especially helpful. Some of these suggestions may require a different approach that may *complement*, but not fit directly, within the existing ELOHA framework. However, either approach (linking fitness or abundance) would fit into a structured decision framework where decisions could be made and revisited as uncertainty about those decisions is reduced.

Conclusions

The results of these efforts show that available ecological and hydrologic data can be used to support the development of science-based regional flow-ecology hypotheses. However, due to the limitations found in our analyses, we suggest improving the available sources of information would advance instream-flow science in the GCP LCC region. The ecological data were limited in type and extent. However, assessing ecological responses to flow alteration from a regional perspective would benefit from a more comprehensive exploration of existing data before resources are directed to additional data collection. Future efforts to test flow-ecology hypotheses will benefit from the expanded fish-community data from Texas and Louisiana. However, the fish-assemblage data are also limited by the lack of sampling and data reporting protocols. Future sampling efforts should be coordinated with an effort to define and implement the necessary protocols to improve the use of the data. It may be beneficial to collect current fish data using an efficient methodology and completing spatial replication over regions of different flow alterations (rather than using historic data). In addition, the development of the regional hydrologic model by the U.S. Geological Survey holds promise for expanded use of existing fish-assemblage data on ungaged streams. Assessment is needed of how errors from different sources of hydrologic data transfer to applications based on different hydrologic model output.

Table 1. U.S. Geological Survey stream gages used to test the GCPLCC regional flow-ecology hypotheses.

State	Stream name	USGS Gage Id	Reference Period		Altered Period		Fish sampling
			Start date	End date	Start date	End date	
LA	Pearl River	02489500	1940	1960	1985	2005	2005
LA	Bogue Chitto River	02492000	1940	1960	1967	1987	1987
LA	Tensas River	07369500	1940	1960	1979	1999	1999
LA	Big Creek	07373000	1942	1962	1965	1985	1985
LA	Tchefuncta River	07375000	1944	1963	1965	1972	1972
LA	Natalbany River	07376500	1943	1963	1963	1979	1979
LA	Amite River	07377000	1951	1970	1970	1990	1990
LA	Comite River	07377500	1943	1958	1958	1973	1973
LA	Amite River	07378500	1940	1960	1960	1980	1980
LA	Calcasieu River	08013000	1943	1963	1965	1981	1981
LA	Calcasieu River	08013500	1940	1960	1989	2009	2009
LA	Ouiska Chitto Creek	08014500	1940	1960	1988	2008	2008
LA	Calcasieu River	08015500	1939	1957	1962	1981	1981
LA	Sabine River	08028500	1940	1960	1983	2003	2003
LA	Sabine River	08030500	1940	1960	1983	2003	2003
OK	Cimarron River	07154500	1951	1970	1985	2005	2005
OK	Cimarron River	07156900	1966	1985	1989	2009	2009
OK	Crooked Creek	07157500	1943	1962	1989	2009	2009
OK	Cimarron River	07158000	1940	1960	1989	2009	2009
OK	Cimarron River	07159100	1974	1993	1995	2009	2009
OK	Cimarron River	07160000	1940	1960	1989	2009	2009
OK	Verdigris River	07171000	1940	1960	1988	2008	2008
OK	Caney River	07175500	1946	1983	1989	2009	2009
OK	Verdigris River	07176000	1940	1960	1990	2010	2010
OK	Bird Creek	07176500	1946	1965	1986	2006	2006
OK	Neosho River	07185000	1940	1960	1990	2010	2010
OK	Spring River	07188000	1940	1960	1989	2009	2009

OK	Illinois River	07195500	1956	1967	1967	1979	1979
OK	Flint Creek	07196000	1956	1976	1982	2004	2004
OK	Illinois River	07196500	1940	1960	1961	1981	1981
OK	Baron Fork	07197000	1949	1968	1985	2005	2005
OK	Little River	07230000	1953	1965	1972	1992	1992
OK	Little River	07231000	1943	1962	1965	1987	2009
OK	Beaver River	07234000	1940	1960	1986	2006	2006
OK	North Canadian River	07237500	1940	1960	1985	2005	2005
OK	North Canadian River	07238000	1947	1966	1989	2009	2009
OK	Deep Fork	07243500	1940	1960	1990	2008	2008
OK	Salt Fork Red River	07300500	1940	1960	1984	2004	2004
OK	North Fork Red River	07301500	1945	1962	1985	2005	2005
OK	East Cached Creek	07311000	1940	1960	1970	1989	1989
OK	Washita River	07316500	1940	1960	1972	1992	1992
OK	Washita River	07325000	1940	1960	1985	2005	2005
OK	Cobb Creek	07325800	1969	1983	1983	1998	1998
OK	Cobb Creek	07326000	1940	1960	1974	1994	1994
OK	Washita River	07328500	1940	1960	1986	2006	2006
OK	Washita River	07331000	1940	1960	1984	2004	2004
OK	Blue River	07332500	1940	1960	1985	2005	2005
OK	Muddy Boggy Creek	07334000	1940	1960	1970	1990	1990
OK	Kiamichi River	07335700	1966	1985	1985	2005	2005
OK	Kiamichi River	07335790	1981	1995	1996	2009	2009
OK	Kiamichi River	07336200	1973	1987	1990	2005	2005
OK	Glover River	07337900	1962	1981	1987	2007	2007
OK	Little River	07338500	1947	1965	1988	2008	2008
OK	Mountain Fork	07339000	1940	1960	1989	2009	2009

Table 2. Ecological flow components used in the GCPLCC flow-ecology hypotheses. IHA metrics in bold were those used to test flow-ecology hypotheses. IHA metrics are defined in the IHA User’s Manual.⁹ Flow-sensitive species codes refer to species identified in the flow-ecology hypotheses in Chapters 3-11 of Davis and Brewer 2014.

Ecological Flow Component	Flow Attribute	Index of Hydrologic Alteration metrics	Flow-Sensitive Species Codes *
Extreme low flow ¹⁰	Magnitude	Median 7-day minimum cfs; Jun-Sep (c)	M.1
	Duration	Median number of days; per year	F.4
		Median number of days; Jun-Sep	M.1
		Median number of days; Mar-Oct	R.1
Base flow	Magnitude	Median cfs; Jun-Aug (g)	F.2
		Median monthly cfs; Jun-Aug (f,h)	
		Median 7-day minimum cfs; Jun-Aug	
		Median cfs per year (a)	F.2
		Median monthly cfs per year	
		Mean cfs per year (e)	F.4, F.6
		Mean monthly cfs per year	
		Median 7-day minimum cfs; Mar-Apr	F.6
Median monthly cfs; Mar-Apr			

⁹ <https://www.conservationsgateway.org/Files/Pages/indicators-hydrologic-alt.aspx47.aspx>

¹⁰ Note: IHA cannot be used to compare monthly EFC values for reference and altered periods.

	Durations	Median daily low flow number of days; Jun-Sep	F.5
		Median monthly low flow number of days; Jun-Sep	
	Variability	Median daily flow coefficient of variation; Jun-Aug	F.2
		Median monthly coefficient of dispersion, Jun-Aug	
High flow pulse (< 1.5 return)	Magnitude	Median annual 3-day maximum cfs	F.6, B.1
	Duration	Median number of days; Jun-Aug	B.1
	Frequency	Median count; Apr-Jun	F.3, F.4
		Median count per year	R.1
Overbank flow (>1.5 yr return)	Magnitude	Median daily cfs; Apr-Jun	F.1
		Median monthly cfs; Apr-Jun	
		Median 7-day maximum cfs; Apr-Jun	
	Frequency	Median count per year	F.6, R.1
	Timing	Median ordinal date; Apr-Jun	F.1

* **Flow-Sensitive species:**

Fish – Alligator gar (F1); Arkansas River (F2); Freckled madtom (F3); Guadalupe bass (F4); Orangethroat darter (F5); and Suckers (F6) Mussels – Steamboat mucket (M1)

Birds – Interior Least Tern (B1)

Vegetation – Riparian woody species (R1)

Table 3. Ecological response metrics associated with species, guilds, and biotic assemblages used in the GCP LCC Flow-Ecology Hypotheses. The Flow-Ecology Hypotheses Codes refer to the hypotheses found in Chapters 3-11 in Davis and Brewer 2014.

