

# Muleshoe Cooperative Management Area Native Fish Restoration: 2012 Monitoring

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## INTRODUCTION

During 2007 through 2011 four federally-listed fish species were stocked into seven waters within the Muleshoe Ranch Cooperative Management Area (CMA), near the Galiuro Mountains of Arizona (Appendix 1, Figures 1 thru 3; Robinson 2008; Robinson et al. 2010, Robinson and Crowder 2012). Spikedace *Meda fulfida* and loach minnow *Rhinichthys cobitis* were stocked into Redfield Canyon upstream of the confluence with Swamp Springs Canyon. Both species were stocked into Hot Springs Canyon, primarily within a 500-m reach just upstream of the confluence of Wildcat Canyon. Stock of both species originated from Aravaipa Creek, directly so in 2007, but in 2008 thru 2011 from fish propagated at Bubbling Ponds Native Fish Conservation Facility (BPNFCF). Gila topminnow *Poeciliopsis occidentalis* and desert pupfish *Cyprinodon macularius* were stocked into Swamp Springs Canyon, Cherry Spring Canyon, Secret Spring, and Headquarters Spring, and desert pupfish were stocked into Larry & Charlie Tank on the hillside above the Muleshoe Ranch headquarters. Gila topminnow were of Bylas Springs lineage, and originated from Arizona State University in 2007 and from The Nature Conservancy's (TNC) Lower San Pedro River Preserve (Dudleyville) ponds near Dudleyville, Arizona during 2008. Desert pupfish stocked in 2007, 2008 and 2010 originated from TNC's Dudleyville ponds, whereas those stocked in 2009 were acquired from BPNFCF but originated from Dexter National Fish Hatchery (Santa Clara Slough lineage).

The first post-stocking annual monitoring was conducted on September 15 and 16, 2008 (Robinson 2008). Gila topminnow were present in each of the three sites where they were stocked the previous year. Desert pupfish were present in Secret Springs and Swamp Springs Canyon, but none were found in Cherry Spring Canyon. Both loach minnow and spikedace were present in Redfield Canyon and Hot Springs Canyon. Additional fish of all species were stocked into the same waters on September 17, 2008, except that no additional Gila topminnow or desert pupfish were stocked into Secret Spring. Also, a new site, Headquarters Spring, was stocked with desert pupfish and Gila topminnow.

The second post-stocking annual monitoring was conducted on September 14 and 15, 2009 (Robinson et al. 2010). Gila topminnow were captured in all of the sites where they had been stocked. Desert pupfish were only captured in Secret Spring and Cherry Spring. Both loach minnow and spikedace were present in Redfield Canyon and Hot Springs Canyon. On October 28, 2009 more fish were stocked into some of the locations. Gila topminnow had reproduced and had increased in abundance in all of the locations where they had been stocked, so no additional topminnow were stocked in 2009. Source stocks of desert pupfish were low in abundance, so only one site, Larry & Charlie Tank, was stocked in 2009. Stocks of Aravaipa lineage spikedace and loach minnow at BPNFCF produced relatively few offspring, so all were stocked into Hot Springs Canyon, which was thought to have the best and most habitat for the two species.

The third post-stocking annual monitoring was conducted during September 12 thru 14, and October 7 and 8, 2010 (Robinson et al. 2011). Gila topminnow, of both size classes ( $\leq 10$  and  $> 10$  mm TL) were captured in all of the locations where they had been stocked. Desert pupfish were captured in Larry & Charlie Tank, Secret Spring, and Cherry Spring Canyon (one fish), but none were captured in Swamp Springs Canyon or Headquarters Spring. Loach minnow and spikedace were captured in Hot Springs Canyon, but only one loach minnow and no spikedace were captured in Redfield Canyon.

The fourth post-stocking annual monitoring was conducted during September 16 thru 19, 2011 (Robinson and Crowder 2012). Gila topminnow, of two size classes ( $\leq 20$  and  $>20$  mm TL) were captured in three of the four locations where they had been stocked. Desert pupfish were captured in Larry & Charlie Tank, Secret Spring, but none were captured in Cherry Spring Canyon, Swamp Springs Canyon or Headquarters Spring. Loach minnow and spikedeace were captured in Hot Springs Canyon, but only one loach minnow and one spikedeace were captured in Redfield Canyon.

This report summarizes the results of the native fish monitoring within the Muleshoe Ranch CMA during September 2012. The goal of the stocking program is to establish populations within the systems where the species are stocked (i.e., to repatriate the species to the systems). A population is considered to have established (a successful repatriation) when it is reproducing to the point where it is self-sustaining (Griffith et al. 1989, Bright and Smithson 2001, Armstrong and Seddon 2007). The objectives of monitoring were to: 1) verify persistence of fish species post stocking; 2) detect recruitment of young (and hence reproduction) into the population; 3) evaluate if relative abundance (measured as catch-per-unit effort) increases over time (i.e., from the starting point of zero); 4) determine if species have dispersed outside of the stocking area; 5) assess population viability per recovery plans; and 6) report any non-native fish species captured during monitoring.

As originally planned, native fish would be stocked as necessary for up to five years (Bureau of Land Management 1998), at which time each site would be assessed to determine whether or not the species had established a population. Because stockings started in 2007, 2011 was the last year that fish were stocked. Stockings could however continue for population augmentation or genetic maintenance or to establish fish in new locations if agreed upon by the multi-agency team. After stocking, a site needs to be monitored for several years to determine whether or not the species has established a population. For Gila topminnow and desert pupfish, which typically live only 1 to 2 years, two years may be sufficient time to determine if they have established a population. However, AGFD has a three year criterion to determine if a Gila topminnow population has been extirpated (not detected in three consecutive monitoring events), so three years of post-stocking monitoring will be used for these species. Spikedeace and loach minnow typically live about three years, so monitoring for at least three years post-stocking should be sufficient to determine if the species has established a population, because most fish stocked will have died by that time or have grown to adult size. At three years post stocking, any fish captured that is  $< 40$  mm TL would be the result of a fairly recent spawning event, and therefore not a stocked fish.

## **STUDY AREA**

The Muleshoe CMA is located on the southwestern edge of the Galiuro Mountains and west of the Winchester Mountains in southern Arizona. The Muleshoe CMA is jointly managed by U. S. Bureau of Land Management (BLM), U. S. Forest Service, and TNC. The 57,500 acre CMA contains major portions of the Redfield Canyon, Cherry Spring Canyon, and Hot Springs Canyon watersheds, all of which are tributaries to the San Pedro River. The Nature Conservancy's Muleshoe Ranch CMA headquarters is located along Hot Springs Canyon at a location previously known as Hookers Hot Springs.

Before native fish stockings in 2007, fish species reported from Redfield Canyon included Sonora sucker *Catostomus insignis*, desert sucker *Catostomus clarki*, longfin dace *Agosia*

*chrysogaster*, speckled dace *Rhinichthys osculus*, Gila chub *Gila intermedia*, and green sunfish *Lepomis cyanellus* (Griffith and Tiersch 1989, Bureau of Land Management 1998; AGFD Native Fish Database). Gila topminnow were stocked into Redfield Canyon in 1977 (Minckley and Brooks 1985) but did not persist. In Swamp Springs Canyon, longfin dace (SONFISHES and AGFD Native Fish databases) and speckled dace (Bob Rogers, TNC, personal communication) were present. Fish species reported from the Hot Springs Canyon drainage include the same five native fishes found in Redfield Canyon (SONFISHES and AGFD Native Fish databases; Bureau of Land Management 1998). No fish were present in Headquarters Spring, Secret Spring, or Larry & Charlie Tank before stockings in 2007 thru 2009.

Fish monitoring described in this report occurred within the following waters: Redfield Canyon Hot Springs Canyon, and three isolated and unnamed springs (referred to as Secret Spring, Headquarters Spring, and Larry & Charlie Tank) within the Hot Springs Canyon drainage. Fish populations in Swamps Springs Canyon and Cherry Spring Canyon were not monitored in 2012 because of funding issues. Length of perennial water in the streams was previously estimated to be 7.5 km in Redfield Canyon, 2.6 km in Swamp Springs Canyon, 5.1 km in Hot Springs Canyon, and 0.7 km in Cherry Spring Canyon, but during dry periods is likely much less and interrupted (Bureau of Land Management 1998). Secret Spring is an earthen pond below a hillside spring and is located about 600 m northwest of the Muleshoe Ranch headquarters. Headquarters Spring is a pooled area at the edge of but within the Hot Springs Canyon stream bed and is located about 25 m east of and down the hill from the Muleshoe Ranch headquarters' dormitory building; springs originate near the dormitory and drain downhill into the pooled area. During 2009, two small tanks (Larry and Charlie Tanks), about 1 m apart, were dug below a spring located about 25 m west and on the hillside above the casitas at the Muleshoe Ranch headquarters; a shallow (~3 mm) film of water flowed over the ridge between the two tanks. During May 2010 a trench was dug to join the two tanks, which then became known as Larry & Charlie Tank.

