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Final Report

Potential Influence of Exchanges between the Lower Guadalupe River and Oxbow Lakes on Food Web Dynamics

Department of Wildlife and Fisheries Sciences
Texas A&M AgriLife Research
College Station, Texas

Dr. Kirk O. Winemiller, PI
Marcelo Andrade*
Caroline Arantes
Thethela Bokhutlo
Luke Bower
Eduardo Cunha**
Friedrich Keppeler
Edwin Lopez
Yasmin Quintana
David Saenz

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TWDB CONTRACTS

In collaboration with:

Texas Parks and Wildlife Department
Inland Fisheries, River Studies
San Marcos, Texas

Kevin Mayes
Clint Robertson

Initial Submission Date
October 2017

Final Project Report Dated June 2018
Texas Water Development Board Contract No. 1448311791
Texas Parks and Wildlife Department Contract No. 476296

[* visiting scientist from Universidade Federal do Pará, Brazil]

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2 of 2

**Floodplain inundation analysis of the lower Guadalupe River:
linking hydrology and floodplain-dependent resources**

Dr. Kimberly Meitzen
Dr. Jennifer Jensen

Department of Geography
Texas State University
San Marcos, Texas

Dr. Thomas Hardy

Department of Biology
Texas State University
San Marcos, Texas

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Executive Summary

The objective of this project was to contribute to understanding of flow-ecology relationships in the lower Guadalupe River by examining how lateral connectivity may facilitate exchanges of organic material, including aquatic organisms, between the river channel and oxbow lakes to influence food web dynamics. An important function of high-flow pulses in lowland rivers of Texas is the creation of lateral connections between the channel and oxbow lakes that provide important habitat for fishes. Oxbow lakes are highly productive aquatic habitats for many plants and animals and function as nursery areas for many fish species. Fish movement between the river channel and oxbows during periods of lateral connectivity can influence food web dynamics in both habitats. Food web dynamics also are influenced by recruitment dynamics of aquatic organisms that in turn are influenced by ecosystem productivity in relation to flow conditions. In the lower Guadalupe River, there are moderate increases in turbidity during high flow pulses, and the river channel remains net autotrophic (i.e., photosynthesis rate > aquatic respiration rate). According to a previous study, biomass of several common fish species within the lower Guadalupe channel has greater reliance on terrestrial sources of production during high flow pulses. What remains unknown is the extent that food web dynamics affecting fish biomass is affected by exchanges of fine particulate organic matter (of either aquatic or terrestrial origin) and aquatic organisms (fishes in particular) between the Guadalupe River channel and oxbow lakes during high flow pulses. This project, funded by the Texas Parks and Wildlife Department, was designed to reveal relationships between flow, lateral connectivity, and food-web dynamics in the lower Guadalupe River and oxbow lakes within its floodplain. Ratios of stable isotopes of carbon and nitrogen were analyzed to infer the potential for alternative basal production sources to support fish and mussel biomass in two oxbow lakes and the river channel under contrasting flow conditions. Of particular interest is evidence that fishes and mussels in the river channel had assimilated material from basal sources from oxbows, and that fishes and mussels in oxbows had assimilated material originating from the river channel. This study also analyzed spatial and temporal variation in fish assemblage structure in the river channel and oxbow lakes to evaluate between-habitat exchanges in relation to hydrology.

Environmental data were recorded (e.g., water conductivity, pH, dissolved oxygen, temperature, transparency), and fishes and aquatic macroinvertebrates were collected from two oxbow lakes and adjacent sites in the Guadalupe River channel during six surveys conducted over a 12-month period from March 2016 to April 2017. At each location during each survey, samples of fish, mussel, riparian plant leaves, seston (phytoplankton and other particles suspended in the water column), and periphyton (benthic algae and other particles deposited on submerged surfaces) were collected for stable isotope analysis. Guadalupe River discharge data for USGS (United States Geological Survey) streamflow gages nearest each oxbow (Cuero and Victoria gages) were obtained for analysis. The flows associated with establishment of lateral connectivity for each oxbow were estimated based on field observations of wetted channels that were the lowest elevations in the riverbank where oxbows connected. All but one of the six survey dates followed a high flow pulse, and the number of days with lateral connection was estimated for the preceding 15-day and 30-day intervals for each oxbow and survey date. Average values for basal sources and values for individual specimens of seven fish species and mussels (two common species) that commonly were captured from oxbows as well as the river channel were plotted on axes defined by stable isotope ratios of carbon (x-axis) and nitrogen (y-axis). Separate plots were constructed for each fish species and mussels for each survey period. Within each plot, an isospace polygon

was drawn to indicate the location of the space occupied by animals that had assimilated some feasible combination of source material from oxbows. Within the same plots, an isospace polygon also was drawn to indicate the space for animals that had assimilated a feasible combination of source material from the river channel. Vertical positions of animals in isotopic biplots (along the y-axis) indicate trophic position in food chains, and their horizontal positions indicate their likely assimilation of material from alternative sources, with closer vertical alignment above a given source indicating a higher probability and proportion of assimilation of that source. Spatial and temporal variation in the composition of local fish assemblages was analyzed based on abundance of species in survey samples. Multivariate ordination was performed in order to examine potential influences of seasonal environmental variation and hydrology on fish assemblage structure.

Stable isotope data indicated that terrestrial vegetation was the most important basal production source supporting biomass of most fishes and mussels in both oxbows and the river channel during every survey. Isospace polygons indicated that either periphyton or seston could have contributed minimal proportions to consumer biomass in some habitats during certain periods. During several survey periods, seston and/or periphyton samples contained insufficient organic material for measurement of carbon and nitrogen stable isotopes. Given the relatively high turbidity recorded throughout the field study, it appears that many seston and periphyton samples contained mostly inorganic particles, especially silt, and aquatic primary production was low in both the water column and benthos during survey periods when seston and periphyton samples lacked sufficient organic matter to obtain isotopic ratios for carbon and nitrogen. Isotopic evidence of cross-habitat exchange of basal sources or consumers was found for 7% of 316 specimens from oxbows and 11% of 231 specimens from the river channel that were analyzed. Most of the specimens that were analyzed had isotopic signatures that were either 1) consistent with assimilation of material from the habitat from which they were captured, or 2) inconclusive with regard to cross-habitat exchanges. Conclusions regarding hydrology and spatial food web subsidies based on current isotopic evidence should be considered tentative given several issues and assumptions that can affect stable isotope analysis and inferences, as well as the fact that the field study was conducted during a relatively wet period when river discharge and turbidity were higher than for an average year.

Fish assemblage structure was significantly different between oxbows and the river channel and was influenced by hydrology, but seasonal variation was not statistically significant. Fish assemblage structure was more variable in oxbows than the river channel; however, several species were more common in oxbows (e.g., shads, orangespotted sunfish, crappies), and others were more common in the river channel (e.g., burrhead chub, ghost shiner, mimic shiner, red shiner). The greatest similarity in assemblage structure of oxbow and channel fish assemblages occurred during June 30–July 1, 2016, when the surveys were conducted at the end of a large flow pulse of long duration. During that flow pulse, the oxbow near Cuero was connected to the channel for 88 days, the oxbow near Victoria was connected to the channel for 92 days, and there apparently was extensive exchange of fishes between oxbows and the river channel.

Estimates of Guadalupe River discharge (USGS gage at Cuero) that induce lateral connectivity at the oxbow near Cuero indicate that cross-habitat exchange of resources and aquatic organisms would occur for the following flow tiers within the current environmental flow standards adopted by the Texas Commission on Environmental Quality: 2-per-season pulse during spring, summer and fall; and 1-per-season pulse during all four seasons. Given the current environmental flow standards for the Victoria gage, lateral connections of the oxbow near Victoria would be achieved by the following seasonal flow pulses protected by current environmental flow standards: 2-per-

season pulse only during spring and fall; and 1-per-season pulse in all four seasons. However, even with this knowledge, it is not a simple matter to estimate the frequency and duration of lateral connections between the river channel and oxbows under future water management scenarios. This is because projected discharge under a given management scenarios depends on both environmental flow standards as well as implementation rules for regulating diversion or capture by water right holders.

Pending further investigation and evidence, the current environmental flow standards appear likely to sustain some degree of lateral connectivity for ecological processes associated with the oxbows near Cuero and Victoria, but with reduced frequency and duration depending on the degree of water diversion and capture by water right holders under current flow standard implementation rules. With regard to channel–oxbow connections, high flow pulses are probably most beneficial to aquatic organisms during spring, followed by summer and fall. The reduction in lateral connectivity during winter relative to historical long-term hydrology may have less ecological consequence, although this assessment is tentative given the limited ecological research conducted on this system to date. In the case of the oxbow near Victoria, however, the magnitude of the higher-tier flow pulse (1/season) during spring and fall may be insufficient for lateral connections. Reduced frequencies and durations of connections would particularly harmful to during seasons when lateral connectivity is most important for between-habitat exchanges of both pre-spawning adult and juvenile fishes as well as spatial food web subsidies. This project did not evaluate ecological functions of overbanking flows (none occurred during our survey period), which are not protected under the current environmental flow standards.

Introduction

The Texas Wildlife Action Plan outlines major conservation goals that include maintaining adequate water quantity and increasing the knowledge and understanding of aquatic ecosystems. Nested within those goals is the Texas Instream Flow Program (TIFP), mandated by Texas Senate Bill 2 (2001), which calls for state resource agencies to conduct instream flow studies to determine conditions to conserve Texas rivers and streams. The objective of this project was to contribute to TIFP efforts on the lower Guadalupe River by examining how lateral connectivity may facilitate exchanges of organic material, including aquatic organisms, between the river channel and oxbow lakes to influence food web dynamics.

One of the most important functions of high flow pulses in lowland rivers is creation of lateral connectivity between the river channel and aquatic habitats in floodplains. For example, in the lower Brazos River, Texas, periodic high flow pulses result in temporary connections between the river channel and oxbow lakes that provide important habitat for several fish species that normally are uncommon within the active river channel, but often attain high densities in oxbow lakes (Zeug et al. 2005). Oxbow lakes tend to be highly productive aquatic habitats that function as nursery areas for many fish species (Zeug and Winemiller 2008a). In the lower Brazos River, some fish species, including gars (*Lepisosteus* species) and shads (*Dorosoma* species), have been shown to move between the river and oxbow lakes to exploit food resources (Robertson et al. 2008, Zeug et al. 2009).

Fish movement between river channel and floodplain aquatic habitats during periods of lateral connectivity influences food web dynamics in both habitats (Winemiller 2004). Some of the small fishes that move from the river channel into oxbows are consumed by predatory fishes, thus representing a transfer of production from the river channel into the food web of aquatic floodplain habitats. Similarly, when young-of-the-year fishes migrate from oxbow lakes into the river channel,

predators residing in the channel receive a substantial subsidy from highly productive floodplain habitats (Winemiller 1996). Thus, food web dynamics affect, and are influenced by, recruitment dynamics of aquatic organisms that in turn are influenced by ecosystem productivity in relation to flow conditions. In the lower Guadalupe River, turbidity increases during high flow pulses and this has been shown to reduce net aquatic primary production (Roach et al. 2014). According to a prior study (Roach and Winemiller 2015), the biomass of several common fish species inhabiting the lower Guadalupe River channel seems to be more dependent on riparian vegetation than algae during high flow pulses.

What remains unknown is the extent that food web dynamics affecting fish biomass is affected by exchanges of particulate organic matter (algae and detritus of either aquatic or terrestrial origin) and aquatic organisms (fishes in particular) between the Guadalupe River channel and oxbow lakes lying within its floodplain. Exchanges of basal food resources and fishes are facilitated by lateral connections that occur during high flow pulses. This project was designed to explore relationships between flow, lateral connectivity, and food web dynamics in the lower Guadalupe River and associated aquatic habitats in its floodplain. The project collected samples of basal production sources and fish and macroinvertebrate tissues for analysis of ratios of stable isotopes of carbon and nitrogen to infer production sources supporting fish biomass at two sites in the river channel and two nearby oxbows in the floodplain under contrasting flow conditions. When certain assumptions are met, analysis of stable isotope ratios can be an effective method to reveal spatial food web subsidies and animal movement (Fry 2002, Rubenstein and Hobson 2004, Roach et al. 2009, Docmac et al. 2017). Different kinds of plants assimilate carbon, nitrogen and other elements in their tissues with different ratios of heavier to lighter stable isotopes. For example, plants that use the C4-photosynthetic pathway tend to have significantly higher ratios of C13/C12 compared to plants that use the C3-photosynthetic pathway. Likewise, some types of algae may have C13/C12 ratios that are heavier than those of terrestrial C3 plants. Phytoplankton and benthic algae isotopic ratios may be influenced by local environmental conditions, including temperature, water velocity and concentrations of dissolved inorganic carbon compounds.

Spatial variation in the isotopic signatures of basal production sources and other components of aquatic food webs can be exploited to estimate movement of resources or consumers between habitats. For example, the contribution of post-spawn salmon carcasses to the food webs of oligotrophic streams of the Pacific Northwest and Alaska has been estimated by analyzing stable isotopes, and this is possible because salmon biomass is derived from marine production sources that have carbon and nitrogen isotopic ratios that are distinct from freshwater production sources (Naiman et al. 2002). When alternative sources have sufficiently distinct isotopic ratios, mixing models can be used to quantify estimates of proportional contributions of sources to consumer biomass (Fry 2006). When the potential sources contributing to consumer biomass have similar isotopic ratios, it is not a viable strategy to estimate proportional contributions using a mixing model. Other assumptions must be met when estimating sources supporting consumer biomass based on analysis of stable isotope ratios. The time interval for assimilation of consumed materials into tissues must be appropriate given the time interval used for interpreting movement of either sources or consumers among landscape units. Also, the trophic fractionation of isotopic ratios of elements must be estimated in order to estimate proportional contribution of sources to consumer biomass using a mixing model. Both of these factors generally are assumed in ecological research based on analysis of stable isotopes. Given these potential limitations, we collected samples for stable isotope analysis from various basal production sources and consumer taxa from channel and oxbow habitats of the lower Guadalupe River to: 1) determine if there was sufficient isotopic

variation among basal production sources at a given site to discriminate their relative contributions to consumer biomass, 2) determine if there was spatial variation within and among basal production sources and consumer taxa to allow estimation of between-habitat exchanges of material, and 3) determine if there was temporal variation within and among basal production sources and consumer taxa to allow estimation of how hydrology influences exchanges of material between oxbow lakes and the river channel. Contingent on finding sufficient isotopic variation, including discrimination among basal sources within and between habitats, our objective was to evaluate the probability that consumer taxa had either 1) received food web subsidies from an adjacent habitat following one or more high flow pulses that created lateral connections, or 2) migrated from an adjacent habitat during a recent high flow pulse that created a lateral connection. We also estimated the flows (discharge levels) that create connections between oxbow lakes and the river channel, and evaluated the frequency, magnitude and duration of high flow pulses that created lateral connections during our study interval. Finally, we examined spatial and temporal variation in fish assemblage structure to provide an additional means to evaluate exchanges of fishes between the river channel and oxbow lakes in relation to flows.

Methods

Field surveys

We consulted technical staff for the TPWD (Texas Parks and Wildlife Department) River Studies and Biowest, Inc. for assistance in determining locations with access to oxbow lakes in the floodplain of the lower Guadalupe River. After reviewing Google Earth imagery and consulting these individuals, two oxbow lakes (Oxbow 1 near Cuero, Texas, and Oxbow 2 near Victoria, Texas) were selected for study (Figures 1, 2 and 3). Two channel sites (River 1, River 2) also were selected for study. Each of the two channel sites was a stretch of up to 100 m that was adjacent to the point of connection between the active river channel and one of the two oxbows.

Surveys were conducted at each of the four study sites approximately every other month for one year (6 surveys at each of 4 study sites) between March 2016 and April 2017. For logistical and safety reasons, surveys were limited to periods when the discharge was below 5,000 cfs; therefore, time intervals between sampling periods were not uniform. Small fishes (< 200 mm standard length [SL]) were collected with two seines including a 7.6 × 1.8 m seine with a 3.2 mm mesh as well as a 9.14 × 3.05 m bag seine with a 3.05 m bag, consisting of 6.4 mm mesh in the wings, and 3.2 mm mesh in the bag. Larger fishes were collected using 29.3 × 1.8 m gill nets with four panels of equal length (7.3 m each) consisting of 1.3, 2.5, 5.1, and 7.6-cm bar mesh. Two gill nets were deployed in the main channel and two gill nets were deployed in the adjacent oxbow between approximately 1000 and 1400 hours for a total of 4 hours per survey. During two surveys (August and October 2016), shallow water prevented the deployment of gill nets in Oxbow 2, and during one of those surveys (October 2016), Oxbow 2 contained too little water to enable use of the seine (maximum depth ca. 20 cm). During each survey of each site, we recorded the survey effort associated with the seine sample (i.e., the total area swept by seine hauls) and gillnet sample (i.e., the time interval of gillnet deployment).