Ecological Condition	Category	Ecological metric	Flow-Ecology Hypotheses Codes*	
Habitat	Availability	Nest habitat (Weighted Useable Area)	F.3.b	
	Amount	Spawning habitat (acres)	F.1.b	
	Condition		Habitat quality	F.4.b, F.6.a
			Depth to groundwater	R.1.a
		Formation	Acre/year	B.1.a
			Sediment deposition rate	R.1.f
Population	Presence	Persistence	F.2.c	
	Abundance	Catch per Unit Effort	F.2.b, F.3.a, F.5.a	
	Invasive	Number of individuals	R.1.b, R.1.g, R.1.i	
Physiological	Body condition	Catabolism	M.1.a	
		Weight/Length	F.6.d	
Reproduction	Nesting success	Number per year	B.1.a	
	Reproductive success	Number of YOY	F.1.a, F.2.a, F.4.c	
	Seed dispersal	Percent of area	R.1.h	
Growth	YOY and Juvenile	Growth rate (cm/yr)	F.4.a, F.6.b	
	Seedling roots			
	Trees	Height (cm/yr)	R.1.e	
Survival	Young	Number of individuals	F.6.c, R.1.d	
Mortality	Adults	Number per year	M.1.b, R.1.c	

* **Flow-Sensitive species:** Fish – Alligator Gar (F1), Arkansas River Shiner (F2), Freckled Madtom (F3), Guadalupe Bass (F4), Orangethroat Darter (F5), and suckers (F6); Mussels – Steamboat Mucket (M1); Birds – Interior Least Tern (B1); Vegetation – riparian woody species (R1)

Table 4. Test statistics for simple regression analysis. DF is degrees of freedom. The regression analysis tested the hypothesis that relative abundance of pelagic broadcast spawning cyprinids would decrease with decreases in baseline flow variability during the spawning months (June-August).

Model	Source	DF	Sum of Squares	Mean Square	F-value	P-value	R²	Adj R²
1	model	1	1287.46	1287.48	97.19	<0.01	0.58	0.57
	error	73	967.01	13.25				
	corrected total	74	2254.46					

Table 5. Parameter estimates for simple regression Model 1 with degrees of freedom (DF), standard error (SE), and significance value. The regression analysis tested the hypothesis that relative abundance of pelagic broadcast spawning cyprinids would decrease with decreases in baseline flow variability during the spawning months (June-August).

Model	Variable	DF	Parameter Estimate	SE	P-value
1	Intercept	1	-2.31	0.68	<0.01
	CV June-Aug	1	33.09	3.36	<0.01

Table 6. Parameter estimates for quantile regression (model 1 and model 2): Results are provided for median, 25th and 75th quantiles. Degrees of freedom (DF), standard error (SE) and 90% confidence limits (CL) are provided. Model 1 tested the hypothesis that relative abundance of pelagic broadcast spawning cyprinids would decrease with decreases in baseline flow variability during the spawning months (June-August). Model 2 tested the hypothesis that the abundance of lithophilic fishes would decrease with baseflow duration.

Model	Quantile Test	Parameter	DF	Estimate	SE	90% CL	T-value	P-value
1	50	intercept	1	-2.59	2.45	-6.67-1.49	-1.06	0.29
		b	1	26.75	17.14	-1.79-55.31	1.56	0.12
	75	intercept	1	-4.69	2.54	-8.92--0.46	-1.85	0.07
		b	1	55.07	17.09	26.59-83.55	3.22	0.002
	90	intercept	1	-3.89	3.29	-9.37-1.59	-1.18	0.24
		b	1	61.52	21.19	26.21-96.84	2.9	0.004
2	50	intercept	1	3.20	3.79	-3.21-9.61	0.84	0.4
		g	1	78.78	12.58	57.55-100.01	6.26	<0.01
	75	intercept	1	19.85	10.48	2.15-37.54	1.89	0.07
		g	1	64.62	35.53	4.67-124.57	1.82	0.08
	90	intercept	1	46.82	21.19	11.06-82.58	2.21	0.03
		g	1	29.07	110.35	-157.10-215.24	0.26	0.79

Table 7. Wald and likelihood ratio test statistics for two quantile regressions (Model 1 and Model 2). Model 1 tested the hypothesis that relative abundance of pelagic broadcast spawning cyprinids would decrease with decreases in baseline flow variability during the spawning months (June-August). Model 2 tested the hypothesis that the abundance of lithophilic fishes would decrease with baseflow duration.

Model	Quantile test	Test Statistic	DF	Chi-Square	P-value
1	0.5	Wald	1	2.44	0.11
	0.5	Likelihood	1	48.22	<0.01
		Ratio			
	0.75	Wald	1	10.38	<0.01
	0.75	Likelihood	1	34.78	<0.01
		Ratio			
	0.9	Wald	1	8.43	<0.01
	0.9	Likelihood	1	26.34	<0.01
Ratio					
2	0.5	Wald	1	39.22	<0.01
	0.5	Likelihood	1	28.83	<0.01
		Ratio			
	0.75	Wald	1	3.31	0.07
	0.75	Likelihood	1	8.59	<0.01
		Ratio			
	0.9	Wald	1	0.07	0.79
	0.9	Likelihood	1	1.93	0.16
Ratio					

