AQUATIC CONSERVATION: MARINE AND FRESHWATER ECOSYSTEMS

Aquatic Conserv: Mar. Freshw. Ecosyst. **22**: 206–219 (2012)

Published online 23 February 2012 in Wiley Online Library
(wileyonlinelibrary.com). DOI: 10.1002/aqc.2226

Long-term changes in unionid assemblages in the Rio Grande, one of the World's top 10 rivers at risk

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ABSTRACT

1. According to the World Wildlife Fund, the Rio Grande is the most endangered river system in the North American continent and one of the World's top 10 rivers at risk, but is globally important for freshwater biodiversity. Unionid bivalves of the Rio Grande river basin used to be represented by a unique assemblage, including four endemic species (*Truncilla cognata, Potamilis metnecktayi*, *Popenaias popeii*, and *Quadrula couchiana*); however, surveys from 1998–2001 failed to recover any live endemic unionid species suggesting a sharp decrease in their populations and potential of extinction.

2. Intensive surveys (162 sites sampled) conducted by the authors from 2001–2011 on the Rio Grande and its tributaries in Texas recovered live *T. cognata, P. metnecktayi*, and the largest population of *P. popeii* ever reported. Overall the unionid assemblage of the Rio Grande basin has changed considerably during the last century.

3. Decline in species diversity, range fragmentation, local extirpations, and introduction of widespread common species were documented. Two species (*Q. couchiana* and *Quincuncina mitchelli*) are locally extinct. *Potamilus metnecktayi* and *T. cognata* have been extirpated from the Pecos River and their ranges in the Rio Grande have been reduced. *Popenaias popeii* has been extirpated from the Pecos River and Las Moras Creek along with the reduction and fragmentation of its range in the Devils River and Rio Grande.

4. Among the environmental factors responsible for the degradation of unionid assemblages in the Rio Grande river basin, the most important are impoundments, habitat degradation, salinization, pollution, and over-extraction of water.

Received 24 July 2011; Revised 18 November 2011; Accepted 04 January 2012

KEY WORDS: river; biodiversity; distribution; rare species; invertebrates; impoundments agriculture; salinity; Unionidae; threats

INTRODUCTION

A continuing dramatic increase in pollution, habitat destruction and introduction of invasive species is resulting in simplification and homogenization of ecosystems and a loss of biodiversity worldwide (Mckinney and Lockwood, 1999). Biodiversity loss is especially large in fresh waters, where many species are far more imperilled than their marine or terrestrial counterparts (Jackson *et al.*, 2001; Strayer and Dudgeon, 2010). This loss of diversity results from widespread habitat degradation, pollution, flow regulation, and water extraction, and these activities are predicted to increase in the

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future (Naiman and Turner, 2000; Jackson *et al.*, 2001; Strayer and Dudgeon, 2010). The opportunity to conserve much of the remaining biodiversity in fresh waters may vanish if trends in human demands for fresh water remain unaltered and species losses continue at present rates (Dudgeon *et al.*, 2006).

The Rio Grande is a globally important river for freshwater biodiversity, supporting numerous endemic fishes, birds, and molluscs (Groombridge and Jenkins, 1998; Revenga *et al.*, 1998, 2000; Johnson, 1999); however, because of the level of impacts affecting the Rio Grande at present (Dahm *et al.*, 2005), many of these species are now extinct and others are facing a sharp decrease in their population density or fragmentation in their range. Focusing analysis on river basins with high ecological importance and those with large human populations, the World Wildlife Fund recognized the Rio Grande River as the most endangered river in the North American continent, and one of the world's top 10 rivers at risk (Wong *et al.*, 2007).

The Rio Grande/Rio Bravo River (length: 2830 km, river basin area: 870 000 km^2) is one of the longest in North America, flowing from its headwaters in Colorado through New Mexico and then forming the shared border between Texas and Mexico before it empties into the Gulf of Mexico near Brownsville, Texas (Dahm *et al.*, 2005). It traverses seven physiographic provinces with a variety of habitats, but most of the basin is arid or semiarid with either desert shrubland or desert grassland (Dahm *et al.*, 2005). The Rio Conchos, the Pecos River and the Devils River historically contributed the main flow of the Rio Grande in the stretch between their confluences and Amistad Reservoir, although these flows have been reduced substantially and are stored at Amistad International Reservoir. Amistad Dam (completed in 1969) and Falcon Dam (completed in 1953) impound the Rio Grande along the border for irrigation and flood control. Evaporation from major reservoirs has been estimated to exceed the quantity of water used for municipal purposes in the basin, which constitute up to 5% of the agricultural consumption. From Laredo to the mouth of the Rio Grande, the river constitutes the primary source of drinking water for communities in both Mexico and the USA (Dahm *et al.*, 2005). Over 10 million people live in the Rio Grande basin, and urban areas are growing fast, particularly in border towns between the USA and Mexico. By 2060 the area from Eagle Pass to Brownsville is projected to almost triple in population (Texas Water Development Board, 2007). Irrigated agriculture is the primary use of the Rio Grande surface flow throughout the basin and accounts for more than 80% of all water taken from the river (Dahm *et al.*, 2005). The river bed between El Paso and Presidio frequently is dry, owing to water over-extraction for irrigation and domestic consumption, and since 2001 the river often fails to reach the Gulf of Mexico (Edwards and Contreras-Balderas, 1991; Contreras-Balderas *et al.*, 2002; Dahm *et al.*, 2005; Wong *et al.*, 2007; Douglas, 2009). Many other factors have contributed to the recent status of the Rio Grande, including persistent drought, increase in border populations, and subsequent declines in water quantity and quality (Dahm *et al.*, 2005; Wong *et al.*, 2007; Douglas, 2009).

Freshwater bivalves in the order Unionoida are considered to be one of the most endangered groups of animals in North America (Bogan, 1993; Lydeard *et al.*, 2004) with over 76% of the North American Unionidae and Margaritiferidae presumed extinct, threatened, endangered, or deemed of special concern (Williams *et al.*, 1993). Unionid bivalves of the Rio Grande river basin represent a unique assemblage and are distinct from the rest of Texas (Neck, 1982; Neck and Metcalf, 1988; Burlakova *et al.*, 2011a, b). The first data on unionid bivalves of the Rio Grande and its tributaries were published at the turn of the 19th century (Singley, 1893; Simpson, 1900, 1914). In the second half of the 20th century, numerous studies conducted on the Rio Grande system were summarized by Johnson (1999), who provided a detailed description of historical records and current distribution of all 15 species of unionids reported from this system. Extensive surveys done by Texas Parks and Wildlife Department in 1998–2001 failed to recover any live endemic unionid species from the Rio Grande, and Howells (2001) suggested that a sharp decrease in their populations may have put them on the edge of extinction. However, subsequent intensive surveys done by the authors in 2001–2011 recovered live *Truncilla cognata, Potamilis metnecktayi*, and the largest population of *Popenaias popeii* ever reported, proving that at least three endemic unionid species are still present in the river. The goals of this paper are: (1) to analyse the changes in the unionid assemblage of the Rio Grande river basin over 100 years; (2) to study the current distribution of the endemic species and estimate, whenever possible, their population densities; (3) to discuss major factors affecting unionid diversity and distribution in the Rio Grande.