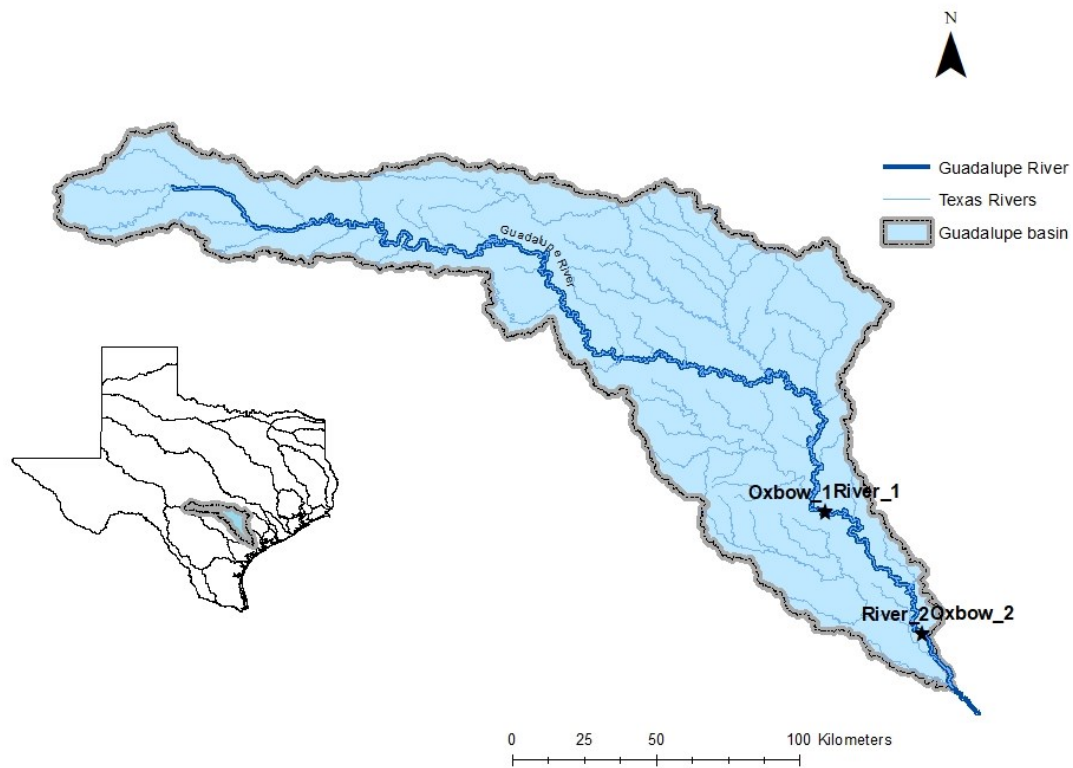


Figure 1. Map showing location of the Guadalupe River Basin in Texas, and locations of the four study sites – River 1, Oxbow 1, River 2, Oxbow 2.

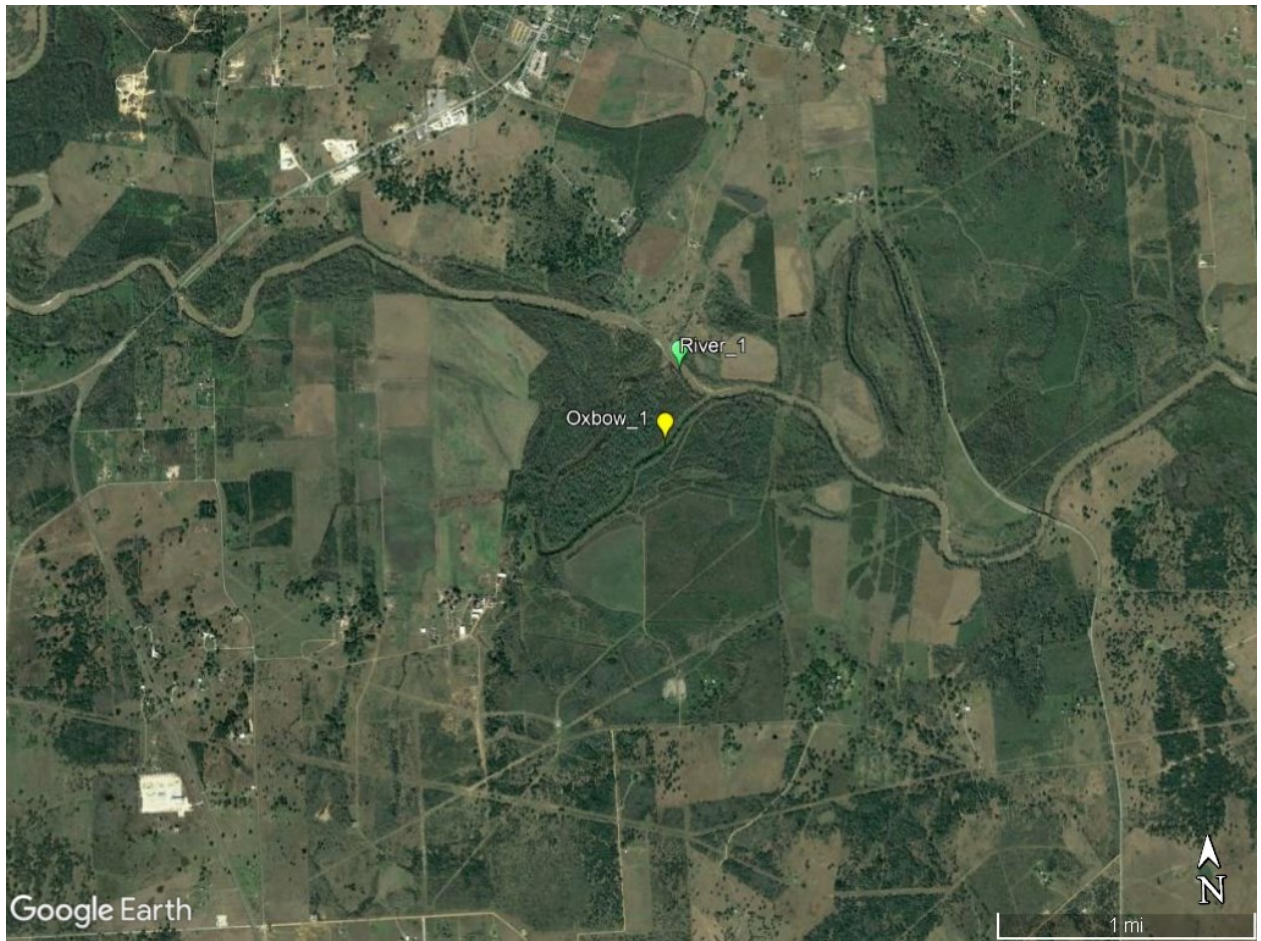


Figure 2. Google Earth image showing locations of survey sites River 1 and Oxbow 1 on the lower Guadalupe River near Cuero, Texas ($29^{\circ} 03' 32.24''$ N; $97^{\circ} 17' 11.15''$ W).

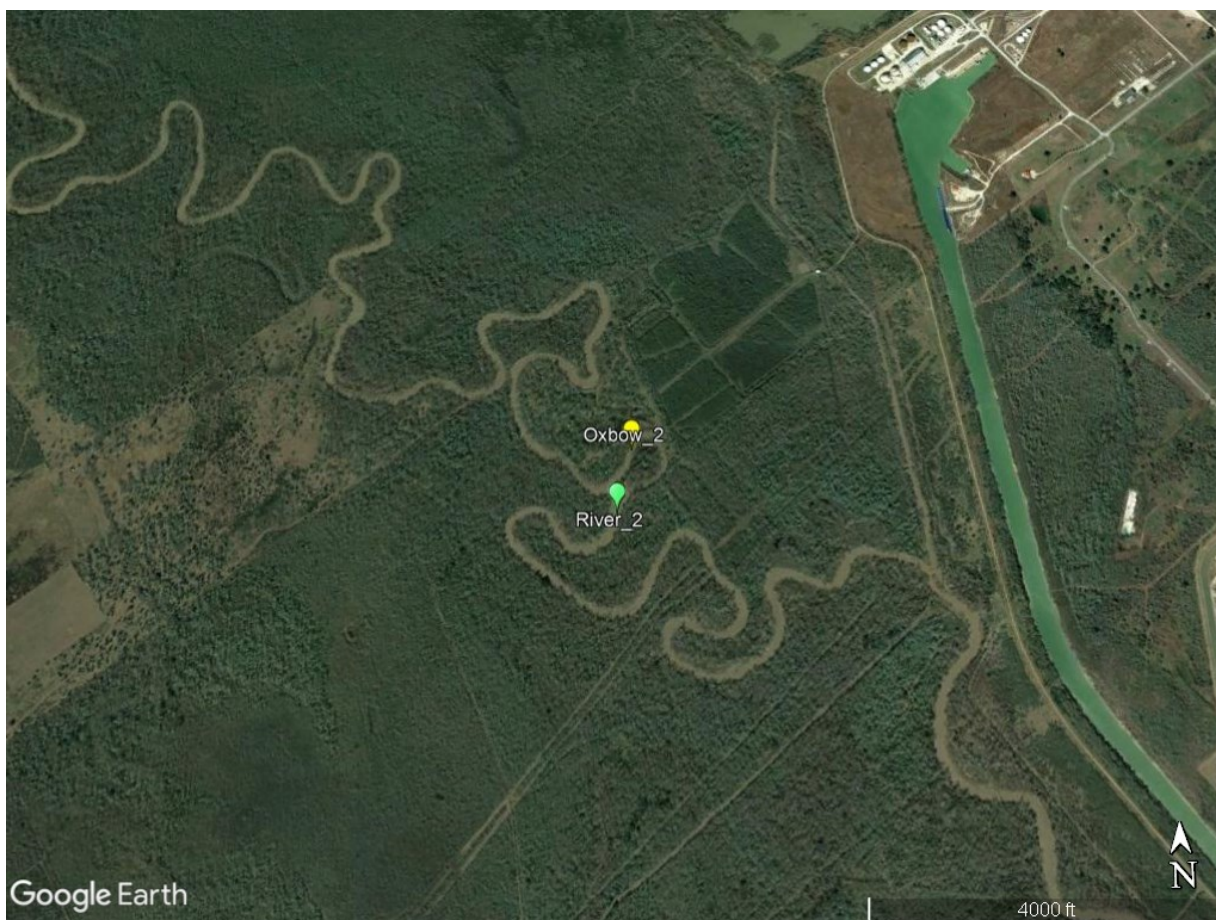


Figure 3. Google Earth image showing locations of survey sites River 2 and Oxbow 2 on the lower Guadalupe River near Victoria, Texas ($28^{\circ} 40' 39.27''$ N; $96^{\circ} 59' 10.69''$ W). The INVICTA chemical plant and ship channel can be seen in the upper right of the image.

Upon removal from the nets, fishes selected for extraction of tissue samples for isotopic analysis or for use as voucher specimens were euthanized according to an approved Texas A&M University animal use protocol (# IACUC 2015-0290). The remaining fishes were identified, counted, measured (SL) or recorded as a size class or life stage (juvenile, adult), and returned to their habitat. When large numbers of small fishes were captured in the seine net, they were placed into 5-gallon buckets filled with river water for holding until they could be identified and counted. A small aquarium net was used to remove fish from the bucket. Standard length was measured for specimens retained for stable isotopic analysis. Voucher specimens were placed into a labeled container with 10% formalin solution and stored in the Aquatic Ecology Lab prior to accession into the Biodiversity Research and Teaching Collection at Texas A&M University. Labels contained the field number, date, location information, and SL if measured.

Muscle and/or fin tissue samples were obtained for stable isotope analysis from common fish species that can be found in both the river channel and oxbows (longnose gar *Lepisosteus osseus*, gizzard shad *Dorosoma cepedianum*, threadfin shad *Dorosoma petenense*, bullhead minnow *Pimephales vigilax*, smallmouth buffalo *Ictiobus bubalus*, white crappie *Pomoxis annularis*, sunfish *Lepomis* spp.) for stable isotope analysis. All but a few samples were muscle tissue, but in a few instances fin tissue was obtained from very large fish in order to avoid harming fish so they could be returned to their habitat. Fish muscle and fin tissue reveals relatively low isotopic differences,

although there can be variation among species and size classes (Kelly et al. 2006). During each survey at each site, the goal was to obtain 3 replicate tissue samples for each taxon. Muscle tissue was obtained from the flanks of fish specimens following euthanasia according to an approved Texas A&M University animal use protocol (IACUC 2015-0290). At each site during each survey, we also collected samples of seston, microphytobenthos, riparian grasses (Poaceae and Cyperaceae species), leaves from dominant riparian vegetation (e.g., American sycamore *Platanus occidentalis*, boxelder *Acer negundo*, black willow *Salix nigra*), snails (superfamily Planorboidea), mussels (three-ridge *Amblema plicata*, yellow sandshell *Lampsilis teres*), and grass shrimp (*Palaemonetes kadiakensis*) for analysis of stable isotope ratios. Muscle tissue was obtained from the foot of snails and mussels, and the tail of grass shrimp. Seston, microphytobenthos, fine particulate organic matter (FPOM) and plant tissue samples consisted of composite (bulk) samples. Samples of benthic algae (microphytobenthos) and samples of FPOM in sediments were obtained by scraping material from submerged branches and logs using a spatula. Riparian macrophyte leaves, snails, and mussels were collected by hand. During each survey, we towed a plankton net (40-cm diameter, 80- μ m mesh) in attempts to obtain samples of zooplankton; however, few or none were collected, and consequently zooplankton were not considered in the analyses. Seston samples were obtained by filtering ca. 1.8 L of water drawn with a hand pump from the water column through a glass fiber filter to collect a sample of material for isotopic analysis. Seston samples were maintained in plastic bags on ice for transport to the lab where they were kept frozen until processing. All other tissue samples were preserved in NaCl in plastic bags for transport and until processing.

During each survey at each site, we recorded basic aquatic habitat parameters and water quality. Temperature ($^{\circ}$ C), conductivity (μ S), salinity (ppm), and dissolved oxygen (mg/L) were measured using a YSI Pro2030, and pH was recorded using an Oakton pH meter. Secchi depth was measured to the nearest centimeter. Substrate of both oxbows was a blend of silt and mud, and substrate in the channel was mostly silt and mud overlying sand and gravel, with some areas of exposed sand and gravel.

Flow data for the Guadalupe River were obtained from the U.S. Geological Survey gauges 08175800 in Cuero, TX, and 08176500 in Victoria, TX. The minimum flow required to connect surface waters of the main channel with a given oxbow was estimated as the flow recorded by the USGS gage on dates when we observed just a few centimeters of water in the channel connecting the oxbow to the river. Our connectivity estimates were corroborated by independent estimates made by Biowest, Inc. based on data for topography, surface water elevation, and USGS gage discharge (Bonner et al. 2017).

Preparation of Tissue Samples for Stable Isotope Analysis

Tissue samples of seston, microphytobenthos, dominant grasses, dominant riparian vegetation, mussels, grass shrimp and fishes were soaked in distilled water for 4-5 h, rinsed, and dried in an oven at 60 $^{\circ}$ C for approximately 48 h. according to the methods used in previous studies (e.g., Zeug and Winemiller 2008b, Roach and Winemiller 2015). After drying, samples were ground into fine powder using mortar and pestle. Samples were weighed (the fish and source samples to the nearest 1.5-3 mg, and seston to the nearest 15-20 mg), loaded into UltraPure tin capsules, and then sent to the Analytical Chemistry Lab at the University of Georgia for analysis of ratios of stable isotopes of carbon and nitrogen using mass spectrometry.

Stable Isotope Analysis

Samples were analyzed for carbon and nitrogen isotope ratios using mass spectrometry at the laboratory at the Analytical Chemistry Laboratory of the Institute of Ecology, University of Georgia, USA. Isotope ratios were reported in parts per thousand (‰) standardized in relation to reference material (Pee Dee Belemnite for C, atmospheric nitrogen for N) and reported as

$$\delta X = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 10^3,$$

where $R = {}^{13}\text{C}/{}^{12}\text{C}$ or ${}^{15}\text{N}/{}^{14}\text{N}$ (the ratio of heavy and light stable isotopes of carbon or nitrogen).

Isotopic ratio data received from the Analytical Chemistry Lab were first examined for reliability based on consistency of values for lab standard materials that were analyzed between every 10 samples from the field study. Second, we examined stable isotope ratios for nitrogen to determine if they reasonably reflected vertical trophic positions (e.g., plants should have lower values than herbivores, such as snails, and herbivores should have lower values than carnivores; see description of trophic position estimates below). Some of our seston samples contained amounts of carbon and nitrogen below the limit of detection for mass spectrometric analysis of stable isotope ratios, apparently because they contained little to no phytoplankton or detritus.

Plots of carbon and nitrogen ratios of sources materials and consumers were used to explore possible exchanges between oxbows and river channel sites. To evaluate the potential for assimilation of material from basal production sources in oxbows and channel habitats, we plotted mean $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of basal production sources and values for individual specimens of macroinvertebrates (mussels only; few snails were obtained and grass shrimp revealed little isotopic variation between channel and oxbows and thus were omitted from further analyses) and fishes from a given oxbow survey and adjacent river channel site on a given date. For each oxbow and river channel survey pair, we plotted mean isotopic ratios of available basal production sources and isotopic ratios of each specimen for each consumer taxon for each of the six survey periods. We then plotted isospace polygons for each habitat type (oxbow vs. channel) with adjustment for the trophic position of the consumer taxon of interest, and evaluated which specimens fell inside or outside the polygon. Those specimens that were inside or very near a given polygon had a reasonable probability of having assimilated some proportion of material from basal resources from the habitat that defined that polygon.

