Gila River Basin Native Fishes Conservation Program:
Arizona Game and Fish Department's Native Fish Conservation Efforts During 2024

Cooperative Agreement R22AC00159
Between Bureau of Reclamation and Arizona Game and Fish Department
Annual Report
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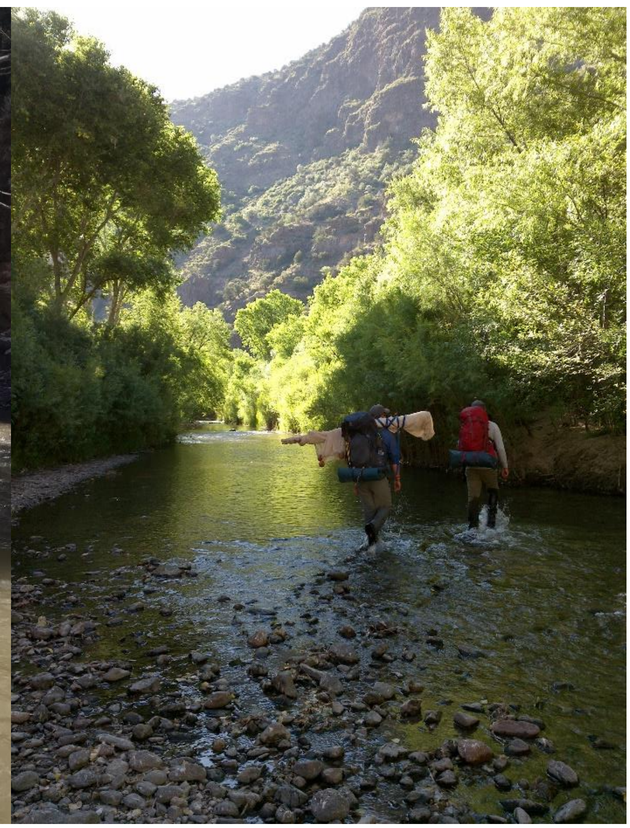

GILA RIVER BASIN NATIVE FISHES CONSERVATION PROGRAM

Program Cooperators:

Arizona Game and Fish Department Mission

To conserve, enhance, and restore Arizona's diverse wildlife resources and habitats through aggressive protection and management programs, and to provide wildlife resources and safe watercraft and off-highway vehicle recreation for the enjoyment, appreciation, and use by present and future generations.

Acknowledgements

The work described in this report was funded through a Cooperative Agreement (R22AC00159) with the U.S. Bureau of Reclamation as part of the Central Arizona Project (CAP) Gila River Basin Native Fishes Conservation Program. Individuals that participated in monitoring, removal, and stocking activities include: Lenna Lough, Brett Montgomery, Skyler Hedden, Cara Waldum, (Department), Betsy Hedden (Reclamation), Neil Dutt (USFS), Scott Bonar, Jessica Rick, Lanie Galland (University of Arizona) and Mark Haberstich (TNC). Individuals from the Department that assisted with fish culture activities at ARCC included Josh Walters, Hudman Evans, Caroline Mallinson, Crystal Castillo, Mattie Diedrick, Trenton Schipper, Dave Rogowski, John Fennell, Crosby Hedden, Ryan Mann, Sarah Taylor, Harley Ough, and Frank Agyagos. We could not have completed the work without their participation and involvement.

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OVERVIEW

The Gila River Basin Native Fishes Conservation Program (Program; previously known as the Central Arizona Project [CAP] Funds Transfer Program) was developed to partially mitigate impacts of the CAP on Threatened and Endangered native fishes of the Gila River basin. The U.S. Fish and Wildlife Service (USFWS) concluded in a 1994 biological opinion that the CAP would be a conduit for transfers of nonnative fishes and other aquatic organisms from the lower Colorado River (where the CAP originates) to waters of the Gila River basin. That opinion identified the spread and establishment of nonnative aquatic organisms as a serious long-term threat to the status and recovery of native aquatic species, following a long history of habitat loss and degradation. Impacts of nonnatives include predation, competition, hybridization, and parasite and pathogen transmission.

The 1994 USFWS opinion concluded that operation of the CAP would jeopardize the continued existence of four native Threatened or Endangered fish species: Gila Topminnow *Poeciliopsis occidentalis occidentalis*, Spikedace *Meda fulgida*, Loach Minnow *Rhinichthys cobitis*, and Razorback Sucker *Xyrauchen texanus*. The Service also concluded that the CAP would adversely modify designated critical habitat of Spikedace, Loach Minnow, and Razorback Sucker. Five reasonable and prudent alternatives were specified: 1) construction and operation of barriers to prevent the spread of nonnative fishes from the CAP to native fish habitats, 2) monitoring of nonnative fish, 3) transfer of funds to USFWS to recover natives, 4) transfer of funds to USFWS to manage nonnatives and research to support that management, and 5) inform and educate the public about native fishes and the impacts caused by nonnative fishes. The transfer of funds under reasonable and prudent alternatives 3 and 4 became known as the CAP Funds Transfer Program. In a 2001 revision of the 1994 opinion, the reasonable and prudent alternatives became conservation measures. In a 2008 revision, the newly-listed endangered Gila Chub[1] *Gila intermedia* and Chiricahua leopard frog *Lithobates chiricahuensis* were added to the Program as species affected by operation of the CAP, and the Santa Cruz River drainage was added to its geographic scope.

The Program is funded by the U.S. Bureau of Reclamation (Reclamation), and is directed by the USFWS and Reclamation in cooperation with the New Mexico Department of Game and Fish (NMDGF) and Arizona Game and Fish Department (Department). Reclamation began administration of Program funding in 2015. The Department and Reclamation finalized an initial agreement (R16AC00077) in 2016, with the present agreement (R22AC00159) initiated in April, 2022. The Program mission is to undertake and support conservation actions (recovery and protection) for federal/state-listed or candidate fish species native to the Gila River basin by implementing existing and future recovery plans for those fishes. There are finalized and approved

[1] In 2016, the American Fisheries Society and the American Society of Ichthyologists and Herpetologists reclassified and merged Roundtail Chub *Gila robusta*, Gila Chub *Gila intermedia*, and Headwater Chub *Gila nigra* into one species, the Roundtail Chub.

recovery plans for four of the five priority species, and a draft recovery plan for the Gila Chub (U.S. Fish and Wildlife Service 1983, 1991a, 1991b, 1998, 2002, 2015). Reports detailing work previously completed by the Department with Program funding from 2007-2023 are listed in Appendix 1.

In addition to the fish and amphibian species specified above, other species mentioned in this report include: Desert Pupfish *Cyprinodon macularius,* Desert Sucker *Pantosteus clarkii*, Longfin Dace *Agosia chrysogaster*, Roundtail Chub *Gila robusta*, Sonora Sucker *Catostomus insignis*, Speckled Dace *Rhinichthys osculus*, Woundfin *Plagopterus argentissimus*, Black Bullhead *Ameiurus melas*, Black Crappie *Pomoxis nigromaculatus*, Bluegill *Lepomis macrochirus*, Brown Trout *Salmo trutta*, Fathead Minnow *Pimephales promelas*, Green Sunfish *Lepomis cyanellus*, Largemouth Bass *Micropterus salmoides*, Red Shiner *Cyprinella lutrensis*, Smallmouth Bass *Micropterus dolomieu*, Western Mosquitofish *Gambusia affinis*, and Yellow Bullhead *Ameiurus natalis*. Other aquatic species mentioned include northern crayfish *Faxonius virilis*, and northern Mexican gartersnake *Thamnophis eques megalops*.

This report summarizes Program work performed by the Department during 2024. For each priority action, work completed during 2024 is presented, followed by recommendations.

PERFORMANCE MEASURES

Cooperative Agreement R22AC00159 between Reclamation and the Department specified the following performance measures.

1. Complete at least three repatriation stockings per calendar year under this agreement, with an overall goal to establish Gila Topminnow into a minimum of 10 surface waters, Loach Minnow into a minimum of 1 surface water, and Spikedace into a minimum of 1 surface water within the five-year performance period.

   Results: During 2024 Department staff completed three repatriation stockings into three waters: Sharp Spring, Hidden Water Spring, and East Fork Sabino Canyon. (Appendix 2).

2. Control or eradicate non-native fish in at least one surface water per calendar year under this agreement, with an overall goal to eradicate or control non-native fish at a minimum of five unique surface waters within the five-year performance period.

   Results: During 2024, Department staff controlled nonnative fish at Harden Cienega Creek, Redfield Canyon, and within the Ash Canyon watershed.

3. Maintain and propagate populations of Loach Minnow, Spikedace, and other rare species at the ARCC (formerly the Bubbling Ponds Native Fishes Conservation Facility)

throughout the five-year performance period unless stakeholders determine that there is no longer a need for captive refuge and propagation at the facility. Also annually acquire Spikedace, Loach Minnow, and other rare species from the wild to maintain broodstocks of each lineage at ARCC.

Results: During 2024, Department staff maintained and propagated three lineages of Spikedace (Aravaipa Creek, upper Gila River, and Gila Forks), four lineages of Loach Minnow (Blue River, Aravaipa Creek, San Francisco River, and Gila Forks) one lineage of Roundtail Chub (Eagle Creek), and one putative lineage of Gila Topminnow (Parker Canyon).

GENERAL ACTIVITIES

Department staff administered and managed Program projects identified in the agreement. Department staff finalized the 2023 annual report, began analyzing data and drafting the 2024 interim and annual reports, revised the FY25 workplan and drafted the FY26 work plan. Staff coordinated with intra-agency staff, other agencies, and private landowners to continue work on existing projects and to develop potential new projects. Staff revised and updated electronic data entry forms and corresponding formatting and summary scripts, entered data into survey and stocking datasets, and checked data for accuracy. Staff purchased and maintained equipment as necessary to carry out fieldwork obligations. Department staff completed the hiring process for the vacant Gila River Basin native fish biologist position. Staff also completed mandatory employee training.

PRIORITY ACTIONS

General Methods

Fish Stockings: The Department coordinates with USFWS regarding locations to stock and sources and lineages of fish appropriate for each translocation. Fish for translocations were collected, transported, and stocked according to Department fish collection, transport, and stocking protocols (best management practice #4; AGFD 2011), and Hazard Analysis and Critical Control Point (HACCP) practices. Fish were collected from select waters inhabited by target lineages using gear appropriate for the given water; typically seines, minnow traps, or electrofishing. Fish were placed into aerated 5-gallon buckets from which they were sorted to confirm species identity and assess condition. Fish were then transferred into transport coolers (100 qt. minimum) equipped with aerators and filled with well water treated with salt and Amquel®. At the translocation site, fish were transferred from the transport cooler back to aerated 5-gallon buckets and carried to the stocking location. Water quality characteristics in the buckets and the stocking location were measured. Conductivity (μS), salinity (mg/L), total dissolved solids (mg/L), pH, and water temperature (°C), were measured using a Hach® Combo meter, and dissolved oxygen (mg/L) using a Sper Scientific® dissolved oxygen meter. Fish were acclimated to stocking site conditions by exchanging 25 to 50% of transport bucket water with stream water, about every 10 minutes, until bucket temperatures are within two degrees of the stream. Fish were sorted a final time to verify species identity, assess condition, and determine a final count before being released into the stocking site. Data recorded for stocking include: site name, date, time of arrival and stocking, participants, type of transport container, water quality in the tanks and site, counts of individuals stocked, condition of fish, fish behavior after release, and number of mortalities.

Fish Surveys: Backpack electrofishing was used at 100 m sub-reaches to survey translocated populations of Spikedace, Loach Minnow, and Roundtail Chub[2], and to assess habitats for fish translocations. The number of sub-reaches sampled was determined by length of target reach, with a minimum of three sub-reaches for short reaches and a goal of at least 10% of the reach length in longer streams (i.e., there are fifteen 100 m sub-reaches in the 14.6 km of the upper Blue River). A backpack electrofisher (Smith-Root; Model LR24) was used to electrofish upstream through each sub-reach in a single pass. Three-pass depletion was carried out between two block nets at select fixed sub-reaches to estimate abundance and capture probability. Stunned fish were netted with dip nets (tear-drop shaped, 0.43 m x 0.37 m with 2 or 3 mm mesh). At the upstream end of each major mesohabitat type (pool, run, riffle, or cascade) within each sub-reach, fish were processed and data were recorded. Captured fish were identified to species and counted. All Spikedace, Loach Minnow, and Roundtail Chub were measured to the nearest millimeter in total length (mm TL). Other species were counted within two size classes for small bodied fishes (≤40 and >40 mm TL for Speckled Dace and Longfin Dace; <20 and ≥20 mm TL for Desert Pupfish and Gila Topminnow) and three size classes for large bodied fish (<50, 50-100, and >100 mm TL; e.g., Sonora Sucker). After processing, fish were released alive just downstream from where they were captured. Fish were released a short distance below the downstream block net during depletion efforts. Data recorded for each sampling effort included: site name, site location (UTM NAD83), length of site, date, time, participants, gear type, gear settings, gear dimensions, seconds shocked, species of fish captured, size class of fish, and counts of individuals within each species-size-class category.

Minnow traps or hoop nets baited with dry Gravy Train® dog food were used to survey for Gila Topminnow, Desert Pupfish, and some Roundtail Chub[3] populations. Promar® collapsible minnow traps (0.46 m long x 0.3 m wide, with 2 mm mesh) were used for Gila Topminnow and Desert Pupfish monitoring, whereas Promar® collapsible mini-hoop nets (0.85 m long x 0.3 m diameter circular hoops, with 9 mm mesh) were used for Roundtail Chub monitoring. Typically, a minimum of 10 traps were set in each location for a minimum soak time of two hours, and fish were processed and released alive back to the location of capture. Data recorded for each sampling effort includes: site name, site location (GPS coordinates, UTM NAD83), date, time, participants, gear type, gear dimensions, set and pull times for each trap set, species of fish captured, size class of fish, and counts of individuals within each species-size-class category.

[2] Including chub populations previously classified as Gila Chub.
[3] Including chub populations previously classified as Gila Chub.

Evaluation of Species Establishment: The goal of translocation efforts is to establish populations of Spikedace, Loach Minnow, Gila Topminnow, and Roundtail Chub to contribute to recovery of these species. A species is considered to have established (a successful translocation) when it is reproducing to the point where it is self-sustaining (Griffith et al. 1989, Bright and Smithson 2001, Armstrong and Seddon 2007). Similarly, the Spikedace recovery plan (USFWS 1991a) describes criteria for establishment with characteristics of abundance, age-class structure, and recruitment in the range of natural variation. To assess this goal, post-stocking monitoring data were collected for each translocated species to evaluate species presence, an index of abundance, population size structure, and dispersion. Arguably, the two most important of these four measures for determining if a species has established are population size structure and an index of abundance.

The objectives of monitoring are to:

1. determine presence of translocated fish species and nonnative fish species;
2. evaluate trends in relative abundance (estimated as catch-per-unit effort) of the translocated species, sympatric native fishes, and nonnative fishes;
3. evaluate size-structure of each population of fish species to detect reproduction and recruitment to the population;
4. determine if translocated species have dispersed from the stocking area.

Presence of individuals during post-stocking monitoring is evidence that the species has persisted. Presence of juvenile fish is evidence of reproduction, and the proportion of the population that are juveniles is evidence of year-class strength. Size structure is used as an indicator of age-structure. Presence of age-0, age-1, and older size classes for several years in a row, and consistently high catch rates for several years in a row is an indication that a population has established. Capture of individuals beyond stocking locations is evidence of dispersal.

After stocking, sites are monitored for several years to determine whether or not the translocated species has established a population. The duration of monitoring varies by target species, and generally exceeds the life span of the species by at least one year. Two years may be sufficient to determine if Gila Topminnow and Desert Pupfish, which typically live only one to two years, have established a population. However, if no fish are detected in three consecutive monitoring events, the population may be considered extirpated (Weedman and Young 1995). Therefore, three years of post-stocking monitoring are used for Gila Topminnow and Desert Pupfish. Spikedace and Loach Minnow have a longer potential lifespan (three to four years), and five years of post-stocking monitoring should be sufficient to determine if the species has established a population. Roundtail Chub[4] typically live about seven years. However, a yearly examination of size structure for five years after stocking is likely sufficient to determine if Roundtail Chub are established. Translocated populations should be monitored periodically after establishment by one or more of the cooperators for at least 10 years to determine population persistence and viability.

[4] Including chub populations previously classified as Gila Chub.

Nonnative Piscivore Removal: Nonnative fishes were typically removed using traps and electrofishing. A variety of traps were used, depending on habitat size: mini-hoop nets (Promar® TR-502 collapsible traps; cylindrical, 0.85 m long x 0.3 m wide, with 9 mm mesh) and minnow traps (Promar® collapsible minnow traps; 0.46 m long x 0.3 m wide, with 2 mm mesh) baited with dry dog food (Gravy Train®). Traps were dispersed throughout the targeted reach and were primarily set in pools or runs that were more than 1 m deep. Traps were retrieved 2 to 22 hours later. For backpack electrofishing, typically the entire targeted reach was shocked, and any nonnative fish captured were removed. A single full pass is defined as electrofishing all water present from the downstream end to the upstream end of the target reach. An initial set of traps in the target reach is considered the first pass, with each reset within the same reach considered a subsequent pass. Nonnative fish are typically measured to the nearest millimeter in total length (mm TL) to assess size structure of the target population.

Evaluation of Nonnative Fish Removal: There are two general goals for nonnative fish removals: suppression or eradication. For situations where barriers to nonnative fish invasion do not exist, the goal is to suppress nonnative populations until barriers can be installed. When barriers to nonnative fish invasion are in place, the goal is eradication. Multiple removals are conducted until goals are achieved. The catch of nonnatives across removal events is examined, and a decrease in abundance of the target nonnative species to low levels or to zero is evidence of control. Absence of target nonnative fishes confirmed by eDNA sampling is evidence of eradication.

Statistical Analyses:
Relative abundance of select fish species in terms of catch per unit effort (CPUE) was calculated as fish per hour (/h or ind/h) of electrofishing effort, soak time of passive gears (i.e., minnow trap, hoop net, gill nets), or fish per square meter sampled for active gears (i.e., dip net, straight seine). Abundance of all fish species encountered at sub-reaches where depletion sampling occurred was estimated via the Carle-Strub method using the "removal" function in the *FSA* package (Carle and Strub 1978, Ogle et al. 2023). Differences in mean relative abundance of native fish between years were evaluated using multiple tests, depending on how data were distributed. The Shapiro-Wilk test of normality (*stats* package) was conducted to determine whether data were normally distributed. A two sample t-test was used for normally distributed data (*stats* package). The non-parametric Wilcoxon rank-sum test was used for non-normal distributed data (*stats* package). Two-sided tests were used for taxa where there was not an obvious change in mean relative abundance between years, and one-sided tests were used for taxa where mean relative abundance appeared to differ between 2022 to 2023. Pearson's product-moment correlation coefficient was used to evaluate trends in mean relative abundance between species pairs across years (*stats* package). All statistical analyses were conducted within Program R (R Development Core Team, 2023).

1. Muleshoe ecosystem stream and spring repatriations (Task AZ-2003-1)

Strategic Plan Goals:

- Preventing Extinction and Managing Toward Recovery
    - Goal 3. Remove nonnative aquatic species threats.
    - Goal 4. Replicate populations and their associated native fish community into protected streams and other surface waters.
    - Goal 7. Monitor to quantitatively measure and evaluate project success in improving the status of target species and their habitats.

Recovery Objectives:

- Spikedace recovery objective 6.4. Monitor success/failure of reintroductions.
- Loach Minnow recovery objective 6.4. Monitor success/failure of reintroductions.
- Gila Topminnow 1999 draft revised recovery plan objective 2.4 Protect habitats of reestablished or potential populations from detrimental nonnative aquatic species.
- Gila Topminnow 1999 draft revised recovery plan objective 3. Monitor natural and reestablished populations and their habitats.
- Gila Chub draft recovery plan objective 1.3.1. Eliminate or control problematic nonnative aquatic organisms
- Gila Chub draft recovery plan objective 2. Ensure representation, resiliency, and redundancy by expanding the size and number of populations within Gila Chub historical range via replication of remnant populations within each RU.
- Gila Chub draft recovery plan objective 7. Monitor remnant, repatriated, and refuge populations to inform adaptive management strategies.

Background: The purpose of this sub-project is to establish Spikedace, Loach Minnow, Gila Topminnow, and Desert Pupfish into various waters on the Muleshoe Ranch Cooperative Management Area (CMA). The Muleshoe Ranch CMA is located on the western slopes of the Winchester and Galiuro mountains. The various waters and stream reaches previously included in this subproject are described in Love-Chezem et al. (2015). Fish stockings began in 2007, when Spikedace and Loach Minnow were stocked into Hot Springs Canyon and Redfield Canyon. Both Spikedace and Loach Minnow failed to establish in Redfield Canyon; however, Gila Topminnow have dispersed downstream from Swamp Springs Canyon and are now established in Redfield Canyon. In 2007, Roundtail Chub[5], Sonora Sucker, and Speckled Dace were translocated upstream of a waterfall in Redfield Canyon to expand their range in the system.

Green Sunfish control in Redfield Canyon started in 2007 and has been conducted annually since initiation. The number of sunfish removed from Reaches 1 and 2 has remained low, and far more sunfish are removed from Reach 3 each year since concerted efforts began there in 2014.

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[5] Chub in Redfield Canyon were previously classified as Gila Chub.

Results: During April 22-23, 2024, Department staff, assisted by University of Arizona researchers, removed Green Sunfish from Redfield Canyon. The crew began electrofishing in Reach 2 near where perennial flow begins in most years, downstream of Swamp Springs Canyon (UTM 562933/3599775), upstream to the waterfall barrier (563872/3589779; Figure 1.1). The crew electrofished for a total of 5,516 seconds but did not capture or observe any Green Sunfish (Figure 1.2). The crew also set six mini-hoop nets in the pool that has supported nearly all of the Green Sunfish captured in Reach 1 the last few years and captured three Sonora Sucker.

Department staff also completed three removal passes in Reach 3. The crew set 10 mini-hoop nets for three consecutive two hour sets in the pools near the wilderness boundary. A total of 51 Green Sunfish were captured during the first set, 3 during the second set, and 5 during the third set for a total of 59 across all three sets. The crew also angled while the traps soaked and captured one additional Green Sunfish and two Roundtail Chub[6]. The range of Green Sunfish total length was similar to 2023, with fish from 61 to 208 mm captured (Figure 1.3)

The number of sunfish removed in Reach 3 is the second lowest total since targeted efforts began in this reach in 2014, with fewer sunfish removed only in 2023. Size structure of Green Sunfish captured in Reach 3 also shifted to a significantly smaller mean size in 2024 (111.52) compared to 2023 (128.70, $W = 877$, $P = 0.029$), although it appears that the shift was driven by the smaller proportion of fish from 120-200 mm TL captured in 2024.

This is only the third time that Green Sunfish have not been captured in Reach 1 or 2 during removal efforts dating back to 2007, with the other two years being 2015 and 2021. Our results seem to suggest that suppression efforts, likely in combination with substantial variation in environmental conditions over the last few years (i.e., record dry and record wet seasons), are effectively meeting our objectives of preventing sunfish from spawning in Reaches 1 and 2, and collapsing the accessible portion of the sunfish population in Reach 3 with only a single removal effort each year.

Recommendations: Department staff will continue to contact the downstream private landowners in Redfield Canyon and attempt to gain permission to access the property and remove sunfish from Reach 3. If permission is granted, the goal of Green Sunfish removal efforts should shift from suppression to eradication, and the frequency and intensity of removal efforts should be increased. If the downstream landowners do not grant permission for access, eradication of Green Sunfish in Redfield Canyon will likely not be feasible. The current level of removal effort (1 removal per year) appears to be sufficient at suppressing the sunfish population in Reaches 1 and 2 and should be continued until the status of the downstream population changes.

[6] Chub in Redfield Canyon were previously classified as Gila Chub

Tables and Figures:

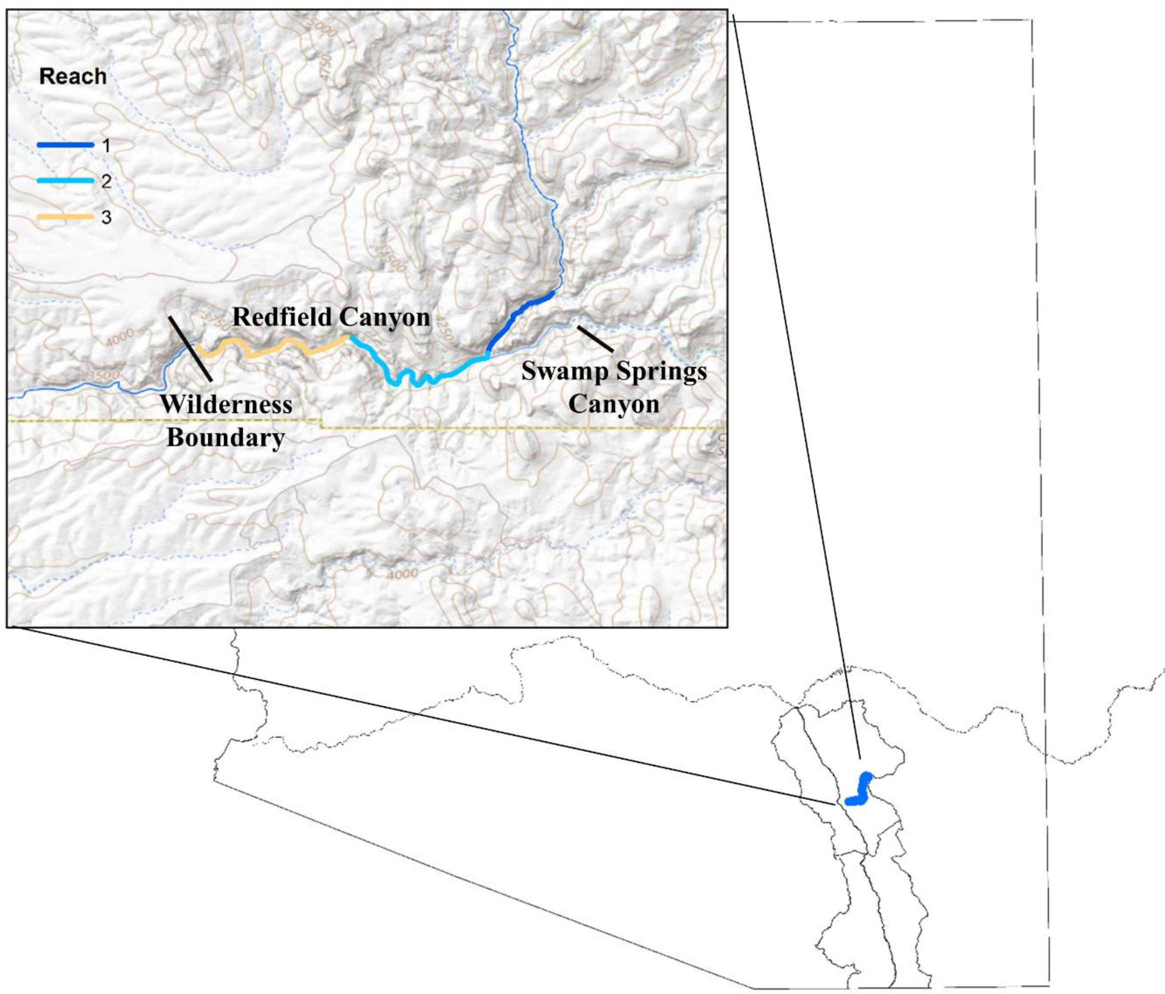

Figure 1.1—Location of Redfield Canyon within the Gila River Basin and San Pedro River sub-basin. Inset map shows the location of sampling Reaches 1 (Swamp Springs Confluence upstream to Barrier), 2 (Rock House tributary upstream to Swamp Springs Confluence), and 3 (Wilderness Boundary upstream to Rock House tributary).

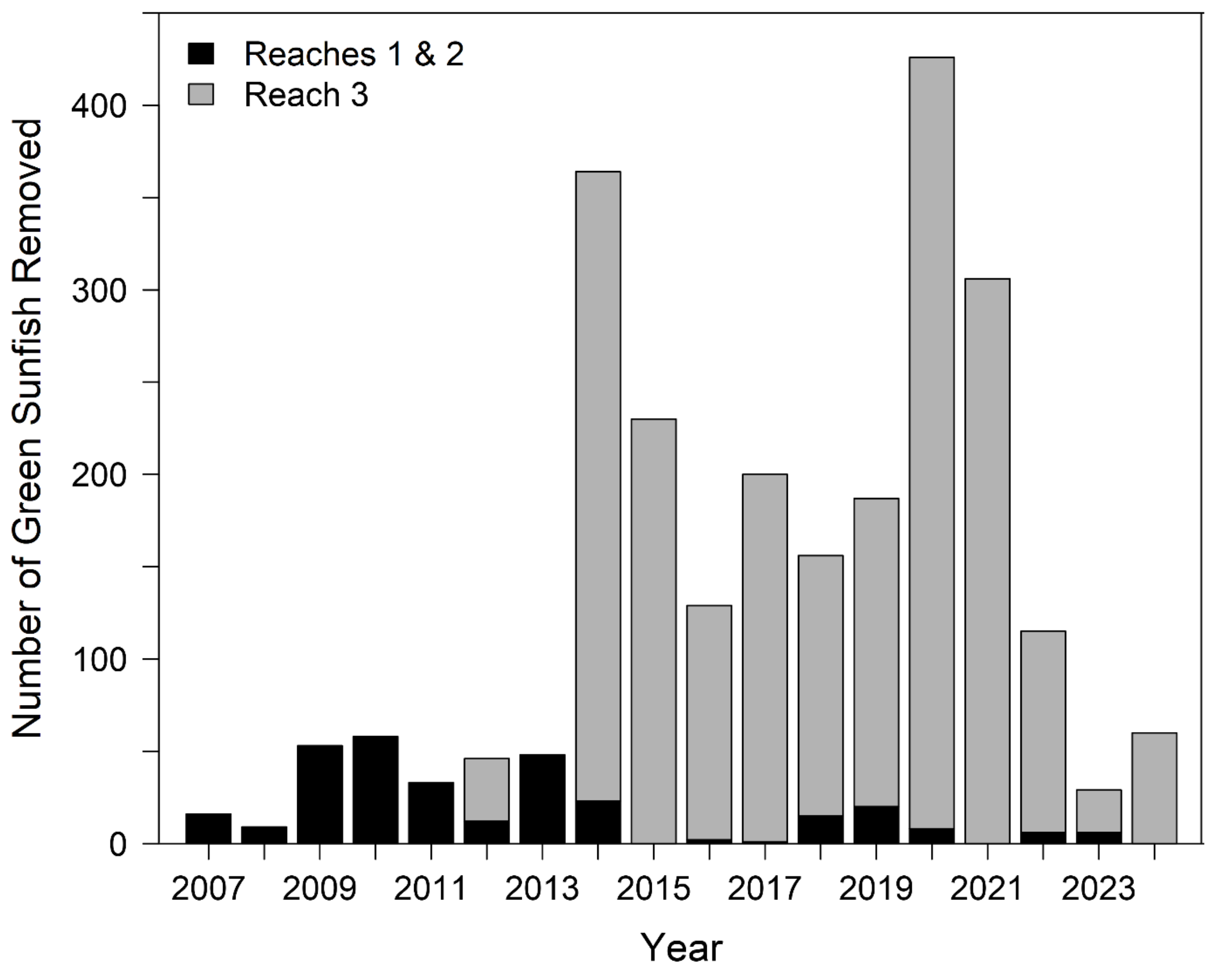

Figure 1.2.—Number of Green Sunfish removed during annual spring removal efforts and autumn monitoring from three reaches of Redfield Canyon, Arizona during 2007 to 2024 Location and description of reaches within Redfield Canyon are included in Figure 1.

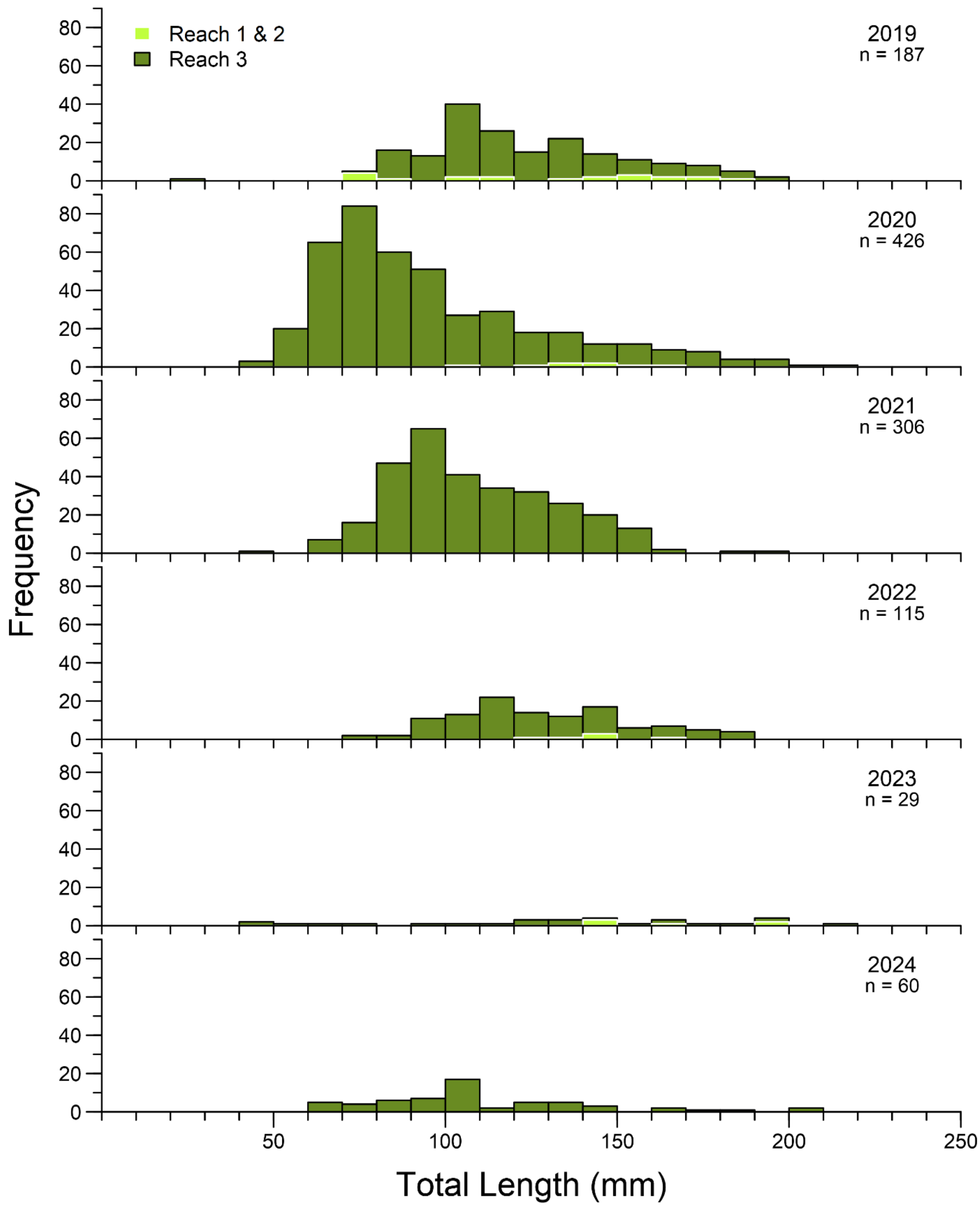

Figure 1.3.—Length frequency distribution of Green Sunfish captured by reach during removal efforts and annual monitoring in Redfield Canyon, 2019 to 2024. Number of fish captured and measured each year is shown in the top right corner of each panel.

2. Gila Topminnow stockings (Task AZ-2002-1)

Strategic Plan Goals:

- Preventing Extinction and Managing Toward Recovery
  - Goal 4. Replicate populations and their associated native fish community into protected streams and other surface waters.
  - Goal 7. Monitor to quantitatively measure and evaluate project success in improving the status of target species and their habitats.

Recovery Objectives:

- Gila Topminnow 1999 draft revised recovery plan objective 2.2. Reestablish Gila Topminnow in suitable habitats following geographic guidelines.
- Gila Topminnow 1999 draft revised recovery plan objective 3. Monitor natural and reestablished populations and their habitats.
- Desert Pupfish recovery objective 2. Re-establish Desert Pupfish populations.
- Desert Pupfish recovery objective 5. Monitor and maintain natural, re-established, and refugia populations.
- Gila Chub draft recovery plan objective 2. Ensure representation, resiliency, and redundancy by expanding the size and number of populations within Gila Chub historical range via replication of remnant populations within each RU.
- Gila Chub draft recovery plan objective 7. Monitor remnant, repatriated, and refuge populations to inform adaptive management strategies.

Overall Background: The purpose of this action is to establish Gila Topminnow populations within the historic range of the species throughout the Gila River Basin in Arizona. Populations of Roundtail Chub[7] may also be established through this project. The Department coordinates with USFWS to determine stocking locations and appropriate donor locations and lineages. The strategy is to stock at least 500 Gila Topminnow initially or for any subsequent augmentations to establish a population. Populations are typically augmented if fewer than 100 fish are captured or observed during monitoring. After stocking, the populations are monitored at 6-months and then annually thereafter for three years after the last stocking event. If a population is considered established after the third post-stocking monitoring, the augmentation and monitoring responsibilities are passed on to other Department programs. Monitoring responsibilities may also be passed along to other agencies. Monitoring techniques are consistent from year to year for a given site, and usually involve setting a minimum of 10 baited minnow trap sets per site, often supplemented with dip netting or seining if habitat conditions allow. Background, results, and recommendations are presented separately for each site.

[7] Populations of Roundtail Chub addressed by this project were formerly classified as Gila Chub.

Sites Monitored or Stocked During 2024:
A table of mean catch per unit effort (CPUE) with standard error, and the proportion of young of year captured for each taxa by gear type at each location can be found in Appendix 3.

Hidden Water Spring.
Background: Hidden Water Spring is located within Cane Spring Canyon, north of Saguaro Lake on the Tonto National Forest. Hidden Water Spring was founded by 350 topminnow from Boyce Thompson Arboretum in 1976 and were continually detected during monitoring through 2010. Three consecutive years of failed detections were then recorded from 2011-2013. Department staff restocked Hidden Water Spring in 2016 with 544 Gila Topminnow from the Phoenix Zoo. Following the Bush Fire in 2020, monitoring failed to detect topminnow for multiple years, suggesting that topminnow were again extirpated in Hidden Water Spring.

Results: On April 10, 2024, Department staff collected 200 Gila Topminnow from Sheepshead Spring and 400 Gila Topminnow (Lower Santa Cruz MU) from the Phoenix Zoo Ranarium Pond. The crew stocked a total of 476 topminnow into Hidden Water Spring with 124 mortalities during transport and stocking.

Recommendations: Hidden Water Spring will continue to be monitored on a three year rotation by Reclamation's monitoring contractor, with the next monitoring effort scheduled for 2026. Hidden Water Spring will be included in the genetic management strategy for Gila Topminnow and will receive periodic managed gene flow in the form of augmentations during implementation of the strategy.

Aravaipa Creek
Background: Aravaipa Creek is a tributary to the San Pedro River about 17 km south of the confluence of the Gila and San Pedro Rivers. It drains the east and north end of the Galuiro Mountains, the southwest portion of the Pinalenos, and the southern portion of the Santa Teresa Mountains. The creek becomes perennial at Aravaipa Spring near Stowe Gulch and flows west to the San Pedro River approximately 35 km. There are two constructed fish barriers (Reclamation funded) at the west end of the creek that prevent upstream movement of nonnative fishes. However, nonnative Green Sunfish, Yellow Bullhead, and Red Shiner were present in the creek before the barriers were constructed. Ongoing nonnative removals led by BLM have largely eliminated Green Sunfish, but Yellow Bullhead were still common as of 2024. Topminnow did not previously exist in the stream, although there was an attempt to establish populations in 1969 and 1977 (Weedman 1999). In April 2022, Department staff stocked 484 Bylas lineage Gila Topminnow into a spring-fed side channel near the upstream end of perennial flow. Major flooding from an above average monsoon season during the summer of 2022 caused the side channel to flood for the first time since 2006. A total of 98 Gila Topminnow were captured during the initial post-stocking monitoring effort in September 2022, and a total of 229 topminnow were captured during monitoring in 2023 (Figure 2.1). Topminnow were first documented in the mainstem of

Aravaipa Creek in September, 2022 with additional captures downstream to near Horse Camp Canyon (approximately 18 km downstream of the original stocking location) in 2023.

<u>Results:</u> On October 1, 2024, Department staff, assisted by Nature Conservancy staff, monitored Gila Topminnow in the spring-fed side channel to Aravaipa Creek. The crew soaked 10 minnow traps for a minimum soak time of two hours and captured 254 Gila Topminnow, 18 Longfin Dace and 15 Speckled Dace. The crew also conducted four opportunistic seine hauls and captured 28 Gila Topminnow, 20 Longfin Dace, and 53 Speckled Dace. An additional 15 dipnet sweeps captured 31 Gila Topminnow, 4 Longfin Dace, and 25 Speckled Dace. Overall, a total of 313 Gila Topminnow (90 < 20 mm; 223 $\geq$ 20 mm) were captured across all sampling methods, although minnow traps were the most effective gear type. Visual observations were made of hundreds of juvenile topminnow in margins of pools that were small enough to evade capture with a dip net. The total capture in 2024 increased relative to 2023 (n = 229), although mean relative abundance estimated from minnow traps was not significantly different between years ($W = 62$, $P = 0.354$, 2023 = 8.11 fish/h, 2024 = 11.09 fish/h). The distribution of topminnow within the side channel appears slightly more continuous compared to 2023 and topminnow have expanded to occupy multiple pools well-beyond the original stocking location (Figure 2.1). Further, Gila Topminnow have continued to be captured in the main channel of Aravaipa Creek. Three detections were made in 2024, one again near Horse Camp Canyon and two further downstream in the vicinity of Javelina Canyon (H. Blasius, BLM, personal communication). The furthest downstream observations of Gila Topminnow at Javelina Canyon represent dispersal of approximately 20.3 km from the initial stocking location.