METHODS

Data collection

To assess the current distribution of unionids, mussels were surveyed at 162 sample sites (subsites) pooled into 28 larger sites within the Rio Grande system during 2001–2011 (Figure 1). Fifteen of these sites were sampled once, while 13 sites were sampled from 2–25 times. Survey sites were often selected within state parks, near public boat ramps, or based on accessibility from roads that either crossed or approached a water body owing to the prevalence of private land in Texas, where only 2% of the lands remain in public ownership (Texas Parks and Wildlife Department, 1974). In addition, numerous sites were reached by canoe or kayak. When site surveys were conducted from private land, a Landowner Permission for wildlife research was acquired from each property owner before entering the property. The work was carried out with an appropriate Scientific Research Permit issued by the Texas Parks and Wildlife Department.

Sampling was completed by hand collection of both live and dead mussels, by wading in low water and by snorkelling or diving. Reconnaissance sampling (timed searches) and random searches were used at most sites to reveal the presence of mussels and species diversity (Strayer *et al.*, 1997; Vaughn *et al.*, 1997) and to compare with historical data. If significant mussel assemblages were present, quantitative methods (randomly placed 0.25 m^2 quadrats, mark-and-recapture surveys, or area searches) were used for assessments of density (Dunn, 2000; Strayer and Smith, 2003). Collected live mussels and shells were taxonomically identified, counted, and measured with calipers to the nearest millimetre. After measurements live mussels were carefully bedded into the sediment from which they were taken. Shell condition of dead mussels was recorded for each specimen.

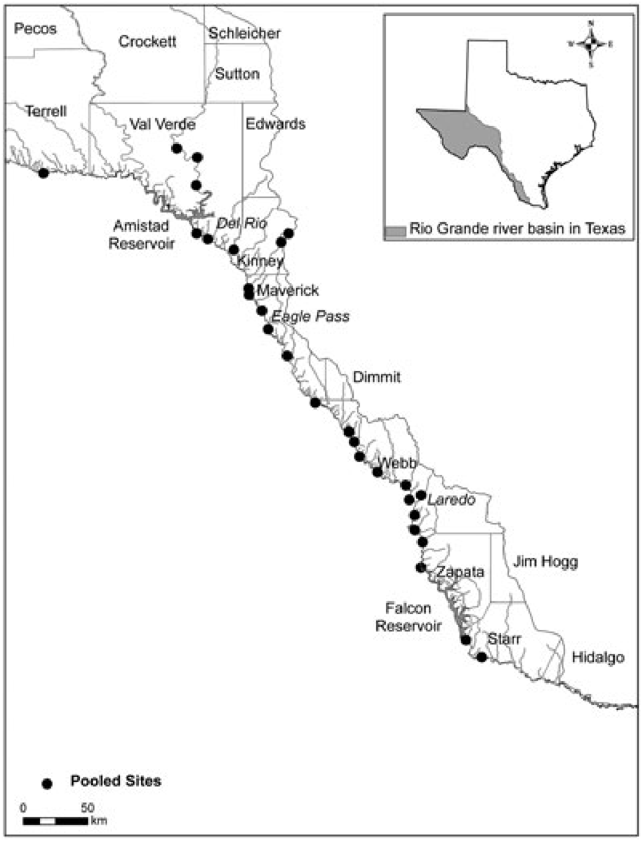

Figure 1. Map of the Rio Grande river basin in Texas with 28 pooled sampling sites surveyed during 2001–2011. Texas counties, major cities (in italics) and reservoirs are indicated.

A mark-and-recapture census was conducted at the La Bota Ranch site in Northern Laredo (Webb County) in March 2011 using methods described by Lang (2001) and Villella *et al.* (2004). Following recommendations by Villella *et al.* (2004), three consecutive days were sampled to estimate capture probabilities using closed population models. All mussels present (new captures, and recaptures) were measured (shell length, width, height ($\pm$ 0.1 mm)), and wet-weighed. First-time captured individuals were marked with unique numbers assigned by embedding oval (4×10 mm) Floy laminated flex tags in Super Glue Gel along the valve hinge posterior to the umbo, to one valve.

Specimens were identified using published taxonomic keys and descriptions (Howells *et al.*, 1996; Johnson, 1998). Voucher specimens were deposited into the Great Lakes Center Invertebrate Collection at Buffalo State College, Buffalo, NY. Each specimen was labelled with a unique number and catalogued in a database with the following information: specimen number, species name, name of person who collected and identified the specimen, date of collection, and detailed site information.

Data analysis

To estimate population density at the mark–recapture site the Schnabel method, an extension of the Petersen method to analyse a series of samples, was used (Krebs, 1999). To evaluate the total size of the *P. popeii* population the average density in the mark–recapture site and the estimation of available habitat area at the La Bota site near Laredo were used.

Differences in community structure were assessed with nonparametric multivariate statistical techniques on data matrices of all live species and their relative densities (as catch-per-unit of effort data, i.e. the number of live mussels for each species found per time search effort at each sampling site (mussels per person per hour). A square root transformation was used to normalize relative densities for the analysis. Similarity of the community composition was summarized by calculating Bray–Curtis distances – a measure of similarity with values ranging from 0 (identical samples) to 1, which is not influenced by rare species as other indices (Bray and Curtis, 1957; Clarke, 1993). To visualize the differences among assemblages, a non-metric multidimensional scaling (NMDS) was used, which calculates a set of metric coordinates for samples, most closely approximating their non-metric distances. Differences among communities were assessed by analysis of similarities (ANOSIM), a resampling technique that uses permutation/randomization methods on Bray–Curtis similarity matrices to identify differences among groups of samples (Clarke, 1993). These analyses were performed using PRIMER 6 software (Plymouth Routines in Multivariate Ecological Research, Version 6.1.6, Primer E-Ltd. 2006). All tests effects were considered significant if $P < 0.05$.