Isospace polygons were plotted by first estimating the isotopic difference between tissue of the consumer taxon and tissue of a primary consumer (mean value for mussels) from the same habitat and survey period, and then adding the assumed value for trophic fractionation (i.e., the shift in isotopic ratio from enrichment of the heavier isotope associated with transfer between adjacent trophic levels in a food chain) of $0.40 \pm 1.3\text{‰}$ (Post 2002) and $2.54 \pm 1.3\text{‰}$ for $\delta^{15}\text{N}$ (Vanderklift and Ponsard 2003) to account for trophic fractionation for the step between basal resources and the primary consumer (mussels). These values were used to move the isospace polygon connecting various basal production sources within a given habitat (oxbow or channel site) an appropriate distance upward and to the right within the isotopic biplot. We were particularly interested in testing the hypothesis that a given consumer captured from an oxbow had assimilated material originating from the river channel, and that a given consumer captured from the river channel had assimilated material originating from an oxbow. Evidence for assimilation of basal production

across habitats is obtained when a consumer's position is well outside the isospace polygon of the habitat where it was captured, while falling inside the polygon for the alternative habitat. Also, the degree to which a consumer is aligned vertically above a particular source indicates the likelihood that the consumer had assimilated material from that source.

A Bayesian mixing model is often used to estimate feasible percent contributions of primary production sources to consumer tissue. These kinds of quantitative estimates are only possible when all reasonable production sources are included in the model, and there is sufficient divergence in the isotopic ratios among production sources to allow for discrimination in the mixing model. In addition, several values must be input into the model as assumptions. These include the trophic fractionation of isotopic ratios of each element and the trophic position (i.e., trophic level) of the consumer. Our stable isotope dataset lacked data for certain production sources at certain sites during certain periods, and in some cases lacked good discrimination among sources from the two habitat types. Consequently, we did not use a mixing model to quantify percent contributions of alternative production sources from the two habitat types in order to infer lateral exchanges of food resources and/or aquatic animals. We instead made qualitative inferences about material exchanges based on interpretation of consumer position within or outside of isospace windows in the isotopic biplots. These inferences were then summarized in a chart to facilitate conclusions about lateral exchanges at the two locations under various flow conditions.

Fish Assemblage Structure

Fish species abundance data were $\log(x+1)$ transformed, and species where only one individual was collected were removed from the dataset. Variation in assemblage structure among sites and seasons was visualized using nonmetric multidimensional scaling (NMDS). Nonmetric multidimensional scaling is a distance based method of ordination, which ordines sample units based on their ranked dissimilarity. Sample units with similar species compositions will fall in close proximity in the ordination space. The NMDS analysis was performed using Bray-Curtis distance and 50 maximum iterations for a stable configuration. To test for significant differences of assemblage structure among sites and seasons, non-parametric permutational multivariate analysis of variance (PERMANOVA) was performed on Bray-Curtis dissimilarity matrices of transformed species abundances as the dependent variable and the sites and seasons as the categorical variables. PERMANOVA is similar in design to a classic MANOVA (multivariate analysis of variance), but has relaxed assumptions by relying on a permutation procedure (Anderson, 2001). The method analyzes between-group dissimilarities over within-group dissimilarities, producing a pseudo-F value that is analogous to the Fisher's F-ratio for each term in the model and assesses significance via permutation (Anderson, 2001). In addition, Canonical correspondence analysis (CCA) was used to explore the relationship among environmental gradients and assemblage structure for all sites. All environmental data input into this analysis can be found in Table 1. Only the first two axes of the CCA were retained for interpretation. NMDS, PERMANOVA and CCA were performed with the VEGAN package in R version 3.1.1 (R Foundation for Statistical Computing).

Estimation of Environmental Flows for Lateral Connectivity

The magnitude of flow for hydrological connectivity was estimated by comparing the discharge at the USGS gage that was nearest to each study oxbows on dates when the oxbow was observed

to have a minimal connection to the river channel. On at least one survey date during the study period, a depth of water ca. 2-10 cm was observed in the channels that connected the oxbows to the river channel. The magnitude and duration of high flow pulses associated with discharges exceeding the discharge for minimum connection was estimated from USGS gage data for the survey period as well as for the previous 10 years. Finally, inferred lateral exchanges of organic material from basal production sources and/or fishes between oxbows and the river channel were evaluated to assess frequency of food web exchanges during the 12-month study period.

Results

Hydrology, Lateral Connectivity and Abiotic Environmental Conditions

According to the discharge record from USGS gage #08175800 Guadalupe River at Cuero and the estimated minimum discharge for water to connect Oxbow 1 (1,630 cfs), there were nine high flow pulses that connected Oxbow 1 between March 1, 2016 until April 30, 2017 (Figure 4). Duration of connections varied from 1 day to 88 days, and maximum discharge during pulses ranged from 838 cfs (March 7, 2016) to 20,900 cfs (June 5, 2016). The pulse with the largest magnitude and duration occurred from April 15 to July 11, 2016, and actually had two peaks (18,400 cfs and 20,900 cfs). Five of the six surveys were conducted at or very near the end of high flow pulses, and only the August 2016 survey did not immediately follow a high flow pulse. Oxbow 1 was connected to the channel for 2–15 days during the 15-day window that preceded survey dates, and for 5–30 days during the 30-day window that preceded survey dates (Figure 4).

According to the discharge record from USGS gage #08176500 Guadalupe River at Victoria and the estimated minimum discharge for water to connect Oxbow 2 (1,580 cfs), there were nine high flow pulses that connected Oxbow 2 between March 1, 2016 until April 30, 2017 (Figure 5). Duration of connections varied from 2 days to 92 days, and maximum discharge during pulses ranged from 786 cfs (April 9, 2016) to 21,300 cfs (June 6, 2016). The pulse with the largest magnitude and duration occurred from April 16 to July 16, 2016, and with two peaks (17,900 cfs and 21,300 cfs). Just as at Oxbow 1, five of the six surveys at Oxbow 2 were conducted at or very near the end of high flow pulses, and only the August 2016 survey did not immediately follow a high flow pulse. Oxbow 2 was connected to the channel for 3–15 days during the 15-day window that preceded survey dates, and for 9–30 days during the 30-day window that preceded survey dates (Figure 5).

Water temperature varied seasonally (channel range = 15.6–32.0° C; oxbow range = 13.9–33.4° C), with both channel sites and oxbows being coldest during the winter survey (January 27–28, 2017) and warmest during summer surveys (July and August, 2016) (Table 1). PH and conductivity in the river channel and oxbows varied in accordance with variation in river discharge. Lowest conductivity (293–405 $\mu\text{S}/\text{cm}$) was recorded at all four sites during March 2016 after a large flow pulse (Figures 4 and 5) and relatively high flows during the preceding four months (USGS gages #08175800 and 08176500). Highest conductivity was recorded in the river channel (587–608 $\mu\text{S}/\text{cm}$) and oxbows (599–873 $\mu\text{S}/\text{cm}$) during August following a period of one month without any flow pulses. Measurements of pH varied from 6.8 (Oxbow 1 during April 2017) to 8.5 (river channels sites on multiple dates and Oxbow 1 in October 2016). Variation in pH had no obvious relationship to temperature or flow conditions. Secchi depth, and indicator of water transparency, was lowest during surveys conducted during spring (March 2016, April 2017) and fall (October 2016) in both oxbows and the river channel (Table 1). This increase in turbidity during spring and

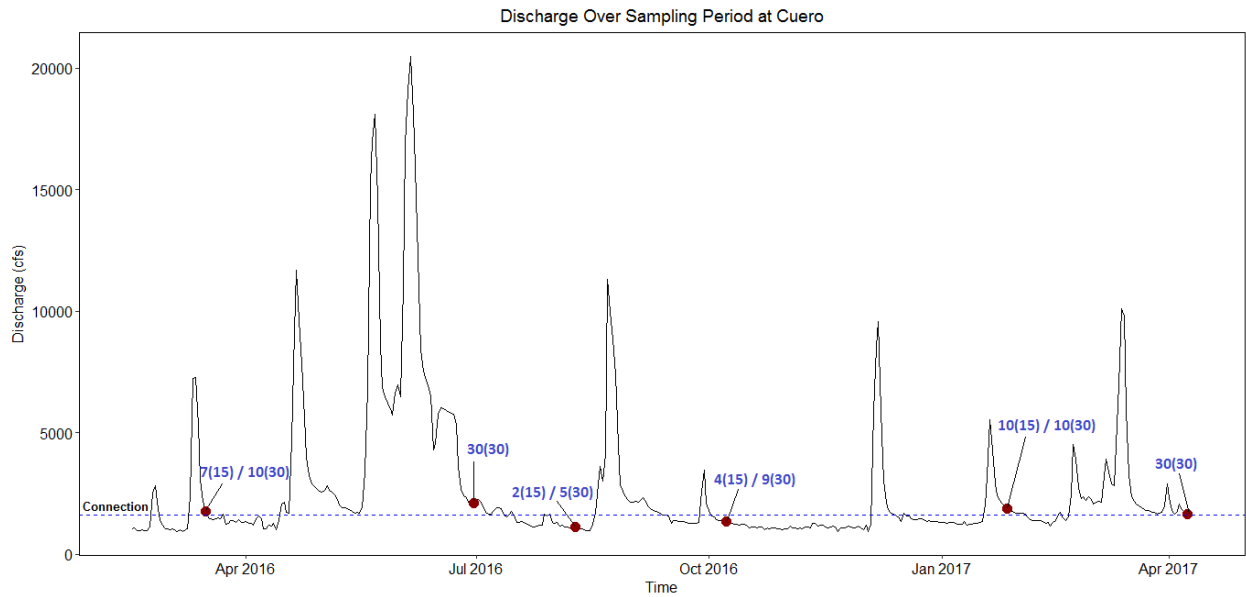


Figure 4. Discharge at USGS gage #08175800 Guadalupe River at Cuero, TX, during the period March 1, 2016 to April 30, 2017. Red dots indicate the six survey dates; blue dotted line indicates the approximate minimum discharge when Oxbow 1 makes a connection with the Guadalupe River channel; blue numbers indicate the estimated number of days Oxbow 1 was connected during the previous 15-day and 30-day intervals.

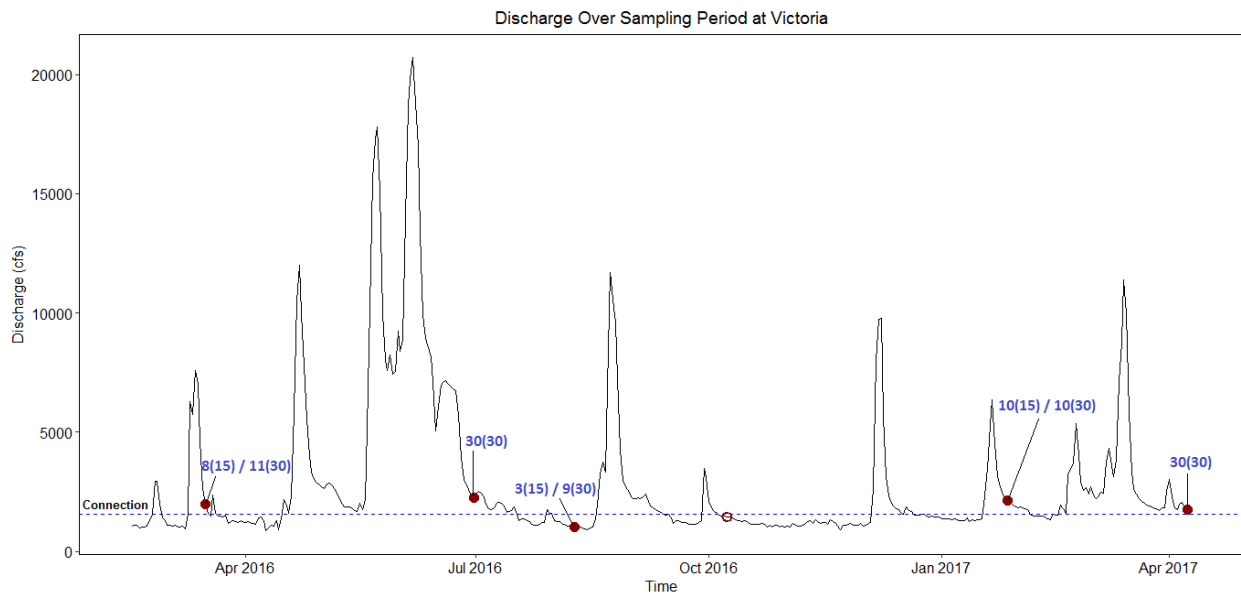


Figure 5. Discharge at USGS gage #08176500 Guadalupe River at Victoria, TX, during the period March 1, 2016 to April 30, 2017. Red dots indicate the six survey dates; open red dot indicates a date when the oxbow contained little water and was not surveyed; blue dotted line indicates the approximate minimum discharge when Oxbow 2 makes a connection with the Guadalupe River channel; blue numbers indicate the estimated number of days Oxbow 2 was connected during the previous 15-day and 30-day intervals.

Table 1. Measures of temperature (°C), pH, Secchi depth (cm), dissolved oxygen (mg/L), and specific conductivity (µS/cm) for each collection habitat, season, and date.

Habitat	Season	Date	Water temp.	pH	Secchi depth	DO	Specific conductivity
Oxbow 1	Winter	1/27/2017	15.0	8.4	20	7.0	628
Oxbow 1	Spring	3/16/2016	22.8	8.2	17	6.9	358
Oxbow 1	Spring	4/8/2017	24.6	6.8	8	6.5	610
Oxbow 1	Summer	7/1/2016	30.2	7.8	50	9.5	640
Oxbow 1	Summer	8/9/2016	33.4	8.3	20	8.2	873
Oxbow 1	Fall	10/8/2016	26.8	8.5	11	9.5	627
Oxbow 2	Winter	1/28/2017	13.9	8.2	15	4.8	610
Oxbow 2	Spring	3/17/2016	22.6	8.2	13	5.6	293
Oxbow 2	Spring	4/9/2017	22.7	7.0	10	7.5	491
Oxbow 2	Summer	6/30/2016	31.0	8.3	40	15.3	428
Oxbow 2	Summer	8/10/2016	32.2	8.1	10	4.3	599
Oxbow 2	Fall	10/9/2016	n/a	n/a	n/a	n/a	n/a
River 1	Winter	1/27/2017	15.8	8.5	25	7.6	504
River 1	Spring	3/16/2016	22.5	8.3	16	8.4	405
River 1	Spring	4/8/2017	22.9	7.3	20	6.8	512
River 1	Summer	7/1/2016	29.8	8.3	25	9.1	500
River 1	Summer	8/9/2016	32.0	8.4	30	7.6	587
River 1	Fall	10/8/2016	26.2	8.4	20	8.8	545
River 2	Winter	1/28/2017	15.6	8.5	19	7.5	573
River 2	Spring	3/17/2016	22.7	8.3	20	8.5	380
River 2	Spring	4/9/2017	23.0	7.0	25	6.6	575
River 2	Summer	6/30/2016	30.8	8.2	25	8.6	546
River 2	Summer	8/10/2016	31.7	8.5	30	7.4	608
River 2	Fall	10/9/2016	26.4	7.0	17	7.9	573

fall surveys was associated with high flow pulses, however, there were surveys conducted following higher flow pulses (e.g., June/July 2016) that recorded greater Secchi depths (i.e., lower turbidity). Dissolved oxygen (DO) values were consistently high in both the river channel and oxbows, including several values that represent supersaturation (i.e., values > 8 mg/L). Oxbow 2 revealed greatest variation in DO, with values ranging from 4.3 mg/L (August 2016) to 15.3 mg/L (June 2016).

Stable Isotope Patterns

Riparian vegetation samples, both C3 and C4 plants, showed relatively low variation in mean $\delta^{13}\text{C}$ (and to a much lesser degree $\delta^{15}\text{N}$) between sites and survey periods (Figures 6-13). One riparian vegetation sample (August 2016) showed a moderate level of within-sample variation for

the river channel. Riparian C3 plants had lowest values for $\delta^{13}\text{C}$, and riparian C4 plants (grasses) had highest values for $\delta^{13}\text{C}$, with one exception during June 2016 and October 2016 when seston from the river channel had higher mean values for $\delta^{13}\text{C}$.

In contrast to leaf tissues of riparian plants, seston and periphyton samples revealed greater variation for $\delta^{13}\text{C}$ (with much less variation for $\delta^{15}\text{N}$) among survey periods and locations. Isotopic data are lacking for seston or periphyton samples at several survey sites during certain periods. Missing values resulted from an inability to find sufficient material for analysis (e.g., periphyton film growing on submerged hard surfaces) or insufficient organic matter in samples resulting in failure of mass spectrometry to estimate values for stable isotopes (Table 2). When a sample failed to produce mass spectrometry results, it was assumed that the sample was comprised mostly of inorganic matter, especially silt that was suspended in the water column (seston samples) or deposited on the substrate (periphyton samples). Samples that did not produce readings often came from the river channel and periods of high turbidity following high flow pulses. Under these conditions, aquatic primary production should be low (Roach et al. 2014), and material from aquatic basal production would not be expected to enter aquatic food chains in substantial amounts (Roach and Winemiller 2015). Stable isotope data were obtained for seston and periphyton samples taken from both channel sites on June 30-July 1, 2016, a survey that followed the largest and longest duration flow pulse during the study period. Turbidity in the channel was high (low Secchi depth) during that survey (Table 1), and those seston samples probably were dominated by fine particulate organic matter in the form of detritus of terrestrial origin. The high $\delta^{13}\text{C}$ values of those seston samples (Figures 6-13) indicate that this material likely was derived from C4 grasses.

