FINAL PERFORMANCE REPORT

As Required by

THE ENDANGERED SPECIES PROGRAM

TEXAS

Grant No. TX E-156-R

(F13AP00671)

Endangered and Threatened Species Conservation

Assessing the Conservation Status of Rare Endemic Mussels Species (Family: Unionidae) in the Lower Guadalupe River, Texas

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29 August 2016

FINAL REPORT

STATE: ____Texas_______________ GRANT NUMBER: ___TX E-156-R-1__

GRANT TITLE: Assessing the Conservation Status of Rare Endemic Mussels Species (Family: Unionidae) in the Lower Guadalupe River, Texas

REPORTING PERIOD: ____1 September 2013 to 31 August 2016_

OBJECTIVE(S). To systematically survey the lower Guadalupe River to determine the location, density, species composition and status of threatened mussel populations.

Segment objectives:

Task 1: Site selection for the Lower Guadalupe River (Sept 2013 – Oct 2013, Year 1)

Task 2: Systematic survey of the lower Guadalupe River from Gonzales Lake to Cuero, Texas (Oct 2013 – Sept 2014, Year 1).

Task 3: Systematic survey of the Lower Guadalupe River from Cuero, Texas to Victoria, Texas (Oct 2014 – May 2015, Year 2).

Task 4: Collect habitat information for significant populations of threatened mussel species in the Lower Guadalupe River (Oct 2013 – May 2015, Years 1 & 2)

Task 5: Create distribution maps, evaluate the conservation status of state threatened mussel species in the Lower Guadalupe River, and submit final report (June 2015– September 2015, Year 2)

Significant Deviations:

None.

Summary Of Progress:

Please see Attachment A.

Location: Along Guadalupe River in Gonzales, De Witt, and Victoria counties, Texas.

Cost: ___Costs were not available at time of this report, they will be available upon completion of the Final Report and conclusion of the project.__

Prepared by: _Craig Farquhar_____________ Date: _29 August 2016

Approved by: ______________________________ Date:___29 August 2016_
C. Craig Farquhar

ATTACHMENT A

Distribution and habitat associations of freshwater mussels (Bivalvia: Unionidae) in the lower Guadalupe River, Texas

Final Report for Section 6 TX E-156-R (TPWD # 442139)

Reporting period: 27 September – 31 August, 2016

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Executive Summary

The Guadalupe River from Gonzales to Victoria, Texas was surveyed in 2014 – 2016 to examine the distribution and habitat associations of common and endemic species of freshwater mussel. We surveyed mussels using random sample design stratified by mesohabtiat: banks, backwaters, behind-of-point-bars, front-of-point-bars, pools and riffles. Both qualitative timed-search and quantitative quadrat methods were conducted at each site, and physical habitat parameters were measured from quadrats to examine microhabitat associations. In total, 21,119 individuals from 13 species were observed across 70 sites in the 6 study reaches surveyed, indicating mussels were generally abundant and relatively diverse in the lower Guadalupe River. In addition, we observed all three threatened species known to occur in the river, including false spike (*Fusconaia mitchelli*), golden orb (*Quadrula aurea*) and Texas Pimpleback (*Quadrula petrina*). Indicator species analysis revealed significant associations for 8 of the 13 species observed. *Amblema plicata*, *C. tampicoensis*, *L. hydiana*, and *L. teres* were associated significantly with banks, *P. grandis* was associated significantly with backwaters and *Q. aurea*, *F. mitchelli* and *Q. petrina* were associated significantly with riffles. Canonical correspondence analysis corroborated these results where the dominant gradient was associated with high values of shear stress, flow and substrate particle size, which are typical of riffle habitats. The secondary gradient reflected increasing percentage of fine particular organic matter and algae, which corresponded to front-of-point-bars and may reflect deposition of organic matter during low flows. Our results provide important information on the distribution and abundance of freshwater mussels, particularly for threatened species occurring in this river and can be targeted for conservation research. Habitat associations observed in our study suggest some mussel species can be used as a tool for predicting future changes to the lower Guadalupe River.

Introduction

Freshwater mussels are in decline worldwide (Lydeard et al., 2004), and in North America, where mussels reach their highest diversity, more than two-thirds of the approximately 300 species require some degree of conservation protection (Williams et al., 1993; Strayer et al., 2004). In response to these declines, there has been a rapid increase of conservation efforts and prioritization of research targeting the ecology of freshwater mussels (NNMCC, 1998; Haag & Williams, 2014; FMCS, 2016). At the most fundamental level, knowledge of current distribution and abundance of mussels, particularly for threatened and endangered species, is needed to understand the status and viability of populations. However, distribution data for many species of mussel occurring in the United States are limited (Lydeard et al., 2004), leaving status assessments and conservation decisions based on incomplete data.

Despite the long-standing goal of advancing the ecology of freshwater mussels, knowledge of physical habitat associations for many species remains unknown. Mussels generally persist in areas where stream flow is neither too high nor too low. Low flows tend to be detrimental to mussels because of increased risk of desiccation, catabolism caused by high temperatures and exposure to poor water quality (Golladay et al., 2004; Haag & Warren, 2008). In contrast, high flows come with increased water velocity and hydraulic forces (e.g., shear stress) acting on the stream bed that lead to scouring and entrainment of mussels from unstable substrates (Strayer, 1999; Hardison & Layzer, 2001; Morales et al., 2006). These physical constraints, coupled with the dynamic nature of rivers systems, create a mosaic of optimal habitat patches (i.e., hydraulic habitat refugia) that influence the distribution and abundance of mussels at multiple scales across riverscapes (Newton et al., 2008).

Texas is faced with an impending conservation crisis regarding the plight of freshwater mussels (Howells et al., 1996, 1997), and unlike other regions in the United States, establishment of conservation priorities have begun only recently. In particular, 15 of 52 species of Texas mussel were listed as state-threatened in 2009 (Texas Register 35, 2010), and shortly thereafter, 12 of those species were petitioned for protection under the U.S. Endangered Species Act (ESA) (Federal Register 76, 2011). Included among those 12 species, 1 has been proposed as endangered and 5 have already advanced to candidacy and are pending review by U.S. Fish and Wildlife Service. As a result, there is an urgent need to study the ecology and conservation of threatened mussels in Texas, especially because many surveys implemented previously have lacked sufficient effort to detect rare species, and knowledge of specific habitat requirements for mussel species occurring in Texas is almost nonexistent (but see Karatayev & Burlakova, 2008; Randklev et al., 2014).

The Guadalupe River, located in south-central Texas, has been inadequately surveyed, despite several threatened species occurring in the river historically. In fact, distributional surveys conducted by the state have described the mussel fauna as being nearly extirpated (Howells, 1995, 1996a, 1997, 1998, 1999, 2000, 2001); however, sampling effort from those surveys were not focused on characterizing mussel faunas from specific rivers, but rather, they were concerned with understanding the distribution of mussels from a range-or state-wide level. Recently, investigators have discovered populations of *Fusconaia mitchelli* (False spike), *Quadrula aurea* (Golden orb) and *Quadrula petrina* (Texas pimpleback) in the Guadalupe River (Randklev et al., 2012), indicating that research regarding the distribution of endemic and common species in this river is needed. Our objectives were to (1) systematic survey freshwater mussels, (2) investigate micro and mesohabitat associations and (3) assess the conservation status of threatened mussel species in the lower Guadalupe River.

Methods

Study area

The Guadalupe River originates in Kerr County, Texas and runs approximately 402 km southeast until reaching the Gulf of Mexico (Huser, 2000). This spring fed river drains 15,539 km^2 of land, which include the Blanco-San Marcos and San Antonio Rivers within parts of the Edwards Plateau region (Huser, 2000). Ten major impoundments for purposes ranging from water supply to hydropower generation occur on the main stem of the Guadalupe River. Canyon Lake is the largest upstream impoundment followed by Lake McQueeney in Guadalupe County (Huser, 2000). Land use in the Guadalupe watershed is characterized by livestock grazing, concentrated animal feeding operations, pecan orchards and oil and gas development. Urban areas (San Marcos and San Antonio) are situated in the upper portion of the watershed along the Interstate 35 corridor. Smaller cities consist of Gonzales, Cuero and Victoria located along lower reaches of the river.