To analyse the historical data a database containing information of unionid species name, water body name, location, recorded date, and the collector's name was created using all available data including published records, museum collections, and web-based searches. Unionid assemblages in the Rio Grande system were analysed using the following time periods: (1) initial reports (before 1931), including collections made by the United States and Mexico Boundary Surveys mostly conducted in 1892 (Taylor, 1967), and data from Singley (1893), Ellis *et al*. (1930), and Strecker (1931); (2) 1968–1990 based mostly on data from Metcalf and Neck (Metcalf, 1974, 1982; Murray, 1975; Neck, 1984; 1987; Neck and Howells, 1984; Neck and Metcalf, 1988); (3) 1992–1999 based on Howells' data (Howells 1994, 1996a, b, 1997a, 1998, 1999, 2000); and (4) 2001–2011 based on the authors' data. Several assumptions were made in the analysis. If the status of a recorded unionid was not reported in the paper used for historical analysis, it was assumed that the specimen was found alive; if the date of collection was not reported in the paper, it was assumed that the mussel was recorded one year earlier preceding the publication year (excluding papers where museum collections were analysed).

RESULTS

Unionid diversity

This study showed that the Rio Grande still supports most of the unionid species previously reported from this river, including the regional endemics *Potamilus metnecktayi, Popenaias popeii*, and *Truncilla cognata* (Table 1). During the current study the most common unionid species were *Cyrtonaias tampicoensis* and *Quadrula*

Table 1. Historical and current records of live unionids (L) and their dead shells (D) from the Rio Grande drainage (excluding the Rio Grande River itself, RGD) and the Rio Grande River (including Falcon and Amistad reservoirs, RG) in Texas. n. r. - not recorded. Total number of species found dead is in parentheses

	Before 1931		1968–1990		1992–1999		2001–2011	
Species	RGD	RG	RGD	RG	RGD	RG	RGD	RG
Cyrtonaias tampicoensis	L	n. r.	L	L	L	L	L	L
Lampsilis teres	L	L	L	L	D	D	n. r.	L
Megalonaias nervosa	L	n. r.	n. r.	D	n. r.	D	n. r.	L
Potamilus metnecktayi[a]	n. r.	n. r.	L	L	n. r.	D	n. r.	L
Popenaias popeii[a]	L	n. r.	L	L	D	D	L	L
Potamilus purpuratus[b]	n. r.	n. r.	n. r.	n. r.	n. r.	L	n. r.	n. r.
Pyganodon grandis[b]	L	n. r.	L	n. r.	n. r.	n. r.	L	n. r.
Quadrula apiculata[b]	n. r.	n. r.	L	L	L	L	L	L
Quadrula couchiana[a]	L	n. r.	n. r.	n. r.	n. r.	n. r.	n. r.	n. r.
Quincuncina mitchelli[a]*	D	n. r.	n. r.	n. r.	n. r.	n. r.	n. r.	n. r.
Toxolasma parvus	L	n. r.	n. r.	n. r.	L	L	n. r.	n. r.
Toxolasma texasensis	L	n. r.	L	n. r.	n. r.	n. r.	L	n. r.
Truncilla cognata[a]	n. r.	n. r.	L	L	n. r.	n. r.	n. r.	L
Uniomerus sp.	n. r.	n. r.	L	n. r.	n. r.	n. r.	n. r.	n. r.
Utterbackia imbecillis	L	n. r.	L	L	L	L	L	L
Total	9 (1)	1	10	7 (1)	4 (2)	5 (4)	6	8

[a]Regional endemics
[b]Introduced species
*Only fossil and greatly weathered specimens are known from Texas part of Rio Grande drainage.

apiculata, found alive at 28.6% of sites sampled (Table 2). The percentage of sites where live molluscs were found compared with the total number of sites where live and dead specimens were found was the greatest for *Q. apiculata* (73%), *Megalonaias nervosa* and *P. popeii* (58% each), and the lowest for *T. cognata* (17%) and *P. metnecktayi* (13%). The rarest species was *P. metnecktayi*, which was found alive at only one location. Other rarely recorded species were *Utterbackia imbecillis* and *Toxolasma texasensis*, which were found mostly in tributaries (Table 2). The highest diversity of unionids was found in a 24 km stretch of the Rio Grande above Laredo (Figure 2(C), 3(B), 4(D)). No live mussels were found below Amistad Reservoir and few below Laredo. Two distinct unionid assemblages depending on the substrate type were found in the Rio Grande above Laredo (Figure 5, $R = 0.942$, $P = 0.001$, one-way ANOSIM). On soft and unconsolidated sediments (silt, sand, small gravel, and combinations of these) unionid assemblages were dominated by *Q. apiculata*, and *C. tampicoensis*; additional species were *M. nervosa* and *T. cognata*. On bedrock and boulders the dominant species was *P. popeii*. This species was most commonly found in crevices under flat boulders resting on the bedrock. Often up to 10 individuals were found under one rock. Additional unionids found in this habitat included *Lampsilis teres, Q. apiculata*, and *T. cognata*.

Endemic species account

Potamilus metnecktayi

Nineteen live *P. metnecktayi* were found in the Rio Grande at the John's Marina site, south of Dryden, Terrell County in 2003–2008 (Figure 2(C)). Mussels were generally found along the shores, in soft sediments (in a mixture of silt and clay) at 0.5–1.2 m depth (at low flows ~30 m^3 s^{-1}). Their size varied from 63 to 124 mm, averaging 87.1 mm (±17.6 standard deviation). Dead shells of *P. metnecktayi* were found at seven more sites. *P. metnecktayi* had the lowest percentage of sites where live mussels were found, from the total number of sites where shells of the species were recorded (13%). At 15 sites below Lake Amistad, only 50 long-dead or sub-fossil valves

Table 2. Occurrence of unionid species in the Rio Grande river drainage, and separately in the river main stem and its tributaries based on 2001–2011 surveys. In total, 28 pooled sites were studied in the Texas part of the drainage, including 21 sites in the Rio Grande River (excluding reservoirs) and seven sites on tributaries. Species occurrence was calculated as a number of sites where the species was found, and percentage occurrence was calculated as the percentage of sites where the species was found. Single valves were counted as half of a shell

