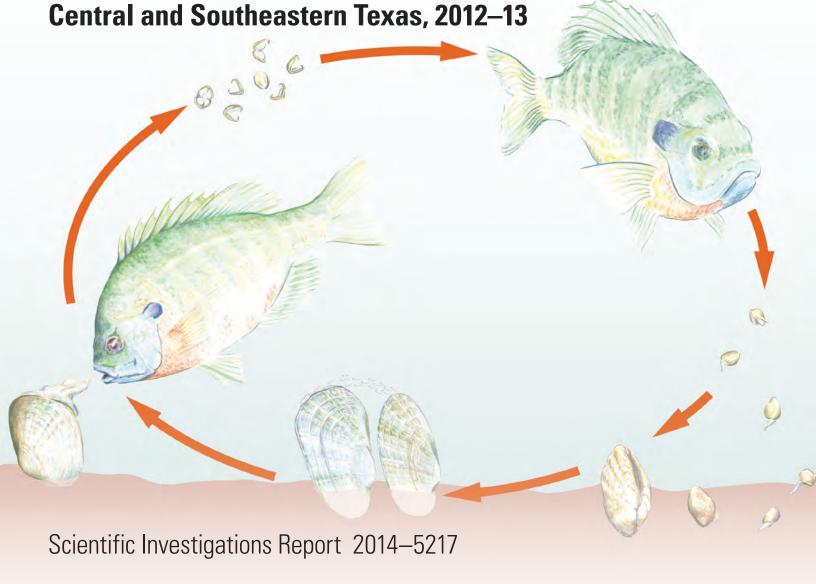


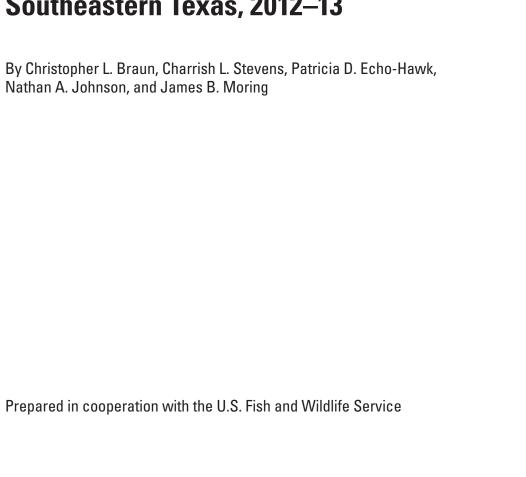
Prepared in cooperation with the U.S. Fish and Wildlife Service

Abundance of Host Fish and Frequency of Glochidial Parasitism in Fish Assessed in Field and Laboratory Settings and Frequency of Juvenile Mussels or Glochidia Recovered from Hatchery-Held Fish,





Abundance of Host Fish and Frequency of Glochidial Parasitism in Fish Assessed in Field and Laboratory Settings and Frequency of Juvenile Mussels or Glochidia Recovered from Hatchery-Held Fish, Central and Southeastern Texas, 2012–13



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Conversion Factors and Datums

SI to Inch/Pound

Multiply	Ву	To obtain
	Length	
millimeter (mm)	0.0394	inch (in.)
centimeter (cm)	0.394	inch (in.)
meter (m)	3.28	foot (ft)
kilometer (km)	0.6215	mile (mi)

Temperature in degrees Celsius (°C) can be converted to degrees Fahrenheit (°F) as °F = (1.8 \times °C) + 32.

Datum

Horizontal coordinate information is referenced to the North American Datum of 1983 (NAD 83).

Abundance of Host Fish and Frequency of Glochidial Parasitism in Fish Assessed in Field and Laboratory Settings and Frequency of Juvenile Mussels or Glochidia Recovered from Hatchery-Held Fish, Central and Southeastern Texas, 2012–13

By Christopher L. Braun,¹ Charrish L. Stevens,² Patricia D. Echo-Hawk,² Nathan A. Johnson¹, and James B. Moring¹

Abstract

In 2012–13, the U.S. Geological Survey (USGS), in cooperation with the U.S. Fish and Wildlife Service (USFWS), completed the first phase of a two-phase study of mussel host-fish relations for five endemic mussel species in central and southeastern Texas that were State-listed as threatened on January 17, 2010: (1) Texas fatmucket (*Lampsilis bracteata*), (2) golden orb (*Quadrula aurea*), (3) smooth pimpleback (*Quadrula houstonensis*), (4) Texas pimpleback (*Quadrula petrina*), and (5) Texas fawnsfoot (*Truncilla macrodon*). On October 6, 2011, the USFWS announced the completion of a status review and determined that the five mussel species warranted listing under the Endangered Species Act; however, listing of these species at that time was precluded by higher priority listing actions, and currently (December 2014), they remained unlisted.

Freshwater mussels are long-lived, sedentary organisms that spend their larval stage as obligate parasites on the gills or fins of fishes, and many of these larvae, which are referred to as "glochidia," can survive only on a narrow range of host-fish species. Results from both study phases are likely to provide information useful for propagation of rare mussels, reintroduction of host fish, population and reproduction monitoring, habitat restoration and enhancement, and adaptive management.

The abundance of host fish, frequency of parasitism in fish, and frequency of juvenile mussels or glochidia recovered from hatchery-held fish was assessed by collecting fish and mussels at 14 sites distributed among seven streams in central and southeastern Texas (juvenile mussels and glochidia were not differentiated in hatchery-held fish). All fish collected and

assessed in this study were wild-caught. Qualitative surveys of the resident mussel communities were made, focusing on the five candidate species. A subsample (3 percent in 2012 and 19 percent in 2013) of the fish collected during aquatic biota surveys was submitted to the USFWS San Marcos National Fish Hatchery and Technology Center to collect juvenile mussels and glochidia recovered from the host fish, which were held for 28 days in holding tanks to allow time for most of the attached glochidia to release from the gills of the fish after transforming into juvenile mussels. All fish not sent to the hatchery were assessed for glochidia in the field or in the USGS Texas Water Science Center laboratory in Austin, Tex. Juvenile mussels and glochidia that were recovered from fish at the hatchery were submitted for use in the second phase of this study, the development of deoxyribonucleic acid (DNA) identification keys to determine mussel and host-fish relationships through DNA-based molecular identification (DNA typing of the juvenile mussels and glochidia). Reporting on the results of DNA-based molecular identification research is beyond the scope of this report.

In 2012, the majority of the fish that were collected, in terms of total number and species types, belonged to the sunfish family Centrarchidae (centrarchids; 1,277 individuals and at least 10 species). Redbreast sunfish (*Lepomis auritus*) was the most common species collected in 2012 (603 individuals), but the largemouth bass (*Micropterus salmoides*) species was caught at all 10 sites. The largest number of species (19) was collected at the San Saba Menard site (San Saba River near Menard, Tex.) on May 22, 2012.

In 2013, most of the fish that were collected, in terms of total number and species types, were centrarchids (763 individuals) and cyprinids (10 species), respectively. Blacktail shiner (*Cyprinella venusta*) was the most common species collected in 2013 (287 individuals), but bluegill (*Lepomis macrochirus*) was the only species that was caught at all nine sites. The largest number of individuals (382) and

¹U.S. Geological Survey.

²U.S. Fish and Wildlife Service.

species (19) was collected from the Colorado Columbus site (Colorado River near Columbus, Tex.) on June 11, 2013.

A minimum of two fish (any species) parasitized with glochidia was collected from each of the 10 sites sampled during 2012. The highest percentage of parasitized fish (19.1 percent) was measured at the Guadalupe Victoria site (Guadalupe River near Victoria, Tex.). The catfish family Ictaluridae (ictalurids) exhibited the highest proportion of parasitized fish (12.1 percent). Of the nine sites sampled in 2013, the Pedernales Fredericksburg site (Pedernales River near Fredericksburg, Tex.) had the highest proportion of parasitized fish at 22.7 percent. Ictalurids again exhibited the highest frequency of parasitism (26.5 percent).

Of the fish that were not sent to the hatchery but assessed for glochidia in the field or in the laboratory in 2012, at least 13 species were parasitized, and longear sunfish (*Lepomis megalotis*) was the species with the highest percentage of parasitized individuals (17.3 percent). Of the fish that were not sent to the hatchery but assessed for glochidia in the field or in the laboratory in 2013, only eight species were parasitized, and flathead catfish (*Pylodictis olivaris*) was the species with the highest percentage of parasitized individuals (42.9 percent).

With the exception of the San Antonio Charco site, fish were submitted to the hatchery from all sampling sites in 2013. During the first sampling period in 2013 (April 1–5), slightly more than half (16 out of 29) of the fish species (on a per site basis) that were submitted to the hatchery released juvenile mussels and glochidia. Compared to the other sampling periods in 2013, substantially fewer glochidia per fish were present on fish submitted to the hatchery during the second sampling period in 2013 (April 29–May 2). Although only two sites were sampled during the third sampling period in 2013 (June 10-11), more juvenile mussels and glochidia were recovered at the hatchery during this sampling period (107) than were recovered during the first two sampling periods in 2013 combined (102). An average of 17 juvenile mussels or glochidia was recovered per largemouth bass submitted to the hatchery from the Guadalupe Victoria site during the third sampling period.

A total of 19 fish species collected at nine sites was submitted to the hatchery in 2013, and 14 of these species had juvenile mussels or glochidia that were recovered at the hatchery. The three most productive species, in terms of the average number of juvenile mussels or glochidia recovered, were longear sunfish, spotted bass, and largemouth bass, each of which averaged more than two juvenile mussels or glochidia recovered per individual.

Introduction

Mussels play an important role in aquatic environments by modifying aquatic habitat to make it more suitable for themselves and other organisms. Mussels capture organic matter from the water column when they siphon, using it to build body and shell, while excreting nutrients that are immediately available to plant life. Mussels deposit any remaining organic material to the sediment, making it available for other invertebrates and fish to consume. During feeding, mussels filter the water they live in by removing phytoplankton as well as the fungi and bacteria attached to the organic particles they have removed from the water column. The mussel shells also act as a vital substrate on which algae and insect larvae attach. Because mussels anchor themselves to stream or lake bottoms, they may actually stabilize the lake or stream bottom, thereby minimizing scouring effects associated with floods and wave action. Mussels are also an important food source for several different kinds of terrestrial and aquatic animals, including muskrats and raccoons, as well as several species of fish (Minnesota Department of Natural Resources, 2014).

About 51 species of freshwater mussels live in Texas (Howells and others, 1996). In 2007 and 2008, the U.S. Fish and Wildlife Service (USFWS) was petitioned by the conservation group WildEarth Guardians (WildEarth Guardians, 2014) to list 12 freshwater mussel species that live in Texas and neighboring States as threatened or endangered with a need for species protection under the Endangered Species Act. Of the 12 mussel species endemic to central and southeastern Texas, 5 were selected as targeted mussel species for the determination of host-fish relationships documented in this report: (1) Texas fatmucket (Lampsilis bracteata), (2) golden orb (*Quadrula aurea*), (3) smooth pimpleback (Quadrula houstonensis), (4) Texas pimpleback (Quadrula petrina), and (5) Texas fawnsfoot (Truncilla macrodon). The five mussel species were State-listed as threatened on January 17, 2010 (Texas Parks and Wildlife Department, 2010), and on October 6, 2011, the USFWS announced the completion of a status review and determined that the mussel species warranted listing under the Endangered Species Act (U.S. Fish and Wildlife Service, 2011c). Their listing at that time, however, was precluded by higher priority listing actions and currently (December 2014), they remained unlisted. Lack of adequate research on Texas freshwater mussels has resulted in little to no information on their biological and ecological needs, including habitat requirements and host-fish use. In some instances, the reduction in size of mussel populations has been caused by shifting fish community dynamics instead of the hydrological environment of the mussels themselves (Haag and Warren, 1998).

In 2012–13, the U.S. Geological Survey (USGS), in cooperation with the USFWS, completed the first phase of a two-phase study of mussel host-fish relations for five endemic mussel species in central and southeastern Texas (fig. 1) that were State-listed as threatened on January 17, 2010. These freshwater mussel species are under a Federal Candidate Notice of Review by the USFWS (U.S. Fish and Wildlife Service, 2013) and were targeted for investigation in this study. The abundance of host fish, frequency of parasitism in fish, and frequency of juvenile mussels or glochidia recovered

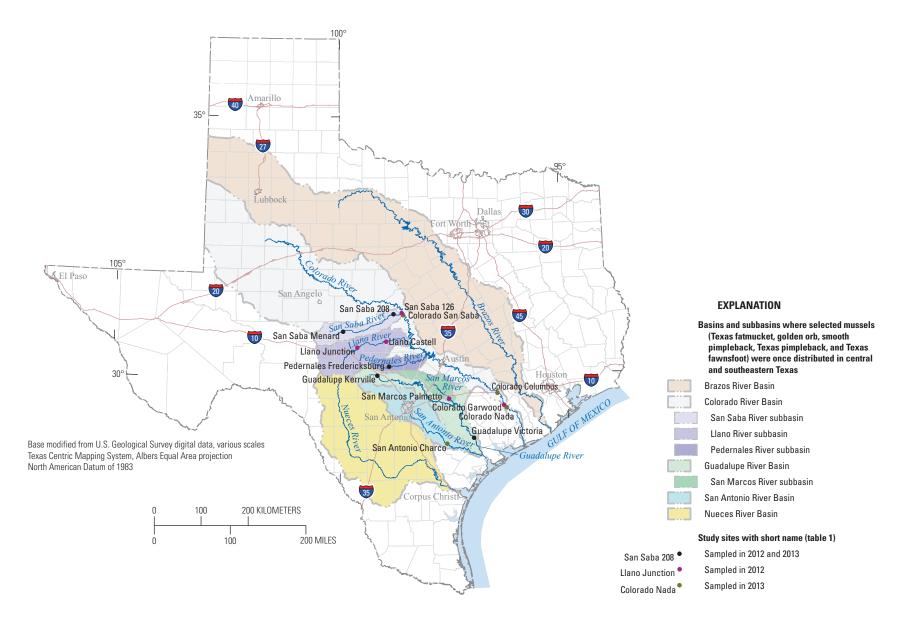


Figure 1. Location of sites sampled for fish and mussels in selected streams as part of a mussel host-fish study, central and southeastern Texas, 2012–13.

from hatchery-held fish was assessed by collecting fish and mussels at 14 sites distributed among seven streams in central and southeastern Texas. All fish collected and assessed in this study were wild-caught. In this report, the short names listed in table 1 are used for referring to sampling sites and are provided on all of the map-based figures.

Methods of investigation for the first phase of this twophase study are outlined in this report and include detailed descriptions of site selection and reconnaissance, mussel surveys, fish collection and field processing, glochidia recovery in the USGS Texas Water Science Center laboratory in Austin, Tex. (hereinafter referred to as "the laboratory"), along with juvenile mussel and glochidia recovery at the USFWS San Marcos National Fish Hatchery and Technology Center in San Marcos, Tex. (hereinafter referred to as "the hatchery"). Results from the first phase of the study are also described in this report and include abundance of host fish, frequency of parasitism in fish assessed for glochidia in the field and laboratory, and frequency of juvenile mussels or glochidia on hatchery-held fish. Juvenile mussels and glochidia that were recovered from fish at the hatchery. collected during the first phase of the study, were submitted for use in the second phase of this study for development of deoxyribonucleic acid (DNA) identification keys to determine mussel and host-fish relationships through DNA-based molecular identification (DNA typing of the juvenile mussels and glochidia) for the five mussel species of concern. Results from both study phases are likely to provide information useful for future recovery efforts that might include propagation of rare mussels, reintroduction of host fish, population and reproduction monitoring, habitat restoration and enhancement, and adaptive management.

Background Information

Native freshwater mussels in the family Unionidae (unionids) have appreciably decreased in numbers and distribution throughout the United States (Randklev and others, 2009). It has been estimated that of the 297 species documented in the United States and Canada, 12 percent are thought to be extinct, 23 percent are considered threatened or endangered (Williams and others, 1993), and approximately 43 percent are in need of conservation (Vaughn and Taylor, 1999). Freshwater mussels possess a suite of biological characteristics that render them susceptible to reductions in habitat range, extirpation (local extinction), and extinction (Vaughn and Taylor, 1999). Unionids are long-lived, sedentary organisms that spend their larval stage as obligate parasites on the gills or fins of fishes, and many of these larvae (fig. 2), which are referred to as "glochidia," can survive only on a few host-fish species (Vaughn and Taylor, 1999; Randklev and others, 2009). As a result, extirpation and extinction of

host-fish species, overharvesting of mussels, urban sprawl, stream impoundments, agriculture practices, introduction of alien species, and varied land-management policies have reduced or eliminated many unionid populations (Neck, 1982; Bogan, 1993; Strayer, 1999; Vaughn and Taylor, 1999; Watters, 1999; Lydeard and others, 2004). A major factor in the demise of freshwater mussels has been the large-scale impoundment of streams during the past 75 years. Impoundments not only affect the free-flowing state of the stream but also affect the ability of fish to migrate longitudinally, which can affect mussel distribution and population structure (Vaughn and Taylor, 1999).