Site selection

Survey sites were selected in the lower Guadalupe River using a stratified random sampling design. We chose this section of the Guadalupe River because its lacks large on-channel reservoirs, which are prevalent throughout the middle and upper sections of the Guadalupe River. We first divided the river into 10 km reaches spanning from Gonzales to Victoria, Texas and selected 8 reaches (i.e., study reaches) randomly to survey mussels. Within each study reach, we identified the following mesohabitat types using aerial imagery (Google Earth™): banks (B), front-of-point-bars (FPB), behind-of-point-bars (BPB), pools (P), backwaters (BW), mid-channels (MC) and riffles (R). Bank habitats were defined by locating the point in the channel where the slope of the bank leveled out, indicating the beginning of mid-channel habitat. Front-of-point-bars and behind-of-point-bars were located on the up-and downstream portion of sand or gravel bars, respectively. Pool habitats were generally characterized by minimal current velocities and relatively deep water. Backwater habitats were areas with minimal velocities and variable water depths and were often located near obstructions or in secondary channels. Mid-channel habitats were located in the middle of the river channel. Riffle habitat was defined as shallow areas with moderate to fast flows, where small hydraulic jumps over rough bed material, cause small ripples, waves and eddies. Once all available mesohabitats suitable for mussels were mapped, we randomly selected two sites per habitat type within each reach. See Randklev et al. (2014) for visual depictions of habitat.

Survey design

We surveyed mussels at each site using both qualitative and quantitative sampling methods. The qualitative timed-search method is favorable for detecting rare species; whereas, the quantitative quadrate method is ideal for detecting smaller species and estimating demographic parameters more accurately (Vaughn et al., 1997). At each site, we selected 20 0.25-m^2 quadrats within a 150-m^2 area randomly and excavated sediment from quadrats to depth of 20 cm using a modified Surber sampler. Sediment excavated from quadrats was passed through a 0.25-inch sieve to separate mussels. After the completion of quadrate surveys, we performed timed-searches using visual and tactile techniques for a total of 4 person-hours (p-h) within the 150-m^2 area. All mussels were identified, measured for shell length (mm) and returned to the study area.

Habitat surveys

Physical characteristics of the river were estimated from a subset of sites to examine mussel-habitat associations. Within each quadrate, we recorded near-bed water velocity and water depth using an electromagnetic flow meter (OTT MF Pro) and near-bed shear stress using Fliesswasserstammtisch (FST) hemispheres. Other habitat variables were estimated from each quadrat visually: substrate size index, substrate sorting index, embeddedness index, algae (%) and fine particulate organic matter (FPOM) (%). Substrate type, sorting and embeddedness indices were qualified following Gordon et al. (2004); see Appendix A and Randklev et al. (2014) for further details. Briefly, each index represents a number where higher values correspond to either coarser substrate material (substrate type), uneven distribution of particle size (substrate sorting) and high proportion of coarse substrate material fixed deeply within surrounding substrate. All habitat measurements were collected prior to mussel sampling.

Data analyses

Several techniques were used to analyze characteristics of mussel assemblages and micro and mesohabiat associations in the lower Guadalupe River. We generated sample-based species accumulation curves to estimate average pooled species richness among study reaches and habitat types (Kindt & Coe, 2005). Species accumulation curves were used in two ways because sampling effort (i.e., number of sites) varied among reaches: to (1) examine for significant differences among groups based on the non-overlapping variances (standard deviation) plotted with curves, and (2) determine if sampling effort was adequate based on whether a curve reached its asymptote (Gotelli & Colwell, 2001). To estimate species richness for the entire study area (i.e., lower Guadalupe River), we used a first-order (nonparametric) Jackknife species richness estimator, which uses a resampling technique (Palmer, 1990; Kindt & Coe, 2005). We plotted mean CPUE and density of mussels by reach and habitat and tested for differences among groups using the nonparametric Kruskal-Wallace rank sum test. Each grouping variable (i.e., reach and habitat) was analyzed separately, and if significant differences were observed, a pairwise Wilcox rank sum test was implemented to identify differences between group levels. Finally, we plotted CPUE against density of mussels and used a smoothing spline scatterplot smoother to examine the relationship between abundance estimates.

We analyzed habitat associations of mussels in two ways. First, indicator species analysis was used to test for significant mesohabitat associations (Dufrene & Legendre, 1997; De Cáceres & Legendre, 2009). This analysis calculates an indicator value (IndVal) index based on differences among proportional occurrence and relative abundance of species within groups (e.g., mesohabitat types) and tests for significant differences among groups using a permutation test (Dufrene & Legendre, 1997). The IndVal statistic represents two parameters, including the probability a site belongs to a specific habitat based on the presence of a species (A), and the probability of finding a species at a site belonging to its associated habitat (B). To ease the interpretation of the results, we restricted the analyses such that each species was only allowed to be associated with 1 habitat type. Second, we complemented these results with canonical correspondence analysis (CCA) of habitat data collected from quadrates. CCA uses reciprocal averaging to constrain and correlate species ordination by the linear combination of environmental variables. Significances of environmental correlates were tested using a permutation test. All analyses were conducted using R version 3.02 (R Foundation for Statistical Computing, Vienna, Austria).

To assess the conservation status of threatened and endemic species occurring in the lower Guadalupe River, we summarized abundance and distribution data and habitat associations for *Q. aurea*, *Q. petrina* and *F. mitchelli*. Survey results obtained for these species were used to create digital biogeographical maps. These maps include all historical locality records of threatened species known from the Guadalupe River, and we compare those data with results presented in this study to provide the most update occurrence and distribution for these species.

Results and Discussion

Distribution and abundance of mussels

We surveyed mussels at 70 sites (6 study reaches) in the lower Guadalupe River (Figure 1) and observed a total of 21,119 individuals from 13 species (Table 1, Appendix B – C). Two study reaches initially selected were not surveyed due to extended periods of high flow that precluded us from entering the river. The two most abundant species recorded were *A. plicata* and *Q. aurea*, which proportionally comprised of 0.58 and 0.22 of total individuals collected, respectively (Table 1). All other species had less than 0.06 proportional abundance (Table 1). Proportional occurrence of species (i.e., proportion of sites a species was observed) across the study area was relatively high for *A. plicata*, *C. tampicoensis*, *L. teres*, *Q. aurea* and *Q. petrina*, which ranged from 0.74 – 0.99 (Table 1). Species with intermediate proportional occurrence (0.34 – 0.50) included *L. hydiana*, *M. nervosa* and *F. mitchelli*; whereas, the remaining species had proportional occurrences less than or equal to 0.13 (Table 1).

Species richness varied by reach with no observable pattern related to stream position (Figure 2A). Reaches II and V had significantly higher species richness, Reaches I, III and VI had relatively moderate levels of species richness and Reach IV had species richness significantly lower than all other reaches (Figure 2A). Sampling effort was generally sufficient; however, species accumulation curves for Reaches I and II failed to asymptote (Figure 2A), indicating richness is expected to be higher if more sites were sampled. Species richness estimated for the entire study area (JACK1 = 14.0 ± 1.0) was higher than observed richness (n = 13), suggesting 1 or 2 additional species might still be present in the lower Guadalupe River, despite the intensity of our survey effort. Species not detected here but are known to occur in the drainage (historically or presently) include *Lampsilis bracteata* (Texas fatmucket), *Leptodea fragilis* (fragile papershell), *Ligumia subrostrata* (pondmussel), *Potamilus purpuratus* (bleufer), *Quadrula apiculata* (southern mapleleaf) and *Utterbackia imbecillis* (paper pondshell) (Strecker, 1931; Howells, 1994, 1995, 1996b; Howells et al., 1996). Though, some of these species (i.e., *Lampsilis bracteata*) are not known to have ever historically occurred in the lower Guadalupe River. Others like *L. subrostrata* and *U. imbecillis* occur primarily in slack water habitats like oxbows and sloughs, which were not surveyed during this study.

CPUE ranged from 42.2 mussels/p-h in Reach I to 110.8 mussels/p-h in Reach III and averaged 63.3 ± 10.5 (SE) mussels/p-h across reaches. Kruskal-Wallis rank sum test failed to detect significant differences in CPUE among study reaches (χ^2 = 6.01, df = 5, p = 0.305). Densities ranged from 6.2 mussels/m^2 in Reach I to 18.6 mussels/m^2 in Reach II and averaged 11.7± 2.2 mussels/m^2 across reaches. Similar to CPUE, Kruskal-Wallis rank sum test failed to detect significant differences in mussel densities among study reaches (χ^2 = 6.10, df = 5, p = 0.297). Interestingly, when compared to estimates from the Brazos River, CPUE was nearly identical (Brazos River: 63.7 ± 14.7 mussels/p-h) but mussel density was 5.1 times higher in the Guadalupe River (Brazos River: 2.3 ± 0.5 mussels/m^2) (Randklev et al., 2014). Furthermore, there was a strong curvilinear relationship between CPUE and density of mussels (r^2 = 0.78) (Figure 3), suggesting that qualitative timed-searches offer abundance estimates comparable to quantitative quadrate sampling. Although this result highlights the adequacy of time-searches, it should not be considered a replacement for quantitative quadrate sampling that offers more robust demographic data (Strayer & Smith, 2003).