Each of the major stream courses was divided into sections (reaches) to facilitate a stratified random sampling design or to encompass stocking locations (Appendix 1, Figures 1 and 2). Reaches in Hot Springs Canyon were: Reach 1 = from Bass Canyon down 1.4 km to Wildcat Canyon, Reach 2 = from Wildcat Canyon down 1.7 km to the second unnamed tributary from the south, and Reach 3 = from the lower end of Reach 2 down 3.7 km to the first unnamed tributary from the south. Reaches in Redfield Canyon were: Reach 1 = from the Swamp Springs Canyon confluence upstream about 1.5 km in Redfield Canyon to the first tributary from the east (the approximate location of the waterfall at UTM coordinates 564212mE, 3590025mN; NAD 83), and Reach 2 = from Swamp Springs Canyon confluence downstream about 2.9 km to an unnamed tributary from the north (561433mE and 3589266mN; NAD 83). Most of Reach 2 is intermittent (Bob Rogers, The Nature Conservancy, personal communication). The two reaches designated in Swamp Springs Canyon were: Lower = from the mouth upstream 1.3 km, but only the uppermost 300 m has perennial water and is sampled, and Upper = an approximately 1.53 km section beginning at an unnamed tributary from the north at about 2.75 km upstream from the mouth, and extending upstream about 100 m past the unnamed tributary from the south that drains Cherry Peak (a small spring is located in this tributary near where it meets Swamp Springs Canyon). The one reach in Cherry Spring Canyon begins at the unnamed tributary containing Cherry Spring and extends upstream for about 2,100 m. The reach is dry except for a small, ~30

m long tinaja about 690 m upstream of the bottom of the reach, and the uppermost 100 m, the latter of which is where topminnow and pupfish were stocked.

## **METHODS**

### **Redfield and Hot Springs Canyons**

#### *Fish Survey*

Annual monitoring of spinedace and loach minnow was completed on October 30 and 31, 2012. Fixed and randomly selected 100-m long sites (transects) were sampled in Redfield Canyon (Figure 1) and Hot Springs Canyon (Figure 2). The three fixed sites in Redfield Canyon and three fixed sites in Hot Springs Canyon were sampled during 2008, 2009, 2010, and 2011 (Robinson 2008, Robinson et al. 2010, Robinson et al. 2011, Robinson and Crowder 2012). A fourth fixed site in Redfield Canyon, located about 2.1 km downstream of the Swamp Springs confluence, was sampled in 2011, but not in other years because it was dry. Four additional sites in Redfield Canyon and six in Hot Springs Canyon were randomly selected in the office by mapping the stream courses (National Geographic TOPO! software), dividing the lengths into 100-m-long segments, and using a random number table to select sites within an identified reach.

Fish were sampled within each 100-m transect using a backpack electrofisher (Smith Root model LR24) moving upstream in a single pass, and stunned fish were netted with 3 mm-mesh dip nets. At the end of each major mesohabitat (pool, run, riffle, cascade) within the 100-m transects, fish were processed and data were recorded. Captured fish were identified to species and counted. All spinedace and loach minnow were measured (mm total length, TL). Other species were counted within two size classes: 20-40 mm and > 40 mm for speckled dace and longfin dace, ≤20 and >20 mm for desert pupfish and Gila topminnow, and 20-100 mm and >100 mm for suckers and Gila chub. Except for topminnow and pupfish, we categorized fish <20 mm TL as larvae. After processing, fish were released alive just downstream from where they were captured. Data recorded for each sampling effort included: site name, site location (GPS coordinates), length of site, date, time, participants, gear type, gear settings, gear dimensions, effort (seconds shocked or length and width of seining or dip netting), species of fish captured, size class of fish, and counts of individuals within each species-size-class category.

#### *Physical Habitat Survey*

Habitat information was typically collected at the 100-m sites. Habitat data recorded included visual estimations of percentage of site composed of each habitat type (cascade, riffle, run, pool), and of each substrate type (clay, silt, sand, gravel, pebble, cobble, boulder). Additionally, because fish were processed at the end of each major mesohabitat type, the length of each mesohabitat type was also recorded. Water quality characteristics measured were: water temperature (°C), pH, and conductivity (µS) using an EXTECH Instruments Inc. ExStik EC500 meter, and dissolved oxygen (mg/L) using an ExStik II dissolved oxygen meter.

### **Muleshoe Headquarters Area Springs**

#### *Fish Survey*

Monitoring for Gila topminnow and desert pupfish was conducted on October 21, 2012 within Secret Spring, Headquarters Spring, and Larry & Charlie Tanks (Figure 2). Fish were sampled using Promar® collapsible minnow traps (0.46 m long x 0.3 m wide, with 2 mm mesh), dip nets (3 mm mesh, 1.5 m long pole), and seines (3 mm mesh, and typically 2 m long by 1.5 m tall).

Collapsible minnow traps were baited with dry dog food. Ten traps were set upon arrival at each site, and were pulled 2-3 h later. An attempt was made to conduct a minimum of 10 dip net sweeps or 10 seine hauls at each site. After each seine haul, dip net sweep, or trap pull, captured fish were held in buckets or the nets in the water until they could be identified, counted and data recorded. After processing, fish were released alive back to the area from which they were captured. Data recorded for each sampling effort included: site name, site location (GPS coordinates), date, time, participants, gear type, gear dimensions, effort (length and width of seining or dip netting, and set and pull times for each trap set from which duration was calculated), species of fish captured, size class of fish ( $\leq 20$  mm or  $>20$  mm), and counts of individuals within each species-size-class category. Water quality characteristics measured were: water temperature ( $^{\circ}\text{C}$ ), pH, and conductivity ( $\mu\text{S}$ ) using an EXTECH Instruments Inc. ExStik EC500 meter, and dissolved oxygen (mg/L) using an ExStik II dissolved oxygen meter.

### **Analyses**

For each water, numbers of fish per size class captured and catch-per-unit effort were calculated. We use catch-per-unit effort (catch rates) as an index of abundance, and not the absolute number of fish captured. Catch rates adjust the number of fish captured by the amount of effort, and so are more correlated with abundance than the total number of fish captured. Catch rates depend on the number of fish present, the number of fish captured, and the amount of and effectiveness of the sampling effort, among other things. Length frequency diagrams for spikedace and loach minnow and length category frequencies for Gila topminnow and desert pupfish were examined to determine if reproduction and recruitment had occurred. Distance between capture location and the stocking reach within Hot Springs Canyon and Redfield Canyon was used as a measure of dispersal distance for spikedace and loach minnow.

## **RESULTS AND DISCUSSION**

### **Redfield and Hot Springs Canyons**

#### *Fish Survey*

Five species of fish were captured in Redfield Canyon during October 2012 (Table 1). Gila chub was the most abundant species captured, followed by Sonora sucker, Gila topminnow, speckled dace, longfin dace, and one each of green sunfish and an unidentified larval fish (Table 2). The one green sunfish was captured in Reach 1 at transect Permanent 2 and was killed and disposed of on site.

No spikedace and loach minnow were captured during the annual monitoring. However, during the March 19, 2012 green sunfish removal effort, the crew observed about 24 fish they thought were spikedace and two fish they thought were loach minnow near the 2010 stocking locations (Clay Crowder, Arizona Game and Fish Department, personal communication). And during the June 12, 2012 green sunfish removal effort, the crew observed about 11 fish they thought were spikedace and three fish they thought were loach minnow near the 2010 stocking locations. Regardless, it is unclear whether or not spikedace or loach minnow persist in Redfield Canyon. Over the years, 12, 20, 0, 1, and 0 spikedace were captured during annual monitoring in 2008 through 2012 respectively, and for loach minnow, 1, 12, 1, 1, and 0 were captured in 2008 through 2012 respectively. For the first few years of monitoring, the numbers captured appeared to be influenced by the numbers stocked the previous year. For example, number of spikedace and loach minnow captured increased from 2008 to 2009, and the number stocked increased

from 2007 to 2008 (in 2007, 213 loach minnow and 192 spikedace were stocked whereas in 2009, 1000 loach minnow and 500 spikedace and were stocked; Appendix 1). In 2009, no fish were stocked, and then in 2010 no spikedace and one loach minnow was captured. However, in 2010 over 730 spikedace and 273 loach minnow were stocked (Appendix 1), but only one of each species was captured in 2011. The decrease in catch from 2009 through 2012 may indicate that the individuals stocked simply did not persist. Also, there was no evidence that spikedace reproduced during that period because all captured were >49 mm TL. There was some evidence that loach minnow had reproduced because the one captured in 2010 was 39 mm TL, and the one captured in 2011 was 35 mm TL, but all 12 captured in 2009 were >47 mm TL. About 33% of Reach 1 was surveyed each year, so it seems unlikely that the decrease in catch was a result of insufficient sampling effort.