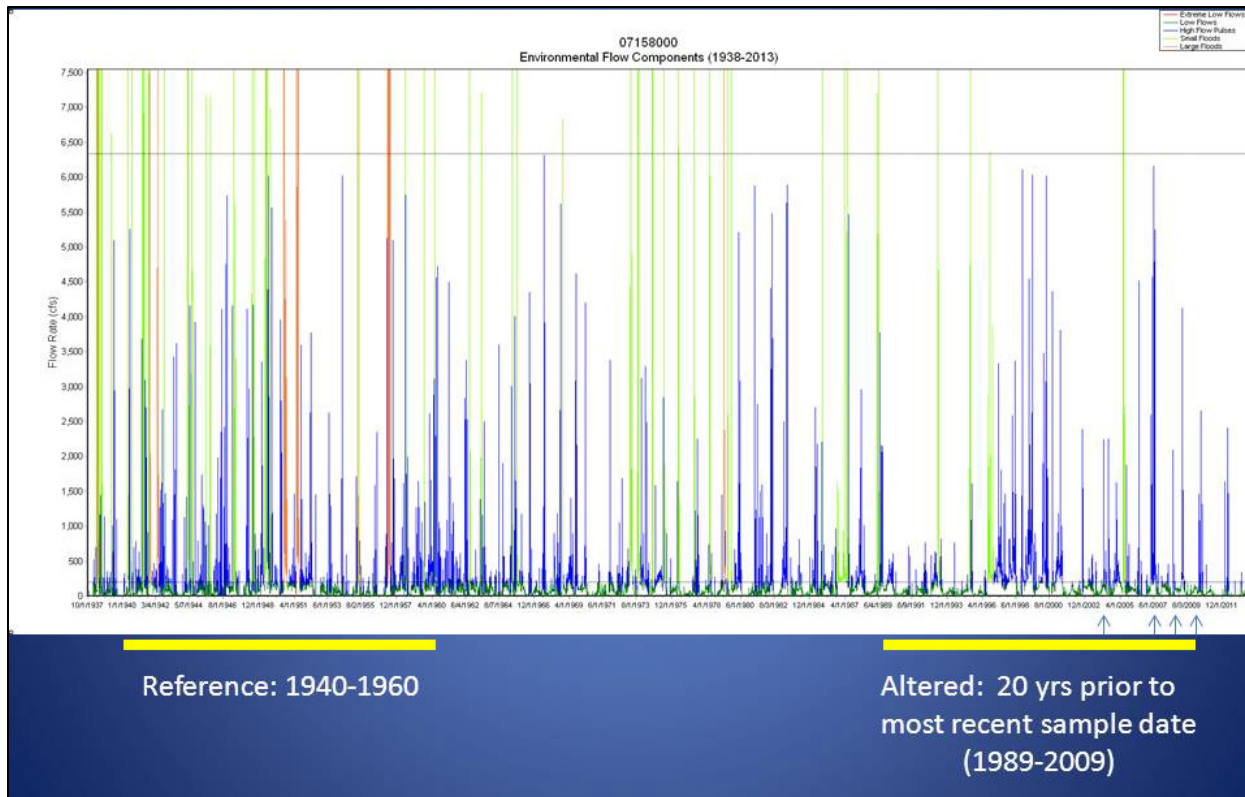


Figure 1. Daily discharge over the period of record for a USGS gage used in calculation of hydrologic metrics. The yellow bars below the graph indicate example reference and altered time periods used to test the GCP LCC flow-ecology hypotheses. Different colors indicate different ecologically significant portions of the flow regime. See text for more information.

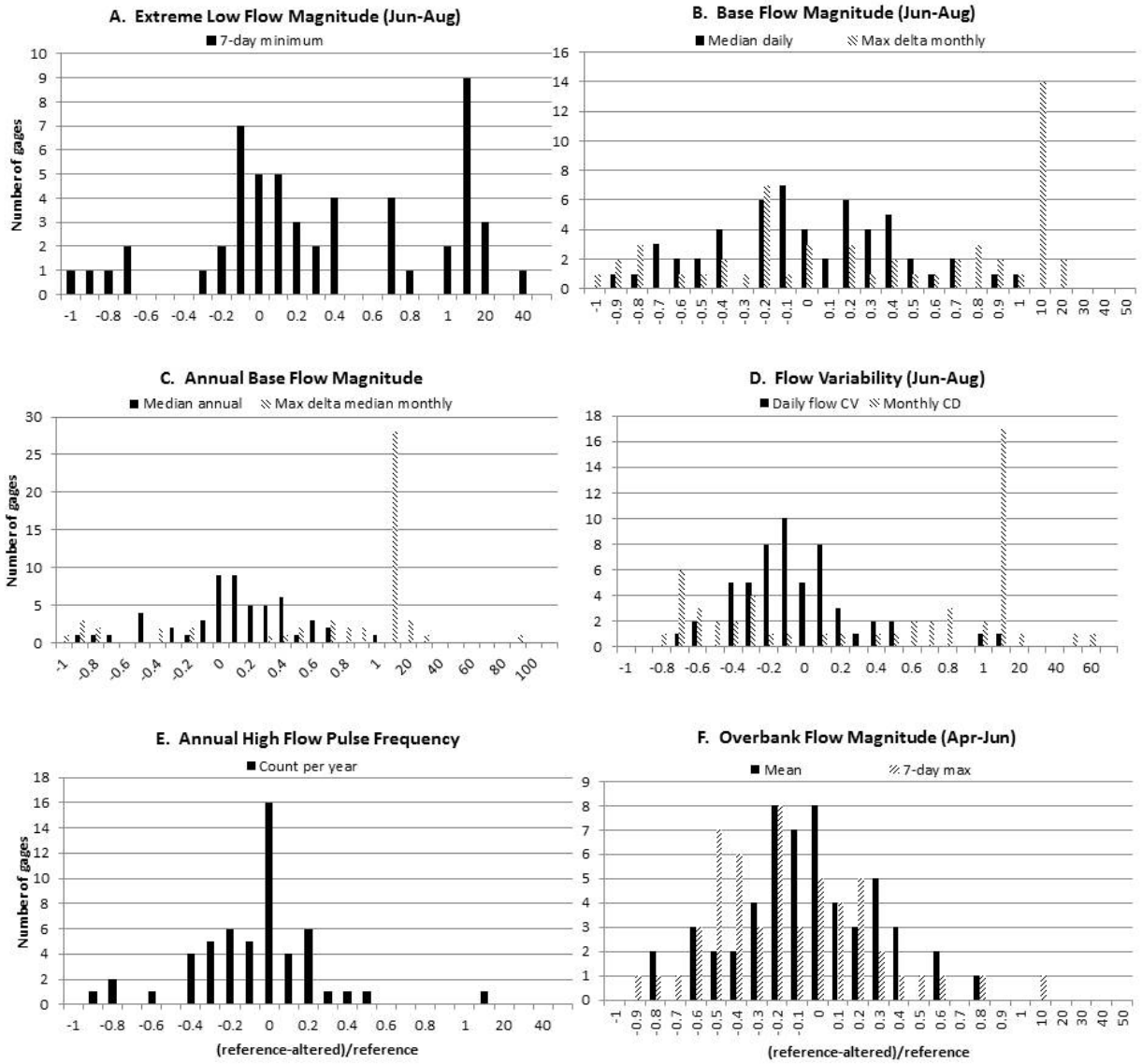


Figure 2. Distribution of degree of flow alteration calculated for reference and altered time periods using select USGS gage records. Note the change in scale for x-axis values >1.0, where flow conditions increased greater than 100% over reference conditions.