Mussel-habitat associations

Mussel species observed in this study varied by mesohabitat types (Table 1), but some species were present and occurred consistently in high abundance in multiple habitat types (e.g., *A. plicata* and *Q. aurea*) (Table 1). Species richness varied significantly by mesohabtiat (Figure 4A). More specifically, bank, behind-of-point-bar, pool and backwaters had significantly higher richness than front-of-point-bars, riffles and mid-channels (Figure 4A). Only some of the species accumulation curves generated by habitat appeared to reach asymptotic growth, including bank, pool and riffle, while species accumulation curves for all other mesohabitats suggest additional species might be present, despite sampling 12 sites for some mesohabitat types.

CPUE ranged from 33.3 mussels/p-h in behind-of-point-bars to 119.1 mussels/p-h in banks (Figure 4B). Kruskal-Wallis test revealed significant differences in CPUE (χ^2 = 17.54, df = 6, p = 0.007), and pairwise comparisons using Wilcoxon rank sum test indicated the only significant difference was between behind-of-point-bars and banks (p = 0.013). Density of mussels ranged from 2.1 mussels/m^2 in behind-of-point-bars to 23.5 mussels/m^2 in riffles (Figure 4C). Similarly, Kruskal-Wallis rank sum test revealed significant differences in density among habitat types (χ^2 = 15.20, df = 6, p = 0.019), which also was driven by differences in density between behind-of-point-bars and banks (p = 0.047), based on Wilcoxon rank sum test. However, a marginally significant difference was observed between riffles and behind-of-point-bars (p = 0.077).

Despite the high occurrence in multiple habitats for some mussel species, indicator species analysis suggested 8 of the 13 species observed in the lower Guadalupe River were associated significantly with one specific mesohabtiat type. *Amblema plicata*, *C. tampicoensis*, *L. hydiana*, and *L. teres* were associated significantly with banks, *P. grandis* was associated significantly with backwaters and *Q. aurea*, *F. mitchelli* and *Q. petrina* were associated significantly with riffles (Table 2). In contrast, *A. confragosa* and *M. nervosa* were associated with banks, *Q. verrucosa* and *T. parva* were associated with front-of-point-bars and *T. texasense* was associated with behind-of-point-bars; however, the strength of these mussel-mesohabtiat associations were not significant (Table 2).

Total inertia (variance) explained of the canonical correspondence analysis of mussel and microhabitat data collected from quadrats was 2.06, 78% of which was explained by unconstrained variance. Eigenvalues, representing the proportion of variance in the community matrix, was 0.47 and 0.32 for the first and second axes, respectively, and permutation test indicated our model was significant (p < 0.001). The first axis explained an environmental gradient in which high shear stress, high flow and coarser substrate were correlated with *F. mitchelli* and, in part, *Q. aurea*, which corresponded with riffles (Table 3, Figure 5). These results were consistent with indicator species analysis, suggesting that these species are most associated with riffles (Table 2). At the other extreme of this gradient, embeddedness increased with decreasing shear stress, flow and substrate size, which correlated with *A. plicata*, *L. teres* and *C. tampicoensis* and corresponded to banks, backwaters and behind-of-point-bars (Table 3, Figure 5). The second axis explained an environmental gradient with high percent of fine particular organic matter (FPOM), high percent of algae and low substrate sorting that was correlated with *M. nervosa* and *T. texasense*, which corresponded to front-of-point-bars (Table 3, Figure 5). Interestingly, these results were somewhat inconsistent with the indicator species analysis, though mesohabtiat associations for those species were not significant (Table 2). No species were significantly correlated with substrate sorting (i.e., unevenness of substrate particles), which corresponded to mid-channel habitats. This result makes sense since mid-channels tend to experience high flows and scouring, which is not suitable for mussels.

Status of threatened species

We observed three species of freshwater mussel that are endemic to central Texas, listed as state-threatened and pending review for federal protection by U.S. Fish and Wildlife Service. The wide distribution of threatened species in the lower Guadalupe River is promising for their conservation. *Fusconaia mitchelli* is known to occur throughout most parts of the lower river; however, we did not detect this species in Reach VI, the most downstream reach in our study area (Figure 7). Given that our sampling effort in Reach VI was adequate (Figure 2A), this suggest that *F. mitchelli*, at the least, occurs in exceptionally low abundance. Surveys conducted by TPWD did find this species within this reach as recent as 2013. Both *Q. aurea* and *Q. petrina* occurred in each of the reaches sampling, confirming their persistence in the lower Guadalupe River (Figure 7).

CPUE and density of threatened species generally followed a similar trend as overall abundance estimates of mussels throughout the study reaches (Figure 6). Mean CPUE was relatively low for *F. mitchelli* (1.9 ± 1.0 mussels/p-h) and *Q. petrina* (2.7 ± 1.0 mussels/p-h); whereas, mean CPUE was high for *Q. aurea* (13.1 ± 3.7 mussels/p-h) (Figure 6A – C) and represented the second most abundance species of all mussels in the lower Guadalupe River (Table 1). Similarly, mean density was relatively low for *F. mitchelli* (0.9 ± 0.5 mussels/m^2) and *Q. petrina* (0.8 ± 0.3 mussels/m^2) and relatively high for *Q. aurea* (2.7 ± 1.1 mussels/m^2) (Figure 6D – F). Given their high abundance, these results suggest these populations can be used in future conservation research.

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Table 1. Species of mussel and their respective abundance, proportional abundance (prop), occurrence (number of times a species occurred at a site) and proportional occurrence. In addition, proportional abundance of species was presented by reach and habitat type. Habitat types are as follows: BH = bank, BPS = behind-of-point-bar, BW = backwater, FPS = front-of-point-bar, MC = midchannel, P = pool and R = riffle.

Species		**Abundance**		**Occurrence**		**Proportional abundance by reach**						**Proportional abundance by habtat**						
		n	**prop**	**n**	**prop**	**I**	**II**	**III**	**IV**	**V**	**VI**	**BH**	**BPS**	**BW**	**FPS**	**MC**	**P**	**R**
Amblema plicata	threeridge	12,233	0.58	69	0.99	0.61	0.39	0.55	0.61	0.64	0.67	0.72	0.43	0.68	0.56	0.69	0.74	0.14
Arcidens confragosa	rock pocketbook	19	0.00	9	0.13	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Cyrtonaias tampicoensis	Tampico pearlymussel	1,345	0.06	52	0.74	0.01	0.01	0.04	0.06	0.16	0.13	0.08	0.11	0.14	0.07	0.04	0.07	0.01
Lampsilis hydiana	Louisiana fatmucket	49	0.00	24	0.34	0.01	0.00	0.00	0.00	0.00	0.00	0.00	0.02	0.00	0.00	0.00	0.00	0.00
Lampsilis teres	yellow sandshell	588	0.03	58	0.82	0.03	0.02	0.02	0.03	0.03	0.05	0.03	0.11	0.05	0.06	0.01	0.03	0.00
Megalonaias nervosa	washboard	573	0.03	35	0.50	0.08	0.03	0.01	0.04	0.01	0.02	0.03	0.01	0.00	0.12	0.02	0.02	0.01
Pyganodon grandis	giant floater	7	0.00	2	0.03	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Quadrula aurea	golden orb	4,624	0.22	65	0.93	0.19	0.33	0.28	0.23	0.10	0.11	0.11	0.20	0.11	0.12	0.20	0.13	0.57
Fusconaia mitchelli	false spike	652	0.03	25	0.36	0.02	0.11	0.05	0.01	0.00	0.00	0.00	0.00	0.00	0.01	0.01	0.00	0.14
Quadrula petrina	Texas pimpleback	893	0.04	50	0.71	0.06	0.10	0.05	0.02	0.01	0.01	0.01	0.06	0.01	0.03	0.03	0.02	0.13
Quadrula verrucosa	pistolgrip	2	0.00	2	0.03	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Toxolasma parva	lilliput	107	0.00	12	0.17	0.00	0.01	0.00	0.00	0.03	0.00	0.00	0.04	0.00	0.04	0.00	0.00	0.00
Toxolasma texasense	Texas lilliput	27	0.00	8	0.11	0.00	0.00	0.00	0.00	0.01	0.00	0.00	0.03	0.00	0.00	0.00	0.00	0.00

Table 2. Species-mesohabtiat associations based on indicator species analysis. IndVal represents the test statistic, *A* is the probability a site belongs to a specific habitat based on the presence of a species, *B* is the probability of finding a species at a site belonging to its associated habitat and p is the level of significant (α = 0.05).

Species	**Habitat type**							**Statistics**			
	BH	**BPS**	**BW**	**FPS**	**MC**	**P**	**R**	***A***	***B***	***IndVal***	***p***
Amblema plicata	×							0.34	1.00	0.58	0.081
Arcidens confragosa	×							0.40	0.27	0.33	0.335
Cyrtonaias tampicoensis	×							0.35	1.0	0.59	**0.036**
Lampsilis hydiana	×							0.57	0.64	0.60	**0.002**
Lampsilis teres	×							0.34	1.00	0.58	**0.023**
Megalonaias nervosa	×							0.31	0.55	0.41	0.695
Pyganodon grandis			×					1.00	0.20	0.45	**0.075**
Quadrula aurea							×	0.53	0.90	0.69	**0.019**
Fusconaia mitchelli							×	0.91	0.90	0.91	**0.001**
Quadrula petrina							×	0.61	0.90	0.74	**0.003**
Quadrula verrucosa				×				1.00	0.09	0.30	1.000
Toxolasma parva				×				0.68	0.18	0.35	0.378
Toxolasma texasense		×						0.64	0.27	0.42	0.107

Table 3. Eigenvalues (loadings) of species and environmental factors associated with the first two axes in the cononical correspondence analysis of mussel abundance and habtiat variables collected from quadrats in the Guadalupe River, Texas.