	Rio Grande drainage			Rio Grande River			Tributaries only		
Species	Total found	Occurrence (number of pooled sites)	Percentage occurrence	Total	Occurrence (number of pooled sites)	Percentage occurrence	Total	Occurrence (number of pooled sites)	Percentage occurrence
Live mussels									
Cyrtonaias tampicoensis	89	8	28.6	29	7	33.3	60	1	14.3
Lampsilis teres	17	2	7.1	17	2	9.5	0	0	0
Megalonaias nervosa	34	7	25.0	34	7	33.3	0	0	0
Popenaias popeii	656	7	25.0	649	5	23.8	7	2	28.6
Potamilus metnecktayi	19	1	3.6	19	1	4.8	0	0	0
Quadrula apiculata	204	8	28.6	129	7	33.3	75	1	14.3
Toxolasma texasensis	11	1	3.6	0	0	0	11	1	14.3
Truncilla cognata	19	2	7.1	19	2	9.5	0	0	0
Utterbackia imbecillis	7	1	3.6	0	0	0	7	1	14.3
Total live mussels	**1056**	**14**	**50.0**	**896**	**11**	**52.4**	**160**	**3**	**42.9**
Shells									
Cyrtonaias tampicoensis	789	20	71.4	788	19	90.5	1	1	14.3
Lampsilis teres	84.5	9	32.1	84.5	9	42.9	0	0	0
Megalonaias nervosa	180.5	12	42.9	180.5	12	57.1	0	0	0
Popenaias popeii	473.5	12	42.9	465	11	52.4	8.5	1	14.3
Potamilus metnecktayi	159.5	8	28.6	159.5	8	38.1	0	0	0
Quadrula apiculata	533.5	11	39.3	533	10	47.6	0.5	1	14.3
Toxolasma texasensis	1	1	3.6	0	0	0	1	1	14.3
Truncilla cognata	291	12	42.9	291	12	57.1	0	0	0
Utterbackia imbecillis	57	10	35.7	17	**7**	33.3	40	3	42.9
Total shells	**2569.5**	**21**	**75.0**	**2518.5**	**19**	**90.5**	**51**	**3**	**42.9**

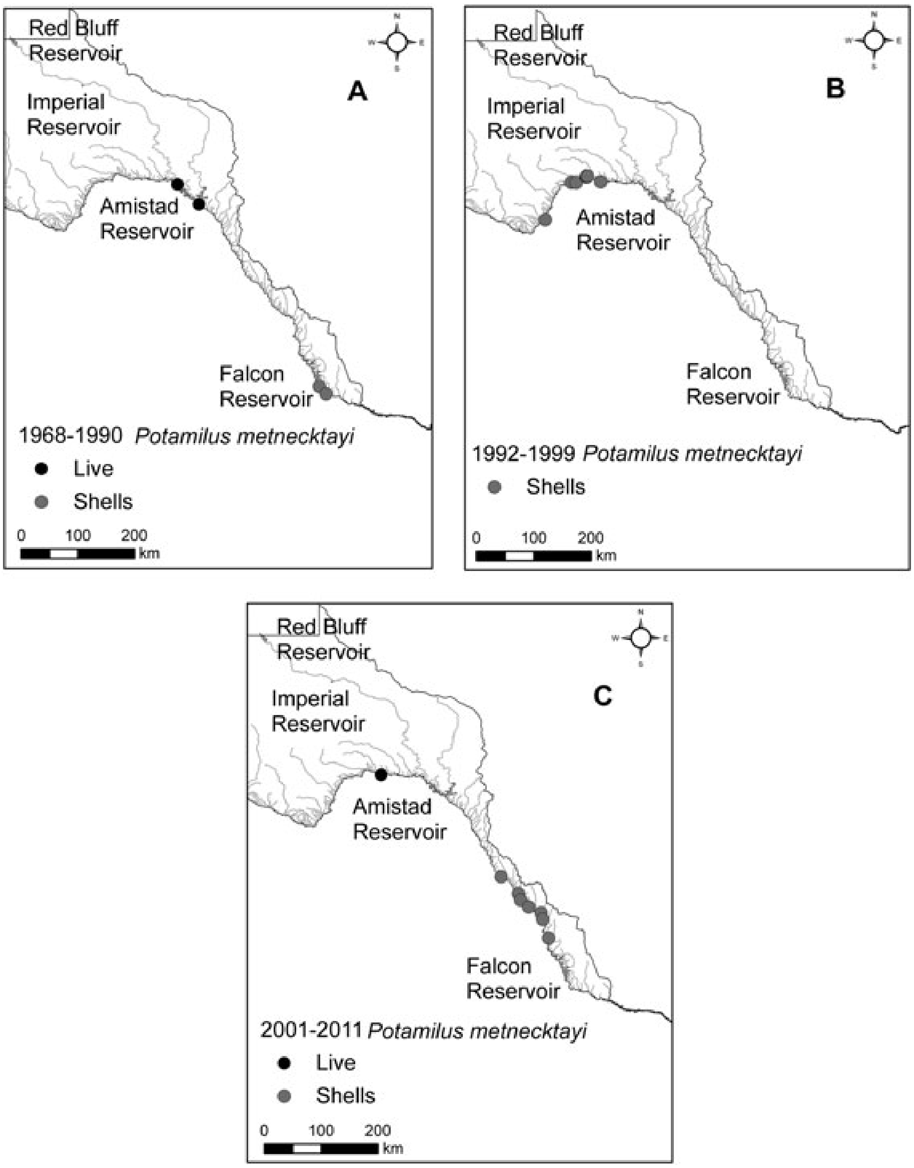

Figure 2. Map of the Rio Grande river basin in Texas with sites where live and/or dead shells of *Potamilus metnecktayi* were found in 1968–1990 (Metcalf, 1974, 1982; Murray, 1975; Neck and Howells, 1984; Neck, 1987; Neck and Metcalf, 1988) (A); in 1992–1999 (Howells 1994, 1996a,b, 1997a, 1998, 1999, 2000) (B); and from 2001 to 2011 (authors' data) (C).

were found, possibly indicating a once widespread population.

Truncilla cognata

In total, 19 live *T. cognata* were found from 2001 to 2011 in the Rio Grande River in Laredo, Webb County. Most molluscs were found down to 15–20 cm deep in a mixture of gravel and sand, and between large boulders. Because of its small size, it was difficult to distinguish *T. cognata* from gravel, adding to the difficulty of detecting this cryptic species. Many excavations were made below the Water Treatment Plant in Laredo, but no live mussels were found there. In 2011 12 *T. cognata* were found at five subsites examined in and above Laredo. Most of them were found in unconsolidated sediments (sand with some silt), captured in shallow protected areas adjacent to gravel riffles. Their size varied from 20.5 mm to 33 mm (average 28.4 ± 4.1 mm). Dead shells of *T. cognata* were found at 12 sites (Figure 3(B)). Very recently dead specimens (i.e. shells with flesh, to 51 mm) were found at four subsites below Laredo into Zapata County. Based on these data, it is likely that additional specimens may be found in Pinto Valle Creek (Webb County) and Dolores Creek (Zapata County). All of the 19 live *T. cognata* from the current study have been found at the confluences of Santa Isabel, Sombrerito, and Zacate Creeks above