<u>Recommendations:</u> Relative abundance of Gila Topminnow has remained stable and reproduction has been documented for three consecutive years without population augmentation. Monitoring results suggest that there is an established population of Gila Topminnow within the side- channel of Aravaipa Creek. While total catch has remained under the target of 500 individuals, topminnow have continued to be documented dispersing out of the side-channel and the total topminnow population size likely greatly exceeds 500 individuals when considering the entire system (i.e., main and side-channels). A final stocking for genetic augmentation of Gila Topminnow in Aravaipa Creek will occur in 2025, and periodic collections and augmentations will occur as part of the topminnow genetic management strategy for the Bylas MU. Future monitoring may be warranted following severe flooding events to confirm Gila Topminnow can persist in the side channel through catastrophic events.

Las Cienegas National Conservation Area – Maternity Wildlife Pond

<u>Background:</u> Maternity Wildlife Pond is located in the Gardner Canyon drainage about 9.6 km upstream of the confluence with Cienega Creek. The pond was improved in 2020 which included dredging and installing a solar well to create a perennial water source for native fish and amphibians. In April 2021, Department staff initially translocated 248 Gila Topminnow from

Empire Tank to Maternity Wildlife Pond, and later augmented the pond with 250 topminnow from Cienega Creek in August, 2023. Post-stocking monitoring from 2021 to 2023 has documented a large, stable population of topminnow (2021 n = 1,554, 2022 n = 1,591, 2023 n = 6,618).

Results: On August 26, 2024, Department staff monitored Gila Topminnow in Maternity Wildlife Pond, by soaking ten minnow traps for a minimum of two hours. A total of 2,078 Gila Topminnow were captured (934 < 20 mm, 1,144 ≥ 20 mm). The catch total for 2024 exhibited over a threefold decrease relative to the 2023 total (2023 = 6,618) and relative abundance estimated from minnow traps significantly decreased (2023 = 250.40 fish/h, 2024 = 75.14 fish/h, $W = 96$, $P < 0.01$). Nearly all adult topminnow were infected with yellow grub (*Clinostomum marginatum*), which may have contributed to the decline in relative abundance. Despite the majority of adult fish carrying a heavy parasite load, the proportion of fish under 20 millimeters in total length increased marginally compared to 2023 (2023 = 33.8%, 2024 = 44.9%) suggesting parasitosis has not strongly influenced spawning activity.

Recommendations: While Maternity Wildlife Pond currently meets the criteria for an established population, the presence of a fish parasite limits the utility of the site as a donor population for future translocations and as a genetic refuge for Cienega Creek lineage Gila Topminnow. However, in October of 2024 it was noted that the pond seal had failed and the water level dropped substantially (C. Perez, BLM, personal communication). Bureau of Land Management personnel are currently trying to resolve the issue; however, the pond may ultimately need to be drained for repair which would require reestablishing the population.

Sabino Canyon

Background: Sabino Canyon is located northeast of Tucson, Arizona within the Coronado National Forest and Sabino Canyon Recreation Area. Sabino Canyon is a tributary to the Santa Cruz River and drains the Santa Catalina Mountains, flowing southwest to its confluence with Tanque Verde Wash in Tucson. Sabino Canyon was chemically treated in 1999 to remove nonnative Green Sunfish, and afterwards was stocked with salvaged Roundtail Chub[8] (Ehret and Dickens 2009). Gila Topminnow were initially stocked in the recreation area near 'The Crack' in 2015 and augmented in 2016. These stockings resulted in the establishment of a population of topminnow mostly below Sabino Lake Dam.

Stream habitat in a reach of Sabino Canyon located approximately 250 m upstream from the confluence with East Fork Sabino Canyon was evaluated in 2017 and 2018 and identified as a candidate for translocation of Gila Topminnow. A total of 557 Gila Topminnow were translocated from the large pools immediately below Sabino Dam to Sabino Canyon upstream of the confluence with East Fork Sabino Canyon in June 2018. The Gila Topminnow population in Sabino Canyon upstream of the East Fork was initially monitored in May, 2019. No topminnow were captured or

[8] Chub stocked into Sabino Canyon were previously classified as Gila Chub.

observed. Immediately following the monitoring effort, a total of 148 Roundtail Chub[1] (>100 mm TL) collected from downstream of Sabino Dam were stocked into a pool just downstream of the topminnow stocking location (520836/3581045). In October 2019, Department staff collected an additional 527 Gila Topminnow in three seine hauls from the pools immediately downstream of Sabino Dam. The fish were translocated to Sabino Canyon upstream of the confluence with East Fork Sabino Canyon (520784/3581144). A total of 350 Gila Topminnow were successfully stocked. During monitoring from 2020 to 2023 no Gila Topminnow were detected but a total of 15, 10, 47 and 3 Roundtail Chub were captured in each year respectively. Despite the relatively low catch in hoop nets, supplemental snorkel surveys indicated that the Roundtail Chub population was much more robust in the vicinity of the stocking location. Surveys downstream of the 2019 stocking location documented the Roundtail Chub population had expanded at least 1.4 km downstream from the original stocking location. Roundtail Chub were first documented in the lower portion of East Fork Sabino Canyon in 2023 when 35 individuals were observed in two pools while snorkeling.

Results: On May 7, 2024, Department staff monitored the population of Roundtail Chub in Sabino Canyon upstream of the East Fork of Sabino Canyon (Figure 2.2). The crew set ten mini-hoop nets for a minimum soak time of two hours and captured eight Roundtail Chub (0.39 fish/h). Two dip net sweeps resulted in the capture of four more Roundtail Chub. Three distinct age classes of fish appeared to be present within the sample (including juveniles and adults), demonstrating that reproduction and recruitment continues to occur within this reach (Figure 2.3). A total of 152 Roundtail Chub were observed during a snorkeling pass in the 2019 stocking pool, suggesting that daytime mini-hoop net sets continue to substantially estimate the number of individuals present.

Department staff coordinated with partners to extend the range of Roundtail Chub in Sabino Canyon upstream to Hutch’s Pool or the upstream extent of designated critical habitat (near the confluence with West Fork Sabino Canyon), and to extend range further upstream in East Fork Sabino Canyon in 2024 (Figure 2.2). The initial plan was to collect chub from near Sabino Dam, or as far downstream as possible within the recreation area for translocation upstream. On May 7, 2024, Department staff failed to observe any Roundtail Chub at planned collection locations within the recreation area reach of Sabino Canyon and only captured 3 individuals in 10 mini-hoop nets set for approximately 7 hours near ‘the crack’ at the upper end of the recreation area. Consequently, the crew decided to postpone the objective of translocating fish upstream to Hutch’s pool, and instead extended the range of the existing population in East Fork Sabino Canyon. Seven of the eight largest chub captured during the monitoring effort were translocated into the most upstream perennial pool in East Fork Sabino Canyon. Mini hoop nets were reset overnight at the monitoring location to collect more fish for translocation to the East Fork Sabino Canyon.

On May 8, 2024, Department, USFS and University of Arizona staff collected 44 additional Roundtail Chub and released an additional 19 individuals back to the site of capture. Translocated fish were of mixed size classes and ranged from 62 to 197 mm TL. Collected fish were translocated into the same pool in East Fork Sabino Canyon that was stocked the previous day. One mortality was recorded during transport due to injury during capture.

Recommendations: The Roundtail Chub population in Sabino Canyon near the confluence with East Fork Sabino Canyon meets all criteria for establishment, as the population has persisted without augmentation for five years, spawning has been documented for the last three consecutive years, and increase in distribution has been documented both upstream into East Fork Sabino Canyon and downstream at least 1.4 km towards the recreation area. An increase in mean relative abundance has been difficult to document with mini-hoop nets at this location due to the low capture probability of short duration daytime sets, however snorkel surveys documented far more individuals were present than the catch data alone suggested. The relative absence of Roundtail Chub at planned collection sites in the recreation reach is likely best explained by the extremely low flow in Sabino Canyon from May to December 2023 (Figure 2.4). Sabino Canyon typically experiences intermittency during the driest time of year (typically May-June), however no flow was detected at the USGS gauge (09484000) in Sabino Canyon for 203 consecutive days, with the next longest streak only being 163 days during 2020-2021. Continuing to translocate fish further upstream into unoccupied critical habitat (including Hutch's pool) is recommended to provide additional drought refugia and dispersal opportunities should additional extreme drought events occur in the future.

Telegraph Canyon

Background: Telegraph Canyon is a tributary to Arnett Creek and drains from the north side of Picketpost Mountain. In 1992, the Department, Tonto National Forest, and USFWS identified an opportunity to reestablish a native fish community in Arnett Creek and its tributary Telegraph Canyon. In the late 1990s, a fish barrier was built on Arnett Creek, the stream was chemically treated to remove nonnative fishes, and native fish were stocked. Unfortunately, those fish did not establish populations likely because too few were stocked and drought greatly reduced the amount of perennial water in the system. The partners re-evaluated the stream in 2007 and determined that the small amount of habitat was probably only suitable for Longfin Dace and Gila Topminnow. Longfin Dace were stocked in 2007 and established a population in Telegraph Canyon. In May 2017, a total of 522 Gila Topminnow were stocked into Arnett Creek just downstream of the Telegraph Canyon confluence. Few topminnow were captured during post-stocking monitoring from 2017-2019, and it is presumed the original population failed after the stream nearly entirely dried in 2018. Following completion of an invasive plant removal project in Telegraph Canyon by USFS staff in 2020, Department staff translocated 389 Redrock Canyon lineage Gila Topminnow from Walnut Spring #392 and one other location to Telegraph Canyon in May, 2021. Despite the Telegraph Fire burning a substantial portion of the upper watershed in June, 2021 post stocking monitoring in October from 2021 to 2023 resulted in the capture of 563, 165, and 147 Gila Topminnow respectively. Dispersal of topminnow to Arnett Creek was first documented in 2021, with topminnow observed each year from 2021 to 2023. A total of 212 topminnow were captured in two minnow traps in 2023 set near the 2017 stocking location.

Results: On October 23, 2024, Department staff monitored Gila Topminnow in Telegraph Canyon. The crew set 10 collapsible minnow traps in pools along a 0.5 km reach from the downstream end of surface water (487132/3680070) to the pool above the original stocking location where 100 fish were moved in 2023 (486899/3679667). Minnow traps were soaked for a minimum of two hours. A total of 452 Gila Topminnow were captured in minnow traps, which is nearly a 12-fold increase compared to the 2023 catch total (2023 n = 38, Figure 2.5). Accordingly, relative abundance increased in 2024 to 15.03 fish/h compared to 1.85 fish/h in 2023, however this difference was not statistically significant ($W = 38$, $P = 0.332$). This is likely an artifact of reduced statistical power resulting from variability in trapping success. For example, 60 % of traps set in 2023 and 40% of traps set in 2024 captured zero fish. Nevertheless, there was a clear qualitative improvement in relative abundance. Additionally, there was evidence of spawning in that 121 captured individuals were <20 mm TL (26% of total catch). Finally, six individuals were captured in the uppermost pool where fish were translocated in 2023 and the crew visually observed more individuals that evaded capture. A single dip net sweep was conducted just below the uppermost pool and a single individual was captured, suggesting individuals may have started to disperse downstream from the uppermost occupied pool.

While traps were soaking in Telegraph Canyon, the crew walked down to Arnett Creek to investigate if topminnow were still present. The crew sampled a large pool near the confluence of Telegraph Canyon and Arnett Creek (487182/ 3680579) that was the original stocking location in 2017. Two minnow traps were set at this location and soaked for 1.5 hours, however no topminnow were captured. The crew also sampled a pool further downstream (486394/ 3681072) using a straight seine. A total of 710 topminnow (267 <20 mm TL 443 >= 20mm TL) were captured in a single haul. This is the fourth consecutive year topminnow have been captured in Arnett Creek and the catch total for 2024 exceeded the total number of individuals initially stocked in 2017.

Recommendations: Measures of relative abundance and evidence of reproduction have substantially improved in Telegraph Canyon and are the highest since 2021. Any future stocking and/or genetic augmentation of the topminnow population in Telegraph Canyon should consider further supplementation of the uppermost occupied pool to bolster the natural dispersal of individuals throughout Telegraph Canyon. The major limiting factor for Gila Topminnow in Telegraph Canyon in the long term will likely be habitat availability, as only a small number of pools continue to be occupied. The crew observed several pools that previously held water were dry in October 2024, possibly related to post-fire and drought effects as some pools continue to fill with sand and gravel while others have scoured out since the Telegraph Fire. Gila Topminnow populations in both Telegraph Canyon and Arnett Creek have persisted for four years, increased in distribution and shown increasing relative abundance since the initial stocking in 2021. Monitoring data to date suggests that populations can be considered established in both of these locations, or at least one metapopulation. Periodic monitoring of both locations by the Department or other partners should continue to occur on a regular basis going forward.

Unnamed Drainage #68B

Background: Unnamed Drainage #68B is located on the Tonto National Forest and is a tributary to Mesquite Creek, which flows into Tortilla Creek just upstream of Canyon Lake. Gila Topminnow were previously stocked in Mesquite Tank #2 (above Unnamed Drainage #68B) in 1982. A valve on the dam of Mesquite Tank #2 was opened, allowing it to drain and completely dry out. As a result, Gila Topminnow washed downstream and established a population in Unnamed Drainage #68B and later dispersed into perennial pools in lower Mesquite Creek and lower Tortilla Creek. The original population occupying Unnamed Drainage 68B was founded with an unknown number of individuals and was never augmented. Despite these challenges, the population persisted until at least 2019, after which topminnow were not detected in three consecutive surveys from 2020 to 2021. In April 2022, Department staff stocked 393 Cottonwood Spring lineage Gila Topminnow into Unnamed Drainage #68B and during post-stocking monitoring in October a total of 990 topminnow were captured. A total of 22 Gila Topminnow were captured in a small isolated pool that appeared to be at immediate risk of drying, and were translocated to a large pool upstream of the stocking location and a natural fish barrier (464691/3711295) in an effort to expand the distribution of the topminnow population (Figure 2.6). A total of 1,422 topminnow were captured during monitoring in 2023, and topminnow were detected in the pools upstream of the barrier. An additional 500 topminnow were translocated from the pools downstream of the barrier to upstream of the barrier to bolster the existing population.

Results: On October 10, 2024, Department staff monitored Gila Topminnow in Unnamed Drainage 68B. The crew set ten minnow traps in three pools near the original stocking location and five additional traps in a series of pools above the natural barrier where fish were translocated in 2023. All traps were soaked for a minimum of two hours. A total of 2,477 Gila Topminnow were captured across all pools (Figure 2.6) including 435 topminnow upstream of the natural barrier.

Relative abundance of Gila Topminnow increased relative to 2023, however the difference was not statistically significant (2023 = 36.32 fish/h; 2024 = 51.96 fish/h, $W = 86.5$, $P = 0.279$). Nevertheless, total catch has continued to increase since initial post-stocking monitoring (2022 = 990; 2023 = 1,434; 2024 = 2,477). Approximately 61.8% of the total catch were juveniles (n = 1,532 < 20 mm TL) indicating substantial spawning is occurring. An additional promising result is that 435 individuals were captured in the pool complex above the natural barrier, which included juveniles. The reach above the first natural barrier represents an expansion of topminnow by roughly 150 meters in the lower reach of Unnamed Drainage 68B. The crew wet/dry mapped the remainder of the drainage up to Mesquite Tank #2 while traps soaked. Despite the tank being dry at the top of canyon, at least 12 pools were present within approximately 400 m of the canyon, with several other likely fish barriers also present (Figure 2.7).

Recommendations: Gila Topminnow in Unnamed Drainage 68B can be considered reestablished because the population has persisted since the most recent stocking in 2022, increased in relative abundance, demonstrated spawning each of the last three years, and has increased in distribution. Unnamed Drainage 68B will be pursued as a primary donor location for the genetic management of Monkey/Cottonwood Springs MU following recommendations in the Gila Topminnow Genetic Management Plan (Mussmann 2024). Therefore, additional stockings will take place following 2025 to establish Unnamed Drainage 68B as the primary donor site for the Monkey/Cottonwood Springs MU and initiate gene flow among populations within the MU.

Rarick Canyon
Background: Rarick Canyon is a tributary to Red Tank Draw and consists of a series of perennial bedrock pools (Figure 2.8). A waterfall barrier (~10 m high) in Rarick Canyon prevents upstream movement of nonnative fishes from the perennial reach of Red Tank Draw. A survey of isolated pools in the Rarick Canyon drainage from 2017 to 2018 detected Black Bullhead. Intensive mechanical removals efforts in 2019 resulted in the eradication of Black Bullhead from the Rarick Canyon drainage. Roundtail Chub from Red Tank Draw were translocated above a natural barrier into four isolated pools in Rarick Canyon in 2019 and augmented in 2020 and 2021. Gila Topminnow (Redrock Canyon lineage) were also translocated to one of the same isolated pools above the barrier in April, 2020 (F18, Figure 2.8). No topminnow were captured during annual monitoring from 2021 to 2023. Roundtail Chub have been consistently captured during monitoring with reproduction and dispersal from stocking locations documented during annual monitoring from 2020 to 2023

Results: During October 15-16, 2024, Department staff monitored the translocated population of Roundtail Chub in Rarick Canyon. A combination of gear types was used that included angling, dip nets, collapsible minnow traps, and mini hoop nets. A total of 11 mini hoop nets and eight minnow traps were set in three pools that were previously stocked (F17, F18, F23, Figure 2.8) and two non-stocked pools (F21, FX2). Dip nets were used to sample an additional unnamed pool that was too shallow to for traps (FX). All traps soaked for a minimum of 24 hours. Traps captured a total of 152 Roundtail Chub and 22 Fathead Minnow, of which 33 chub were contacted in a pool outside of the original stocking locations. An additional 61 juvenile Roundtail Chub were captured by dip netting a shallow pool (FX) and five individuals were captured via angling (totals across all gear types = 218 Roundtail Chub, 33 Fathead Minnow, Figure 2.9). No Gila Topminnow were captured during monitoring.

The total catch for Roundtail Chub increased over 50% compared to 2023 (n = 140) however mean relative abundance did not significantly increase compared to 2023 (2024 = 0.34 fish/h, 2023 = 0.22 fish/h, W = 164.5, P = 0.07, Figure 2.10). Further, this is the third consecutive year that Fathead Minnow relative abundance has remained depressed, suggesting that Roundtail Chub may be exerting strong predation pressure (Figure 2.10). Length distribution of the Roundtail Chub population suggests that spawning has occurred for at least the third time since monitoring began in 2020 (Figure 2.11). Of particular note was a strong juvenile size-class signifying that environmental conditions were favorable for spawning.

Recommendations: Roundtail Chub in Rarick Canyon can now likely be considered established since the population has persisted without augmentation since 2021, has expanded in distribution to pools outside the original stocking locations, has increased in mean relative abundance, and spawning has occurred at least three years. The population is now sufficiently large to support a collection of individuals should it become necessary, making Rarick Canyon an important replicate of the Red Tank Draw Roundtail Chub population. Rarick Canyon should be monitored periodically going forward to confirm that the population continues to persist, particularly following years of extreme drought.

Figures:

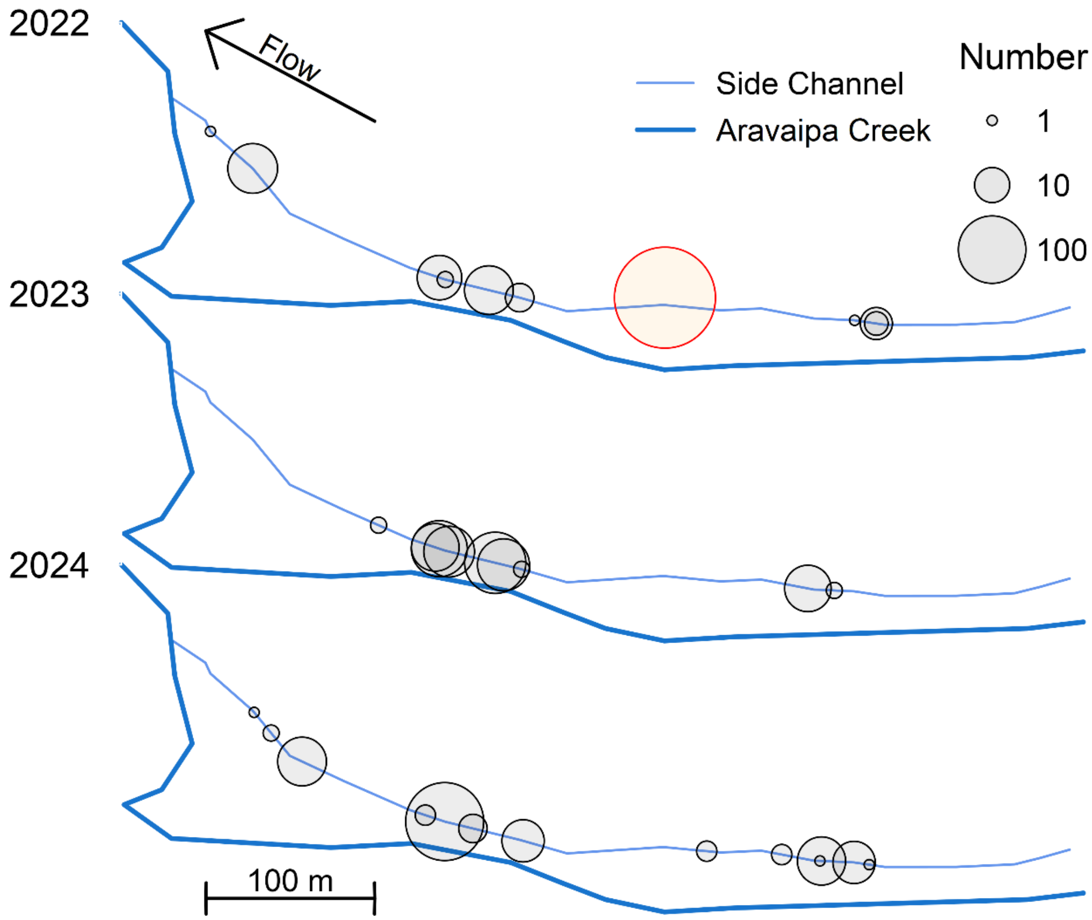

Figure 2.1.—Location and number of Gila Topminnow captured in the Aravaipa Creek side-channel from 2022 to 2024 (top to bottom). The dark blue line represents the main channel of Aravaipa Creek while the light blue line represents the spring fed side-channel. The orange circle depicts the initial stocking location in 2022, while grey circles represent the location topminnow were captured during monitoring. The size of circles reflects the number of Gila Topminnow captured and/or stocked at a given location.

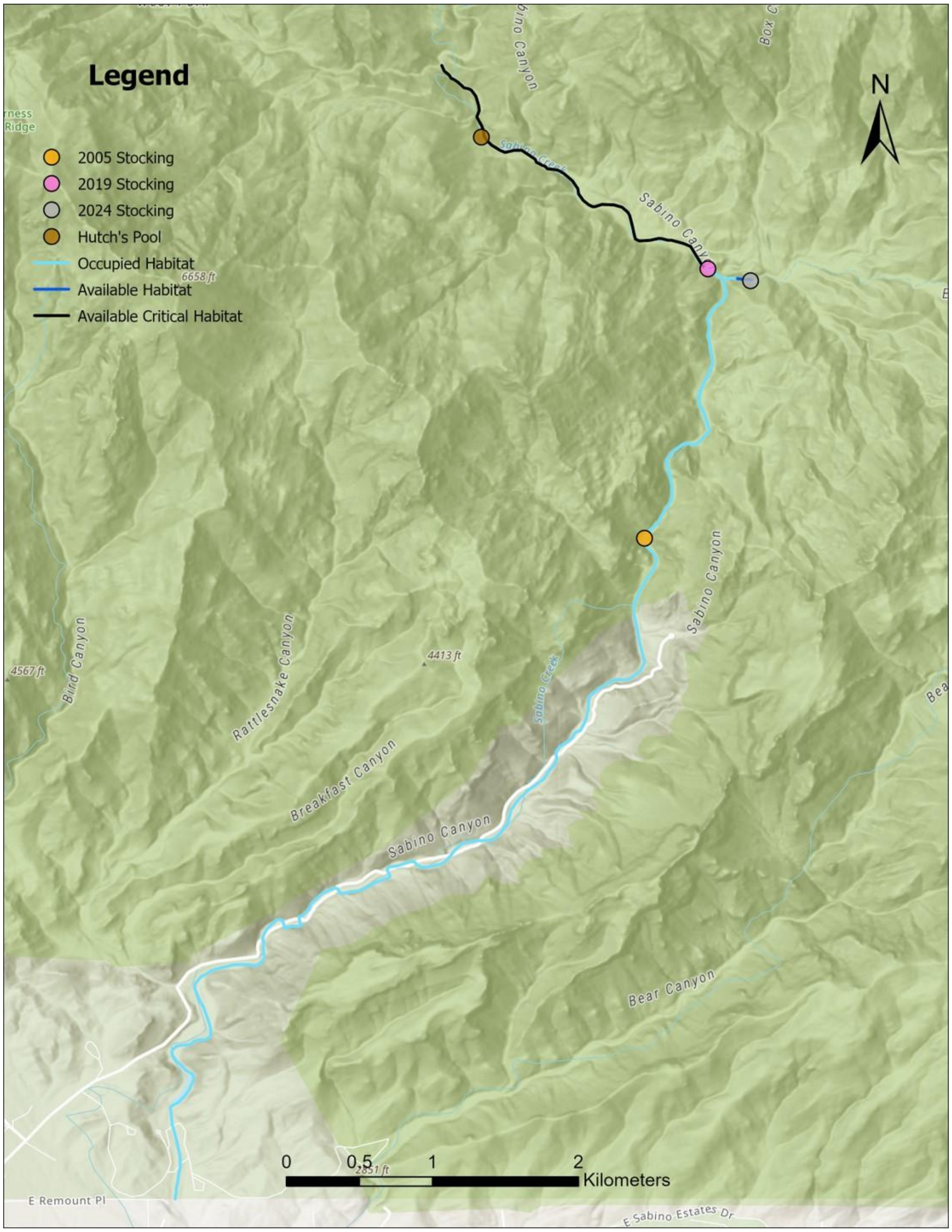

Figure 2.2.—Map detailing the focal area within Sabino Canyon and recent points of translocation. Monitoring data from 2023 found chub to be at low density in the reach between the confluence of East Fork Sabino and the 2005 stocking location. Fish were captured from upstream of the confluence with East Fork Sabino (pink circle) and translocated to East Fork Sabino Canyon (grey circle) to expand the distribution of chub within the system in 2024.

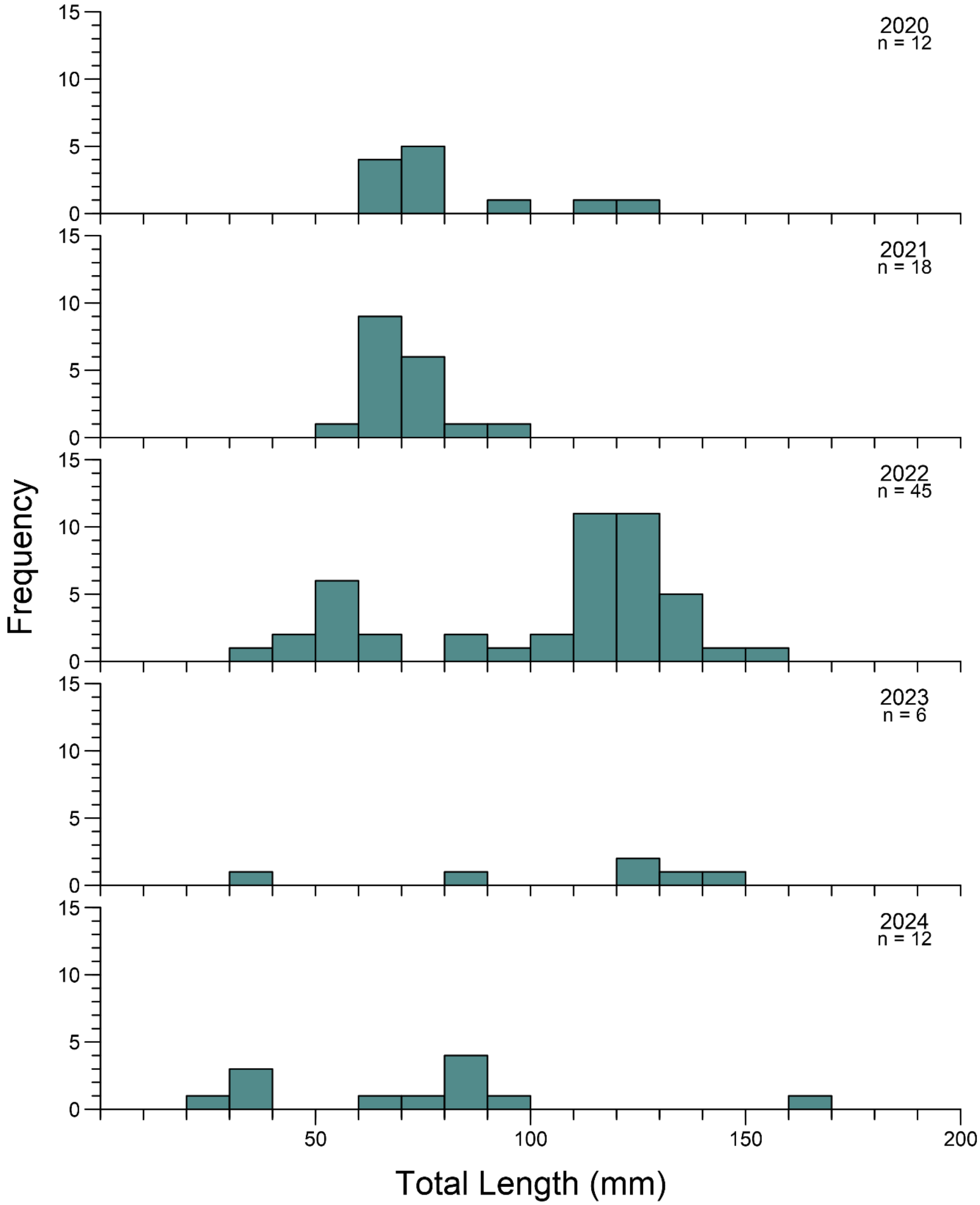

Figure 2.3.—Length frequency distribution of Roundtail Chub captured during annual monitoring in Sabino Canyon in the vicinity of the confluence with East Fork Sabino Canyon from 2020 to 2024. Number of fish captured and measured each year is shown in the top right corner of each panel.

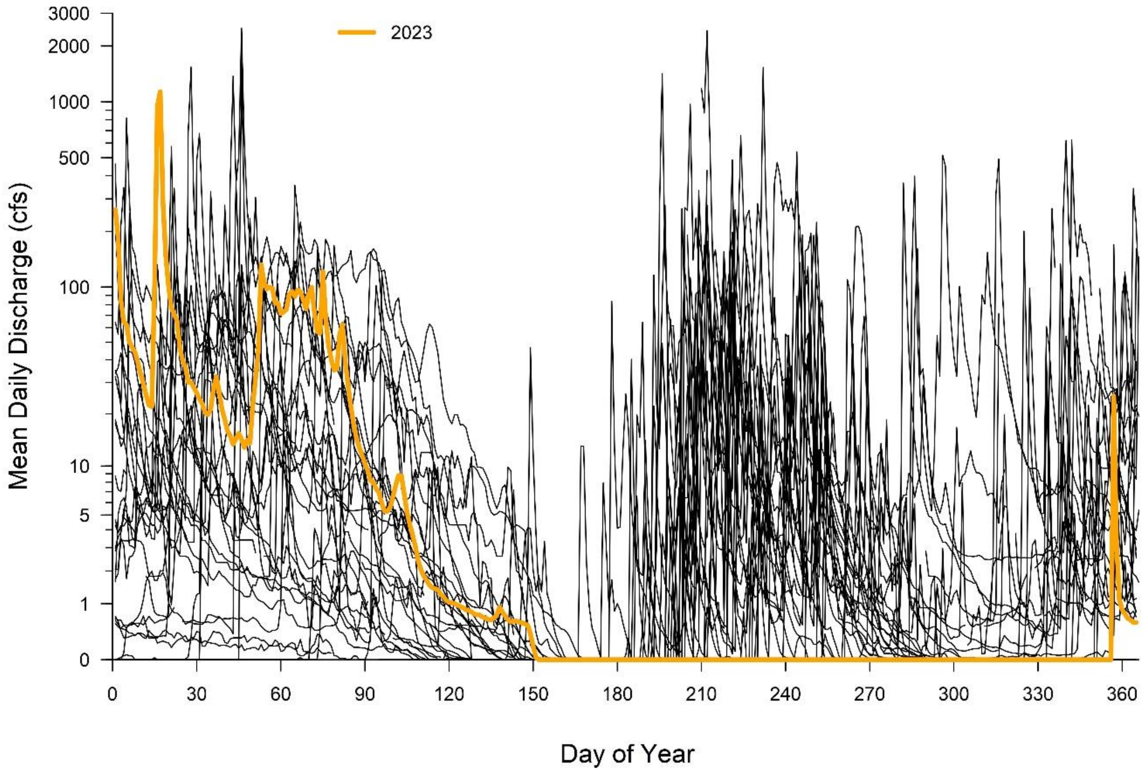

Figure 2.4.—Mean daily discharge for the USGS Gauge (09484000) at Sabino Canyon from 1994 through 2024. Mean daily discharge for 2023 is highlighted with an orange line. Note zero flow was recorded at the gage for 203 consecutive days.

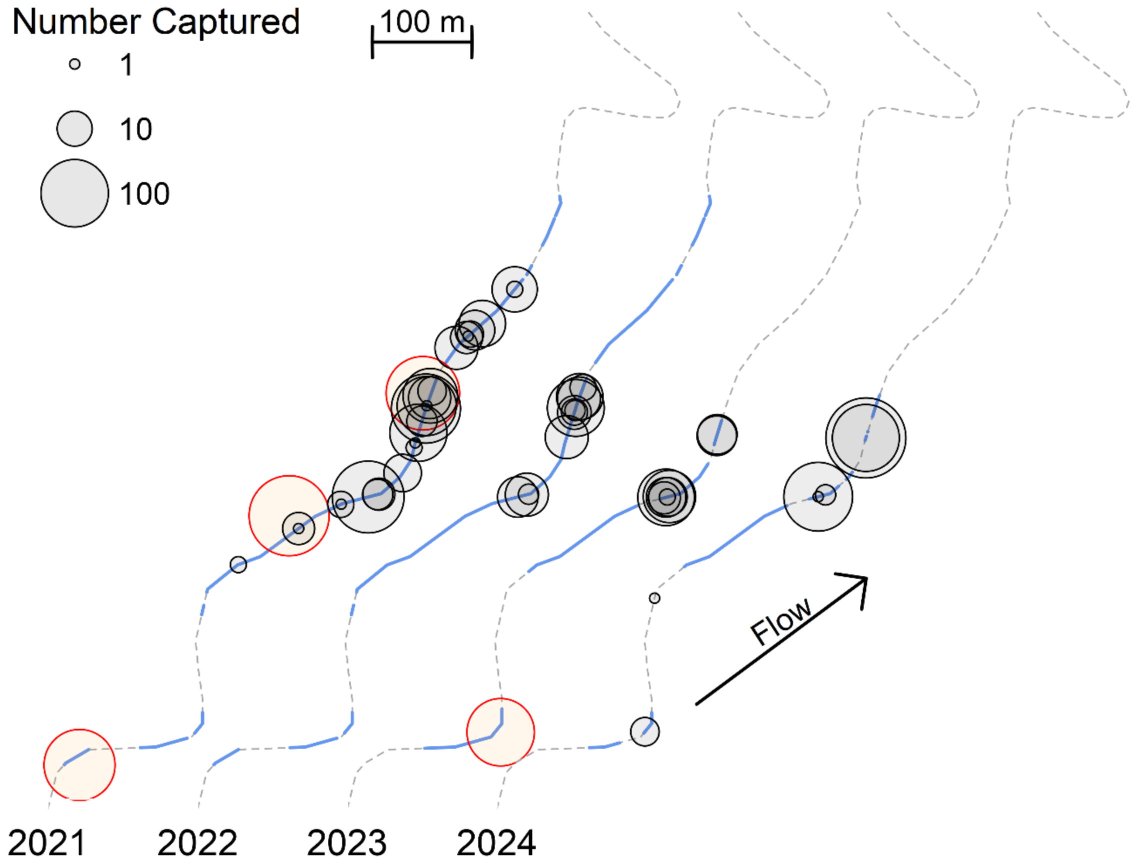

Figure 2.5.—Location and number of Gila Topminnow captured in Telegraph Canyon during annual monitoring from 2021 to 2024 (from left to right). The blue line represents the perennial portion of Telegraph Canyon (based on the most recent wet-dry mapping) while the dashed line represents intermittent portions. Orange circles represent the location and number of fish released the year prior, while grey circles represent the location and number of fish captured during monitoring.

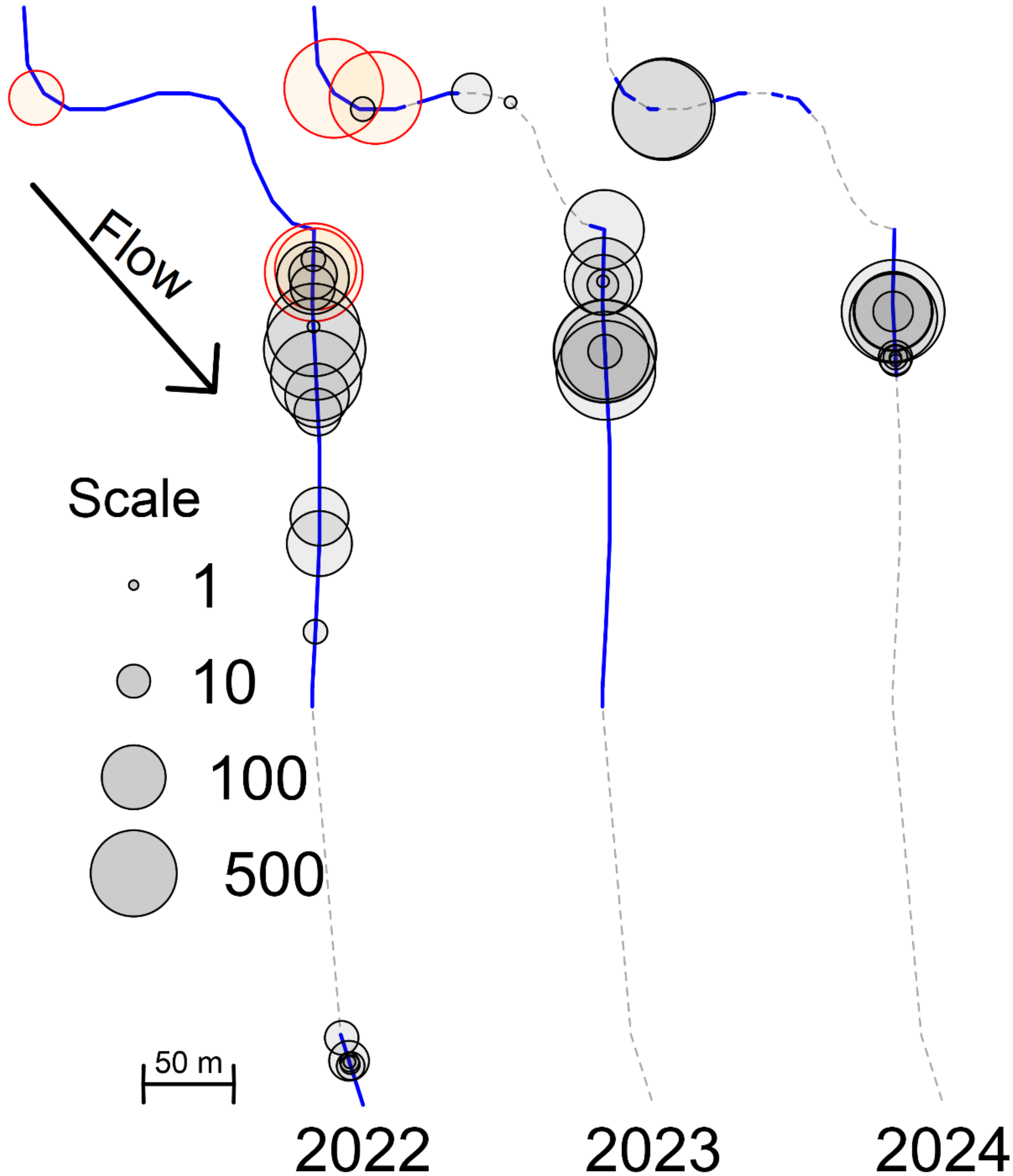

Figure 2.6.—Location and number of Gila Topminnow captured in Unnamed Drainage #68B during annual monitoring from 2022 to 2024 (from left to right). The blue line represents the perennial portion of Unnamed Drainage 68B (based on wet-dry mapping for each year) while the dashed line represents intermittent portions. Orange circles represent the location and number of fish released, while grey circles represent the location and number of fish captured during monitoring.

Figure 2.7.—Site photographs of Unnamed Drainage #68B. The first photographs show Mesquite Tank #2 in 2023 (A) compared to 2024 (B). Examples of pool habitat present below Mesquite Tank #2 in 2024 are shown in panels (C) and (D).