Species or habitat variable	**CCA1**	**CCA2**
Flow	-0.7698	-0.2709
FST	-0.8558	-0.1396
Algae	-0.4371	0.4480
FPOM	-0.2487	0.8218
substrate size	-0.3242	-0.0510
Substrate sorting	0.0931	-0.4003
Substrate embededness	0.6001	-0.1460
Mesohabtiat		
behind-of-point-bar	0.1703	-0.0308
backwater	0.4578	-0.0477
front-of-point-bar	-0.2111	0.8026
mid-channel	-0.1151	-0.2420
riffle	-0.7608	-0.3148
Amblema plicata	0.2500	0.0122
Cyrtonaias tampicoensis	0.9148	-0.2644
Lampsilis teres	0.6902	-0.1768
Megalonaias nervosa	-0.5496	1.3925
Quadrula aurea	-0.4087	-0.2364
Fusconaia mitchelli	-1.5828	-0.7919
Quadrula petrina	-0.4037	0.2845
Toxolasma texasense	0.0031	0.7983

Figure 1. Map of the sample sites on the lower Guadalupe River from Gonzales to Victoria, Texas.

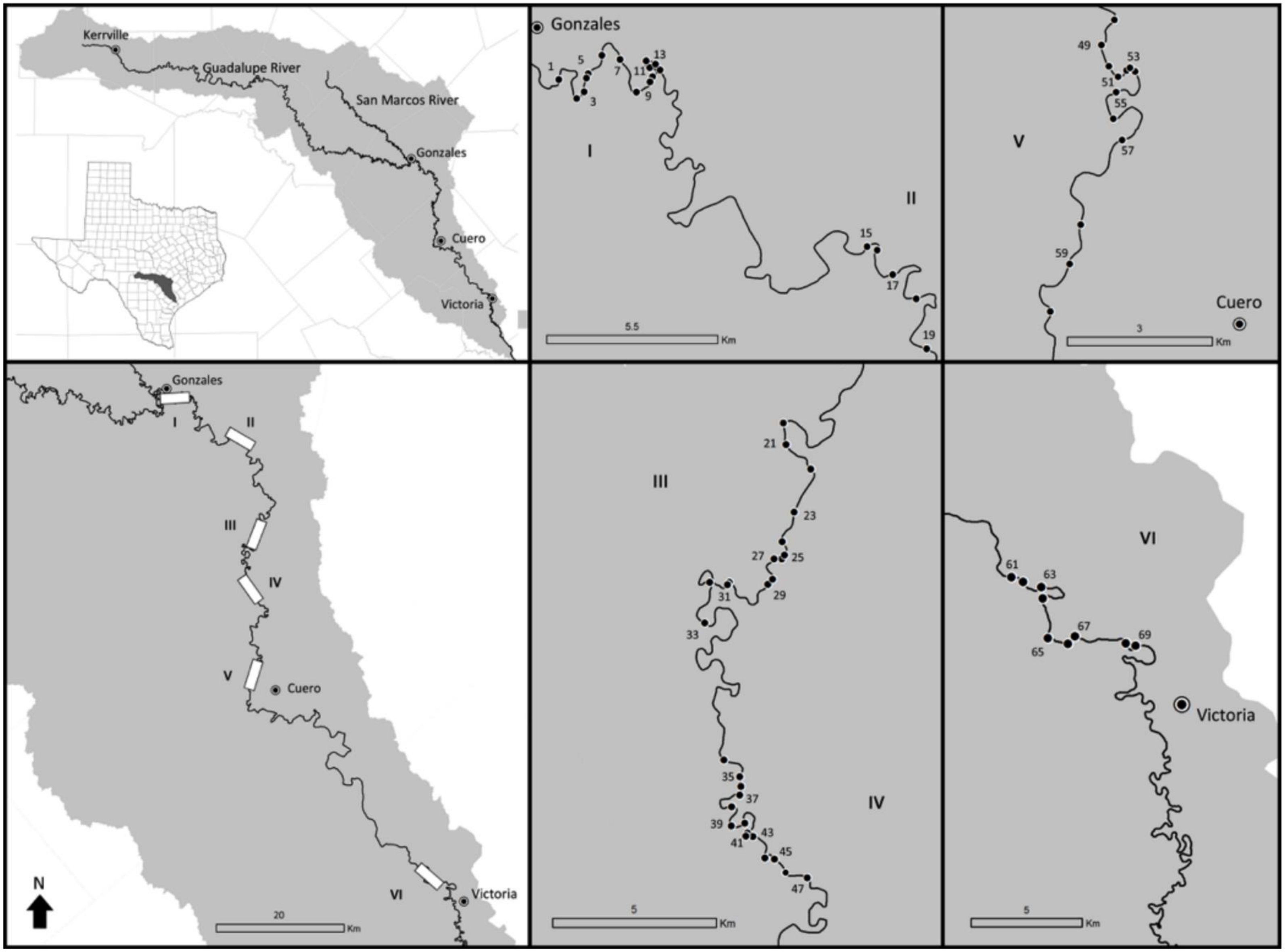


Figure 2. (A) Species accumulation curves, (B) catch-per-unit effort (CPUE) and (C) density of mussels by reach in the lower Guadalupe River, Texas.

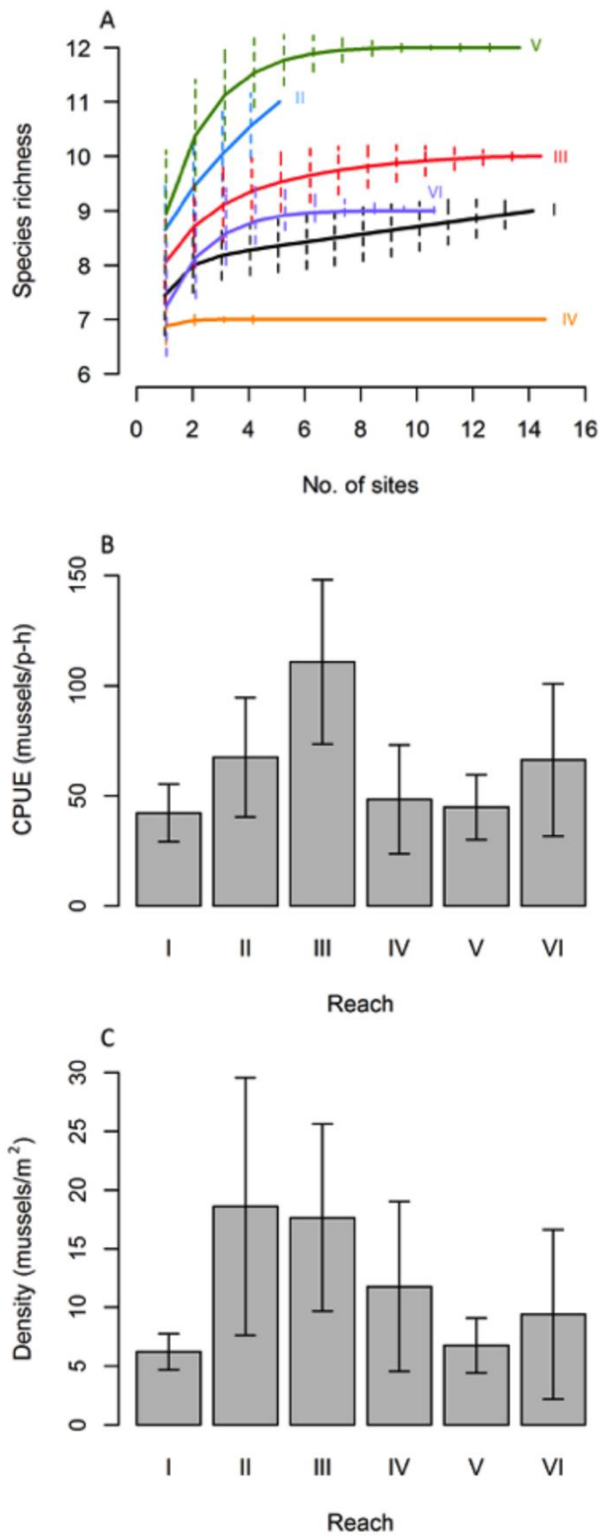


Figure 3. Relationship between catch-per-unit-effort and density of mussels in the lower Guadalupe River. Shaded area indicates 95% confidence intervals.