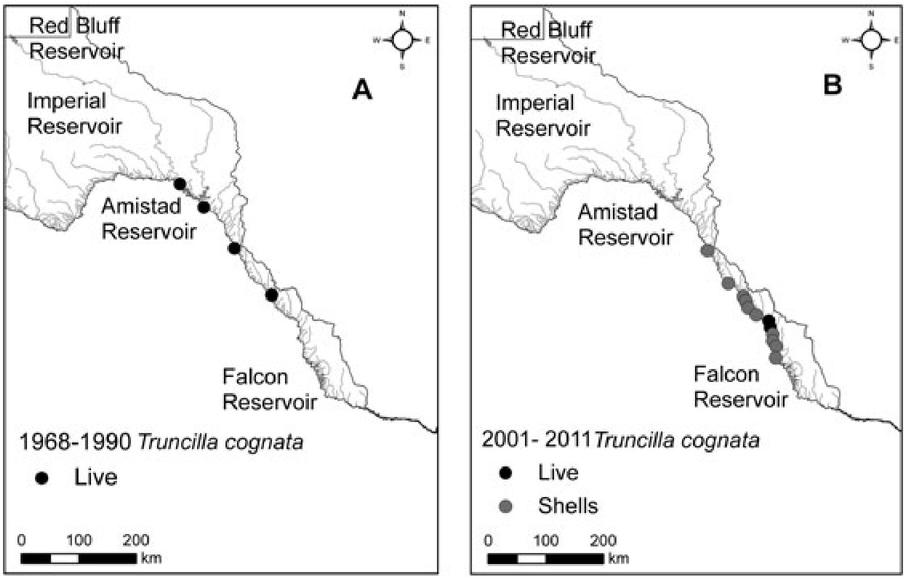

Figure 3. Map of the Rio Grande river basin in Texas with sites where live and/or dead shells of *Truncilla cognata* were found in 1968–1990 (data from Metcalf, 1974, 1982; Murray, 1975, Neck and Howells, 1984; Neck, 1987; Neck and Metcalf, 1988) (A), and from 2001 to 2011 (authors data) (B).

Laredo. Their presumed habitat preference of small gravel/sand/silt mixed substrates is also well known as each of these areas has or had a sand and gravel excavation site nearby.

Popenaias popeii

During 10 years of the current survey, one live *P. popeii* was found in the Rio Grande River in Terrell County (John's Marina), seven live in the Devils River, and 648 live in the Rio Grande near Laredo. Live mussels were found at seven sites, and dead shells were found at a further five sites (Figure 4(D)). Most live mussels were found at the La Bota mark-and-recapture subsite (in Laredo) which had an abundance of low-flow refuges occurring under large boulders, where sand and clay seams provide substrates for mussels. At this mark–recapture site (area sampled *c.* 1000 m^2) 406 live *P. popeii* were found. The recovery rate was 11.7% (18 of 154 mussels marked) on the second day, and was 6.5% (17 of 260 mussels marked) on the third day (9.1% in average). Therefore, the total population may be near 1500 at the site, with a density of ~1.5 m^{-2}. This population consisted of multiple age-classes, with shell lengths ranging from 33.2 to 87 mm (63 ± 1, mean $\pm$ 95% confidence interval). Over a third of the mussels measured were less than 60 mm, and 12 individuals were less than 45 mm in length. Considering that the total area of similar substrate upstream of this site was ~ 3200 m^2, and assuming similar densities, up to 4700 individuals of this species may be in this area. At three other subsites located *c.* 1.6 km downstream from this mark-and-recapture locality, 182 live *P. popeii* were found in 3 person-hours of timed searches. These subsites were located along a 280 m river stretch, and may contain up to 4000 more mussels. Therefore, the total population of *P. popeii* in the La Bota area may contain up to 8700 mussels.

DISCUSSION

Long-term changes in unionid diversity

The unionid assemblage of the Rio Grande drainage has changed significantly over the last century (Table 1, 3). Although the Rio Grande itself still supports the majority of unionid species ever reported alive in this river, its unionid assemblage has faced decline in species diversity, range fragmentation, local extirpations, and introduction of widespread common species. Two species (*Quadrula couchiana* and *Quincuncina mitchelli*) are already extinct from the Texas part of the Rio Grande basin. The most drastic changes were recorded during the last 40 years (Table 3).

Several streams and rivers of the Rio Grande drainage have lost all or a significant number of unionid species, including Las Moras Creek in Fort Clark (Brackettville, Kinney County), the Devils River and the Pecos River (Table 4). Along with the local extirpation of rare and endemic species from the Rio Grande drainage, the unionid