Table 2. Summary of survey periods and sites with autochthonous primary production source samples below detection level for mass spectrometry analysis of stable isotope ratios of C and N. Samples below detection for C and N are assumed to contain mostly inorganic material that was suspended in the water column or deposited on substrates and thus increasing turbidity to levels that hindered light penetration and phytoplankton and benthic alga growth.

	Oxbow 1	Oxbow 2	River 1	River 2
Seston				
March 2016			X	
June 2016				
August 2016	X		X	
October 2016		X	X	X
January 2017	X	X	X	X
April 2017	X	X	X	X
Periphyton				
March 2016	X	X	X	X
June 2016	X			
August 2016	*	X	X	X
October 2016	X		X	*
January 2017		X	X	
April 2017		*	*	X

* indicates a single sample was above detection level and yielded isotopic data

Figures 6–13 are $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ biplots for each major fish taxon and two mussel species (combined for analysis) that were commonly captured from both river channel and oxbow habitats. Seasonal surveys are plotted separately as six biplots for each taxon, with oxbow data printed as orange and red symbols, and channel data printed as blue or black symbols. The mean values of basal production sources appear in each biplot, along with the isospace polygons for oxbow versus river habitats, each of which represents the space that could be occupied by a consumer that assimilates some combination of material derived from production sources in one of these habitats. Given that $\delta^{13}\text{C}$ has relatively low trophic fractionation ($0.40 \pm 1.3\text{‰}$, Post 2002) and $\delta^{15}\text{N}$ has higher trophic fractionation ($2.54 \pm 1.3\text{‰}$, Vanderklift and Ponsard 2003), the position of a consumer in relation to the positions of alternative basal sources in a biplot indicates the probability that the consumer had assimilated material, either via direct consumption or via food chains, from those sources (Fry 2006). If a consumer is positioned directly above and slightly to the right of source A, and other potential sources (sources B and C) are located much further to the left or right in the biplot, then it is probable that the consumer derived most of its carbon from source A and very little from sources B and C. A formal means to estimate probable source contributions to consumer biomass is the isospace polygon defining the feasible space for a consumer deriving biomass from some combination of a set of basal sources. The majority of mussel and fish samples were positioned above riparian (terrestrial) C3 plants, indicating that allochthonous vegetation was an important basal production source for aquatic food webs in both the river channel and oxbow lakes throughout the study period.

The position of consumers relative to isospace polygons for *in situ* habitat (the same habitat type where the consumer was collected) and *ex situ* habitat (the opposite habitat type than where the consumer was collected), can be used to infer the likelihood that the consumer had assimilated matter originating from the oxbow or channel habitat, irrespective of the habitat where it was captured. Of particular interest for the goals of this project were cases in which the consumer fell inside the isospace polygon of the opposite habitat type and outside the isospace polygon of the habitat type from where it was captured. Such cases indicate either a transfer of basal production from one habitat type to the other (i.e., spatial from the food web base), or the movement of consumers from one habitat type to the other (i.e., spatial food web subsidy to higher consumers) (Polis et al. 1996). Twenty-one (7%) consumers captured from oxbows showed this evidence of spatial food web subsidy (Table 3). Twenty-six (11%) consumers captured from the river channel showed evidence of spatial food web subsidy (Table 3). An additional 85 (27%) consumers from oxbows and 142 (61.5 %) consumers from the river channel fell inside both the *in situ* and *ex situ* polygons, and therefore are unresolved in terms of the probability that they could have assimilated material from the opposite habitat type. The remaining consumer samples (65.9% in oxbows, 27% in river channel) fell inside their respective *in situ* isospace polygons, and therefore were likely to have recently assimilated material only from the habitat where they were captured.

The longnose gar did not reveal evidence of assimilation of basal source material from the opposite habitat type, and evidence of spatial food web subsidy was only observed during one survey period for threadfin shad (5 specimens captured from oxbows in October 2016) and smallmouth buffalo (3 specimens captured from the river channel in March 2016) (Table 3). The bullhead minnow only revealed evidence that fish captured from the river channel had assimilated basal source material from oxbows, and this corresponded to half of the six survey periods (March, August 2016 and January 2017). Gizzard shad, sunfishes, white crappie and mussels showed

Lepisosteus osseus- Longnose gar

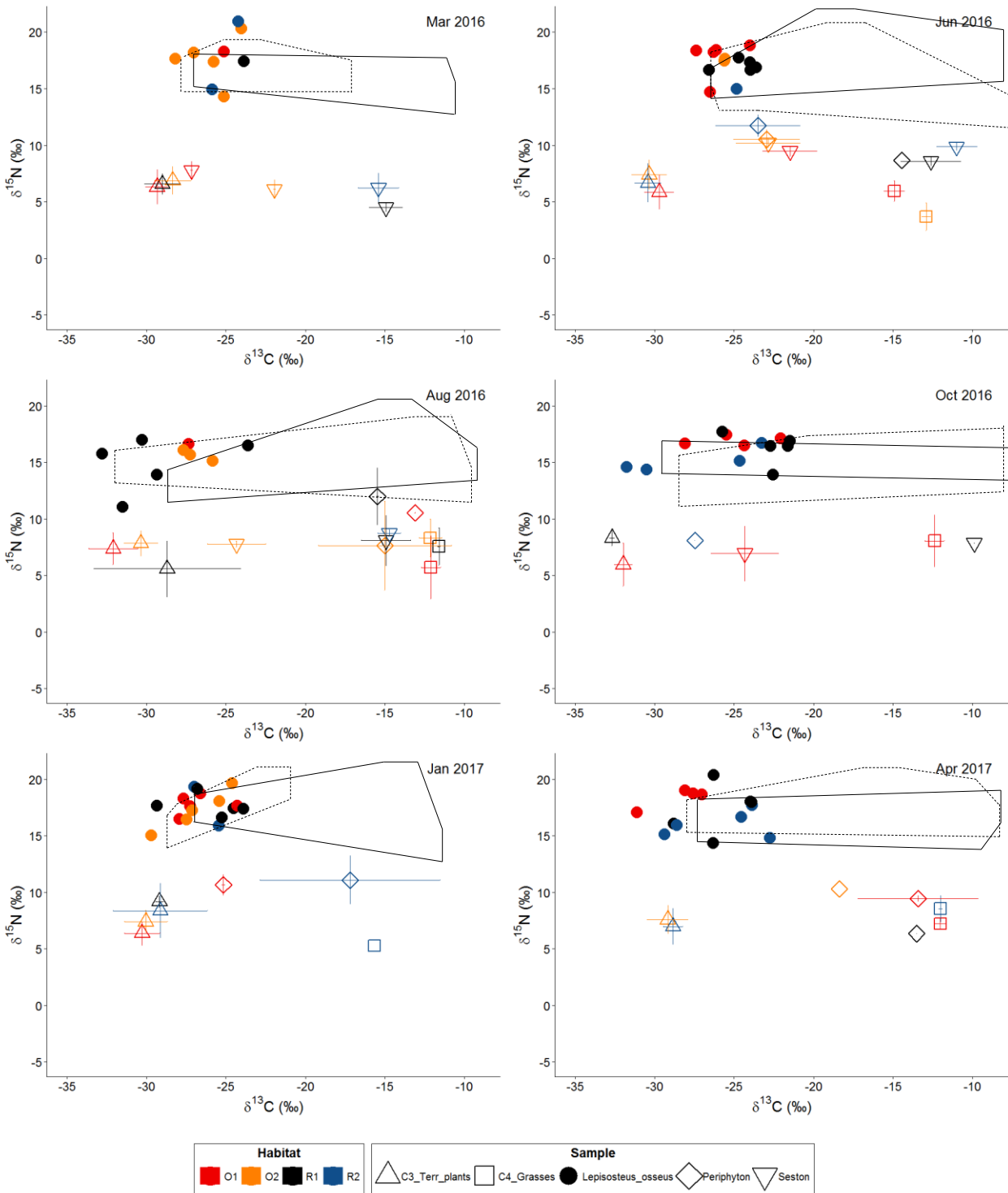


Figure 6. Biplots of carbon and nitrogen stable isotope ratio values for tissue samples of basal production sources and longnose gar from oxbow and river channel sites. The isospace polygon defining feasible locations for a consumer assimilating some combination of material from basal production sources is shown for oxbows (dotted lines) and river channel (solid lines).

Dorosoma cepedianum - Gizzard shad

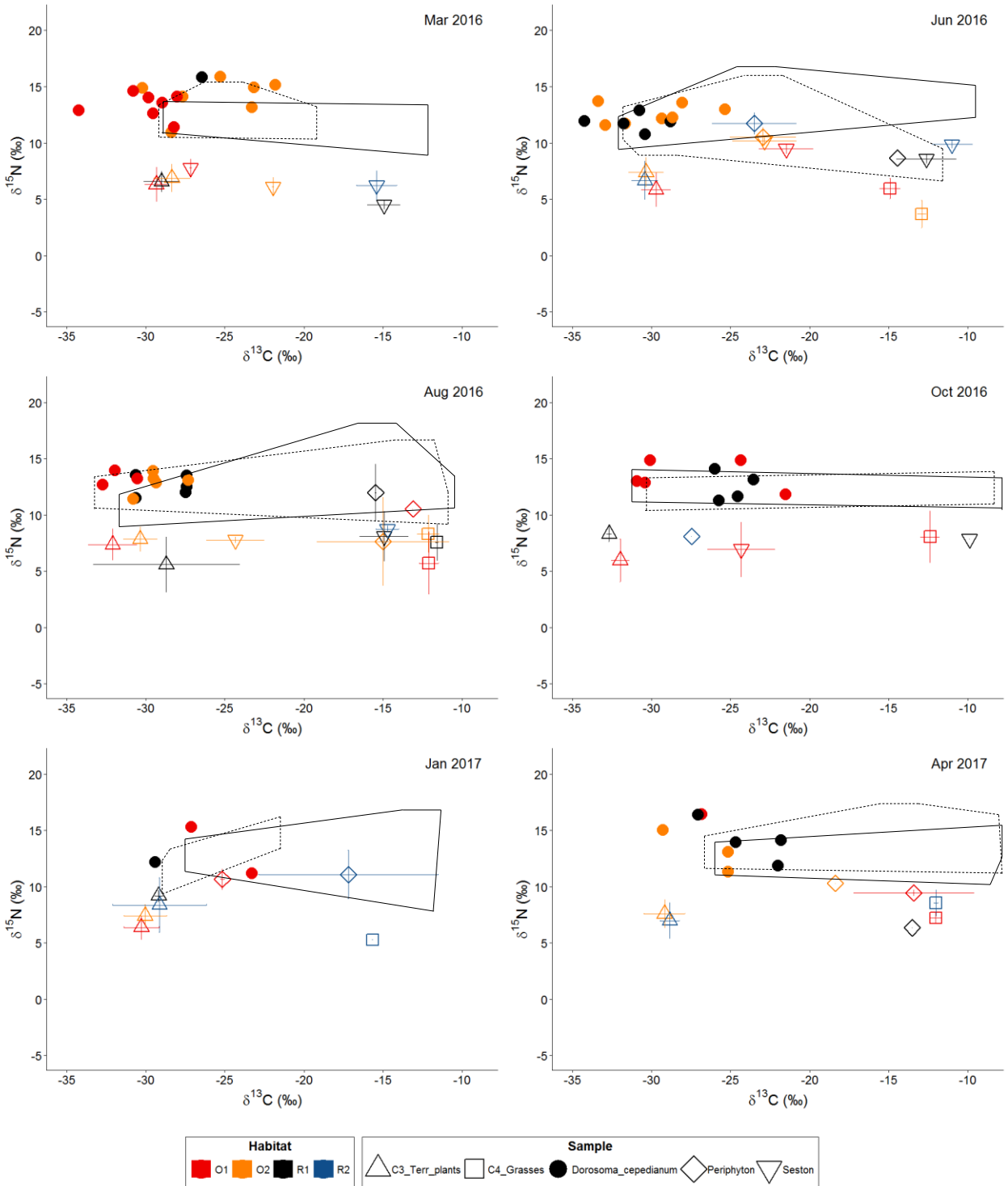


Figure 7. Biplots of carbon and nitrogen stable isotope ratio values for tissue samples of basal production sources and gizzard shad from oxbow and river channel sites. The isospace polygon defining feasible locations for a consumer assimilating some combination of material from basal production sources is shown for oxbows (dotted lines) and river channel (solid lines).

Dorosoma petenense- Threadfin shad

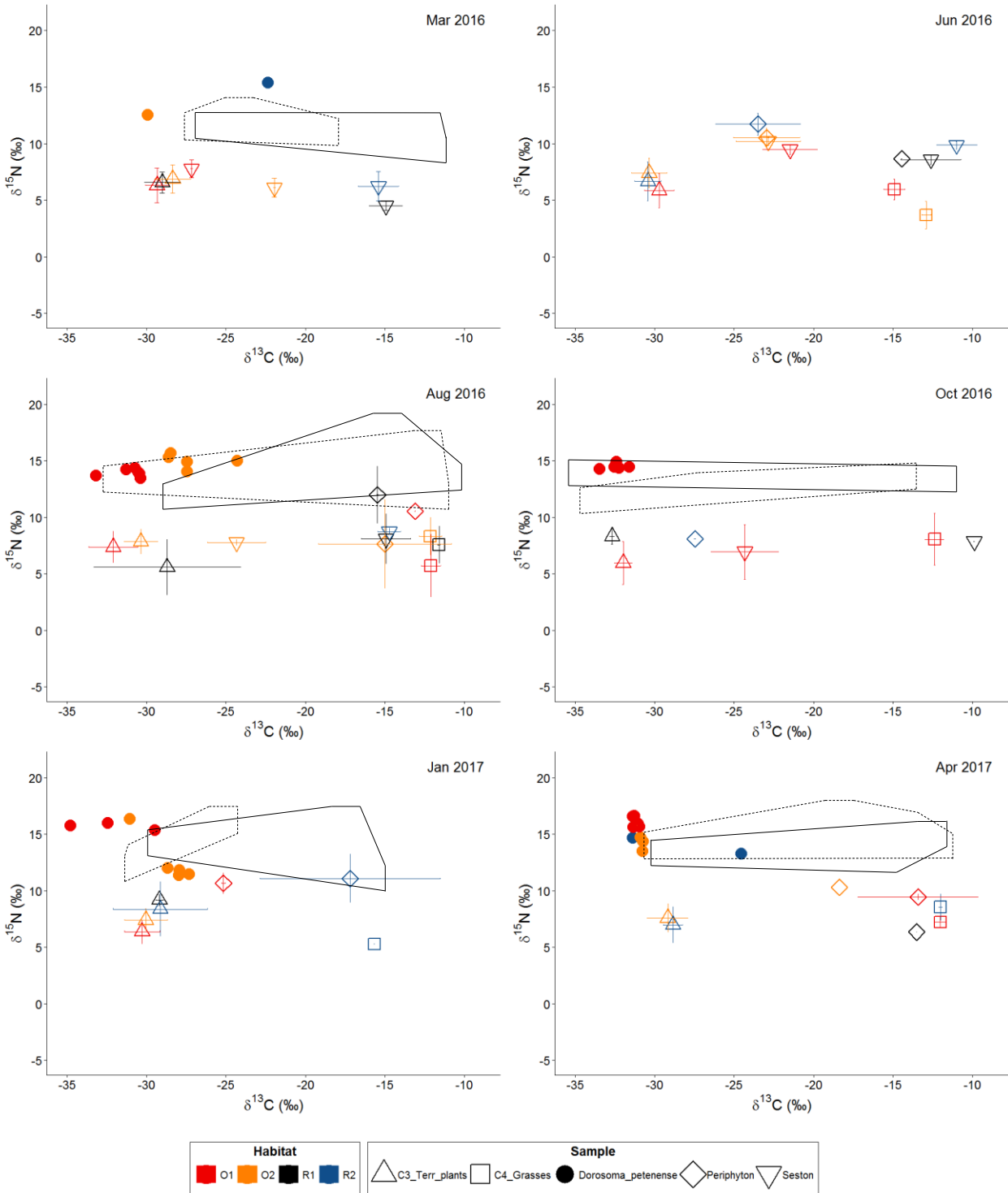


Figure 8. Biplots of carbon and nitrogen stable isotope ratio values for tissue samples of basal production sources and threadfin shad from oxbow and river channel sites. The isospace polygon defining feasible locations for a consumer assimilating some combination of material from basal production sources is shown for oxbows (dotted lines) and river channel (solid lines).