Knowledge of host fish is essential for understanding both mussel abundance and patterns of mussel distribution; this knowledge is also essential for the development of effective conservation programs (Haag and Warren, 1998). Poor recruitment in some mussel populations has been attributed to the lack of proper fish hosts caused by humaninduced changes in the fish assemblage (Haag and Warren, 1998). If populations of the primary host fish for mussels decline, it is likely that mussel populations that depend on these hosts will also decline (Ziuganov and others, 2001). An increase in nonnative fish or changes in the presence or abundance of native host fish can cause changes in fish community structure and composition, which can alter mussel and host-fish relationships. Conservation plans for native freshwater mussels should include detailed information about the host fish that are critical for completion of the life cycle of the mussel (Martel and Lauzon-Guay, 2005).

In the second phase of the study, and only mentioned in this report to provide additional context for the results described in this report, the collection of tissue samples to develop a DNA library from adult mussels began in 2009 and continues to the present (2014). This library has many applications and can be used to determine host-fish use in the wild for the five candidate mussel species by capturing naturally parasitized fish and identifying juvenile mussels or glochidia by using DNA-based molecular identification data. Juvenile mussels and glochidia were submitted for DNA typing that were collected from 1 of 2 sources, either hatcheryheld fish or fish that were assessed for glochidia in the field or the laboratory (only glochidia that were still attached to fish were collected in the field or laboratory). Previous studies in different parts of the United States have used a DNA-typing approach to identify relations between mussels and their host fish in the Pacific Northwest (Gustafson and Iwamoto, 2005), Maine (Kneeland and Rhymer, 2008), and more recently, the St. Croix River in Minnesota (Boyer and others, 2011). Plans are that all DNA data obtained from this assessment of five threatened mussels and their host fish in Texas will be submitted to public DNA databanks, and all specimens will be vouchered in public museums to ensure open access to data.

		Short name				Water	Date(s) sampled
USGS station number	USGS station name	(unique site identifier assigned to each USGS station name) (figs. 1, 3, 4–10, 12, and 15)	Reach midpoint latitude (decimal degrees)	Reach midpoint longitude (decimal degrees)	Approxi- mate reach length (meters)	temperature collected with a continuous temperature probe?	2012	2013
292652096225300	Colorado River below Farm Road 950 near Garwood, Tex.	Colorado Gar- wood	29.4478	96.3813	1,600	no	May 31	
311221098340800	Colorado River below Highway 190 near San Saba, Tex.	Colorado San Saba	31.2057	98.5689	200	yes		April 29
292341096192900	Colorado River near County Road 475 near Nada, Tex.	Colorado Nada	29.3946	96.3247	1,200	no		May 2
294040096310200	Colorado River near Kleimann Road near Columbus, Tex.	Colorado Columbus	29.6779	96.5173	610	no		June 11
284952097033200	Guadalupe River at Highway 77 near Victoria, Tex.	Guadalupe Vic- toria	28.8311	97.059	200	no	May 30	April 4 and June 10
300300099093300	Guadalupe River near Concho Drive at Kerrville, Tex.	Guadalupe Ker- rville	30.05	99.1592	350	yes	May 29	April 2 and April 30
304212098573100	Llano River at Farm Road 2768 at Castell, Tex.	Llano Castell	30.7032	98.9586	400	no	August 22	
303520099355100	Llano River at County Road 385 near Junction, Tex.	Llano Junction	30.589	99.5976	460	no	August 22	
301316098540200	Pedernales River at Boos Lane near Fredericksburg, Tex. ¹	Pedernales Fred- ericksburg	² 30.2203, ³ 30.2223	² 98.9003, ³ 98.9033	² 200; ³ 530	no	August 23	May 1
08188200	San Antonio River near Charco, Tex.	San Antonio Charco	28.7339	97.6431	210	no		April 3
293519097350700	San Marcos River at Palmetto State Park, Tex.	San Marcos Pal- metto	29.5886	97.5852	330	no	August 24	
311418098360700	San Saba River at County Road 126 near San Saba, Tex.	San Saba 126	31.2384	98.602	230	no	May 21	
311323098470500	San Saba River at County Road 208 near San Saba, Tex.	San Saba 208	31.2231	98.7846	200	yes	August 21	April 1
305358099543800	San Saba River below Beyer Crossing near Menard, Tex.	San Saba Menard	30.8995	99.9107	1,250	yes	May 22	April 5

¹Study reach was downstream from Boos Lane bridge in 2012 and upstream from Boos Lane bridge in 2013.

²In 2012.

³In 2013.

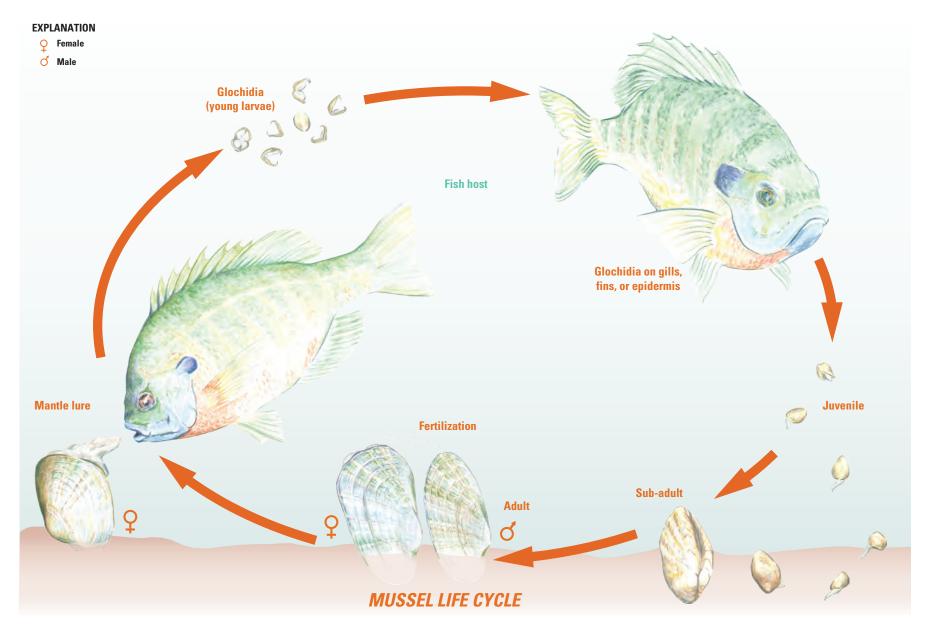


Figure 2. Life cycle of unionid mussels native to Texas.

Historical Range and Recent (2011) Occurrence of Target Mussel Species

Texas fatmucket, golden orb, smooth pimpleback, Texas pimpleback, and Texas fawnsfoot were once widely distributed throughout the major river basins of central and southeastern Texas (U.S. Fish and Wildlife Service, 2011b). As of the status review in 2011, each of the species was found in low densities at reduced locations. Texas fatmucket was found historically in large sections of the Colorado, Guadalupe, and San Antonio River Basins, but its known range has been reduced to include only parts of eight counties and no longer includes the San Antonio River or its tributaries (fig. 3). Golden orb was found historically in large sections of the San Antonio and Guadalupe River Basins and in a small section of the Nueces River Basin, but its known range has been reduced to include only parts of seven counties (fig. 4). Smooth pimpleback was found historically in large parts of the Brazos and Colorado River Basins, but its known range has been reduced to include only parts of 16 counties located almost exclusively in the lower halves of each basin (fig. 5). Texas pimpleback was found historically in most of the Guadalupe River Basin in addition to large parts of the Colorado and San Antonio River Basins, but its known range has been reduced to include only parts of five counties (fig. 6). Texas fawnsfoot was found historically in most of the Brazos and Colorado River Basins, but its known range has been reduced to 12 counties on the upper and lower Brazos River and middle and lower Colorado River (fig. 7).

Purpose and Scope

The purpose of this report is to describe the abundance of host fish, frequency of parasitism in fish assessed for glochidia in the field and in laboratory settings, and frequency of juvenile mussels or glochidia on hatchery-held fish at 14 stream sites in central and southeastern Texas during 2012–13. Qualitative surveys of the resident mussel communities were made, focusing on the five candidate species for listing under the Endangered Species Act (Texas fatmucket, golden orb, smooth pimpleback, Texas pimpleback, and Texas fawnsfoot). Reporting on the results of DNA-based molecular identification of juvenile mussels and glochidia is beyond the scope of this report.

Methods of Investigation

The Colorado, Guadalupe, and San Antonio River Basins were selected for fish and mussel sampling on the basis of historical ranges and known counties of recent (2011) occurrence for each of the target mussel species (U.S. Fish and Wildlife Service, 2011a; figs. 3–7). Sampling locations on streams in each river basin were chosen on the basis of the number of candidate mussels likely present at each site,

accessibility to the sites, and satellite imagery. Satellite imagery provides evidence of desirable site characteristics, such as overhanging vegetation (which provides fish cover) and the presence of multiple mesohabitat types, and helps to confirm site accessibility.

A subsample of fish collected during aquatic biota surveys was submitted to the hatchery to collect juvenile mussels from host fish, which were held in holding tanks to allow time for most of the attached glochidia to mature and release from the gills of the fish. The fish that were not sent to the hatchery were assessed for glochidia in the field or in the USGS laboratory in Austin, Tex., and encysted glochidia were extracted in the laboratory, if present. Juvenile mussels and glochidia recovered at the hatchery were submitted to Nathan Johnson (research biologist with the USGS Southeast Ecological Science Center in Gainesville, Florida) for use in the second phase of this study, the development of DNA identification keys to determine mussel and host-fish relationships through DNA typing of the juvenile mussels and glochidia.

Site Selection and Reconnaissance

Preliminary sites were selected on the basis of recommendations from experts in the field of freshwater mussels in Texas (Lyubov Burlakova, State University of New York at Buffalo, oral commun., 2012; Robert G. Howells, BioStudies, oral commun., 2012; Alexander Karatayev, State University of New York at Buffalo, oral commun., 2012; and Charles Randkley, Texas A&M Institute of Renewable Natural Resources, oral commun., 2012–13), in addition to professional judgment of the authors. After the preliminary sites were selected, field reconnaissance of the sites was done to reduce the number of final sampling sites. Attempts were made to find representative sites from the upper and lower parts of the Colorado, Guadalupe, and San Antonio River Basins; however, this was not attainable for the upper part of the San Antonio River, so only the lower part of the San Antonio River was sampled.

Established populations of the five candidate mussel species are distributed among the Brazos (smooth pimpleback and Texas fawnsfoot), Colorado (Texas fatmucket, smooth pimpleback, Texas pimpleback, and Texas fawnsfoot), Guadalupe (Texas fatmucket, golden orb, and Texas pimpleback), San Antonio (Texas fatmucket, golden orb, and Texas pimpleback), and Nueces (golden orb) River Basins (Howells and others, 1996). All five candidate species were considered initially because of the uncertainty associated with historical records and recent (2011) records on locations where these species have been found. Historical records on occurrence and follow-up site reconnaissance were used to locate candidate mussel populations. Mussel populations were considered acceptable for study if indicators of successful recruitment were present. Indicators of successful recruitment include a broad range in size classes (and presumably ages) (Darr and others, 2013) and presence of subadult mussels (fig. 2).

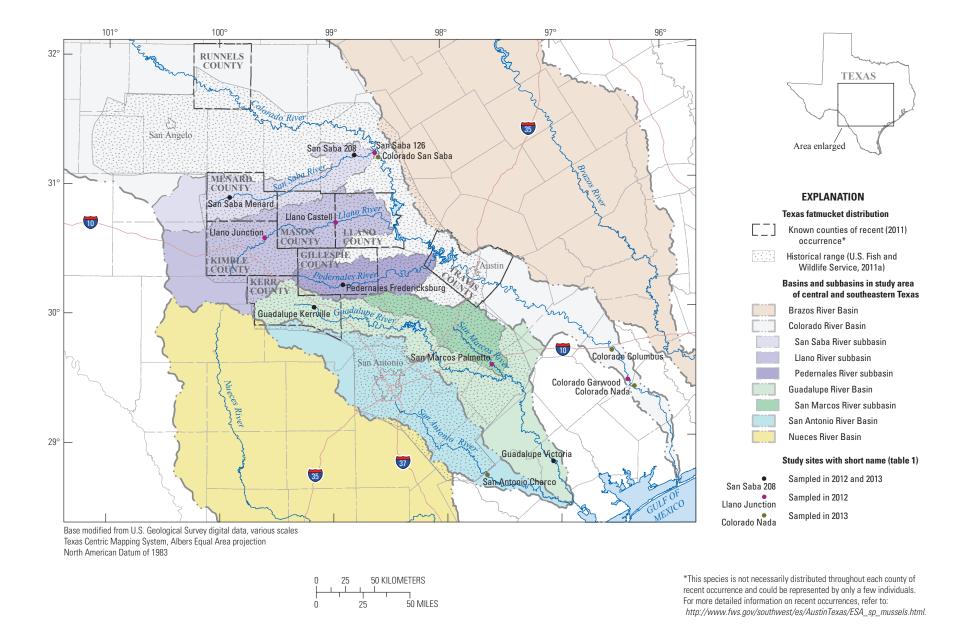
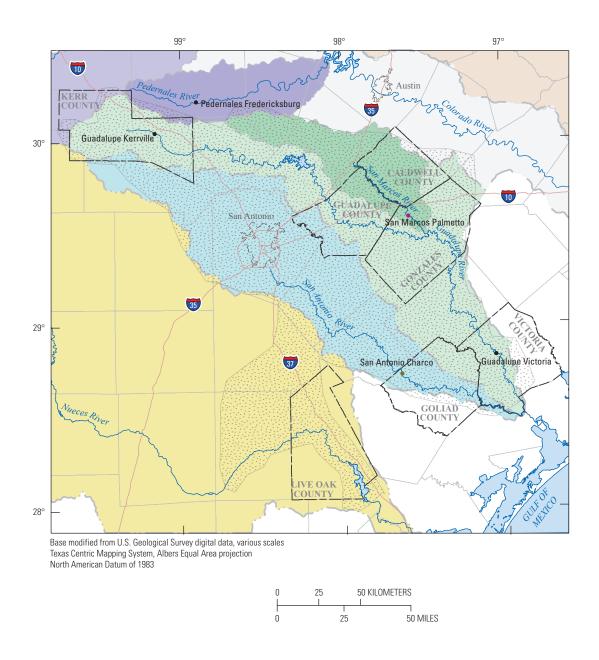


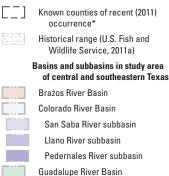
Figure 3. Historical range for the Texas fatmucket (Lampsilis bracteata) and counties of known recent (2011) occurrence as part of a mussel host-fish study, central and southeastern Texas.





EXPLANATION





Study sites with short name (table 1)

San Marcos River subbasin San Antonio River Basin Nueces River Basin

Guadalupe Victoria
San Marcos Palmetto
San Antonio Charco

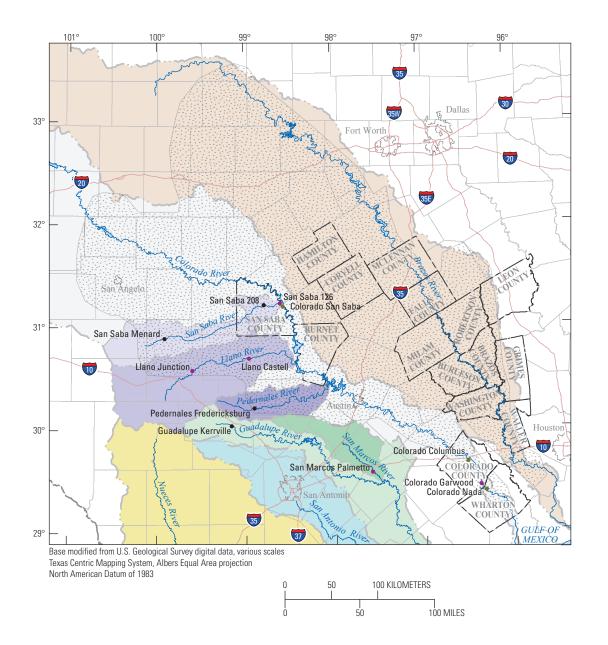
Sampled in 2012 and 2013

Sampled in 2012

Sampled in 2013

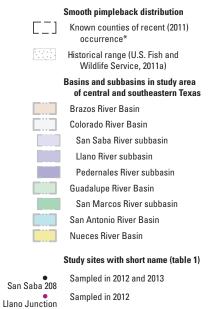
Figure 4. Historical range for the golden orb (*Quadrula aurea*) and counties of known recent (2011) occurrence as part of a mussel host-fish study, central and southeastern Texas.

^{*}This species is not necessarily distributed throughout each county of recent occurrence and could be represented by only a few individuals. For more detailed information on recent occurrences, refer to: http://www.fws.gov/southwest/es/AustinTexas/ESA_sp_mussels.html.





EXPLANATION



Sampled in 2013

Colorado Nada

Figure 5. Historical range for the smooth pimpleback (*Quadrula houstonensis*) and counties of known recent (2011) occurrence as part of a mussel host-fish study, central and southeastern Texas.

^{*}This species is not necessarily distributed throughout each county of recent occurrence and could be represented by only a few individuals. For more detailed information on recent occurrences, refer to: http://www.fws.gov/southwest/es/AustinTexas/ESA_sp_mussels.html.