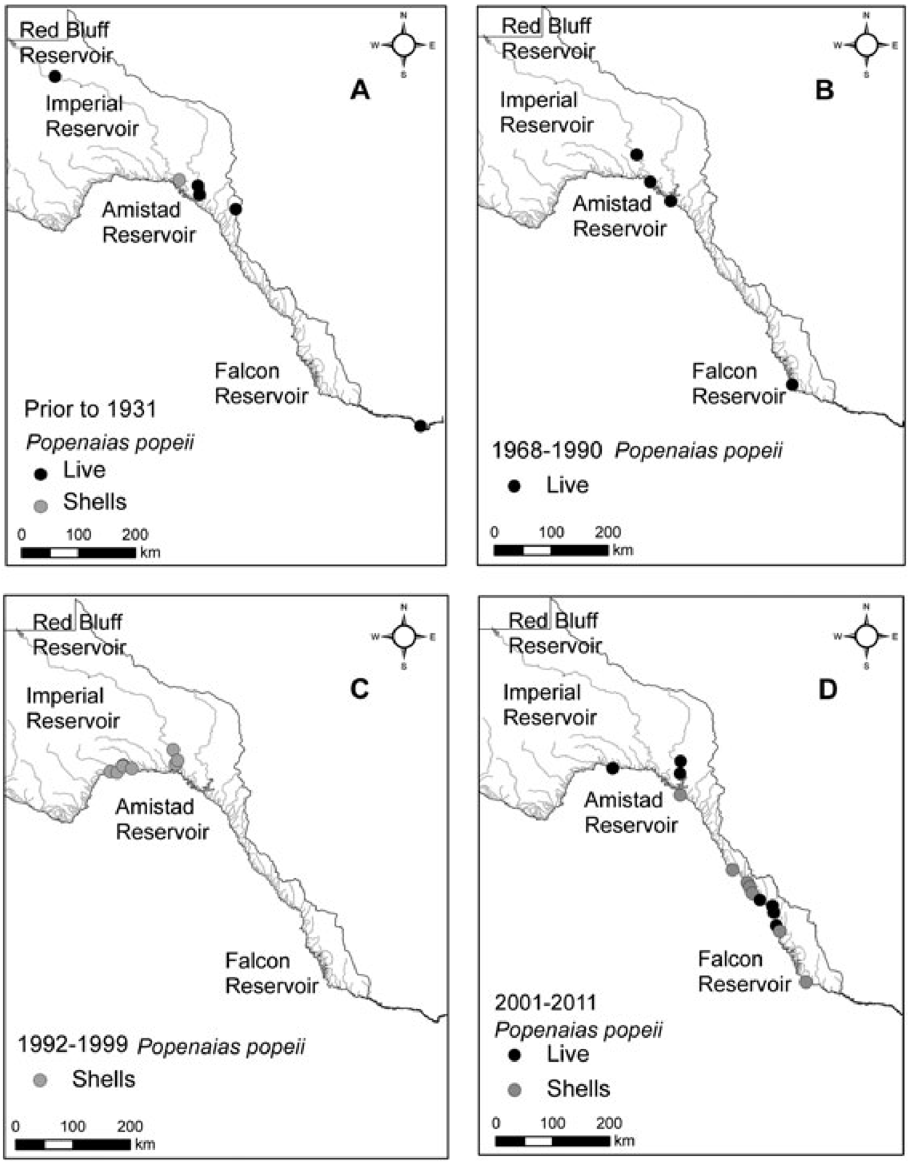

Figure 4. Map of the Rio Grande river basin in Texas with sites where live and/or dead shells of *Popenaias popeii* were found before 1931 (based on data from Singley, 1893; Ellis *et al.*, 1930; Strecker, 1931; Taylor, 1967) (A); in 1968–1990 (Metcalf, 1974, 1982; Murray, 1975, Neck and Howells, 1984; Neck, 1987; Neck and Metcalf, 1988) (B); in 1992–1999 (Howells 1994, 1996a, b 1997a, 1998, 1999, 2000) (C); and from 2001 to 2011 (authors' data) (D).

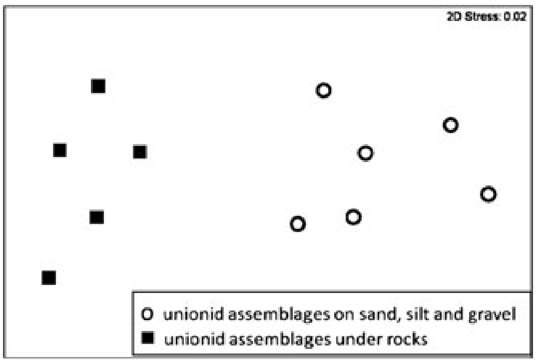

Figure 5. NMDS ordination plot of the unionid assemblages in the Rio Grande near Laredo found on sand, silt and gravel and under rocks. Relative density data (mussels per person per hour) for live molluscs collected at all sampled sites (excluding sites where fewer than two species were collected) were square-root transformed and converted to a similarity matrix using the Bray–Curtis similarity index. There was a significant difference in assemblage structure among the two substrates (Global R = 0.942, $P = 0.001$, one-way ANOSIM).

assemblage was reshaped by the introduction of common species (*Q. apiculata, P. purpuratus*, and *P. grandis*) non-native to this drainage (Metcalf and Smart, 1972; Metcalf, 1982; Johnson, 1999). In the 20th century *Q. apiculata* became very common in the Rio Grande and its tributaries. Previous research noted a lack of fossil *Q. apiculata* (Metcalf, 1982), and no fossil specimens were found during this study. Current data indicate slow, upriver range extension of *Q. apiculata* with greater abundance in Casa Blanca and Falcon reservoirs. *Potamilus purpuratus* has been recorded in the Amistad Reservoir in 1994, 1995 and 1998 (Howells, 1997b, 1999). Historical records of *P. purpuratus* from the Rio Grande river basin (Singley, 1893) have been shown to be

Table 3. Long-term changes in unionid diversity in the Texas part of the Rio Grande drainage

Time period	Changes
Before 1900	Extinction of *Quadrula couchiana* from the Rio Grande drainage
	Introduction of *Pyganodon grandis*
1900–1970	Extinction of *Q. mitchelli* from the Rio Grande drainage
	Introduction of *Q. apiculata*
1970–2010	Local extirpations of *Popenaias popeii, Potamilus metnecktayi, Truncilla cognata*
	Range fragmentation of *P. popeii, P. metnecktayi, T. cognata*
	Introduction of *Potamilus purpuratus*
	Range expansion of *Q. apiculata*

misidentified specimens of *C. tampicoensis* (Neck and Metcalf, 1988; Johnson, 1999). Another introduced species, *P. grandis*, was reported from the Granjeno Lake in 1892 (Singley, 1893) and canals in Hidalgo County (Ellis *et al*., 1930), from the El Toro Cement Agency Lake in El Paso in 1969 (Johnson, 1999), and in the Topaz Power Plant cooling pond, Laredo in 2006 (T. Miller unpublished data).

Endemic species accounts

Potamilus metnecktayi

This regional endemic was reported to be extremely rare in the Rio Grande in Texas (Neck and Metcalf, 1988), uncommon even at the fossil localities sampled in New Mexico and Mexico (Metcalf, 1982), and it has been recently added to the state's list of threatened species (Texas Register 35, 2010). Live specimens in the USA were collected in Texas only by Metcalf on the Rio Grande 9.7 km west of Del Rio in 1972, and by Taylor in the Pecos River 1.28 km above its mouth at the former crossing of US Hwy 90 in 1968 (Metcalf, 1982) (Table 4, Figure 2). No live or dead *P. metnecktayi* were found in the Del Rio area during sampling in 2008. Only dead shells of *P. metnecktayi* were found in Texas since the mid-1970s (Howells, 1994, 1999, 2000; Howells *et al*., 1997; Figure 2(B)). Our discovery of 19 live and numerous shells of *P. metnecktayi* in the Rio Grande by Johnson Marina, Terrell County, proves that this species still exists in the middle Rio Grande, although its distribution range was significantly reduced during the 20th century. Additional studies are urgently needed to estimate the current distribution and population size of *P. metnecktayi* in the Rio Grande considering the subsequent catastrophic floods in 2008 and 2010, and to develop appropriate measures for the species' conservation.