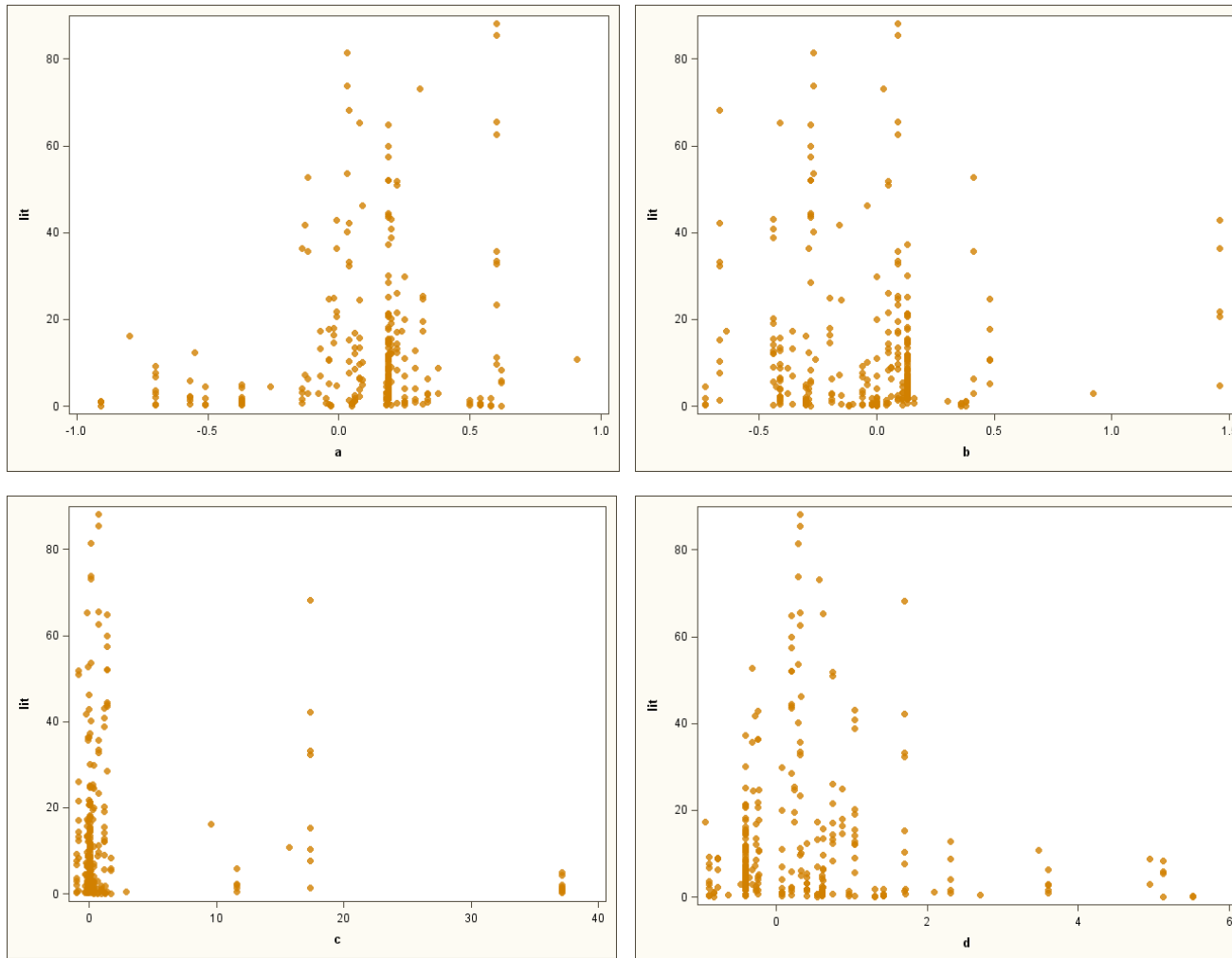


Figure 3. Scatterplot showing relative abundance of lithophilic fishes (lit) as related to different hydrologic metrics (a= median annual flow, b= daily CV (June-Aug), c=7 d minimum (June-Sept), d= April-June median monthly flow). Negative values on the x-axis indicate a decrease and positive values an increase, respectively, in the metric relative to reference conditions.

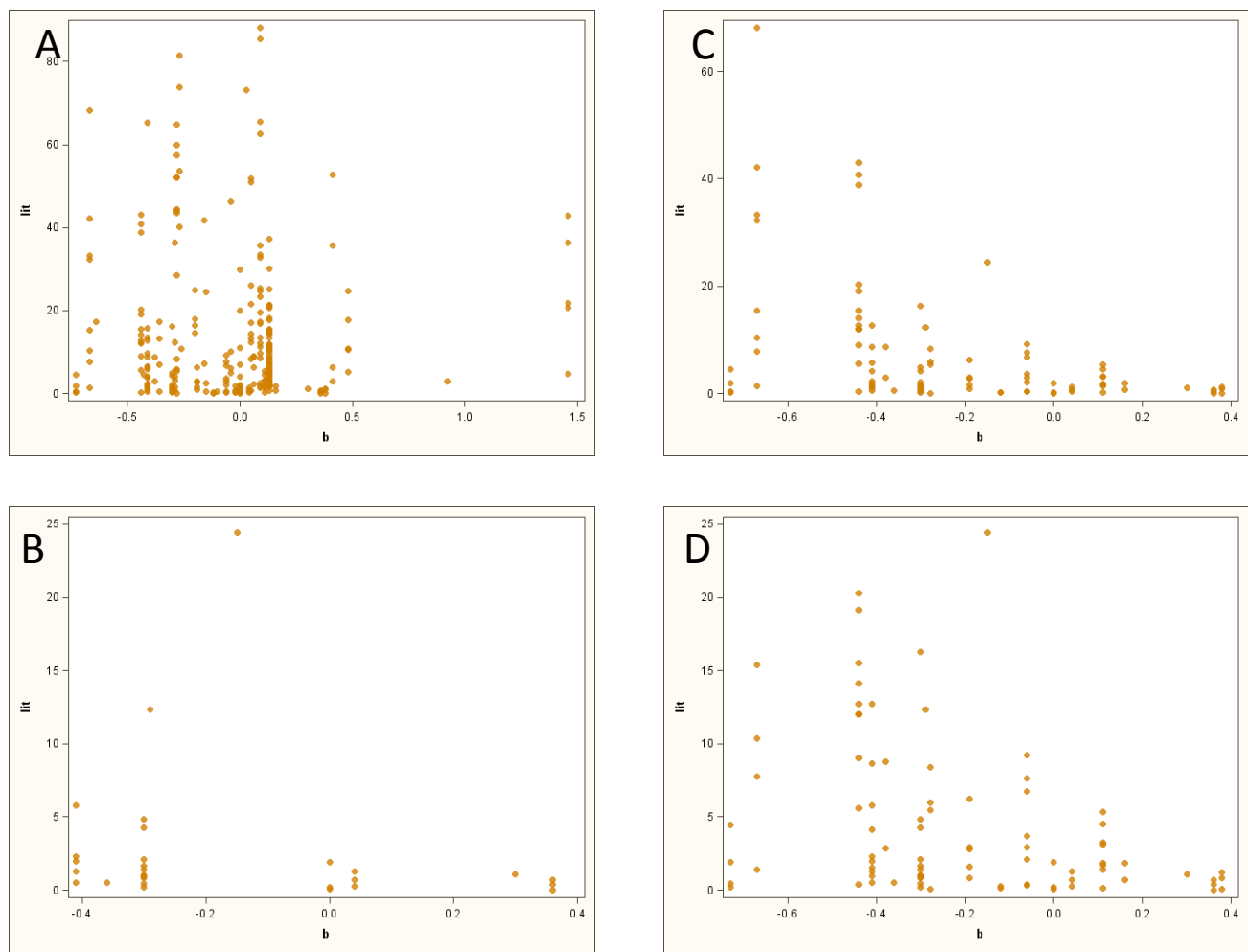


Figure 4. Scatterplot of lithophilic spawner (lit) relative abundance and daily flow coefficient of variation (June-Aug). Panels represent: A= all data, B= EPA Great Plains Ecoregion, C= combined ecoregion of the Southern Plains, and D= C with omitted relative abundance > 25. Negative values on the x-axis indicate a decrease and positive values an increase, respectively, in the daily flow coefficient of variation relative to reference conditions