Gila topminnow were captured within both transects in Reach 2 (175 in Random 2-1 and 38 in Random 2-2) in Redfield Canyon and many others were observed in isolated pools within that reach. The number captured was much greater than previous years (34 in 2009, 9 in 2010, and 1 in 2011), and electrofishing catch rates were correspondingly greater in 2012 than in 2010 or 2011 (Robinson 2008, Robinson et al. 2010, Robinson and Crowder 2011); they were only captured in dip nets in 2009. Of the 213 topminnow captured in 2012, 81 were categorized into length class with 31%  $\leq 20$  mm TL. Therefore, given that they have been captured for four consecutive years, and multiple age classes were present in 2012, Gila topminnow are now considered established in Redfield Canyon near and downstream of the Swamp Springs confluence. Topminnow entered Redfield Canyon from Swamp Springs Canyon and will likely continue to during floods.

Similar to previous years, seven species of fish, all native, were captured in Hot Springs Canyon during October 2012 (Table 1). Longfin dace was the most abundant species followed by speckled dace, desert sucker, loach minnow, spikedace, Gila chub, and Sonora sucker (Table 1). Catch rates for each species are summarized in Table 2. Numbers of fish captured were substantially greater than in 2011 but similar to 2010. It is possible that the flooding that occurred a few days before the monitoring in 2011 washed fish downstream and caused lower catch rates that year.

Fifty-six spikedace and 85 loach minnow were captured in Hot Springs Canyon (Tables 1 and 2), which is more than captured during any previous year for either species. For spikedace, 4, 24, 29, 4, and 56 were captured in 2008 through 2012 respectively, and for loach minnow, 12, 23, 65, 26, and 85 were captured in 2008 through 2012 respectively. The backpack electrofishing catch rates for spikedace declined from 2009 through 2011, but then increased in 2012 (Figure 3); a variety of techniques were used in 2008, so catch rates for that year were not included in the analysis. In contrast, the electrofishing catch rates for loach minnow increased from 2009 to 2010, then decreased in 2011, and then increased in 2012 (Figure 3). There does not seem to be a direct correlation between the electrofishing catch rates in a given year and the number stocked the previous year (Figure 3). The decrease in catch rates from 2010 to 2011 may have been a result of the flooding that occurred days before sampling occurred in 2011. Robinson and Crowder (2012) hypothesized that another possible reason why fewer loach minnow were captured in 2011 compared to previous years was because the only sampling method used in 2011 was backpack electrofishing, whereas in previous years, riffles had been sampled by a

combination of kick seining and backpack electrofishing; the latter method is more efficient for capturing loach minnow. However, 2012 had the highest catch rates and only single-pass backpack electrofishing was used, so the 2012 data do not support this hypothesis.

All of the spinedace captured in Hot Springs Canyon were greater than 50 mm TL (Figure 4), providing no clear evidence that spinedace reproduced during 2012. However, there was evidence that loach minnow reproduced in 2012 because two were likely age-0 (37 mm TL and 44 mm TL; Figure 4). An additional four loach minnow were between 45 and 49 mm TL, and given that the sampling was completed at the end of October, may also have been age-0.

Our catch information indicates that both species have dispersed downstream of the stocking reach (Reach 1; Figure 2) in Hot Springs Canyon. Similar to 2010 and 2011, spinedace were captured in Reach 3 during 2012, and were captured in all three sampling sites. Similar to previous years, loach minnow were captured in both Reach 1 and Reach 2 (Figure 2).

#### *Physical Environment*

Similar to previous years, mesohabitat in Hot Springs Canyon was mostly riffles and runs during September 2012, (Figure 5). In contrast, Redfield Canyon mesohabitat was comprised mostly of pools with a moderate amount of riffles and few runs (Figure 5). Gradient over 5 km (determined from National Geographic TOPO) is about 1.5% in Hot Springs Canyon and about 1.9% in Redfield Canyon. Substrate types were reflective of the habitat types and gradients in the two streams. Substrate in Hot Springs Canyon was mostly of smaller sizes (sand, gravel, pebbles), whereas substrate in Redfield Canyon was of mostly larger sizes (pebble, cobble, boulder, bedrock; Figure 6). Water quality characteristics were only measured in Hot Springs Canyon and Redfield Canyon and were unremarkable (Table 4).

### **Muleshoe Headquarters Area Springs**

#### *Fish Survey*

Gila topminnow were stocked into two of the three sites that were monitored in October 2012, and were captured in both, whereas desert pupfish, which were stocked into all three of the sites, were only captured in two of the sites. At Larry & Charlie Tank where only desert pupfish were stocked, 11 desert pupfish were captured; mean = 0.4 fish/hr $\pm$ 0.25 SE. One of the 11 pupfish captured was <20 mm TL. Therefore it can be concluded that desert pupfish are persisting and reproducing in Larry & Charlie Tank. Mean catch rates of desert pupfish declined each year from 2010 thru 2012 (Figure 5), indicating the population may be in decline. Alternatively, the fish may for some reason be avoiding the traps.

At Secret Spring, 1,884 Gila topminnow and two desert pupfish were captured (Table 3). Gila topminnow catch rates (61.8 fish/hr $\pm$ 17.61) were lower than previous annual collections, but catch rates have varied considerably over the years (Figure 5). About 20% (381 fish) of the topminnow were <20 mm TL. Similar to what was concluded in previous annual reports, based on the abundance of fish and evidence of reproduction during every annual monitoring since 2008, it can be concluded that Gila topminnow are established in Secret Spring. Similar to previous years, few desert pupfish were captured and minnow trap catch rates remain low (Figure 5). Both of the desert pupfish captured in 2012 were categorized as  $\geq$ 20 mm TL.

Therefore, based on the evidence reported, desert pupfish continue to persist in Secret Spring, but likely have a very small population that may be vulnerable to extirpation.

At Headquarters Spring, 59 Gila topminnow but no desert pupfish were captured in minnow traps during 2012 (Table 3). An additional 61 Gila topminnow, but no desert pupfish, were captured in 24 dip net sweeps. About 47% (56 fish) of the Gila topminnow captured were <20 mm TL. Gila topminnow were stocked into Headquarters Spring in September 2008, and were detected in low numbers during monitoring in 2009, 2010, and 2012 (Figure 5); a few were detected in 2011 about one month after monitoring. Therefore, we conclude that Gila topminnow have established a small population in Headquarters Spring. This is the fourth year that no desert pupfish were captured, even with a second stocking in 2010. Because of the second stocking in 2010, we cannot yet conclude that desert pupfish are extirpated, based on the criteria of Weedman and Young (1995): three consecutive, intensive surveys with no fish captured.

## **RECOMMENDATIONS**

Threatened and endangered fish species were stocked into Muleshoe CMA waters during 2007 thru 2011 (Appendix 1), but for a variety of reasons, not all waters were stocked every year with each species planned for that water. Reasons included the unavailability of stock of some species, the likely extirpation of a species from sites, and the likely establishment of a species at sites. Partners (AZGFD, BLM, BOR, FWS, and TNC) that comprise the Muleshoe Native Fishes Team discussed results after 2011 and agreed that no more fish would be stocked into the sites already attempted, but that monitoring would continue until 2016 to better determine if species had established in those sites. The Nature Conservancy indicated that a new pond could be created near a spring near Bass Canyon, and that site will likely be stocked with desert pupfish. A few other waters that have been discussed in the past, but have still not been stocked with fish, may still be stocked with fish. For instance, Gila topminnow may still be stocked into Wildcat Canyon and possibly in Bass and Double R canyons. One or two locations in Wildcat and Double R canyons may also be suitable for desert pupfish.

To date, the attempt to establish Gila topminnow in waters within the Muleshoe CMA has been successful. Gila topminnow are considered established in Cherry Spring and Swamp Springs canyons (Robinson and Crowder 2012), Secret Spring, Redfield Canyon (dispersed from Swamp Springs Canyon), and Headquarters Spring. With the exception of Headquarters Spring, where the population was nearly decimated by a flood, further stockings of Gila topminnow at these locations are not necessary, except to occasionally introduce fresh genetic material.