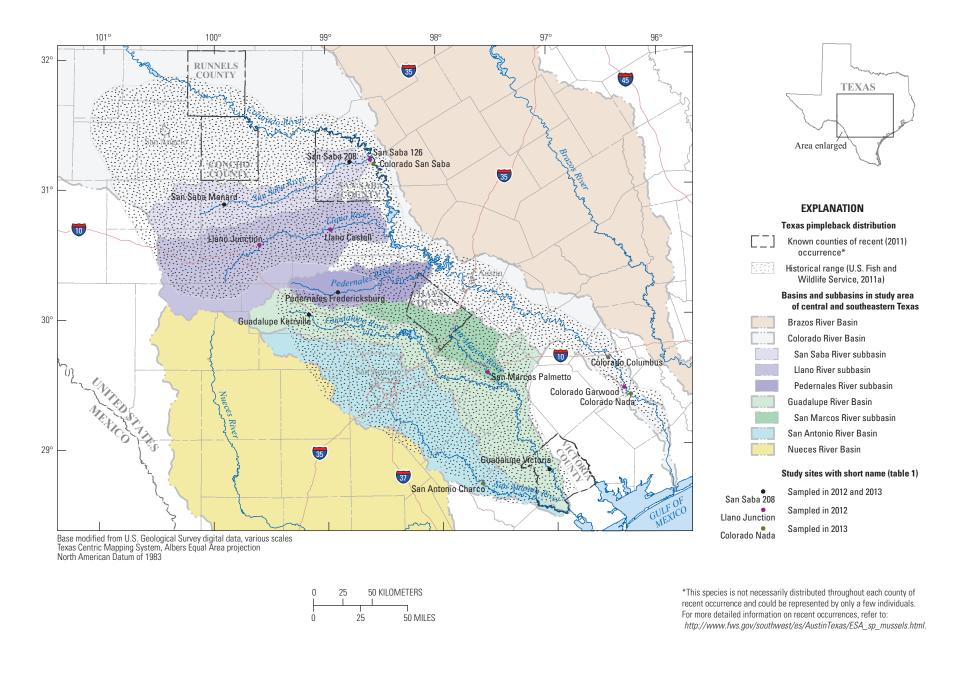
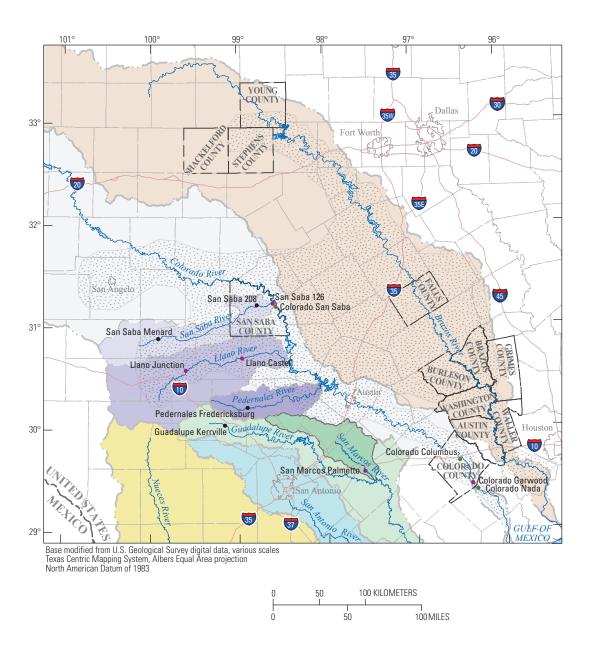


Figure 6. Historical range for the Texas pimpleback (*Quadrula petrina*) and counties of known recent (2011) occurrence as part of a mussel host-fish study, central and southeastern Texas.





EXPLANATION

Texas fawnsfoot distribution

Known counties of recent (2011) occurrence*

Historical range (U.S. Fish and Wildlife Service, 2011a)

Basins and subbasins in study area of central and southeastern Texas

Brazos River Basin
Colorado River Basin

San Saba River subbasin
Llano River subbasin

Pedernales River subbasin

Guadalupe River Basin

San Marcos River subbasin
San Antonio River Basin

Nueces River Basin

Study sites with short name (table 1)

San Saba 208 Llano Junction Sampled in 2012 and 2013

Sampled in 2012

Colorado Nada Sampled in 2013

Figure 7. Historical range for the Texas fawnsfoot (*Truncilla macrodon*) and counties of known recent (2011) occurrence as part of a mussel host-fish study, central and southeastern Texas.

^{*}This species is not necessarily distributed throughout each county of recent occurrence and could be represented by only a few individuals. For more detailed information on recent occurrences, refer to: http://www.fws.gov/southwest/es/AustinTexas/ESA_sp_mussels.html.

Ten sites on six different streams were sampled in 2012 (table 1); five sites were resampled in 2013 because they offered a combination of living target mussel species and large proportions of fish parasitized with glochidia. In five cases, sites that had been selected for sampling in 2012 had to be replaced with alternate sites in 2013 for a variety of reasons. U.S. Geological Survey station names are listed for all sites sampled in 2012 and 2013 in table 1.

Sites on the lower Colorado River (Colorado Garwood, Colorado Nada, and Colorado Columbus; fig. 1, table 1) were each sampled only once. The Colorado Garwood site was replaced by the Colorado Nada site in hopes of collecting fish that were parasitized with Texas fawnsfoot glochidia because of the presence of abundant Texas fawnsfoot shell material at the Colorado Nada site. The Colorado Nada site was then replaced by the Colorado Columbus site in an effort to collect a larger proportion of fish that were parasitized with Texas fawnsfoot glochidia. Sites on the Llano River (Llano Castell and Llano Junction; fig. 1, table 1) and the San Marcos River (San Marcos Palmetto; fig. 1, table 1) were not revisited in 2013. Relatively low proportions of parasitized fish were found at the sites on the Llano River, and none of the target mussel species were found at the Llano Castell site. Only one target mussel species (Texas fatmucket) was found at the site on the San Marcos River, a species that was well represented by the sites on the Guadalupe River.

One of the San Saba River sites (San Saba 126; fig. 1, table 1) had to be discontinued after it was sampled only once because mussels from another site on the San Saba River near County Road 340 were translocated to the San Saba 126 site and scheduled for long-term monitoring by Texas A&M University. The Texas Department of Transportation, Texas Parks and Wildlife, and Texas A&M University were involved in the translocation effort. The San Saba 126 site was replaced with a second site on the San Saba River (San Saba 208; fig. 1, table 1), which was located approximately 24 miles upstream from the San Saba 126 site to minimize disturbance to translocated mussels.

In 2013, a site was added on the San Antonio River (San Antonio Charco site; fig. 1, table 1) to provide coverage of an additional targeted river basin. A site was also added on the Colorado River (Colorado San Saba site; fig. 1, table 1) to provide additional coverage of the upper part of the Colorado River Basin.

As part of the reconnaissance process, continuous monitoring temperature probes were installed at four study sites (table 1) to measure and store water temperature at 2-hour intervals. Water temperature data were collected on the basis that there might be a relation between water temperature and the number of parasitized fish collected (no relation was evident). The water temperature data were published in the USGS National Water Information System (U.S. Geological Survey, 2014) but are not further discussed in this report.

Mussel Surveys

At each sampling site, a search of all habitat types within the prescribed reach length (table 1) was done. Reach lengths varied from site to site on the basis of the need to sample the maximum number of mesohabitat (visually distinct units of habitat within a stream; [Pardo and Armitage, 1997] with unique depths, velocities, slopes, substrates, and cover) types. Sampling followed the qualitative sampling methods outlined in Strayer and Smith (2003), specifically, random timed searches utilizing snorkeling, wading, and hand grubbing to approximately 15 centimeters (cm) deep; the utilization of viewing buckets where clear water conditions were present; or both. Search times varied at each site but generally were standardized to approximately 3 person-hours per site. Shorelines were searched for shell material to identify additional species that were not found along the prescribed reach length for a given search. Shell material was classified as fresh dead (periostracum present, nacre pearly, and soft tissue present); recent dead (periostracum mostly present, little fading of nacre, some algae stains, no tissue present); long dead (periostracum worn, nacre fading, lots of algae stains); or subfossil (little to no periostracum, white and chalky nacre, algae stains) (Texas Parks and Wildlife Department, 2014). The periostracum is the outermost, external layer of the shell, whereas the nacre is the pearly interior of the shell (Great Plains Nature Center, 2014). A species was considered extant at a site if it was represented by live or recently dead shell material (Szafoni, 2001). All mussels were identified by using standard taxonomic references (Howells and others, 1996; Neil Ford, University of Texas at Tyler, written commun., 2010; Howells, 2010; Charles Randklev, Texas A&M Institute of Renewable Natural Resources, written commun., 2010), along with the USFWS reference collection housed at the Texas Coastal Ecological Services Field Office in Houston, Tex. After identification, mussels were returned to the stream as close as possible to where they were collected. Presence and absence data for different mussel species are shown in table 2.

Fish Collection and Field Processing

A fish assemblage survey was completed at each site during the spawning periods of targeted mussels, which varied by species. Timing of fish sampling was based on the best available knowledge and information gained during the initial mussel community surveys. All mesohabitats were sampled in a given reach, and efforts were made to sample for fish in areas where the targeted mussel species had been observed.

Sampling methods were dependent on site conditions, fish assemblages, and abundance of target fish species. The same reaches that were sampled for mussels were also sampled for fish. To ensure that fish diversity and size-class

 Table 2.
 Mussel survey results, including condition as part of a mussel host-fish study, central and southeastern Texas, 2012–13.

[USGS, U.S. Geological Survey; --, not collected; L, live; RD¹, recent dead; LD², long dead; SF³, subfossil; FD⁴, fresh dead]

			Mussel sur	vey results, includi	ng condition	
	-		Mu	ssel species of inte	erest	
USGS station name	Short name	Texas fatmucket (<i>Lamsilis</i> <i>bracteata</i>)	Golden orb (<i>Quadrula</i> <i>aurea</i>)	Smooth pimpleback (<i>Quadrula</i> houstonensis)	Texas pimpleback (<i>Quadrula</i> <i>petrina</i>)	Texas fawnsfoot (<i>Truncilla</i> <i>macrodon</i>)
Colorado River below Farm Road 950 near Garwood, Tex.	Colorado Garwood			L/RD-LD	L/RD-LD	L/RD-SF
Colorado River below Highway 190 near San Saba, Tex.	Colorado San Saba			L/RD-LD	L/RD-LD	RD-SF
Colorado River near County Road 475 near Nada, Tex.	Colorado Nada			L/RD	L/RD-LD	RD-SF
Colorado River near Kleimann Road near Columbus, Tex.	Colorado Columbus			RD-LD	RD-SF	LD-SF
Guadalupe River at Highway 77 near Victoria, Tex.	Guadalupe Victoria		L/RD-LD		L/RD-LD	
Guadalupe River near Concho Drive at Kerrville, Tex. ⁵	Guadalupe Kerrville	L/RD-LD	L/RD		L/RD-LD	
Llano River at Farm Road 2768 at Castell, Tex.	Llano Castell					
Llano River at County Road 385 near Junction, Tex.	Llano Junction	FD-RD				
Pedernales River at Boos Lane near Fredericksburg, Tex. ⁶	Pedernales Fredericksburg	FD-LD				
San Antonio River near Charco, Tex.	San Antonio Charco		L/RD-LD			
San Marcos River at Palmetto State Park, Tex.	San Marcos Palmetto		L/RD-D			
San Saba River at County Road 126 near San Saba, Tex.	San Saba 126			L/RD-LD	L/RD-LD	
San Saba River at County Road 208 near San Saba, Tex.	San Saba 208			L/RD-LD	L/RD-LD	
San Saba River below Beyer Crossing near Menard, Tex.	San Saba Menard	L/RD-LD			FD-RD	

 Table 2.
 Mussel survey results, including condition as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

[USGS, U.S. Geological Survey; --, not collected; L, live; RD1, recent dead; LD2, long dead; SF3, subfossil; FD4, fresh dead]

						Mus	sel survey r	esults, incl	uding condi	tion					
							Additional	native muss	sel species						
Short name	Three- ridge (<i>Amblema</i> <i>plicata</i>)	Rock pock- etbook (Arcidens confrago- sus)	Tampico pearly mussel (<i>Cyrtona-</i> ias tampi- coensis)	Southern mapleleaf (<i>Quadrula</i> <i>apiculata</i>)	False spike (Qua- drula mitch- elli)	Pistolgrip (<i>Tritogo-</i> nia verru- cosa)	Louisiana fatmucket (<i>Lampsilis</i> <i>hydiana</i>)	Yellow sandshell (<i>Lamp-</i> silis teres)	Fragile paper- shell (<i>Leptodea</i> fragilis)	Wash- board (<i>Meg-</i> alonaias nervosa)	Bleufer (Pota- milus purpura- tus)	Giant floater (<i>Pygan-</i> odon gran- dis)	Lilliput (Toxo- lasma parvum valis)	Texas lilliput (Toxo- lasma texas- iense)	Paper pondshell (Utter- backia imbecil- lis)
Colorado Gar- wood	L/RD-LD	L/RD	L/RD-LD	L/RD-LD				L/RD-LD	L/RD-LD		L/RD-LD	L/RD	L/RD-LD	L/RD-LD	
Colorado San Saba	L/RD-LD		L/RD-LD	L/RD-LD		L/RD-LD			L/RD-LD		L/RD-LD				
Colorado Nada	L/RD - LD			LD-SF				L/RD-LD	L/RD - LD				SF	SF	
Colorado Colum- bus	RD-LD							RD-LD	L/RD-LD				LD-SF		
Guadalupe Vic- toria	L/RD-LD	L/RD	L/RD-LD	L/RD-LD			FD-RD	L/RD-LD	L/FD-RD	L/RD	L/RD				
Guadalupe Ker- rville			L										L/RD	L/RD	
Llano Castell															
Llano Junction															
Pedernales Fred- ericksburg															
San Antonio Charco	L/RD-LD	L/RD				L/RD-LD		L/RD-LD		RD					
San Marcos Pal- metto	L/RD-LD		RD		SF	LD		L/RD-LD							
San Saba 126	L/RD - LD					L/RD - LD									
San Saba 208	L/RD - LD					L/RD - LD			L/RD-LD						
San Saba Menard	RD		L/RD	L/RD		L									L/RD-LD

¹Periostracum mostly present; little fading of nacre; some algae stains; no tissue present.

²Periostracum worn; nacre fading; lots of algae stains.

³Little to no periostracum; white and chalky nacre; algae stains.

⁴Periostracum present; nacre pearly; soft tissue present.

⁵Mussel survey area also includes small impoundment upstream from the Guadalupe Kerrville site.

⁶Study reach was moved downstream from Boos Lane bridge in 2012 to upstream from Boos Lane bridge in 2013.

variability were surveyed efficiently at each site, available mesohabitat types were sampled by using a combination of boat and barge electrofishing and seining. Most sites were sampled primarily by using a barge that was followed closely by a second barge carrying multiple species-specific holding tanks. Fish were identified by a fish taxonomist as soon as they were netted and separated by species, when possible, into appropriate species-specific, aerated holding tanks to avoid potential glochidia loss or cross-contamination between species. Because of limited space, it was not always possible to keep very large fish separated by species. To ensure retention of glochidia that detached from fish during handling and transport, water from holding tanks was transferred to appropriate species-specific coolers that were used to transfer fish to the hatchery. Sites with wide channels and deep pools that could not be waded effectively were electrofished from a cataraft (raft with two pontoons); this included all of the sites on the Colorado River except the Colorado San Saba site and the San Saba Menard site (fig. 1, table 1). Seining was also done in many cases to supplement electrofishing efforts by using a 1.0- by 3.0-meter (m), 0.17-cm mesh, flat-panel seine. Seining was used when certain target families, particularly Percidae (percids), were not collected by electrofishing. To minimize holding times and stress during the transfer of potential host fish to the hatchery in 2013, fish collection activity was often curtailed after suitable quantities of target fish species had been collected; therefore, seining was infrequently part of the 2013 fish-sampling efforts.

After fish assemblage surveys were completed, the number and species of fish were selected for submittal to the hatchery. Members of different families were selected to provide a diverse selection of potential host fish. Fish species that had a high proportion of glochidia parasitism in 2012 were targeted for submittal in 2013. Tanks were aerated during transport, and fish survival rate was high (greater than 95 percent).

Fish that were not submitted to the hatchery were measured to the nearest millimeter (mm) to determine total length. Fish that could be identified in the field and assessed for glochidia without a microscope (typically fish with total lengths greater than 100 mm) were returned to the stream if no glochidia were present. Forceps were used to check for glochidia on anterior and posterior surfaces of gill filaments on each arch and between gill filaments. If glochidia were observed, then the gill arch was removed and retained. If the fish was heavily infested, either the head or gill arches were preserved. Fish that could not be identified in the field or that could not be checked for glochidia effectively without a microscope were preserved in a 95-percent ethanol solution, labeled by site and date, and returned to the USGS Texas Water Science Center for preliminary identification. Ethanol solution was used in place of formalin because formalin degrades DNA and makes tissues unsuitable for analysis (Nathan A. Johnson, U.S. Geological Survey, written commun., 2014). All small fish (total lengths less than about 100 mm) that were not submitted to the hatchery were retained to be assessed for glochidia at the USGS laboratory in Austin, Tex.