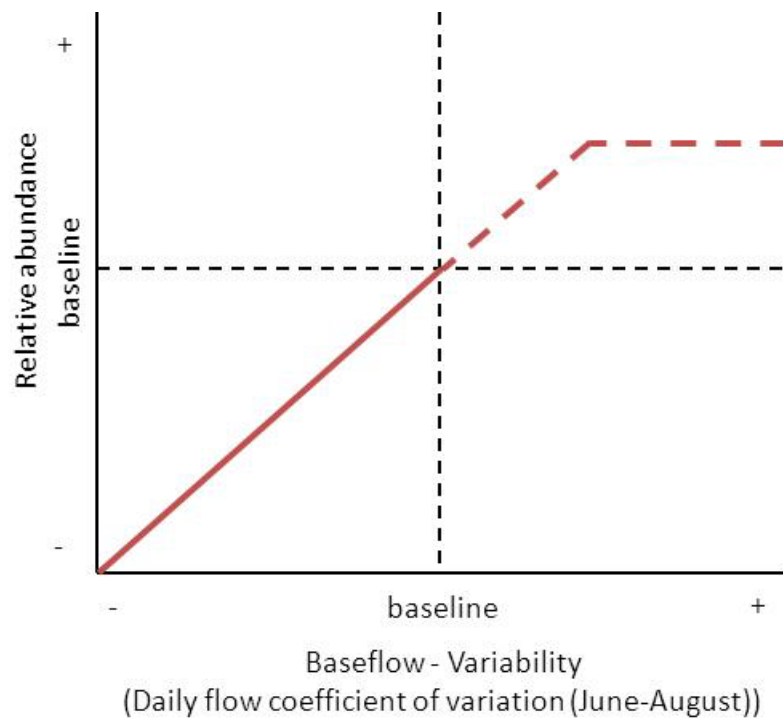


Figure 5. Regional Flow-Ecology Hypothesis for Arkansas River Shiner. Abundance of Arkansas River Shiner (ARS) will decrease with decrease in baseline flow variability during spawning months (June-August) due to mechanisms that are not known. An increase in abundance with increases in flow variability above baseline may result in above baseline abundance of ARS (Worthington and Brewer 2014).

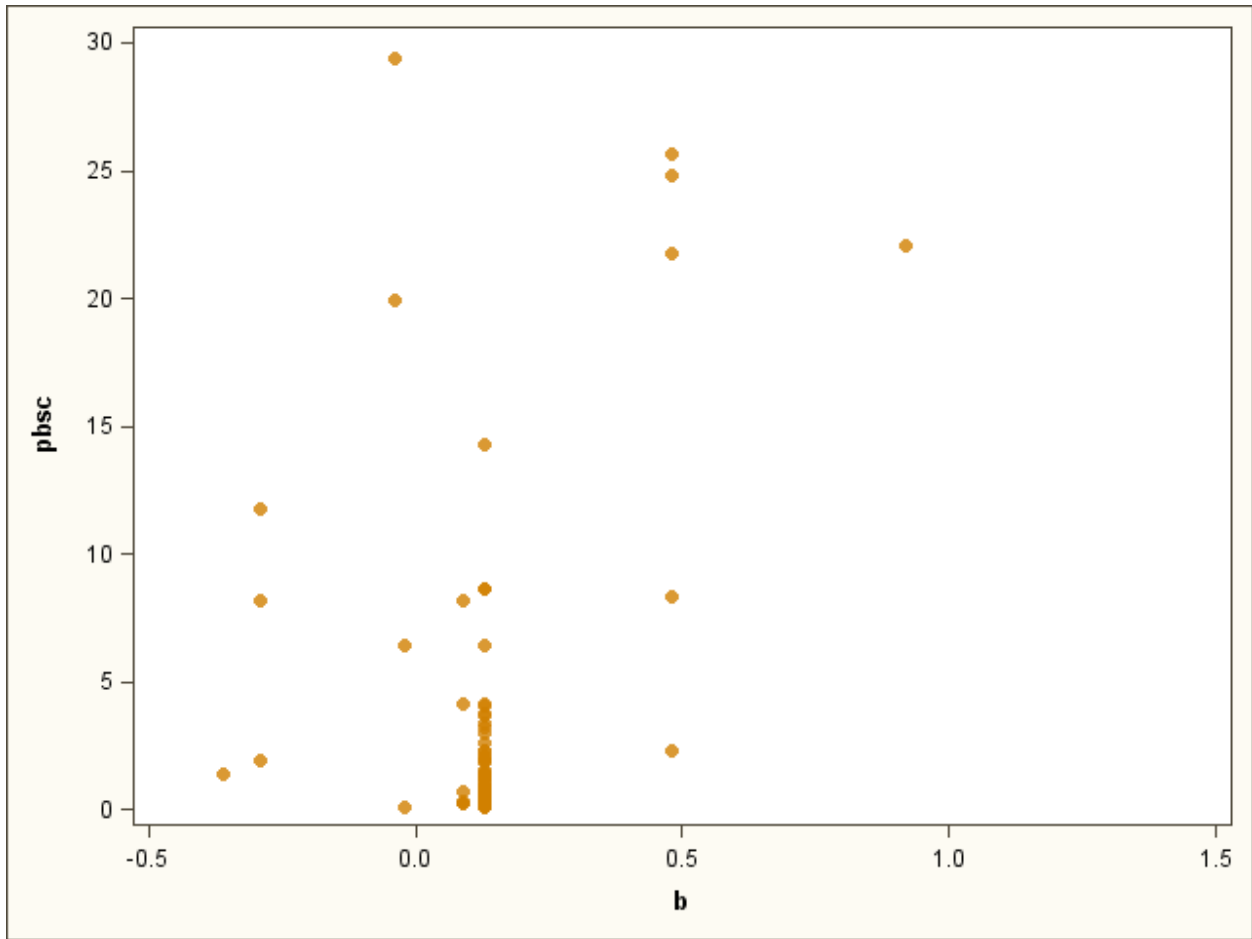


Figure 6. Relative abundance of pelagic broadcast spawning cyprinids (pbsc) related to the degree of alteration of the daily coefficient of variation of discharge from June-August (b) in the Southern Plains Ecoregion.