The attempted establishment of desert pupfish in most of the same waters was much less successful than for Gila topminnow. Desert pupfish appear to have established a small population in Secret Spring and have likely established a population in Larry & Charlie Tank. However, desert pupfish are extirpated from Swamp Springs Canyon, Cherry Spring Canyon, and Headquarters Spring. It is unknown why desert pupfish failed to establish in these locations, but it could have been a result of negative interactions with Gila topminnow (Robinson and Ward 2011). Habitat, or some aspect of it, may have been unsuitable for desert pupfish. Water quality seems unlikely to be the cause of desert pupfish extirpation because water quality characteristics are within the range of other sites where desert pupfish persist (USFWS 1993). It

may be that the stream environment was unsuitable, because an examination of stocking data revealed that pupfish have not persisted in any stream setting where they have been reintroduced in Arizona (Voeltz and Bettaso 2003, USFWS 2010). It may be that flood events in some of these streams resulted in the extirpation of pupfish. If another pond is dug near a spring near Bass Canyon, it is recommended that only desert pupfish be stocked in that location.

Spikedace and loach minnow may have established populations in Hot Springs Canyon. Both species have been captured during monitoring each year (2008 thru 2012) and putative YOY spikedace were captured in 2010 and putative YOY loach minnow in 2008, 2010, and 2012 indicating reproduction had occurred. However, the two species have been stocked each year from 2007 thru 2011, which has undoubtedly contributed to recruitment. It is unclear whether natural recruitment is sufficient to allow the species to establish and persist. It is unclear if the lack of capture of spikedace <40 mm TL indicates lack of recruitment or that the size class for the species is not vulnerable to electrofishing. Two more years of post-stocking monitoring should provide sufficient evidence of establishment of these species or lack thereof. If multiple size classes of the two species continue to be detected during monitoring thru 2014 then the species can be considered established in Hot Springs Canyon.

It is less likely that spikedace and loach minnow have established populations in Redfield Canyon. Loach minnow were detected during monitoring in 2008 thru 2011 (4, 12, 1, and 1 respectively). None were captured in 2012, but during green sunfish removal efforts in 2012, the crew observed two fish thought to be loach minnow in March, and three thought to be loach minnow in June (Clay Crowder, Arizona Game and Fish Department, personal communication). Putative YOY loach minnow were detected in 2010 and 2011, possibly indicating that reproduction had occurred, although some of the fish stocked during 2010 were small (<20 mm TL), so the one loach minnow captured in 2011 may have been an individual that was stocked the previous year. Spikedace have been detected in three of the five years of monitoring (12 in 2008, 20 in 2009, 0 in 2010, 1 in 2011, and 0 in 2012), but putative YOY individuals were only captured during 2008. However, during green sunfish removal efforts in 2012, the crew observed about 24 fish thought to be spikedace in March and about 11 fish thought to be spikedace in June (Clay Crowder, Arizona Game and Fish Department, personal communication). It is unclear whether natural recruitment is sufficient for the two species to establish populations in Redfield Canyon. In addition, suitable habitat for the two species is limited in Redfield Canyon. The perennial reach where the two species were stocked is only about 1.5 km long, and mesohabitat is mostly pools or runs (Figure 5) in that reach. If neither species is captured for two more years they can probably be considered to have not established populations in Redfield Canyon.

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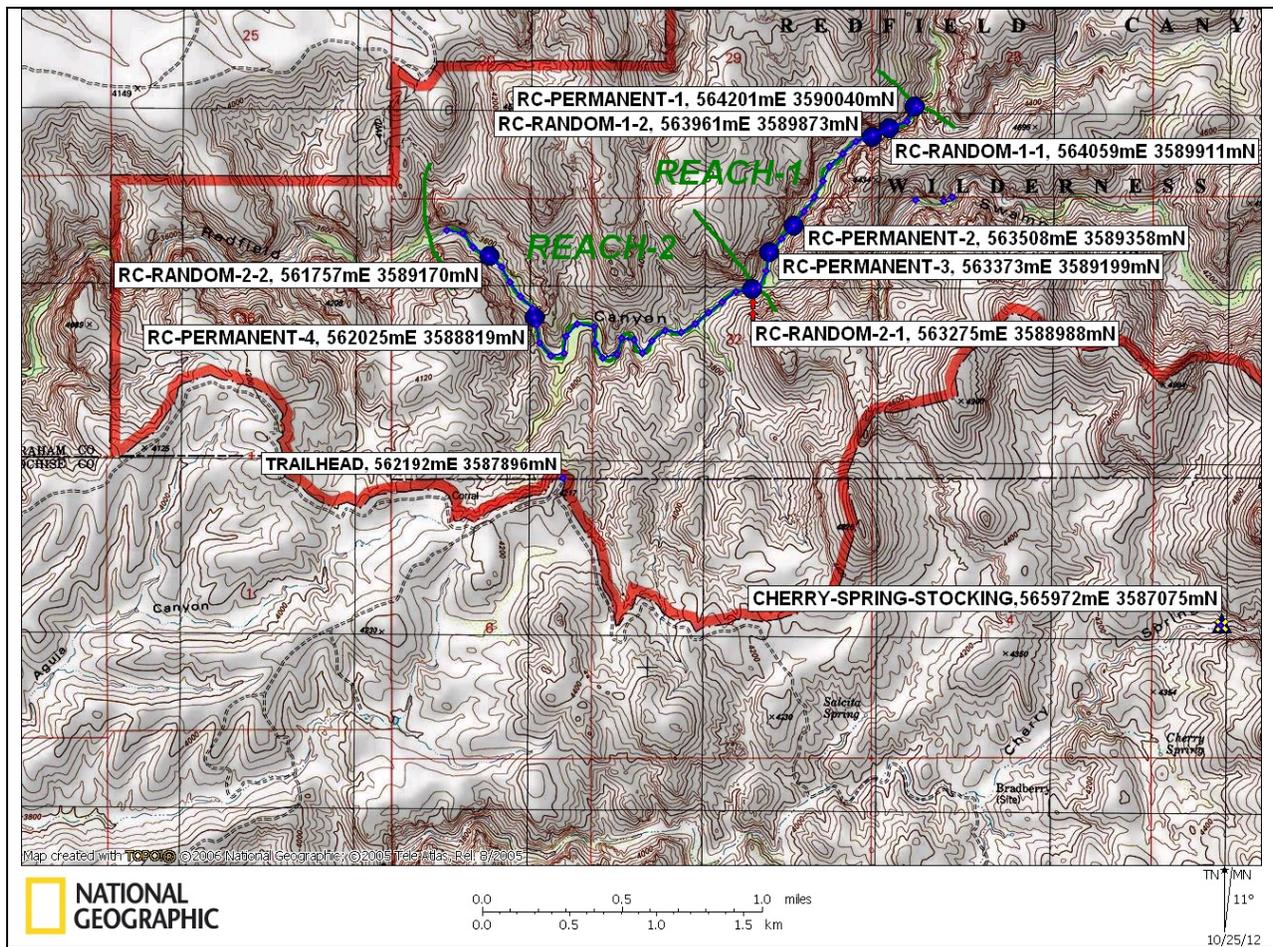


Figure 1. Map showing locations (blue circles) of fish monitoring transects in Redfield Canyon within the Muleshoe Cooperative Management Area, southwest of the Galiuro Mountains, Arizona during 2012. The map shows the locations of permanent and randomly selected 100-m long transects within two reaches (delineated with green lines); spikedeace and loach minnow were stocked into Reach 1 during 2007, 2008, and 2010. The thick red line is the boundary of Redfield Canyon Wilderness.