Fish that were identified in the field during sampling were identified to the lowest taxonomic level possible (generally species). All retained fish specimens were preserved in a 70-percent ethanol solution and deposited with the Curator of Ichthyology at the University of Texas Natural Science Center in Austin, Tex., for taxonomic verification and permanent deposition (University of Texas, 2012).

Glochidia Recovery at U.S. Geological Survey Laboratory

Fish that were not submitted to the hatchery and could not be assessed for glochidia without a microscope were retained for evaluation by USGS personnel in a laboratory setting. Each fish was identified to species and measured for total length prior to examining both sets of gills for glochidia by using a microscope. When observed, glochidia were removed with a sterilized needle and placed into a microplate and preserved with 95 percent ethyl alcohol. A microplate is a series of small test tubes (wells) that are arranged on a plastic plate in a regular matrix pattern, most commonly with 96 wells (Medical Museion, 2010). Site name, collection date, fish species, total length, and glochidia extraction location were recorded, and a unique sample identifier was assigned that corresponded to an individual well in the microplate. During the reconnaissance phase of the project (2012), excessive amounts of fish gill tissue were often included with the glochidia in the wells, which contaminated some of the samples prior to DNA typing, rendering them unusable. In 2013, care was taken to reduce the amount of gill material that was attached to glochidia placed in each well.

Juvenile Mussel and Glochidia Recovery at the U.S. Fish and Wildlife Service Hatchery

At the hatchery operated by the USFWS, fish were separated by species and placed into holding tanks. Larger fish were placed in individual holding tanks, whereas smaller fish (such as cyprinids) of the same species were often grouped in a single holding tank. Fish were held at temperatures ranging from 21 to 24 degrees Celsius (°C). Results from a previous passive infestation trial (Johnson and others, 2012) indicated that most transformations from glochidia to juvenile mussels occurred between 17 and 26 days on four different species of fish for the Texas fatmucket, one of the target mussel species in this study, so holding tanks were siphoned daily for the first 28 days of each trial. Shell length, height, and hinge length were measured to the nearest 0.01 mm for juvenile mussels and glochidia (fig. 2), and the

number of juvenile mussels or glochidia recovered per fish was tabulated. Not all glochidia transformed into juvenile mussels, and the hatchery did not differentiate glochidia recovered from host fish from the juvenile mussels that had transformed from glochidia. Only juvenile mussels that have detached from the host fish are viable; the glochidia that have detached from the host fish are not viable and will not develop into juvenile mussels (Hambrook and Eberle, 2000). Each juvenile mussel or glochidium that was recovered was preserved individually in 95 percent ethyl alcohol for DNAbased molecular identification (DNA results are not discussed in this report). Because a small number of the glochidia did not release from the host fish during the 28-day trials, not all glochidia were recovered at the hatchery. In these cases, fish were returned to the USGS to be examined in a laboratory for additional glochidia. Identifying the species of glochidia that were still attached to the fish returned to the USGS laboratory was not completed and would not indicate mussel-to-host-fish relations, because the glochidia will attach to nonhosts but will not complete metamorphosis to a free-living juvenile mussel. Studies have shown that glochidia will attach to nonhosts without complete metamorphosis (Lellis and others, 2013) and, in some cases, will attach to inanimate objects (Haag, 2012). In addition, some glochidia that were observed on fish that were returned by the hatchery for examination in the USGS laboratory may not have had sufficient time to reach maturity over the course of the 28-day trial.

Quality Assurance

Field identification of fish was done with the aid of appropriate taxonomic keys by trained biologists familiar with the types of fish taxa found in central and southeastern Texas. Unidentified fish taxa retained for laboratory identification were provided to the Curator of Ichthyology at the University of Texas Natural Science Center in Austin, Tex., for taxonomic verification and permanent deposition (University of Texas, 2012). Field identification of mussels was done by trained biologists familiar with the freshwater species present in central and southeastern Texas with the aid of field guides for mussel species in Texas or with the help of other expert mussel biologists from Texas Parks and Wildlife and Texas A&M University. All fish and mussel voucher specimens were labeled onsite and will be retained at the USGS laboratory in Austin or the University of Texas Natural Science Center in Austin for a minimum of 5 years.

Abundance of Host Fish

In 2012, fish belonging to 12 families (fig. 8; composite of data from tables 3 and 4) were collected as potential hosts. The majority of the fish that were collected, in terms of total

number and species type, belonged to the sunfish family Centrarchidae (centrarchids) and the carp and minnow family Cyprinidae (cyprinids). A total of 1,277 individuals and at least 10 different species of centrarchids (the number of different species could be higher depending on the identity of the unknown members of the Micropterus genus that were collected) and a total of 623 individuals and at least 9 different species of cyprinids (the number of different species could be higher depending on the identity of the unknown members of the Cyprinidae family that were collected) were collected in 2012 (fig. 8; composite of data from tables 3 and 4). Redbreast sunfish (Lepomis auritus) was the most common species collected in 2012 (603 individuals) (fig. 8; composite of data from tables 3 and 4). Other common species were blacktail shiner (Cyprinella venusta, 353 individuals) and longear sunfish (Lepomis megalotis, 220 individuals). The largest number of individuals (416) was collected at the Llano Castell site on August 22, 2012, whereas the largest number of species (19) was collected at the San Saba Menard site on May 22, 2012 (composite of data from tables 3 and 4). The fewest number of individuals (62) and species (7) was collected at the Guadalupe Kerrville site (fig. 1, table 1) on May 29, 2012 (composite of data from tables 3 and 4). Fewer than 10 individuals of a given species were collected for 20 out of the 38 species (excluding the unknown species that were collected) across all 10 sites sampled for fish during 2012. Largemouth bass (Micropterus salmoides) were caught at all 10 sites, and four other species (bluegill [Lepomis macrochirus], longear sunfish, blacktail shiner, and channel catfish [Ictalurus punctatus]) were caught at 9 of the 10 sites.

In 2013, fish belonging to 11 families (fig. 9; composite of data from tables 5 and 6) were collected as potential hosts. The majority of the fish that were collected in 2013, in terms of total number and species types, were centrarchids and cyprinids, respectively. A total of 686 individuals and 10 species of centrarchids and a total of 763 individuals and 6 species of cyprinids were collected in 2013 (fig. 9; composite of data from tables 5 and 6). Blacktail shiner was the most common species collected in 2013 (287 individuals) (fig. 9; composite of data from tables 5 and 6). Other common species were red shiner (Cyprinella lutrensis, 263 individuals) and longear sunfish (238 individuals). The largest number of individuals (382) and species (19) was collected from the Colorado Columbus site on June 11, 2013. The fewest number of individuals (53) was collected from the San Saba Menard site on April 5, 2013, whereas the fewest number of species (n=5) was collected at the Pedernales Fredericksburg site (fig. 1, table 1) on May 1, 2013 (composite of data from tables 5 and 6). Fewer than 10 individuals of a given species were collected for 13 out of the 34 species that were collected across all 10 sites. Bluegill was the only species that was caught at all 9 sites that were sampled in 2013, whereas largemouth bass, longear sunfish, and bullhead minnow (Pimephales vigilax) were caught at 7 out of 9 sites.

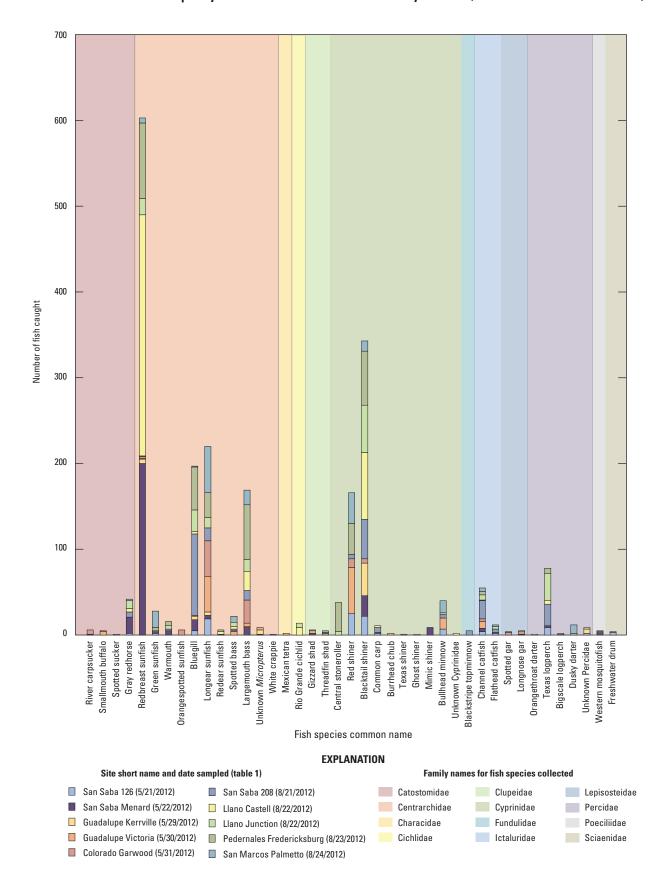


Figure 8. Abundance of potential host fish from sites sampled in 2012 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.

Abundance of Host Fish

Table 3. Number of fish collected and glochidial parasitism by species from sites sampled in 2012 as part of a mussel host-fish study, central and southeastern Texas, 2012–13. [No., number; Avg., average; mm, millimeters; shading indicates fish species are members of the same family; cross-hatching indicates fish species were not collected]

			Short name (fig. 1) with 2012 sampling date in parentheses											
		Species	Sa	an Saba 126	6 (5/21)	Sar	saba Men	ard (5/22)	Gua	dalupe Ker	rville (5/29)			
Family	Genus and species	common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations			
Catostomidae	Carpiodes carpio	River carpsucker				1	30	0						
	Ictiobus bubalus	Smallmouth buffalo												
	Minytrema melanops	Spotted sucker												
	Moxostoma congestum	Gray redhorse	2	438	0	19	415	0						
Centrarchidae	Lepomis auritus	Redbreast sunfish				200	122	5	5	100	1			
	Lepomis cyanellus	Green sunfish	2	43	0	2	153	0						
	Lepomis gulosus	Warmouth				5	91	0						
	Lepomis humilis	Orangespotted sunfish												
	Lepomis macrochirus	Bluegill	5	54	2	13	68	1	4	68	1			
	Lepomis megalotis	Longear sunfish	19	68	9	4	61	0	4	69	0			
	Lepomis microlophus	Redear sunfish				1	145	0						
	Micropterus punctulatus	Spotted bass				/////								
	Micropterus salmoides	Largemouth bass	1	158	0	8	217	0	1	190	0			
	Unknown members of	<i>Micropterus</i> genus	/////			1	35	0	5	37	0			
	Pomoxis annularis	White crappie												
Characidae	Astyanax mexicanus	Mexican tetra												
Cichlidae	Herichthys cyanoguttatum	Rio Grande cichlid												
Clupeidae	Dorosoma cepedianum	Gizzard shad	1	310	0	1	410	0						
	Dorosoma petenense	Threadfin shad				/////								
Cyprinidae	Campostoma anomalum	Central stoneroller												
	Cyprinella lutrensis	Red shiner	25	45	0									
	Cyprinella venusta	Blacktail shiner	22	61	0	24	49	2	38	62	0			
	Cyprinus carpio	Common carp	2	593	0	1	680	0						
	Macrhybopsis marconis	Burrhead chub	/////	///////		/////	///////							

Table 3. Number of fish collected and glochidial parasitism by species from sites sampled in 2012 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

					Short name	(fig. 1) wi	th 2012 sam	pling date in p	arenthes	es	
		Species	S	an Saba 12	6 (5/21)	Sar	ı Saba Men	ard (5/22)	Gua	dalupe Ker	rville (5/29)
Family	Genus and species	common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations
	Notropis amabilis	Texas shiner									
	Notropis buchanani	Ghost shiner									
	Notropis volucellus	Mimic Shiner				9	41	0			
	Pimephales vigilax	Bullhead minnow	7	43	1						
	Unknown members o	of Cyprinidae family	/////								
Fundulidae	Fundulus notatus	Blackstripe topminnow									
Ictaluridae	Ictalurus punctatus	Channel catfish	4	354	0	4	348	0			
	Pylodictis olivaris	Flathead catfish	2	340	1	1	315	0			
Lepisosteidae	Lepisosteus oculatus	Spotted gar		///////							
	Lepisosteus osseus	Longnose gar				1	360	0			
Percidae	Etheostoma spectabile	Orangethroat darter				/////					
	Percina carbonaria	Texas logperch	9	83	1	2	58	0			
	Percina macrolepida	Bigscale logperch	1	40	0						
	Percina sciera	Dusky darter									
	Unknown members	of Percidae family	2	43	0				5	34	0
Poeciliidae	Gambusia affinis	Western mosquitofish				3	32	0			
Sciaenidae	Aplodinotus grunniens	Freshwater drum	3	458	0						
		Totals	107	106	14	300	137	8	62	64	2
	Percentage of fish	infested with glochidia			13.1			2.7			3.2
	Т	Total number of species			16			19			7

Abundance of Host Fish

Table 3. Number of fish collected and glochidial parasitism by species from sites sampled in 2012 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with 2012 sampling date in parentheses											
F 11	Genus and species	Species	Gua	dalupe Vict	oria (5/30)	Col	orado Garw	ood (5/31)	S	an Saba 20	8 (8/21)			
Family	denus and species	common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations			
Catostomidae	Carpiodes carpio	River carpsucker				5	62	0						
	Ictiobus bubalus	Smallmouth buffalo	4	518	0	1	610	0						
	Minytrema melanops	Spotted sucker				1	350	0						
	Moxostoma congestum	Gray redhorse							6	300	0			
Centrarchidae	Lepomis auritus	Redbreast sunfish	1	45	0	2	60	1	1	160	0			
	Lepomis cyanellus	Green sunfish												
	Lepomis gulosus	Warmouth							2	153	0			
	Lepomis humilis	Orangespotted sunfish				6	103	0						
	Lepomis macrochirus	Bluegill				1	50	0	95	89	13			
	Lepomis megalotis	Longear sunfish	41	63	14	42	61	7	15	110	0			
	Lepomis microlophus	Redear sunfish	.////											
	Micropterus punctulatus	Spotted bass	4	150	0	1	160	1	2	153	0			
	Micropterus salmoides	Largemouth bass	4	96	2	27	185	3	11	122	3			
	Unknown members o	f <i>Micropterus</i> genus	3	33	2									
	Pomoxis annularis	White crappie	.////						1	90	0			
Characidae	Astyanax mexicanus	Mexican tetra	2	40	0									
Cichlidae	Herichthys cyanoguttatum	Rio Grande cichlid												
Clupeidae	Dorosoma cepedianum	Gizzard shad	3	77	0									
	Dorosoma petenense	Threadfin shad				2	63	0	1	175	0			
Cyprinidae	Campostoma anomalum	Central stoneroller												
	Cyprinella lutrensis	Red shiner	54	49	2	10	48	0	5	71	0			
	Cyprinella venusta	Blacktail shiner				5	50	0	46	78	0			
	Cyprinus carpio	Common carp							6	500	0			
	Macrhybopsis marconis	Burrhead chub	2	55	0									
	Notropis amabilis	Texas shiner	/////						1	50	0			

Table 3. Number of fish collected and glochidial parasitism by species from sites sampled in 2012 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with 2012 sampling date in parentheses											
		Species	Gua	dalupe Vict	oria (5/30)	Cole	orado Garw	ood (5/31)	Sa	an Saba 20	8 (8/21)			
Family	Genus and species	common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations			
	Notropis buchanani	Ghost shiner												
	Notropis volucellus	Mimic Shiner												
	Pimephales vigilax	Bullhead minnow	13	47	3				4	59	0			
	Unknown members	of Cyprinidae family	1////		/////////				/////					
Fundulidae	Fundulus notatus	Blackstripe topminnow							5	66				
Ictaluridae	Ictalurus punctatus	Channel catfish	8	239	4	3	303	1	21	138	0			
	Pylodictis olivaris	Flathead catfish				/////			4	170	0			
Lepisosteidae	Lepisosteus oculatus	Spotted gar				3	427	0						
	Lepisosteus osseus	Longnose gar				3	537	0						
Percidae	Etheostoma spectabile	Orangethroat darter				/////		////////						
	Percina carbonaria	Texas logperch							25	102	0			
	Percina macrolepida	Bigscale logperch	1	60	0									
	Percina sciera	Dusky darter	./////											
	Unknown members	of Percidae family												
Poeciliidae	Gambusia affinis	Western mosquitofish												
Sciaenidae	Aplodinotus grunniens	Freshwater drum	1	440	0									
		Totals	141	84	27	112	129	13	251	111	16			
	Percentage of fi	sh infested with glochidia			19.1			11.6			6.4			
		Total number of species			14			15			18			