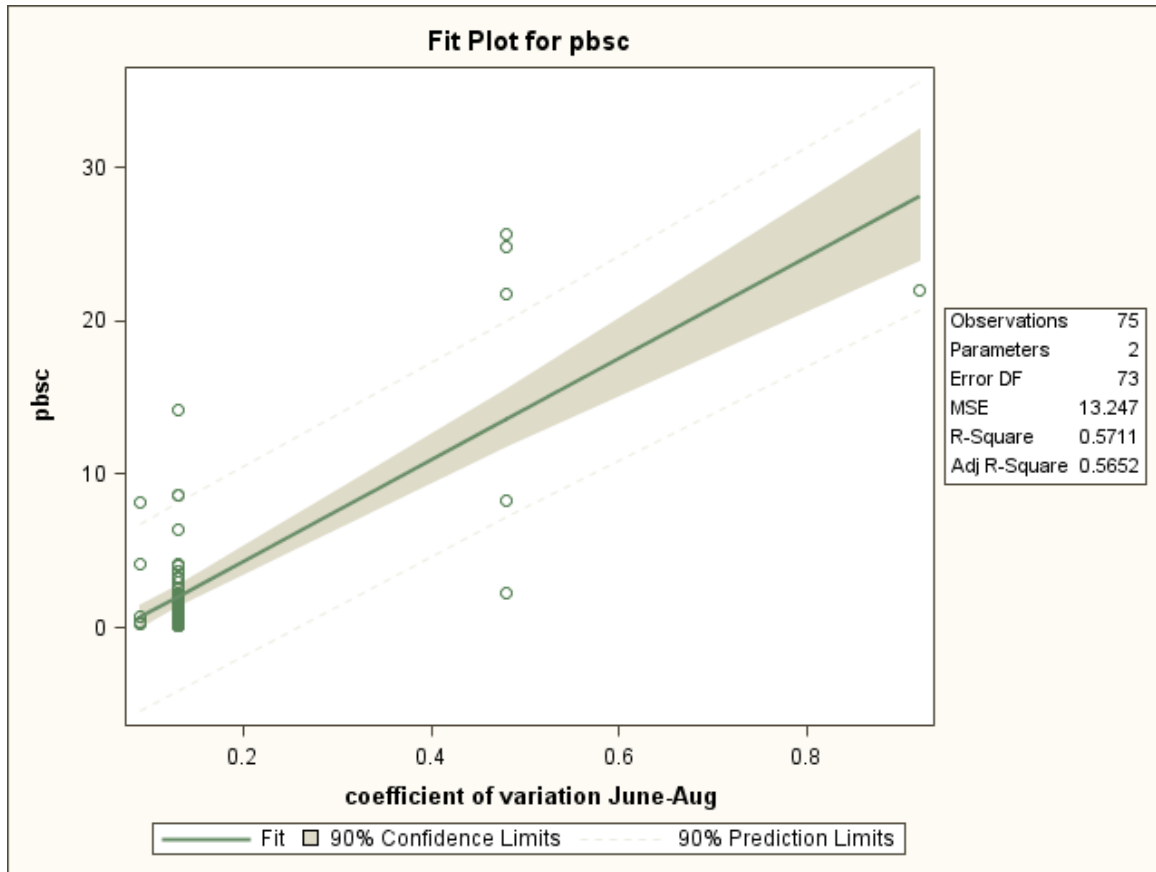


Figure 7. Simple linear relationship between the relative abundance of pelagic broadcast spawning cyprinids (pbsc) and the degree of alteration of the daily coefficient of variation of discharge from June-August.

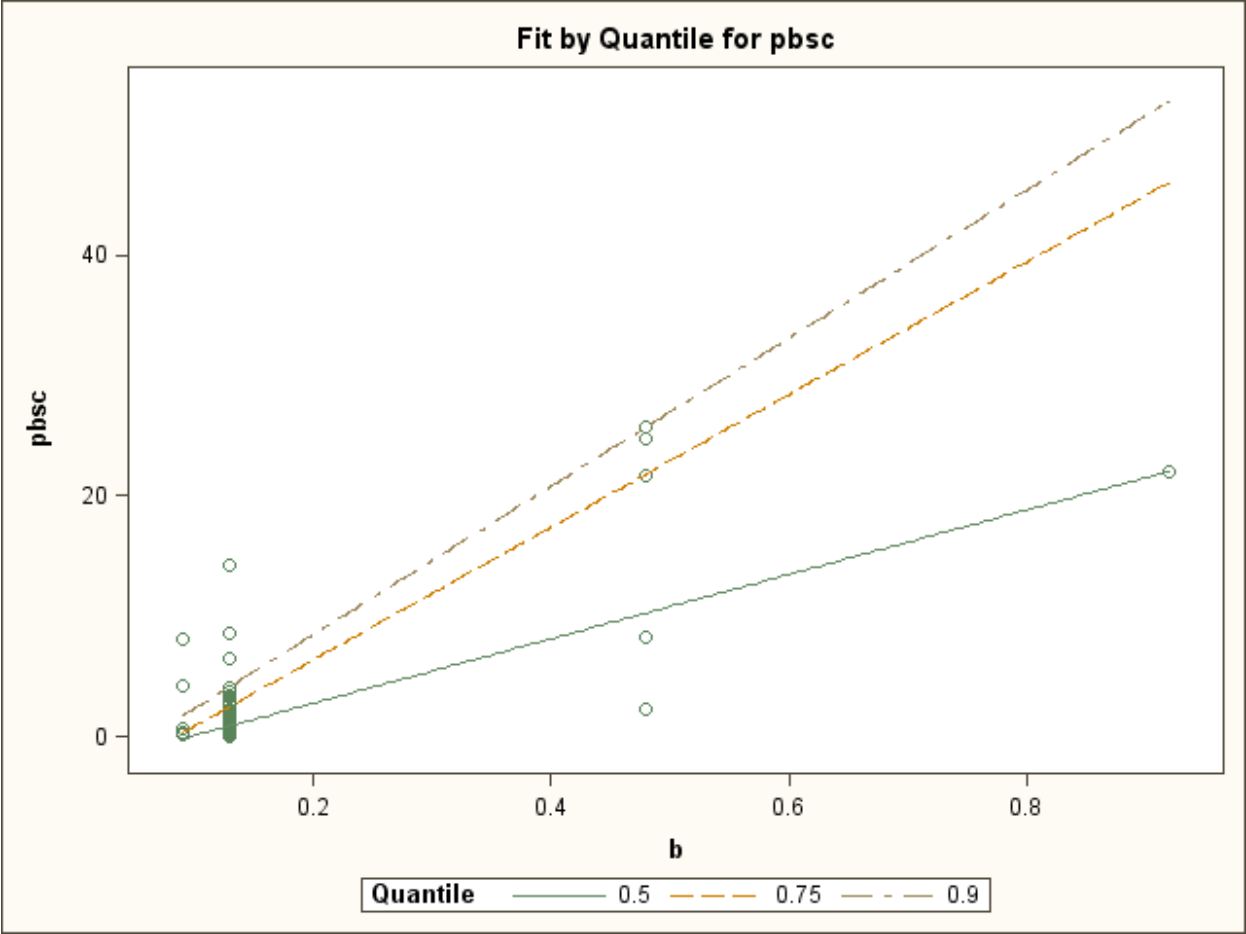


Figure 8. Simple quantile regression showing the relationship between the relative abundance of pelagic broadcast spawning cyprinids (pbsc) and the degree of alteration of the daily coefficient of variation of discharge from June-August. The model was fit using the simplex algorithm in SAS.

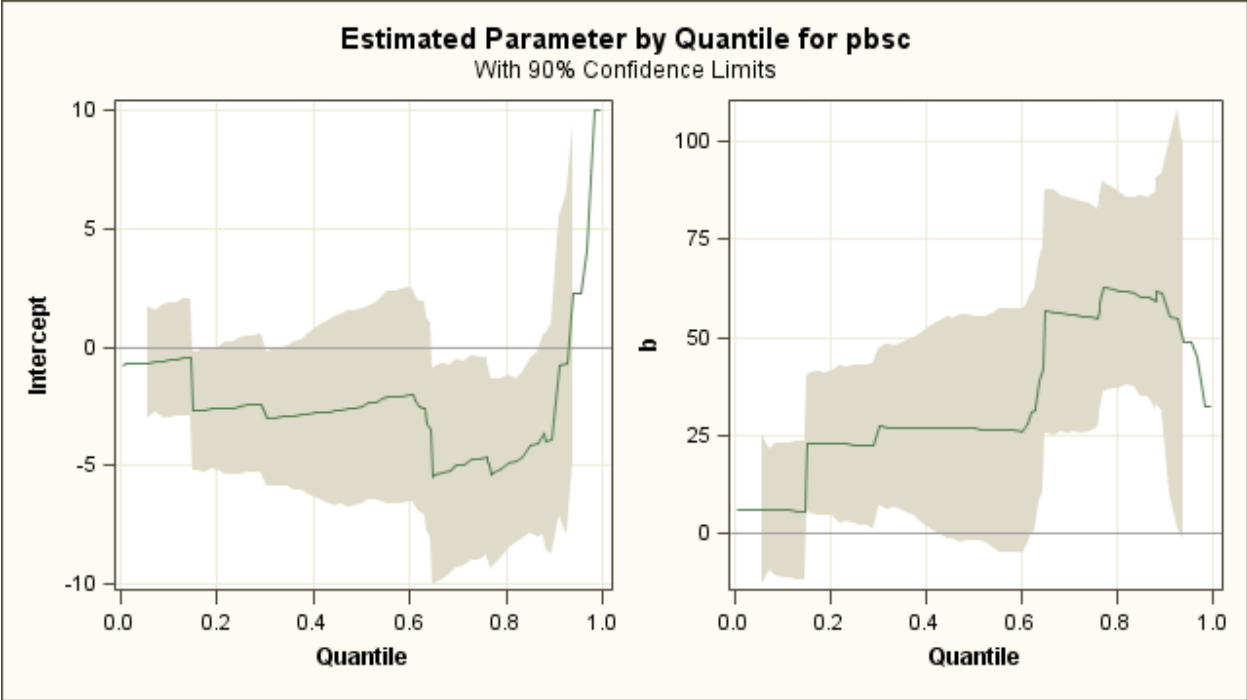


Figure 9. Relationship between the regression intercept and slope parameter (b) to quantiles. Notice the sharp increase in the slope parameter at about 0.6 where the slope parameter begins to differ from zero.