Truncilla cognata

Truncilla cognata is another regional endemic that was described from the Devils River, Texas, and Rio Salado, Nuevo Leon, Mexico (Lea, 1857; Johnson, 1999). This species has a NatureServe

Table 4. Historical and current records of live *Potamilus metnecktayi, Truncilla cognata*, and *Popenaias popeii* in the Texas part of the Rio Grande drainage

Water body	Historical collections	Current status
Potamilus metnecktayi		
Rio Grande, 9.7 km West of Del Rio	1972 (Metcalf, 1982)	No live mussels were found
Rio Grande, Johnson Marina, Terrell County	No historical records from this location	19 live specimens were collected by the authors 2003–2008
Pecos River, 1.28 km above its mouth at the former US Hwy 90 crossing	1968 (Metcalf, 1982)	Flooded by Amistad Reservoir. No live mussels were found
Truncilla cognata		
Rio Grande, 9.7 km West of Del Rio	1972 (Metcalf, 1982)	No live mussels found
Rio Grande, Laredo	No historical records from this location	19 mussels total were found by the authors at two sites 2001–2011
Pecos River, 1.28 km above its mouth at the former US Hwy 90 crossing	1968 (Metcalf, 1982)	Flooded by Amistad Reservoir (population probably extirpated)
Popenaias popeii		
Las Moras Creek, Kinney County	1892 (Taylor, 1967)	No live mussels were found. Population extirpated (Murray, 1975)
Devils River, Val Verde County	1892 (Singley, 1893)	7 live mussels were found by authors 2008–2011
Pecos River, Val Verde County	1903, 1968, 1972, 1973 (Metcalf, 1982)	Flooded by Amistad Reservoir. No live mussels were found
Rio Grande, 9.7 km West of Del Rio	1972 (Metcalf, 1982)	No live mussels were found
Rio Grande, 2.3 km downstream of Falcon Dam, Starr County	1975 (Neck and Metcalf, 1988)	No live mussels were found
Rio Grande, Laredo	No historical records from this location	645 live mussels were found by the authors in 2002–2011
Rio Grande, Johnson Marina, Terrell County	No historical records from this location	1 live specimen was collected by the authors in 2008

global status of 'critically imperilled' (NatureServe, 2009), is considered endangered by the American Fisheries Society, and has recently been added to the state's list of threatened species (Texas Register 35, 2010). *Truncilla cognata* is currently under consideration for federal listing by the US Fish and Wildlife Service (USFWS) (Federal Register 74, 2009). In the USA, *T. cognata* was reported only from a few sites in Texas (Table 4, Figure 3) with no living or dead specimens collected since 1972 (Howells *et al.*, 1997; Howells, 2001). Again it is likely that the Pecos River population of *T. cognata* is already extirpated and the 19 live specimens that were found in the Rio Grande near Laredo in 2001–2011 represent the only known population of this species left in the US.

Popenaias popeii

Popenaias popeii is known from the Rio Grande drainage in Texas (Singley, 1893; Taylor, 1967; Neck, 1987), Black River in New Mexico (Lang, 2001; Carman, 2007), and several Mexican tributaries of the Rio Grande (Simpson, 1914; Johnson, 1999; Strenth *et al.*, 2004). Strecker (1931) reported that *P. popeii* 'seems to be rather scarce', Stansbery (1971) listed this species as 'rare and endangered', and Neck (1984) included it in his list of restricted and declining species of Texas. NatureServe ranks *P. popeii* as critically imperilled across its range (NatureServe, 2009). This species has recently been added to the state's list of threatened species (Texas Register 35, 2010), and is currently considered a candidate for listing (priority 8) under the federal Endangered Species Act.

In Texas, live *P. popeii* were reported from Las Moras Creek (Taylor, 1967), the Devils (Singley, 1893) and Pecos Rivers (Metcalf, 1982), and from two distinct areas in the Rio Grande (Metcalf, 1982; Neck and Metcalf, 1988) (Table 4, Figure 4). Only two dead shells of *P. popeii* were reported in Texas outside the Rio Grande drainage in the South Concho and Llano Rivers (Strenth *et al.*, 2004). There is no evidence that these records represent extant populations of *P. popeii*.

No live *P. popeii* had been found in the Rio Grande since the mid-1970s (Howells, 2001). Our discovery of seven live *P. popeii* in the Devils River in 2008–2011, and 45 live *P. popeii* in 2002–2008 in the Rio Grande River confirmed that the species was still present in Texas. However, more significant was the discovery of a large population (604 live specimens recorded) of *P. popeii* in 2011 in Laredo. The conservative estimate of more than 8000 individuals made this Laredo population by far the largest ever reported from Texas, New Mexico or Mexico. This population consisted of multiple age-classes suggesting the recruitment of juvenile mussels and thus a healthy reproducing population. This also implies that a healthy host fish population occurs in this reach of the river, which is very important for unionid reproduction, and future population survival.

These particular refuges in upper Laredo may be vulnerable to excess water fluctuations including periods of low water and flood. During a low-flow period (22.6 $m^3 s^{-1}$) in December, 2002, snowy egrets (*Egreta thula*) were observed feeding on *P. popeii*. Another site on Zacate Creek (Las Palmas Park, a TPWD mussel sanctuary) where more than 50 live mussels of six species (including numerous *P. popeii*) were found over the years, has been smothered by cobble deposited by the July 2010 flood. No live mussels were recorded at this site since this last flood. Specimens of *P. popeii* do not appear to survive well in the Rio Grande downstream of Zacate Creek (Las Palmas Park, Laredo). Only one live mussel and two shells have been found in numerous shore surveys along the 80 km downstream reach of the river to Falcon Lake.

Another important finding was suitable habitat for *P. popeii* in the Rio Grande. This is similar to the preferred habitat for this species in the Black River: low-flow refuges characterized by aggregations of mussels under large boulders of limestone conglomerates, where clay seams provide stable substrates for mussels in low-velocity microhabitats (Lang, 2010). This habitat is different from the soft substrate type preferred by other species such as *C. tampicoensis*, *T. cognata*, *M. nervosa*, and *Q. apiculata* (Figure 5).

Environmental factors affecting unionids

The Rio Grande is at present one of the most impaired rivers in the world, with both water quantity and water quality issues being the major concerns (Dahm *et al.*, 2005). We suggest that among various types of human activities on the Rio Grande drainage, most destructive for unionid assemblages are impoundments, habitat degradation, salinization, pollution, and over-extraction of water (Table 5).

Impoundments

Some species may now be extinct in the Pecos system because of impoundment of its lowermost part by Amistad Reservoir (Metcalf and Stern, 1976). Creation of Falcon Reservoir most likely decimated the lotic habitat of the bivalves in the lower Rio Grande (Neck and Metcalf, 1988). In south-eastern New Mexico, the construction of impoundments (Lake MacMillan, Brantley and Avalon reservoirs) was one of the many factors responsible for extirpation of *P. popeii* from the Pecos River mainstem (Taylor, 1967). The construction of reservoirs also facilitated the introduction and range expansion of common species (*Q. apiculata, P. purpuratus*, and *P. grandis*) non-native to the Rio Grande river drainage (Metcalf and Smart, 1972; Metcalf, 1982; Johnson, 1999). Low-head dams on the Black River apparently preclude opportunities for recolonization by *P. popeii* in upstream riverine reaches and with downstream recolonization potentially limited by altered physicochemical (salinity gradient) and flow regimes (Lang, 2001). Any future projects to construct a new dam, or to modify existing low-head dams and associated water diversion structures, both on the Black River or in the Rio Grande River in Laredo, could potentially have impacts on *P. popeii*.