Pimephales vigilax - Bullhead minnow

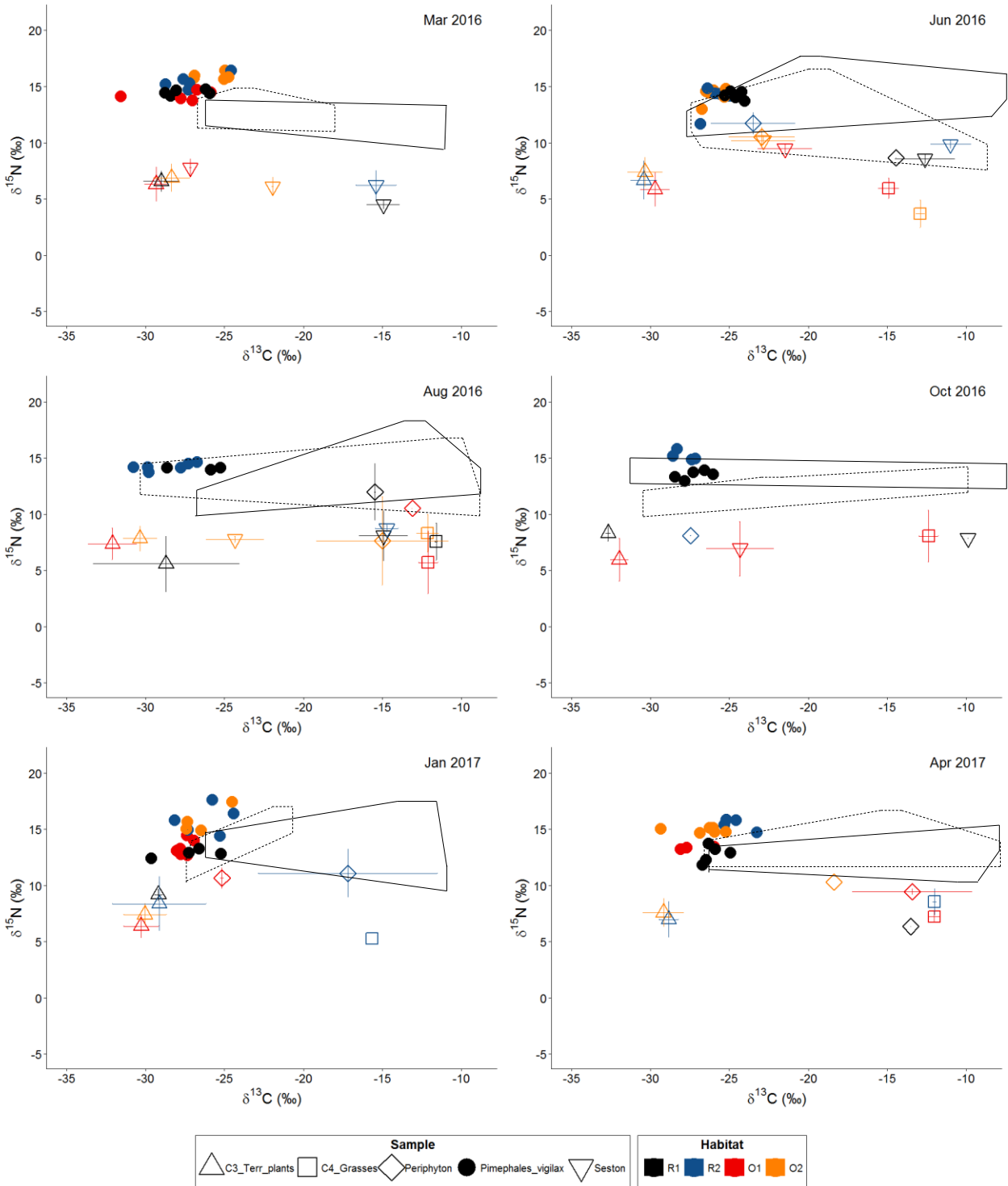


Figure 9. Biplots of carbon and nitrogen stable isotope ratio values for tissue samples of basal production sources and bullhead minnow from oxbow and river channel sites. The isospace polygon defining feasible locations for a consumer assimilating some combination of material from basal production sources is shown for oxbows (dotted lines) and river channel (solid lines).

Ictiobus bubalus - Smallmouth buffalo

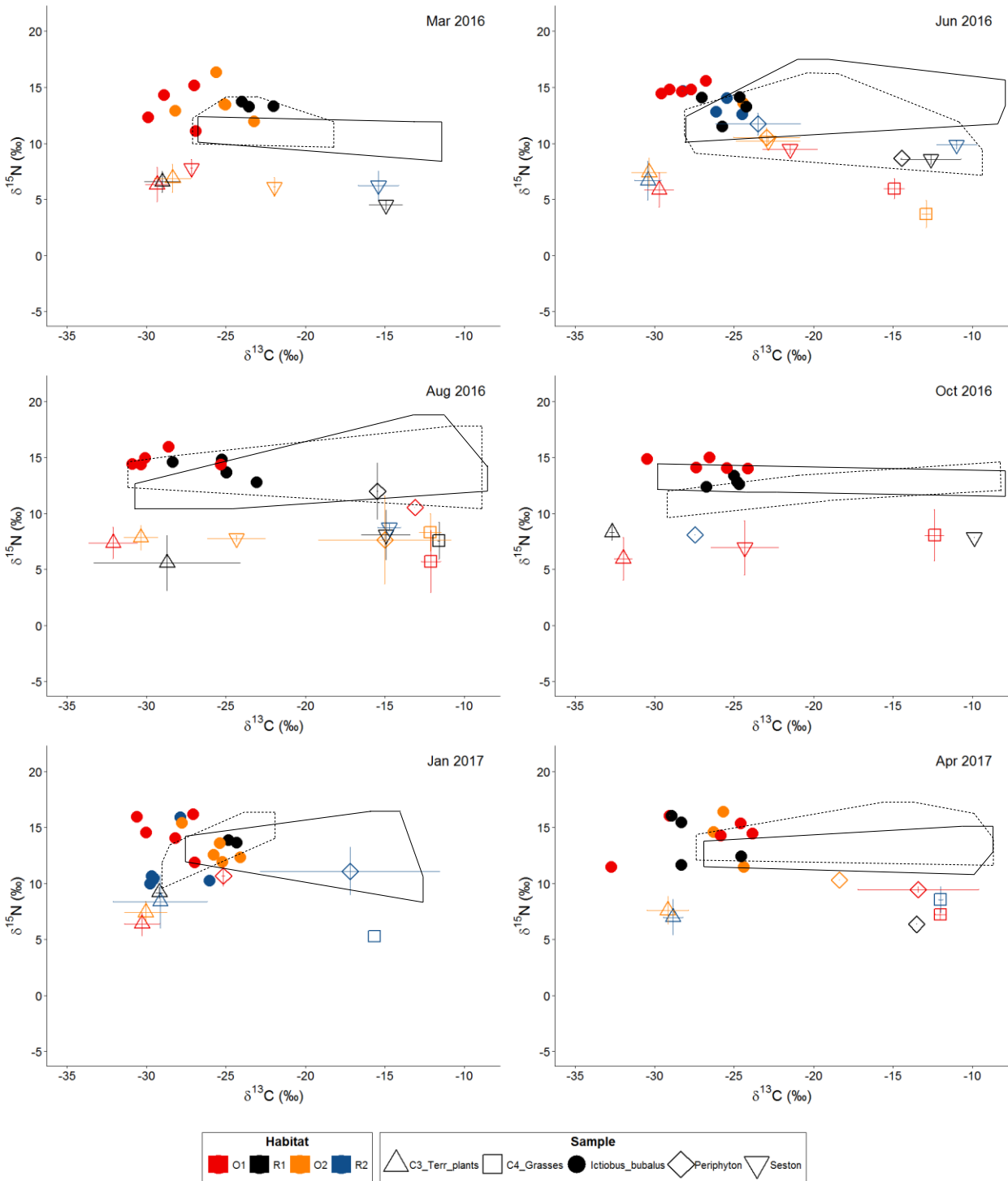


Figure 10. Biplots of carbon and nitrogen stable isotope ratio values for tissue samples of basal production sources and smallmouth buffalo from oxbow and river channel sites. The isospace polygon defining feasible locations for a consumer assimilating some combination of material from basal production sources is shown for oxbows (dotted lines) and river channel (solid lines).

Lepomis spp. - Sunfishes

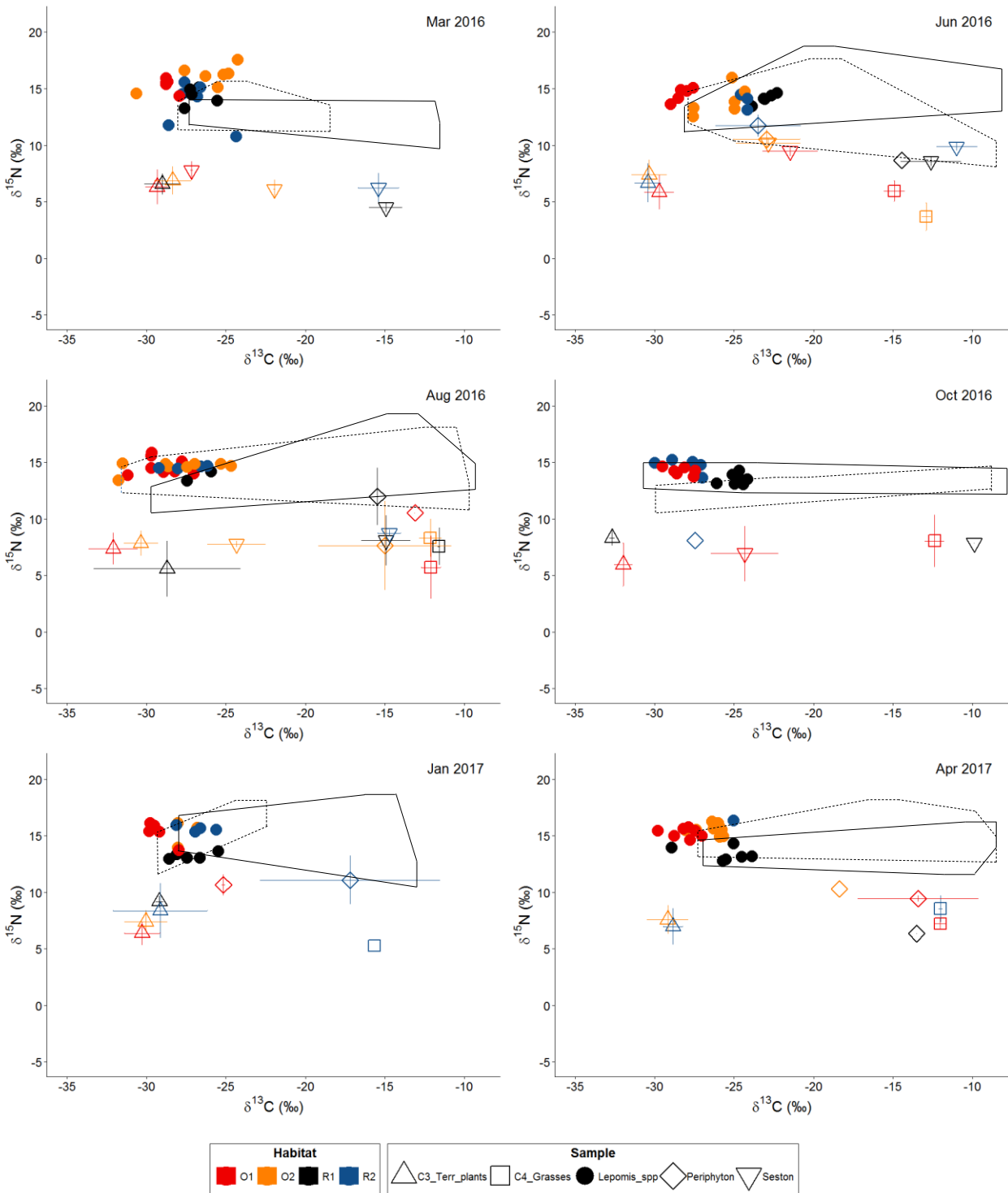


Figure 11. Biplots of carbon and nitrogen stable isotope ratio values for tissue samples of basal production sources and sunfishes from oxbow and river channel sites. The isospace polygon defining feasible locations for a consumer assimilating some combination of material from basal production sources is shown for oxbows (dotted lines) and river channel (solid lines).

Pomoxis annularis- White crappie

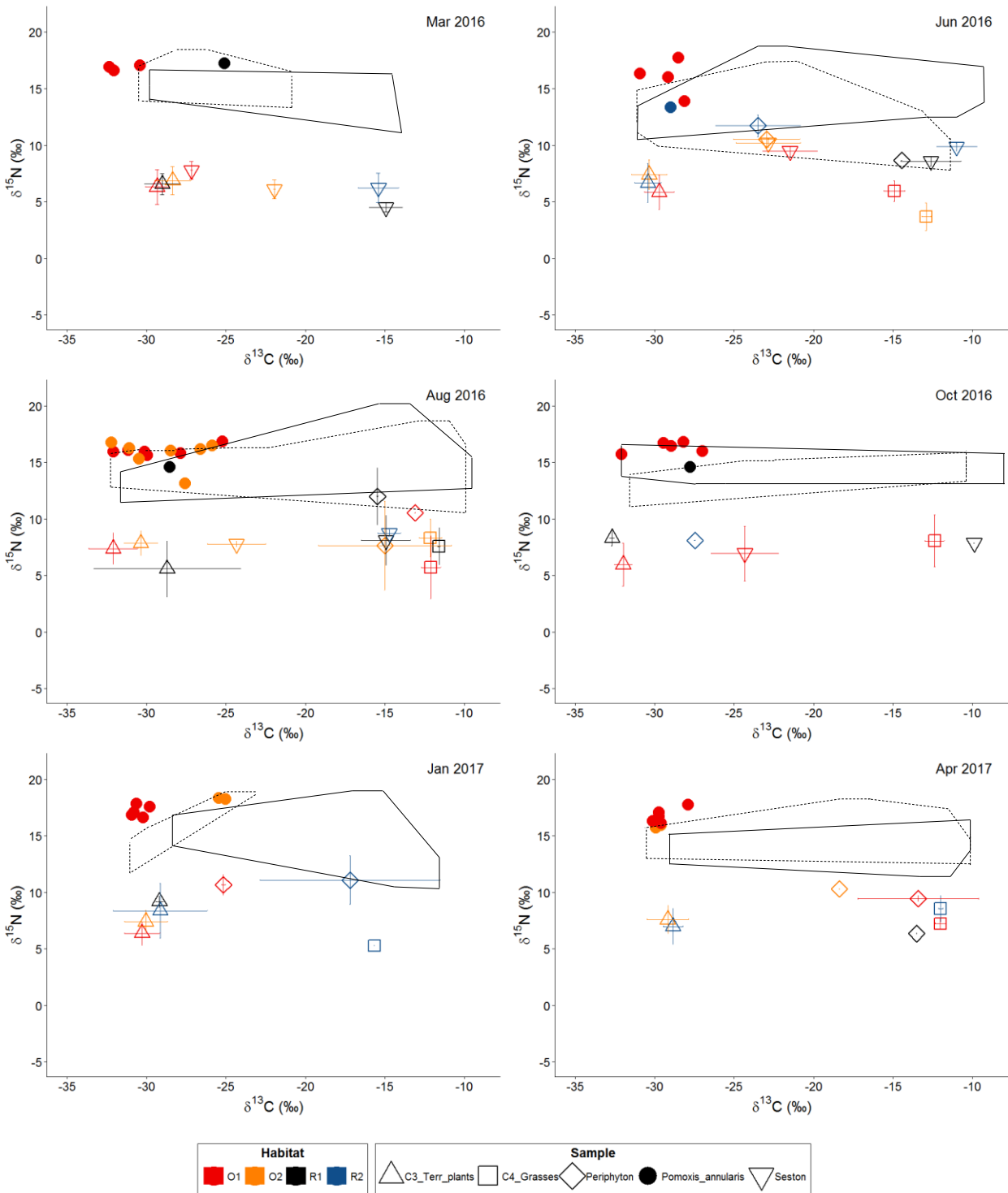


Figure 12. Biplots of carbon and nitrogen stable isotope ratio values for tissue samples of basal production sources and white crappie from oxbow and river channel sites. The isospace polygon defining feasible locations for a consumer assimilating some combination of material from basal production sources is shown for oxbows (dotted lines) and river channel (solid lines).