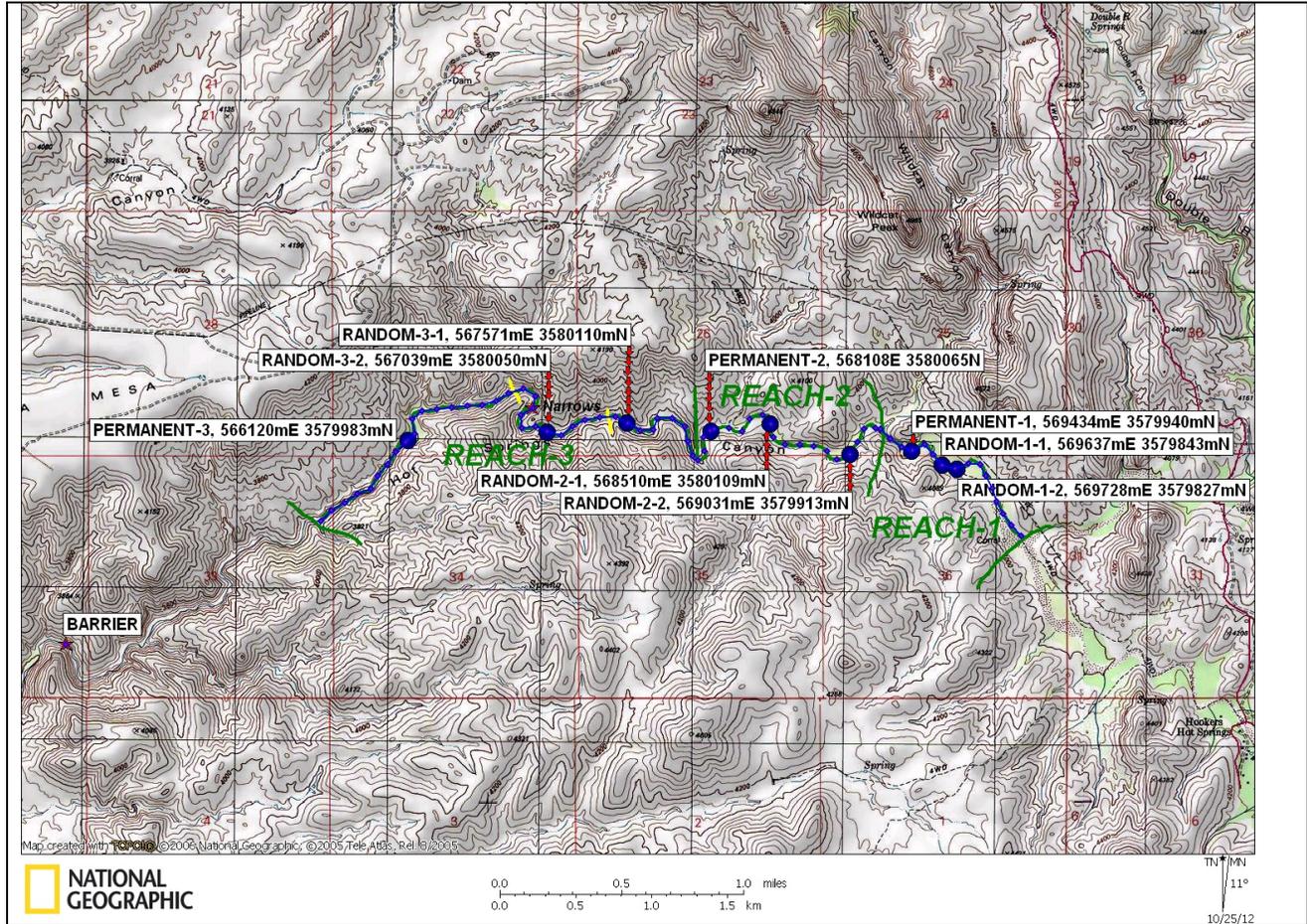


Figure 2. Map showing locations (blue circles) of fish monitoring sites in Hot Springs Canyon and near the Muleshoe Ranch Headquarters (Hookers Hot Springs) within the Muleshoe Cooperative Management Area, west of the Winchester Mountains, Arizona during 2012. In Hot Springs Canyon downstream (north and west) of Bass Canyon, the map shows the locations of permanent and randomly selected 100-m long transects within three reaches (delineated with green lines); spikedace and loach minnow were stocked into Reach 1 during 2007, 2008, 2009, and 2010, and into Reach 1 and 2 in 2011.

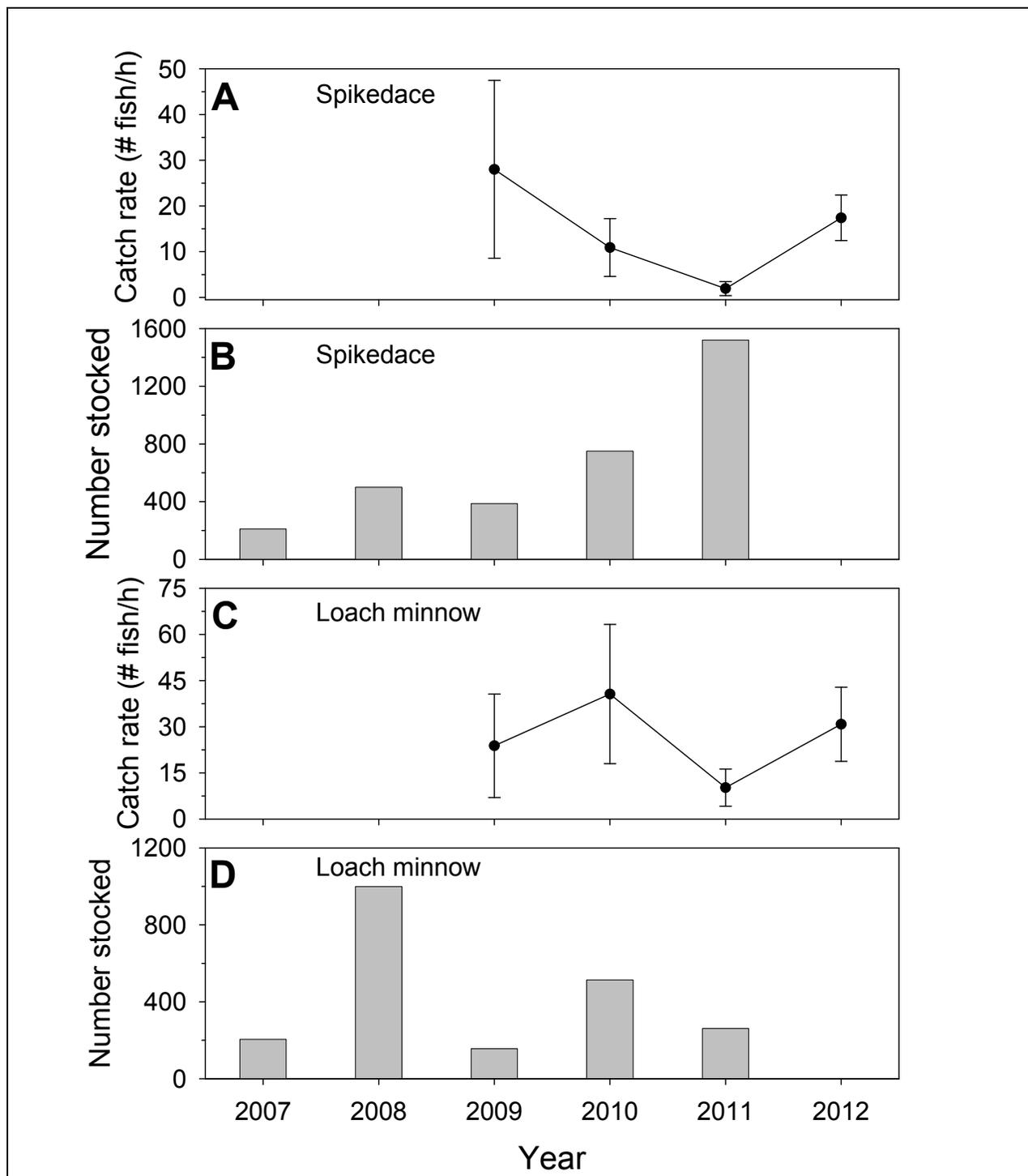
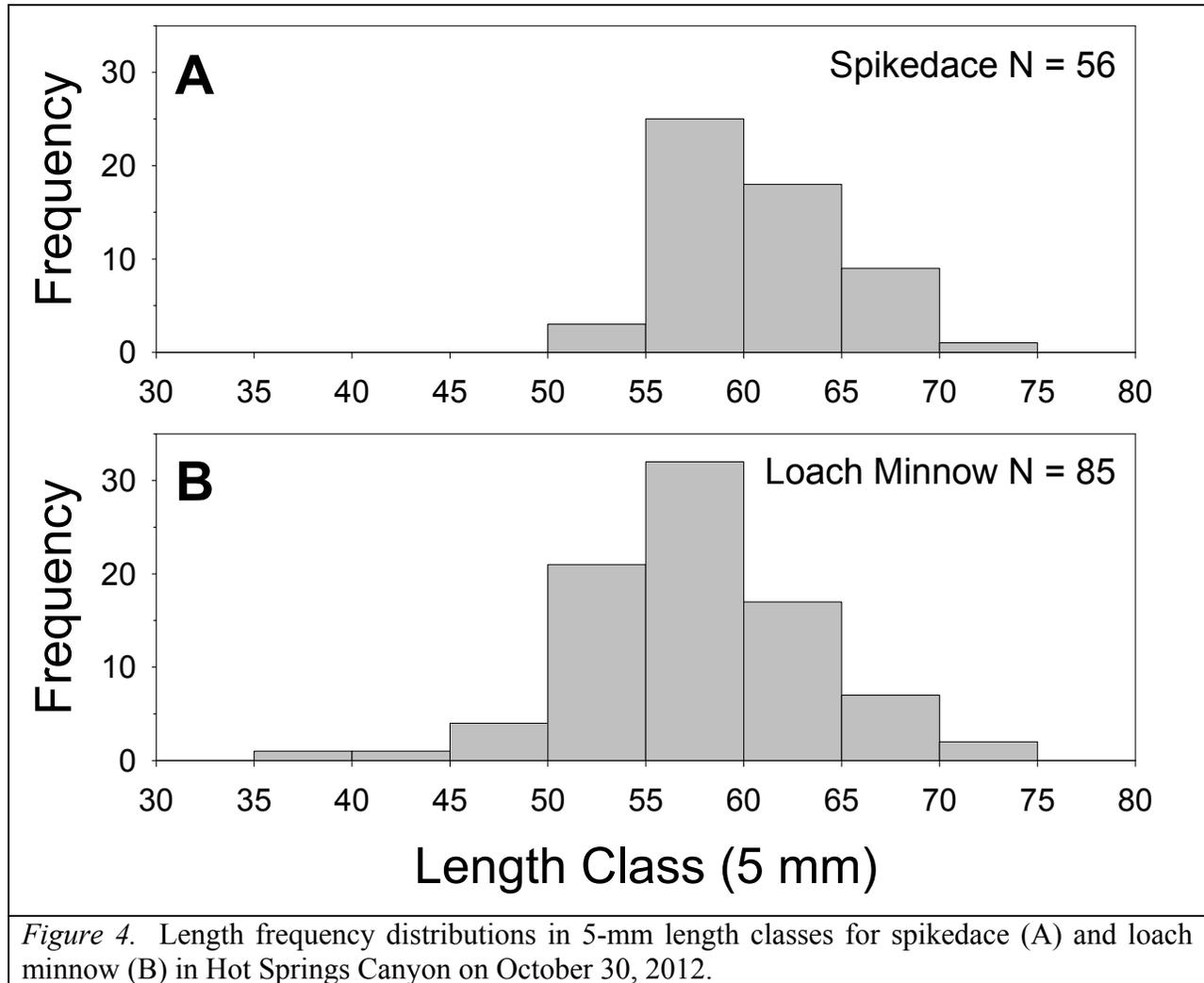