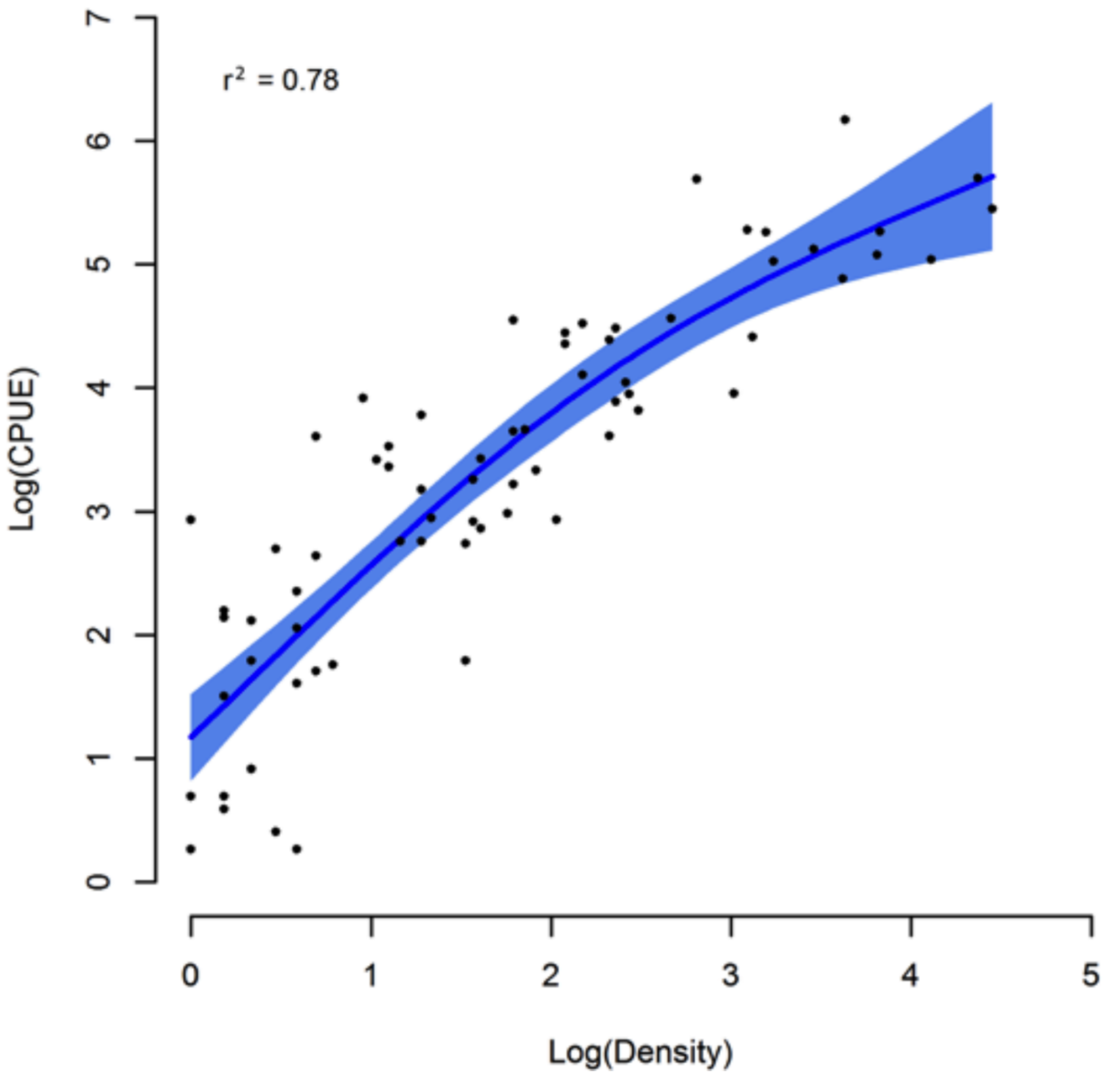


Figure 4. (A) Species accumulation curves, (B) catch-per-unit effort (CPUE) and (C) density of mussels by habitat type in the lower Guadalupe River, Texas.

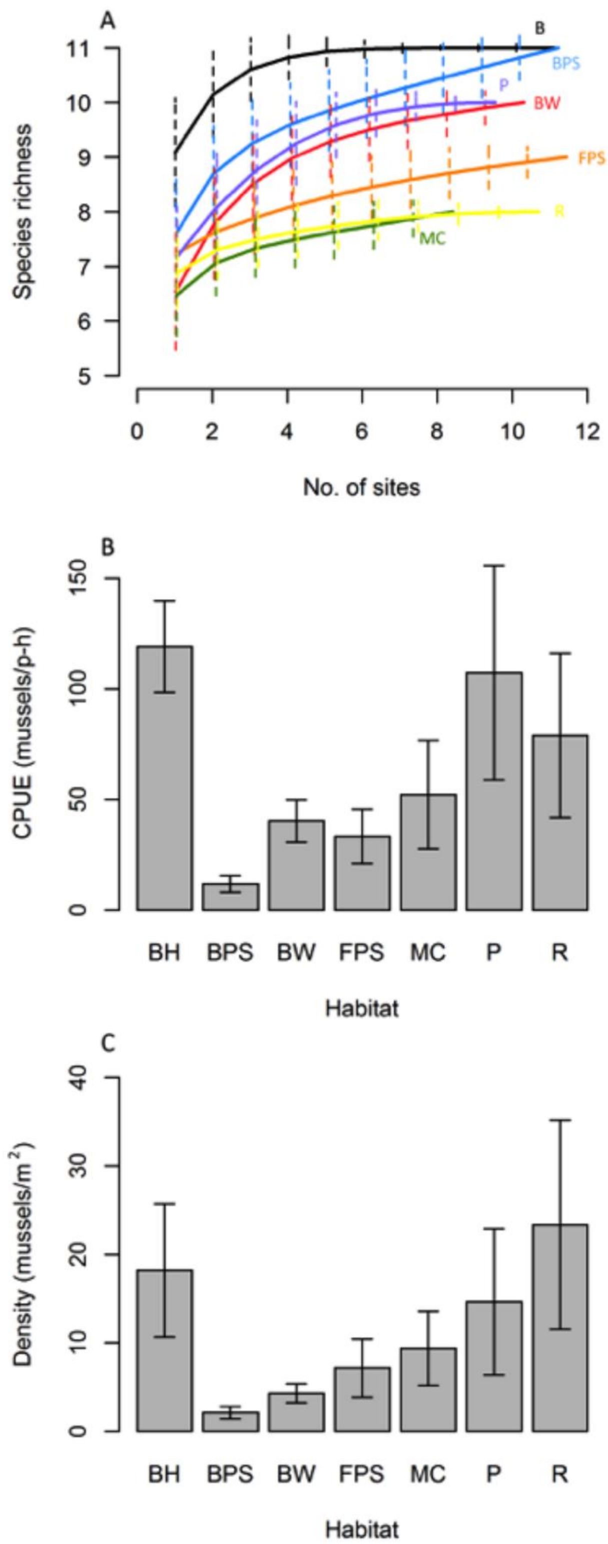


Figure 5. Canonical correspondence analysis of mussel and microhabitat data collected from quadrats in the lower Guadalupe River, Texas. Black points represent scores derived from mussel data in quadrats and red initials represent species scores: AP = *A. plicata*, CT = *C. tampicoensis*, FM = *F. mitchelli*, GO = *Q. aurea*, LT = *L. teres*, QP = *Q. petrina*, MN = *M. nervosa* and TX = *T. texasense*. Arrows point to the direction of increasing values of habitat, including algae, embeddedness (embed), fine particulate organic matter (FPOM), flow (water velocity), shear stress (FST), substrate sorting index (sorting), substrate size index (substrate) and substrate embeddedness index (embed). Blue initials indicate mesohabitat scores: B = bank, BPS = behind-of-point-bar, BW = backwater, FPS = front-of-point-bar, MC = mid-channel and R = riffle.