Abundance of Host Fish

Table 3. Number of fish collected and glochidial parasitism by species from sites sampled in 2012 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with 2012 sampling date in parentheses											
F 11	Genus and species	0	L	lano Caste	I (8/22)	LI	ano Junctio	on (8/22)	Pedernales Fredericksburg (8/23)					
Family		Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations			
Catostomidae	Carpiodes carpio	River carpsucker												
	Ictiobus bubalus	Smallmouth buffalo												
	Minytrema melanops	Spotted sucker												
	Moxostoma congestum	Gray redhorse	4	385	0	9	334	2						
Centrarchidae	Lepomis auritus	Redbreast sunfish	281	113	4	19	107	1	88	118	0			
	Lepomis cyanellus	Green sunfish				1	140	0	4	140	0			
	Lepomis gulosus	Warmouth				4	140	0	5	147	0			
	Lepomis humilis	Orangespotted sunfish												
	Lepomis macrochirus	Bluegill	3	98	0	25	96	3	50	100	1			
	Lepomis megalotis	Longear sunfish				12	98	0	23	107	0			
	Lepomis microlophus	Redear sunfish	3	188	0				2	135	0			
	Micropterus punctulatus	Spotted bass	3	192	0	5	210	0						
	Micropterus salmoides	Largemouth bass	22	132	0	14	121	0	58	98	4			
	Unknown members	of <i>Micropterus</i> genus	/////											
	Pomoxis annularis	White crappie												
Characidae	Astyanax mexicanus	Mexican tetra												
Cichlidae	Herichthys cyanoguttatum	Rio Grande cichlid	9	111	0	5	83	0						
Clupeidae	Dorosoma cepedianum	Gizzard shad				1	330	0						
	Dorosoma petenense	Threadfin shad												
Cyprinidae	Campostoma anomalum	Central stoneroller				4	80	0	34	78	0			
	Cyprinella lutrensis	Red shiner							26	70	0			
	Cyprinella venusta	Blacktail shiner	78	70	0	55	77	0	63	80	0			
	Cyprinus carpio	Common carp	2	500	0	/////			/////	//////				
	Macrhybopsis marconis	Burrhead chub	1////	///////										
	Notropis amabilis	Texas shiner												

Table 3. Number of fish collected and glochidial parasitism by species from sites sampled in 2012 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with 2012 sampling date in parentheses										
Family	Comus and anasias	Cuasias assumas usus	L	lano Castel	II (8/22)	LI	lano Junctio	on (8/22)	Pedernales Fredericksburg (8/23)				
	Genus and species	Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations		
	Notropis buchanani	Ghost shiner	1	70	0								
	Notropis volucellus	Mimic Shiner											
	Pimephales vigilax	Bullhead minnow				2	60	0					
	Unknown members	of Cyprinidae family	2	73	0		.//////						
Fundulidae	Fundulus notatus	Blackstripe topminnow											
Ictaluridae	Ictalurus punctatus	Channel catfish	1	115	0	6	230	2	4	95	0		
	Pylodictis olivaris	Flathead catfish				3	135	0					
Lepisosteidae	Lepisosteus oculatus	Spotted gar											
	Lepisosteus osseus	Longnose gar	1	500	0								
Percidae	Etheostoma spectabile	Orangethroat darter	1	40	0								
	Percina carbonaria	Texas logperch	4	68	0	32	88	0	6	83	0		
	Percina macrolepida	Bigscale logperch							./////				
	Percina sciera	Dusky darter											
	Unknown member	s of Percidae family				2	58	0					
Poeciliidae	Gambusia affinis	Western mosquitofish	1	55	0				1	55	0		
Sciaenidae	Aplodinotus grunniens	Freshwater drum											
		Totals	416	111	4	199	112	8	364	98	5		
	Percentage of	fish infested with glochidia			1.0			4.0			1.4		
		Total number of species			16			17			13		

Abundance of Host Fish

Table 3. Number of fish collected and glochidial parasitism by species from sites sampled in 2012 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with 2012 sampling date in page 1									arentheses					
Family		Succion common	San M	arcos Pa	lmetto (8/24)	To	Total - all sites (2012 only)				Totals - by family (2012 only)						
	Genus and species	Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infesta- tions	No. of fish	Avg. total length (mm)	No. of glochidia infesta- tions	Percent- age of fish infested	No. of	Avg. total length (mm)	No. of glochidia infesta- tions	Percent- age of fish infested				
Catostomidae	Carpiodes carpio	River carpsucker		/////		6	57	0	0								
	Ictiobus bubalus	Smallmouth buffalo				5	536	0	0		2.52		2 =				
	Minytrema melanops	Spotted sucker				1	350	0	0	54	352	2	3.7				
	Moxostoma congestum	Gray redhorse	2	253	0	42	372	2	4.8								
Centrarchidae	Lepomis auritus	Redbreast sunfish	1////	/////		597	116	12	2.0								
	Lepomis cyanellus	Green sunfish	19	777	0	28	91	0	0								
	Lepomis gulosus	Warmouth	1///	/////		16	129	0	0								
	Lepomis humilis	Orangespotted sunfish				6	103	0	0								
	Lepomis macrochirus	Bluegill	1	100	0	197	90	21	10.7								
	Lepomis megalotis	Longear sunfish	48	106	6	208	83	36	17.3	1,247	109	87	7.0				
	Lepomis microlophus	Redear sunfish	1////	/////		6	163	0	0								
	Micropterus punctulatus	Spotted bass	7/7/	239	0	22	198	1	4.5								
	Micropterus salmoides	Largemouth bass	11	152	3	157	132	15	9.6								
	Unknown members o	f <i>Micropterus</i> genus	1////		///////	9	35	2	22.2								
	Pomoxis annularis	White crappie				1	90	0	0								
Characidae	Astyanax mexicanus	Mexican tetra				2	40	0	0	2	40	0	0				
Cichlidae	Herichthys cyanoguttatum	Rio Grande cichlid				14	101	0	0	14	101	0	0				
Clupeidae	Dorosoma cepedianum	Gizzard shad				6	214	0	0	1.1	177	0	0				
	Dorosoma petenense	Threadfin shad	2	185	0	5	134	0	0	11	177	0	0				

Table 3. Number of fish collected and glochidial parasitism by species from sites sampled in 2012 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with 2012 sampling date in parentheses										
	Genus and species	Species common name	San Marcos Palmetto (8/24)			To	otal - all s	sites (2012 (only)	Totals - by family (2012 only)			
Family			No. of fish	Avg. total length (mm)	No. of glochidia infesta- tions	No. of fish	Avg. total length (mm)	No. of glochidia infesta- tions	Percent- age of fish infested	No. of fish	Avg. total length (mm)	No. of glochidia infesta- tions	Percent- age of fish infested
Cyprinidae	Campostoma anomalum	Central stoneroller				38	78	0	0				
	Cyprinella lutrensis	Red shiner	36	72	0	156	58	2	1.3				
	Cyprinella venusta	Blacktail shiner	2	70	0	333	71	2	0.6				
	Cyprinus carpio	Common carp	////	/////		11	533	0	0				
	Macrhybopsis marconis	Burrhead chub				2	55	0	0				
	Notropis amabilis	Texas shiner				1	50	0	0	581	75	8	1.4
	Notropis buchanani	Ghost shiner				1	70	0	0				
	Notropis volucellus	Mimic Shiner				9	41	0	0				
	Pimephales vigilax	Bullhead minnow	2	68	0	28	50	4	14.3				
	Unknown members	of Cyprinidae family	////	/////	////////	2	73	0	0				
Fundulidae	Fundulus notatus	Blackstripe topminnow				5	66	0	0	5	66	0	0
Ictaluridae	Ictalurus punctatus	Channel catfish	3	213	0	54	204	7	13.0		207	0	10.1
	Pylodictis olivaris	Flathead catfish	2	260	0	12	217	1	8.3	66	207	8	12.1
Lepisosteidae	Lepisosteus oculatus	Spotted gar	1	510	0	4	448	0	0	•			
	Lepisosteus osseus	Longnose gar	1////	/////		5	494	0	0	9	474	0	0
Percidae	Etheostoma spectabile	Orangethroat darter				1	40	0	0				
	Percina carbonaria	Texas logperch				78	90	1	1.3				
	Percina macrolepida	Bigscale logperch				2	50	0	0	100	83	2	2.0
	Percina sciera	Dusky darter	10	77	1	10	77	1	10.0				
	Unknown members	s of Percidae family	/////		////////	9	41	0	0				
Poeciliidae	Gambusia affinis	Western mosquitofish				5	41	0	0	5	41	0	0
Sciaenidae	Aplodinotus grunniens	Freshwater drum				4	454	0	0	4	454	0	0
		Totals	146	111	10	2,098	110	107	5.1	2,098	110	107	5.1
	Percentage of fi	sh infested with glochidia			6.8								
		Total number of species			14								

Table 4. Juvenile mussels or glochidia recovered from hatchery-held fish collected in 2012 in central and southeastern Texas and submitted to U.S. Fish and Wildlife Service National Fish Hatchery and Technology Center in San Marcos, Tex., as part of a mussel host-fish study, 2012–13.

Short name	Date	Family	Species common name¹	Number of fish submitted to hatchery	Number of juvenile mussels or glochidia recovered from hatchery-held fish
Pedernales Fredericksburg	8/23/2012	Centrarchidae	Redbreast sunfish	6	0
Pedernales Fredericksburg	8/23/2012	Centrarchidae	Longear sunfish	6	0
Pedernales Fredericksburg	8/23/2012	Centrarchidae	Largemouth bass	6	0
Pedernales Fredericksburg	8/23/2012	Cyprinidae	Red shiner	12	2
Pedernales Fredericksburg	8/23/2012	Cyprinidae	Blacktail shiner	10	1
Pedernales Fredericksburg	8/23/2012	Ictaluridae	Channel catfish	1	0
San Marcos Palmetto	8/24/2012	Centrarchidae	Longear sunfish	6	1
San Marcos Palmetto	8/24/2012	Centrarchidae	Largemouth bass	6	2
San Marcos Palmetto	8/24/2012	Cyprinidae	Red shiner	10	4
San Marcos Palmetto	8/24/2012	Cyprinidae	Blacktail shiner	10	1
San Marcos Palmetto	8/24/2012	Percidae	Dusky darter	2	0
			Totals	75	11

¹All of the species listed in table 4 are among those listed in table 3.

In 2012, an average of 217 individuals and an average of 14.9 species were caught per site. In 2013, the average number of individuals and species caught decreased to 151 and 11.7, respectively, per site (and per sampling period in the cases of the Guadalupe Victoria site [fig. 1, table 1] and the Guadalupe Kerrville site, which were each sampled twice) (tables 3–6). Reasons for discrepancies in the number of fish and the number of species caught between 2012 and 2013 include changes in sampling methodology. To minimize holding times, stress, and mortality during the transfer of fish to the hatchery in 2013, fish-collection activity was often suspended once sufficient quantities of target fish species were collected. Additionally, only five of the sites that were sampled in 2012 were resampled in 2013, so direct comparisons of fish abundance between 2012 and 2013 should not be made. The largest number of individuals (416) was collected on August 22, 2012, from the Llano Castell site, a site that was not resampled in 2013. In addition, fish populations might have declined in the stream reaches at sites that were sampled in 2012 and again in 2013; fish collection activities might have caused a reduction in the number and types of fish available for collection at sites resampled in 2013 relative to 2012.

Frequency of Parasitism in Fish Assessed for Glochidia in Field and Laboratory Settings

Only a small percentage (3 percent in 2012 and 19 percent in 2013) of the fish that were collected over the course of the study was sent to the hatchery for glochidia recovery. Any fish that were not submitted to the hatchery were assessed for parasitism by glochidia in the field or in the laboratory. The number of parasitized fish was used as the basis for comparison between fish assessed for glochidia in field and laboratory settings rather than the number of glochidia observed because of concerns that glochidia could be mistaken for other types of parasites. There was also some concern that glochidia counts made on live fish in the field would be less accurate than assessing the presence or absence of glochidia. References to parasitism in fish that were assessed for glochidia in the field or in the laboratory will refer to instances of suspected parasitism by glochidia because of the potential for mistaking other types of parasites with glochidia. It is important to note that a fish parasitized with glochidia is not necessarily parasitized with

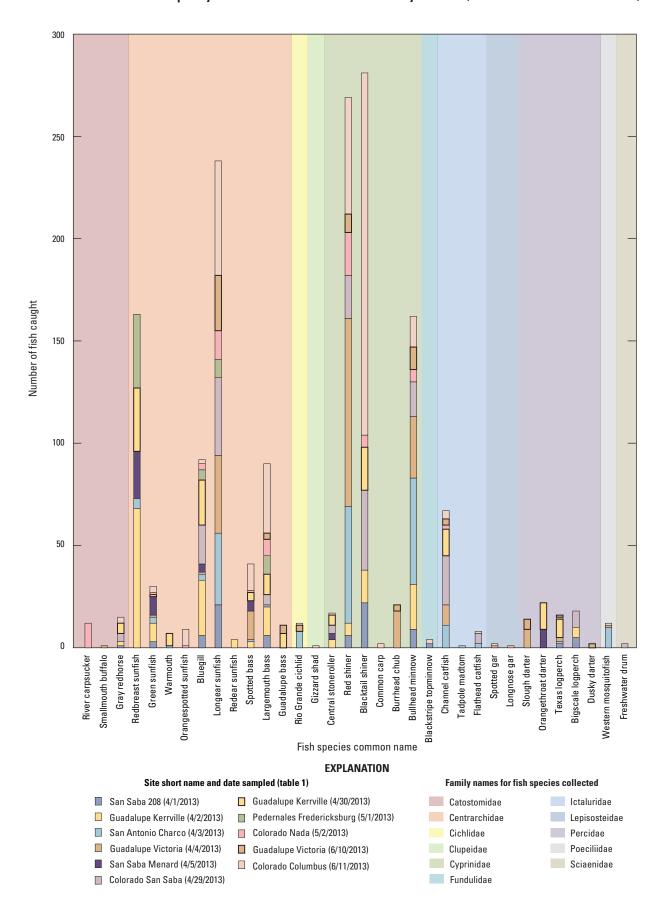


Figure 9. Abundance of potential host fish from sites sampled in 2013 in central and southeastern Texas, 2012–13.

glochidia from one of the target mussel species. In addition, the presence of glochidia on a fish does not necessarily indicate that the fish is a host because glochidia will attach to nonhosts. Additionally, fish that were sent to the hatchery for glochidia recovery were not included in any calculations of average total length because these fish were not measured prior to being sent. These measurements were not taken in order to maximize fish survival by minimizing handling and stress to the fish.

A minimum of two fish (any species) parasitized with glochidia was collected from each of the 10 sites sampled during 2012 (table 3). The highest percentage of parasitized fish (19.1 percent) was measured at the Guadalupe Victoria site (27 instances of parasitism out of 141 fish), whereas the lowest percentage (1.0 percent) was measured at the Llano Castell site (4 instances of parasitism out of 416 fish) (fig. 10). More than 10 percent of the fish were parasitized at three sites (Colorado Garwood, Guadalupe Victoria, and San Saba 126). These three sites plot above the dashed line depicting the relation between the number of fish parasitized with glochidia by site compared to the number of fish collected by site in fig. 10*B*.

Parasitized individuals were collected from five families of fish in 2012 (fig. 11, table 3). The highest percentage of parasitized individuals (12.1 percent) was measured in the catfish family Ictaluridae (ictalurids), followed by centrarchids at 7.0 percent (fig. 11A). Centrarchids had the highest number of parasitized individuals (87), followed by cyprinids and the ictalurids (each with eight individuals) (fig. 11*B*). Each of the seven families that did not have any parasitized individuals was represented by 14 or fewer individuals from all 10 sites combined. The seven families of fish without any documented parasitized individuals may be an artifact of the small number of individuals representing these families that were collected during the study. Other factors that could have contributed to the lack of parasitism in these seven families include fish morphology, behavior, and mesohabitat preference. The two families with the largest average total length (Lepisosteidae and Sciaenidae) and the three families with the smallest average total length (Characidae, Fundulidae, and Poeciliidae, each of which is represented by a single collected species) had no parasitized individuals, so the size of the individual was not a consistent predictor of the frequency of parasitism.

Nine sites were sampled in 2013, and 2 of the 9 sites (Guadalupe Victoria and Guadalupe Kerrville) were sampled twice (table 1). No parasitized fish were observed at 1 of the 9 sites sampled in 2013 (San Saba Menard), and no parasitized fish were recorded at another site (Guadalupe Victoria) the second time it was sampled on June 10, 2013 (fig. 12, table 5). The highest percentage of parasitized fish (22.7 percent) was recorded at the Pedernales Fredericksburg site (10 instances of parasitism out of 44 fish), followed by the Guadalupe Victoria site on April 4, 2013, at 13.5 percent

(22 instances of parasitism out of 163 fish) and the San Saba 208 site on April 1, 2013, at 13.0 percent (7 instances of parasitism out of 54 fish) (fig. 12, table 5). The percentage of parasitized fish increased substantially at the Pedernales Fredericksburg site between 2012 (1.4 percent) and 2013 (22.7) percent), likely because the sampled area was upstream from the bridge at this site in 2013, whereas it was downstream from the bridge in 2012 (table 1). The area that was sampled upstream from the bridge was wider than the downstream area, and it included runs and pools more than 2 feet deep. which provide better habitat for larger fish (average total lengths increased from 98 mm in 2012 to 158 mm in 2013). Three sites (Pedernales Fredericksburg, San Saba 208, and Guadalupe Victoria) plot above the dashed line in figure 12B used to identify sites where more than 10 percent of fish were parasitized with glochidia.