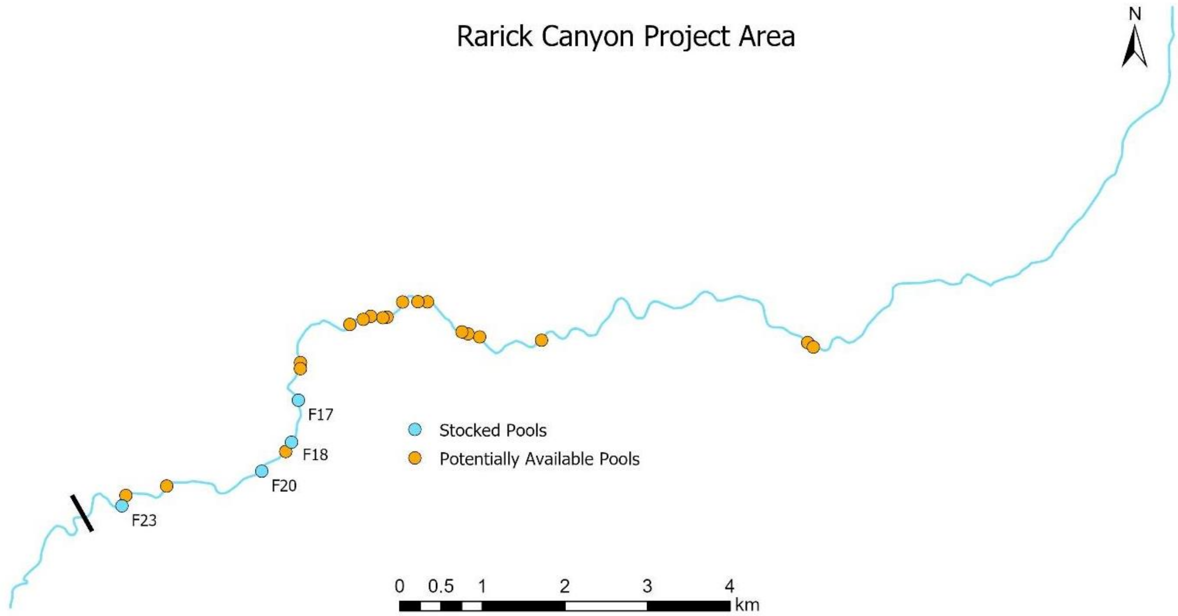

Figure 2.8.—Map of Rarick Canyon (blue line) including the location of the barrier falls (thick black line), names and locations of pools stocked with fish (blue dots), and the remaining potentially habitable pools in Rarick Canyon (orange dots).

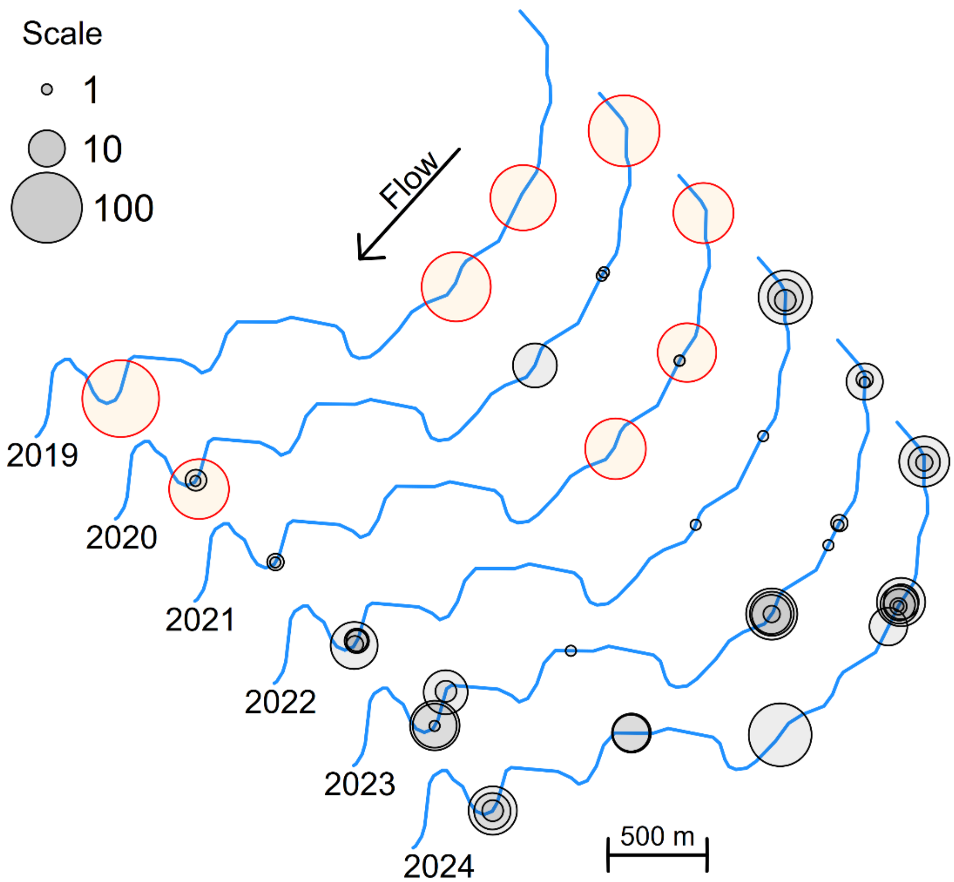

Figure 2.9.—Location and number of Roundtail Chub stocked and captured in Rarick Canyon from 2019 to 2024 (from top left to bottom right). Orange circles represent the location and number of fish stocked, while grey circles represent the location and number of fish captured during monitoring.

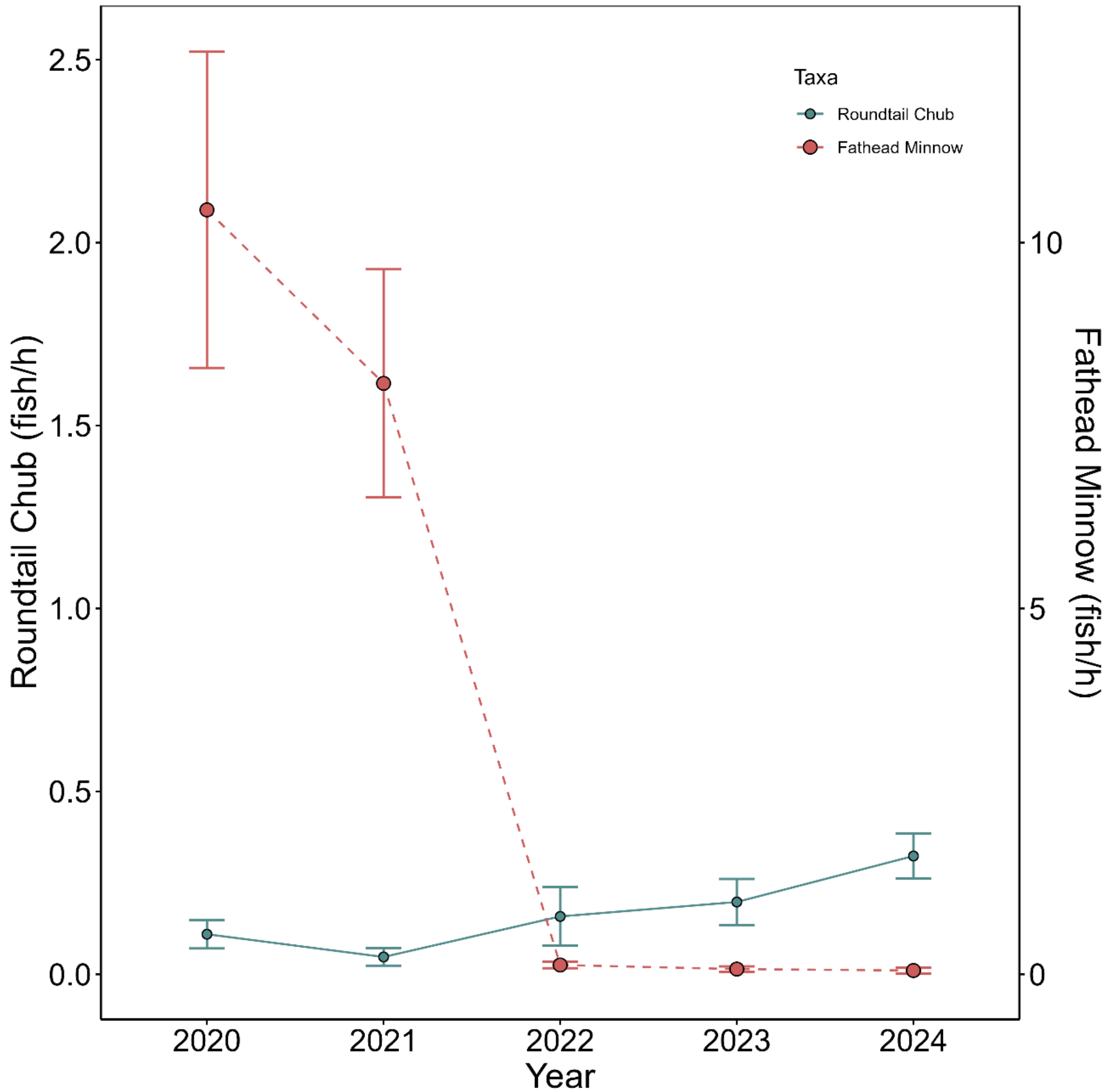

Figure 2.10.—Mean relative abundance of Roundtail Chub and Fathead Minnow captured in Rarick Canyon from 2020 to 2024. Data reflect the mean of mini-hoop net and minnow trap catch per unit effort (fish/h) and include standard error. Note that relative abundance is on a different scale for each taxon.

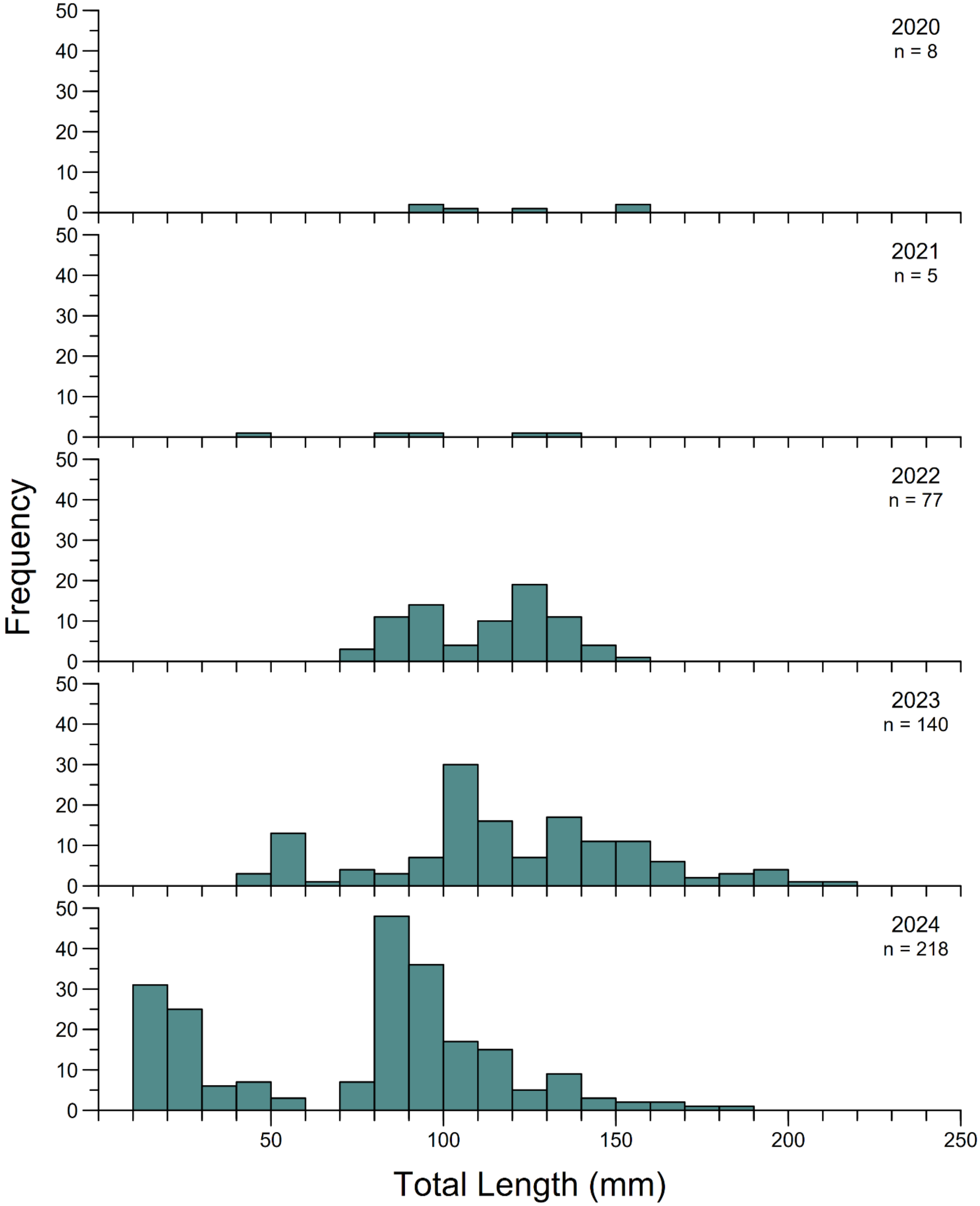

Figure 2.11.—Length frequency distribution of Roundtail Chub captured during annual monitoring in Rarick Canyon 2020 to 2024. Number of fish captured and measured each year is shown in the top right corner of each panel.

3. Spring Creek (Oak Creek tributary) repatriations (Task AZ-2013-1)

Strategic Plan Goals:

- Preventing Extinction and Managing Toward Recovery
  - Goal 3. Remove nonnative aquatic species threats.
  - Goal 4. Replicate populations and their associated native fish community into protected streams and other surface waters.
  - Goal 7. Monitor to quantitatively measure and evaluate project success in improving the status of target species and their habitats.

Recovery Objectives:

- Spikedace recovery objective 6.2.5 Reclaim as necessary to remove non-native fishes.
- Spikedace recovery objective 6.3. Reintroduce Spikedace to selected reaches.
- Spikedace recovery objective 6.4. Monitor success/failure of reintroductions.
- Gila Topminnow 1999 draft revised recovery plan objective 2.2. Reestablish Gila Topminnow in suitable habitats following geographic guidelines.
- Gila Topminnow 1999 draft revised recovery plan objective 3. Monitor natural and reestablished populations and their habitats.
- Gila Chub draft recovery plan objective 1.3.1. Eliminate or control problematic nonnative aquatic organisms.
- Gila Chub draft recovery plan objective 7. Monitor remnant, repatriated, and refuge populations to inform adaptive management strategies.

Background: Spring Creek is a tributary to Oak Creek in the Verde River drainage, and contains Roundtail Chub[9], Speckled Dace, Longfin Dace, Desert Sucker, and northern Mexican gartersnake. A small diversion dam about 0.95 km upstream from the confluence with Oak Creek purportedly prevented most nonnative fishes from invading upstream, but there are historic records of Smallmouth Bass and Fathead Minnow in the system. Green Sunfish were detected below the diversion dam in 2011, and in May 2014 Green Sunfish were captured 2.5 km above the dam. Department staff began removal efforts immediately and completed seven removals during the summer of 2014, after which the Department's Conservation and Mitigation Program staff successfully eradicated Green Sunfish above the diversion dam. Reclamation finished construction of a fish barrier about 1.1 km upstream from Oak Creek in April 2015.

Gila Topminnow were initially stocked in 2015 and were considered established above the barrier by 2019. However, topminnow were not detected during topminnow-specific monitoring by Reclamation's long-term monitoring contractor in 2022 (Shollenberger et al. 2023). In 2023 Department staff translocated 492 Gila Topminnow from Sheepshead Canyon to the pool immediately above the fish barrier in Spring Creek. Spikedace were initially stocked above the

[9] Chub in Spring Creek were previously classified as Gila Chub.

barrier in 2015, with additional stockings occurring in 2017, 2018, 2020, 2021, and 2022 (Figure 3.1). In total, 3,788 Spikedace have been translocated into Spring Creek upstream of the barrier. Spikedace relative abundance was low during the first three years of annual monitoring, and remained low until reproduction was first documented in 2018, with peak relative abundance documented in 2019 (Figure 3.1).

Results: On September 10, 2024, Department staff monitored Spikedace in Spring Creek by electrofishing one fixed 100 m sub-reach and two randomly selected 100 m sub-reaches. The crew captured a total of 25 Spikedace, 177 Roundtail Chub, 105 Desert Sucker, 100 Longfin Dace, 307 Speckled Dace, and 3 Gila Topminnow (Table 3.1) during the first pass. The Gila Topminnow were captured in the reach upstream of the barrier in the vicinity of the 2023 stocking location. The second randomly selected reach, which abuts private land, is where the majority of Spikedace were captured (n = 23). Relative abundance increased compared to 2023, however the difference was not statistically significant (2023 = 10.09 fish/h, 2024 = 26.88 fish/h, $W = 5$, $P = 0.827$). Relative abundance and total number of Spikedace captured in Spring Creek were both the second highest recorded since monitoring began in 2015, despite no stocking occurring since spring of 2022 (Figure 3.1). This suggests that the Spikedace population in Spring Creek may be able to sustain itself without supplemental stockings, given prevailing environmental conditions allow spawning to occur.

No juvenile Spikedace were captured in 2024, suggesting that spawning continues to be sporadic with spawning success likely dependent on elevated springtime flows, as was the case in 2019 and 2023 when the strongest wild spawned year classes were documented (Figure 3.2). Mean total length of captured Spikedace was 56.0 mm (min = 48 mm, max = 63 mm) demonstrating that the 2023 year-class was well represented by individuals that successfully recruited (Figure 3.2). Consequently, spawning success may have been substantially better than the handful of juveniles captured in 2023 may have initially suggested. Three-pass depletion sampling was conducted at the fixed site near the Willow Point Road crossing. Estimated abundance of Spikedace using the Carle-Strub method was 14 fish per 100 meters with a capture probability of 0.26 (Table 3.2, Figure 3.3). This abundance estimate is nearly two and half times greater than the abundance estimate calculated in 2023 (N = 6; CI = 3.29 – 8.71), although the relatively wide confidence intervals around the estimates makes further inference difficult.

There were not any notable changes in mean relative abundance between 2023 and 2024 for any of the other native fish taxa present in Spring Creek, as numbers appeared to be stable or increasing for all taxa (Figure 3.4)

Recommendations: A small population of Spikedace is likely established in Spring Creek. The population has been detected each year since 2017, spawned at least three years since monitoring began in 2015, individuals have been captured throughout the project area, and has increased in relative abundance since the most recent stocking effort, meeting criteria for population establishment. Because of the relatively small size of the population, the documented inconsistent spawning, and close proximity to ARCC, this population should be augmented every three years to ensure population persistence and maintain genetic diversity. Lastly, Spikedace might be more abundant within reaches of Spring Creek located on private lands that have not been accessed for sampling. Future monitoring efforts should seek permission from private property owners to better understand distribution and abundance of Spikedace throughout the entire perennial portion of the system above the fish barrier.

Tables and Figures:

Table 3.1.—Summary of fish captured during the first pass at three 100 m electrofishing sub-reaches in Spring Creek during annual monitoring on September 10, 2024. Shown are the number of fish captured in each sub-reach (#Ind) and the mean number of fish captured per hour of electrofishing effort (#Ind/h) for the first pass. Standard error (SE) is included for the total estimate that reflects the first three passes.

Sub-reach	Statistic	Desert Sucker	Gila Topminnow	Longfin Dace	Roundtail Chub	Speckled Dace	Spikedace
Random-16	#Ind	1	3		58	58	
	#Ind/h	2.93	8.80		170.17	170.17	
Random-8	#Ind	32		12	44	54	23
	#Ind/h	101.67		38.12	139.80	171.57	73.08
Fixed-2	#Ind	72		88	75	195	2
	#Ind/h	272.84		333.47	284.21	738.95	7.59
Total	#Ind	105	3	100	177	307	25
	#Ind/h	125.82	2.93	123.87	198.06	360.23	26.89
	SE	78.85	2.93	105.38	43.96	189.36	23.20

Table 3.2.—Multi-pass depletion estimates of abundance from the fixed reach near the Willow Road Point Road Crossing from 2024. Results include the number of individuals captured in each pass (C1:C3), Carle-Strub 3-pass abundance estimate (N) with 95% confidence intervals of the abundance estimate in parentheses and estimated capture probability (p) with 95% confidence intervals of the estimate of capture probability in parentheses.

Taxa	C1	C2	C3	N	p
Desert Sucker	72	39	17	144 (128.09 – 159-91)	0.51 (0.40 – 0.63)
Speckled Dace	195	94	65	426 (385.26 – 466.74)	0.45 (0.37 – 0.52)
Roundtail Chub	75	29	44	225 (149.27 – 300.73)	0.30 (0.16 – 0.44)
Longfin Dace	88	83	33	278 (220.44 – 335.56)	0.35 (0.24 – 0.47)
Spikedace	2	3	4	14 (0 – 39.65)	0.26 (0 – 0.89)

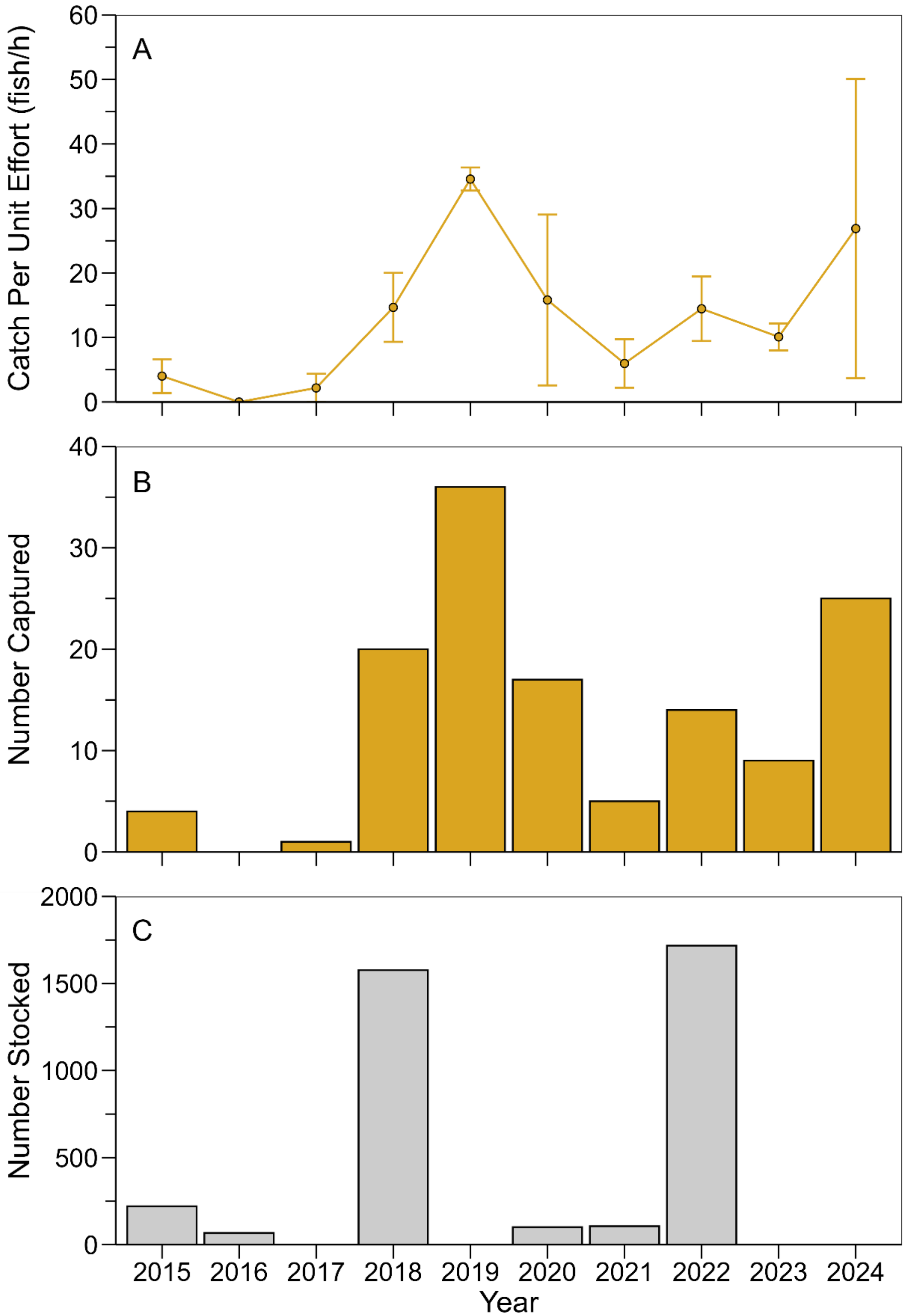

Figure 3.1.—Summary of Spikedace captured and stocked in Spring Creek, AZ, annually from 2015 to 2024 with (A) mean annual backpack electrofishing catch per unit effort (fish/h; first pass only) with standard error bars, (B) total number of fish captured (first pass), and (C) total number of fish stocked.

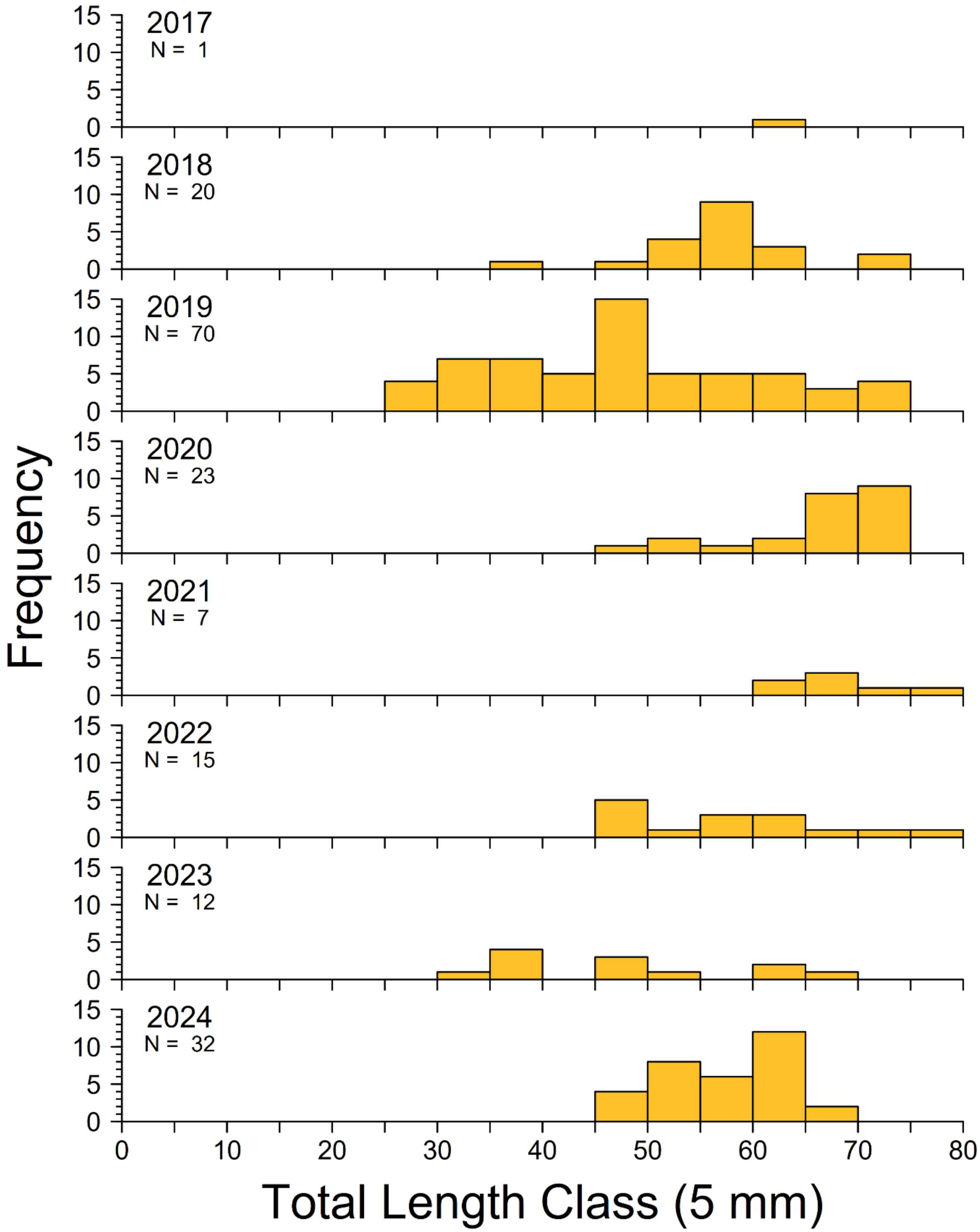

Figure 3.2.—Length frequency distribution of Spikedace captured during annual monitoring in Spring Creek, 2017 through 2024. Total number of fish captured and measured among all passes each year is shown in the top left corner of each panel. Note depletion sampling did not occur in 2017 and 2018.

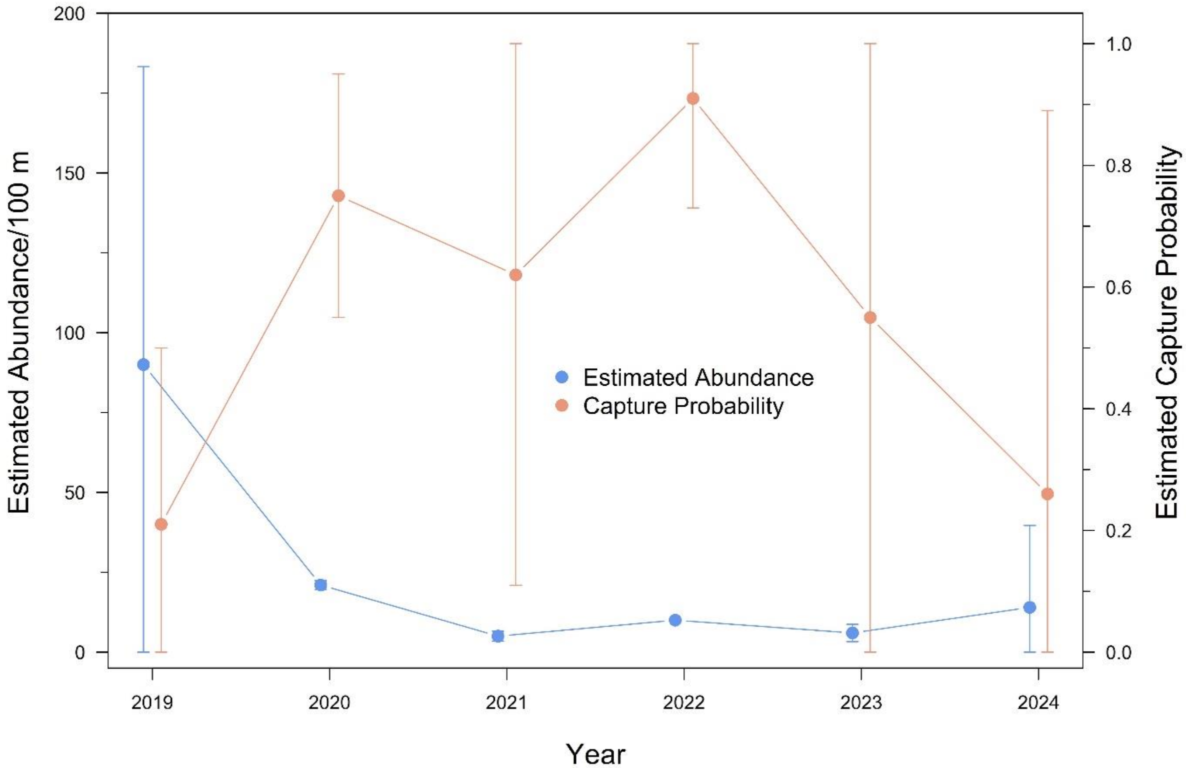

Figure 3.3.—Three-pass depletion estimates of Spikedace abundance per 100 m (blue points and lines) and capture probability (orange points and lines) at the fixed sub-reach in Spring Creek during annual monitoring from 2019 to 2024. Error bars represent the lower and upper bounds of the 95% confidence interval of each estimate.

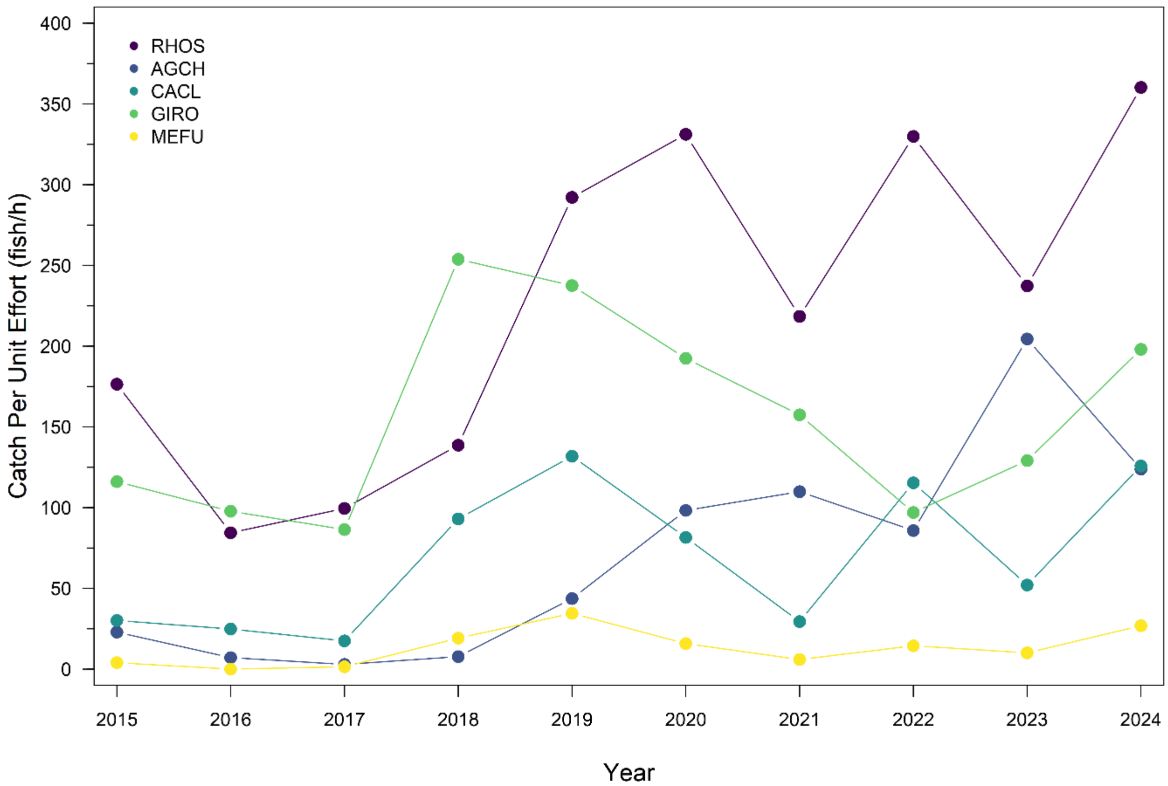

Figure 3.4—Mean annual backpack electrofishing catch per unit effort (fish/h) of select native fish species (AGCH = Longfin Dace, CACL = Desert Sucker, GIRO = Roundtail Chub, MEFU = Spikedace, RHOS = Speckled Dace) captured in Spring Creek, AZ from 2015 to 2024. Standard error bars are not shown to improve clarity of mean catch per unit effort trends.

4. Blue River native fish restoration (Task AZ-2002-3)

Strategic Plan Goals:

- Preventing Extinction and Managing Toward Recovery
  - Goal 3. Remove nonnative aquatic species threats.
  - Goal 4. Replicate populations and their associated native fish community into protected streams and other surface waters.
  - Goal 7. Monitor to quantitatively measure and evaluate project success in improving the status of target species and their habitats.

Recovery Objectives:

- Spikedace recovery objective 6.2.5. Reclaim as necessary to remove non-native fishes.
- Spikedace recovery objective 6.3. Reintroduce Spikedace to selected reaches.
- Spikedace recovery objective 6.4. Monitor success/failure of reintroductions.
- Loach Minnow recovery objective 6.2.5 Reclaim as necessary to remove non-native fishes.
- Loach Minnow recovery objective 6.3. Reintroduce Loach Minnow to selected reaches.
- Loach Minnow recovery objective 6.4. Monitor success/failure of reintroductions.

General Background: The Blue River Native Fish Restoration Project is implemented by the Department, U.S. Forest Service, Reclamation, and USFWS, with the goal of protecting and restoring the entire assemblage of native fishes within the Blue River drainage and benefiting their conservation status within the Gila River Basin (Reclamation 2010). The major components of the project included construction of a fish barrier, mechanical removal of non-native fishes, and translocation and monitoring of federally listed warm-water fishes in the Blue River. The initial focus of the project was the lower 18 km of the Blue River, from Fritz Ranch to the confluence with the San Francisco River (lower Blue River; Figure 4.1). A synthesis of conservation efforts leading to the eradication of nonnative fishes and establishment of Spikedace and Roundtail Chub populations in the lower Blue River through 2019 can be found in Hickerson et al. (2021a). The entire native fish community in the lower Blue River experienced substantial decline following impacts from the Bringham and Cow Canyon fires in 2020 (Shollenberger et al. 2021, Shollenberger et al. 2022). Additional efforts to establish populations of Spikedace and Roundtail Chub are now taking place in the middle and upper Blue River (Figure 4.1).

Middle Blue River

The middle Blue River project area includes 5.5 km of the Blue River from the confluence with McKittrick Creek upstream to The Box (Figure 4.1). The downstream most 1.3 km of the project area is located on private property where permission to sample has not been granted since 2020. Roundtail Chub were initially stocked in the middle Blue River in 2016 (n = 1,194), with subsequent augmentations in 2019 (n = 100) and 2023 (n = 2,249; Figure 4.2). Spikedace were initially stocked in 2017 (n = 448), with subsequent augmentations in 2018 (n = 291) and 2023 (n = 517; Figure 4.3). Monitoring of the Roundtail Chub population began in 2017 when only hoop nets were set. In September, 2018, a monitoring approach was implemented that involved electrofishing ten randomly selected and two fixed 100 m sub-reaches within three river reaches: McKittrick Creek to KP Creek (n = 4), KP Creek to Cole Flat (n = 4), and Cole Flat to the natural waterfall barrier at The Box (n = 4). Hoop netting was dropped from the monitoring protocol following the 2018 monitoring effort.

Upper Blue River

The upper Blue River project area includes 14.8 km of the Blue River from Blue Crossing upstream to the New Mexico state line (Figure 4.1). Spikedace and Roundtail Chub were initially stocked in 2020 when 826 Spikedace and 226 Roundtail Chub were translocated from the lower Blue River due to concerns over post-fire impacts from the Brigham Fire, and stocked at Bobcat Flat and near Upper Blue Campground respectively. The upper Blue River was subsequently augmented with 500 Spikedace and 1,000 Roundtail Chub in 2023. In September 2021, a monitoring approach was implemented that involved electrofishing 12 randomly selected and three fixed 100 m sub-reaches within three river reaches: Blue Crossing to Swafford Canyon (n = 5), Swafford Canyon to Upper Blue Campground (n = 5), and Upper Blue Campground to the New Mexico state line (n = 5).

<u>Results:</u>

Middle Blue River: During September 23-25, 2024, Department staff monitored the native fish community in the middle Blue River. The crew electrofished 10 randomly selected sites and two fixed sites, all of which were 100 meters in length. A total 471 Spikedace, 13 Loach Minnow, 76 Roundtail Chub, 44 Longfin Dace, 1,170 Speckled Dace, 1,928 Desert Sucker, 310 Sonora Sucker, 1 Brown Trout, 1 Fathead Minnow, and 1 narrow-headed gartersnake were captured during the first pass of electrofishing across all sites (Table 4.1).

Total catch and mean relative abundance of Spikedace reached new highs in 2024, although the increase in mean relative abundance was not statistically significant (Table 4.2, Figure 4.3). The strong 2023 year class appeared to survive well and recruited successfully, contributing to the positive population metrics (Figure 4.4). In addition, it appeared that there was some limited reproduction with a few juvenile individuals captured, although far fewer were captured than in 2023 (Figure 4.4). Estimated abundance of Spikedace at the lower fixed site remained high with 129 individuals/100 m (CI 97.13 – 160.87), while capture probability remained relatively low (p = 0.39 [CI 0.23-0.54]; Table 4.3, Figure 4.5). Consistently low estimates of capture probability (<0.5) over the years suggests that single pass electrofishing continues to substantially undercount the true number of Spikedace present (Figure 4.5). Spikedace continue to be absent from the uppermost reaches of the project area, likely due to the relatively steep gradient near The Box (Figure 4.6). Given available data, the Spikedace population likely numbers in the thousands within this reach of the Blue River.

Captures of Roundtail Chub more than doubled compared to 2023 (2023 = 31, 2024 = 76) and mean relative abundance also increased, but not significantly so (Table 4.2, Figure 4.2). No young of year Roundtail Chub were captured in 2024, but there is a relatively strong age class of individuals from 70-120 mm TL that likely comprises some individuals stocked in 2023 that continue to persist within the project area (Figure 4.7). It is not clear why Roundtail Chub appear to have spawned only once or twice within the project area, with 2020 producing the only strong wild year class (Figure 4.7). Estimated abundance of Roundtail Chub remains consistently low at both depletion sites, with estimates of fewer than 20 individuals per 100 m (Figure 4.8). Unlike Spikedace, Roundtail Chub appear to be well distributed throughout the project area, just at lower abundance than other species (Figure 4.9).

Loach Minnow catch remained low through 2024 and was similar to the 2023 catch total (2023 = 17, 2024 = 13), as was relative abundance (Figure 4.10). Juvenile Loach Minnow were not detected in 2024 (Figure 4.11). The Loach Minnow population appears to be struggling to recover fully to the strength seen in 2020, which may be an anomalous baseline for comparison given that mean relative abundance in 2018 and 2019 was also below 20 fish/h (Figure 4.10). Estimated Loach Minnow abundance at the downstream fixed site was over 100 individuals per 100 m in 2020, but no Loach Minnow have been captured at the site since 2022, suggesting that Loach Minnow are also now more patchily distributed within the project area (Figure 4.12, Figure 4.13). Interestingly, Loach Minnow relative abundance from 2018 to 2024 is highly and significantly correlated with Longfin Dace relative abundance ($r = 0.987$, $t = 13.54$, $P < 0.001$). A habitat link that might result in these two species responding to environmental conditions similarly isn't well established in the literature, however it is likely that both species are struggling with lingering post-fire impacts from the Cow Canyon Fire and the substantial variability in flow regime since 2020, either individually or in combination.