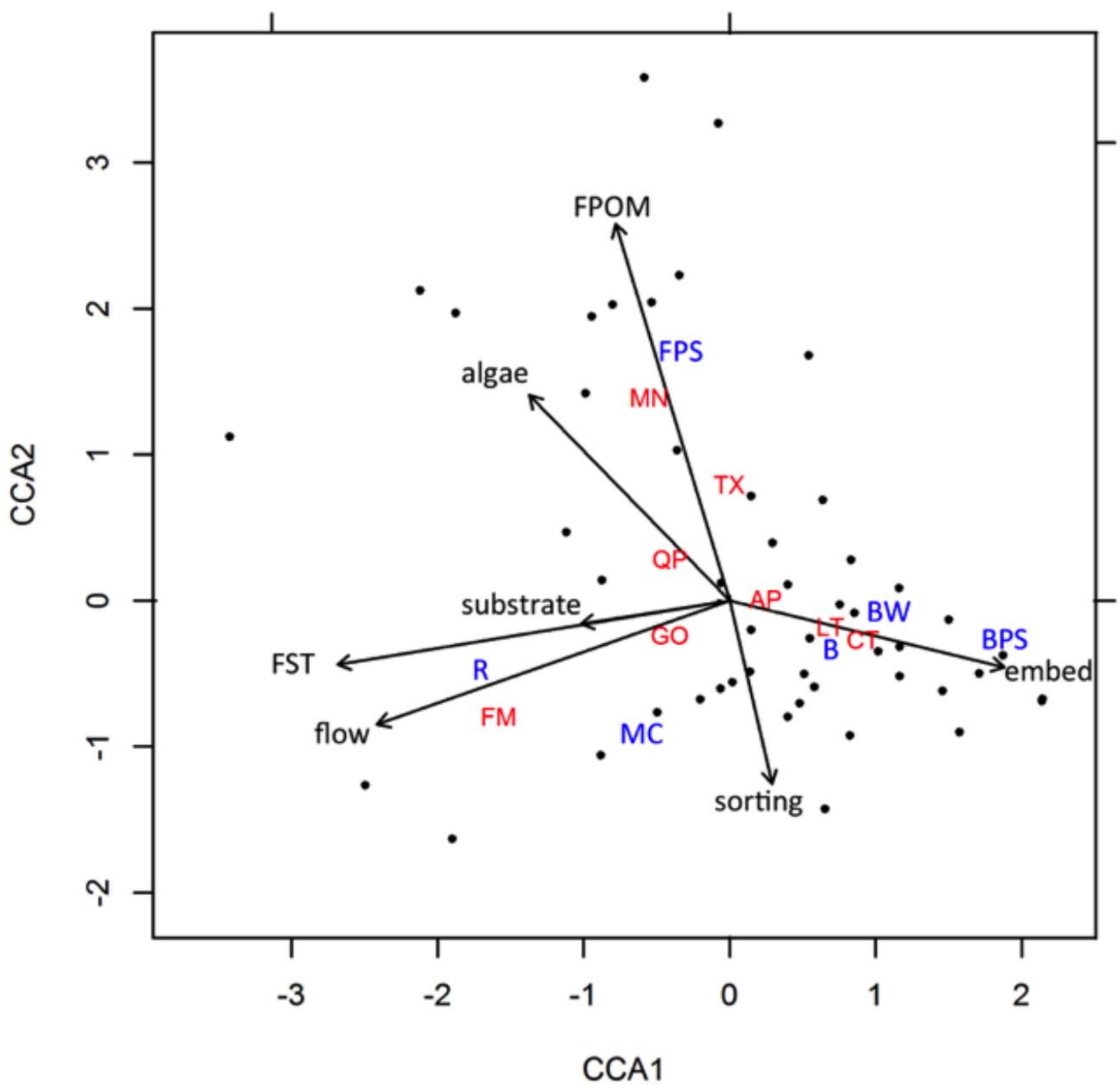


Figure 6. CPUE and density of state-threatened species of mussel (*Fusconaia mitchelli*, *Quadrula aurea* and *Quadrula petrina*) among reaches in the lower Guadalupe River.

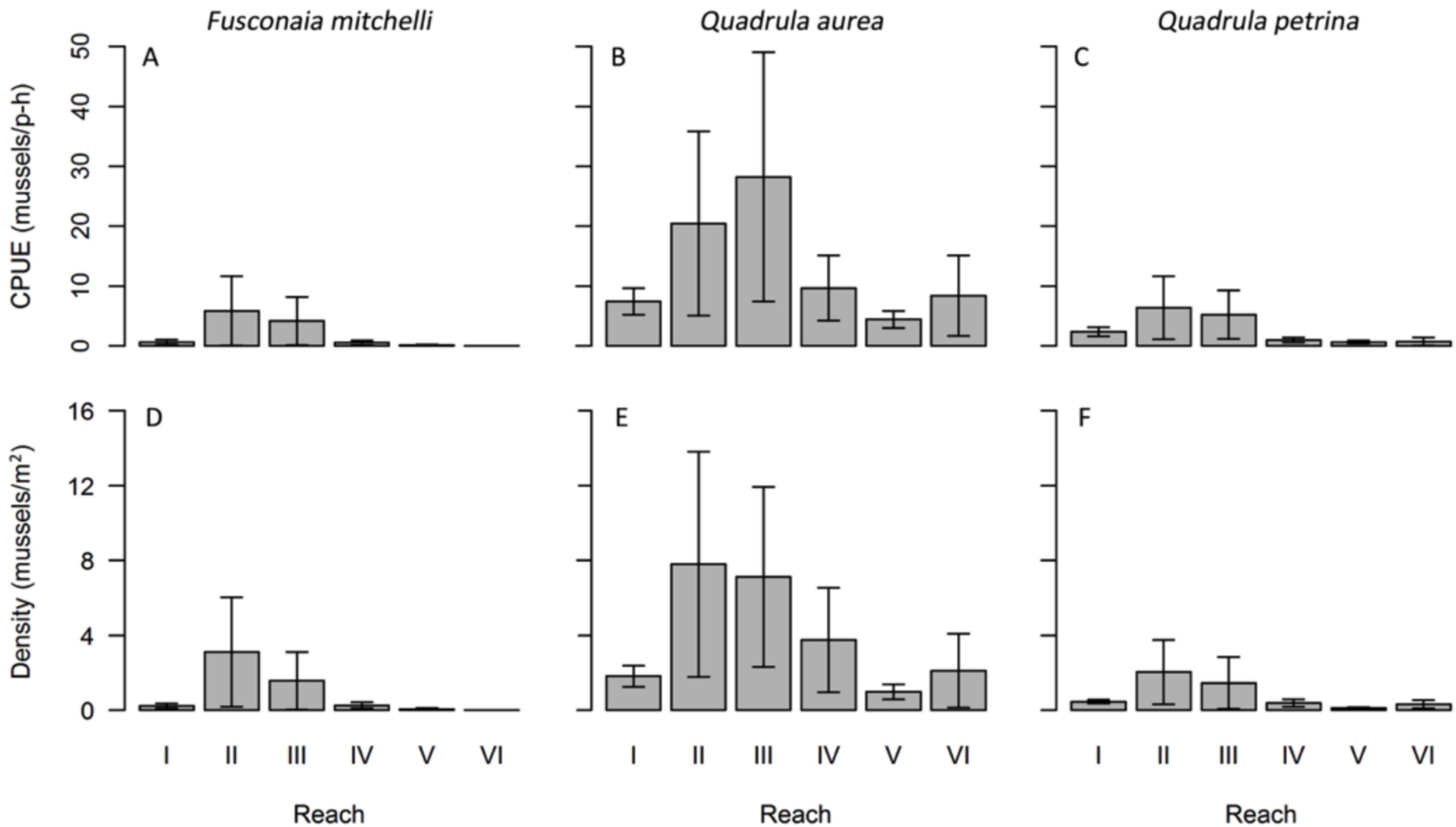
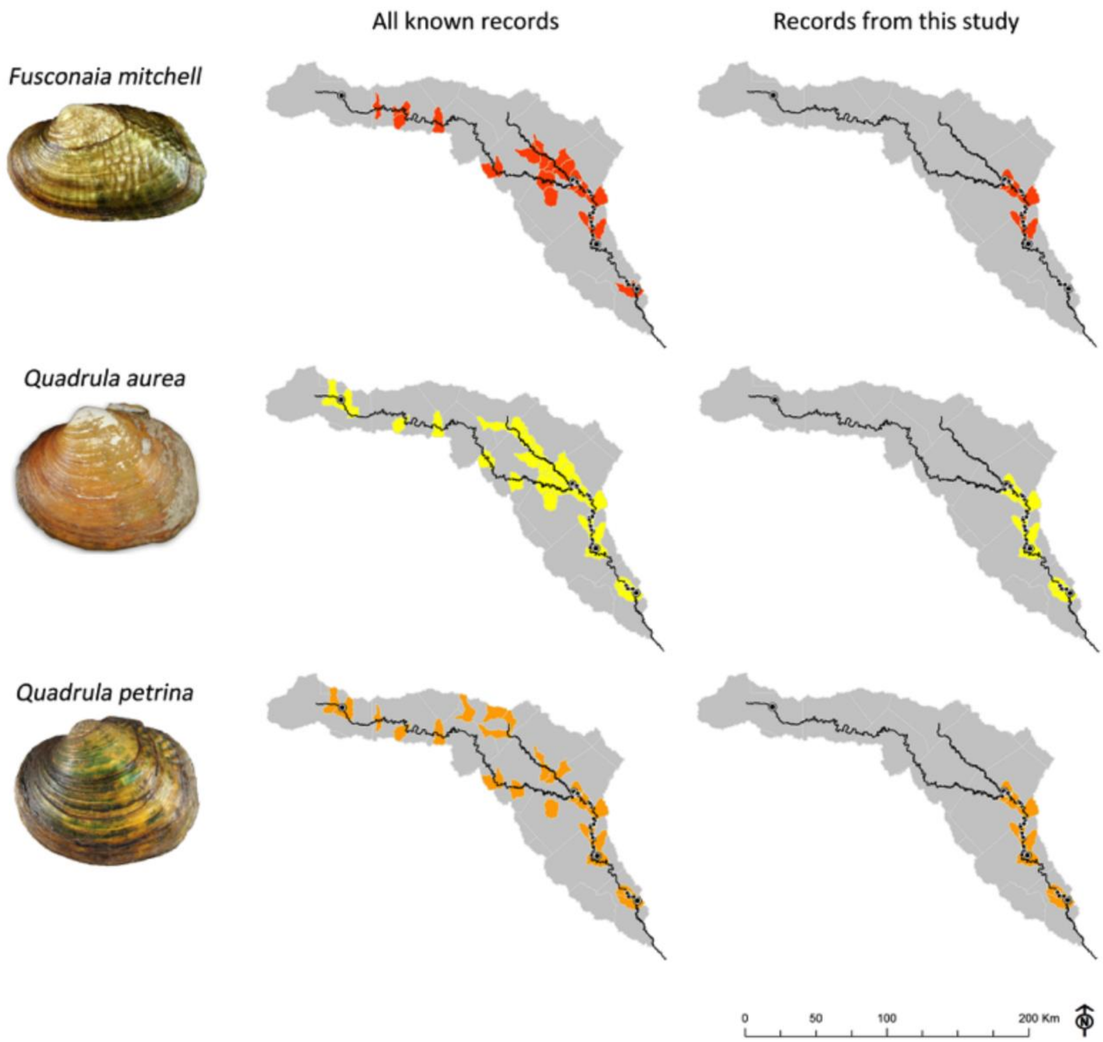