Parasitized individuals were only collected from three families of fish in 2013 (fig. 13, table 5). Ictalurids exhibited the highest frequency of parasitism (26.5 percent), followed by centrarchids at 7.1 percent. Centrarchids had the highest number of parasitized individuals (40), followed by ictalurids (13). Of the eight families that did not have any parasitized individuals, it is possible that insufficient numbers of individuals were collected to accurately assess the frequency of parasitism. For example, at most only 25 individuals were collected representing each of these eight families from all nine sites combined. Although collecting large sample sizes can put additional survival stress on species that are not common, there is some evidence underscoring the importance of large sample sizes in determining a representative proportion of parasitized individuals in a family. Members of the two families that were collected most frequently, centrarchids (1,247 individuals in 2012 and 564 individuals in 2013) and cyprinids (581 individuals in 2012 and 654 individuals in 2013), were parasitized in almost the same percentages in 2012 and 2013. In 2012, 7.0 percent of centrarchids and 1.4 percent of cyprinids were parasitized, whereas in 2013, 7.1 percent of centrarchids were parasitized and 1.1 percent of cyprinids were parasitized.

Of the fish that were not sent to the hatchery but assessed for glochidia in the field or in the laboratory, at least 13 species were parasitized in 2012; this does not include two parasitized individuals of the genus *Micropterus* that were collected at the Guadalupe Victoria site whose species could not be identified because they were too small and immature (fig. 14, table 3). Five of the 13 species of parasitized fish identified in 2012 were centrarchids, and 3 were cyprinids. Other than the unknown *Micropterus* genus individuals, the longear sunfish was the species with the highest percentage of parasitized individuals in 2012 (17.3 percent; 36 of 208 individuals), followed by bullhead minnow (14.3 percent; 4 of 28 individuals) and channel catfish (13.0 percent; 7 of 54 individuals). Of the fish that were not sent to the hatchery but assessed for glochidia in the field or in the laboratory,



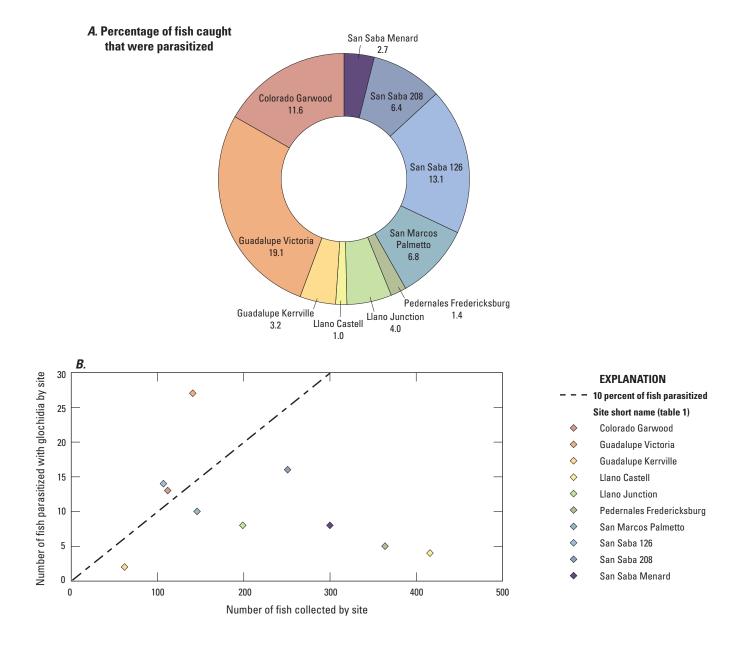


Figure 10. A, percentage, and B, number of parasitized fish that were assessed in the field or laboratory for glochidia at 10 sites in 2012 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.

only eight species were parasitized in 2013. Four of the eight species of parasitized fish were centrarchids, two were cyprinids, and two were ictalurids. Flathead catfish (Pylodictis olivaris) was the species with the highest percentage of parasitized individuals in 2013 (42.9 percent), although sample size was small (3 parasitized individuals out of 7). Channel catfish was the second most parasitized species in 2013 (24.4 percent) (10 of 41 individuals), followed by longear sunfish (11.6 percent) (25 of 216 individuals) and redbreast sunfish (9.1 percent) (13 of 143 individuals) (table

5). The number of parasitized species based on field and laboratory observations was lower in 2013 than in 2012, likely because more fish were sent to the hatchery in 2013 (321) than in 2012 (75). In many cases in 2013, all of the individuals of a given species that were collected at a site were submitted to the hatchery. When data from the hatchery are included, the total number of parasitized species in 2013 (16) surpasses the number of parasitized species collected in 2012 (likely 13 but could be as many as 15 depending on the species of the two unknown parasitized individuals of Micropterus genus).

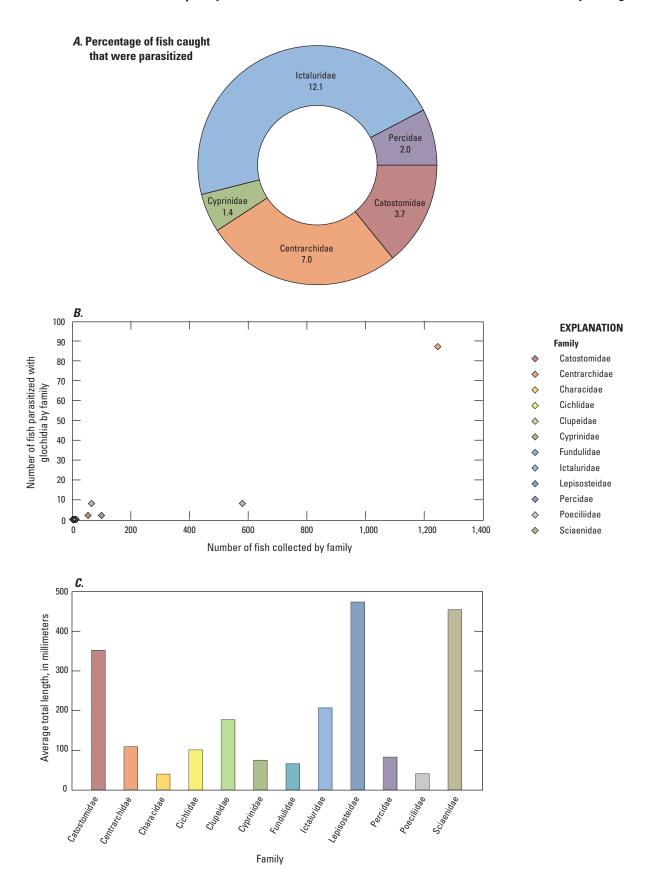


Figure 11. A, percentage; B, number; and C, total length of fish grouped by family that were assessed in the field or laboratory for glochidia in 2012 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.

A. Percentage of fish caught that were parasitized Guadalupe Victoria (6/10/2013) 0.0 - not shown San Saba Menard 0.0 - not shown Colorado San Saba Colorado Nada Colorado Columbus 0.5 San Saba 208 13.0 Guadalupe San Antonio Charco Victoria (4/4/2013) 13.5 Guadalupe Kerrville (4/2/2013) Pedernales Fredericksburg 2.0 22.7

Guadalupe Kerrville (4/30/2013) 1.7

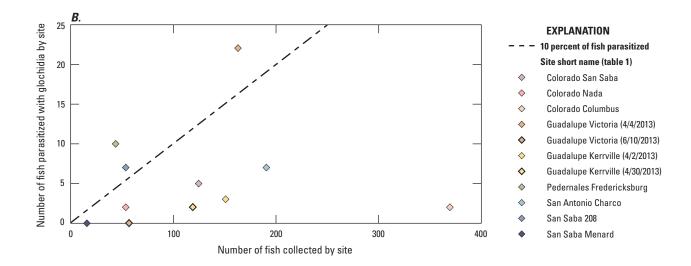


Figure 12. A, percentage, and B, number of parasitized fish that were assessed in the field or laboratory for glochidia at nine sites (two of which were sampled twice) in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.

Table 5. Number of fish collected and glochidial parasitism by species from sites sampled in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13. [No., number; Avg., average; mm, millimeters; shading indicates fish species are members of the same family; cross-hatching indicates fish species were not collected]

			Short name (fig. 1) with sampling date in parentheses										
			;	San Saba 20	8 (4/1)	Gua	dalupe Kerr	ville (4/2)	Sa	n Antonio C	narco (4/3)		
Family	Species	Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations		
Catostomidae	Carpiodes carpio	River carpsucker											
	Ictiobus bubalus	Smallmouth buffalo											
	Moxostoma congestum	Gray redhorse				1	430	0					
Centrarchidae	Lepomis auritus	Redbreast sunfish				68	111	3	5	53	0		
	Lepomis cyanellus	Green sunfish	3	97	0	9	97	0	3	65	0		
	Lepomis gulosus	Warmouth							1	65	0		
	Lepomis humilis	Orangespotted sunfish											
	Lepomis macrochirus	Bluegill				21	74	0	3	72	0		
	Lepomis megalotis	Longear sunfish	21	68	7				35	89	1		
	Lepomis microlophus	Redear sunfish				4	133	0					
	Micropterus punctulatus	Spotted bass				3	115	0	1	180	0		
	Micropterus salmoides	Largemouth bass	3	93	0	11	154	0	1	225	0		
	Micropterus treculii	Guadalupe bass											
Cichlidae	Herichthys cyanoguttatum	Rio Grande cichlid							8	59	0		
Clupeidae	Dorosoma cepedianum	Gizzard shad											
Cyprinidae	Campostoma anomalum	Central stoneroller				4	81	0					
	Cyprinella lutrensis	Red shiner							57	51	2		
	Cyprinella venusta	Blacktail shiner	19	83	0	13	78	0					
	Cyprinus carpio	Common carp											
	Macrhybopsis marconis	Burrhead chub											
	Pimephales vigilax	Bullhead minnow	4	43	0	17	56	0	52	52	0		

Table 5. Number of fish collected and glochidial parasitism by species from sites sampled in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with sampling date in parentheses									
				San Saba 20	08 (4/1)	Gua	dalupe Kerr	ville (4/2)	Sai	n Antonio Cl	ıarco (4/3)	
Family	Species	Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	
Fundulidae	Fundulus notatus	Blackstripe topminnow	2	60	0							
Ictaluridae	Ictalurus punctatus	Channel catfish							11	202	4	
	Noturus gyrinus	Tadpole madtom							1	65	0	
	Pylodictis olivaris	Flathead catfish							2	180	0	
Lepisosteidae	Lepisosteus oculatus	Spotted gar										
	Lepisosteus osseus	Longnose gar										
Percidae	Etheostoma spectabile	Orangethroat darter										
	Percina carbonaria	Texas logperch	2	48	0				1	55	0	
	Percina sciera	Dusky darter										
Poeciliidae	Gambusia affinis	Western mosquitofish							10	42	0	
Sciaenidae	Aplodinotus grunniens	Freshwater drum										
		Totals	54	73	7	151	101	3	191	71	7	
	Percentage of fi	sh infested with glochidia			13.0			2.0			3.7	
Total ı	number of species collected (i	includes fish from table 6)			11			12			15	
	Species not shown in this tal	ole but included in table 6		Gray redho	orse		Channel ca	atfish				
				Bluegill			Orangethro	oat darter				
				Red shiner								
				Bigscale lo	ogperch							

Table 5. Number of fish collected and glochidial parasitism by species from sites sampled in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with sampling date in parentheses										
			Gua	adalupe Vic	toria (4/4)	Sar	Saba Mer	nard (4/5)	Col	orado San S	Saba (4/29)		
Family	Species	Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations		
Catostomidae	Carpiodes carpio	River carpsucker											
	Ictiobus bubalus	Smallmouth buffalo	1	480	0								
	Moxostoma congestum	Gray redhorse											
Centrarchidae	Lepomis auritus	Redbreast sunfish				13	130	0					
	Lepomis cyanellus	Green sunfish	1	100	0	2	98	0					
	Lepomis gulosus	Warmouth	1////			/////							
	Lepomis humilis	Orangespotted sunfish											
	Lepomis macrochirus	Bluegill	1	90	0	1	55	0	13	84	0		
	Lepomis megalotis	Longear sunfish	33	95	16	/////			32	89	0		
	Lepomis microlophus	Redear sunfish	1////						/////				
	Micropterus punctulatus	Spotted bass	11	165	1								
	Micropterus salmoides	Largemouth bass	1////		////////								
	Micropterus treculii	Guadalupe bass											
Cichlidae	Herichthys cyanoguttatum	Rio Grande cichlid											
Clupeidae	Dorosoma cepedianum	Gizzard shad											
Cyprinidae	Campostoma anomalum	Central stoneroller							1	65	0		
	Cyprinella lutrensis	Red shiner	82	61	4				21	59	0		
	Cyprinella venusta	Blacktail shiner	1////						27	80	0		
	Cyprinus carpio	Common carp							1/////		////////		
	Macrhybopsis marconis	Burrhead chub	8	60	0								
	Pimephales vigilax	Bullhead minnow	20	61	0				10	58	0		
Fundulidae	Fundulus notatus	Blackstripe topminnow											

Table 5. Number of fish collected and glochidial parasitism by species from sites sampled in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with sampling date in parentheses										
			Gu	adalupe Vic	toria (4/4)	Sai	n Saba Men	ard (4/5)	Col	orado San S	Saba (4/29)		
Family	Species	Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations		
Ictaluridae	Ictalurus punctatus Noturus gyrinus Pylodictis olivaris	Channel catfish Tadpole madtom Flathead catfish	3	340	1				14 ///// 5	191 /////// 111	2//////////////////////////////////////		
Lepisosteidae	Lepisosteus oculatus Lepisosteus osseus	Spotted gar Longnose gar											
Percidae	Etheostoma spectabile Percina carbonaria Percina sciera	Orangethroat darter Texas logperch Dusky darter	///// 2 /////	60 60									
Poeciliidae Sciaenidae	Gambusia affinis	Western mosquitofish Freshwater drum	1	//////// 35 ////////	////////////// 0 /////////////////////				2	313			
Sciacinuae	Aplodinotus grunniens	Totals	163	83	22	16	121	0	125	95	5		
	_	sh infested with glochidia			13.5			0.0			4.0		
Total ı	number of species collected (i	ncludes fish from table 6)			13			6			12		
	Species not shown in this tal	ole but included in table 6		Spotted ga	r		Spotted ba	ass		Gray redh	orse		
				Slough da	ter		Central st	oneroller		Largemou	th bass		
							Orangethr	oat darter		Bigscale l	ogperch		

Table 5. Number of fish collected and glochidial parasitism by species from sites sampled in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with sampling date in parentheses									
			Guad	lalupe Kerr	ville (4/30)	Pedern	ales Freder	icksburg (5/1)	(Colorado Na	da (5/2)	
Family	Species	Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	
Catostomidae	Carpiodes carpio	River carpsucker							12	201	0	
	Ictiobus bubalus	Smallmouth buffalo										
	Moxostoma congestum	Gray redhorse	2	428	0							
Centrarchidae	Lepomis auritus	Redbreast sunfish	26	130	0	31	159	10				
	Lepomis cyanellus	Green sunfish	1	85	0	1////		////////	1	80	0	
	Lepomis gulosus	Warmouth	6	183	0							
	Lepomis humilis	Orangespotted sunfish	1////						1	110	0	
	Lepomis macrochirus	Bluegill	17	97	0							
	Lepomis megalotis	Longear sunfish	1////			9	137	0	9	87	1	
	Lepomis microlophus	Redear sunfish				/////		////////	/////			
	Micropterus punctulatus	Spotted bass	4	111	0				1	355	0	
	Micropterus salmoides	Largemouth bass	5	247	0	4	196	0	5	125	1	
	Micropterus treculii	Guadalupe bass	7	259	0				/////			
Cichlidae	Herichthys cyanoguttatum	Rio Grande cichlid										
Clupeidae	Dorosoma cepedianum	Gizzard shad										
Cyprinidae	Campostoma anomalum	Central stoneroller	5	108	0							
	Cyprinella lutrensis	Red shiner							21	58	0	
	Cyprinella venusta	Blacktail shiner	21	79	0				1	65	0	
	Cyprinus carpio	Common carp										
	Macrhybopsis marconis	Burrhead chub										
	Pimephales vigilax	Bullhead minnow							1	70	0	
Fundulidae	Fundulus notatus	Blackstripe topminnow							/////			
Ictaluridae	Ictalurus punctatus	Channel catfish	8	204	2				1	380	0	

Table 5. Number of fish collected and glochidial parasitism by species from sites sampled in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

					Short na	me (fig. 1) with sampling date in parentheses					
			Guad	dalupe Keri	ville (4/30)	Pedern	ales Freder	icksburg (5/1)	(Colorado Na	da (5/2)
Family	Species	Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations
	Noturus gyrinus	Tadpole madtom									
	Pylodictis olivaris	Flathead catfish									
Lepisosteidae	Lepisosteus oculatus	Spotted gar									
	Lepisosteus osseus	Longnose gar							1	570	0
Percidae	Etheostoma spectabile	Orangethroat darter	13	43	0						
	Percina carbonaria	Texas logperch	4	119	0						
	Percina sciera	Dusky darter									
Poeciliidae	Gambusia affinis	Western mosquitofish									
Sciaenidae	Aplodinotus grunniens	Freshwater drum									
		Totals	119	130	2	44	158	10	54	123	2
	Percentage of	fish infested with glochidia			1.7			22.7			3.7
Tot	al number of species collected	(includes fish from table 6)			13			5			12
	Species not shown in this t	able but included in table 6					Bluegill			Bluegill	
							Texas log	perch			