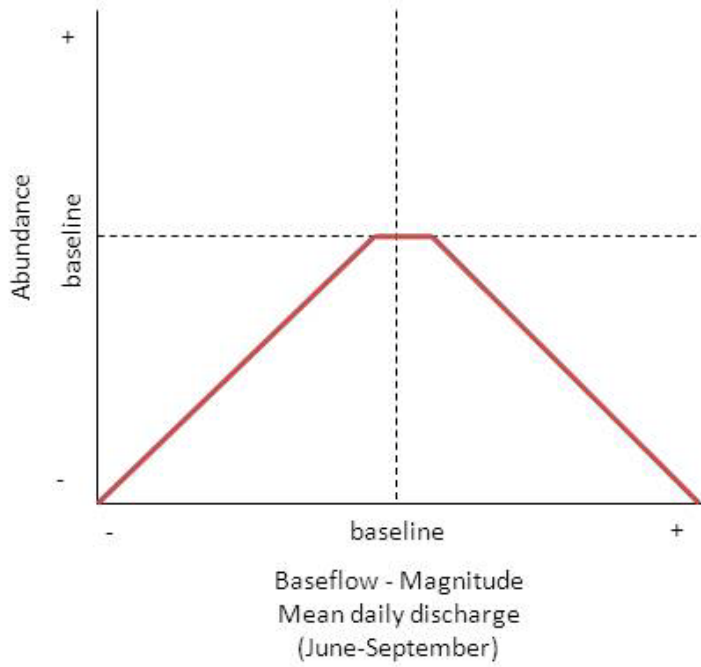


Figure 10. Orangethroat Darter Baseflow Magnitude Hypothesis: The abundance of Orangethroat Darter declines with decreased monthly low-flow magnitude during the summer as water temperatures increase, aeration of substrates decrease, and availability of habitat declines. Increases in monthly low-flow magnitude during summer may result in decreased abundance due to competition with non-native species (Taylor et al. 2014).

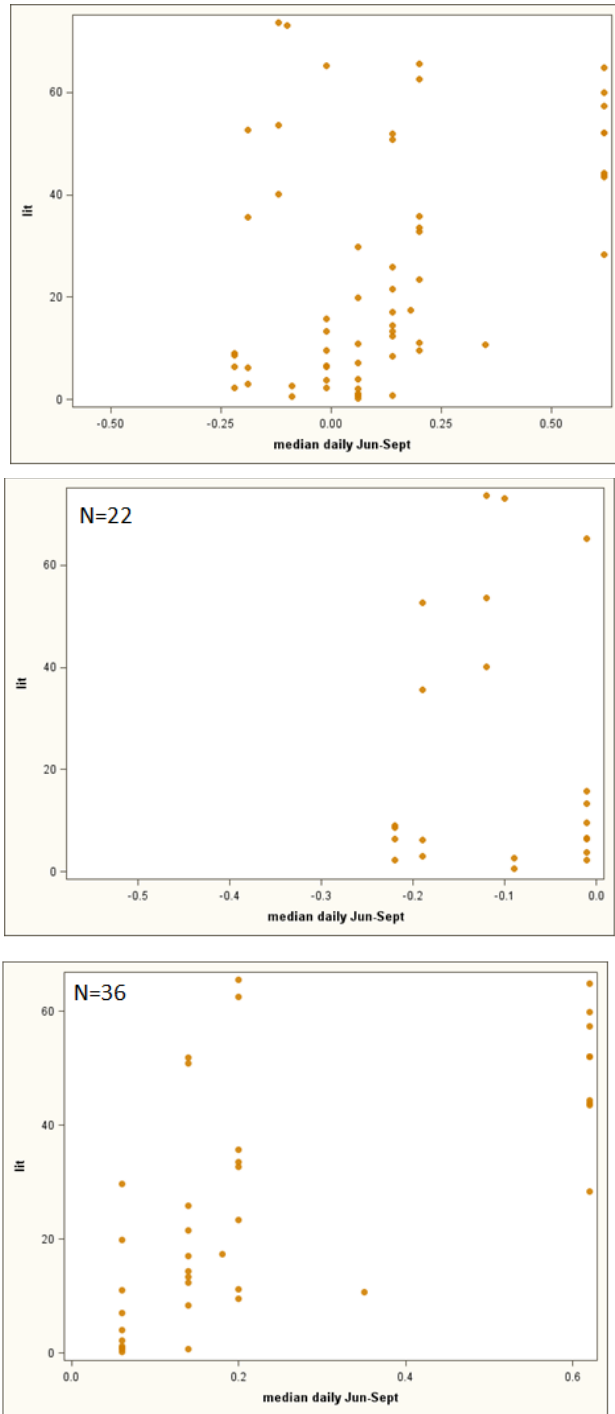


Figure 11. Scatterplot of data showing the relationship between relative abundance of lithophilic fishes (lit) and median daily discharge (June-September). The upper panel shows all data whereas the lower two only show increases or decreases of the hydrologic metric from baseline.