Other native fish taxa in the middle Blue River did not significantly change in relative abundance from 2023 to 2024 (Table 4.2, Figure 4.14). All other native fish taxa remained broadly distributed within the project area (Figure 4.15). However, the lack of a strong 2024 year class for Spikedace, Loach Minnow, and Roundtail Chub was also consistent with the other native fish taxa where relatively few young of year individuals were captured.

Upper Blue River: During September 16-19, 2024, Department staff, assisted by Reclamation staff, monitored the native fish community in the upper Blue River, between the New Mexico state line and Blue Crossing Campground. The crew sampled 12 randomly selected sites and three fixed sites, all of which were 100 meters in length. A total of 23 Loach Minnow, 1,219 Desert Sucker, 621 Longfin Dace, 349 Sonora Sucker, 1,857 Speckled Dace, and 55 Brown Trout during the first pass of electrofishing across all sites (Table 4.4). Spikedace were not captured (Figure 4.16) and a single Roundtail Chub was captured during the second pass of electrofishing at a fixed site (Figure 4.17, Table 4.5). The single Roundtail Chub captured was an adult (251 mm TL) that is likely from the initial stocking in 2020, suggesting at least some individuals have been able to persist for five years. The stocking of Spikedace and Roundtail Chub appears to have failed to bolster the existing populations in the upper Blue River as none of the fish stocked in 2023 were recaptured in 2024. Interannual survival of both species has been documented within the project area, and spawning has been documented for Spikedace. It seems likely that environmental conditions within the project area since the initial stocking have been sufficient for survival of some individuals, but marginal enough to preclude the widespread persistence and reproduction of stocked individuals. The upper Blue River was not impacted by either of the 2020 fires, so it seems most likely that the extreme variability in flow regime, between extreme drought and extreme floods, has proven difficult for these taxa to establish populations in this headwaters reach of the Blue River (Figure 4.18).

Loach Minnow catch also declined in 2024 relative to 2023, with mean relative abundance significantly decreasing from 31.78 fish/h in 2023 to 4.00 fish/h in 2024 ($W = 58$, $P = 0.009$; Table 4.6, Figure 4.19). Spawning appeared to occur in 2024 as several juvenile individuals were captured, however the strong 2023 year class didn't seem to recruit as well as expected (Figure 4.20). Distribution of Loach Minnow was also more patchy than in the past, with Loach Minnow only captured at 7 of 15 subreaches (Figure 4.21). Interestingly, three Loach Minnow were captured in just a few minutes of electrofishing effort near Blue Crossing Campground in July for a fish health assessment, so it is possible a flood (or some other disturbance) event between July and September negatively impacted the Loach Minnow population. Much like the translocated taxa, it is difficult to explain why Loach Minnow have generally declined since monitoring began in 2021, other than the hydrologic extremes experienced over the same period (Figure 4.18). Similar to the middle Blue River reach, Loach Minnow relative abundance is highly, but not significantly, correlated with Longfin Dace relative abundance ($r = 0.921$, $t = 3.392$, $P = 0.08$), suggesting that there is likely a related mechanism acting on the relative abundance of these taxa throughout the river.

Relative abundance for other native fish taxa in the upper Blue River did not significantly differ from 2023, with the exception of Speckled Dace which significantly declined (Table 4.6, Figure 4.22). The 2023 year class was strong for all native fish taxa, presumably because of the above average spring discharge, however that didn't translate to broad increases in native fish relative abundance in this reach, suggesting that environmental conditions either impacted survival of fish over the last year, facilitated dispersal from the monitoring reach, or potentially both.

Lastly, the number of Brown Trout captured exhibited a five-fold increase relative to 2023 (2023 = 11, 2024 = 55). Mean relative abundance also significantly increased from 2.27 fish/h in 2023 to 9.75 fish/h in 2024 (Table 4.6). The majority of Brown Trout captured were between 60 – 135 mm in total length (mean 127.79 mm TL), which represented a significant decrease in mean total length compared to 2023 (mean = 206.82 mm TL; $W = 132$, $P = 0.001$; Figure 4.23). The apparent increase in Brown Trout relative abundance is notable given that none of the native fish taxa in the upper Blue River experienced increases in relative abundance between 2023 and 2024. Potential explanations for the increase in Brown Trout relative abundance could be that primarily juvenile fish dispersed downstream from Campbell Blue Creek since 2023, or that prevailing environmental conditions since the last monitoring effort in 2023 favored Brown Trout at the expense of native fish taxa.

Recommendations: The Spikedace and Roundtail Chub populations in the middle Blue River are continuing to recover from the Cow Canyon fire, and are on track for establishment in the near future if consistent recruitment can be documented. Additional monitoring should occur in 2025 to confirm the trajectory of these populations and monitor the response of Loach Minnow.

Translocated populations of Spikedace and Roundtail Chub both appear to be struggling in the upper Blue River, as few individuals have been recaptured during monitoring over the last few years despite substantial stocking in 2023. Additional translocations of Spikedace are required in the upper Blue River to evaluate whether a population of Spikedace can establish under less extreme hydrologic conditions (both floods and drought) than those experienced since Spikedace were initially stocked in 2020. Without stocking, there is little opportunity for the Spikedace population to become established in the near term, given how few adults have been captured to date.

Evidence from monitoring throughout all three reaches of the Blue River suggests that Roundtail Chub reproduction occurs sporadically. It takes several years, likely five or more, to recruit into the population which makes assessing short-term population trends difficult. The continued persistence of adult Roundtail Chub in both the upper and middle Blue River reaches through extreme hydrologic conditions of the last few years is evidence that chub are resilient to these extreme conditions in both reaches and will likely be able to establish in time. Continued monitoring is needed to document reproduction and determine if additional supplemental stockings will be necessary in order for populations of chub to establish.

Preliminary reports from monitoring in the lower Blue River in 2024 indicates that Loach Minnow have been detected for the first time since 2020 (P. Reap, Marsh and Associates, personal communication). This detection suggests that Loach Minnow are again able to disperse to and persist in the lower Blue River. Translocation of Loach Minnow to the lower Blue River from ARCC should be considered, as similar efforts following the Wallow Fire helped to quickly reestablish the Loach Minnow population. Reestablishment of the Loach Minnow population in the lower Blue River is important because of the apparent struggles of the Loach Minnow population in the middle and upper reaches of the river. Supplemental stocking of the existing Loach Minnow population in the upper and middle Blue River should also be considered if population trends don’t improve in the near future.

Tables and Figures:

Table 4.1—Summary of fish captured during the first pass of backpack electrofishing within each monitoring reach in the middle Blue River during annual monitoring from September 2024. Shown for each reach is the number of sub-reaches sampled (N), number of fish captured (#Ind), the mean relative abundance (number of fish captured per hour of electrofishing effort; #Ind/h) and standard error of mean relative abundance (SE).

Reach	N	Statistic	Loach Minnow	Roundtail Chub	Spikedace	Desert Sucker	Longfin Dace	Sonora Sucker	Speckled Dace	Fathead Minnow	Brown Trout
1	4	#Ind	3	30	385	998	29	90	553	1.00	
		#Ind/h	1.29	13.00	181.90	480.16	13.79	39.95	268.84	0.39	
		SE	0.81	6.26	39.90	41.39	5.67	13.40	42.40	0.39	
2	4	#Ind	6	18	86	527	4	120	353		
		#Ind/h	4.48	12.46	54.65	363.48	2.57	82.81	241.83		
		SE	3.04	2.39	32.72	55.59	1.83	12.16	64.05		
3	4	#Ind	4	28		403	11	100	264		1
		#Ind/h	3.47	22.55		331.15	8.79	80.08	210.52		0.72
		SE	1.77	3.52		45.28	3.72	3.99	31.00		0.72
Total	12	#Ind	13	76	471	1928	44	310	1170	1	1
		#Ind/h	3.01	15.21	94.01	404.01	8.79	64.27	245.19	0.16	0.18
		SE	1.14	2.94	29.65	31.99	2.81	8.94	27.17	0.16	0.18

Table 4.2.—Wilcoxon signed rank test (non-parametric) and two-sample t-test (parametric) statistics evaluating changes in mean relative abundance of all fish taxa captured in the middle Blue River between 2023 and 2024. Significant values are indicated with an asterisk (*).

Taxa	Test Type	Alternative	*t*	*W*	*P*
Spikedace	Non-parametric	Two-sided		76.5	0.81
Roundtail Chub	Non-parametric	Two-sided		103.0	0.08
Loach Minnow	Non-parametric	Two-sided		70.0	0.93
Longfin Dace	Non-parametric	One-sided		35.0	0.02*
Speckled Dace	Parametric	Two-sided	-1.53		0.14
Desert Sucker	Parametric	Two-sided	0.51		0.62
Sonora Sucker	Parametric	Two-sided	-0.60		0.55

Table 4.3.—Three-pass depletion estimates of abundance for all fish species captured per 100 m at each fixed sub-reach in the middle Blue River during annual monitoring in 2024. Results include the number of individuals captured during each pass (C1, C2, C3), Carle-Strub three pass abundance estimate (N) and capture probability (*p*). The 95% confidence intervals for the estimate of abundance and capture probability are included in parentheses.

Sub-reach	Species	C1	C2	C3	N	*p*
1F28	Spikedace	51	27	22	129 (97.13 - 160.87)	0.39 (0.23 - 0.54)
	Roundtail Chub	4	2	2	8 (5.31 - 10.69)	0.57 (0.12 - 1.00)
	Speckled Dace	125	50	28	222 (206.39 - 237.61)	0.55 (0.47 - 0.64)
	Desert Sucker	249	98	72	479 (447.20 - 510.80)	0.50 (0.43 - 0.56)
	Sonora Sucker	25	8	7	43 (36.47- 49.53)	0.56 (0.37 - 0.76)
	Longfin Dace	4	5	5	23 (0.00 - 57.77)	0.25 (0.00 - 0.75)
3F04	Roundtail Chub	13	5	2	20 (17.95 - 22.05)	0.69 (0.46 - 0.92)
	Loach Minnow	0	1	0	1 (0 - 2.44)	0.50 0.00 - 1.00)
	Speckled Dace	88	18	6	113 (110.31 - 115.69)	0.77 (0.69 - 0.85)
	Desert Sucker	111	24	24	168 (158.57 - 177.43)	0.62 (0.53 - 0.71)
	Sonora Sucker	40	15	5	62 (57.61 - 66.39)	0.66 (0.52 - 0.80)
	Longfin Dace	7	5	3	17 (9.89 - 24.11)	0.47 (0.10 - 0.84)

Table 4.4.—Summary of fish captured during the first pass of backpack electrofishing within each monitoring reach in the upper Blue River during annual monitoring in September 2024. Shown for each reach is the number of sub-reaches sampled (N), number of fish captured (#Ind), the mean relative abundance (number of fish captured per hour of electrofishing effort; #Ind/h) and standard error of mean relative abundance (SE).

Reach	N	Statistic	Loach Minnow	Desert Sucker	Longfin Dace	Sonora Sucker	Speckled Dace	Brown Trout
1	5	#Ind	10	367	289	110	932	5
		#Ind/h	6.22	228.74	185.80	68.14	575.83	3.15
		SE	4.76	92.53	82.70	26.32	263.34	2.46
2	5	#Ind	10	394	130	127	408	25
		#Ind/h	4.59	179.94	58.84	61.55	185.43	11.07
		SE	3.34	46.25	9.28	18.77	47.99	3.93
3	5	#Ind	3	458	202	112	517	25
		#Ind/h	1.36	234.49	97.89	62.41	265.66	17.04
		SE	0.86	22.74	33.99	12.62	62.24	8.11
Total	15	#Ind	23	1219	621	349	1857	55
		#Ind/h	4.06	214.39	114.18	64.03	342.31	10.42
		SE	1.89	33.33	31.16	10.74	96.00	3.26

Table 4.5.—Three-pass depletion estimates of abundance for all fish species captured per 100 m at each fixed sub-reach in the upper Blue River during annual monitoring in 2024. Results include the number of individuals captured during each pass (C1, C2, C3), Carle-Strub three pass abundance estimate (N) and capture probability (*p*). The 95% confidence intervals for the estimates of abundance and capture probability are included in parentheses.

Sub-reach	Taxa	C1	C2	C3	N	*p*
1F46	Roundtail Chub	0	1	0	1 (0.00 - 2.44)	0.50 (0.00 - 1.00)
	Speckled Dace	78	53	37	239 (176.50 - 301.50)	0.33 (0.20 - 0.46)
	Desert Sucker	59	34	19	134 (111.44 - 156.56)	0.45 (0.31 - 0.58)
	Sonora Sucker	36	12	14	73 (57.42 - 88.58)	0.46 (0.28 - 0.64)
	Longfin Dace	27	6	4	37 (34.81 - 39.19)	0.73 (0.57 - 0.88)
	Brown Trout	4	1	1	6 (4.70 - 7.30)	0.67 (0.23 - 1.00)
2F47	Speckled Dace	66	17	12	99 (92.81 - 105.19)	0.64 (0.53 - 0.75)
	Desert Sucker	66	20	14	106 (97.86 - 114.14)	0.60 (0.49 - 0.72)
	Sonora Sucker	10	3	6	23 (11.48 - 34.52)	0.41 (0.06 - 0.77)
	Longfin Dace	29	5	5	40 (36.99 - 43.01)	0.68 (0.52 - 0.85)
	Brown Trout	12	2	0	14 (13.66 - 14.34)	0.88 (0.70 - 1.00)
3F44	Loach Minnow	2	1	1	4 (2.10 - 5.90)	0.57 (0.00 - 1.00)
	Speckled Dace	134	81	44	317 (278.44 - 355.56)	0.43 (0.34 - 0.52)
	Desert Sucker	114	40	29	202 (185.92 - 218.08)	0.54 (0.45 - 0.64)
	Sonora Sucker	15	16	4	41 (28.99 - 53.01)	0.45 (0.21 - 0.70)
	Longfin Dace	56	34	19	133 (108.11 - 157.89)	0.43 (0.29 - 0.57)
	Brown Trout	0	1	0	1 (0 - 2.44)	0.5 (0.00 - 1.00)

Table 4.6.—Wilcoxon signed rank test (non-parametric) and two-sample t-test (parametric) statistics evaluating changes in mean relative abundance of all fish taxa captured in the upper Blue River between 2023 and 2024. Significant values are indicated with an asterisk (*).

Taxa	Test Type	Alternative	*t*	*W*	*P*
Spikedace	Non-parametric	Two-sided		97.5	0.16
Roundtail Chub	Non-parametric	Two-sided		96.0	0.26
Loach Minnow	Non-parametric	One-sided		58.0	<0.01*
Longfin Dace	Non-parametric	Two-sided		81.0	0.20
Speckled Dace	Non-parametric	Two-sided		53.0	0.01*
Desert Sucker	Parametric	Two-sided	-0.38		0.71
Sonora Sucker	Parametric	Two-sided	-1.58		0.13
Brown Trout	Non-parametric	One-sided		169.5	<0.01*

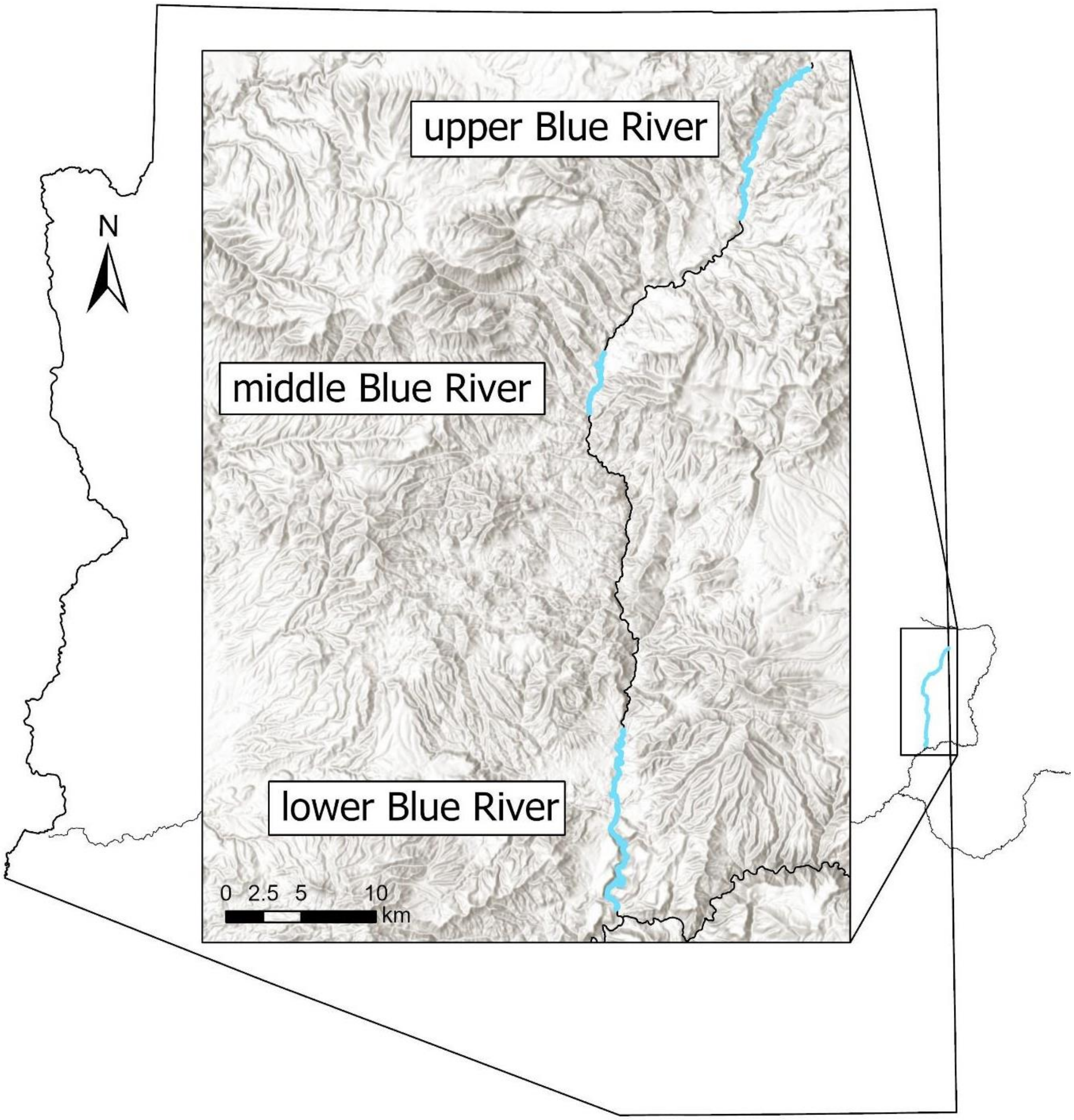

Figure 4.1.—Map showing the upper (New Mexico border downstream to Blue Crossing Campground), middle (The Box downstream to Fritz Ranch), and lower (Fritz Ranch downstream to the barrier) project areas of the Blue River.

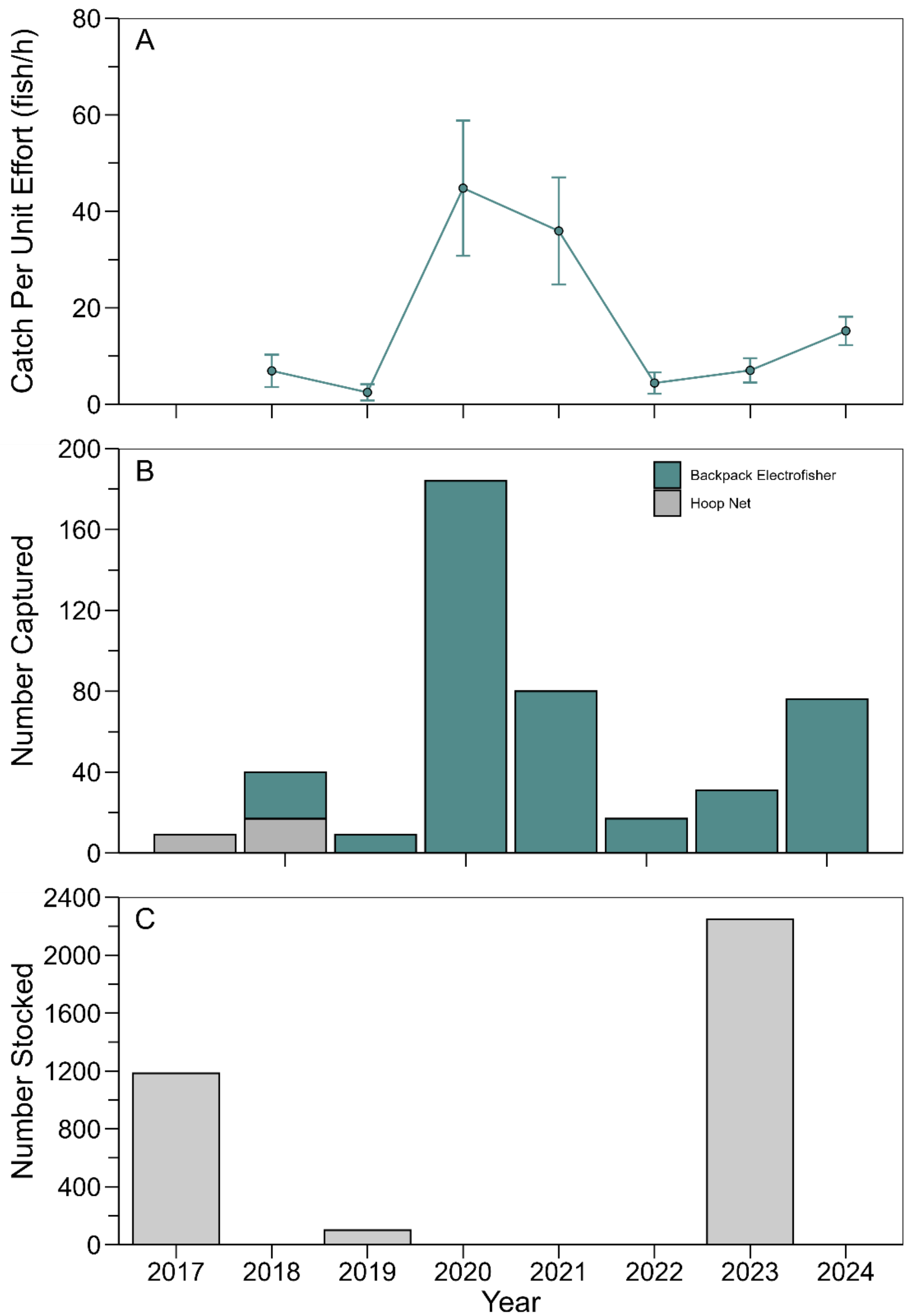

Figure 4.2.—Summary of Roundtail Chub captured and stocked in the middle Blue River, annually from 2017 to 2024 with (A) mean catch per unit effort (fish/h; first pass only) for backpack electrofishing with standard error bars, (B) total number of fish captured by gear type (hoop nets in gray, backpack electrofishing in black; first pass only), and (C) total number of fish stocked. Catch per unit effort is not displayed for hoop nets in panel A because it was less than one fish per hour.

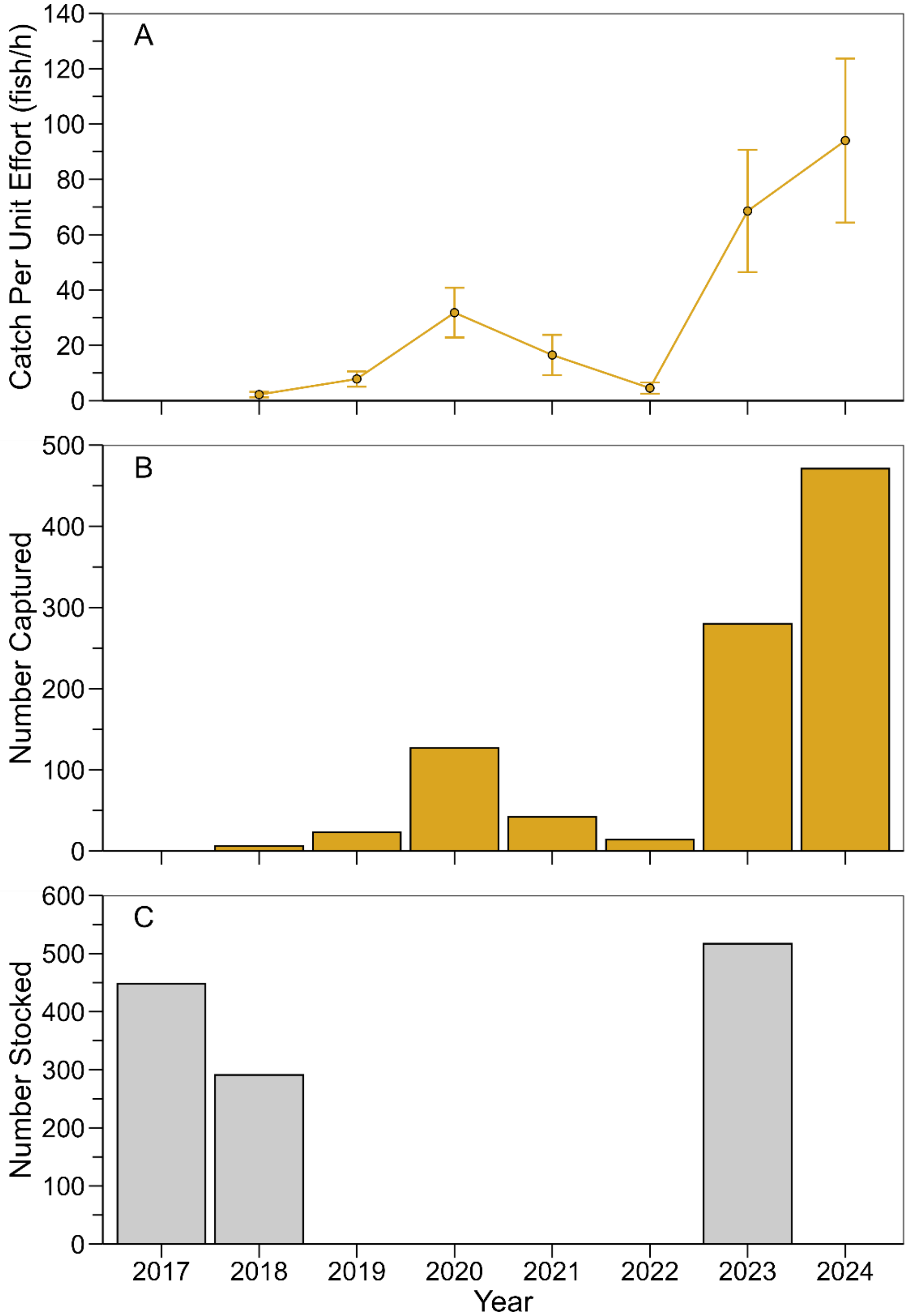

Figure 4.3.—Summary of Spikedace captured and stocked in the middle Blue River, annually from 2017 to 2024 with (A) mean annual backpack electrofishing catch per unit effort (fish/h; first pass only) with standard error bars, (B) total number of fish captured (first pass), and (C) total number of fish stocked.

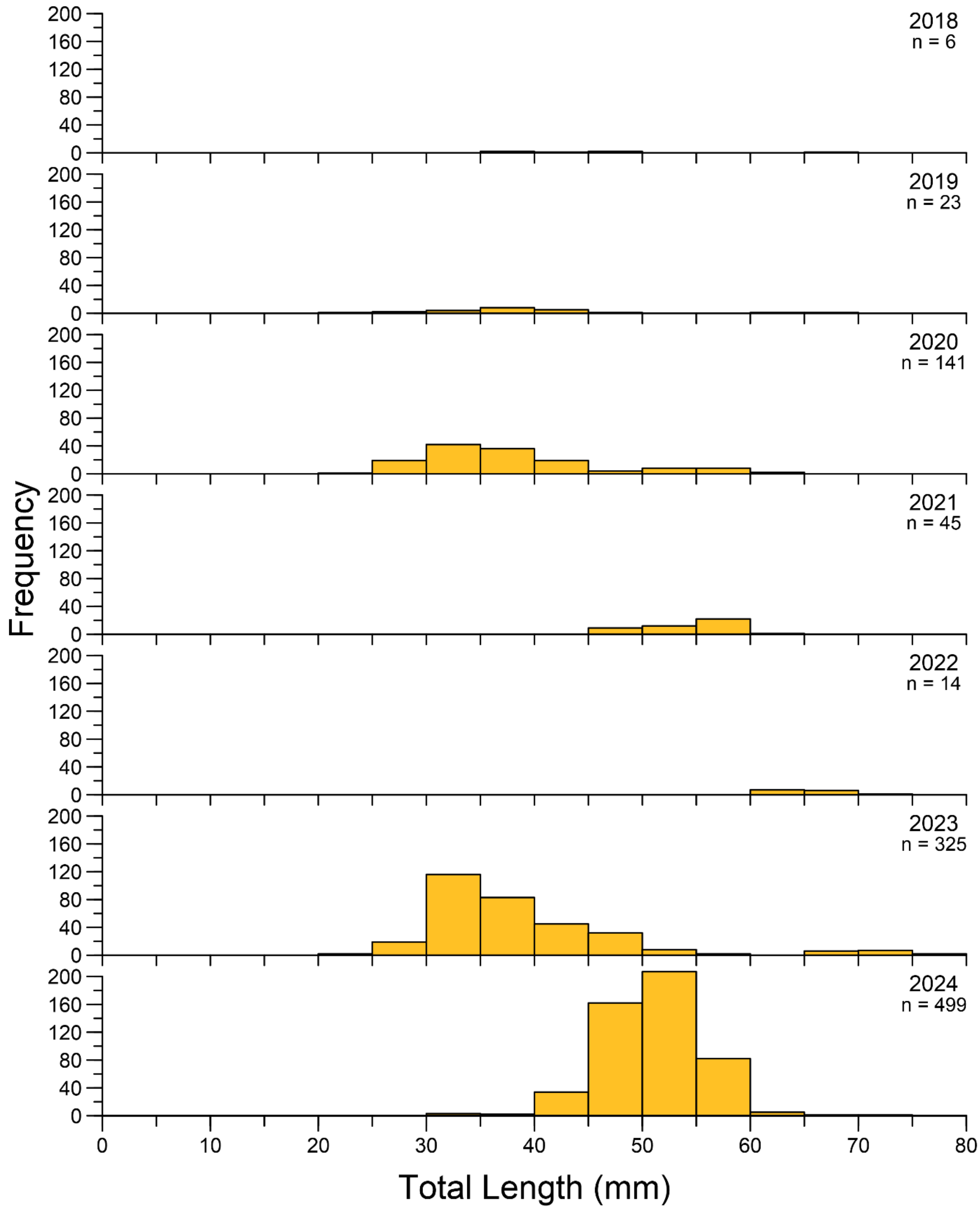

Figure 4.4.—Length frequency distribution of Spikedace captured during annual monitoring in the middle Blue River, from 2018 to 2024. Total number of fish captured and measured among all passes each year is shown in the top right corner of each panel. Note depletion sampling did not occur in 2018.

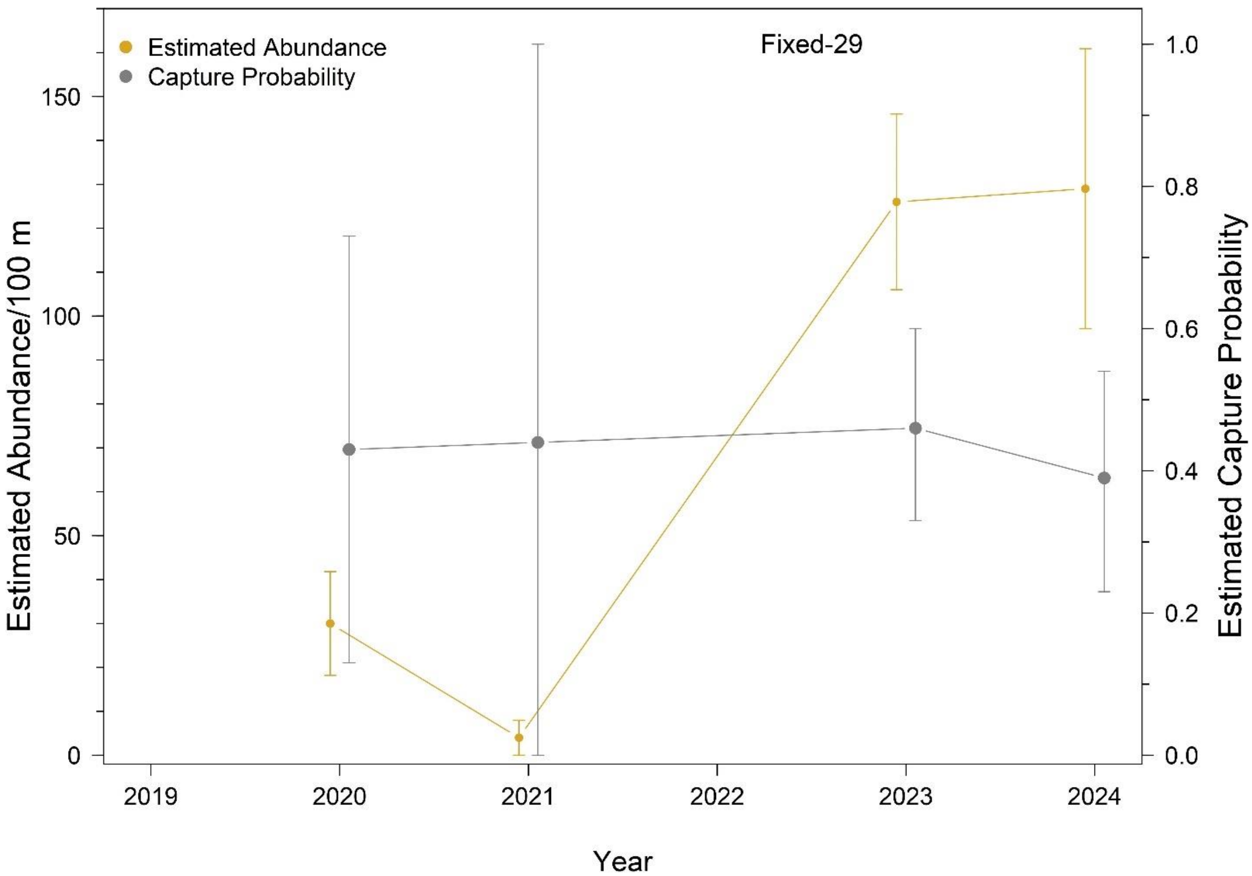

Figure 4.5.—Three-pass depletion estimates of Spikedace abundance per 100 m (yellow points and lines) and capture probability (grey points and lines) at the downstream (Fixed-29) sub-reach in the middle Blue River during annual monitoring from 2019 to 2024. Bars represent the lower and upper bounds of the 95% confidence interval of each estimate.

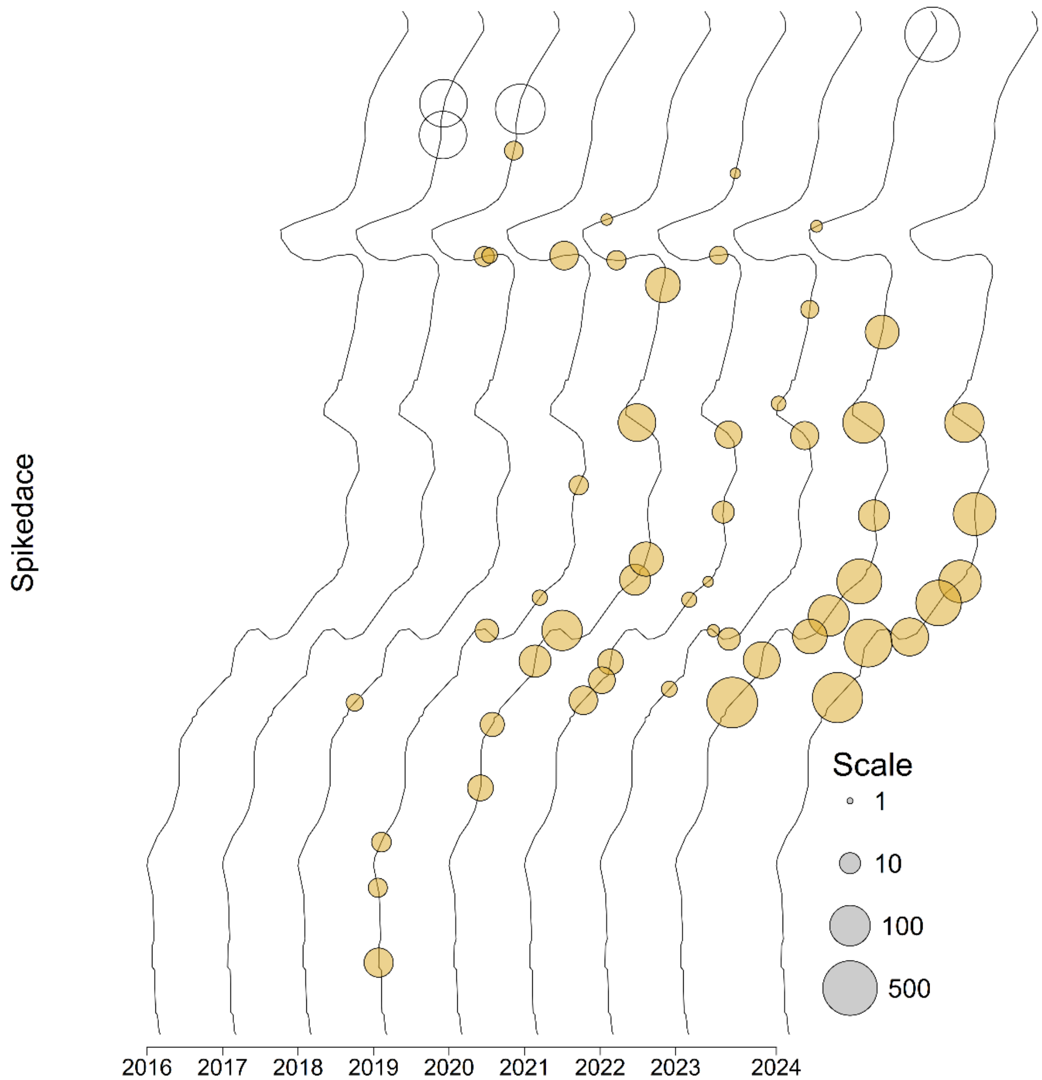

Figure 4.6.—Spikedace stocking locations (open circles) and mean backpack electrofishing relative abundance (CPUE, fish/h) at each monitoring site in the middle Blue River from 2016 to 2024. Size of points indicates either the number of fish stocked or the relative abundance during monitoring at a particular location.

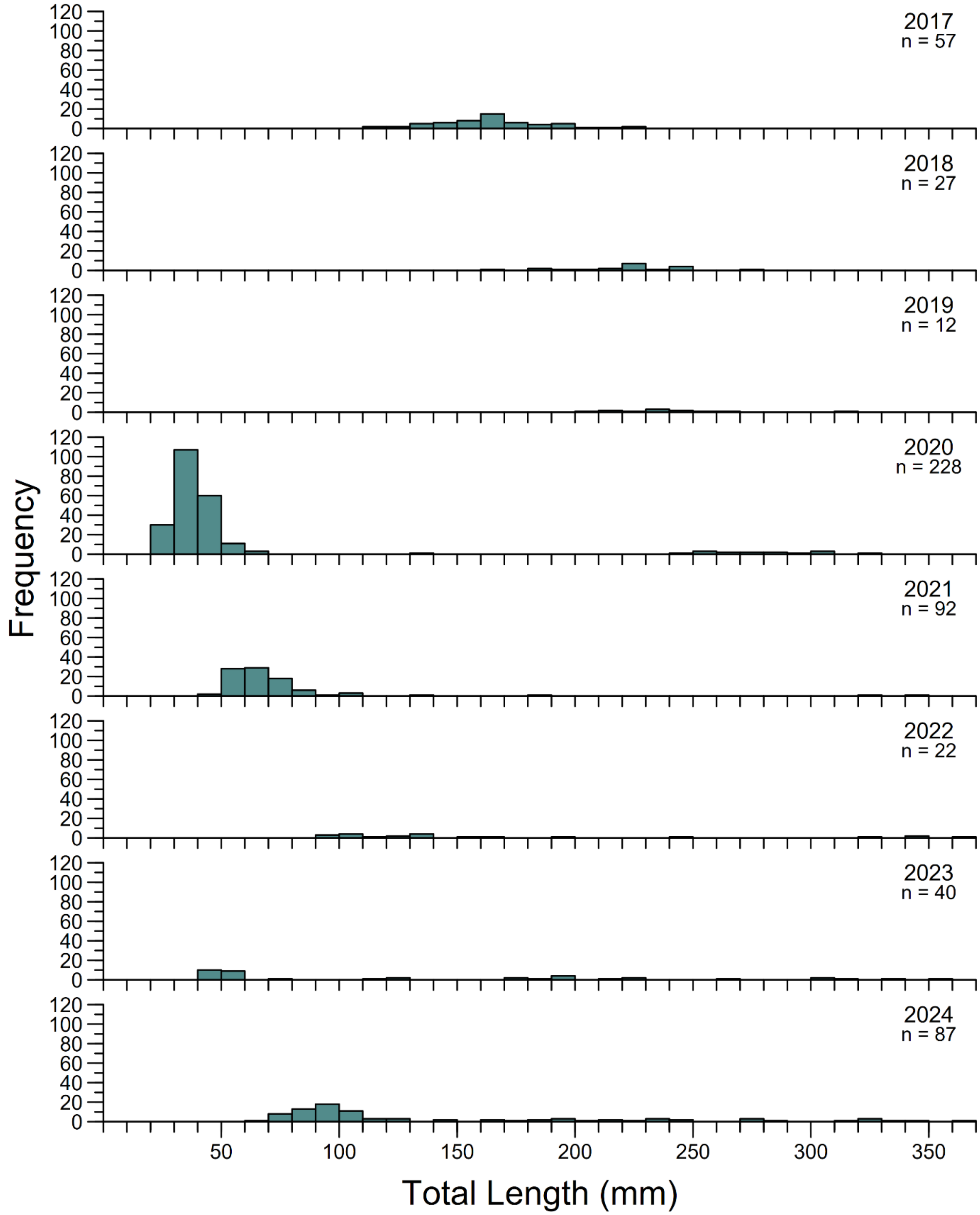

Figure 4.7.—Length frequency distribution of Roundtail Chub captured during annual monitoring in the middle Blue River, from 2017 to 2024. Total number of fish captured and measured among all passes each year is shown in the top right corner of each panel. Note depletion sampling did not occur in 2017 and 2018.