Figure 3. Yearly mean backpack electrofishing catch rates within 100-m transects and numbers of fish stocked in Hot Springs Canyon, Arizona, showing A) spikedace catch rates, B) number of spikedace stocked, C) loach minnow catch rates, and D) number of loach minnow stocked. Note that catch rates for 2009 and 2010 are a combination of upstream single-pass electrofishing and downstream-riffle electrofishing kick seining, whereas catch rates for 2011 and 2012 are just for upstream single pass electrofishing because that was the only method used those years.



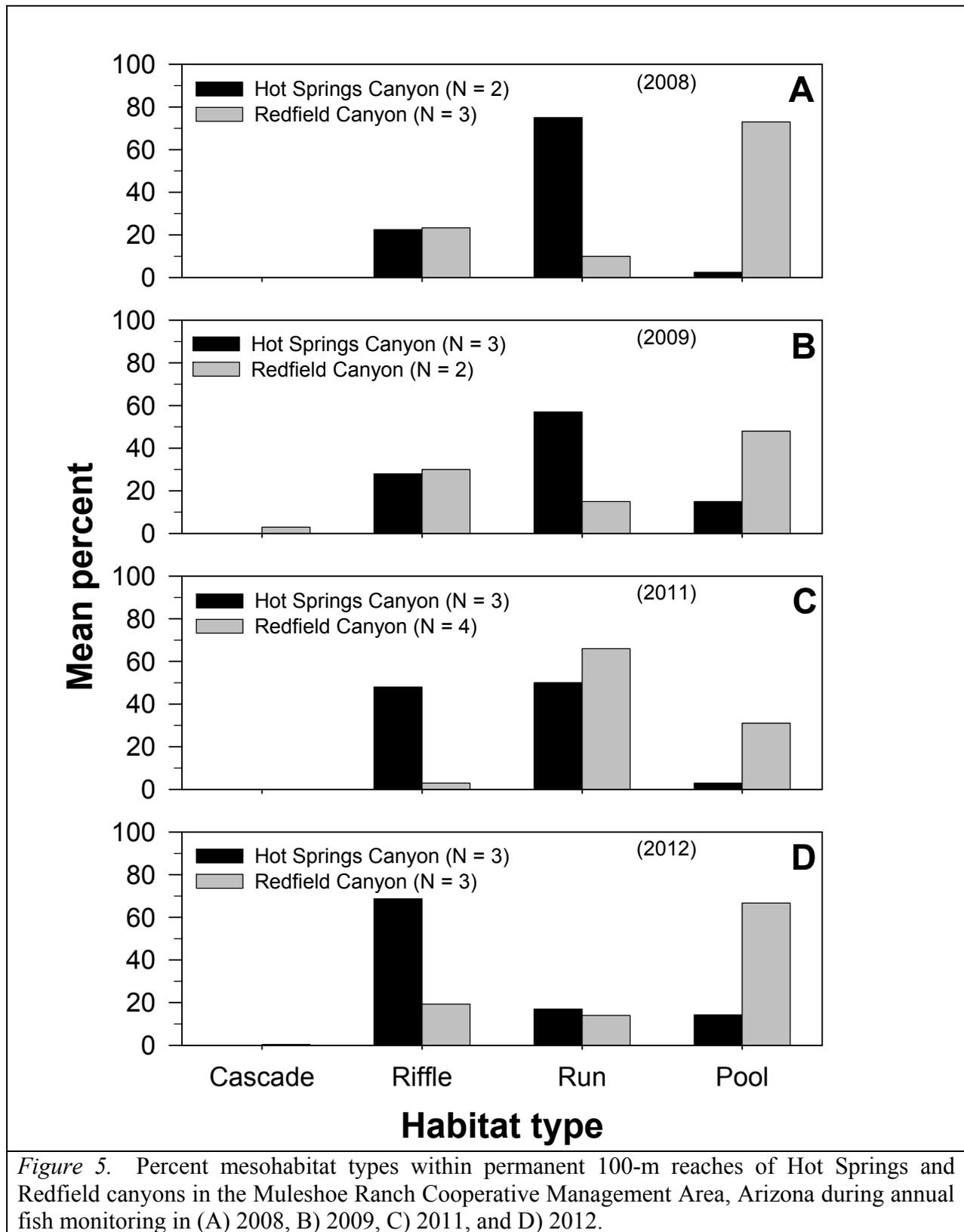


Figure 5. Percent mesohabitat types within permanent 100-m reaches of Hot Springs and Redfield canyons in the Muleshoe Ranch Cooperative Management Area, Arizona during annual fish monitoring in (A) 2008, B) 2009, C) 2011, and D) 2012.