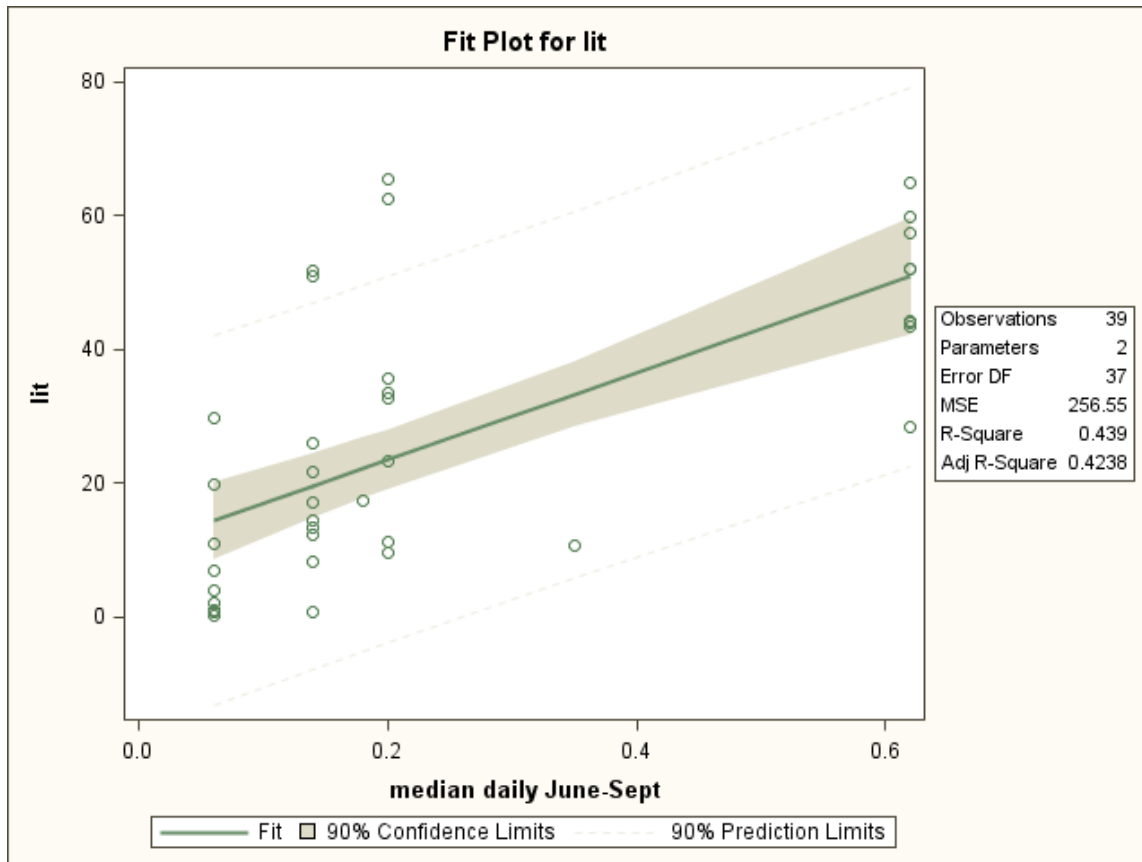


Figure 12. Linear regression and 90% confidence limits around the relationship between relative abundance of lithophilic fishes (lit) and median daily discharge (June-September). Parameter estimates and R square provided on the figure.

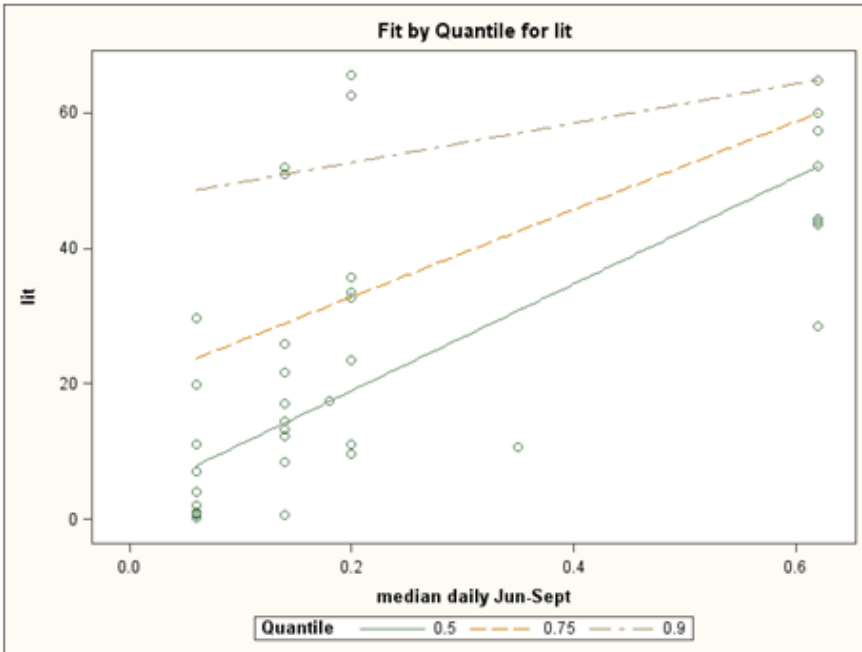


Figure 13. Quantile regression for median, 75th and 90th quantiles showing the relationship between relative abundance of lithophilic fishes (lit) and median daily discharge (June-September). Parameter estimates provided in Table 5.

Ecoregion 3 Lithophyl RA

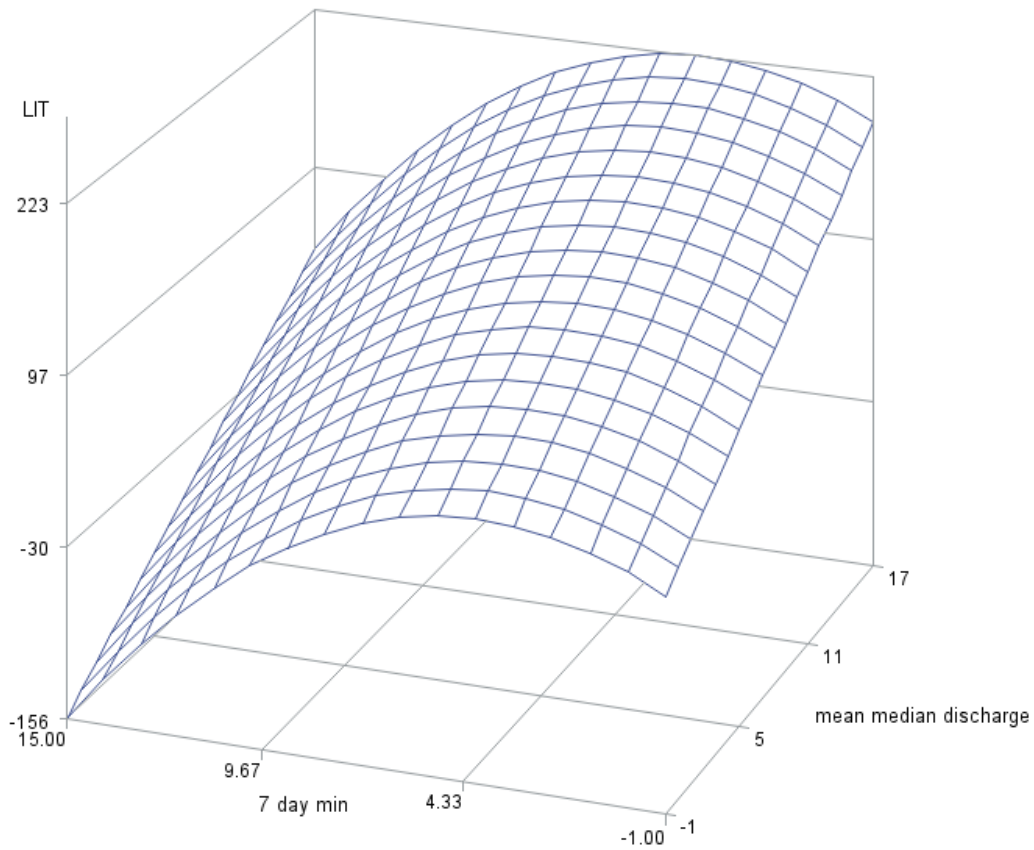


Figure 14. Surface plot showing the relationship among two hydrologic variables and the relative abundance of lithophilic -spawning species. This figure should not be interpreted to be ecologically meaningful but is simply an example of a biplot for testing two variable flow-ecology hypotheses.

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Appendix A: Default parameters used in IHA to calculate alteration of hydrologic metrics to test the GCP LCC flow-ecology hypotheses.

The following parameters were used to set up the IHA analysis project:

1. Analysis years: Gage-specific 20 year period for reference and altered time periods
2. Analysis days: Hypothesis-specific days of the year (e.g., April 1 – June 30)
3. Statistics: Non-parametric, where the high flow and low flow pulse thresholds are the median plus or minus 25%
4. Environmental Flow Components:
 - For the initial high flow/low flow separation, all flows that exceed 75% of the daily flows for the period will be classified as high flows. All other flows are classified as low flows.
 - A small flood event is defined as an initial high flow with a peak flow greater than 1.5 year return interval event
 - A large flood event is defined as an initial high flow with a peak flow greater than 10 year return interval event
 - An extreme low flow is defined as an initial low flow below 10% of the daily flows for the period. All initial low flows not classified as extreme low flows are classified as low flow.