Salinity

Salinity concentrations in the Rio Grande are the result of both human activities and natural conditions: the naturally salty waters of the Pecos River are a major source of the salts that flow into Amistad Reservoir and continue downstream. Salinity may be the major factor limiting *P. popeii* distribution in the Pecos River and in the Rio Grande below its confluence with the Pecos River. In laboratory studies *P. popeii* shows behavioural signs of physiological stress, followed by death, at a salinity of 7.0 psu (Lang, 2001). Salinity in the area of the Black River occupied by *P. popeii* is approximately 0.9 psu. It increases significantly downstream to 2.8 psu and, in the Pecos River, ranges from 6.0–7.0 psu downstream of the confluence with the Black River (Lang, 2001). This increased salinity may have prevented populations becoming established in the main stem of the Pecos River even before its impoundment.

Table 5. Environmental pressures affecting unionid assemblages in the Texas part of the Rio Grande drainage

Environmental pressure	Effect
Impoundments	• Extirpation of *P. metnecktayi, T. cognata* and *P. popeii* from the lower Pecos River flooded by Amistad Reservoir • Decreased range of *P. metnecktayi* and *T. cognata* in the Rio Grande • Introduction of *P. grandis*, and *P. purpuratus*
Habitat degradation and pollution	• Extirpation of all unionids, including *Q. mitchelli* and *P. popeii* from Las Moras Creek • Decreased or fragmented range of all unionids, including *P. popeii, P. metnecktayi*, and *T. cognata* in the Rio Grande
Salinization	• Extirpation of all unionids, including *P. popeii* from the Pecos River
Over-extraction of water	• Decreased or fragmented range of all unionids, including *P. popeii, P. metnecktayi*, and *T. cognata* in the Rio Grande

Over-extraction, habitat destruction, and pollution

Water diversion from the middle Rio Grande is so high that the river bed between El Paso and Presidio/Ojinaga often lies dry (Dahm *et al*., 2005; Wong *et al*., 2007; Douglas, 2009). Evapotranspiration, groundwater recharge, and human appropriation of Rio Grande water has resulted in less than 1% of basin precipitation reaching the mouth, and failures to reach the Gulf of Mexico were recorded in much of 2002 and 2003 (Dahm *et al*., 2005). Growth in water demand from agricultural economic activity near the Mexican border and regional maquiladoras (manufacturing or export assembly plants in northern Mexico that produce parts and products for the USA) resulted in more than a 5-fold loss of lower Rio Grande stream flow between 1905–1934 and 1951–1980 (reviewed in Douglas, 2009). The population in the basin was about 13 million inhabitants in 1990, and increased along the Texas border by 27% between 1980 and 1990, and by 26% on the Mexican side. As a result of low water levels, the concentration of pollutants is very high; salinization has already displaced 32 native freshwater fish species, while marine fish species are invading as far as 400 km upstream (Contreras and Lozano, 1994). In addition to salinization, water quality problems include elevated nutrients, bacteria, metals, pesticides, herbicides, and organic solvents (Dahm *et al*., 2005). Another major change in the Rio Grande in recent years has been the disconnection of the river from the floodplain (Molles *et al*., 1998);

the fragmentation of river channels by dams, diversions and depletions has eliminated the natural flood pulse, reducing productivity and altering the structure of riparian ecosystems.

Protection

All three endemic species (*P. metnecktayi, T. cognata*, and *P. popeii*) have been added by the Texas Parks and Wildlife Department (TPWD) to the list of state-threatened species in 2010 (Texas Register 35, 2010). However, the state protection only prohibits the 'take' of a state-threatened species. Since 2009 these species are under consideration for federal listing by the USFWS (Federal Register 74, 2009), but listing of these species has not yet been warranted (Federal Register 76, 2011). *Popenaias popeii* was petitioned to the Candidate list as a Federally Endangered Species with Critical Habitat in 2004, and is currently considered a Candidate Species under the Federal Endangered Species Act (Federal Register 71, 2006). Although USFWS encourages conservation of these species, candidate species receive no statutory protection under the Endangered Species Act.

In 1978, a 315 km stretch of the United States side of the Rio Grande along the Mexican border was designated as a National Wild and Scenic River (National Parks and Recreation Act of 1978, Public Law 95–625, November 10, 1978). The river segment begins in, and is administered from, the Big Bend National Park in Brewster County and continues to the Terrell and Val Verde County border, thereby encompassing the area where the extremely rare *P. metnecktayi* and a few specimens of *P. popeii* were found. The Wild and Scenic Rivers Act prohibits federal support for construction of dams, water conduits, reservoirs, or other instream activities that would harm the river's free-flowing condition, water quality, or outstanding resource values. However, the designation neither prohibits development nor gives the federal government control over private property, and does not affect existing water rights. Although the part of the Rio Grande in and above Laredo where we found the only large known population of *P. popeii* has the status of a mussel sanctuary (where mussel harvest is prohibited) (Texas Register 31, 2006), additional protection is urgently necessary as any activity associated with water flow alteration could potentially damage the remaining habitat of *P. popeii*.

ACKNOWLEDGEMENTS

Surveys were conducted in conjunction with the US Geological Survey. Many surveys utilizing multi-day kayak or canoe trips were accomplished with the assistance of Dr Tom Vaughan (Texas A&M International). This study was funded in part by the US Fish and Wildlife Service, Texas Parks and Wildlife Department (TPWD), and New Mexico Department of Game and Fish (NMDGF) (Joint Traditional Section 6 Project 407348 to LEB, AYK, B. Lang (NMDGF) and M. May (TPWD) and by the US FWS State Wildlife Grant Program through the TPWD (PIs LEB, AYK, M. E. May, M. D. Warriner, and B. Gottfried, TPWD). LEB was also supported by the Research Foundation of SUNY. We thank Mary F. Perrelli (Buffalo State College, Geography and Planning Department) for help with GIS. This study would not have succeeded without the assistance of B. Lang (NMDGF), D. J. Berg and K. Inoue (Miami University, Oxford, OH), and Y. Zhang and T. Noble (Texas State University – San Marcos).

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