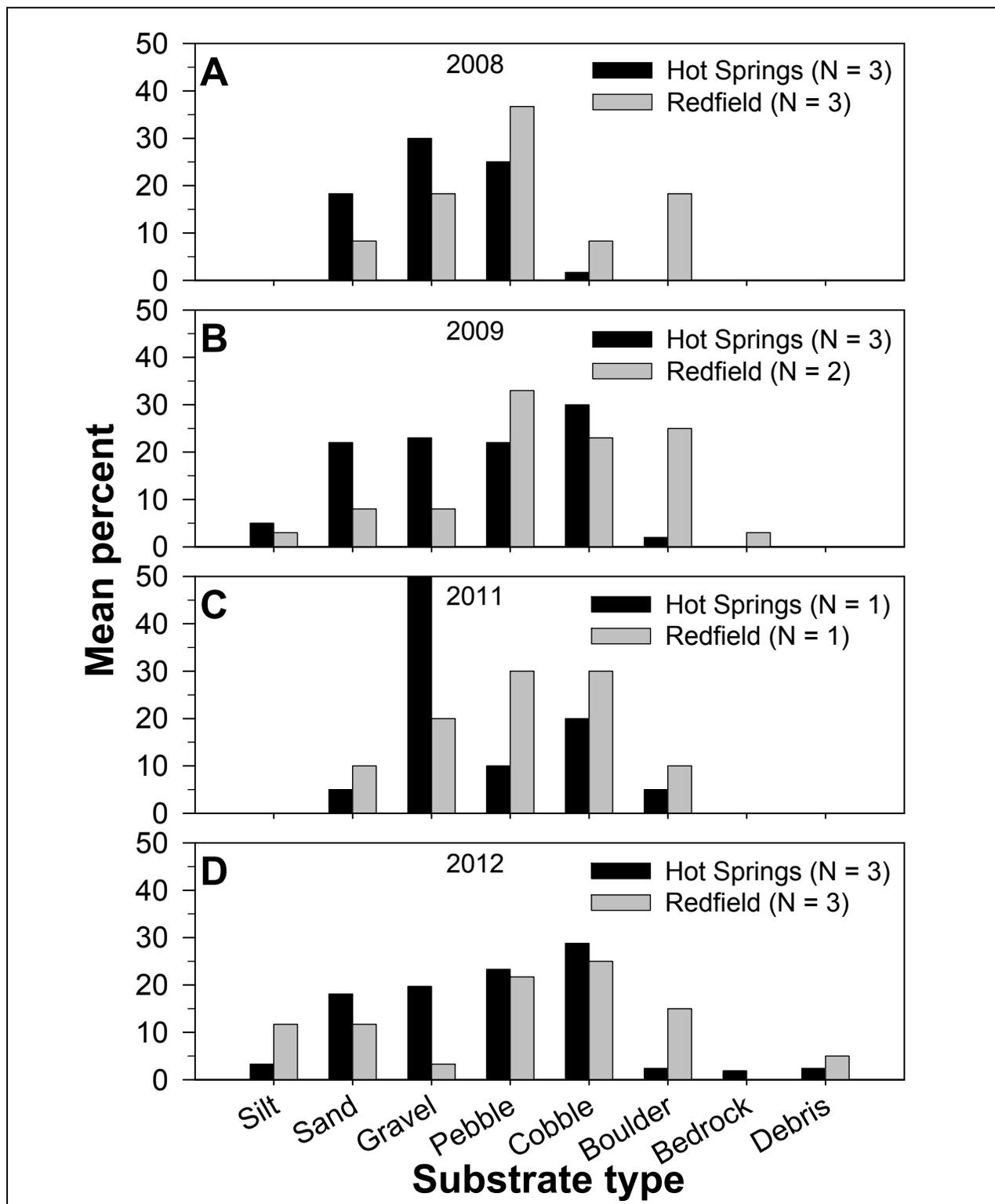


Figure 6. Percent substrate types within permanent 100-m reaches of Hot Springs and Redfield canyons in the Muleshoe Ranch Cooperative Management Area, Arizona during annual fish monitoring in (A) 2008, (B) 2009, (C) 2011, and (D) 2012.

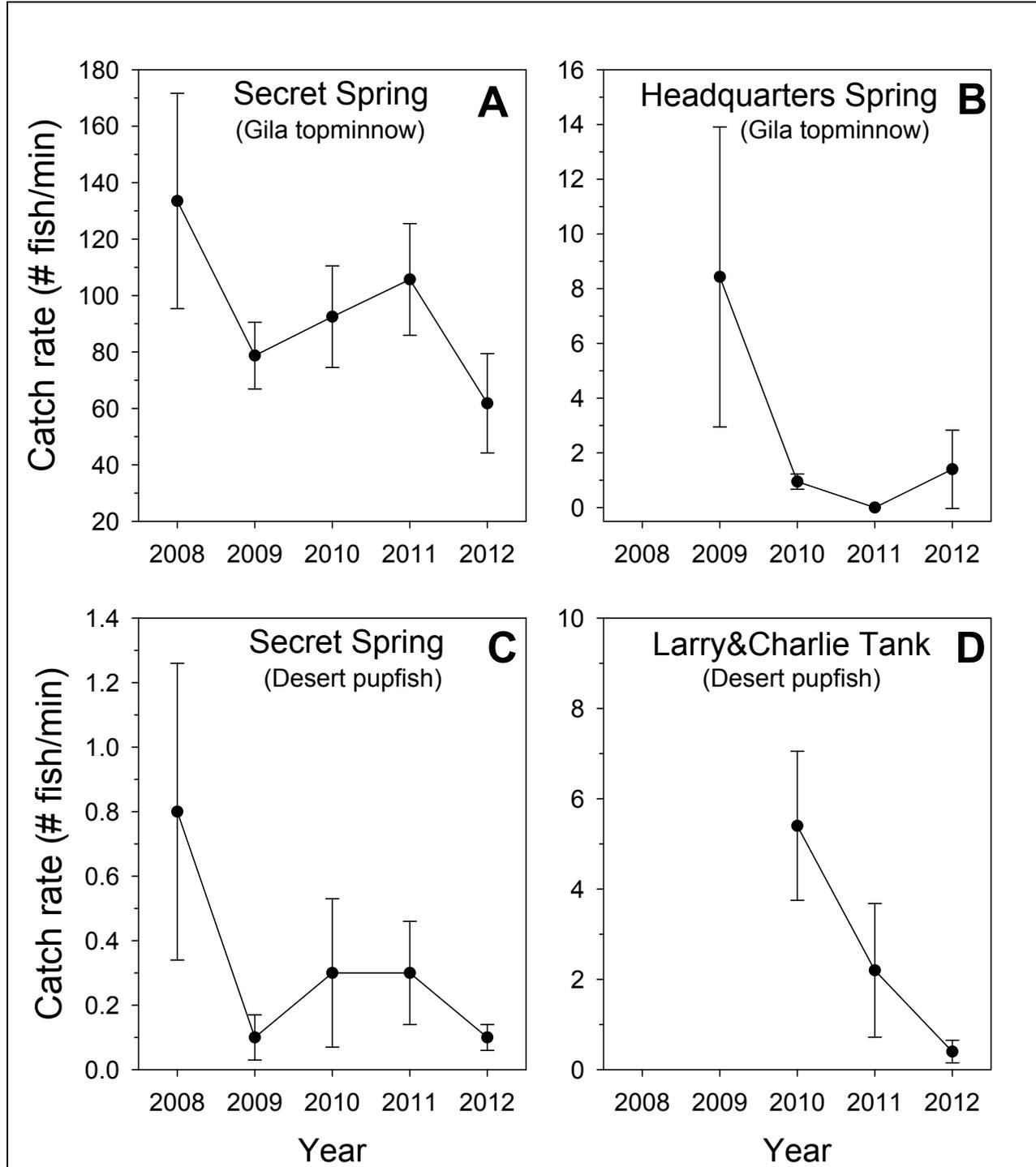


Figure 7. Minnow trap catch rates for Gila topminnow or desert pupfish in three springs within the Muleshoe Cooperative Management Area, Arizona from 2008 through 2012: A) Gila topminnow in Secret Spring, B) Gila topminnow in Headquarters Spring, C) desert pupfish in Secret Spring, and D) desert pupfish in Larry & Charlie Tank.

Table 1. Numbers of fish of each species captured within 100-m backpack electrofishing transects in Hot Springs Canyon and Redfield Canyon, Muleshoe Cooperative Management Area, Arizona during October 2012. Reach descriptions are presented in the Study Area section.

Water	Reach	Transect	Spikedace	Loach minnow	Speckled dace	Longfin dace	Gila chub	Desert sucker	Sonora sucker	Gila topminnow	Green sunfish	Larval	Total		
Redfield Canyon	1	Permanent 1			52			55		50		1	158		
		Permanent 2						8		16			25		
		Permanent 3						25		14			39		
		Random 1-1			58			53		63			174		
		Random 1-2			25			64		71			160		
		Reach total			135			205		214		0	1	556	
	2	Random 2-1			1	13		37		12		175		238	
		Random 2-2										38		38	
		Reach total			1	13		37		12		213		276	
	Stream total	All			136	13		242		226		213	1	1	832
		% Total			16.3	1.6		29.1		27.2		25.6	0.1	0.1	100
	Hot Springs Canyon	1	Permanent 1	9	23	35	22		13	2				104	
			Random 1-1			117	46	3	3					169	
Random 1-2				4	68	9								81	
Reach total			9	27	220	77	3	16	2					354	
2		Permanent 2	17	10	109	66	9	23	4					238	
		Random 2-1	8	12	82	63	4	22	1					192	
		Random 2-2	5	36	85	73		18						217	
		Reach total	30	58	276	202	13	63	5					647	
3		Permanent 3	8		54	82	2							146	
		Random 3-1	4		15	49	4	36						108	
		Random 3-2	5		16	277	14	12						324	
		Reach total	17		85	408	20	48						578	
Stream total		All	56	85	581	687	36	127	7					1579	
		% Total	3.5	5.4	36.8	43.5	2.3	8.0	0.4					100	

Table 2. Number of fish captured (# fish) and mean and standard error (SE) catch rates for backpack electrofishing (#/min) by reach and by stream for Hot Springs Canyon and Redfield Canyon, September, 2011. Reach definitions are presented in the Study Area section. N is the number of transects per reach, from which the mean catch rates were calculated.