Figure 7. Maps of all known locality records and locality records obtained from the present study for threatened species of mussel in the lower Guadalupe River Texas. Highlighted areas indicate hydrological units in which mussels have been observed.



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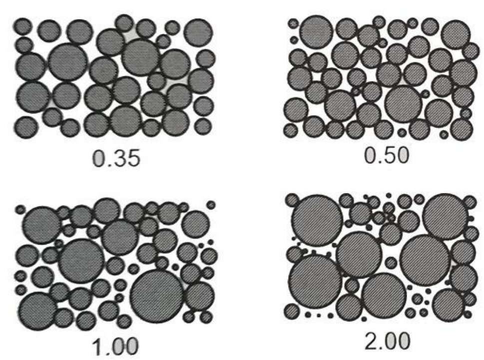
Appendix A. Indicies used to qualify habtiat variables visually within quadrats sampled for mussels in the lower Guadalupe River, Texas. Substrate size consisted of a three number index: (1) dominate substrate size, (2) sub-dominant substrate size and (3) percent of fine particles (represented as 0 – 9). Methods and images adapted from Gordon et al. (2004).

Substrate size

Index	Description
1	Fines (sand and smaller)
2	Small gravel (4 - 25 mm)
3	Medium gravel (25 - 50 mm)
4	Large gravel (50 - 75 mm)
5	Small cobble (75 - 150 mm)
6	Medium cobble (150 - 225 mm)
7	Large cobble (225 - 300 mm)
8	Small blouder (300 - 600 mm)
9	Large blouder (> 600 mm)

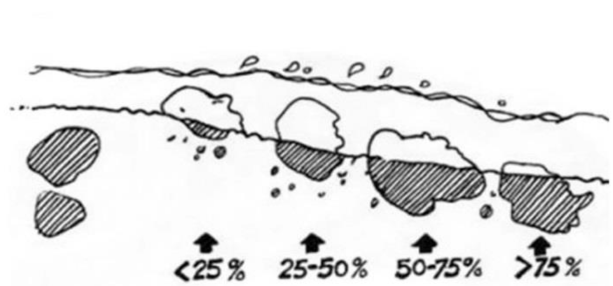
Substrate sorting

Index	Despcription
1	< 0.35 Very well sorted
2	0.35 - 0.50 Well sorted
3	0.50 - 1.00 Moderately sorted
4	1.00 - 2.00 Poorly sorted
5	> 2.00 Very poorly sorted



Substrate embededness

Index	Description
1	< 25% embeded
2	25 - 50% embeded
3	50 - 75% embeded
4	>75% embeded



Appendix B. Raw data collected from qualitative timed-search surveys in the lower Guadalupe River, Texas.

Site	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25
Reach	I	I	I	I	I	I	I	I	I	I	I	I	I	I	II	II	II	II	II	III	III	III	III	III	III
Habitat	BW	FPS	FPS	BH	R	MC	BH	BPS	P	BPS	R	P	BW	MC	BPB	FPB	MC	RFL	BH	P	R	MC	BH	R	BW
Amblema plicata	11	173	37	183	41	626	97	50	38	71	14	73	2	65	13	228	9	40	312	1699	62	239	574	74	265
Arcidens confragosus	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0	0
Cyrtonaias tampicoensis	0	1	0	3	1	0	5	2	1	4	0	2	0	1	8	2	1	3	3	43	4	21	89	0	14
Fusconaia mitchelli	1	1	1	0	10	5	0	0	0	1	14	0	0	2	0	1	0	116	0	3	160	1	2	67	0
Lampsilis hydiana	0	0	0	2	0	0	4	5	2	1	0	1	0	0	0	0	0	0	1	0	0	0	6	1	0
Lampsilis teres	5	5	2	4	1	4	10	11	7	4	0	12	0	0	9	5	1	0	7	0	2	0	56	2	29
Megalonaias nervosa	1	151	8	0	11	17	4	2	0	3	0	0	0	2	0	16	3	21	0	23	6	11	4	2	2
Pyganodon grandis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Quadrula aurea	0	41	3	20	90	104	23	1	6	46	25	30	0	27	12	62	2	324	9	125	815	57	27	398	8
Quadrula petrina	0	7	1	12	24	26	7	1	5	21	17	1	0	11	5	11	0	109	3	20	142	20	3	94	0
Quadrula verrucosa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
Toxolasma parvum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	9	0	0	0	2	0	0	0	2	0	0
Toxolasma texasense	0	0	0	0	0	0	0	0	0	1	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0
Total	18	379	52	224	178	782	150	72	59	152	70	119	2	108	58	326	16	613	337	1913	1191	349	766	638	318
Species richness	4	7	6	6	7	6	7	7	6	9	4	6	1	6	7	8	5	6	7	6	7	6	10	7	5
CPUE	4.5	94.8	13.0	56.0	44.5	195.5	37.5	18.0	14.8	38.0	17.5	29.8	1.0	27.0	14.5	81.5	4.0	153.3	84.3	478.3	297.8	87.3	191.5	159.5	79.5

Appendix B. (*continued*)

Site	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50
Reach	III	III	III	III	III	III	III	III	IV	IV	IV	IV	IV	IV	IV	IV	IV	IV	IV	IV	IV	IV	V	V	V
Habitat	FPS	BPS	MC	BW	P	BPS	FPS	BH	FPS	MC	BH	BPS	BH	R	FPS	BW	P	R	P	BW	BPS	MC	P	BH	MC
Amblema plicata	14	7	361	77	80	25	15	109	20	0	574	1	359	29	168	163	12	20	294	67	15	0	56	311	110
Arcidens confragosus	0	0	1	2	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Cyrtonaias tampicoensis	0	0	55	20	3	8	1	5	4	1	13	0	60	0	22	16	4	0	7	39	0	0	29	26	12
Fusconaia mitchelli	0	0	2	0	0	0	0	0	0	0	14	0	1	10	0	0	0	6	0	0	0	0	1	0	0
Lampsilis hydiana	0	1	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Lampsilis teres	15	2	2	1	3	18	11	8	5	0	17	3	14	0	5	3	2	4	1	9	5	0	2	6	0
Megalonaias nervosa	0	0	3	0	0	0	0	9	0	0	55	0	68	2	0	0	0	0	0	0	0	0	0	0	2
Pyganodon grandis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Quadrula aurea	3	9	92	10	27	6	2	1	24	2	236	0	92	50	8	21	2	39	2	48	17	1	50	17	16
Quadrula petrina	0	1	8	0	4	1	1	0	2	0	17	0	9	9	2	1	0	2	4	8	1	0	5	1	4
Quadrula verrucosa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Toxolasma parvum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	3	0
Toxolasma texasense	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total	32	20	524	111	118	59	30	132	55	3	926	4	603	100	205	204	20	71	308	171	38	1	143	364	144
Species richness	3	5	8	6	6	6	5	5	5	2	7	2	7	5	5	5	4	5	5	5	4	1	6	6	5
CPUE	8.0	5.0	131.0	27.8	29.5	14.8	7.5	33.0	13.8	1.0	231.5	2.0	150.8	25.0	51.3	51.0	5.0	17.8	77.0	42.8	12.7	0.3	35.8	91.0	36.0