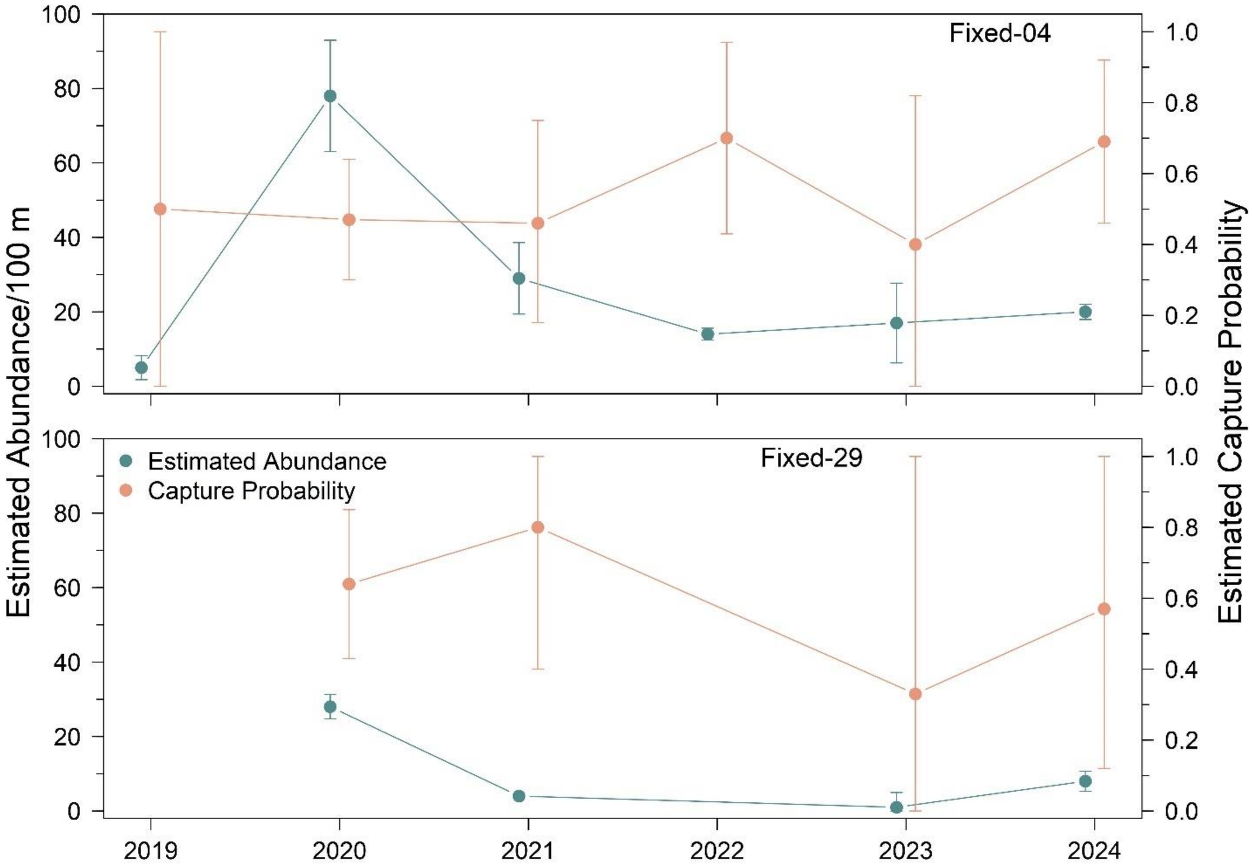

Figure 4.8.—Three-pass depletion estimates of Roundtail Chub abundance per 100 m (blue points and lines) and capture probability (orange points and lines) at the upstream (Fixed-04) and downstream (Fixed-29) sub-reaches in the middle Blue River during annual monitoring from 2019 to 2024. Bars represent the lower and upper bounds of the 95% confidence interval of each estimate.

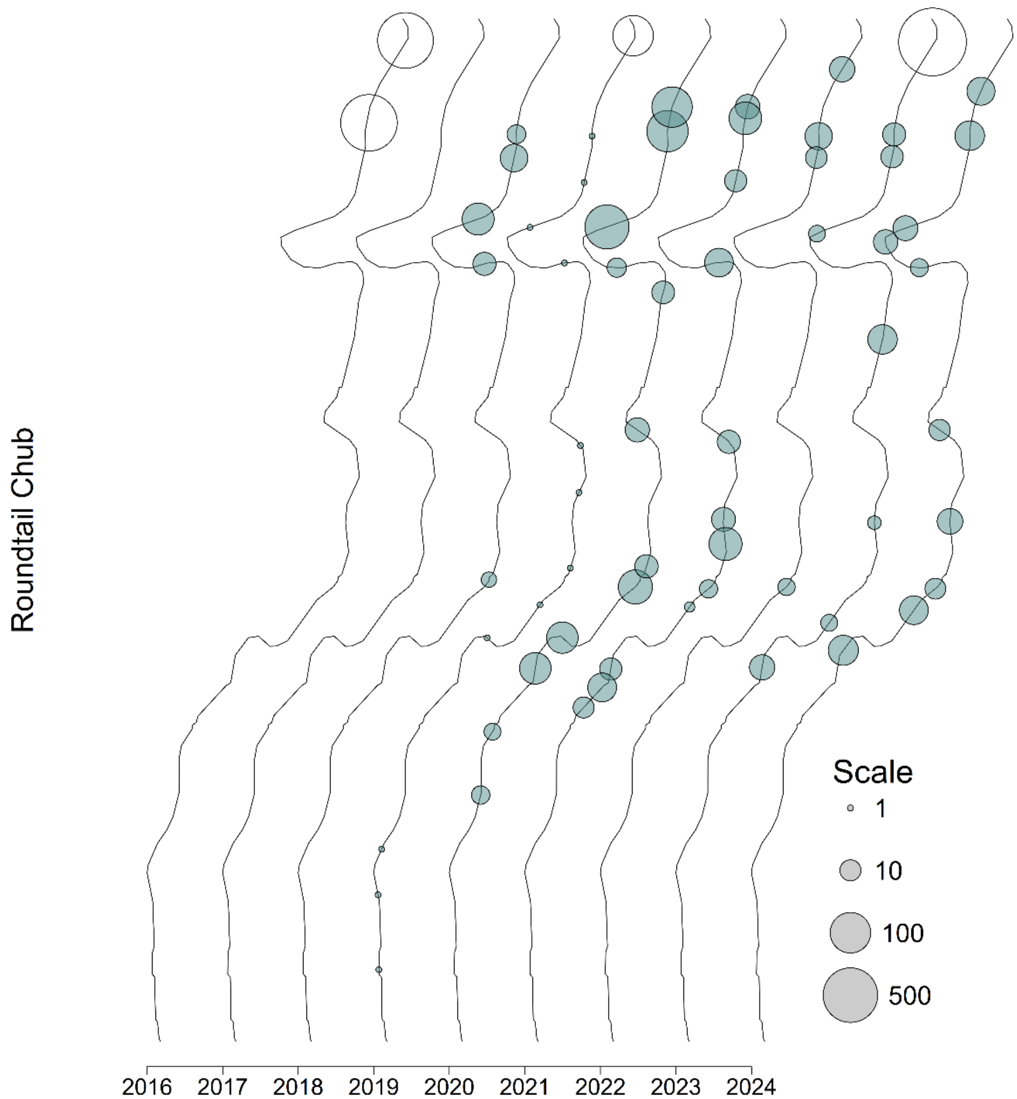

Figure 4.9.—Roundtail Chub stocking locations (open circles) and mean backpack electrofishing relative abundance (CPUE, fish/h) at each monitoring site in the middle Blue River from 2016 to 2024. Size of points indicates either the number of fish stocked or the relative abundance during monitoring at a particular location.

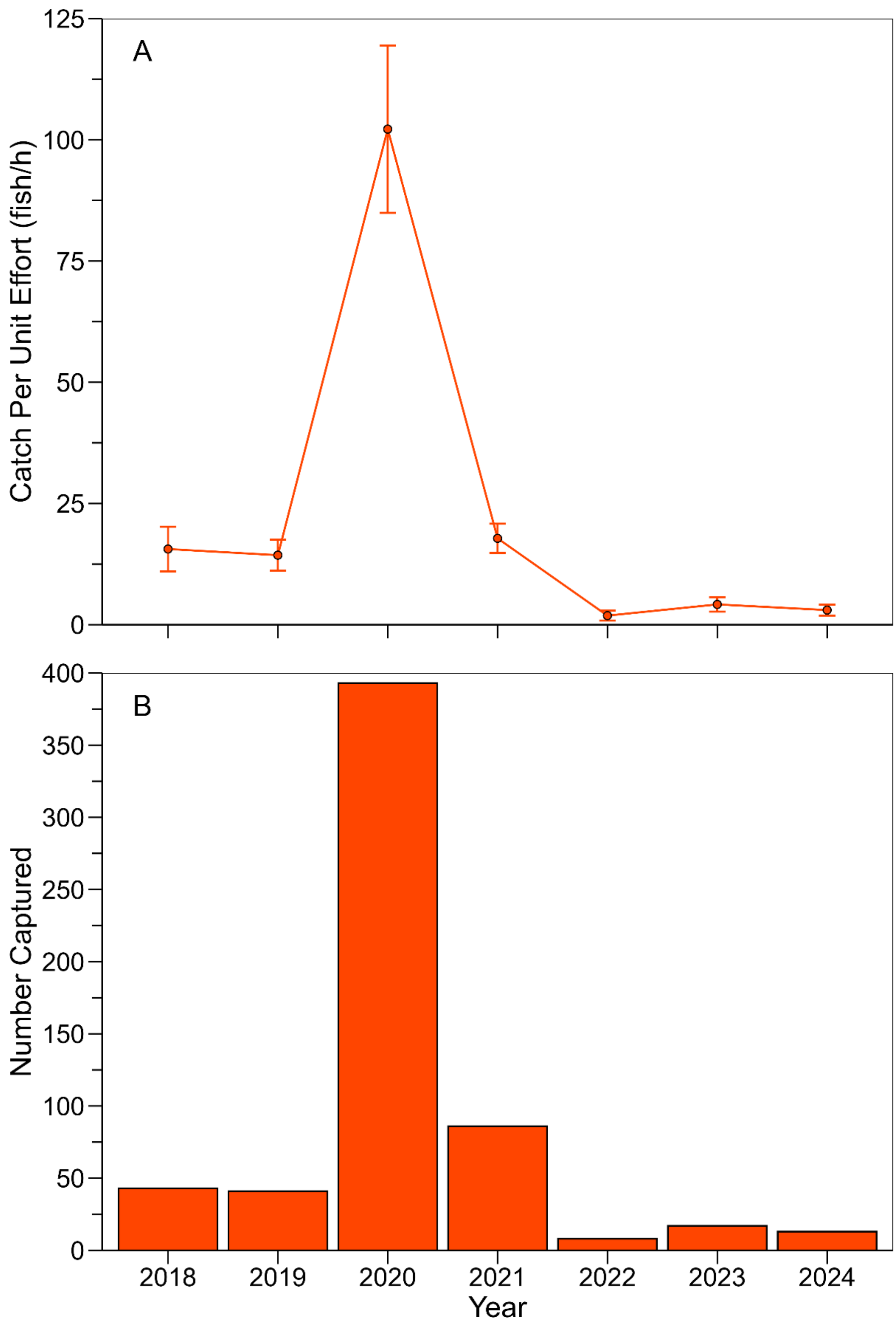

Figure 4.10.—Summary of Loach Minnow captured in the middle Blue River, annually from 2018 to 2024 with (A) mean annual backpack electrofishing catch per unit effort (fish/h; first pass only) with standard error bars, and (B) total number of fish captured (first pass).

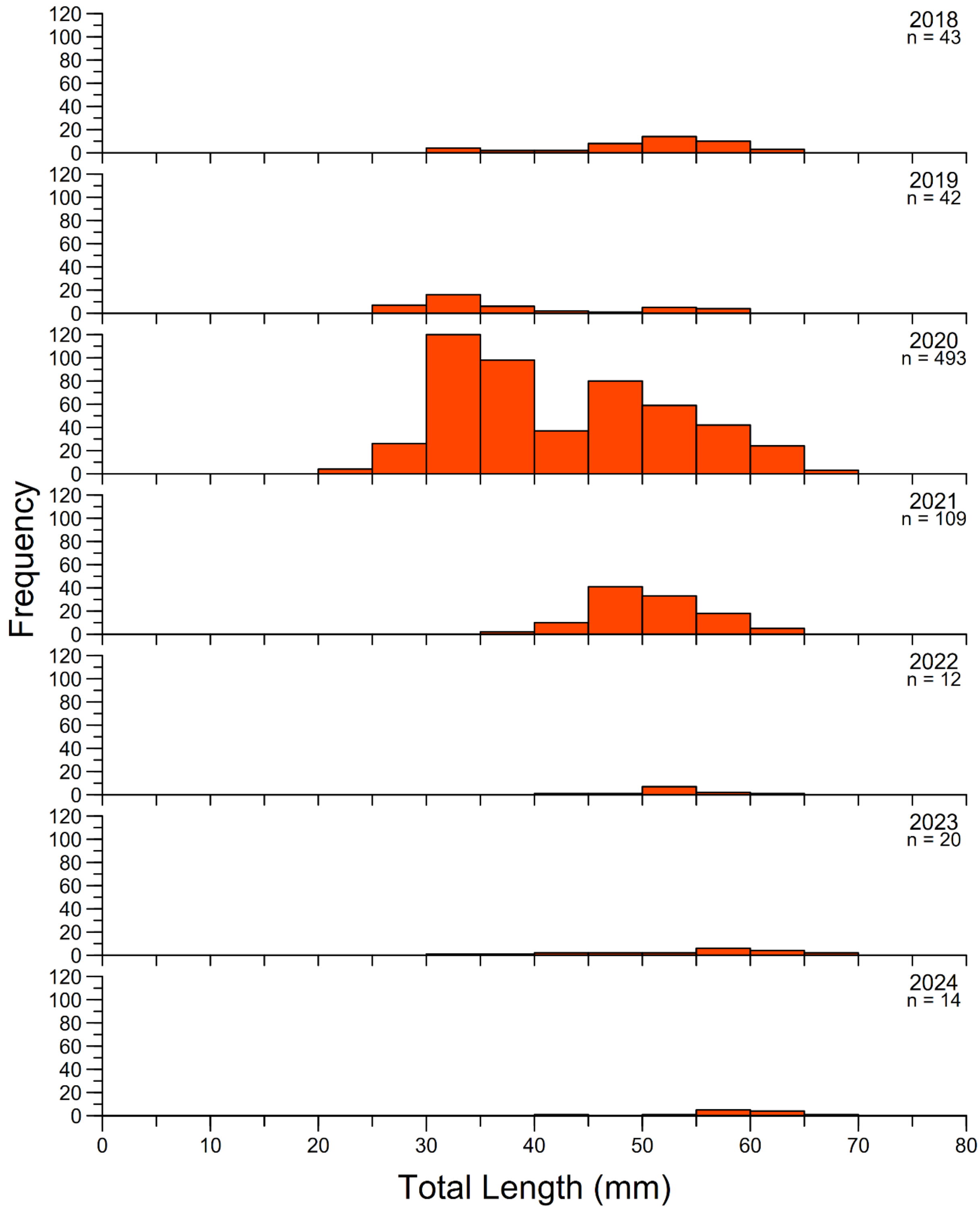

Figure 4.11.—Length frequency distribution of Loach Minnow captured during annual monitoring in the middle Blue River, from 2018 to 2024. Total number of fish captured and measured among all passes each year is shown in the top right corner of each panel. Note depletion sampling did not occur in 2018.

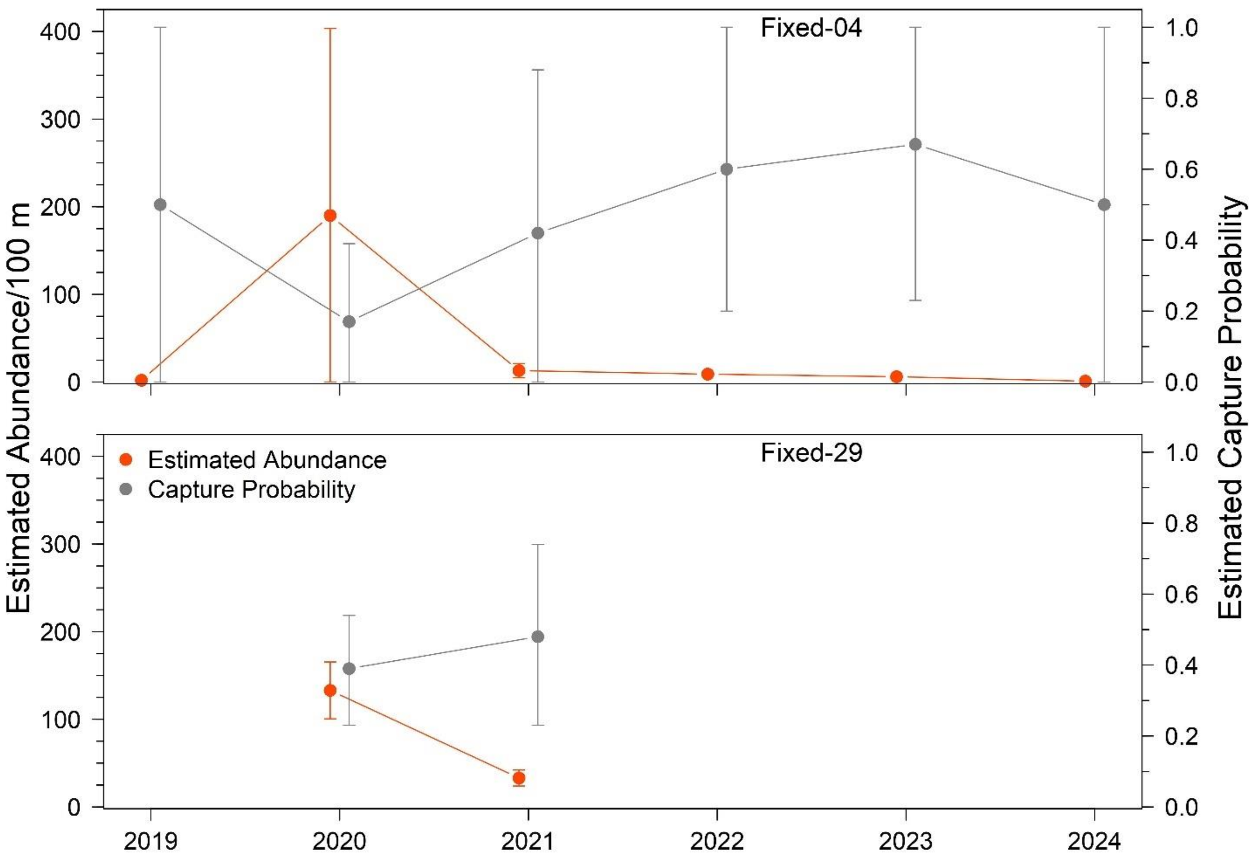

Figure 4.12.—Three-pass depletion estimates of Loach Minnow abundance per 100 m (orange points and lines) and capture probability (grey points and lines) at the upstream (Fixed-04) and downstream (Fixed-29) sub-reaches in the middle Blue River during annual monitoring from 2019 to 2024. Bars represent the lower and upper bounds of the 95% confidence interval of each estimate.

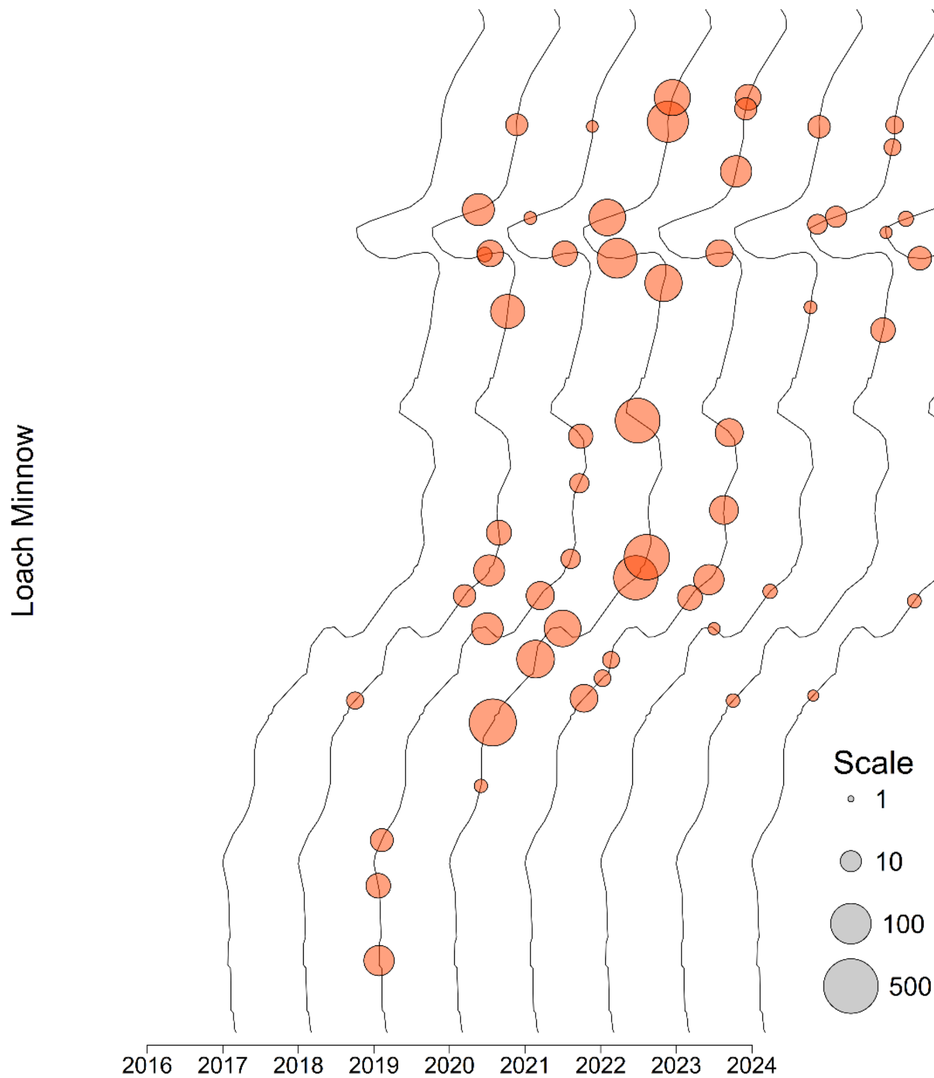

Figure 4.13.—Loach Minnow mean backpack electrofishing relative abundance (CPUE, fish/h) at each monitoring site in the middle Blue River from 2016 to 2024. Size of points indicates the relative abundance during monitoring at a particular location.

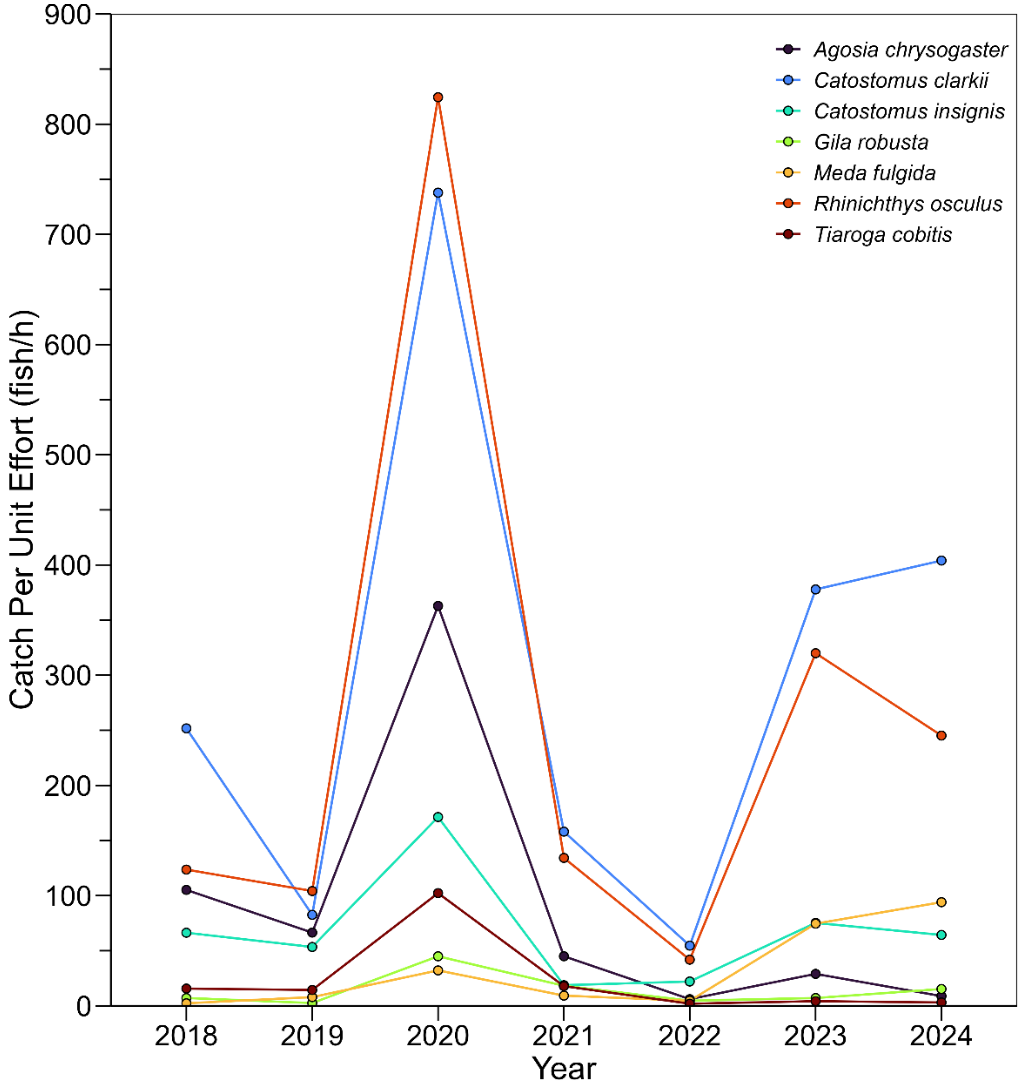

Figure 4.14.—Mean annual backpack electrofishing catch per unit effort (fish/h) of all native fish species captured in the middle Blue River, AZ from 2018 to 2024. Standard error bars are not shown to improve clarity of mean catch per unit effort trends.

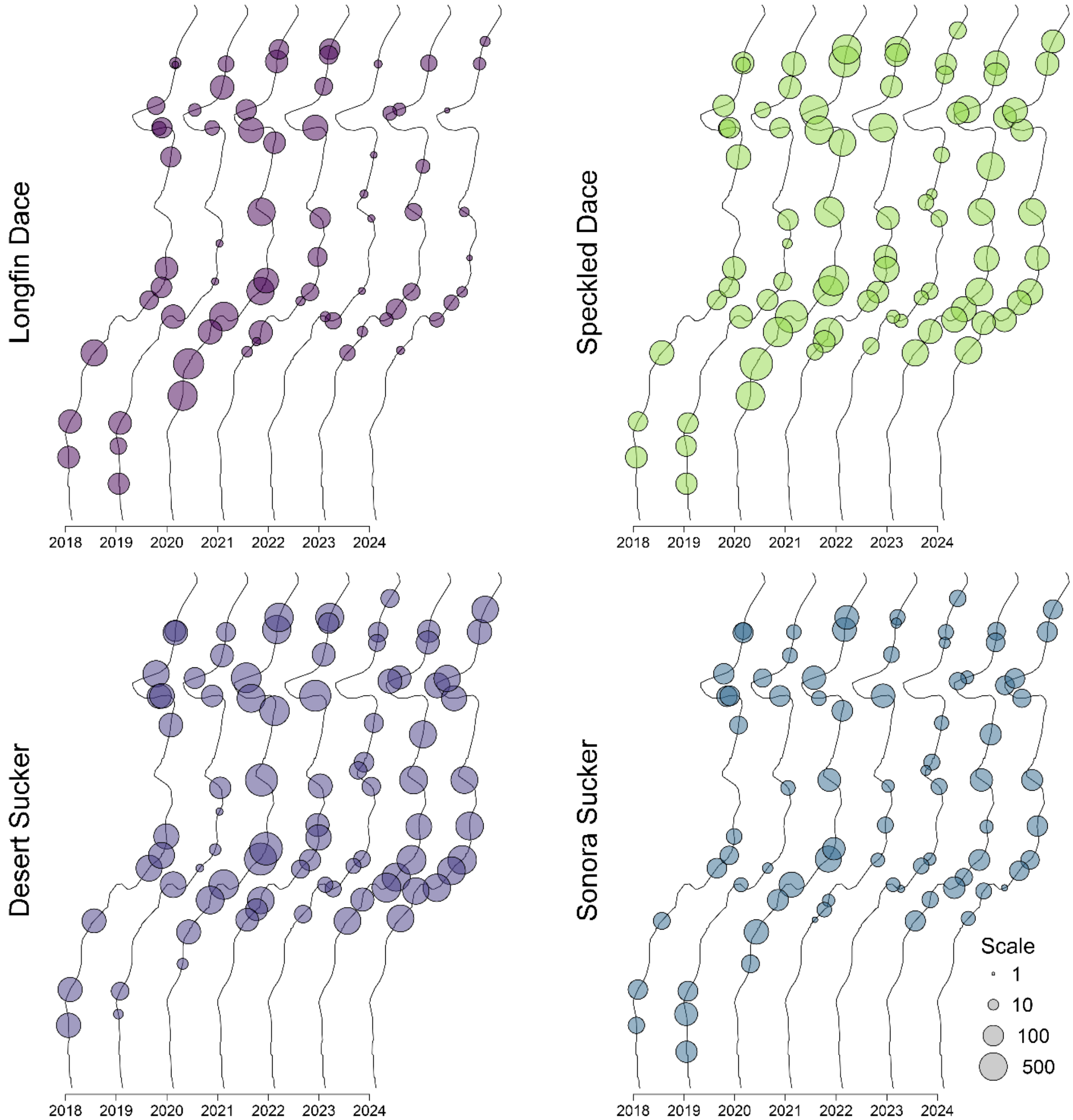

Figure 4.15.—Mean backpack electrofishing relative abundance (CPUE, fish/h) of Longfin Dace (top left), Speckled Dace (top right), Desert Sucker (bottom left), and Sonora Sucker (bottom right) at each monitoring sub-reach in the middle Blue River from 2018 to 2024. Size of points indicates the relative abundance during monitoring at a particular location.

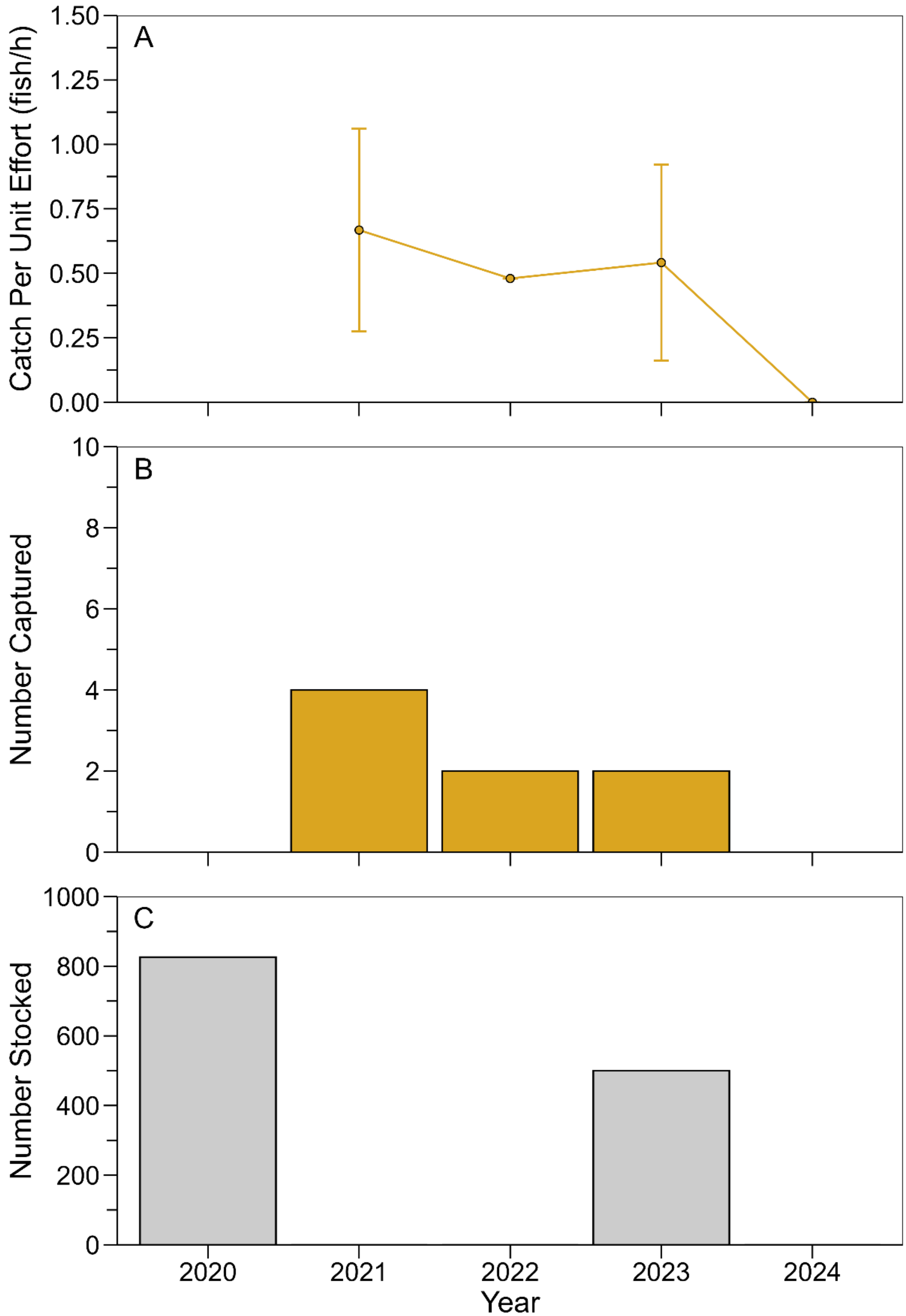

Figure 4.16.—Summary of Spikedace captured and stocked in the upper Blue River, annually from 2020 to 2024 with (A) mean annual backpack electrofishing catch per unit effort (fish/h; first pass only) with standard error bars, (B) total number of fish captured (first pass), and (C) total number of fish stocked.

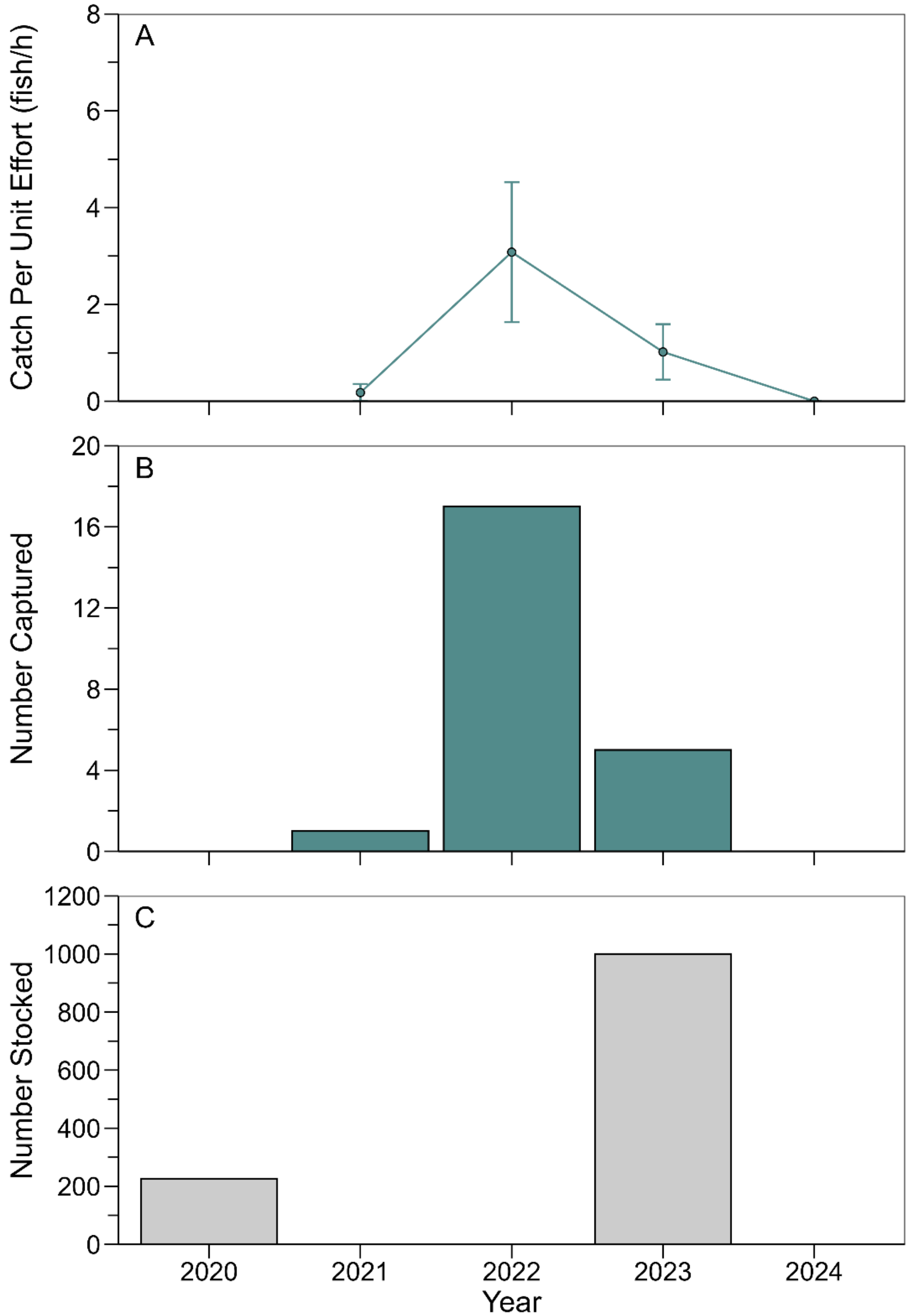

Figure 4.17.—Summary of Roundtail Chub captured and stocked in the upper Blue River, annually from 2020 to 2024 with (A) mean annual backpack electrofishing catch per unit effort (fish/h; first pass only) with standard error bars, (B) total number of fish captured (first pass), and (C) total number of fish stocked.

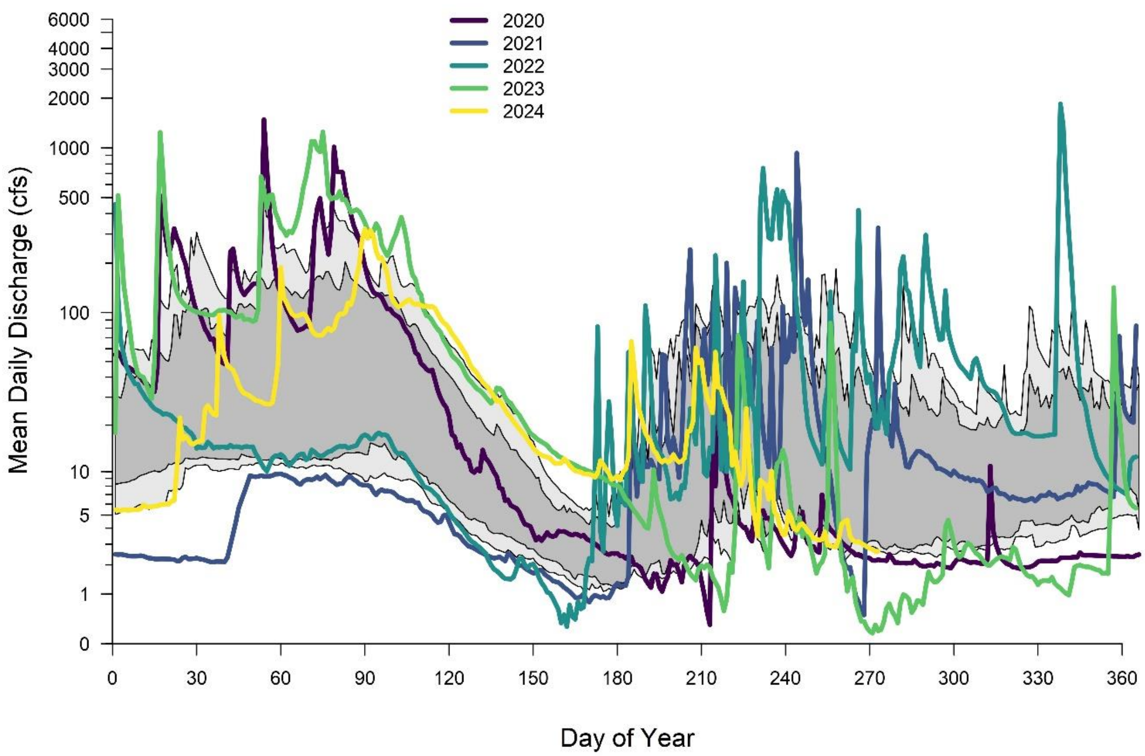

Figure 4.18.—Mean daily discharge (cfs) of the Blue River at the USGS gage at Juan Miller Crossing (09444200) from 2020 to 2024. Dark grey polygon indicates the 20th to 80th percentile of discharge, while the light grey polygon represents 10th to 90th percentile of discharge values from 1999-2024 (period that complete records exist).