Water	Reach	N	Statistic	Spikedace	Loach minnow	Speckled dace	Longfin dace	Gila chub	Desert sucker	Sonora sucker	Gila topminnow	Green sunfish	Total
Hot Springs Canyon	1	3	# fish	9	27	220	77	3	16	2	0	0	354
			Mean #/min	0.2	0.7	7.1	2.1	0.1	0.3	0.0	0.0	0.0	10.6
			SE	(0.18)	(0.41)	(2.50)	(0.80)	(0.08)	(0.24)	(0.04)	(0.00)	(0.00)	(2.18)
	2	3	# fish	30	58	276	202	13	63	5	0	0	647
			Mean #/min	0.5	0.9	4.3	3.2	0.2	1.0	0.1	0.0	0.0	10.1
			SE	(.15)	(0.33)	(0.38)	(0.26)	(0.11)	(0.15)	(0.05)	(0.00)	(0.00)	(0.61)
	3	3	# fish	17	0	85	408	20	48	0	0	0	578
			Mean #/min	0.2		1.1	4.7	0.2	0.6				6.9
			SE	(0.06)		(0.59)	(1.96)	(0.10)	(0.44)				(1.63)
	All	9	# fish	56	85	581	687	36	127	7	0	0	1579
			Mean #/min	0.3	0.5	4.2	3.3	0.2	0.7	0.0	0.0	0.0	9.2
			SE	(0.08)	(0.20)	(1.14)	(0.72)	(0.05)	(0.18)	(0.02)	(0.00)	(0.00)	(1.00)
Redfield Canyon	1	5	# fish	0	0	135	0	205	0	214	0	1	555
			Mean #/min			1.1		1.8		1.9		0.0	4.8
			SE			(0.51)		(0.28)		(0.28)		(0.02)	(0.95)
	2	2	# fish	0	0	1	13	37	0	12	213	0	276
			Mean #/min			0.0	0.4	1.0		0.3	7.1		8.8
			SE			(0.03)	(0.37)	(1.04)		(0.34)	(2.79)		(4.56)
	All	7	# fish	0	0	136	13	242	0	226	213	1	831
			Mean #/min			0.8	0.1	1.6		1.5	2.0	0.0	6.0
			SE			(0.40)	(0.10)	(0.33)		(0.36)	(1.44)	(0.01)	(1.40)

*Table 3.* Number of Gila topminnow and desert pupfish captured (#), mean catch-per-unit effort (#/h) and standard error (SE) of the mean in three waters within the Muleshoe Cooperative Management Area, Arizona during monitoring in October 2012.

Water	# Traps	Statistic	Gila topminnow	Desert pupfish	Total
Headquarters Spring	10	# Fish	59	0	59
		Mean #fish/h	1.4	0.0	1.4
		SE	(1.43)	(.00)	(1.43)
Larry&Charlie Tank	10	# Fish	0	11	11
		Mean #fish/h	0.0	0.4	0.4
		SE	(.00)	(.25)	(.25)
Secret Spring	10	# Fish	1884	2	1886
		Mean #fish/h	61.8	0.1	61.9
		SE	(17.61)	(.04)	(17.64)

*Table 4. Water quality characteristics measured in Hot Springs Canyon (transect Permanent-1) and Redfield Canyon (transect Permanent-1), Muleshoe Cooperative Management Area, Arizona during monitoring on October 30 and 31, 2012.*

Water Quality Characteristic	Hot Springs Canyon	Redfield Canyon
Time (military; hh:mm)	09:21	10:25
Water temperature (°C)	15.9	14.4
Dissolved oxygen (mg/L)		5.63
Ph	7.5	8.12
Conductivity (µS)	321	376
Total dissolved solids (mg/L)		254
Salinity (ppm)		184

*Appendix 1. Summary of species stocked and stocking location information for seven waters within the Muleshoe Cooperative Management Area, Arizona during 2007-2011. Reach definitions are given in the Study Area section.*

Water	Reach	Species	Date	Site name	Meters from bottom of reach	Easting (NAD 83)	Northing (NAD 83)	Number Stocked			
Redfield Canyon	1	Loach minnow	10/4/2007		613	563625	3589507	77			
			10/4/2007		904	563760	3589433	44			
			10/4/2007		1134	563932	3589858	92			
			9/17/2008		1212	564011	3589842	200			
			9/17/2008		1258	564013	3589918	200			
			9/17/2008		924	563768	3589918	120			
			9/17/2008		881	563732	3589744	120			
			9/17/2008		813	563688	3589683	120			
			9/17/2008		746	563674	3589617	120			
			9/17/2008		501	563674	3589414	120			
			10/28/2010		825	563707	3589690	273			
					Loach minnow or spikedace	10/28/2010		825	563707	3589690	279
					Spikedace	10/4/2007		1180	564027	3589881	192
						9/17/2008		924	563768	3589918	100
						9/17/2008		881	563732	3589744	100
			9/17/2008		813	563688	3589683	100			
			9/17/2008		746	563674	3589617	100			
			9/17/2008		134	563354	3589126	100			
			10/28/2010		960	563803	3589781	346			
			10/28/2010		930	563793	3589743	261			
			10/28/2010		915	563784	3589737	123			
Hot Springs Canyon	1	Loach minnow	10/4/2007		160	569319	3579964	205			
			9/17/2008		717	569706	3579826	250			
			9/17/2008		609	569620	3579844	249			
			9/17/2008		476	569592	3579986	250			
			9/17/2008		255	569422	3579933	250			

Water	Reach	Species	Date	Site name	Meters from bottom of reach	Easting (NAD 83)	Northing (NAD 83)	Number Stocked
			10/28/2009		160	569334	3579986	87
			10/28/2009		609	569624	3579847	69
			10/28/2010		465	569590	3579935	130
			10/28/2010		420	569580	3579983	45
			10/28/2010		350	569501	3579998	88
			10/28/2010		215	569378	3579948	63
			10/28/2010		200	569366	3579961	130
			10/28/2010		195	569359	3579961	58
			10/18/2011		180	569384	3579951	99
	2		10/18/2011		323	568224	3580084	162
		Spikedace	10/4/2007		68	569265	3580026	105
			10/4/2007		255	569424	3579941	105
			9/17/2008		717	569706	3579826	125
			9/17/2008		609	569620	3579844	125
			9/17/2008		476	569592	3579986	125
			9/17/2008		255	569422	3579933	125
			10/28/2009		68	569277	3580018	212
			10/28/2009		609	569624	3579847	174
			10/28/2010		465	569590	3579935	73
			10/28/2010		420	569580	3579983	171
			10/28/2010		350	569501	3579998	91
			10/28/2010		215	569378	3579948	197
			10/28/2010		200	569366	3579961	114
			10/28/2010		195	569359	3579961	104
			10/18/2011		225	569413	3579938	176
			10/18/2011		246	569420	3579939	162
			10/18/2011		360	569513	3580017	496
			10/18/2011		314	569458	3579980	30
	2		10/18/2011		381	568282	3580056	656

Water	Reach	Species	Date	Site name	Meters from bottom of reach	Easting (NAD 83)	Northing (NAD 83)	Number Stocked
	2		10/18/2011		178	568078	3580060	485
Swamp Springs Canyon	Lower	Gila topminnow	10/4/2007		1275	564394	3589487	249
		Desert pupfish	10/4/2007		1275	564394	3589487	248
	Upper	Gila topminnow	9/17/2008		4245	566576	3589070	225
			9/17/2008		4267	566603	3589070	50
		Desert pupfish	9/17/2008		4245	566576	3589070	225
			9/17/2008		4267	566603	3589070	48
Cherry Spring Canyon	1	Gila topminnow	10/4/2007	Lower pool	1310	565977	3587082	130
			10/4/2007	Upper pool	1320	565981	3587090	134
			9/17/2008	Lower pool	1310	565977	3587082	275
		Desert pupfish	10/4/2007	Lower pool	1310	565977	3587082	148
			10/4/2007	Upper pool	1320	565981	3587090	98
			9/17/2008	Lower pool	1310	565977	3587082	275
Secret Spring		Gila topminnow	10/4/2007			571100	3578303	499
		Desert pupfish	10/4/2007			571100	3578303	496
			10/28/2010			571100	3578303	311
Headquarters Spring		Gila topminnow	9/17/2008			571624	3577960	275
		Desert pupfish	9/17/2008			571624	3577960	290
			10/28/2010			571624	3577960	374
Larry & Charlie Tank		Desert pupfish	10/28/2009			571603	3577909	196