Table 5. Number of fish collected and glochidial parasitism by species from sites sampled in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with sampling date in parentheses									
				Guadalupe Vic	toria (6/10)		Colorado Colu	mbus (6/11)				
Family	Species	Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations				
Catostomidae	Carpiodes carpio	River carpsucker										
	Ictiobus bubalus	Smallmouth buffalo										
	Moxostoma congestum	Gray redhorse				3	290	0				
Centrarchidae	Lepomis auritus	Redbreast sunfish										
	Lepomis cyanellus	Green sunfish				3	113	0				
	Lepomis gulosus	Warmouth				///////						
	Lepomis humilis	Orangespotted sunfish				8	89	0				
	Lepomis macrochirus	Bluegill				2	90	0				
	Lepomis megalotis	Longear sunfish	24	89	0	53	85	0				
	Lepomis microlophus	Redear sunfish	1/////			///////						
	Micropterus punctulatus	Spotted bass				13	117	0				
	Micropterus salmoides	Largemouth bass				31	68	0				
	Micropterus treculii	Guadalupe bass	4	196	0							
Cichlidae	Herichthys cyanoguttatum	Rio Grande cichlid	3	98	0	1	70	0				
Clupeidae	Dorosoma cepedianum	Gizzard shad				1	220	0				
Cyprinidae	Campostoma anomalum	Central stoneroller				1	70	0				
	Cyprinella lutrensis	Red shiner	9	51	0	57	48	0				
	Cyprinella venusta	Blacktail shiner				172	65	1				
	Cyprinus carpio	Common carp				2	403	0				
	Macrhybopsis marconis	Burrhead chub	3	60	0							
	Pimephales vigilax	Bullhead minnow	11	49	0	15	51	0				
Fundulidae	Fundulus notatus	Blackstripe topminnow				2	55	0				
Ictaluridae	Ictalurus punctatus	Channel catfish				4	483	1				
	Noturus gyrinus	Tadpole madtom										
	Pylodictis olivaris	Flathead catfish										

Table 5. Number of fish collected and glochidial parasitism by species from sites sampled in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with sampling date in parentheses								
				Guadalupe Vic	etoria (6/10)		Colorado Colu	mbus (6/11)			
Family	Species	Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	No. of fish	Avg. total length (mm)	No. of glochidia infestations			
Lepisosteidae	Lepisosteus oculatus	Spotted gar				1	540	0			
	Lepisosteus osseus	Longnose gar				///////					
Percidae	Etheostoma spectabile	Orangethroat darter									
	Percina carbonaria	Texas logperch	1	100	0						
	Percina sciera	Dusky darter	2	43	0						
Poeciliidae	Gambusia affinis	Western mosquitofish	//////			1	30	0			
Sciaenidae	Aplodinotus grunniens	Freshwater drum									
		Totals	57	80	0	370	78	2			
	Percentage	of fish infested with glochidia			0.0			0.5			
To	otal number of species collect	ed (includes fish from table 6)			11			19			
	Species not shown in thi	s table but included in table 6		Largemouth ba	ass		Flathead catfish				
				Channel catfish	1						
				Slough darter							

Table 5. Number of fish collected and glochidial parasitism by species from sites sampled in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with sampling date in parentheses										
				Total - all	sites (2013 onl	у)		Totals - by	family (2013 or	ıly)			
Family	Species	Species common name	No. of fish	Avg. total length (mm)	No. of glochidia infestations	Percentage of fish infested	No. of fish	Avg. total length (mm)	No. of glochidia infestations	Percentage of fish infested			
Catostomidae	Carpiodes carpio	River carpsucker	12	201	0	0.0							
	Ictiobus bubalus	Smallmouth buffalo	1	480	0	0.0	19	266	0	0.0			
	Moxostoma congestum	Gray redhorse	6	359	0	0.0	1)	200	V	0.0			
Centrarchidae	Lepomis auritus	Redbreast sunfish	143	125	13	9.1							
	Lepomis cyanellus	Green sunfish	23	94	0	0.0							
	Lepomis gulosus	Warmouth	7	166	0	0.0							
	Lepomis humilis	Orangespotted sunfish	9	91	0	0.0							
	Lepomis macrochirus	Bluegill	58	83	0	0.0							
	Lepomis megalotis	Longear sunfish	216	89	25	11.6	564	108	40	7.1			
	Lepomis microlophus	Redear sunfish	4	133	0	0.0							
	Micropterus punctulatus	Spotted bass	33	141	1	3.0							
	Micropterus salmoides	Largemouth bass	60	116	1	1.7							
	Micropterus treculii	Guadalupe bass	11	236	0	0.0							
Cichlidae	Herichthys cyanoguttatum	Rio Grande cichlid	12	70	0	0.0	12	70	0	0.0			
Clupeidae	Dorosoma cepedianum	Gizzard shad	1	220	0	0.0	1	220	0	0.0			
Cyprinidae	Campostoma anomalum	Central stoneroller	11	91	0	0.0							
	Cyprinella lutrensis	Red shiner	247	55	6	2.4							
	Cyprinella venusta	Blacktail shiner	253	70	1	0.4	651	62	7	1.1			
	Cyprinus carpio	Common carp	2	403	0	0.0	654	02	/	1.1			
	Macrhybopsis marconis	Burrhead chub	11	60	0	0.0							
	Pimephales vigilax	Bullhead minnow	130	54	0	0.0							

Table 5. Number of fish collected and glochidial parasitism by species from sites sampled in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.—Continued

			Short name (fig. 1) with sampling date in parentheses										
		Species common name		Total - all	sites (2013 onl	y)	Totals - by family (2013 only)						
Family	Species		No. of fish	Avg. total length (mm)	No. of glochidia infestations	Percentage of fish infested	No. of fish	Avg. total length (mm)	No. of glochidia infestations	Percentage of fish infested			
Fundulidae	Fundulus notatus	Blackstripe topminnow	4	58	0	0.0	4	58	0	0.0			
Ictaluridae	Ictalurus punctatus	Channel catfish	41	240	10	24.4							
	Noturus gyrinus	Tadpole madtom	1	65	0	0.0	49	221	13	26.5			
	Pylodictis olivaris	Flathead catfish	7	131	3	42.9							
Lepisosteidae	Lepisosteus oculatus	Spotted gar	1	540	0	0.0	2	555	٥	0.0			
	Lepisosteus osseus	Longnose gar	1	570	0	0.0	2	555	0	0.0			
Percidae	Etheostoma spectabile	Orangethroat darter	13	43	0	0.0							
	Percina carbonaria	Texas logperch	10	85	0	0.0	25	60	0	0			
	Percina sciera	Dusky darter	2	43	0	0.0							
Poeciliidae	Gambusia affinis	Western mosquitofish	12	40	0	0.0	12	40	0	0.0			
Sciaenidae	Aplodinotus grunniens	Freshwater drum	2	313	0	0.0	2	313	0	0.0			
		Totals	1,344	91	60	4.5	1,344	91	60	4.5			

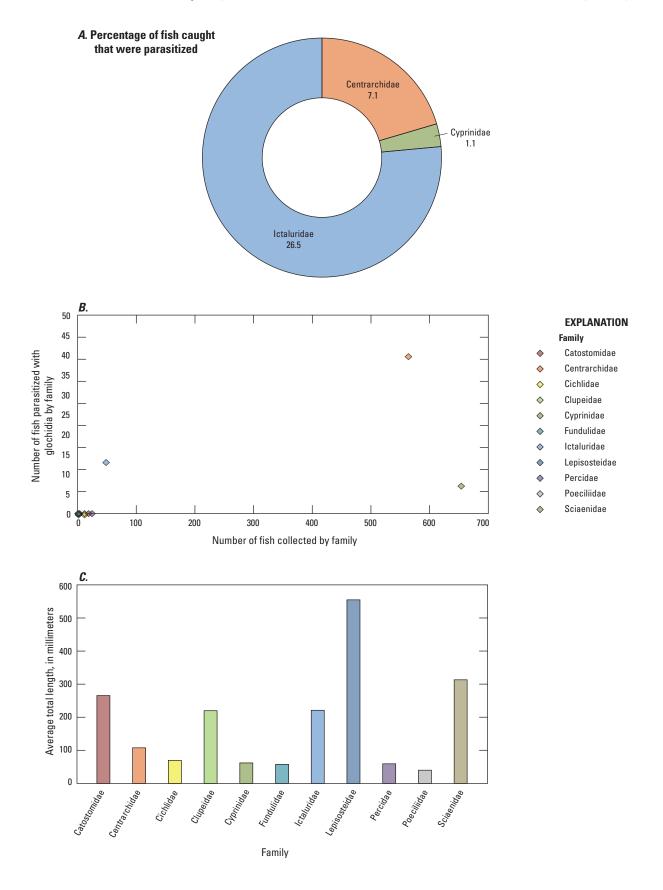


Figure 13. A, percentage; B, number; and C, average total length of fish grouped by family that were assessed in the field or laboratory for glochidia in 2013 as part of a mussel host-fish study, central and southeastern Texas, 2012–13.

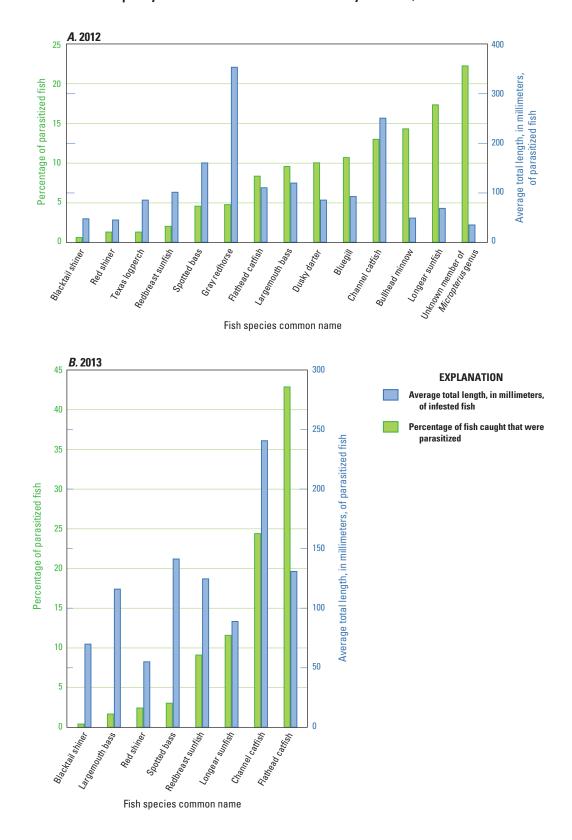


Figure 14. Percentage of parasitized fish and average total length of fish grouped by species that were assessed in the field or laboratory for glochidia in *A*, 2012; and *B*, 2013; as part of a mussel host-fish study, central and southeastern Texas, 2012–13.

Frequency of Juvenile Mussels or Glochidia Recovered from Hatchery-Held Fish

Multiple individuals of the same fish species were frequently placed in a single holding tank at the hatchery. In these cases, attributing juvenile mussels or glochidia to specific individuals and quantifying how many juvenile mussels or glochidia were recovered from each individual fish was not possible; therefore, parasitism in fish held at the hatchery is discussed as the average number of juvenile mussels or glochidia by fish species. As described in the "Methods" section of this report, juvenile mussels and glochidia were not differentiated in hatchery-held fish; therefore, when discussing study results pertaining to fish held at the hatchery, juvenile mussels and glochidia are hereinafter collectively referred to as "juvenile mussels." Because glochidia observed in the USGS laboratory on fish returned by the hatchery remained attached to the gills following the 28-day trials, these glochidia are referred to as "glochidia" rather than juvenile mussels. References to parasitism in fish that released juvenile mussels that were recovered at the hatchery refer to actual parasitism, whereas references to parasitism in fish that were returned by the hatchery and assessed for glochidia in the USGS laboratory refer to suspected parasitism.

In 2012, fish were only submitted to the hatchery from the Pedernales Fredericksburg site on August 23 and the San Marcos Palmetto site on August 24 (table 4). Of the six species collected at the Pedernales Fredericksburg site that were submitted to the hatchery, only red shiners and blacktail shiners released any juvenile mussels; the red shiners (n=12) released 2 juvenile mussels, and the blacktail shiners (n=10) released 1 juvenile mussel (table 4). Of the five species submitted to the hatchery that were collected at the San Marcos Palmetto site, all except for the dusky darters (*Percina sciera*) released juvenile mussels. The longear sunfish (n=6) and the blacktail shiners (n=10) released one juvenile mussel apiece. The largemouth bass (n=6) released 2 juvenile mussels, and the red shiners (n=10) released 4 juvenile mussels.

With the exception of the San Antonio Charco site, fish were submitted to the hatchery from all sampling sites in 2013, including fish from both sample collection dates at the Guadalupe Victoria and Guadalupe Kerrville sites. Fish collected on April 3, 2013, from the San Antonio Charco site were not submitted to the hatchery because of a lack of available holding tanks at that time; fish collected in early April 2013 at sites which had previously been sampled in 2012 (the Guadalupe Victoria, Guadalupe Kerrville, San Saba 208, and San Saba Menard sites) were given priority. The average number of juvenile mussels or glochidia per fish is presented by species in figures 15 and 16 as the sum of the proportion of juvenile mussels per fish by species recovered at the hatchery and the proportion of glochidia per fish extracted in the laboratory from fish returned by the hatchery. In most

cases, the proportion of juvenile mussels recovered at the hatchery represented the majority of the average number of juvenile mussels or glochidia per fish by species depicted in figures 15 and 16.

During the first sampling period in 2013 (April 1–5), slightly more than half (16 out of 29) of the fish species (on a per site basis) that were submitted to the hatchery released juvenile mussels, and individuals from 5 of the 16 fish species retained glochidia after completing trials at the hatchery (fig. 15). The most species that released juvenile mussels (7 out of 8 species) were collected at the Guadalupe Victoria site. There were at least two species that released juvenile mussels that were collected at each of the remaining three sites sampled during the first sampling period in 2013. Largemouth bass collected from the Guadalupe Kerrville site and spotted bass (*Micropterus punctulatus*) collected from the Guadalupe Victoria site released more than four juvenile mussels per individual on average.

Compared to the other sampling periods in 2013, substantially fewer glochidia per fish were present on fish submitted to the hatchery during the second sampling period in 2013 (April 29–May 2). Only longear sunfish (collected at the Colorado San Saba site) averaged more than one juvenile mussel per fish (fig. 15). Less than a third (7 out of 25) of the fish species (on a per site basis) released juvenile mussels that were recovered at the hatchery; individuals from 3 of these 7 fish species retained glochidia after completing trials at the hatchery. One largemouth bass collected at the Colorado Nada site retained a single glochidium that was not recovered at the hatchery. None of the fish collected at the Pedernales Fredericksburg and Colorado Nada sites released juvenile mussels at the hatchery.

Although only two sites were sampled during the third sampling period in 2013 (June 10–11), more juvenile mussels were recovered at the hatchery during this sampling period (107) than were recovered during the first two sampling periods in 2013 combined (102) (table 6). Four out of six fish species (from the two sites) that were submitted to the hatchery released juvenile mussels, and individuals from 2 of the 4 fish species retained glochidia after completing trials at the hatchery (fig. 15). An average of 17 juvenile mussels was recovered per largemouth bass submitted to the hatchery from the Guadalupe Victoria site during the third sampling period (fig. 15) on June 10, 2013, and the 14 fish (representing 4 species) submitted from the Guadalupe Victoria site on that date released a total of 102 juvenile mussels at the hatchery for an average of 7.3 juvenile mussels per fish (table 6).

A total of 19 fish species collected at nine sites (including both 2013 sampling periods at Guadalupe Victoria and Guadalupe Kerrville) was submitted to the hatchery in 2013, and 14 of these species released juvenile mussels that were recovered at the hatchery (fig. 16*A*). The three most productive species, in terms of average juvenile mussels recovered, were longear sunfish, largemouth bass, and spotted bass, each of which averaged more than two glochidia recovered per individual. Centrarchids released the highest number of

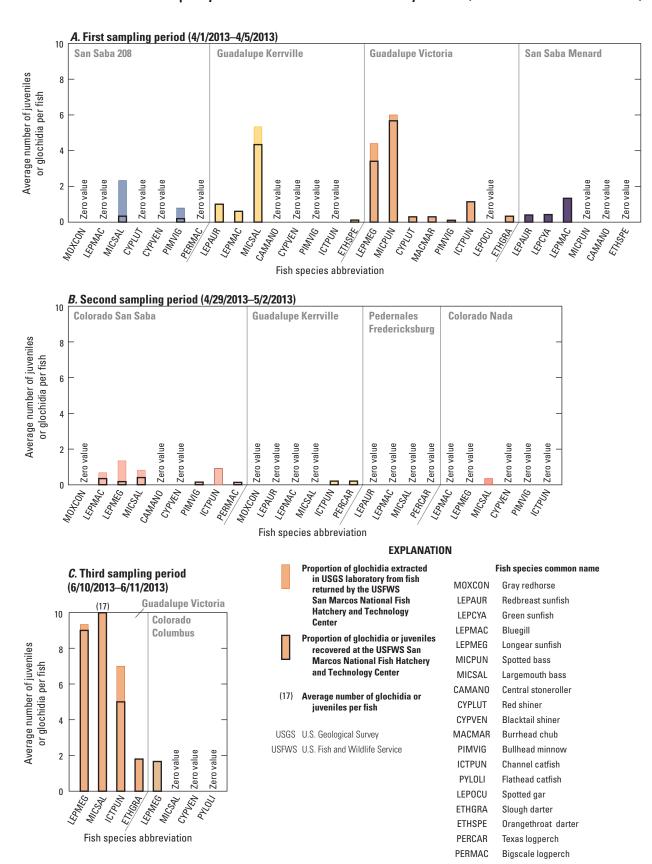


Figure 15. Average number of juvenile mussels or glochidia collected per fish species by site from fish held at the U.S. Fish and Wildlife Service San Marcos National Fish Hatchery and Technology Center in 2013 (*A*, First sampling period; *B*, second sampling period; and *C*, third sampling period), as part of a mussel host-fish study, central and southeastern Texas, 2012–13.