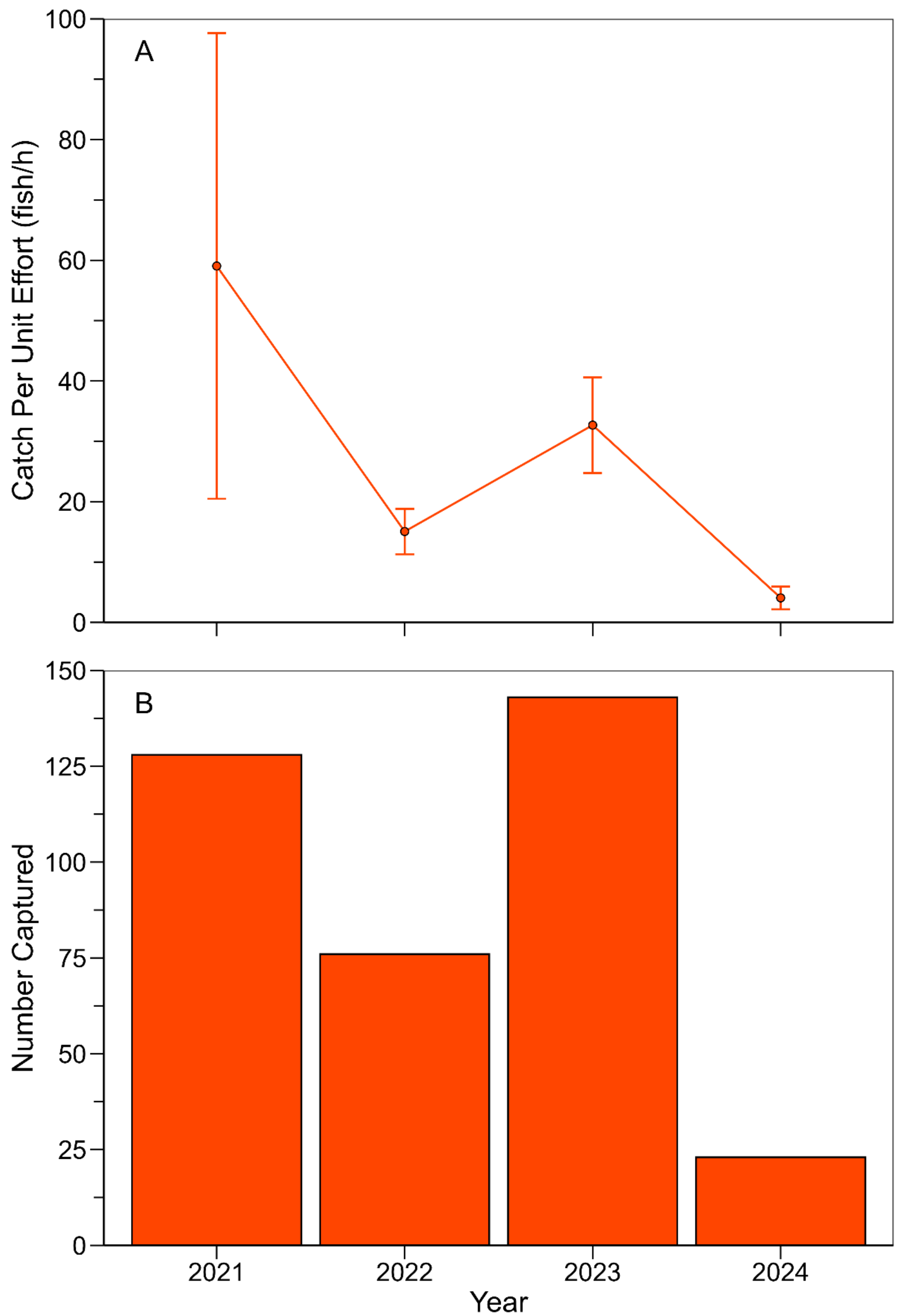

Figure 4.19.—Summary of Loach Minnow captured in the upper Blue River, annually from 2021 to 2024 with (A) mean annual backpack electrofishing catch per unit effort (fish/h; first pass only) with standard error bars, and (B) total number of fish captured (first pass only).

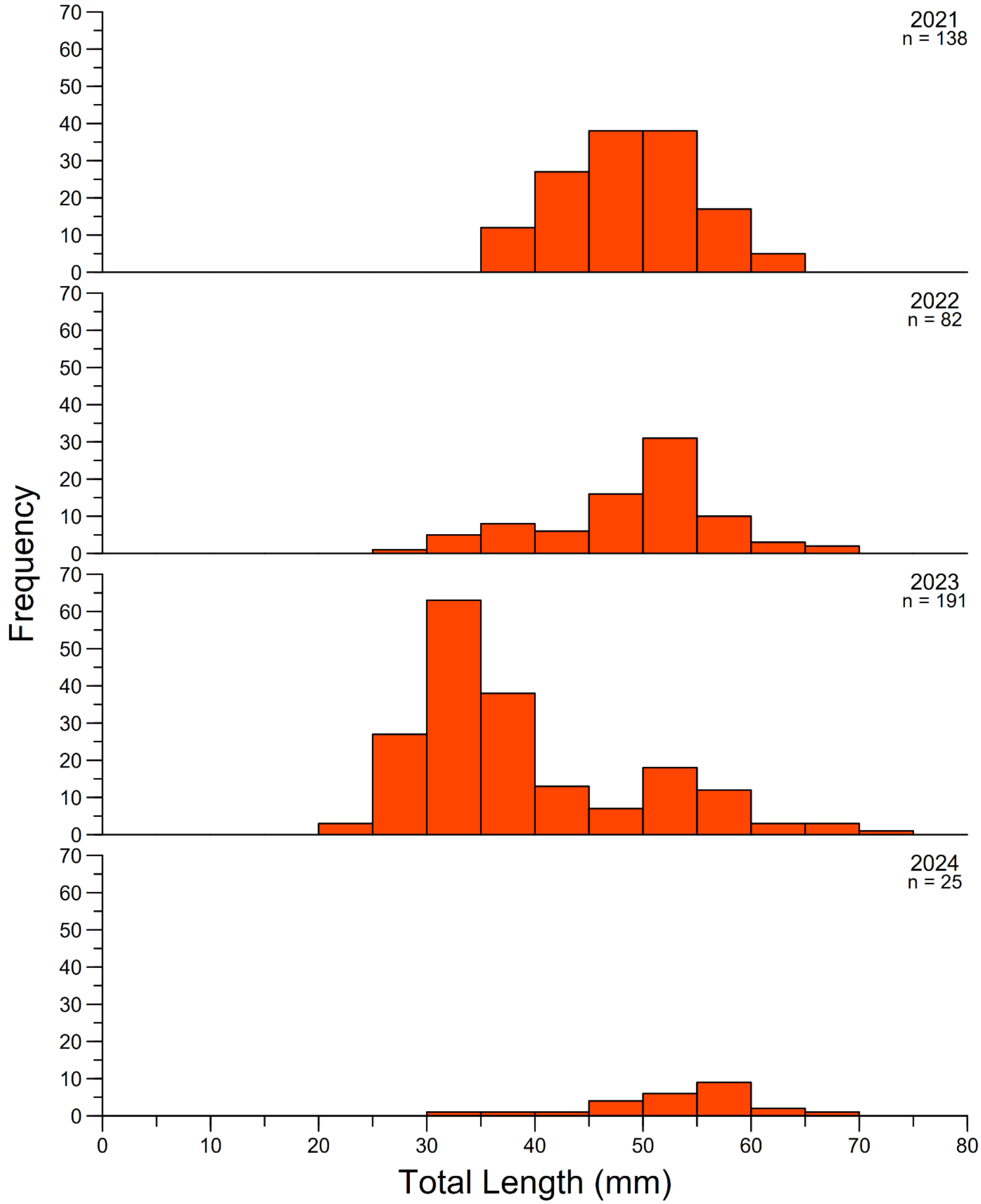

Figure 4.20.—Length frequency distribution of Loach Minnow captured during annual monitoring in the upper Blue River, from 2021 to 2024. Number of fish captured and measured each year is shown in the top right corner of each panel.

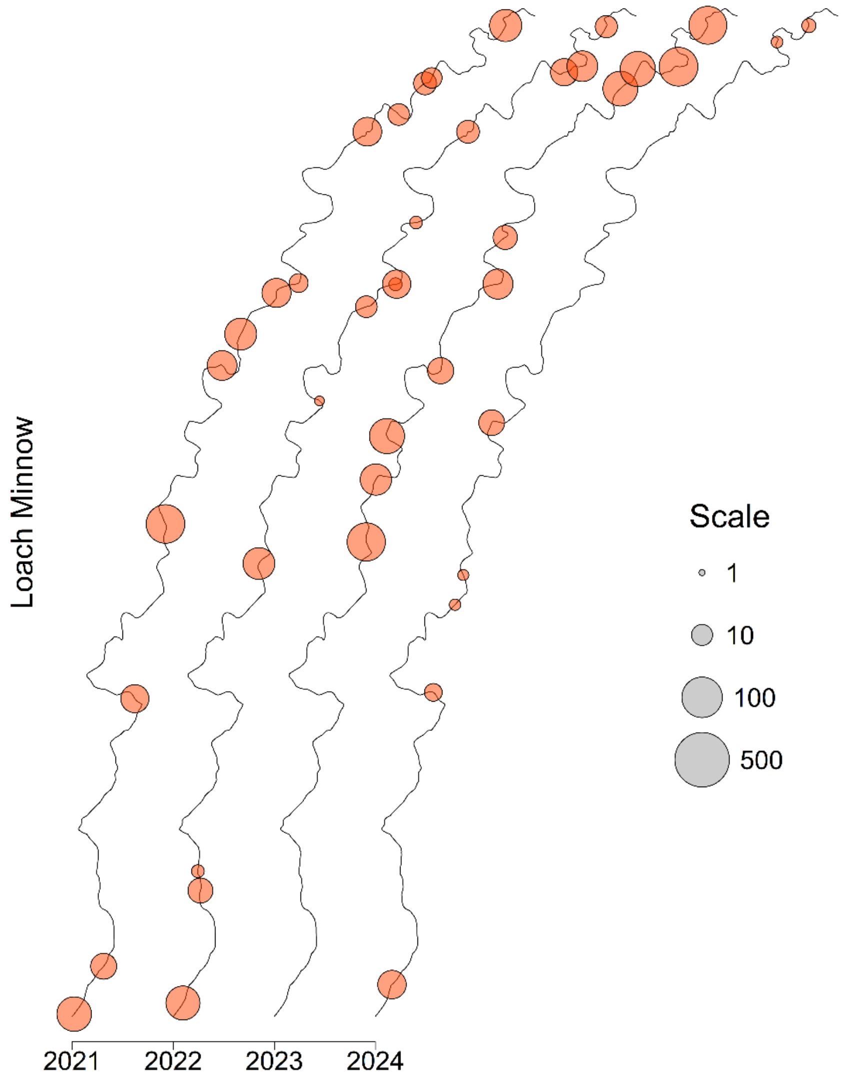

Figure 4.21.—Loach Minnow mean backpack electrofishing relative abundance (CPUE, fish/h) at each monitoring site in the upper Blue River from 2021 to 2024. Size of points indicates the relative abundance during monitoring at a particular location.

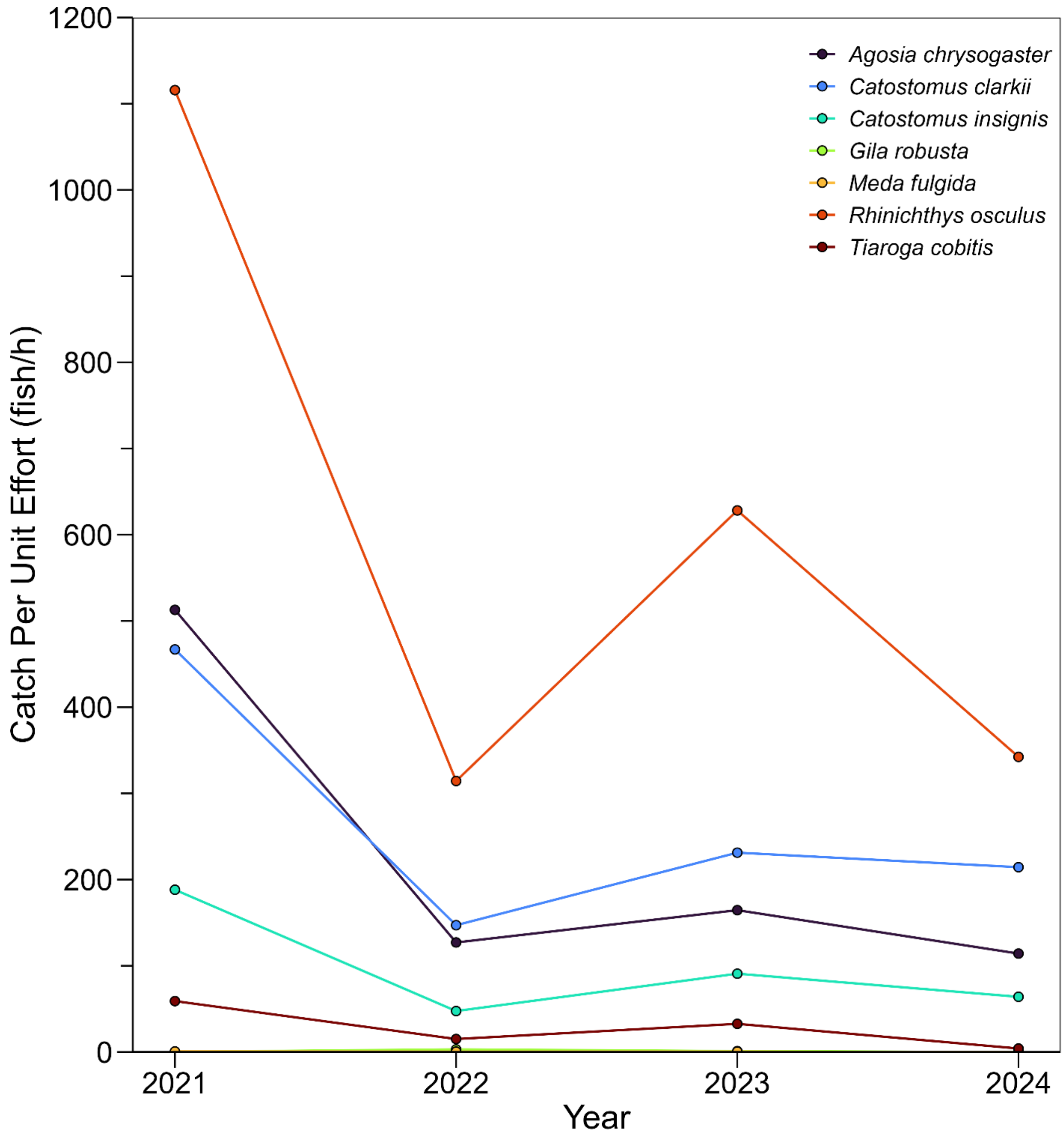

Figure 4.22.—Mean annual backpack electrofishing catch per unit effort (fish/h) of all native fish species captured in the upper Blue River, AZ from 2021 to 2024. Standard error bars are not shown to improve clarity of mean catch per unit effort trends.

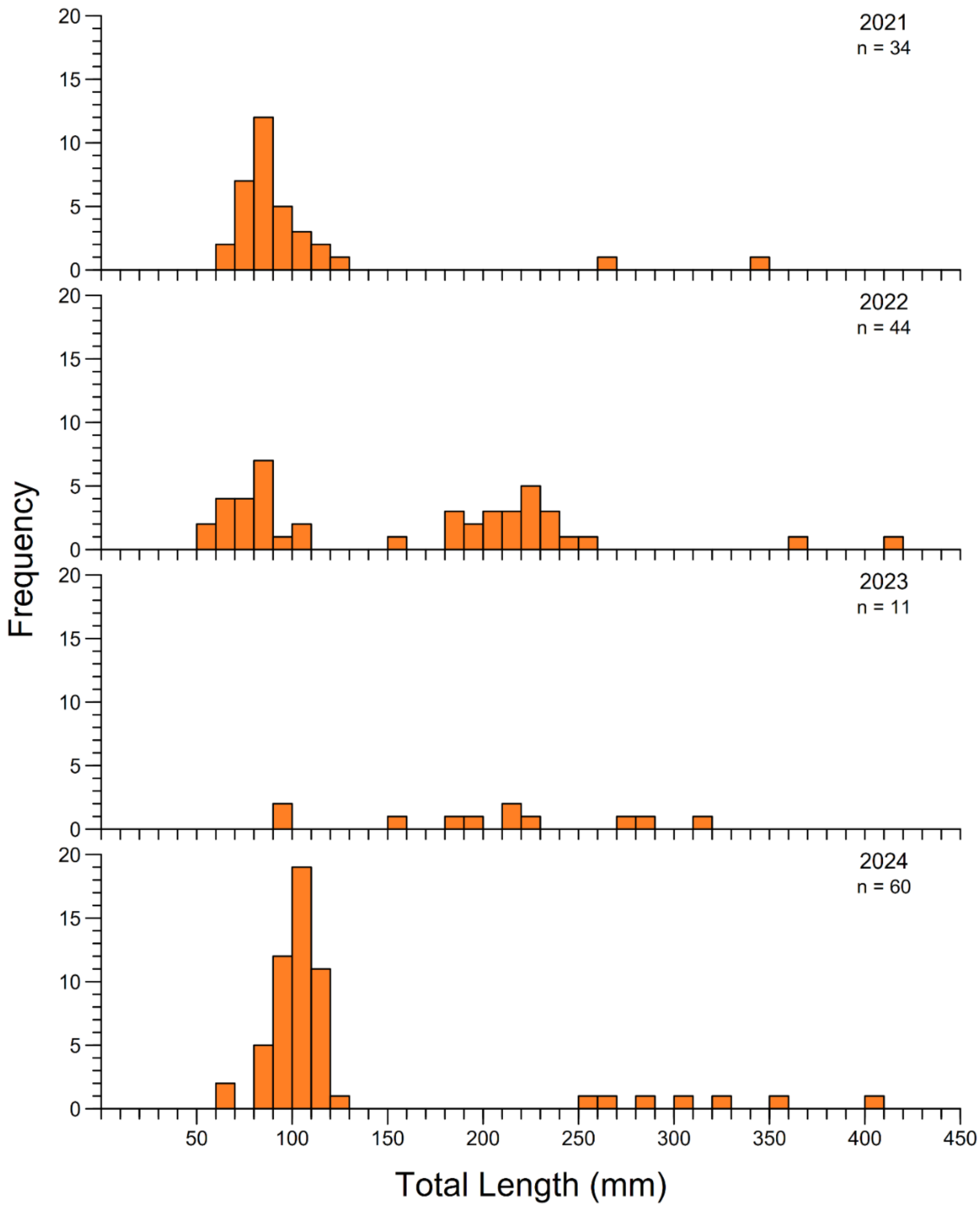

Figure 4.23.—Length frequency distribution of Brown Trout captured during annual monitoring in the upper Blue River, from 2021 to 2024. Number of fish captured and measured each year is shown in the top right corner of each panel.

5. Harden Cienega Creek Native Fish Restoration (Task AZ-2014-1)

Strategic Plan Goals:

- Preventing Extinction and Managing Toward Recovery
  - Goal 3. Remove nonnative aquatic species threats.
  - Goal 4. Replicate populations and their associated native fish community into protected streams and other surface waters.
  - Goal 7. Monitor to quantitatively measure and evaluate project success in improving the status of target species and their habitats.

Recovery Objectives:

- Gila Topminnow 1999 draft revised recovery plan objective 2.2. Reestablish Gila Topminnow in suitable habitats following geographic guidelines.
- Gila Topminnow 1999 draft revised recovery plan objective 2.4 Protect habitats of reestablished or potential populations from detrimental nonnative aquatic species.
- Gila Topminnow 1999 draft revised recovery plan objective 3. Monitor natural and reestablished populations and their habitats.
- Gila Chub draft recovery plan objective 1.3.1. Eliminate or control problematic nonnative aquatic organisms
- Gila Chub draft recovery plan objective 2. Ensure representation, resiliency, and redundancy by expanding the size and number of populations within Gila Chub historical range via replication of remnant populations within each RU.

Background: Harden Cienega Creek is a tributary to the San Francisco River near the New Mexico state line. Roundtail Chub[10] occupy the stream, although their distribution was historically limited to approximately 2 km of stream below a natural waterfall barrier. In April 2013, Department staff surveyed above the waterfall and determined that about 1.4 km of perennial water existed upstream that was suitable for Roundtail Chub. Chub were initially translocated from lower Harden Cienega Creek to the previously unoccupied reach upstream of the waterfall in 2015, with genetic augmentations occurring in 2018 and 2019. Monitoring from 2017 to 2020 detected several hundred chub representing all size classes upstream of the waterfall barrier. Gila Topminnow (n = 631; Bylas Spring lineage) were first stocked in lower Harden Cienega Creek downstream of the waterfall barrier in 2019 and augmented in 2021. Green Sunfish were detected above the barrier during post-stocking monitoring, with one removed in 2017 and two in 2018. Four Green Sunfish were captured and removed downstream of the barrier in 2019, suggesting the population was more abundant and broadly distributed within Harden Cienega Creek. A removal plan was drafted (Hickerson et al. 2020a) and Green Sunfish removal efforts were initiated in 2020. Because Green Sunfish were captured well upstream of the barrier on multiple occasions, it was concluded that an upstream source of Green Sunfish exists in the Harden Cienega Creek drainage. Surveys of all

[10] Chub in Harden Cienega Creek were previously classified as Gila Chub.

43 stock tanks in the Arizona portion of the Harden Cienega Creek watershed failed to detect any fish. However, Green Sunfish were detected in at least three tanks in New Mexico during surveys in 2021.

Results:

Gila Topminnow Monitoring

On June 4, 2024, Department staff set ten minnow traps in the vicinity of the Gila Topminnow stocking location at the mouth of the canyon bound reach and captured 28 Speckled Dace, 2 Sonora Sucker, and 11 Roundtail Chub. The crew failed to capture or observe any Gila Topminnow in about 6.5 hours of soak time. At least three nets were disturbed (including two that were totally destroyed) by a bear that discovered the traps. This is the fifth consecutive year of monitoring in which no Gila Topminnow have been captured, so it is likely that the population failed to establish.

Green Sunfish Removal

During June 4-5, 2024, Department staff began the first pass of electrofishing in Harden Cienega Creek to remove Green Sunfish. The crew shocked for a total of 6,906 seconds up to the pool where the majority of Green Sunfish have been captured in the past. Unfortunately, the remainder of the pass had to be aborted due to failure of the electrofisher. The crew walked the majority of the way up to the confluence with Prospect Canyon to visually search for any Green Sunfish. No Green Sunfish were observed downstream or upstream of the falls. No Green Sunfish were captured in seven mini-hoop nets (five in the sunfish pool, two in the pool where most sunfish were captured in 2019) during a daytime soak of approximately four hours. The nets were reset overnight and still did not capture any Green Sunfish. A total of 51 Roundtail Chub, 11 Desert Sucker, and 6 Sonora Sucker were captured between the two sets, with one Roundtail Chub mortality that was eaten by a crayfish. This is the first year since 2017 that no Green Sunfish were encountered in the perennial reach both downstream and upstream of the barrier (Figure 5.1).

On June 12, 2024, Department staff hiked approximately 11.5 kilometers of Harden Cienega Creek to search for perennial pools potentially occupied by Green Sunfish. There was little surface water present, with a total of 19 pools or short flowing reaches of surface water within the sampled reach (Figure 5.2). The crew set a total of nine mini-hoop nets in pools that were too deep to confidently assess the presence of fish or where fish were observed. A total of 66 Green Sunfish were captured in two pools, roughly halfway through the surveyed reach. The crew also carried out two dip net sweeps and captured 12 newly hatched Green Sunfish in the pool that supported most of the Green Sunfish. The size class distribution of all Green Sunfish captured suggest that potentially up to three age-classes may be present in Harden Cienega Creek (Figure 5.3). One juvenile Roundtail Chub (<50 mm TL) was captured in a pool approximately 2 km upstream from the perennial reach and approximately 50 more juvenile chub were observed but were not captured. This detection represents a natural range expansion of the Roundtail Chub population in Harden Cienega Creek from the perennial reach to the intermittent reach.

Recommendations: Topminnow have not been captured for a third consecutive year following the last augmentation in 2021. As a result, the Department does not recommend future stocking of Gila Topminnow at Harden Cienega Creek. There are likely biotic and abiotic factors limiting success at this location. Direct predation by Roundtail Chub has been documented following the 2021 stocking event. Additionally, the quality of pool habitat in the vicinity of the stocking location declined after flooding in 2023.

Removal of Green Sunfish from the intermittent reach is recommended given this is likely to be the immediate source for downstream dispersal of Green Sunfish to the perennial reach. The observation of multiple size classes of Green Sunfish, including recently spawned individuals, suggests that populations are established and have been persisting for multiple years. As such, invasion from these pools to the perennial reach will continue to remain a threat to downstream populations of Roundtail Chub unless addressed. The pools occupied by Green Sunfish were relatively small and removal by mechanical methods in a short period of time would be feasible unless sunfish disperse to additional pools in the interim.

Tables and Figures:

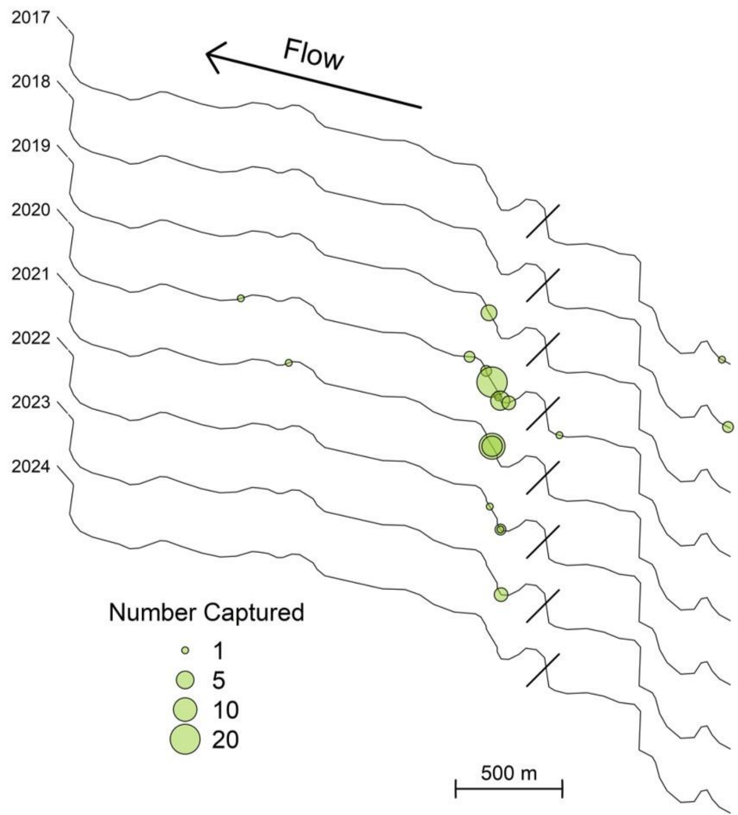

Figure 5.1.—Locations of Green Sunfish captured in the perennial reach of Harden Cienega Creek from 2017 to 2024. The barrier location is indicated by a diagonal line. Size of points indicates number of fish captured at a particular location during nonnative removal or monitoring efforts. Monitoring during 2017 to 2019 was only upstream of the barrier.

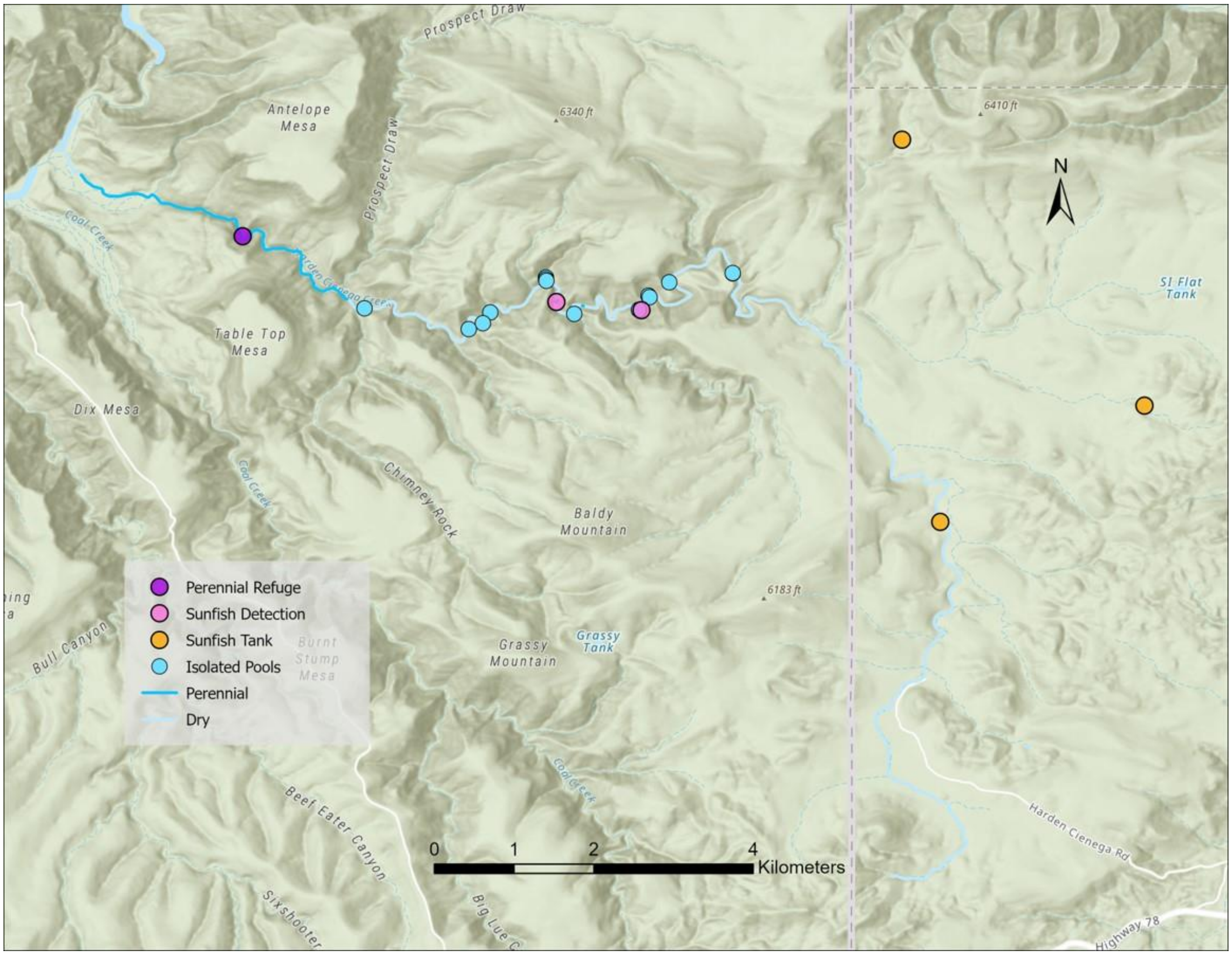

Figure 5.2.—Map depicting the location of pools in the intermittent reach of Harden Cienega Creek. The survey route started at the New Mexico state line and continued to the beginning of the perennial reach (near Chimney Rock Canyon). Pools where Green Sunfish were observed are shown in pink while fishless pools are shown in blue. Stock tanks in New Mexico where Green Sunfish were detected in 2021 are also displayed (Ditch, Distill and California Tanks).

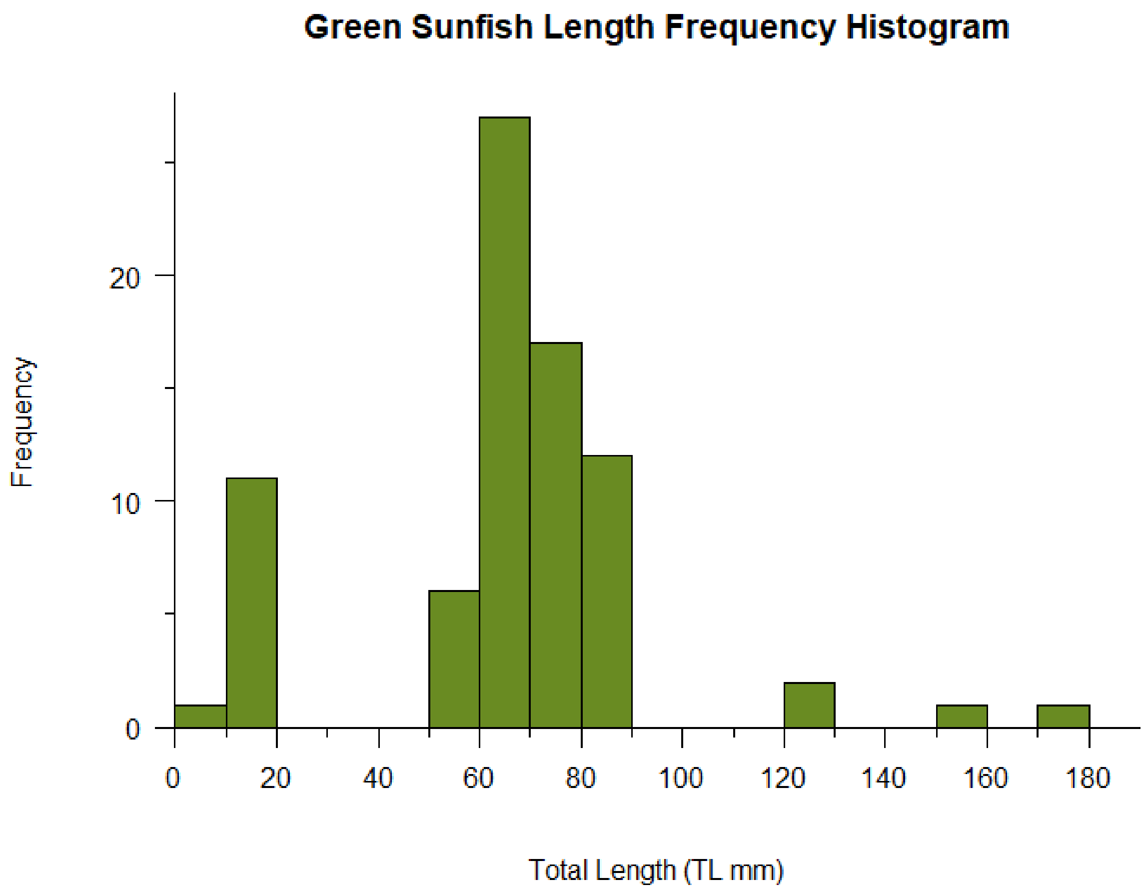

Figure 5.3.—Length frequency histogram of all Green Sunfish (n = 66) captured in isolated pools in upper Harden Cienega Creek on June 12, 2024.

6. George Wise Spring Nonnative Fish Removals (Task AZ-2024-1)

Strategic Plan Goals:

- Preventing Extinction and Managing Toward Recovery
  - Objective 3a. Eradicate or suppress nonnative aquatic species from a minimum of five surface waters to prepare them for repatriations of native fishes
  - Objective 4a. Replicate Gila Topminnow stocks into a minimum of 10 surface waters.

Recovery Objectives:

- Gila Topminnow 1999 draft revised recovery plan objective 2.2. Reestablish Gila Topminnow in suitable habitats following geographic guidelines.
- Gila Topminnow 1999 draft revised recovery plan objective 2.4 Protect habitats of reestablished or potential populations from detrimental nonnative aquatic species.
- Gila Topminnow 1999 draft revised recovery plan objective 3. Monitor natural and reestablished populations and their habitats.

Background: Ash Canyon is a largely ephemeral tributary to Sonoita Creek (now impounded by Patagonia Lake). Perennial waters in the Ash Canyon watershed includes a few stock tanks, some bedrock pools, and George Wise Spring, which provides perennial flow to about 300 meters of stream in Ash Canyon. The perennial reach supported by George Wise Spring is located on Arizona Game and Fish Commission owned lands. The Department has implemented habitat improvements at George Wise Spring using Desert Fish Habitat Partnership funds, including the installation of pipe-rail fencing to keep trespass cattle out of the entire riparian area. The property was purchased by the Department for native species conservation generally, and Gila Topminnow specifically, due to its proximity to important topminnow populations in Coal Mine and Fresno Canyons. Gila Topminnow of unknown origin were detected in Ash Canyon approximately 1.7 km upstream from George Wise Spring in 2006, but were never detected since. The site was assessed for the potential to reintroduce Gila Topminnow in March 2020, but Green Sunfish were found throughout the perennial reach and in the same pool where topminnow were detected in 2006. Green Sunfish were not observed in Ash Canyon upstream of the topminnow location to Mata Siete Spring, which was also fishless.

The Ash Canyon watershed upstream of George Wise Spring contains six tanks that could serve as a potential source of Green Sunfish to perennial waters downstream (Figure 6.1). Two tanks are located on Coronado National Forest Lands (Henry Tank, Ash Canyon #1). Ash Canyon #1 was completely dry when visited during stock tank surveys conducted by Ehret and Dickens (2009) and no fish were observed in a visual encounter survey conducted in 2020. Two tanks are located on private lands (Mine Tank, Private Unnamed Tank) and the final two tanks are located on Department owned lands (Cieneguita Canyon Tank, Department Unnamed Tank). The goal of this project is to identify whether any sources of Green Sunfish exist in the Ash Canyon watershed upstream of the Montezuma Road crossing, and evaluate the feasibility of mechanically removing the existing Green Sunfish population in Ash Canyon, including George Wise Spring.

Results: During May 20-22, 2024, Department staff removed Green Sunfish from the reach of Ash Canyon inside the cattle exclosure at George Wise Spring. Surface water within the exclosure was interrupted within a 250 m reach and mostly restricted to two long narrow pools that contained fish, along with a few small (< 5 m long), shallow off-channel pools that had water. Only one of the small off-channel pools contained fish. A total of 11 passes of backpack electrofishing were carried out throughout all surface water and a total of 99 Green Sunfish were captured and removed. The initial Green Sunfish population size of the George Wise Reach was estimated at 103 individuals (95% CI = 97.1 – 108.9) with an estimated captured probability of 0.25 (95% CI = 0.19 – 0.31; Table 6.1). Heavy duckweed cover made observing and capturing fish difficult during the first several passes. The crew spent some time after the first few passes removing as much duckweed as possible with dip nets to improve visibility. The depletion estimates of abundance suggest that nearly all, or possibly all, of the fish present in the George Wise Spring reach were captured in just a few days of backpack electrofishing effort.

During June 17-19, 2024, Department staff surveyed the Ash Canyon watershed upstream of George Wise Spring. Three of the six tanks were surveyed using a bag seine, which included Cieneguita Tank, Ash Canyon #1, and Department Unnamed Tanks (Figure 6.1). Two hauls with a bag seine were made in each tank respectively and no fish were captured. The remaining three tanks were not sampled as Henry and Private Unnamed Tanks were dry during visits in both May and June. Mine Tank was not sampled as the landowner confirmed that no fish had ever been observed and that the tank occasionally dries up.

The crew also surveyed the main channel within Ash Canyon below Cieneguita Tank and within side drainages below Department Unnamed Tank (Figure 6.1). The majority of the route surveyed was dry and only two pools were found. The first was near the Montezuma Well/Coal Mine Canyon road (hereafter Road Crossing Pool) and the second pool was located between the Road Crossing Pool and George Wise Spring (hereafter Canyon Pool, Figure 6.1). Road Crossing Pool was sampled with a bag seine given its relatively small size at the time and nearly the entirety of the pool could be sampled in a single haul. Ten hauls were completed with a total of 937 Green Sunfish captured and removed. The initial population size of the Road Crossing Pool was estimated at 966 individuals (95% CI = 952.02 – 979.98) with an estimated captured probability of 0.29 (95% CI = 0.27 – 0.31; Table 6.1). The removal estimates suggest that nearly all of the Green Sunfish were removed from the pool in just a few hours of effort, so mechanical removal from Road Crossing Pool is likely feasible within a relatively short period of time. Canyon Pool was sampled with two mini hoop nets that soaked for just over two hours. A total of 42 Green Sunfish were captured and subsequently removed. A dense growth of filamentous algae in the pool prevented easy observation of sunfish, and precluded other sampling methods like backpack electrofishing or seining. The maximum depth of the pool appeared to be no more than 0.5 m.

The mean size of Green Sunfish significantly increased downstream in each of the three locations where sunfish were captured (Figure 6.2). Mean total length of Green Sunfish was only 39.26 mm in the Road Crossing Pool, which was significantly less than the mean length in the Canyon Pool (82.14 mm TL) just downstream ($W = 174.5$, $P < 0.001$). Similarly, the mean length in the George Wise Spring reach (114.58), was significantly greater than the Canyon Pool just upstream ($W = 3592$, $P < 0.001$). The Road Crossing Pool was dominated by recently spawned fish from (10-28 mm TL), while relatively few (n = 6) individuals greater than 100 mm TL were captured. The Canyon Pool lacked the recently hatched young of year size class that dominated the Road Crossing Pool, but also lacked the larger adults (>150 mm TL) that were present in the George Wise Spring reach. Similarly, the George Wise Spring reach also lacked young of year fish despite the presence of many adults.