Mussels

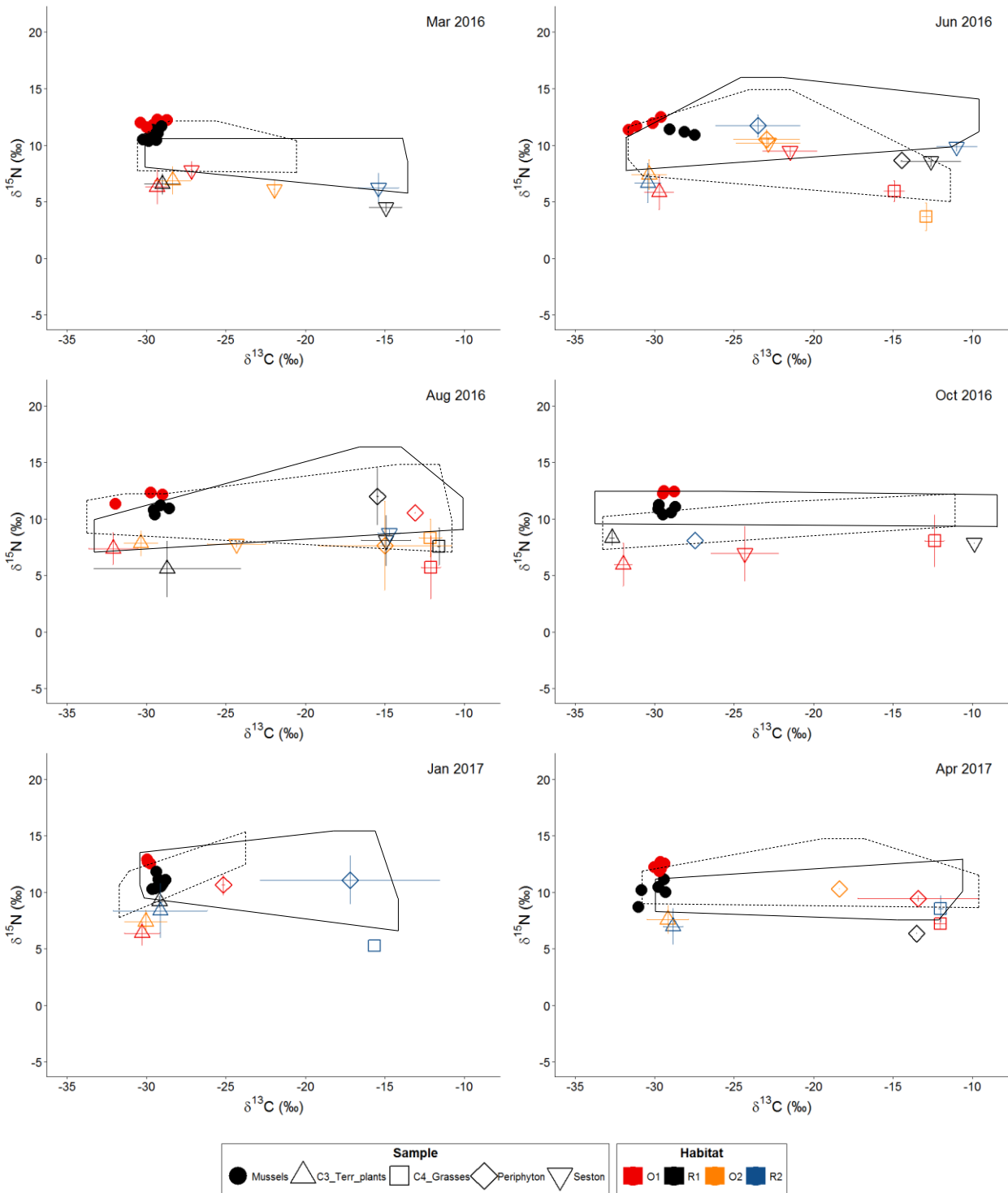


Figure 13. Biplots of carbon and nitrogen stable isotope ratio values for tissue samples of basal production sources and mussels from oxbow and river channel sites. The isospace polygon defining feasible locations for a consumer assimilating some combination of material from basal production sources is shown for oxbows (dotted lines) and river channel (solid lines).

evidence of spatial food web subsidy for specimens captured from oxbows as well as specimens captured from the river channel, but these findings varied according to survey period.

The most inferred cases of spatial food web subsidy (N= 20) were for oxbow resident organisms (threadfin shad, gizzard shad, sunfishes, white crappie, mussels) during October 2016 (Table 3), a period that followed one large and one small flow pulse that would have connected both oxbows (Figures 4 and 5). October 2016 was the only survey date that revealed oxbow organism having received a food web subsidy from the river channel. The second-most cases of inferred spatial food web subsidy (N= 12) were for organisms captured from the river channel (gizzard shad, bullhead minnow, sunfishes) during the August 2016 survey, and the third most cases (N= 9) were for organisms captured from the river channel (gizzard shad, bullhead minnow, smallmouth buffalo, sunfishes, white crappie) during the March 2016 survey. The other survey periods showing evidence of channel-resident organisms having assimilated basal source material originating from oxbows were April 2017 (3 cases involving threadfin shad, mussels) and January 2017 (2 cases involving bullhead minnow). The only survey period that did not reveal any strong inference of spatial food web subsidy was June-2016 (in addition, there were 25 unresolved cases among oxbow-resident organisms, and 35 unresolved cases among channel-resident organisms). The June 2016 survey followed the period with the recorded highest discharge and longest period of continuous lateral connection between oxbows and river channel (Figures 4 and 5).

Table 3. Summary of interpretations of basal production source contributions to consumers based on stable isotope biplots for fishes and mussels captured from oxbow lakes and river channel habitats of the lower Guadalupe River over six survey periods. *In situ* aquatic production refers to periphyton and/or seston from the same habitat type where the fish or mussel was captured; *ex situ* aquatic production refers to periphyton and/or seston from the opposite habitat type where the fish or mussel was captured. Numbers represent the number of specimens that fell inside (Y) or outside (N) of the polygon defining the isospace estimated to indicate assimilation of some proportion of basal sources from a given habitat as illustrated in Figures 8–13. In the case of *ex situ* sources, “Y” represents cases in which the consumer signatures fell inside the polygon for *ex situ* sources and outside the polygon for *in situ* sources; “N” represents cases in which the consumer signatures fell outside the polygon for *ex situ* sources; and “?” represents cases in which the consumer signatures fell inside both the *ex situ* and *in situ* sources polygons, and therefore the location of source material contributing to consumer biomass is undetermined.

Species	Survey	<i>In situ</i> aquatic production		<i>Ex situ</i> aquatic production	
		Oxbow	River	Oxbow	River
Longnose gar (<i>Lepisosteus osseus</i>)	Mar-16	5Y, 1N	2Y, 1N	2N, 4?	1N, 2?
	Jun-16	6Y, 1N	6Y	3N, 4?	6?
	Aug-16	4Y	1Y, 4N	3N, 1?	2N, 3?
	Oct-16	2Y, 2N	6Y, 3N	4?	3N, 6?
	Jan-17	9Y, 1N	6Y, 1N	5N, 5?	1N, 6?
	Apr-17	2Y, 2N	5Y, 4N	3N, 1?	5N, 4?
Gizzard shad (<i>Dorosoma cepedianum</i>)	Mar-16	10Y, 4N	1N	9N, 5?	1Y
	Jun-16	5Y, 2N	4Y, 1N	2N, 5?	1N, 4?
	Aug-16	8Y	4Y, 1N	4N, 4?	1Y, 4?
	Oct-16	2Y, 3N	4Y	1Y, 2N, 2?	1N, 3?
	Jan-17	2N	1N	1Y, 1N	1?
	Apr-17	2Y, 2N	3Y, 1N	2N, 2?	1N, 3?
Threadfin shad (<i>Dorosoma petenense</i>)	Mar-16	1N	1N	1N	1N
	Jun-16	-	-	-	-
	Aug-16	11Y	-	9N, 2?	-
	Oct-16	5N	-	5Y	-
	Jan-17	2Y, 7N	-	8N, 1?	-
	Apr-17	3Y, 5N	1Y, 1N	8N	1Y, 1N
Bullhead minnow (<i>Pimephales vigilax</i>)	Mar-16	2Y, 8N	10N	10N	1Y, 9N
	Jun-16	5Y	8Y, 1N	1N, 4?	1N, 8?
	Aug-16	-	9N	-	9Y
	Oct-16	-	9Y	-	9N
	Jan-17	3Y, 8N	4Y, 5N	11N	2Y, 5N, 2?
	Apr-17	3Y, 7N	6Y, 3N	10N	2Y, 3N, 4?
Smallmouth buffalo (<i>Ictiobus bubalus</i>)	Mar-16	3Y, 5N	3N	6N, 2?	3Y
	Jun-16	1Y, 5N	6Y, 2N	5N, 1?	2N, 6?
	Aug-16	4Y, 1N	3Y, 1N	4N, 1?	4?
	Oct-16	5N	3Y, 1N	3Y, 2?	1N, 3?
	Jan-17	6Y, 4N	2Y, 5N	5N, 5?	5N, 2?
	Apr-17	5Y, 3N	1Y, 3N	7N, 1?	3N, 1?

Table 3, continued

Sunfishes (<i>Lepomis</i> spp.)	Mar-16	2Y, 8N	5Y, 5N	10N	3Y, 2N, 5?
	Jun-16	9Y, 2N	7Y	5N, 6?	7?
	Aug-16	16Y	2Y, 4N	13N, 3?	2Y, 4?
	Oct-16	6N	11Y	6Y	5N, 6?
	Jan-17	6Y, 2N	8Y, 1N	4N, 4?	9?
	Apr-17	6Y, 8N	5Y, 2N	11N, 3?	1N, 6?
White crappie (<i>Pomoxis annularis</i>)	Mar-16	1Y, 2N	1N	3N	1Y
	Jun-16	2Y, 2N	1Y	3N, 1?	1?
	Aug-16	11Y, 2N	1Y	8N, 5?	1?
	Oct-16	5N	1Y	5Y	1?
	Jan-17	2Y, 5N	-	7N	-
	Apr-17	4Y, 3N	-	7N	-
Mussels (<i>Amblema plicata</i>) & (<i>Lampsilis teres</i>)	Mar-16	5Y	6Y, 3N	5N	9?
	Jun-16	4Y	3Y	4?	3?
	Aug-16	3Y	5Y	3N	5?
	Oct-16	3N	5Y	3Y	5?
	Jan-17	3Y	5Y	3?	5?
	Apr-17	5Y	3Y, 2N	5N	2Y, 3?
	Total Y	181	152	21	26
	Total N	130	79	205	63
	Total ?	-	-	85	142

Fish Assemblage Structure

Ordination of fish samples using NMDS revealed a strong pattern of separation of fish assemblage samples based on habitat type, with river samples more tightly clustered with the two locations overlapping, and two oxbow sites separated in assemblage space (Figure 14). The stress value of 0.167 indicates that the NMDS ordination was fairly robust with few misleading distance estimates. The NMDS ordination did not separate fish assemblage samples according to seasons (Figure 15). PERMANOVA indicated that there was a statistically significant effect of habitat type on assemblage structure, but no significant effect of season or the interaction of habitat and season (Table 4). Oxbow assemblage samples tended to have more spotted gar, gizzard shad, threadfin shad, inland silverside, orangespotted sunfish, black crappie, white crappie, and bluntnose darter; whereas, river channel assemblage samples tended to have more channel catfish, Rio Grande cichlid, green sunfish, burrhead chub, ghost shiner, mimic shiner, red shiner, Texas shiner, and dusky darter (Table 5). Differences in the fish assemblages of Oxbow 1 and Oxbow 2 were strongly influenced by greater abundance of inland silverside and bluntnose darter in the former, and greater abundance of pugnose minnow, blue catfish and black crappie in the latter.

CCA computed using matrices of species abundance x survey site/period data and environmental variables x survey site/period yielded a different ordination pattern than NMDS based on species abundance x site/period alone. CCA axis 1 accounted for 0.37 percent of the variance modeled by explanatory variables, and CCA axis 2 accounted for 0.24 percent (total percent of variance modeled by the two axes = 0.61). Similar to NMDS results, the CCA separated surveys according to habitat type, with greater overlap between the two river channel sites compared to the oxbow sites (Figure 16). However, a difference between the two ordination results was that CCA produced several extreme outliers (NMDS uses a ranking technique that tends to reduce outlier effects), especially for oxbow 2. Oxbow 2 was the site that underwent greatest changes in water depth and water quality factors over the course of the field study. River site 1 also had one outlier (June 2016, the survey that followed the largest flow pulse of longest duration). Unlike NMDS ordination, CCA resulted in strong separation of surveys according to seasons (Figure 17). Clearly, this seasonal separation was strongly influenced by abiotic variables, such as temperature and dissolved oxygen concentration, that varied according to seasons.

Samples that clustered near the origins of NMDS axes represent species assemblages with greatest similarity. High similarity between oxbow and river assemblages would be expected during periods of extensive lateral hydrologic connectivity during or immediately following high flow pulses. Of the five samples clustered nearest to the origins of the two NMDS axes, four were taken from the four sites during survey period that occurred during the end of the largest flood pulse with longest duration and longest period of lateral connectivity for both oxbows. This indicates extensive exchange of fishes between the river channel and oxbow lakes during this period of lateral connectivity. The other survey that had high similarity between the river and oxbow samples was Oxbow 2 during March 2016, a period that occurred at the end of a moderate flow pulse that had connected the oxbow for 8 of the previous 15 days.

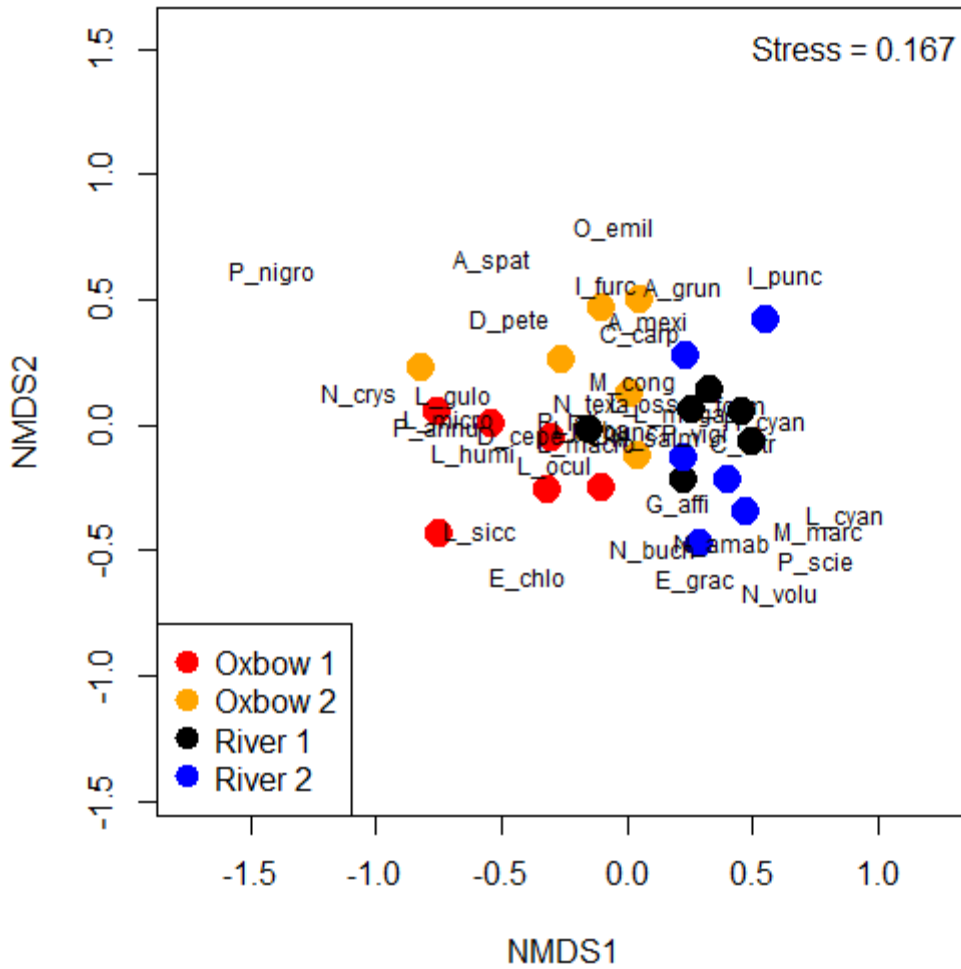


Figure 14. NMDS ordination plot showing relative positions of fish samples from four locations within assemblage space. Abbreviations indicate fish species, and locations of abbreviations in the ordination space indicate the strength of association with the two NMDS axes. Abbreviations are defined in Table 6.

Table 5. The abundance (A) and relative abundance (R) for fish species by survey site and across all survey periods.

Family	Species	Oxbow 1		Oxbow 2		River 1		River 2	
		A	R	A	R	A	R	A	R
Atherinopsidae	<i>Labidesthes sicculus</i>	18	0.006	2	0.001	1	<0.001	0	0
Catostomidae	<i>Moxostoma congestum</i>	0	0	2	0.001	1	<0.001	0	0
	<i>Ictiobus bubalus</i>	95	0.031	29	0.020	28	0.004	17	0.015
Centrarchidae	<i>Lepomis cyanellus</i>	0	0	0	0	1	<0.001	1	0.001
	<i>Lepomis gulosus</i>	27	0.009	30	0.021	4	0.001	2	0.002
	<i>Lepomis humilis</i>	904	0.293	32	0.022	12	0.002	7	0.006
	<i>Lepomis macrochirus</i>	700	0.227	12	0.085	67	0.009	31	0.027
	<i>Lepomis megalotis</i>	10	0.003	36	0.025	28	0.004	42	0.036
	<i>Lepomis microlophus</i>	47	0.015	7	0.005	13	0.002	0	0
	<i>Micropterus punctulatus</i>	1	<0.001	4	0.003	2	<0.001	1	0.001
	<i>Micropterus salmoides</i>	2	0.001	8	0.006	1	<0.001	4	0.003
	<i>Pomoxis annularis</i>	357	0.116	32	0.224	3	<0.001	1	0.001
	<i>Pomoxis nigromaculatus</i>	0	0	15	0.010	0	0	0	0
Characidae	<i>Astyanax mexicanus</i>	21	0.007	88	0.061	35	0.005	24	0.021
Cichlidae	<i>Herichthys cyanoguttatus</i>	0	0	1	0.001	3	<0.001	2	0.002
Clupeidae	<i>Dorosoma cepedianum</i>	363	0.117	14	0.103	22	0.003	0	0
	<i>Dorosoma petenense</i>	79	0.026	15	0.110	0	0	9	0.008
Cyprinidae	<i>Cyprinella lutrensis</i>	77	0.025	66	0.046	664	0.886	394	0.337
	<i>Cyprinus carpio</i>	0	0	1	0.001	0	0	0	0
	<i>Macrhybopsis marconis</i>	0	0	0	0	2	<0.001	9	0.008
	<i>Notemigonus crysoleucas</i>	1	<0.001	2	0.001	0	0	0	0
	<i>Notropis amabilis</i>	2	0.001	0	0	0	0	22	0.019
	<i>Notropis buchanani</i>	11	0.004	1	0.001	0	0	15	0.013
	<i>Notropis shumardi</i>	1	<0.001	0	0	0	0	0	0
	<i>Notropis texanus</i>	0	0	2	0.001	18	0.002	0	0
	<i>Notropis volucellus</i>	0	0	11	0.008	0	0	90	0.077
	<i>Opsopoedus emiliae</i>	0	0	5	0.003	0	0	0	0
	<i>Pimephales vigilax</i>	130	0.042	20	0.144	223	0.030	293	0.251
Fundulidae	<i>Fundulus notatus</i>	0	0	0	0	0	0	1	0.001
Ictaluridae	<i>Ictalurus furcatus</i>	3	0.001	24	0.017	1	<0.001	1	0.001
	<i>Ictalurus punctatus</i>	0	0	0	0	1	<0.001	15	0.013
Lepisosteidae	<i>Atractosteus spatula</i>	1	<0.001	10	0.007	0	0	0	0
	<i>Lepisosteus oculatus</i>	9	0.003	6	0.004	0	0	3	0.003
	<i>Lepisosteus osseus</i>	53	0.017	26	0.018	63	0.008	31	0.027
Percidae	<i>Etheostoma chlorosomum</i>	79	0.026	1	0.001	0	0	0	0
	<i>Etheostoma gracile</i>	2	0.001	0	0	0	0	4	0.003
	<i>Percina sciera</i>	0	0	0	0	0	0	1	0.001
Poeciliidae	<i>Gambusia affinis</i>	81	0.026	62	0.043	300	0.040	147	0.126
	<i>Poecilia formosa</i>	1	<0.001	0	0	4	0.001	0	0
	<i>Poecilia latipinna</i>	15	0.005	2	0.001	17	0.002	1	0.001
Sciaenidae	<i>Aplodinotus grunniens</i>	0	0	7	0.005	2	<0.001	0	0

Table 6. List of fish scientific and common names and species abbreviations.