Table 6. Juvenile mussels or glochidia recovered from hatchery-held fish collected in 2013 in central and southeastern Texas and submitted to U.S. Fish and Wildlife Service National Fish Hatchery and Technology Center in San Marcos, Tex., as part of a mussel host-fish study, 2012–13.

Short name	Date	Family	Species common name	Number of fish submitted to hatchery	Number of juvenile mussels or glochidia recovered from hatcheryheld fish	Number of juvenile mussels or glochidia collected in laboratory from fish returned by the hatchery
San Saba 208	4/1/2013	Catostomidae	Gray redhorse	1	0	0
San Saba 208	4/1/2013	Centrarchidae	Bluegill	6	0	0
San Saba 208	4/1/2013	Centrarchidae	Largemouth bass	3	1	6
San Saba 208	4/1/2013	Cyprinidae	Red shiner	6	0	0
San Saba 208	4/1/2013	Cyprinidae	Blacktail shiner	3	0	0
San Saba 208	4/1/2013	Cyprinidae	Bullhead minnow	5	1	3
San Saba 208	4/1/2013	Percidae	Bigscale logperch	5	0	0
Guadalupe Kerrville	4/2/2013	Centrarchidae	Redbreast sunfish	2	2	0
Guadalupe Kerrville	4/2/2013	Centrarchidae	Bluegill	5	3	0
Guadalupe Kerrville	4/2/2013	Centrarchidae	Largemouth bass	3	13	3
Guadalupe Kerrville	4/2/2013	Cyprinidae	Central stoneroller	6	0	0
Guadalupe Kerrville	4/2/2013	Cyprinidae	Blacktail shiner	9	0	0
Guadalupe Kerrville	4/2/2013	Cyprinidae	Bullhead minnow	10	0	0
Guadalupe Kerrville	4/2/2013	Ictaluridae	Channel catfish	3	0	0
Guadalupe Kerrville	4/2/2013	Percidae	Orangethroat darter	9	1	0
Guadalupe Victoria	4/4/2013	Centrarchidae	Longear sunfish	5	17	5
Guadalupe Victoria	4/4/2013	Centrarchidae	Spotted bass	3	17	1
Guadalupe Victoria	4/4/2013	Cyprinidae	Red shiner	10	3	0
Guadalupe Victoria	4/4/2013	Cyprinidae	Burrhead chub	10	3	0
Guadalupe Victoria	4/4/2013	Cyprinidae	Bullhead minnow	10	1	0
Guadalupe Victoria	4/4/2013	Ictaluridae	Channel catfish	7	8	0
Guadalupe Victoria	4/4/2013	Lepisosteidae	Spotted gar	1	0	0
Guadalupe Victoria	4/4/2013	Percidae	Slough darter	9	3	0
San Saba Menard	4/5/2013	Centrarchidae	Redbreast sunfish	10	4	0
San Saba Menard	4/5/2013	Centrarchidae	Green sunfish	7	3	0
San Saba Menard	4/5/2013	Centrarchidae	Bluegill	3	4	0
San Saba Menard	4/5/2013	Centrarchidae	Spotted bass	5	0	0
San Saba Menard	4/5/2013	Cyprinidae	Central stoneroller	3	0	0
San Saba Menard	4/5/2013	Percidae	Orangethroat darter	9	0	0
		Totals	(first sampling period)	168	84	18

Table 6. Juvenile mussels or glochidia recovered from hatchery-held fish collected in 2013 in central and southeastern Texas and submitted to U.S. Fish and Wildlife Service National Fish Hatchery and Technology Center in San Marcos, Tex., as part of a mussel host-fish study, 2012–13.—Continued

Short name	Date	Family	Species common name	Number of fish submitted to hatchery	Number of juvenile mussels or glochidia recovered from hatchery- held fish	Number of juvenile mussels or glochidia collected in laboratory from fish returned by the hatchery
Colorado San Saba	4/29/2013	Catostomidae	Gray redhorse	4	0	0
Colorado San Saba	4/29/2013	Centrarchidae	Bluegill	6	2	2
Colorado San Saba	4/29/2013	Centrarchidae	Longear sunfish	6	1	7
Colorado San Saba	4/29/2013	Centrarchidae	Largemouth bass	5	2	2
Colorado San Saba	4/29/2013	Cyprinidae	Central stoneroller	3	0	0
Colorado San Saba	4/29/2013	Cyprinidae	Blacktail shiner	12	0	0
Colorado San Saba	4/29/2013	Cyprinidae	Bullhead minnow	7	1	0
Colorado San Saba	4/29/2013	Ictaluridae	Channel catfish	10	9	0
Colorado San Saba	4/29/2013	Percidae	Bigscale logperch	8	1	0
Guadalupe Kerrville	4/30/2013	Catostomidae	Gray redhorse	3	0	0
Guadalupe Kerrville	4/30/2013	Centrarchidae	Redbreast sunfish	5	0	0
Guadalupe Kerrville	4/30/2013	Centrarchidae	Bluegill	5	0	0
Guadalupe Kerrville	4/30/2013	Centrarchidae	Largemouth bass	5	0	0
Guadalupe Kerrville	4/30/2013	Ictaluridae	Channel catfish	5	1	0
Guadalupe Kerrville	4/30/2013	Percidae	Texas logperch	5	1	0
Pedernales Fredericksburg	5/1/2013	Centrarchidae	Redbreast sunfish	5	0	0
Pedernales Fredericksburg	5/1/2013	Centrarchidae	Bluegill	5	0	0
Pedernales Fredericksburg	5/1/2013	Centrarchidae	Largemouth bass	5	0	0
Pedernales Fredericksburg	5/1/2013	Percidae	Texas logperch	1	0	0
Colorado Nada	5/2/2013	Centrarchidae	Bluegill	3	0	0
Colorado Nada	5/2/2013	Centrarchidae	Longear sunfish	5	0	0
Colorado Nada	5/2/2013	Centrarchidae	Largemouth bass	3	0	1
Colorado Nada	5/2/2013	Cyprinidae	Blacktail shiner	5	0	0
Colorado Nada	5/2/2013	Cyprinidae	Bullhead minnow	5	0	0
Colorado Nada	5/2/2013	Ictaluridae	Channel catfish	1	0	0
		Totals (se	cond sampling period)	127	18	12
Guadalupe Victoria	6/10/2013	Centrarchidae	Longear sunfish	3	27	1
Guadalupe Victoria	6/10/2013	Centrarchidae	Largemouth bass	3	51	0
Guadalupe Victoria	6/10/2013	Ictaluridae	Channel catfish	3	15	6
Guadalupe Victoria	6/10/2013	Percidae	Slough darter	5	9	0
Colorado Columbus	6/11/2013	Centrarchidae	Longear sunfish	3	5	0
Colorado Columbus	6/11/2013	Centrarchidae	Largemouth bass	3	0	0
Colorado Columbus	6/11/2013	Cyprinidae	Blacktail shiner	5	0	0
Colorado Columbus	6/11/2013	Ictaluridae	Flathead catfish	1	0	0
		Totals (t	hird sampling period)	26	107	7
			(all sampling periods)	321	209	37

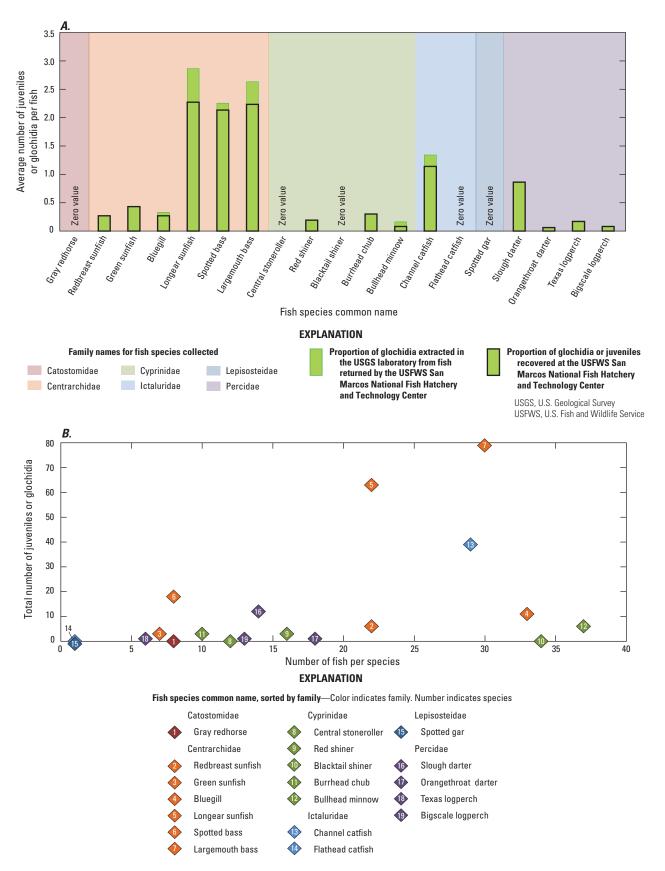


Figure 16. Composite from all sites sampled in 2013 of *A*, average number of juvenile mussels or glochidia collected per fish species; and *B*, total number of glochidia collected per fish species from fish held at the U.S. Fish and Wildlife Service San Marcos National Fish Hatchery and Technology Center as part of a mussel host-fish study, central and southeastern Texas, 2012–13.

juvenile mussels on average (1.48), followed by ictalurids (1.3) and percids (0.29). Centrarchids also retained the most glochidia per individual on average from fish returned by the hatchery (0.23) followed by ictalurids (0.20). All centrarchid and percid species that were submitted to the hatchery released juvenile mussels (fig. 164, table 6).

Summary

In 2012–13, the U.S. Geological Survey (USGS), in cooperation with the U.S. Fish and Wildlife Service (USFWS), completed the first phase of a two-phase study of mussel host-fish relations for five endemic mussel species in central and southeastern Texas that were State-listed as threatened on January 17, 2010: (1) Texas fatmucket (*Lampsilis bracteata*), (2) golden orb (*Quadrula aurea*), (3) smooth pimpleback (*Quadrula houstonensis*), (4) Texas pimpleback (*Quadrula petrina*), and (5) Texas fawnsfoot (*Truncilla macrodon*). On October 6, 2011, the USFWS announced the completion of a status review and determined that the five mussel species warranted listing under the Endangered Species Act; however, listing of these species at that time was precluded by higher priority listing actions, and currently (December 2014), they remained unlisted.

Freshwater mussels are long-lived, sedentary organisms that spend their larval stage as obligate parasites on the gills or fins of fishes, and many of these larvae, which are referred to as "glochidia," can survive only on a narrow range of host-fish species. Results from both study phases are likely to provide information useful for propagation of rare mussels, reintroduction of host fish, population and reproduction monitoring, habitat restoration and enhancement, and adaptive management.

The abundance of host fish, frequency of parasitism in fish, and frequency of juvenile mussels or glochidia recovered from hatchery-held fish was assessed by collecting fish and mussels at 14 sites distributed among seven streams in central and southeastern Texas. All fish collected and assessed in this study were wild-caught. Qualitative surveys of the resident mussel communities were made, focusing on the five candidate species. A subsample (3 percent in 2012 and 19 percent in 2013) of the fish collected during aquatic biota surveys were submitted to the USFWS San Marcos National Fish Hatchery and Technology Center to collect juvenile mussels or glochidia recovered from host fish, which were held in holding tanks. All fish not sent to the hatchery were assessed for glochidia in the field or in the USGS Texas Water Science Center laboratory in Austin, Tex. Juvenile mussels and glochidia that were recovered from fish at the hatchery were submitted for use in the second phase of this study for the development of deoxyribonucleic acid (DNA) identification keys to determine mussel and host-fish relationships through DNA-based molecular identification (DNA typing of the juvenile mussels and glochidia). Reporting on the results of

DNA-based molecular identification research is beyond the scope of this report.

In 2012, the majority of the fish that were collected, in terms of total number and species types, belonged to the sunfish family Centrarchidae (centrarchids; 1,277 individuals and at least 10 species). Redbreast sunfish (*Lepomis auritus*) was the most common species collected in 2012 (603 individuals), but largemouth bass (*Micropterus salmoides*) were caught at all 10 sites. The largest number of individuals (416) was collected at the Llano Castell site (Llano River at Castell, Tex.) on August 22, 2012, whereas the largest number of species (19) was collected at the San Saba Menard site (San Saba River near Menard, Tex.) on May 22, 2012.

In 2013, the majority of the fish that were collected, in terms of total number and species types, were centrarchids (763 individuals) and cyprinids (10 species), respectively. Blacktail shiner (*Cyprinella venusta*) was the most common species collected in 2013 (287 individuals), but bluegill (*Lepomis macrochirus*) was the only species that was caught at all nine sites. The largest number of individuals (382) and species (19) was collected from the Colorado Columbus site (Colorado River near Columbus, Tex.) on June 11, 2013.

A minimum of two fish (any species) parasitized with glochidia was collected from each of the 10 sites sampled during 2012. The highest percentage of parasitized fish (19.1 percent) was measured at the Guadalupe Victoria site (Guadalupe River near Victoria, Tex.). Parasitized individuals were collected from five families of fish in 2012. The catfish family Ictaluridae (ictalurids) exhibited the highest proportion of parasitized fish (12.1 percent), but the centrarchids had the highest number of parasitized individuals (87).

Nine sites were sampled in 2013, and the Pedernales Fredericksburg site (Pedernales River near Fredericksburg, Tex.) had the highest proportion of parasitized fish at 22.7 percent. The percentage of parasitized fish increased substantially at the Pedernales Fredericksburg site between 2012 (1.4 percent) and 2013 (22.7 percent), likely because the sampled area was relocated. Parasitized individuals were only collected from three families of fish in 2013. Ictalurids exhibited the highest frequency of parasitism (26.5 percent), but centrarchids had the highest number of parasitized individuals (40).

Of the fish that were not sent to the hatchery but assessed for glochidia in the field or in the laboratory in 2012, at least 13 species were parasitized, and longear sunfish (*Lepomis megalotis*) was the species with the highest percentage of parasitized individuals (17.3 percent). Of the fish that were not sent to the hatchery but assessed for glochidia in the field or in the laboratory in 2013, only eight species were parasitized, and flathead catfish (*Pylodictis olivaris*) was the species with the highest percentage of parasitized individuals (42.9 percent). The number of parasitized species based on field and laboratory observations was lower in 2013 than in 2012, but when data from the hatchery are included, the total number of parasitized species in 2013 surpasses the number of parasitized species collected in 2012.

With the exception of the San Antonio Charco site, fish were submitted to the hatchery from all sampling sites in 2013. During the first sampling period in 2013 (April 1–5), slightly more than half (16 out of 29) of the fish species (on a per site basis) that were submitted to the hatchery released juvenile mussels or glochidia (hereinafter juvenile mussels; juvenile mussels and glochidia were not differentiated in hatcheryheld fish), and individuals from 5 of the 16 fish species retained glochidia after completing trials at the hatchery. Largemouth bass collected from the Guadalupe Kerrville site and spotted bass (Micropterus punctulatus) collected from the Guadalupe Victoria site released more than four juvenile mussels per individual on average. Compared to the other sampling periods in 2013, substantially fewer glochidia per fish were present on fish submitted to the hatchery during the second sampling period in 2013 (April 29–May 2); only longear sunfish (collected at the Colorado San Saba site [Colorado River near San Saba, Tex.]) averaged more than one juvenile mussel per fish. None of the fish collected at the Pedernales Fredericksburg and Colorado Nada (Colorado River near Nada, Tex.) sites released juvenile mussels at the hatchery. Although only two sites were sampled during the third sampling period in 2013 (June 10–11), more juvenile mussels were recovered at the hatchery during this sampling period (107) than were recovered during the first two sampling periods in 2013 combined (102). An average of 17 juvenile mussels were recovered per largemouth bass submitted to the hatchery from the Guadalupe Victoria site during the third sampling period, and all four species of fish submitted from the Guadalupe Victoria site released juvenile mussels at the hatchery, which equals an average of 7.3 juvenile mussels

A total of 19 fish species collected at nine sites was submitted to the hatchery in 2013, and 14 of these species released juvenile mussels at the hatchery. The three most productive species, in terms of average juvenile mussels recovered, were longear sunfish, spotted bass, and largemouth bass, each of which averaged more than two juvenile mussels recovered per individual.

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