On May 21, 2024, Department staff visited the adjacent Fresno Canyon to verify recent observations of Green Sunfish made by Reclamation's monitoring contractor and to better understand the distribution, relative abundance, and size structure of the Green Sunfish population. A single pass was made with a backpack electrofisher through the perennial reach. Total effort was 3,193 seconds shocked and 35 Green Sunfish were captured (CPUE = 39.46 fish/h). Green Sunfish were concentrated in two pools (507746/3485732 and 507733/ 3485925). The majority of captured fish were young of year fish (n = 34) between 20-50 mm TL that were likely spawned in the spring. One adult was captured (215 mm TL) and an additional adult of similar of size was observed but evaded capture (Figure 6.3). Longfin Dace and Gila Topminnow were abundant during the survey and likely numbered in the several thousands, but no attempt was made to capture and count them during the removal pass. Crayfish and bullfrog tadpoles were also abundant throughout the surveyed reach.

Recommendations: Results from survey efforts in Ash Canyon indicate that the distribution of Green Sunfish is limited and populations are currently restricted to three small perennial waters on Department owned lands. Survey results also suggest that the source of Green Sunfish to the perennial waters of Ash Canyon was either an upstream population of Green Sunfish that has since become extirpated, or more likely, invasion from Patagonia Lake. Removal data demonstrates that Green Sunfish could feasibly be removed with mechanical methods from all three perennial portions of the Ash Canyon watershed with relatively little effort. A removal plan was drafted outlining a Green Sunfish removal strategy for the Ash Canyon watershed and provided to Reclamation as part of this project. The Department recommends that removal efforts should be pursued in 2025. After confirmation of eradication Gila Topminnow should be translocated to each of the perennial waters, as well as Cieneguita and Department Unnamed Tank. Cieneguita Tank may be able to serve as a suitable primary donor location for the Fresno/Coalmine Canyon topminnow lineage during implementation of the Gila Topminnow Genetic Management Plan in 2025 or 2026 (Mussmann 2024).

Results from Fresno Canyon point a very recent invasion that was limited to a few large individuals. However, these data clearly show Green Sunfish successfully spawned in spring of 2024, therefore addressing the issue prior to additional spawning is imperative. Currently, the Green Sunfish population could likely be eradicated with minimal mechanical removal effort, assuming additional dispersal from upstream has not occurred in the interim.

Tables and Figures:

Table 6.1.—K-pass depletion estimates of abundance for all Green Sunfish captured in the George Wise Spring (GWS) and Road Crossing Pool (RCP) reaches of Ash Canyon in 2024. Included is the number of fish caught in each pass (C1:C11), Carle-Strub abundance estimate (N) and capture probability (*p*). The 95% confidence intervals of are provided for estimated abundance and capture probability.

Location	C1	C2	C3	C4	C5	C6	C7	C8	C9	C10	C11	N	p
GWS	25	14	22	14	7	0	1	7	6	3	0	103 (97.1 – 108.9)	0.25 (0.19 – 0.31)
RCP	179	330	152	82	77	24	48	16	12	17	NA	966 (952.02 – 979.98)	0.29 (0.27 – 0.31)

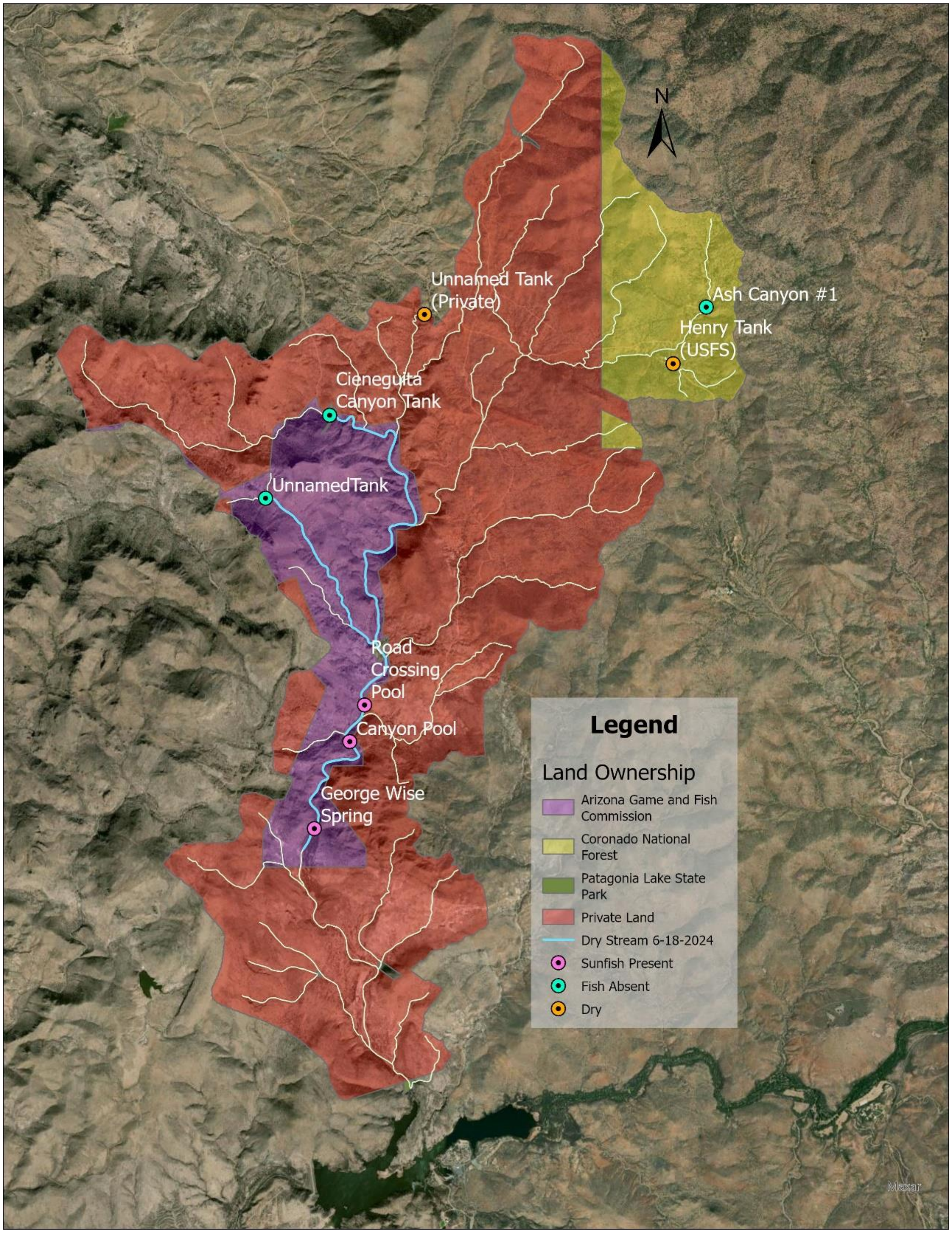

Figure 6.1.—Extent of the Ash Canyon Watershed by land ownership type. Included is the location of ephemeral tributaries (thin white lines), the stream channels that were wet/dry mapped during June 18-19, 2024 (blue lines), locations where Green Sunfish were captured (pink dots), stock tanks where water was present but fish were absent (teal dots), and stock tanks that were dry (orange dots) in 2024.

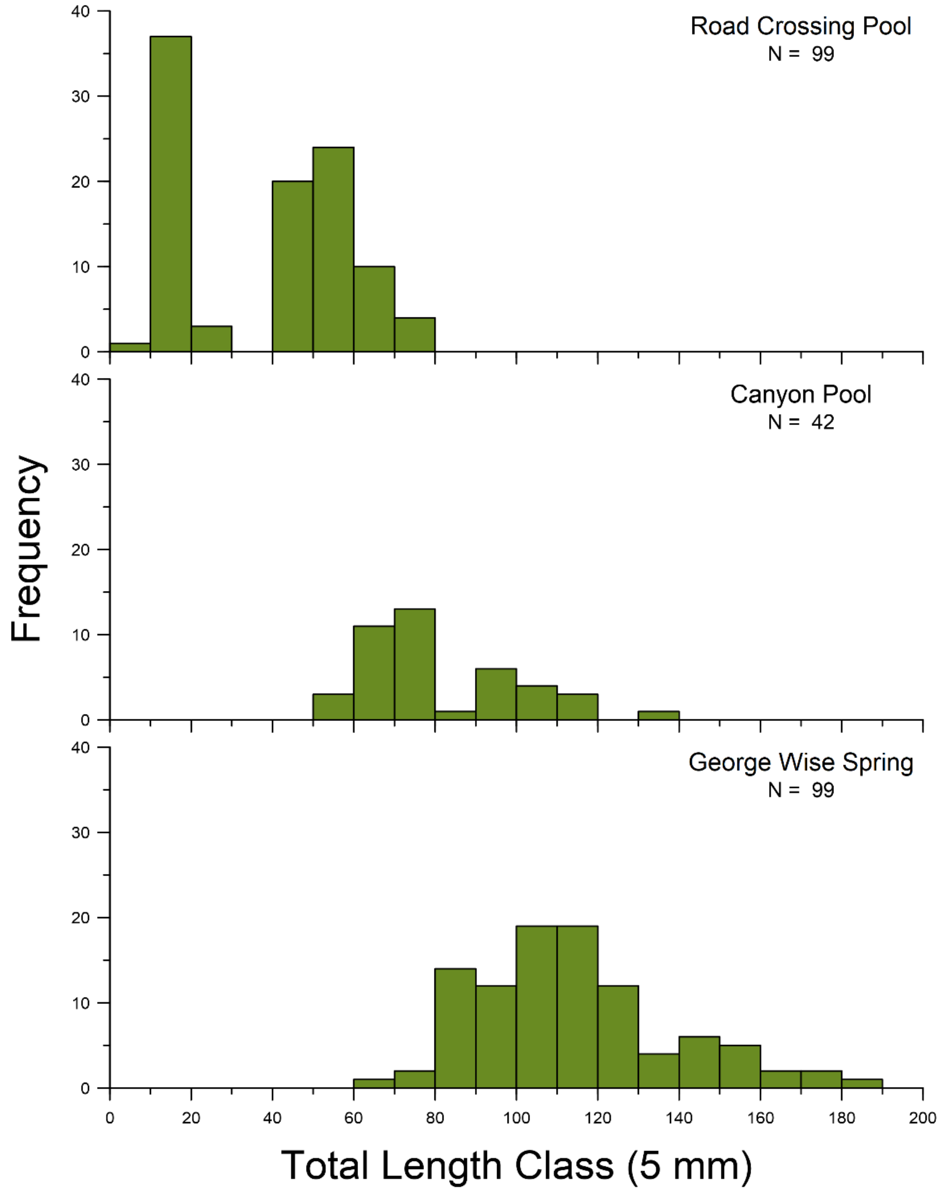

Figure 6.2.—Length frequency distribution of Green Sunfish captured during removal efforts in the Ash Canyon watershed from May 20-22 and on June 18, 2024. Total number of fish captured and measured among all passes at each location is shown in the top right corner of each panel. Note only the first 99 individuals were measured for total length in the Road Crossing Pool.

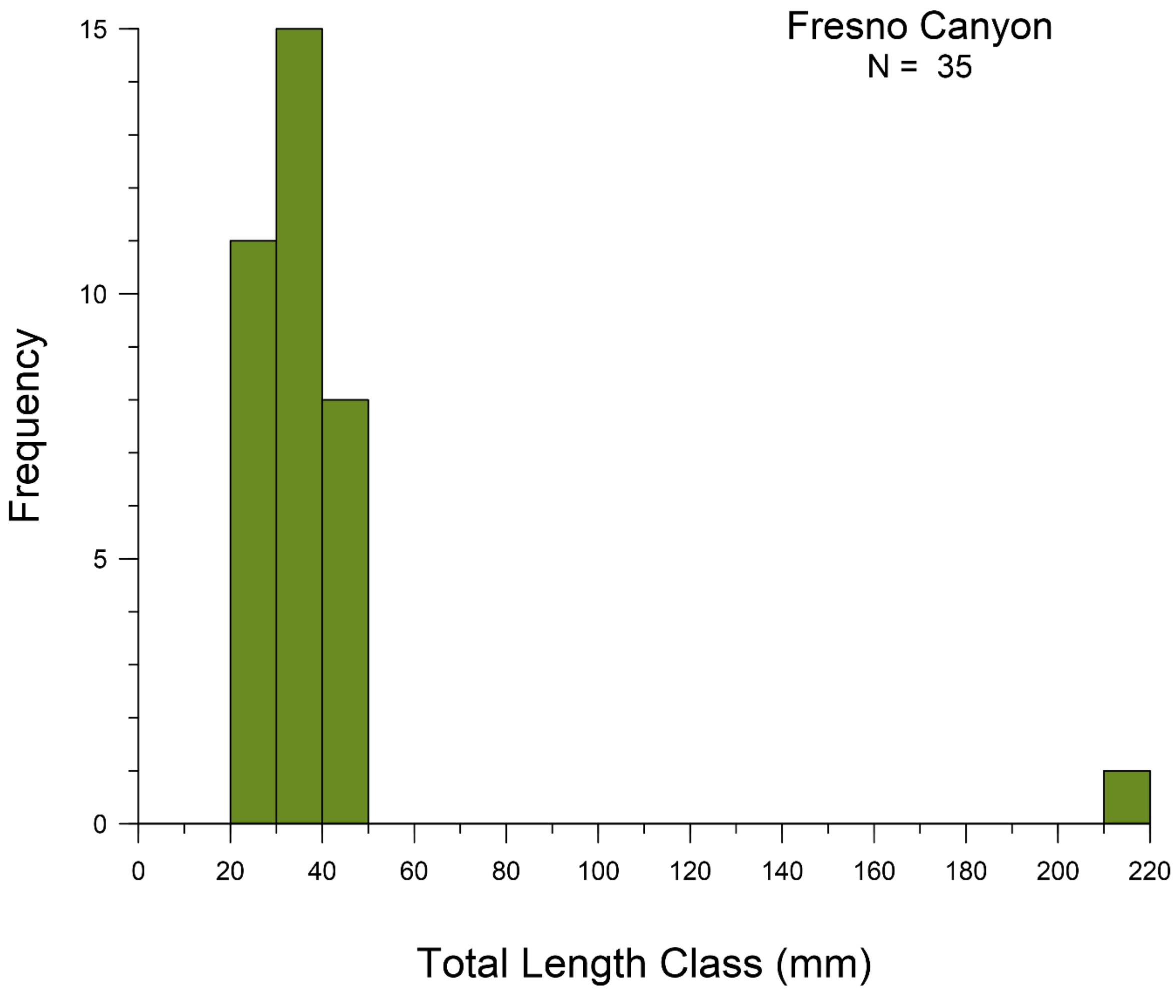

Figure 6.3.—Length frequency distribution of Green Sunfish captured during removal efforts in Fresno Canyon on May 21, 2024. Total number of fish removed is shown in the top right corner.

7. Sharp Spring native fish restoration (Task AZ-2016-3)

Strategic Plan Goals:

- Preventing Extinction and Managing Toward Recovery
  - Goal 3. Remove nonnative aquatic species threats.
  - Goal 4. Replicate populations and their associated native fish community into protected streams and other surface waters.
  - Goal 7. Monitor to quantitatively measure and evaluate project success in improving the status of target species and their habitats.

Recovery Objectives:

- Gila Chub draft recovery plan objective 1.3.1. Eliminate or control problematic nonnative aquatic organisms.
- Gila Chub draft recovery plan objective 2. Ensure representation, resiliency, and redundancy by expanding the size and number of populations within Gila Chub historical range via replication of remnant populations within each RU.
- Gila Chub draft recovery plan objective 7. Monitor remnant, repatriated, and refuge populations to inform adaptive management strategies.
- Gila Topminnow 1999 draft revised recovery plan objective 2.2. Reestablish Gila Topminnow in suitable habitats following geographic guidelines.
- Gila Topminnow 1999 draft revised recovery plan objective 2.4 Protect habitats of reestablished or potential populations from detrimental nonnative aquatic species.
- Gila Topminnow 1999 draft revised recovery plan objective 3. Monitor natural and reestablished populations and their habitats.

Background: Sharp Spring is a series of perennial cienega pools located on San Rafael State Natural Area, which is owned by Arizona State Parks and Trails (AZSP, Figure 7.1). The drainage is tributary to the Santa Cruz River, about 2 km from the international border with Mexico. Sharp Spring was historically occupied by a relict population of Gila Topminnow. Nonnative Western Mosquitofish were first detected in Sharp Spring in 1979. Monitoring by the Department staff and partners documented the gradual decline and eventual disappearance of Gila Topminnow, which was last detected in 2001. Extirpation of topminnow has primarily been attributed to predation by and competition with nonnative mosquitofish. In June 2013, Department staff attempted to eradicate Western Mosquitofish by draining the pools in Sharp Spring with gasoline powered pumps, but the effort was abandoned because the pools could not be completely dried. The purpose of this project is to eradicate Western Mosquitofish from Sharp Spring and reintroduce Sharp Spring lineage Gila Topminnow. Eradication of Western Mosquitofish will also create an opportunity to potentially replicate a population of upper Santa Cruz River Roundtail Chub[11] in Sharp Spring.

[11] Roundtail Chub in the upper Santa Cruz River previously classified as Gila Chub.

Department staff conducted a rotenone treatment in June 2022 and successfully eradicated Western Mosquitofish. Following confirmation of eradication, 659 Gila Topminnow from two donor locations were stocked into two pools of Sharp Springs in July 2022. Nine additional topminnow were stocked in October 2022. A total of 135 Gila Topminnow were captured in the two pools that were initially stocked during the first post-stocking monitoring effort in October 2022. In May of 2023, Department staff stocked 200 Gila Topminnow into three pools (SP02, SP04, SP10) and 286 fish into SP13, which had not been stocked since treatment. Monitoring in August 2023 revealed topminnow only persisted in pool SP02 where 117 fish were captured.

Results: On May 29, 2024, Department staff collected 1,000 Gila Topminnow from Cottonwood Tank at Robbins Butte in a single seine haul and a few dip net sweeps with four topminnow mortalities and one Desert Pupfish mortality during the collection effort. The crew transported topminnow in two aerated coolers to Sharp Springs later the same afternoon. The fish were stocked between three pools that have previously been stocked with topminnow and held Western Mosquitofish prior to the rotenone treatment in 2022 (SP10 = 318, SP13 = 253, SP02 = 429). There were seven mortalities during transport and stocking.

On May 30, 2024, HOBO dissolved oxygen (DO) and temperature loggers were installed in two pools, one that has supported topminnow during both monitoring efforts since the initial restocking effort in 2022 (SP02), and one where topminnow have not been recaptured despite multiple stocking attempts (SP10).

On August 28, 2024, Department staff revisited Sharp Spring. The crew set a total of 24 minnow traps across seven pools that soaked for a minimum of two hours. A single male Gila Topminnow was captured in SP13. Importantly, monitoring data continues to confirm eradication of Western Mosquitofish following rotenone treatment in 2022. The crew also downloaded DO and temperature data from the loggers in SP02 and SP10. Dissolved oxygen data from SP02 and SP10 revealed near anoxic conditions in both pools from May to August (Figure 7.2). Anoxic conditions were also confirmed by observations of asphyxiated macroinvertebrates and bullfrog tadpoles in traps when they were retrieved. Loggers were redeployed to monitor dissolved oxygen levels over the fall, winter and early spring.

The exact cause for the limited DO in Sharp Spring remains unclear. One potential explanation is that changes in the light regime resulting from a reduction in the riparian gallery has enabled duckweed to proliferate more extensively relative to historical conditions. Duckweed inhibits the diffusion of oxygen into the water. Given duckweed now covers nearly all surface water, it is possible that there are only small pockets of oxygenated water within pools. In addition to low DO, the water level in the pools has declined substantially relative to historic conditions in the 1980s and 1990s and is now 1-1.5 meters below historical staff gauges (Figure 7.3). Potential climactic change that might explain lower water levels was explored by using estimates of annual precipitation (PRISM, 4km resolution, Daly et al. 2008) averaged for the San Rafael Valley catchment for each year from 1981-2023. A generalized linear model of mean annual precipitation by year revealed that mean annual precipitation has significantly decreased in the San Rafael Valley by about 13.8 cm over the 42 year period ($t = -2.459$, $P = 0.018$; Figure 7.4). Reduced rainfall may be slowing the replenishment of groundwater, resulting in drying and lowering of the water table, which might contribute to lower oxygen conditions in Sharp Spring.

Recommendations: Gila Topminnow should continue to be stocked into Sharp Spring as a means to monitor whether environmental conditions improve consistently enough for widespread survival of topminnow. Continued data collection of DO over fall and winter will inform avenues of further investigation for potential solutions to address water chemistry issues. Consultation with a hydrologist may be warranted to better identify problematic areas and identify targeted solutions that could improve cienega function. Department staff will work with partners at Arizona State Parks to identify potential habitat improvement options that can be implemented. Monitoring of Sharp Spring should continue in order to verify the continued persistence of Gila Topminnow and document the outcome of any additional translocation events.

Tables and Figures:

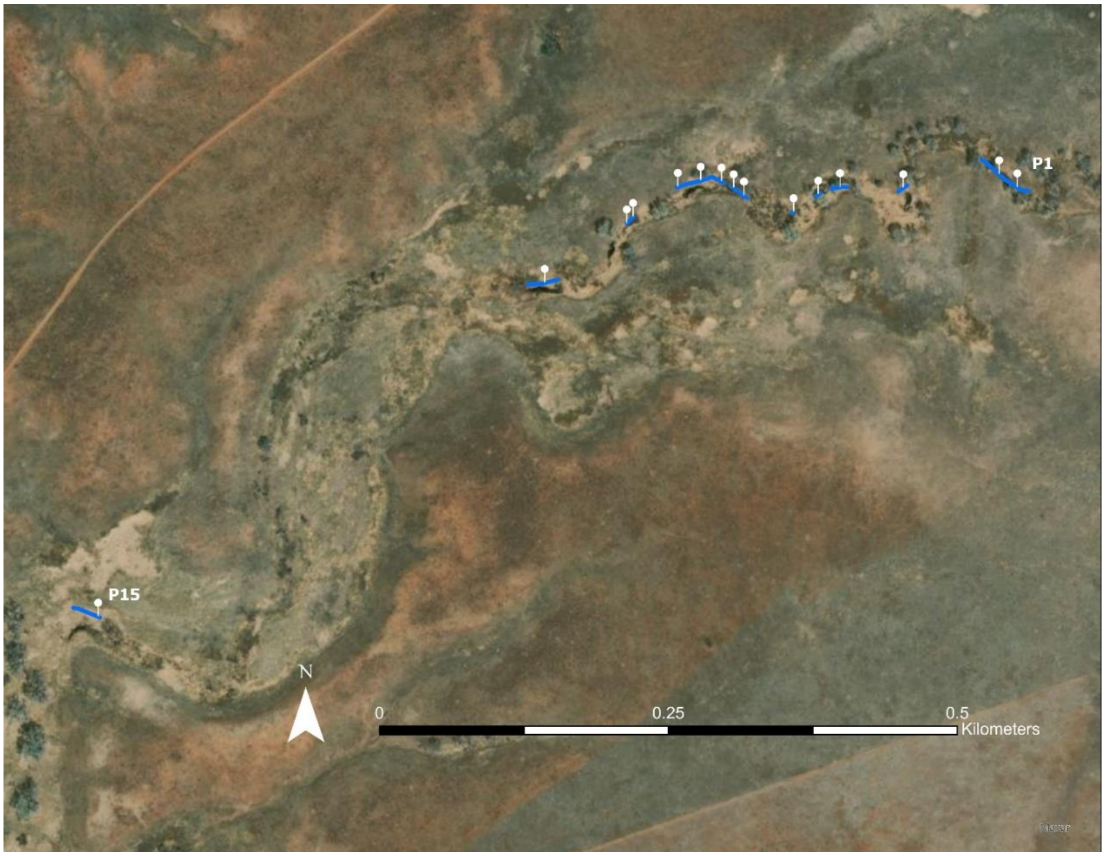

Figure 7.1.—Overview of the project area at Sharp Spring. Approximate location of each of the 15 pools (white dots) from upstream (P1) to downstream (P15) is shown.

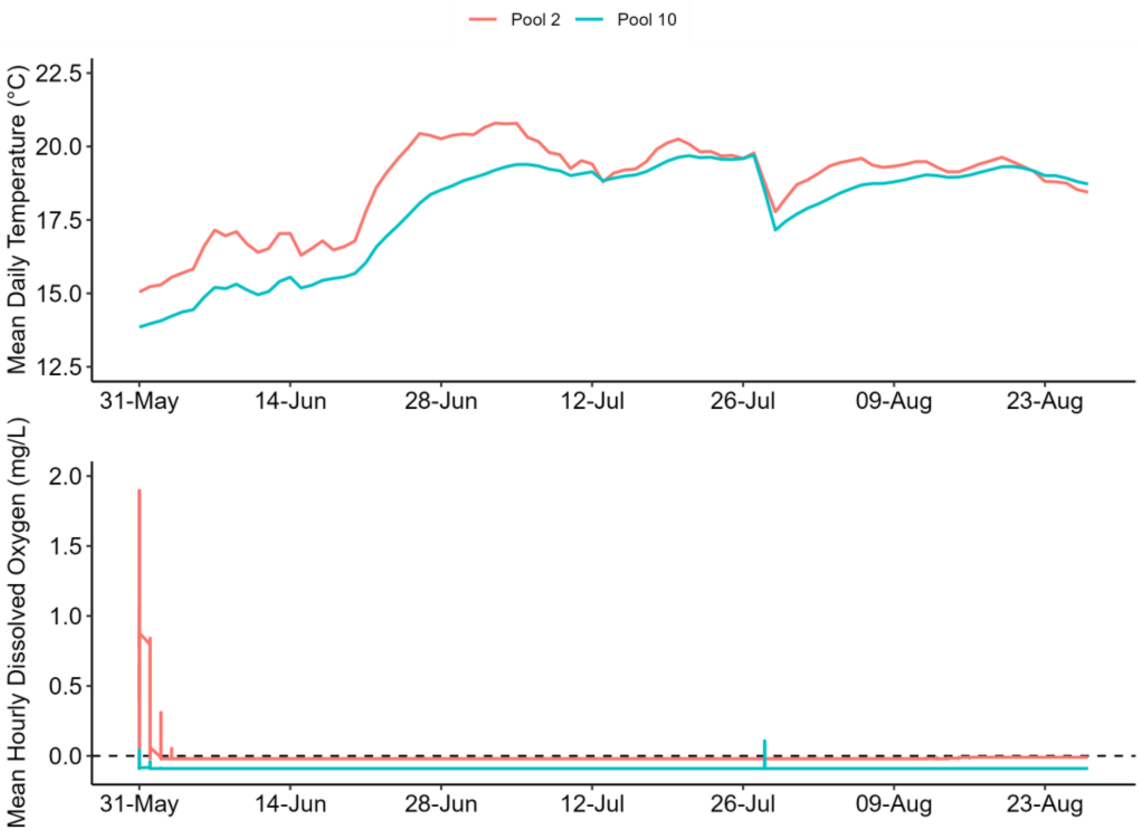

Figure 7.2.—Mean daily temperature (top) and Mean hourly dissolved oxygen (bottom) from pool 2 and 10 at Sharp Spring. Data loggers were deployed from May 30 to August 28, 2024.

Figure 7.3.—Photos of pool SP14 at Sharp Spring from 1992 (top), 2001 (middle) and 2023 (bottom) demonstrating water level and habitat change in Sharp Spring. Note the staff gauge (identified with orange circle) is completely out of the water in 2023.

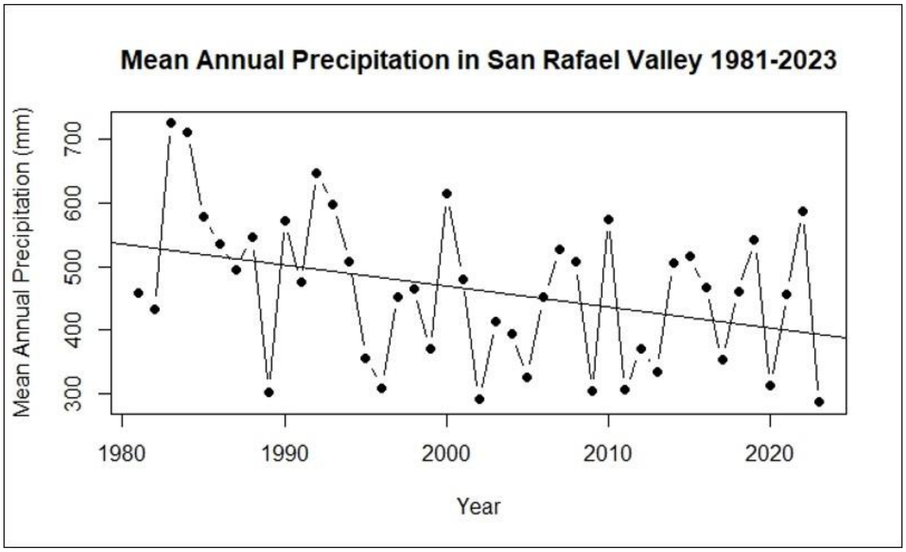

Figure 7.4.—Mean annual precipitation in the San Rafael Valley from 1981 to 2023. The regression line shows a decrease of 137.7 mm in mean annual precipitation over a 42-year period (intercept = 7027.61, slope = -3.28).

8. Upper Verde River native fish restoration (Task AZ-2020-1)

Strategic Plan Goals:

- Preventing Extinction and Managing Toward Recovery
  - Goal 3. Remove nonnative aquatic species threats.
  - Goal 4. Replicate populations and their associated native fish community into protected streams and other surface waters.
  - Goal 7. Monitor to quantitatively measure and evaluate project success in improving the status of target species and their habitats.

Recovery Objectives:

- Spikedace recovery objective 6.2.5. Reclaim as necessary to remove non-native fishes.
- Spikedace recovery objective 6.3. Reintroduce Spikedace to selected reaches.
- Spikedace recovery objective 6.4. Monitor success/failure of reintroductions.
- Loach Minnow recovery objective 6.2.5 Reclaim as necessary to remove non-native fishes.
- Loach Minnow recovery objective 6.3. Reintroduce Loach Minnow to selected reaches.
- Loach Minnow recovery objective 6.4. Monitor success/failure of reintroductions.
- Gila Topminnow 1999 draft revised recovery plan objective 2.2. Reestablish Gila Topminnow in suitable habitats following geographic guidelines.
- Gila Topminnow 1999 draft revised recovery plan objective 2.4 Protect habitats of reestablished or potential populations from detrimental nonnative aquatic species.
- Gila Topminnow 1999 draft revised recovery plan objective 3. Monitor natural and reestablished populations and their habitats.
- Razorback Sucker recovery objective 1.3 Reduce adverse biological impacts
- Razorback Sucker recovery objective 2.6 Augment or reintroduce XYTE in recovery areas
- Razorback Sucker recovery objective 2.6.2.3 Monitor reestablishment and augmentation efforts

Background: The upper Verde River Native Fish Restoration Project is a multi-agency effort focused on protecting and restoring the native fish assemblage within the upper Verde River drainage in central Arizona. The project is currently focused on assessing the distribution and assemblage of nonnative fish communities within the watershed to inform potential planning for nonnative fish control efforts should Reclamation construct fish barriers on the Verde River. In 2019, Department staff assessed the feasibility of surveying stock tanks in the upper Verde River drainage for presence of nonnative fishes. Tanks most likely to support nonnative fish were identified using an automated approach developed in Program R to classify tanks as wet or dry using normalized difference water index values, and a scoring system based on perennial status, previous nonnative fish records, and distance to the Verde River. A total of 146 tanks received a score of 2 or greater which corresponded to the greatest risk categories. A total of 87 tanks were surveyed, with 19 containing nonnative fishes. The remaining 59 tanks were either dry, or access for sampling was denied. Green Sunfish is the most common species detected in stock tanks to date, with Yellow Bullhead, Black Crappie, Largemouth Bass, Western Mosquitofish, Bluegill and Western Mosquitofish also detected.

With the detection of nonnative fish in a relatively high proportion of tanks sampled, and the observation of Green Sunfish actively dispersing through several ephemeral stream channels during the summer of 2021, surveys of ephemeral and intermittent stream channels that could be utilized as conduits for dispersal by nonnative fish (particularly downstream of nonnative fish occupied stock tanks) was identified as a critical information gap. Priority streams identified for sampling in 2023 were lower Hell Canyon (from the Verde River confluence to Hell Canyon Tank), which has at least 12 nonnative fish occupied stock tanks within the watershed, and Big Chino Wash and Williamson Valley Wash where permission to survey stock tanks within each watershed was largely denied. Nonnative fish appear to be able to readily move within tributaries to the upper Verde River, specifically Williamson Valley Wash, Big Chino Wash, and Hell Canyon. Survey results from Hell Canyon in 2023 confirm that ephemeral or intermittent stream channels downstream of stock tanks with known nonnative fish populations should be considered potentially occupied by nonnative fishes (Hickerson et al. 2023). Surveys in 2024 focused on additional Verde River tributaries that were identified as having a high probability of being occupied by nonnative fishes based on the number and proximity of occupied stock tanks in the watershed. These included MC Canyon and Grindstone Wash in the Hell Canyon watershed (Figure 8.1).

Results: During August 12-13, 2024, Department staff, assisted by U.S. Bureau of Reclamation staff, completed surveys in MC Canyon and Grindstone Wash. On August 12, 2024, the crew hiked 7.2 miles up MC Canyon from the Drake Road crossing. No surface water was encountered until the middle reach of MC Canyon where a total of 12 pools were located. Three mini hoop nets were set in each of the two largest pools that soaked for over two hours. The remaining pools were sampled using a combination of dip net sweeps and visual observation. All 12 pools in this section of MC canyon were fishless, however the crew observed well developed canyon treefrog tadpoles in the majority of pools suggesting that they held water for a prolonged period. The crew also hiked from the Drake Road crossing 6 miles downstream in MC Canyon to the confluence with Hell Canyon. No surface water was encountered during the hike. Deep scours were present along the margins of the canyon but there was no indication that scours had recently held water. The crew also noted a large natural barrier to upstream movement in the lower reach of MC Canyon. After reaching the confluence, the crew hiked up Grindstone Wash to the Drake Road crossing and noted no surface water. The substrate in both Grindstone Wash and lower MC Canyon was primarily boulder and large cobble with very little exposed bedrock, whereas the pools documented in the upper section of MC Canyon were almost entirely associated with exposed bedrock.

On August 21, 2024, Department staff surveyed the upper reach of MC Canyon and East Fork MC Canyon (Figure 8.1). Angling, dip nets and visual observation were used to collect presence absence data. The crew started at DT Tank and hiked down East Fork MC Canyon and sampled four pools, finding Green Sunfish in two. Two additional two pools were located but were inaccessible without climbing gear and the confluence with MC Canyon could not be reached following the stream channel because of a steep cliff (387918/3879967). The crew hiked up MC Canyon (starting at 387757/3879880) and found several large bedrock pools that were occupied by Green Sunfish. A total of 18 pools were marked in upper MC Canyon and 16 were found to be occupied by Green Sunfish (Figure 8.2). The crew visually observed the presence of young of year in multiple pools suggesting that Green Sunfish are likely established in the upper reach of MC Canyon. A total of 312 sunfish were either captured or visually observed during surveys of East Fork MC Canyon and upper MC Canyon. Total length of captured individuals ranged from 39 – 152 mm TL.

Recommendations: Results provide additional documentation of intermittent tributaries to the Verde River supporting populations of nonnative fishes. Colonization is likely occurring out of upstream stock tanks given that pools in East Fork MC Canyon and upper MC Canyon were often isolated complexes with barriers that would prevent upstream movement. Future surveys should prioritize visiting the upper reaches of tributaries first, where topographic complexity increases the probability of encountering exposed bedrock. Department staff plan to continue surveys in 2025 and will visit upper Hell Canyon (Upstream of Hell Canyon Tank) and possibly Bear Canyon (tributary directly east of MC).

Tables and Figures:

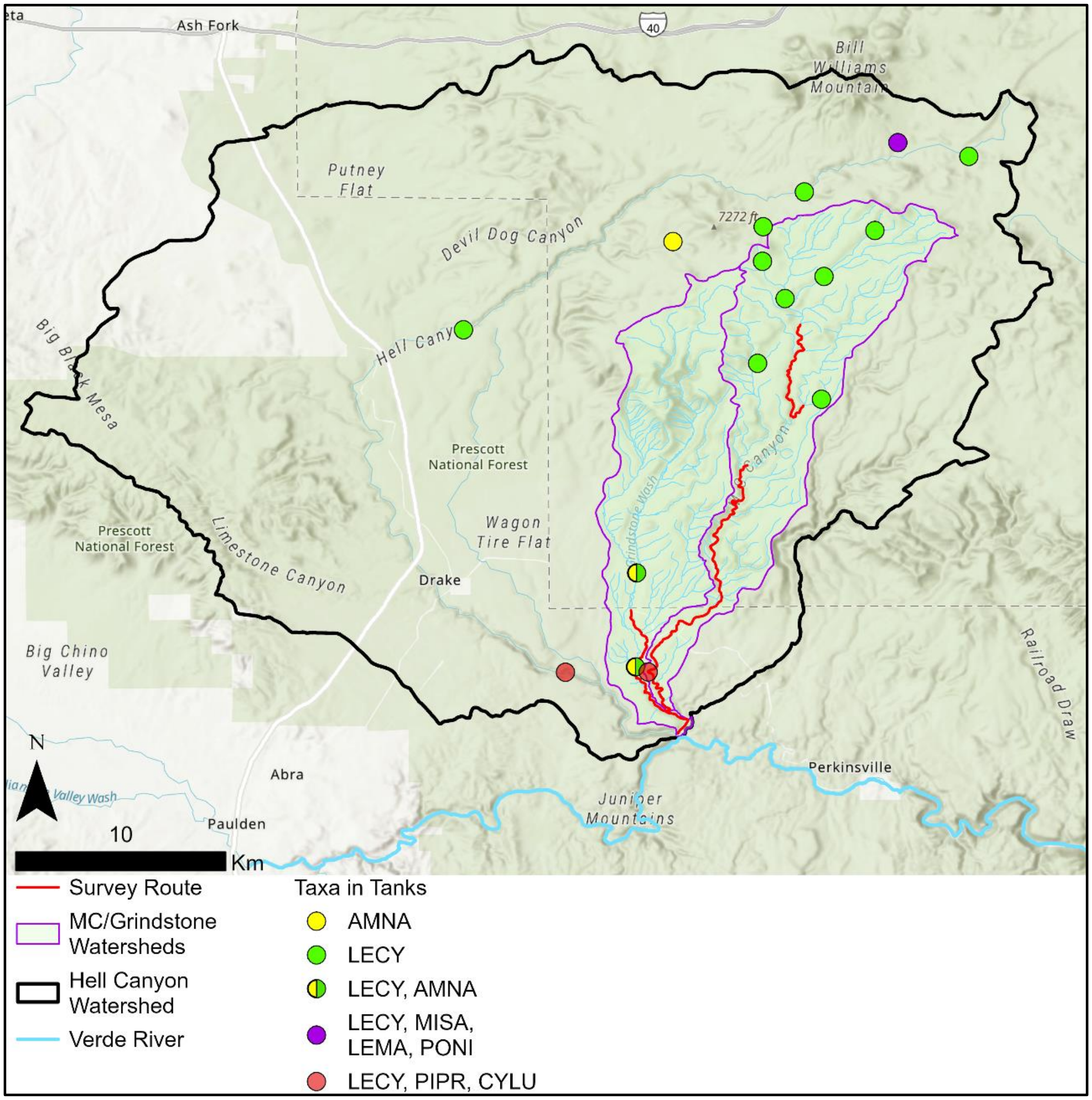

Figure 8.1.—Overview of Verde River tributary surveys conducted in 2024. Survey routes within Grindstone Wash (left) and MC Canyon (right) are shown in red. Also included are a subset of locations in the Hell Canyon watershed where nonnative fish were detected by Department Staff. Color indicates what species of nonnatives were recorded in tanks.

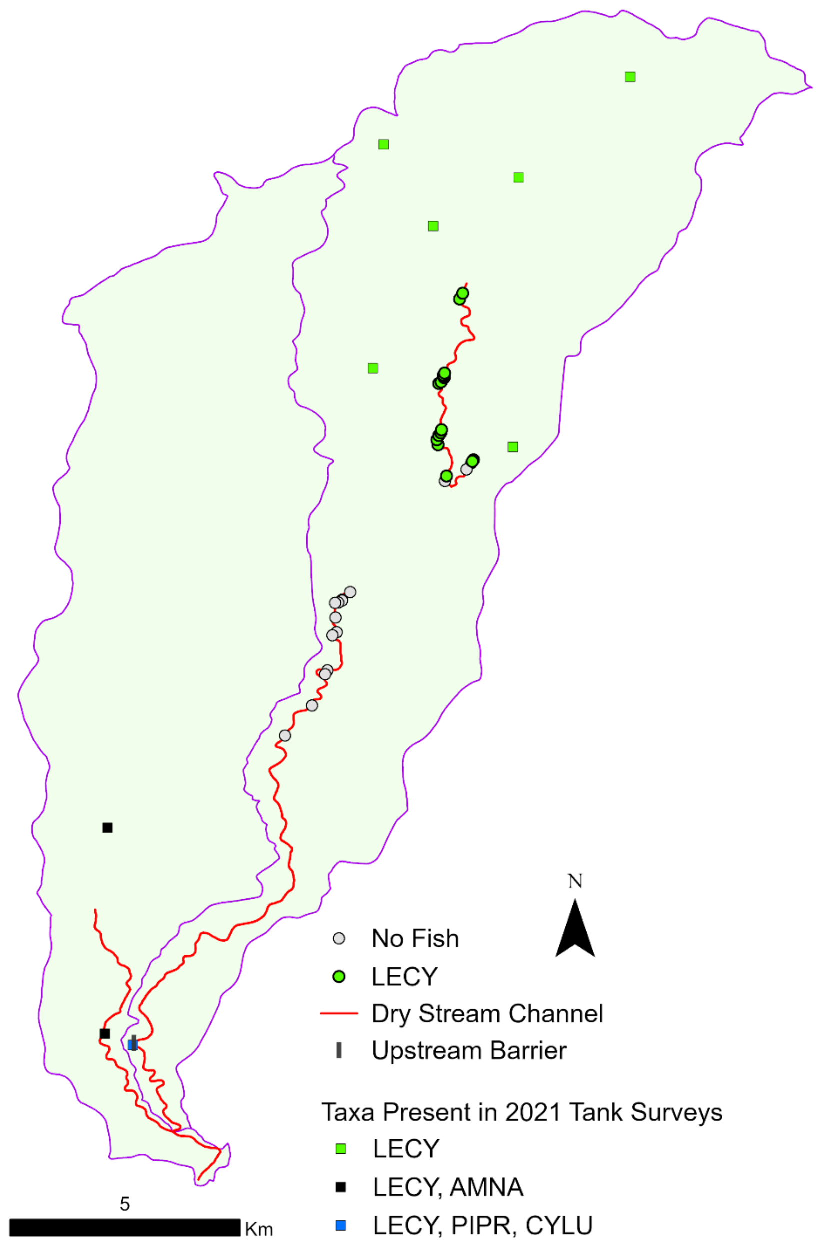

Figure 8.2.—Map depicting the Grindstone Wash (left) and MC Canyon (right) watershed boundaries. The red line illustrates dry stream channel and circles mark the location of pools. Pools are coded with presence/absence information whereby grey circles represent fishless pools and green circles are pools with Green Sunfish were detected. Results from 2021 stock tank surveys conducted in Grindstone Wash and MC Canyon are also shown.