Species name	Common name	Abbreviation
<i>Aplodinotus grunniens</i>	Freshwater drum	A_grun
<i>Astyanax mexicanus</i>	Mexican tetra	A_mex
<i>Atractosteus spatula</i>	Alligator gar	A_spat
<i>Cyprinella lutrensis</i>	Red Shiner	C_lutr
<i>Cyprinus carpio</i>	Common carp	C_carp
<i>Dorosoma cepedianum</i>	American gizzard shad	D_cepe
<i>Dorosoma petenense</i>	Threadfin shad	D_pete
<i>Etheostoma chlorosomum</i>	Bluntnose darter	E_chlo
<i>Etheostoma gracile</i>	Slough darter	E_grac
<i>Gambusia affinis</i>	Western Mosquitofish	G_affi
<i>Herichthys cyanoguttatus</i>	Texas cichlid	H_cyan
<i>Ictalurus furcatus</i>	Blue catfish	I_furc
<i>Ictalurus punctatus</i>	Channel catfish	I_punc
<i>Ictiobus bubalus</i>	Smallmouth buffalo	I_buba
<i>Labidesthes sicculus</i>	Brook silverside	L_sicc
<i>Lepisosteus oculatus</i>	Spotted gar	L_ocul
<i>Lepisosteus osseus</i>	Longnose gar	L_osse
<i>Lepomis cyanellus</i>	Green sunfish	L_cyan
<i>Lepomis gulosus</i>	Warmouth	L_gulo
<i>Lepomis humilis</i>	Orangespotted sunfish	L_humi
<i>Lepomis macrochirus</i>	Bluegill	L_macro
<i>Lepomis megalotis</i>	Longear sunfish	L_mega
<i>Lepomis microlophus</i>	Redear sunfish	L_micro
<i>Macrhybopsis marconis</i>	Burrhead chub	M_marc
<i>Micropterus punctulatus</i>	Spotted bass	M_punc
<i>Micropterus salmoides</i>	Largemouth bass	M_salm
<i>Moxostoma congestum</i>	Gray redhorse	M_cong
<i>Notemigonus crysoleucas</i>	Golden shiner	N_crys
<i>Notropis amabilis</i>	Texas shiner	N_amab
<i>Notropis buchmanani</i>	Ghost shiner	N_buch
<i>Notropis texanus</i>	Weed shiner	N_texa
<i>Notropis volucellus</i>	Mimic shiner	N_volu
<i>Opsopoeodus emiliae</i>	Pugnose minnow	O_emil
<i>Percina sciera</i>	Dusky darter	P_scie
<i>Pimephales vigilax</i>	Bullhead minnow	P_vigi
<i>Poecilia formosa</i>	Amazon molly	P_form
<i>Poecilia latipinna</i>	Sailfin molly	P_lati
<i>Pomoxis annularis</i>	White crappie	P_annu
<i>Pomoxis nigromaculatus</i>	Black crappie	P_nigro

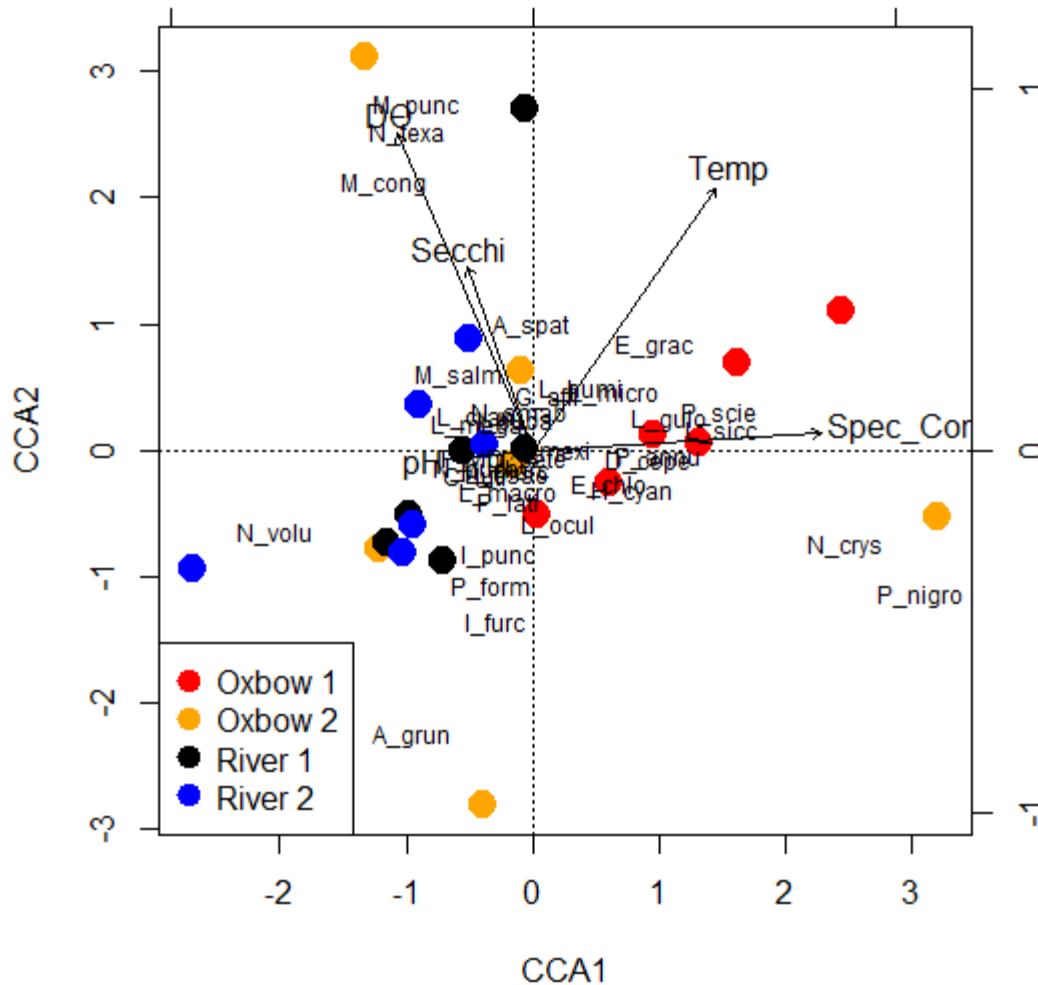


Figure 16. CCA ordination plot showing relative positions within assemblage-environment space of fish samples from four habitats. Abbreviations indicate fish species and environmental variables; locations of abbreviations and tips of vectors (the latter only for environmental variables) in the ordination space indicate the loadings of the variables on the two axes. Abbreviations are defined in Table 6.

Discussion

Our field study was performed during a 12-month period that had higher precipitation and discharge compared to the long-term average for Victoria, Texas. Rainfall was above average during March, April, May, August, November and December 2016, as well as January–April 2017). As a result, there were frequent high flow pulses in the lower Guadalupe River during the study period, and all but one of our six surveys were conducted at the termination of a high flow pulse that connected both oxbows that were selected for study. Only the August 2016 survey was conducted following a period of baseflow, and even for that survey, the two oxbows were estimated to have been connected to the river channel with at least a few centimeters of water during 5 of the previous 30 days (Oxbow 1 near Cuero) and 9 of the previous 30 days (Oxbow 2 near Victoria). The longest continuous period of baseflow without lateral connections estimated for the two oxbows occurred from October 5 to December 5, 2016.

Analysis of fish assemblage data indicated differences in the composition between oxbow and river channel habitats, and greater variation within and between samples from oxbows. This finding is consistent with findings from research on the lower Brazos River, Texas, wherein fish assemblages were shown to be more divergent among oxbow lakes compared to sites surveyed in the river channel (Winemiller et al. 2000, Zeug et al. 2005). Fish assemblages in oxbows also varied more over time compared to those in the river channel, and this was observed in both the lower Guadalupe (current report) and lower Brazos (Zeug et al. 2005). In both of these studies, the greatest similarity in fish assemblage composition occurred during or immediately following high flow pulses that connected the oxbows to the river. This is interpreted as exchange of fishes, with some moving from the channel into the flooded oxbows, and others exiting from oxbows and entering the river channel. In the lower Brazos, oxbows were found to be much more productive habitats than the river channel, and the Guadalupe is likely to be the same.

Roach et al. (2014) examined aquatic ecosystem primary production in the lower Guadalupe River between March 2010 and March 2011, a period that encompassed a severe drought and relatively low flows. Turbidity in the lower Guadalupe River was consistently low during that period (maximum turbidity was 367 FTU), and littoral habitats were consistently net autotrophic. In contrast, the Brazos River had turbidity ranging as high as 1,474 FTU, and the littoral areas were net heterotrophic during periods of high flow and turbidity. They inferred that light attenuation under conditions of high turbidity reduced algal production, with benthic algae affected to a greater degree than phytoplankton. Roach and Winemiller (2015) analyzed stable isotopes of hydrogen deuterium using a Bayesian mixing model to estimate contributions of algae versus terrestrial plants to consumers during different hydrologic phases in the Neches, Brazos, and Guadalupe rivers in Texas. In the Brazos and Guadalupe rivers, flow pulses increase turbidity, and assimilation of basal source material by consumers varied according to discharge stage. In these rivers, algae made greater contributions to macroinvertebrates and fish biomass during low-flow periods, and terrestrial plants made greater contributions during high-flow pulses. During flow pulses, lower algal biomass and production, combined with increased inputs of terrestrial organic matter from watersheds and riparian habitats, appears to result in greater inputs of terrestrial material into aquatic food chains. These patterns are consistent with predictions of the River Wave Concept (Humphries et al. 2014), which posits that flow is the key process determining the source of organic matter assimilated by higher consumers in rivers.

Although we did not measure aquatic primary production and respiration rates during the current project, shallow oxbow lakes generally have higher aquatic production than river channel

habitats (Hunt et al. 2012). Oxbows are shallow and generally lack flow, the exception being periods of high flow pulsing when water enters from the river channel. As in previous studies of oxbow fish assemblages in the Brazos River (Winemiller et al. 2000, Zeug et al. 2005, Zeug and Winemiller 2007, 2008a) and other parts of North America (Miranda 2011) and the world (Jardine et al. 2012), we found that these lentic floodplain habitats support high densities of juvenile fishes of several species. Therefore, floodplain lakes appear to have an important nursery function for many fishes. Productive oxbow habitats export large numbers of young fishes into river channels where they likely play a significant role in stock recruitment. The significance of oxbows and other lentic backwater habitats for recruitment dynamics of regional fish stocks would depend on the magnitude, duration and frequency of flow pulses causing lateral connections of varying duration, the productivity of the habitats, and the number of habitats in the floodplains.

Our project did not perform a comprehensive survey of oxbow lakes in the lower Guadalupe River; however, a recent project performed by Biowest, Inc. (Bonner et al. 2017) investigated five oxbow lakes between Gonzales and Victoria, Texas. Their findings for fish assemblage composition in oxbows were largely consistent with findings from the current study, with relatively lentic-adapted species (e.g., pugnose minnow, orangespotted sunfish, white crappie, bluntnose darter) frequently captured from oxbows, while being uncommon in samples from littoral habitats of the river channel. The Biowest study also emphasized the apparent role of oxbows as nursery habitats that enhance stock recruitment. For lentic-adapted fish species of the lower Guadalupe River, oxbows might support source subunits of regional metapopulations, in which case, it would be essential to have periodic flooding of oxbows to maintain habitat as well as connections that allow entry of spawners or larvae from the river and export of young fish back to the river. The river channel is the ultimate source pool for aquatic organisms, and this is because oxbows occasionally dry up. Nonetheless, these oxbows eventually are recolonized during the next high flow pulse that reconnects and reestablishes aquatic habitat.

Food web subsidies were apparent from our analysis of stable isotope data – at a minimum 7% of fishes and mussels captured from oxbows and 11% of those captured from the river channel. However, these estimates should be considered conservative, because many consumer data were inconclusive (i.e., the consumer's isotopic signature positioned it within both the *in situ* and *ex situ* isospace polygons). According to our graphical analysis of stable isotope data, most fishes and mussels biomass appeared to be derived from riparian plants; and among the available aquatic production sources (seston and/or periphyton), *in situ* sources were inferred to be more important in most cases.

Inference of production sources supporting consumer biomass based on stable isotope analysis relies on several assumptions. There are several potential confounding factors for analyses of stable isotopes to infer assimilation of sources from different spatial units. These include 1) assumptions for the trophic fractionation of isotopic ratios of elements (Bunn et al. 2013, Bastos et al. 2017), 2) the possibility that important sources are missing from the dataset (Rubenstein and Hobson 2004), assumptions about tissue turnover time (Fry 2006, Rubenstein and Hobson 2004, Bunn et al. 2013), insufficient sample sizes given levels of between-sample isotopic variation (Rubenstein and Hobson 2004, Jardine et al. 2005, 2014), and other factors (Hopkins and Ferguson 2012). These potentially confounding issues could have affected our findings. Given these uncertainties, we did not use an isotopic mixing model to make quantitative estimates of consumer assimilation of alternative sources. The Bayesian modeling approach yields probability distributions for percent contributions of sources – and our dataset was certain to yield extremely broad distributions that overlap extensively. Therefore, we adopted a graphical approach (Fry 2006) to evaluate the

potential for assimilation of material from *in situ* versus *ex situ* basal sources.

It is important to note that our field study was conducted during a period of relatively high regional rainfall and discharge in the lower Guadalupe River, and the river channel and most oxbows were turbid with suspended sediments during every survey. Consequently, the aquatic food webs of both habitats apparently were supported predominantly by basal production in the form of riparian vegetation, as discussed above and consistent with other studies as well as the River Wave Concept (Humphries et al. 2014). During periods of base-flow conditions, it is likely that oxbow lakes are more productive than littoral habitats of similar depths within the river channel. Thus, the importance of aquatic production of oxbows for supporting fish and macroinvertebrate biomass in the river channel was probably greatly underestimated by our study.

The research approach adopted here should be used to investigate the issue of spatial food web subsidies during intervals that encompass more extended periods of base-flow conditions with lower turbidity and greater aquatic primary production. Future studies also could examine more oxbows, and, building on the findings from the current study, develop specific hypotheses that target certain taxa and locations. Inferences about spatial food web subsidies would be enhanced by simultaneous measurement of aquatic ecosystem primary production and respiration (to infer net ecosystem productivity) in the water column and benthos within oxbow and river channel habitats. In addition, estimates of fish and mussel growth rates could establish relationships between hydrology, food web dynamics and organism performance in various habitats. Growth rate history can be estimated from analysis of hard structures, including fish otoliths (Rodger et al. 2016) and mussel shells in cross section (Rypel et al. 2009). Of course, more analysis of hydrology, geomorphology (bank, oxbow and connecting channel elevation), and lateral connectivity (magnitude, frequency and timing of connections) is needed to better inform inferences from ecological research. With more findings on hydrology, lateral connectivity and aquatic community and food web dynamics, computer models could be developed to simulate ecosystem dynamics. Such models would allow scientists and policymakers to evaluate alternative water management scenarios.

Implications for an Environmental Flow Regime for Lower Guadalupe River

Given the uncertainties associated with our analysis of stable isotope data, together with the fact that our study was conducted during a relatively wet period with multiple high flow pulses, high turbidity, and aquatic food webs dominated by inputs from riparian plants, any recommendations to revise current environmental flow standards for the lower Guadalupe River would be tenuous. Conservative inferences of spatial food web subsidies, in both directions, were made based on the isotopic analysis. Our analysis of fish assemblage structure corroborates the findings and supports the recommendations from the recent study by Biowest (Bonner et al. 2017).