Appendix B. (*continued*)

Site	51	52	53	54	55	56	57	58	59	60	61	62	63	64	65	66	67	68	69	70
Reach	V	V	V	V	V	V	V	V	V	V	VI	VI	VI	VI	VI	VI	VI	VI	VI	VI
Habitat	FPB	BPB	P	RFL	FPB	BPB	BW	BH	RFL	BW	BH	R	BW	FPB	P	BH	FPB	BPB	BW	BPB
Amblema plicata	17	8	62	148	37	24	123	535	18	84	955	6	20	1	355	23	4	9	294	0
Arcidens confragosus	0	1	0	0	0	0	0	2	0	2	2	0	0	0	2	0	0	0	0	0
Cyrtonaias tampicoensis	29	2	2	6	29	29	48	65	15	80	168	0	0	0	132	1	0	0	16	0
Fusconaia mitchelli	0	0	0	5	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0
Lampsilis hydiana	0	0	0	0	0	0	1	2	1	1	7	0	0	0	0	1	0	0	0	0
Lampsilis teres	11	5	0	2	11	2	0	17	2	6	30	0	0	0	59	1	0	1	13	0
Megalonaias nervosa	0	0	1	2	0	0	0	20	1	0	0	0	1	0	37	0	0	0	2	0
Pyganodon grandis	0	0	0	0	0	0	6	0	0	1	0	0	0	0	0	0	0	0	0	0
Quadrula aurea	0	3	5	66	0	1	15	23	21	16	12	0	5	0	166	3	0	4	46	1
Quadrula petrina	0	0	1	10	0	0	4	2	6	1	1	0	1	0	16	0	0	0	2	0
Quadrula verrucosa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Toxolasma parvum	39	0	0	0	15	9	0	1	0	0	0	0	0	0	0	0	0	0	0	0
Toxolasma texasense	0	0	0	0	0	10	0	1	0	0	4	0	0	0	2	0	0	0	0	0
Total	96	19	71	239	92	75	197	668	66	191	1179	6	27	1	769	29	4	14	373	1
Species richness	4	5	5	7	4	6	6	10	8	8	8	1	4	1	8	5	1	3	6	1
CPUE	24.0	4.8	17.8	59.8	23.0	18.8	49.3	167.0	16.5	47.8	294.8	1.5	6.8	0.3	192.3	7.3	1.0	3.5	93.3	0.3

Appendix C. Raw data collected from quantitative quadrate surveys in the lower Guadalupe River, Texas.

Site	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25
Reach	I	I	I	I	I	I	I	I	I	I	I	I	I	I	II	II	II	II	II	III	III	III	III	III	III
Habitat	BW	FPS	FPS	BH	R	MC	BH	BPS	P	BPS	R	P	BW	MC	BPS	FPB	MC	RFL	BH	P	R	MC	BH	R	BW
Amblema plicata	2	33	2	32	21	53	17	7	7	11	5	10	3	15	3	56	4	16	20	150	26	21	92	30	34
Arcidens confragosus	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Cyrtonaias tampicoensis	0	0	0	0	0	0	0	0	1	1	0	0	0	0	1	1	0	4	2	7	0	4	7	0	1
Fusconaia mitchelli	0	1	1	0	3	3	0	0	0	1	6	0	0	1	0	4	0	74	0	0	66	0	0	42	1
Lampsilis hydiana	0	0	0	0	0	1	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0	0	2	0	0
Lampsilis teres	1	2	2	2	0	1	1	1	0	1	1	0	0	1	3	5	0	1	1	1	0	0	5	0	8
Megalonaias nervosa	1	13	0	0	4	4	0	0	0	0	0	0	0	0	0	3	0	3	1	5	2	1	3	2	0
Pyganodon grandis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Quadrula aurea	1	11	0	14	24	36	4	4	1	8	4	9	0	11	5	32	0	157	1	21	229	22	7	117	2
Quadrula petrina	0	6	0	2	3	7	3	2	1	3	3	1	0	1	0	7	0	44	0	0	68	0	1	30	0
Quadrula verrucosa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0
Toxolasma parvum	0	0	0	0	0	0	0	0	1	0	0	0	0	0	6	0	0	0	10	0	0	0	0	0	0
Toxolasma texasense	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total	5	67	5	51	55	105	25	14	11	27	19	20	3	29	18	108	4	300	35	184	391	48	117	221	46
Density (m^2)	1	13.4	1	10.2	11	21	5	2.8	2.2	5.4	3.8	4	0.6	5.8	3.6	21.6	0.8	60	7	36.8	78.2	9.6	23.4	44.2	9.2

Appendix C. (*continued*)

Site	26	27	28	29	30	31	32	33	34	35	36	37	38	39	40	41	42	43	44	45	46	47	48	49	50
Reach	III	III	III	III	III	III	III	III	IV	IV	IV	IV	IV	IV	IV	IV	IV	IV	IV	IV	IV	IV	V	V	V
Habitat	FPS	BPS	MC	BW	P	BPS	FPS	BH	FPS	MC	BH	BPS	BH	R	FPS	BW	P	R	P	BW	BPS	MC	P	BH	MC
Amblema plicata	1	0	82	5	2	6	0	6	1	1	200	0	71	3	66	37	11	5	32	4	1	1	1	30	26
Arcidens confragosus	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Cyrtonaias tampicoensis	0	0	5	3	2	3	0	0	0	0	2	0	8	0	10	4	2	2	0	5	0	0	0	3	5
Fusconaia mitchelli	0	0	1	0	0	0	0	0	0	0	11	0	0	5	1	0	0	1	0	0	0	0	0	0	0
Lampsilis hydiana	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	1	1
Lampsilis teres	0	1	4	0	0	0	0	0	0	0	6	0	11	0	6	4	1	3	0	0	0	0	0	0	1
Megalonaias nervosa	0	0	0	0	0	0	0	3	0	0	16	0	2	0	1	0	0	0	0	0	0	0	0	1	0
Pyganodon grandis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Quadrula aurea	0	1	87	2	5	4	0	1	2	0	178	0	26	8	9	5	4	22	1	2	3	3	3	3	12
Quadrula petrina	0	0	3	0	0	0	0	0	0	0	11	0	4	3	4	2	0	0	2	1	0	0	1	0	1
Quadrula verrucosa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Toxolasma parvum	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
Toxolasma texasense	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total	1	2	182	10	9	13	1	10	3	1	424	0	122	19	97	52	18	33	35	13	4	4	5	39	46
Density (m^2)	0.2	0.4	36.4	2	1.8	2.6	0.2	2	0.6	0.2	84.8	0	24.4	3.8	19.4	10.4	3.6	6.6	7	2.6	0.8	0.8	1	7.8	9.2

Appendix C. (*continued*)

Site	51	52	53	54	55	56	57	58	59	60	61	62	63	64	65	66	67	68	69	70
Reach	V	V	V	V	V	V	V	V	V	V	VI	VI	VI	VI	VI	VI	VI	VI	VI	VI
Habitat	FPS	BPS	P	R	FPS	BPS	BW	BH	RFL	BW	BH	R	BW	FPS	P	BH	FPB	BPS	BW	BPS
Amblema plicata	13	5	0	17	6	13	5	104	7	21	56	0	3	0	95	1	1	0	18	0
Arcidens confragosus	0	0	0	0	0	0	0	0	0	1	1	0	0	0	1	0	0	0	0	0
Cyrtonaias tampicoensis	5	0	0	2	3	8	3	29	0	13	15	0	0	0	28	0	0	0	1	0
Fusconaia mitchelli	0	0	0	2	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0	0
Lampsilis hydiana	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
Lampsilis teres	3	1	0	0	1	0	0	3	0	6	2	0	0	0	24	1	0	0	1	0
Megalonaias nervosa	0	0	0	0	0	0	0	8	1	0	1	0	0	0	8	0	0	0	0	0
Pyganodon grandis	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Quadrula aurea	1	0	0	16	1	0	0	6	9	6	1	0	1	0	60	0	0	1	4	0
Quadrula petrina	0	0	0	2	0	0	0	2	1	0	1	2	0	0	7	0	0	0	0	0
Quadrula verrucosa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Toxolasma parvum	3	0	0	0	2	0	0	1	0	1	1	0	0	0	0	0	0	0	1	0
Toxolasma texasense	0	0	0	0	0	3	0	1	0	0	0	0	0	0	1	0	0	0	0	0
Total	25	6	0	39	13	24	8	154	20	48	78	2	4	0	225	2	1	1	25	0
Density (m^2)	5	1.2	0	7.8	2.6	4.8	1.6	30.8	4	9.6	15.6	0.4	0.8	0	45	0.4	0.2	0.2	5	0