We estimated nine high flow pulses that significantly connected the two oxbow lakes to the river channel during the 12 months of our field study. Our estimates of the discharge (1,630 cfs) at the USGS gage at Cuero that connects Oxbow 1 with the river channel closely match the estimates of Biowest for this same oxbow ('Cuero 2' in their report). They estimated that based on long-term hydrological records for the USGS gage at Cuero, Oxbow 1 should connect with the river channel 6.6 times per year on average. Biowest did not survey our Oxbow 2 near Victoria; however, their study surveyed two other oxbows near Victoria. They estimated that both of those oxbows would be connected under base-flow conditions (290 and 144 cfs). In contrast, Oxbow 2 in our study had a lateral connection only at flows exceeding about 1,700 cfs.

According to the current environmental flow standards adopted by the state for the Cuero gage (TCEQ 2012), 1,630 cfs is achieved by the following seasonal flow pulses recommended for protection (Figure 18): 2-per-season pulse in spring, summer and fall; and 1-per-season pulse in all four seasons. Given the current environmental flow standards for the Victoria gage, 1,700 cfs (the flow that connects this oxbow to the river channel) is achieved by the following seasonal flow pulses recommended for protection (Figure 19): 2-per-season pulse only during spring and fall; and 1-per-season pulse in all four seasons.

The TCEQ essentially adopted the recommendations of the science team that studied environmental flow needs of the river (Guadalupe, San Antonio, Mission, and Aransas Rivers and Mission, Copano, Aransas and San Antonio Bay and Basin Expert Science Team 2011) for the Cuero gage, with the exception that no overbanking flows were protected for environmental functions (Figure 20). These high magnitude and duration overbanking flow pulses have much lower frequencies, and also occur less predictably than lower pulse tiers within environmental flow regime. The TCEQ did not fully adopt the environmental flow regime recommendations of the basin's expert science team for the Victoria gage. The current TCEQ standards have lower magnitude 1-per-season flows than those recommended by the science team (Figure 20), especially for spring (from 9020 cfs to 3240 cfs) and also for fall (from 4620 cfs to 3240 cfs) and winter (from 5370 cfs to 3240 cfs). Whereas the flow pulses in the current standards provide for lateral connectivity of Oxbow 2, the frequency and duration of connections likely would be significantly reduced relative to the duration that would be provided by the flow pulses recommended by the basin's science team. Of course, this inference assumes that future infrastructure for water diversion or storage would be capable of significantly impacting flows. Hydrologic modeling is required to obtain estimates of discharge under various scenarios involving flow standards and implementation rules for water storage or diversion by water right holders. This project did not evaluate ecological functions of overbanking flows, which are not protected under the current environmental flow standards. High flow pulses at tiers lower than those currently classified as "overbanking" are probably most beneficial to aquatic organisms with regard to lateral connectivity during spring, followed by summer and fall. The small reduction in lateral connection during winter relative to historical long-term hydrology may have little ecological consequence, although this assessment is tentative given the limited ecological research conducted on this system to date. The flow standards for the USGS gage at Victoria would provide for multiple annual connections of Oxbow 2; however, the magnitude and duration of the higher-tier flow pulse (1/season) during spring may be insufficient and likely would result in reduced frequencies, magnitudes and durations of connections during a season when lateral connectivity is most important for between-habitat exchanges of pre-spawning and post-spawning adults and juveniles. The one-per-season flow pulse during fall in the current flow standards for the Victoria gage also may be insufficient, especially with regard to lateral connectivity facilitating movement of juvenile fishes to the river and spatial food web subsidies for the river channel. The small reduction in lateral connection during winter relative to historical long-term hydrology may have relatively less ecological consequence, although this is merely conjecture given the limited ecological research conducted on this system to date, and the need for hydrologic modeling to estimate the potential influence of flow standards implementation.

Season	Subsistence	Base	Small Seasonal Pulse (2 per season)	Large Seasonal Pulse (1 per season)
Winter	130 cfs	980 cfs	Trigger: 1,610 cfs Volume: 14,100 af Duration: 13 days	Trigger: 4,610 cfs Volume: 55,300 af Duration: 26 days
Spring	120 cfs	940 cfs	Trigger: 3,370 cfs Volume: 31,800 af Duration: 18 days	Trigger: 8,870 cfs Volume: 100,000 af Duration: 30 days
Summer	130 cfs	800 cfs	Trigger: 1,050 cfs Volume: 8,300 af Duration: 12 days	Trigger: 2,110 cfs Volume: 19,300 af Duration: 17 days
Fall	86 cfs	870 cfs	Trigger: 1,730 cfs Volume: 14,100 af Duration: 13 days	Trigger: 5,200 cfs Volume: 54,700 af Duration: 23 days

Figure 18. Current environmental flow standards adopted by the Texas Commission on Environmental Quality for the lower Guadalupe River at the USGS gage at Cuero (from TCEQ 2012).

Season	Subsistence	Base	Small Seasonal Pulse (2 per season)	Large Seasonal Pulse (1 per season)
Winter	160 cfs	975 cfs	Trigger: 1,690 cfs Volume: 14,400 af Duration: 13 days	Trigger: 3,240 cfs Volume: 33,000 af Duration: 18 days
Spring	130 cfs	945 cfs	Trigger: 3,240 cfs Volume: 33,000 af Duration: 18 days	Trigger: 3,240 cfs Volume: 43,500 af Duration: 25 days
Summer	150 cfs	795 cfs	Trigger: 1,040 cfs Volume: 8,570 af Duration: 11 days	Trigger: 2,060 cfs Volume: 19,200 af Duration: 16 days
Fall	110 cfs	865 cfs	Trigger: 1,880 cfs Volume: 15,600 af Duration: 13 days	Trigger: 3,240 cfs Volume: 35,500 af Duration: 23 days

Figure 19. Current environmental flow standards adopted by the Texas Commission on Environmental Quality for the lower Guadalupe River at the USGS gage at Victoria (from TCEQ 2012).

Table 6.1-8. – Environmental Flow Regime Recommendation, Guadalupe River at Cuero

Overbank Flows	Qp: 45,400 cfs with Average Frequency 1 per 5 years Regressed Volume is 869,000 Duration Bound is 91											
	Qp: 24,700 cfs with Average Frequency 1 per 2 years Regressed Volume is 406,000 Duration Bound is 64											
	Qp: 16,600 cfs with Average Frequency 1 per year Regressed Volume is 247,000 Duration Bound is 50											
High Flow Pulses	Qp: 4,610 cfs with Average Frequency 1 per season Regressed Volume is 55,300 Duration Bound is 26			Qp: 8,870 cfs with Average Frequency 1 per season Regressed Volume is 110,000 Duration Bound is 32			Qp: 2,110 cfs with Average Frequency 1 per season Regressed Volume is 19,300 Duration Bound is 17			Qp: 5,200 cfs with Average Frequency 1 per season Regressed Volume is 54,700 Duration Bound is 23		
	Qp: 1,610 cfs with Average Frequency 2 per season Regressed Volume is 14,100 Duration Bound is 13			Qp: 3,370 cfs with Average Frequency 2 per season Regressed Volume is 31,800 Duration Bound is 18			Qp: 1,050 cfs with Average Frequency 2 per season Regressed Volume is 8,300 Duration Bound is 12			Qp: 1,730 cfs with Average Frequency 2 per season Regressed Volume is 14,100 Duration Bound is 13		
Base Flows (cfs)	980			940			800			870		
	760			680			600			670		
	550			410			390			480		
Subsistence Flows (cfs)	130			120			130			86		
	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
	Winter			Spring			Summer			Fall		
Flow Levels	High (75th %ile)											
	Medium (50th %ile)											
	Low (25th %ile)											
	Subsistence											

Table 6.1-9. – Environmental Flow Regime Recommendation, Guadalupe River at Victoria

Overbank Flows	Qp: 48,000 cfs with Average Frequency 1 per 5 years Regressed Volume is 971,000 Duration Bound is 96											
	Qp: 25,500 cfs with Average Frequency 1 per 2 years Regressed Volume is 438,000 Duration Bound is 66											
	Qp: 16,700 cfs with Average Frequency 1 per year Regressed Volume is 257,000 Duration Bound is 51											
High Flow Pulses	Qp: 4,620 cfs with Average Frequency 1 per season Regressed Volume is 56,100 Duration Bound is 26			Qp: 9,020* cfs with Average Frequency 1 per season Regressed Volume is 119,000 Duration Bound is 34			Qp: 2,060 cfs with Average Frequency 1 per season Regressed Volume is 19,200 Duration Bound is 16			Qp: 5,370 cfs with Average Frequency 1 per season Regressed Volume is 57,800 Duration Bound is 23		
	Qp: 1,690 cfs with Average Frequency 2 per season Regressed Volume is 14,400 Duration Bound is 13			Qp: 3,300 cfs with Average Frequency 2 per season Regressed Volume is 33,000 Duration Bound is 18			Qp: 1,040 cfs with Average Frequency 2 per season Regressed Volume is 8,570 Duration Bound is 11			Qp: 1,880 cfs with Average Frequency 2 per season Regressed Volume is 15,600 Duration Bound is 13		
Base Flows (cfs)	1,050			1,020			870			940		
	800			710			630			720		
	580			450			420			510		
Subsistence Flows (cfs)	160			130			150			110		
	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec
	Winter			Spring			Summer			Fall		
Flow Levels	High (75th %ile)											
	Medium (50th %ile)											
	Low (25th %ile)											
	Subsistence											

Figure 20. Environmental flow regime recommendations of the Guadalupe, San Antonio, Mission, and Aransas Rivers and Mission, Copano, Aransas and San Antonio Bay and Basin Expert Science Team 2011 for the lower Guadalupe River at USGS gages at Cuero (top) and Victoria (bottom).

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Appendix A

Responses to Required Changes to Task 2 Report

1. Please reference “TWDB Contract No. 1448311791” on the cover of the report.

Response: addressed

2. Please check the report for typos such as the following and correct as necessary:
 - a. Page 1, 1st paragraph, 5th sentence, “food web also are” should be “food web dynamics are also.”
 - b. Page 1, 1st paragraph, last sentence, “this studied also analyzed” should be “this study also analyzed.”
 - c. Page 5, 1st paragraph, 4th sentence, “would achieved by” should be “would be achieved by.”
 - d. Page 6, 3rd paragraph, 4th sentence, “are influence by” should be “are influenced by.”
 - e. Page 7, 2nd paragraph, 3rd sentence, “for analysis ratios” should be “for analysis of ratios.”
 - f. Page 8, 1st paragraph, 2nd to last sentence, “evaluate the frequency” should be “evaluated the frequency.”
 - g. Page 12, 3rd paragraph, 1st sentence, “found both the river” should be “found in both the river.”
 - h. Page 12, 3rd paragraph, 4th sentence, “replicate tissues samples” should be “replicate tissue samples.”
 - i. Page 13, 1st paragraph, 2nd sentence, “samples fine particulate” should be “samples of fine particulate.”
 - j. Page 14, 4th paragraph, 1st sentence, “consumers and were used” should be “consumers were used.”
 - k. Page 16, 1st paragraph, last sentence, “exchanges during at the two locations” should be “exchanges at the two locations.”
 - l. Page 16, 2nd paragraph, 1st sentence, “canonical correspond analysis” should be “canonical correlation analysis.”
 - m. Page 16, 2nd paragraph, 2nd sentence, “among of environmental gradients” should be “among environmental gradients.”
 - n. Page 17, 1st paragraph, 2nd sentence, “a pseudo-F values” should be “a pseudo-F value.”
 - o. Page 21, 1st paragraph, 8th sentence, “that that recorded” should be “that recorded.”
 - p. Page 22, 1st paragraph, 2nd sentence, “from an ability” should be “from an inability.”
 - q. Page 33, 2nd paragraph, last sentence, “according survey” should be “according to survey.”
 - r. Page 41, Table 6 title, “list fish” should be “list of fish.”
 - s. Page 45, 2nd paragraph, 3rd sentence, “have had been” should be “have been.”
 - t. Page 45, 2nd paragraph, last sentence, “occurred during from” should be “occurred from.”
 - u. Page 48, 3rd paragraph, 3rd sentence, “bas-flow” should be “base-flow.”
 - v. Page 50, 2nd paragraph, 1st sentence, “according the current” should be “according to the current.”
 - w. Page 50, 2nd paragraph, 2nd sentence, “connections with Oxbow 2” should be connections with Oxbow 1.”

Response: all addressed

3. Please provide definitions of all acronyms before they are used in the text, for example:
 - a. “USGS” on page 3.
 - b. “TPWD” on page 9.
 - c. “AUP” on page 12.
 - d. “FPOM” on page 13.

Response: all addressed

4. Please provide references for the following statements:
 - a. Page 6, 2nd paragraph: “For example, in the lower Brazos River, Texas, periodic high flow pulses result in temporary connections between the river channel and oxbow lakes that provide important habitat for several fish species that normally are uncommon within the active river channel, but often attain high densities in oxbow lakes.”
 - b. Page 6, 3rd paragraph: “In the lower Guadalupe River, turbidity increases during high flow pulses and this has been shown to reduce aquatic primary productivity.”
 - c. Page 6, 3rd paragraph: “According to a prior study, the biomass of several common fish species inhabiting the lower Guadalupe River channel seems to be more dependent on riparian vegetation than algae during high flow pulses.”

Response: all addressed

5. On page 17, 3rd paragraph, the authors estimate the discharge required to connect Oxbow 1 as 1,630 cfs. On page 49, 3rd paragraph, they give an estimate of 1,710 cfs to connect Oxbow 1. Please reconcile these two different estimates for the flow required to provide connectivity to the oxbow.

Response: addressed (corrected to 1,630 cfs on p. 49)

6. An assessment of the current standards, as attempted on pages 50 to 54 of the report, is beyond the scope of work of this project. However, it would be beneficial to have the insight of the authors (who are very familiar with river-floodplain ecosystems and their requirements) regarding the ability of the current standards to protect a sufficient number of oxbow-main channel connections to maintain the current status of the lower Guadalupe River. Unfortunately, they appear to have greatly exaggerated the number of days of oxbow connection protected by the current standards.

In their assessment of the standards, the authors have neglected two important factors. First, the standards have a volume cut-off (displayed in Figure 18 and 19). As stated by the standard: *The water right holder shall not divert or hold water except during times that streamflow at the applicable measurement point exceeds the applicable high flow pulse trigger level and until either the applicable volume amount has passed the measurement point or the applicable duration time has passed since the high flow pulse trigger level occurred* (30 Texas Administrative Code 298.375d1). In practice, if flows remain above the trigger level of the pulse values in Figures 18 and 19, the volume criteria is met well before the duration is reached. For example, for the large seasonal pulse for the winter season at Cuero (see Figure 18), the volume requirement of 55,300 acre-feet is met in less than seven days at a continuous flow rate of 4,610 cfs (1 cfs per day is equal to 1.983 acre-feet). The authors erroneously state that in average or wet years with sufficient rainfall the standards would protect 26 days of connection for Oxbow 1 during the winter season. In reality, if flows stayed above the trigger level, only 7 days of connection would be protected by the standards. Second, the standards do not always protect three pulses per season (two small and one large). In many cases, the standard is met

with only two pulses (one small and one large) per season. As stated by the standard: *if a pulse flow requirement for a large seasonal pulse is satisfied for a particular season, one of the smaller pulse requirements is also considered to be satisfied* (30 TAC 298.375d6).

The impact of these two factors on the number of days of connectivity for Oxbow 1 can be illustrated using 20 years of data (1997-2016) from the Cuero gage (USGS gage no. 08175800). With an estimated connection flow of 1,700 cfs, there were 102 days per year of oxbow connection on average during the twenty year period from 1997 to 2016. Applying the current standards (see Figure 18) (with both factors mentioned above) to the hydrology time series results in protection of an average of 21 days per year of connectivity for the same time period. On page 52, the authors of the report suggest that including overbank events in the standards (as recommended by the basin's science team) would provide a significant improvement in the number of days of connection. With overbank flows like those in Figure 53 included, the number of days of connection protected by the standards increases to about 35 days per year on average for the 1997-2016 time period. However, this is still less than half the connections currently provided in the hydrologic record. Analysis of connectivity and standards for the Victoria site provides similar results.

Please provide a more accurate evaluation of the flows protected by the current standards or remove this section from the report.

Response: We deleted the text that lies beyond the scope of the contract. We only retained the following statements:

“Whereas the flow pulses in the current standards provide for lateral connectivity of Oxbow 2, the frequency and duration of connections likely would be significantly reduced relative to the duration that would be provided by the flow pulses recommended by the basin’s science team. Of course, this inference assumes that future infrastructure for water diversion or storage would be capable of significantly impacting flows. Hydrologic modeling is required to obtain estimates of discharge under various scenarios involving flow standards and implementation rules for water storage or diversion by water right holders.”

7. In the *References Cited* section, please provide the following references:
 - a. (IACUC 2015) mentioned on page 12.
 - b. (Post 2002) mentioned on pages 15 and 23.
 - c. (Vanderklift and Ponsard 2003) mentioned on pages 15 and 23.
 - d. (TCEQ 2012) mentioned on page 50.

Also, the following references are mentioned in the *References Cited* section but not in the report. Please refer to them at an appropriate place in the report or remove them from the *References Cited* section.

- e. Polis, et al. 1997
- f. Robertson, et al. 2008
- g. Winemiller, 1996
- h. Winemiller, 2004
- i. Winemiller, 2007
- j. Zeug, et al. 2009

Response: all addressed