9. Aquatic Research and Conservation Center O&M (Task HA-2006-2)

Strategic Plan Goals**:**

- Scientific Foundation
  - Goal 3. Improve propagation techniques for Spikedace and Loach Minnow
- Preventing Extinction and Managing Toward Recovery
  - Goal 1. Maintain the Aquatic Research and Conservation Center (ARCC) and explore alternative locations for establishment of hatchery stocks of upper Gila and San Francisco River lineages of Spikedace and Loach minnow.

Recovery Objectives:

- Spikedace recovery objective 8. Plan and conduct investigations on captive holding, propagation and rearing.
- Spikedace recovery objective 8.1. Determine wild stocks suitable for contribution to hatchery stocks.
- Spikedace recovery objective 8.2. Collect and transfer wild stocks to suitable facility.
- Loach Minnow recovery objective 8. Plan and conduct investigations on captive holding, propagation and rearing.
- Loach Minnow recovery objective 8.1. Determine wild stocks suitable for contribution to hatchery stocks.
- Loach Minnow recovery objective 8.2. Collect and transfer wild stocks to suitable facility.
- Gila Topminnow 1999 draft revised recovery objective 1.1. Maintain refugia populations of natural populations to ensure survival of the species.
- Desert Pupfish recovery objective 2. Reestablish Desert Pupfish populations.
- Gila Chub draft recovery plan objective 4. Establish and maintain refuge populations in protected ponds or hatcheries as appropriate.

Background: Reclamation funded construction of the Aquatic Research and Conservation Center (ARCC) on the grounds of the Department's Bubbling Ponds Hatchery. The main purposes of the facility were to develop propagation techniques for Loach Minnow and Spikedace, to establish refuge populations of all lineages, and to propagate fish for translocations. A number of improvements were made to the facility between 2000 and 2007. Beginning in 2014, Reclamation began providing funds (through USFWS) for a variety of improvements to ARCC, including new spawning raceways between existing structures, a new quarantine building, and new ponds.

Spikedace and Loach Minnow from all extant lineages were previously acquired under a separate sub-project (Task AZ-2003-1: Acquire Spikedace, Loach Minnow and rare populations of other native fish) and brought to ARCC, to establish refuge populations and support propagation efforts. The goal is to have each lineage represented by 500 adults. There are few natural populations left, and removing too many fish at a time could have negative impacts. The number of fish to collect from a given population is a coordinated decision between USFWS and state wildlife agencies, and is usually based on estimated number of fish in the stream derived from the most recent monitoring. New individuals should be brought into ARCC every year to maintain the population size and genetic diversity with wild stock. Counts of Spikedace and Loach Minnow brought into ARCC, brood stock, fish produced, and fish stocked each year since 2015 are presented in Table 9.1.

At various times Woundfin, Gila Topminnow, and Desert Pupfish were also brought to the facility to propagate fish in support of translocation efforts. Eagle Creek Roundtail Chub were brought to the facility in 2010 to establish a refuge population and support propagation efforts for the Blue River project. The putative Parker Canyon lineage of Gila Topminnow was brought to the facility to establish a refuge population in 2022, pending further genetic investigations. The facility also holds various other species for research or educational purposes.

In 2017 and 2018, ARCC staff began testing the influence of spawner density on propagation success of captive Spikedace and Loach Minnow, using the Aravaipa Creek lineage in replicates. ARCC staff planned to continue studying spawner density trials in 2019; however, not enough Aravaipa Creek lineage fish existed in broodstock to fill all six spawning raceways (i.e., two replicates of three densities) allocated to the experiment. Results from 2017 and 2018 showed that lowest spawner density tested produced the highest progeny per spawner on average. This finding, combined with the limited number of Aravaipa broodstock, led to all spawning raceways being setup identical to one another at the low densities identified in 2018 (i.e., Loach Minnow: 7.5 fish/m^2, Spikedace: 0.025 fish/L). Due to COVID-19 and subsequent restrictions, ARCC staff continued with this raceway setup for the 2020 and 2021 spawning seasons. The number of raceways used for each lineage is dependent on the overall brood stock size (a minimum of one raceway is allocated to each lineage) and the need for larval fish to supplement broodstock or to stock during repatriations, with each raceway having 32 adult fish and 13 nest sites for Loach Minnow and 34 adults for Spikedace. Loach Minnow were once again given nest sites consisting of medium sized cobbles arranged in 15 cm circles spaced 38 cm from edge of nest to edge of nest on a bed of small chip gravel. Beginning in 2022, we began to test the effect of nest spacing on larval fish production in Loach Minnow, using the Blue River lineage. Two replicate tanks each of 25 cm and 50.8 cm nest spacings were tested against the control spacing of the traditional 38 cm. Spikedace and other Loach Minnow lineages have used identical setups to those from 2019-2021, again, with the number of raceways allocated to lineages based on broodstock needs or with those with stocking locations identified for repatriation. For both species, larval fish were manually removed and counted once per week and placed in holding tanks. Algae were carefully removed as needed to minimize the potential effects of high algal biomass on spawning.

<u>Results</u>:

ARCC O&M

The Department continued to operate ARCC and maintain refuge populations of three lineages of Spikedace (Aravaipa Creek, upper Gila River, Gila River Forks) and four lineages of Loach

Minnow (Blue River, Aravaipa Creek, San Francisco River, and Gila River Forks). Unfortunately, the remaining individuals of the Eagle Creek lineage of Roundtail Chub were lost to a disease outbreak in 2024. The Department also continued to maintain Parker Canyon lineage Gila Topminnow at ARCC until more explicit genetic information can be obtained to inform management of this putative lineage.

During the 2024 spawning season, ARCC staff successfully spawned each Spikedace and Loach Minnow lineage held on station despite dealing with unexpected staffing shortages throughout the year (Table 9.1). Overall production numbers were at or near average levels for the last 10 years for most stocks despite broodstock numbers generally being at the lowest numbers since 2014 for most stocks (noting that the number of fish going into each spawning raceway is standardized; Figure 9.1, Figure 9.2). Gila River Spikedace brood was the second lowest in 2024, while production was the fourth highest on record. Gila Forks Spikedace brood was the lowest since 2014, while production was the fourth highest. Aravaipa Spikedace brood was the lowest while production was the sixth highest. Gila Forks Loach Minnow brood was the six lowest number while production was the third highest. San Francisco River Loach Minnow brood was the second lowest while production was the fifth highest. Blue River Loach Minnow brood was the lowest while production was the fourth highest. Aravaipa Creek Loach Minnow brood lowest while production sixth highest.

No new large-scale physical improvements to ARCC were completed in 2024, although minor updates to infrastructure, including plumbing, occur regularly.

Acquire Spikedace, Loach Minnow, and rare populations of other native fish

On July 1, 2024, Department staff collected 30 Speckled Dace from the Blue River in the vicinity of Blue Crossing Campground for a fish health assessment as a surrogate for Loach Minnow. No parasites or pathogens of concern were detected. Three Loach Minnow and one Roundtail Chub were captured and returned to the stream.

On September 3, 2024, Department staff assisted by Reclamation staff, collected 30 Longfin Dace from the west end of Aravaipa Creek upstream of the fish barrier for a fish health assessment. All fish were transported to the fish health laboratory at Department headquarters. Longfin Dace served as a surrogate for Spikedace and Loach Minnow.

On December 16, 2024, Department staff visited the east end of Aravaipa Creek to collect Loach Minnow and Spikedace for ARCC broodstock augmentation. The crew made a total of 16 seine hauls and captured 204 Spikedace, 51, Loach Minnow, 199 Longfin Dace, 48 Desert Sucker, 24 Speckled Dace, 8 Sonora Sucker, and 8 Roundtail Chub. A total of 51 Spikedace and 45 Loach Minnow were collected and transported to ARCC in an aerated cooler with no mortalities during capture or transport.

Recommendations: Recommendations for acquiring wild fish in 2024 include continuing to collect Spikedace and Loach Minnow from remnant populations for all lineages held at ARCC. Collections should continue to try to be made at regular intervals while minimizing impacts on the remnant population with the goal of maintaining broodstocks of 500 adults, subject to changes advised by the forthcoming genetic management plans. Continued wild fish collection is critical to maintaining ARCC's broodstocks as refuge populations and for consideration of the genetic health of our broodstocks. ARCC staff should coordinate with NMDGF regarding acquiring Loach Minnow and Spikedace from respective New Mexico lineages.

Tables and Figures:

Table 9.1.—Summary of number of broodstock (#B), number of offspring produced (#P), number of offspring stocked (#S), and number of wild fish brought in to augment existing broodstock (#A), for each species and lineage held at the Aquatic Research and Conservation Center, from 2016 through 2024. Data for years prior to 2016 can be located in Hickerson et al. (2021b; Table 1, Table 12). Numbers stocked do not include fish transferred to New Mexico.

Taxa	Extant Lineage		2016	2017	2018	2019	2020	2021	2022	2023	2024
Spikedace	upper Gila River	#B	531	267	159	254	219	176	131	72	108
		#P	0	384	352	2404	408	914	466	0	662
		#S	0	327	0	0	0	0	0	1667	0
		#A	0	0	0	0	0	0	0	0	0
Spikedace	Gila River Forks	#B	138	122	83	71	76	151	120	96	41
		#P	0	1183	195	1132	833	203	1252	0	949
		#S	0	1000	0	0	0	0	705	0	0
		#A	0	0	1	0	0	52	0	0	0
Spikedace	Aravaipa Creek	#B	262	382	331	523	529	379	158	180	98
		#P	120	1347	3214	4250	2182	1032	393	0	684
		#S	67	0	2234	0	2897	106	1707	0	0
		#A	80	160	0	322	49	0	27	67	51
Loach Minnow	Gila River Forks	#B	96	128	97	169	121	0	58	40	91
		#P	220	7	1207	665	15	0	475	0	584
		#S	0	159	0	0	0	0	0	0	0
		#A	0	110	145	0	0	102	0	0	0
Loach Minnow	San Francisco R.	#B	215	314	318	231	208	173	92	51	44
		#P	26	177	1627	601	3	541	310	0	422
		#S	0	243	0	0	0	0	0	205	0
		#A	0	0	0	0	0	0	0	0	0
Loach Minnow	Blue River	#B	214	156	117	290	266	364	244	172	111
		#P	426	47	6	713	16	919	278	0	310
		#S	390	0	0	0	500	400	0	0	0
		#A	12	0	223	80	269	130	4	45	0
Loach Minnow	Aravaipa Creek	#B	297	490	439	354	337	261	200	174	49
		#P	265	305	1848	1398	57	504	168	0	207
		#S	0	0	0	0	300	0	0	0	0
		#A	200	100	0	57	82	0	23	56	45
Roundtail Chub	Eagle Creek	#B	101	99	99	99	98	84	81	58	0
		#P	0	57	0	0	0	0	7405	0	0
		#S	1194	0	0	0	0	0	0	6640	0
		#A	0	0	0	0	0	0	0	0	0

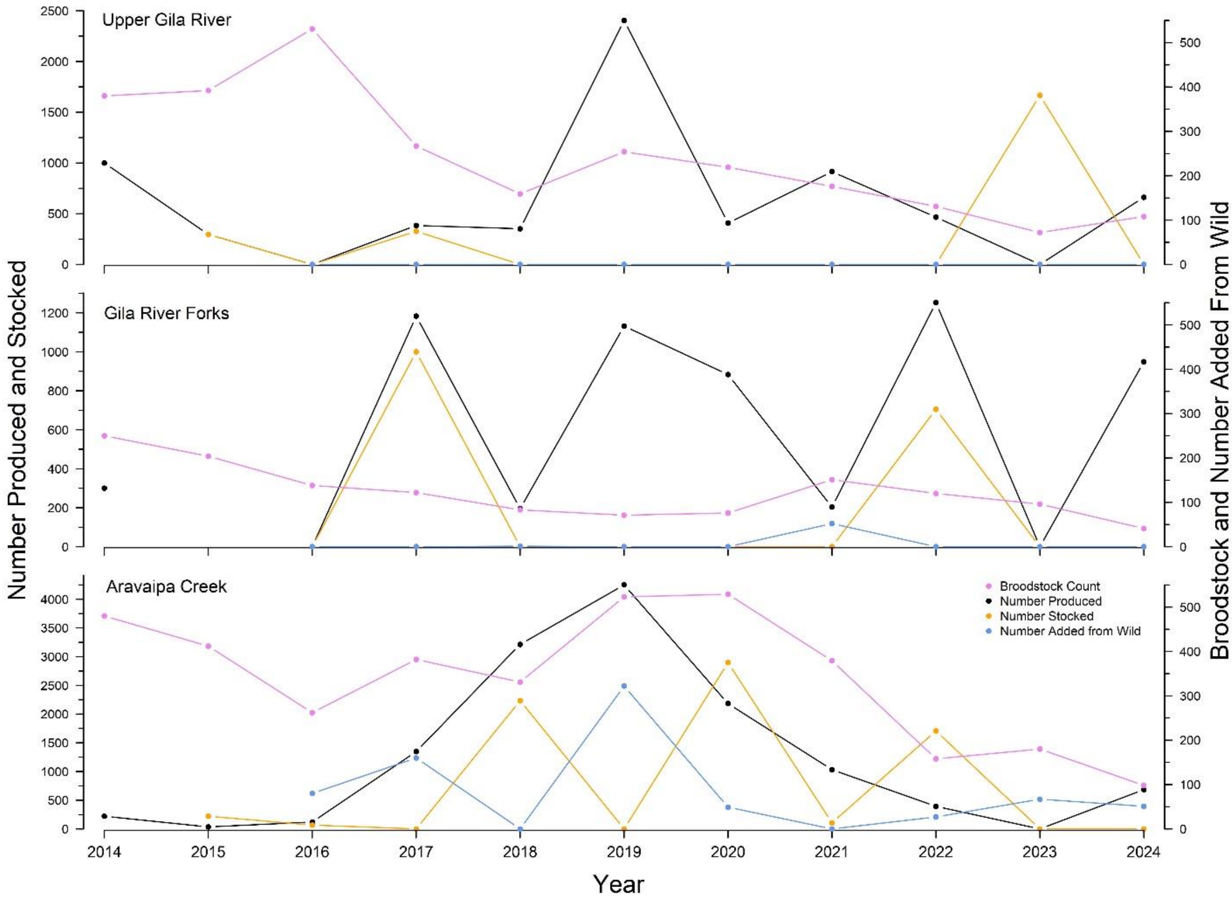

Figure 9.1.—Trends in the broodstock count (pink), number of fish produced (black), number of fish stocked out (orange), and number of fish added from the wild (blue) for each Spikedace lineage (top to bottom: Upper Gila River, Gila Forks, Aravaipa Creek) currently held at ARCC from 2014 to 2024.

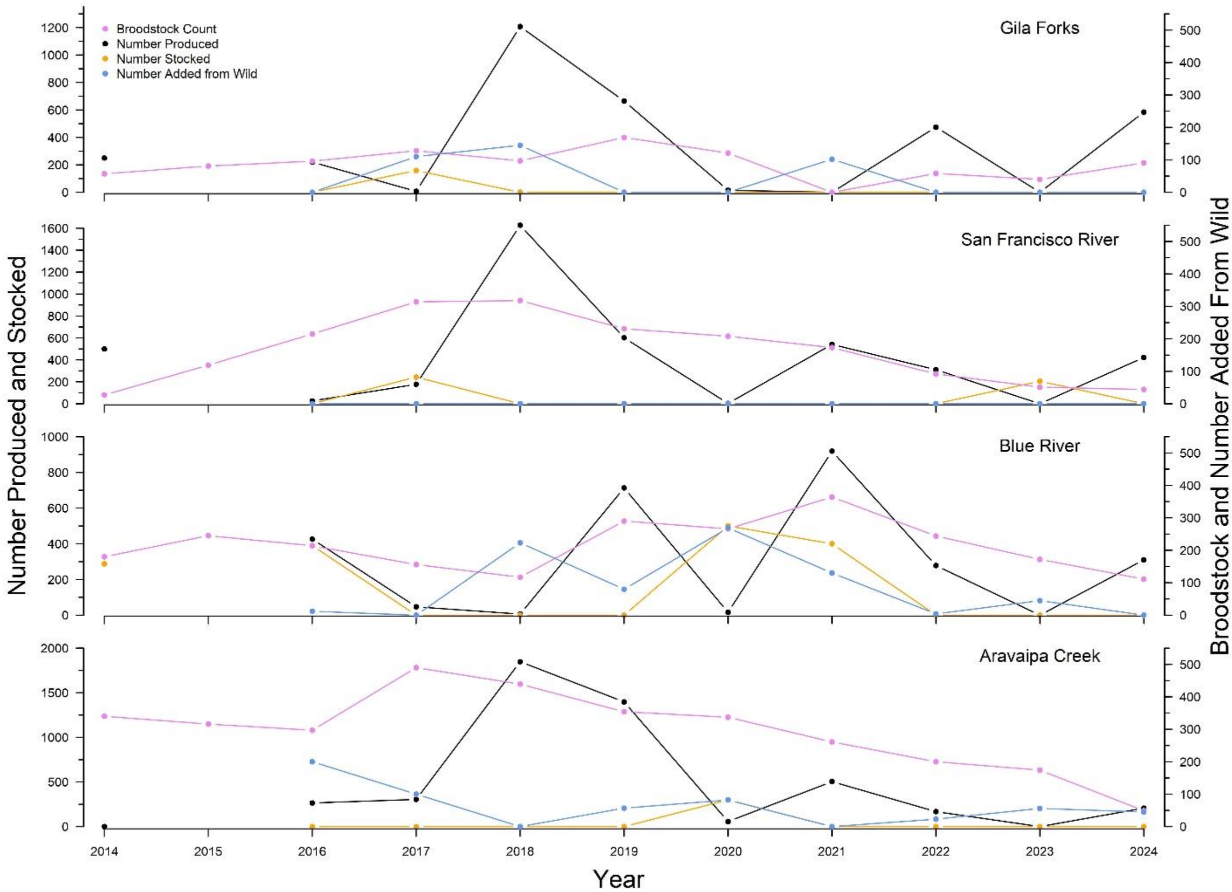

Figure 9.2.—Trends in the broodstock count (pink), number of fish produced (black), number of fish stocked out (orange), and number of fish added from the wild (blue) for each Loach Minnow lineage (top to bottom: Gila Forks, San Francisco River, Blue River, Aravaipa Creek) currently held at ARCC from 2014 to 2024.

LITERATURE CITED

Anderson, R., and P. Turner. 1977. Stream Survey of the San Francisco River. Department of Fishery and Wildlife Sciences, New Mexico State University, Las Cruces, NM.

Arizona Game and Fish Department. 2011. Fish Collection, transport, and stocking protocol: best management practice (BMP #4). Arizona Game and Fish Department, Phoenix.

Armstrong, D. P., and P. J. Seddon. 2007. Directions in reintroduction biology. Trends in Ecology and Evolution 23:20-25.

Bright, P. W., and T. J. Smithson. 2001. Biological invasions provide framework for reintroductions: selecting areas in England for pine martin releases. Biodiversity and Conservation 10:1247-1265.

Carle, F. L., and M. R. Strub. 1978. A new method for estimating population size from removal data. Biometrics 34:621-360.

Daly, C., M. Halbleib, J. I. Smith, W. P. Gibson, M. K. Doggett, G. H. Taylor, J. Curtis, and P. P. Pasteris. 2008. Physiographically sensitive mapping of climatological temperature and precipitation across the conterminous United States. International Journal of Climatology 28:2031-2064.

Ehret, S., and B. Dickens. 2009. Sabino Canyon survey trip report, June 3-4, 2009. Arizona Game and Fish Department, Region V, Tucson.

Ferguson, B.D., and M. P. Zeigler. 2021. Gila River Basin Native Fishes Conservation Program: New Mexico Department of Game and Fish Native Fish Conservation Efforts 2020 Annual Report. New Mexico Department of Game and Fish, Fisheries Management Division, Santa Fe.

Gill, C., and C. Carveth. 2006. Fish Creek Trip Report. Regional Trip report submitted to the Arizona Game and Fish Department. Mesa, Arizona.

Griffith, B., J. M. Scott, J. W. Carpenter, and C. Reed. 1989. Translocation as a species conservation tool: status and strategy. Science 245:477-480.

Hendrickson, D. A., and W. L. Minckley. 1984. Cienegas—vanishing climax communities of the American Southwest. *Desert Plants* 6(3):1984 (issued early 1985).

Hickerson, B. T., E. R. Grube, and A. T. Robinson. 2020a. Harden Cienega Creek Fish Restoration Project: Nonnative Fish Removal Plan. Draft.

Hickerson, B.T., E.R. Grube, J. Walters, and A.T. Robinson. 2020b. Gila River Basin Native Fishes Conservation Program: Arizona Game and Fish Department's Native Fish Conservation Efforts During 2019. An Arizona Game and Fish Department Annual Report for Cooperative Agreement No. R16AC00077 submitted to U.S. Bureau of Reclamation, Phoenix Area Office. Arizona Game and Fish Department, Aquatic Wildlife Branch, Phoenix.

Hickerson, B. T., E. R. Grube, K. T. Mosher, A. T. Robinson. 2021a. Successful Restoration of a Native Fish Assemblage in the Blue River, Arizona. North American Journal of Fisheries Management 41(3):746-756.

Hickerson, B. T., J. Walters and A. T. Robinson. 2021b. Gila River Basin Native Fishes Conservation Program: Arizona Game and Fish Department's native fish conservation efforts during 2020. An Arizona Game and Fish Department Annual Report for Cooperative Agreement No. R16AC00077 submitted to U.S. Bureau of Reclamation, Phoenix Area Office. Arizona Game and Fish Department, Aquatic Wildlife Branch, Phoenix.

Hickerson, B. T., J. E. Cleveland, and J. Walters. 2022. Gila River basin Native Fishes Conservation Program: Arizona Game and Fish Department's native fish conservation efforts during 2021. An Arizona Game and Fish Department Annual Report for Cooperative Agreement No. R16AC00077 submitted to U.S. Bureau of Reclamation, Phoenix Area Office. Arizona Game and Fish Department, Aquatic Wildlife Branch, Phoenix.

Love-Chezem, T. S., A. T. Robinson, and C. D. Crowder. 2015. Muleshoe Cooperative Management Area native fish restoration: 2014 activities. A Gila River Basin Native Fishes Conservation Program Progress Report for Task 3-75f; U.S. Fish and Wildlife Service Cooperative Agreement No. F09AC00084. Arizona Game and Fish Department, Nongame Wildlife Branch, Phoenix.

Meffe, G. K., D. A. Hendrickson, and J. N. Rinne. 1982. Description of a new topminnow population in Arizona, with observations on topminnow/mosquitofish co-occurrence. *The Southwestern Naturalist* 27(2):226–228.

Meffe, G. K., D. A. Hendrickson, W. L. Minckley, and J. N. Rinne. 1983. Factors resulting in decline of the endangered Sonoran topminnow *Poeciliopsis occidentalis* (Atheriniformes: Poeciliidae) in the United States. *Biological Conservation* 25(2):135–159.

Mussmann, S. M. 2024. Gila Topminnow Genetics Management Plan. Report submitted to the U.S. Bureau of Reclamation, Dexter, NM.

Ogle, D. H., J. C. Doll, A. P. Wheeler, and A. Dinno. 2023. FSA: Simple Fisheries Stock Assessment Methods. R Package version 0.9.4. R Foundation for Statistical Computing, Vienna.

R Development Core Team. 2023. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. Available: https://R-project.org/.

Reclamation (U.S.D.I. Bureau of Reclamation). 2010. Final environmental assessment, Blue River native fish restoration project, Apache-Sitgreaves National Forests, Greenlee and Apache Counties, Arizona. U. S. Department of the Interior, Bureau of Reclamation, Phoenix Area Office.

Shollenberger, K. R., B. R. Kesner, and P. C. Marsh. 2021. Gila River Basin Native Fish Monitoring 2020 Annual Report. An Annual Report for Agreement No. R17PC00108 submitted to U.S. Bureau of Reclamation, Phoenix Area Office. Marsh and Associates, Tempe.

Shollenberger, K. R., B. R. Kesner, and P. C. Marsh. 2022. Gila River Basin Native Fish Monitoring 2021 Annual Report. An Annual Report for Agreement No. R17PC00108 submitted to U.S. Bureau of Reclamation, Phoenix Area Office. Marsh and Associates, Tempe.

Shollenberger, K. R., P. C. Reap, B. R. Kesner, and P. C. Marsh. 2023. Gila River Basin Native Fish Monitoring 2022 Annual Report. An Annual Report for Agreement No. 140R3022F0030 submitted to U.S. Bureau of Reclamation, Phoenix Area Office. Marsh and Associates, Tempe.

U.S. Fish and Wildlife Service. 1983. Gila and Yaqui Topminnow Recovery Plan. U.S. Fish and Wildlife Service, Albuquerque, New Mexico. 56 pp.

U.S. Fish and Wildlife Service. 1991a. Spikedace Recovery Plan. Albuquerque, New Mexico. 38 pp.

U.S. Fish and Wildlife Service. 1991b. Loach Minnow Recovery Plan. Albuquerque, New Mexico. 38 pp.

U. S. Fish and Wildlife Service. 1998. Razorback sucker (*Xyrauchen texanus*) recovery plan. U. S. Fish and Wildlife Service, Denver, CO. 81 pp.

U.S. Fish and Wildlife Service. 2002. Razorback sucker (*Xyrauchen texanus*) Recovery Goals: amendment and supplement to the Razorback Sucker Recovery Plan. U.S. Fish and Wildlife Service, Mountain-Prairie Region (6), Denver, Colorado.

U.S. Fish and Wildlife Service. 2015. Gila chub (Gila intermedia) Draft Recovery Plan. U.S. Fish and Wildlife Service, Southwest Region, Albuquerque, New Mexico. 118 pp. + Appendices A-C.

Weedman, D. A. 1999. Gila topminnow, *Poeciliopsis occidentalis,* revised recovery plan. Draft. August 1999. U.S. Fish and Wildlife Service, Phoenix, AZ.

Weedman, D. A. and K. L. Young. 1995. Gila topminnow extirpation sites in Arizona: 1994-95 field season results. Nongame and Endangered Wildlife Program Technical Report 80. Arizona Game and Fish Department, Phoenix, AZ.

Weedman, D.A., A.L. Girmendonk, and K.L. Young. 1996. Status review of the Gila chub, *Gila intermedia,* in the United States and Mexico. Nongame and Endangered Wildlife Program Technical Report 91. Arizona Game and Fish Department, Phoenix, Arizona.

APPENDICIES

Appendix 1.—Reports detailing work completed by the Arizona Game and Fish Department under the Gila River Basin Native Fishes Conservation Program from 2007-2024.

Year	Citation
2007	C. Carter. 2007. Three Forks Loach Minnow Survey, August 28-30, 2007. *Arizona Game and Fish Department*, Phoenix, AZ.
2008	Robinson, A.T. 2008. Arnett Creek and Telegraph Canyon 1-Year Post-Stocking Monitoring, July 23, 2008. *Arizona Game and Fish Department*, Phoenix, AZ.
2008	Robinson, A.T., J. Bahm, and C. Carter. Loach Minnow Survey in the Three Forks Area, East Fork Black River, July - August, 2008. *Arizona Game and Fish Department*, Phoenix, AZ.
2008	Robinson, A.T. 2008. Mineral Creek Fish Survey, April 21-22, 2008. *Arizona Game and Fish Department*, Phoenix, AZ.
2008	Robinson, A.T. 2008. Mud Springs #18: Gila Topminnow and Desert Pupfish Monitoring on November 6, 2008. *Arizona Game and Fish Department*, Phoenix, AZ.
2009	Robinson, A.T. 2009. Muleshoe Cooperative Management Area Native Fish Repatriations, One-Year Post-Stocking Monitoring and First Augmentation Stocking September 15-17, 2008. *Arizona Game and Fish Department*, Phoenix, AZ.
2009	Robinson, A.T., D. Orabutt, and C. Crowder. 2009. Loach Minnow Survey of East Fork Black River and Tributaries during July 2009. *Arizona Game and Fish Department*, Phoenix, AZ.
2009	Robinson, A.T. 2009. Repatriation of Native Fishes to Fossil Creek: Summary of Monitoring and Stocking During 2008. *Arizona Game and Fish Department*, Phoenix, AZ.
2010	Robinson, A.T. 2010. Arizona Native Fish Recovery and Nonnative Fish Control. *Arizona Game and Fish Department*, Phoenix, AZ.
2010	Robinson, A.T., C. Crowder, and D. Boyarski. 2010. Mechanical removal of Nonnative Fishes from the Blue River During June 1-3, 2009. *Arizona Game and Fish Department*, Phoenix, AZ.

2010	Robinson, A.T., R. Timmons, D. Boyarski, and C. Crowder. Muleshoe Cooperative Management Area Native Fish Restoration: Monitoring and Stocking During 2009. *Arizona Game and Fish Department*, Phoenix, AZ.
2010	Boyarski, D.E., A.T. Robinson, and C.D. Crowder. 2010. Repatriation of Native Fishes to Fossil Creek: Summary of Monitoring and Stocking During 2009. *Arizona Game and Fish Department*, Phoenix, AZ.
2010	Robinson, A.T., D. Orabutt, and C. Crowder. 2010. Devils Canyon and Mineral Creek Fish Surveys During 2009. *Arizona Game and Fish Department*, Phoenix, AZ.
2010	Crowder, C.D., D.E. Orabutt, and A.T. Robinson. 2010. Gila Topminnow and Desert Pupfish Repatriations to Morgan City Wash and Chalky Spring, Monitoring and Stocking During 2010. *Arizona Game and Fish Department*, Phoenix, AZ.
2010	Robinson, A.T. 2010. Mud Springs #18: Gila Topminnow and Desert Pupfish Monitoring on August 26, 2009. *Arizona Game and Fish Department*, Phoenix, AZ.
2011	Robinson, A.T., R. Timmons, and C. Crowder. 2011. Muleshoe Cooperative Management Area Native Fish Restoration: Monitoring and Stocking During 2010. *Arizona Game and Fish Department*, Phoenix, AZ.
2011	Robinson, A.T. and C.D. Crowder. 2011. Repatriation of Native Fishes to Fossil Creek: Summary of Monitoring and Stocking During 2010. *Arizona Game and Fish Department,* Phoenix, AZ.
2011	Crowder, C.D. and A.T. Robinson. Devils Canyon Drainage Stock Tank Surveys During 2010 and 2011. *Arizona Game and Fish Department,* Phoenix, AZ.
2011	Robinson, A.T. and C.D. Crowder. 2011. Gila Topminnow and Desert Pupfish Repatriations to Morgan City Wash and Chalky Spring, Monitoring and Stocking During 2010. *Arizona Game and Fish Department*, Phoenix, AZ.
2011	Robinson, A.T. 2011. Mud Springs #18: Gila Topminnow and Desert Pupfish Monitoring on July 26, 2010. *Arizona Game and Fish Department*, Phoenix, AZ.

2011	Mosher, K.R., C.D. Crowder, and A.T. Robinson. 2011. O'Donnell Canyon and Turkey Creek Fish Surveys During 2010. *Arizona Game and Fish Department*, Phoenix, AZ.
2011	Mosher, K.R., C.D. Crowder, and A.T. Robinson. 2011. Robbins Butte Wildlife Area Gila Topminnow and Desert Pupfish Stockings and Monitoring During 2009 and 2010. *Arizona Game and Fish Department*, Phoenix, AZ.
2011	Yarush, K., A.T. Robinson, and C.D. Crowder. 2011. Attempted Establishment of Desert Pupfish in Walnut Springs, Mazatzal Mountains, Arizona, 2008-2011. *Arizona Game and Fish Department*, Phoenix, AZ.
2012	Robinson, A.T. and C. Crowder. 2012. Muleshoe Cooperative Management Area Native Fish Restoration: Monitoring and Stocking During 2011. *Arizona Game and Fish Department*, Phoenix, AZ.
2012	Robinson, A.T. and C.D. Crowder. 2012. Repatriation of Native Fishes to Fossil Creek: Summary of Monitoring and Stocking During 2011. *Arizona Game and Fish Department*, Phoenix, AZ.
2012	Crowder, C.D. and A.T. Robinson. 2012. O'Donnell Canyon and Turkey Creek Fish Surveys During 2011. *Arizona Game and Fish Department*, Phoenix, AZ.
2013	Robinson, A.T., C. Crowder, and D. Pearson. 2013. Blue River Native Fish Restoration Project: 2012 Annual Report. *Arizona Game and Fish Department*, Phoenix, AZ.
2013	Robinson, A.T., C. Crowder, and D. Pearson. 2013. Muleshoe Cooperative Management Area Native Fish Restoration: 2012 Monitoring. *Arizona Game and Fish Department*, Phoenix, AZ.
2013	Robinson, A.T., C.D. Crowder, and N. Robb. 2013. Fish Habitat Survey of Mescal Creek, Gila River Tributary below San Carlos Reservoir. *Arizona Game and Fish Department*, Phoenix, AZ.
2013	Pearson, D.B., A.T. Robinson, and C.D. Crowder. 2013. Attempted Establishment of Gila Topminnow and Desert Pupfish in Morgan City Wash and Chalky Springs, Lake Pleasant Regional Park, Arizona. *Arizona Game and Fish Department*, Phoenix, AZ.

2013	Pearson, D.B., A.T. Robinson, and C.D. Crowder. 2013. Establishment of Gila Topminnow and Desert Pupfish at Mud Springs (Site #18), Mesa Ranger District, Tonto National Forest. *Arizona Game and Fish Department*, Phoenix, AZ.
2013	Pearson, D.B., A.T. Robinson, and C.D. Crowder. 2013. Establishment of Gila Topminnow and Desert Pupfish at Robbins Butte Wildlife Area. *Arizona Game and Fish Department*, Phoenix, AZ.
2014	Robinson, A.T., C. Crowder, and D. Pearson. 2014. Blue River Native Fish Restoration Project: 2013 Annual Report. *Arizona Game and Fish Department*, Phoenix, AZ.
2014	Robinson, A.T. and C. Crowder. 2014. Muleshoe Cooperative Management Area Native Fish Restoration: 2013 Activities. *Arizona Game and Fish Department, Phoenix, AZ.*
2014	Staffeldt, R.R., D.B., Pearson, A.T. Robinson, and R. Babel. 2014. Attempted Establishment of Gila Topminnow in Buckhorn Spring, Arizona. *Arizona Game and Fish Department*, Phoenix, AZ.
2014	Robinson, A.T., C.D. Crowder, and D.B. Pearson. 2014. Repatriation of Native Fishes to Fossil Creek: Summary of Monitoring and Stocking During 2013. *Arizona Game and Fish Department*, Phoenix, AZ.
2014	Crowder, C.D., T.S. Love-Chezem, and A. S. Makinster. 2014. Mineral Creek Drainage Fish Surveys During 2013. *Arizona Game and Fish Department*, Phoenix, AZ.
2014	Frear, L.R., R.R. Staffeldt, A.T. Robinson, and C.D. Crowder. 2014. Attempted Establishment of Gila Topminnow in Walnut Spring (Site #392), Arizona. *Arizona Game and Fish Department*, Phoenix, AZ.
2015	Robinson, A.T. and T.S. Love-Chezem. 2015. Blue River Native Fish Restoration Project: 2014 Annual Report. *Arizona Game and Fish Department*, Phoenix, AZ.
2015	Robinson, A.T. and C. Crowder. 2015. Spikedace Survey of the Upper Verde River, During July 2011. *Arizona Game and Fish Department*, Phoenix, AZ.

2015	Love-Chezem, T.S., A.T. Robinson, and C. Crowder. 2015. Muleshoe Cooperative Management Area Native Fish Restoration: 2014 Activities. *Arizona Game and Fish Department*, Phoenix, AZ.
2015	Love-Chezem, T.S., A.T. Robinson, and C. Crowder. 2015. Translocation of Gila Chub from Dix Creek and Harden Cienega Creek to New Mexico. *Arizona Game and Fish Department*, Phoenix, AZ.
2015	Love-Chezem, T.S., A.T. Robinson, and C.D. Crowder. 2015. Repatriation of Native Fishes to Fossil Creek: Summary of Monitoring and Stocking During 2014. *Arizona Game and Fish Department*, Phoenix, AZ.
2015	Frear, L.R., R.R. Staffeldt, A.T. Robinson, and C.D. Crowder. 2015. Attempted Establishment of Gila Topminnow in Rock Spring, Arizona. *Arizona Game and Fish Department*, Phoenix, AZ.
2016	Robinson, A.T. and T.S. Love-Chezem. 2016. Blue River Native Fish Restoration Project: Report of 2015 Activities. *Arizona Game and Fish Department*, Phoenix, AZ.
2016	Mosher, K.R., T.S. Love-Chezem, and A.T. Robinson. 2016. Gila Topminnow and Desert Pupfish Stocking Activities During 2015. *Arizona Game and Fish Department*, Phoenix, AZ.
2016	Love-Chezem, T.S. and A.T. Robinson. 2016. Muleshoe Cooperative Management Area Native Fish Restoration: 2015 Activities. *Arizona Game and Fish Department*, Phoenix, AZ.
2016	Mosher, K.R., T.S. Love-Chezem, and A.T. Robinson. 2016. Loach Minnow Survey of East Fork Black River and Tributaries during July 2015. *Arizona Game and Fish Department*, Phoenix, AZ.
2017	Robinson, A.T., K.R. Mosher, and K. Smith. 2017. Gila River Basin Native Fishes Conservation Program: Arizona Game and Fish Department's Native Fish Conservation Efforts During 2016 and 2017 Work Plan. *Arizona Game and Fish Department*, Phoenix, AZ.
2018	Robinson, A.T. and K.R. Mosher. 2018. Gila River Basin Native Fishes Conservation Program: Arizona Game and Fish Department's Native Fish Conservation Efforts During 2017. *Arizona Game and Fish Department*, Phoenix, AZ.

2019	Hickerson, B.T. and A.T. Robinson. 2019. Gila River Basin Native Fishes Conservation Program: Arizona Game and Fish Department's Native Fish Conservation Efforts During 2018. *Arizona Game and Fish Department*, Phoenix, AZ.
2020	Hickerson, B.T., E.R. Grube, J. Walters, and A.T. Robinson. 2020. Gila River Basin Native Fishes Conservation Program: Arizona Game and Fish Department's Native Fish Conservation Efforts During 2019. *Arizona Game and Fish Department*, Phoenix, AZ.
2021	Hickerson, B.T., J. Walters, and A.T. Robinson. 2021. Gila River Basin Native Fishes Conservation Program: Arizona Game and Fish Department's Native Fish Conservation Efforts During 2020. *Arizona Game and Fish Department*, Phoenix, AZ.
2022	Hickerson, B.T., J.E. Cleveland, and J. Walters. 2022. Gila River Basin Native Fishes Conservation Program: Arizona Game and Fish Department's Native Fish Conservation Efforts During 2021. *Arizona Game and Fish Department*, Phoenix, AZ.
2023	Hickerson, B. T., C.K. Hedden, and J. Walters. 2023. Gila River Basin Native Fishes Conservation Program: Arizona Game and Fish Department's Native Fish Conservation Efforts During 2022. *Arizona Game and Fish Department*, Phoenix, AZ.
2024	Hickerson, B. T., C.K. Hedden, and J. Walters. 2023. Gila River Basin Native Fishes Conservation Program: Arizona Game and Fish Department's Native Fish Conservation Efforts During 2023. *Arizona Game and Fish Department*, Phoenix, AZ.

Appendix 2.—Summary of native fish translocated in Arizona during 2024 by the Department under the Gila River Basin Native Fishes Conservation Program. Easting and Northing are in UTMs (NAD 83; zone 12S).

Taxa	Water Name	Easting	Northing	Date	Lineage	# Stocked	# Mortalities
Roundtail Chub	East Fork Sabino Canyon	521043	3580990	5/7/2024	Sabino Canyon	7	0
Roundtail Chub	East Fork Sabino Canyon	521025	3580982	5/8/2024	Sabino Canyon	44	1
Gila Topminnow	Hidden Water Spring	459316	3717238	4/10/2024	Lower Santa Cruz	200	39
Gila Topminnow	Hidden Water Spring	459320	3717244	4/10/2024	Lower Santa Cruz	400	85
Gila Topminnow	Sharp Spring	540165	3468750	5/29/2024	Sharp Spring	253	0
Gila Topminnow	Sharp Spring	540489	3468807	5/29/2024	Sharp Spring	429	5

Appendix 3.—Summary of monitoring results during 2024 for the five-priority species and other target native fish species that were previously stocked into various waters in the Gila River Basin Arizona. Included is the number of sites sampled, number of individuals captured at a particular location (#Ind), the proportion of young of year individuals captured (%YOY), Mean relative abundance (CPUE) and standard error of the mean relative abundance (SE). Note, statistics reflect only the first pass of electrofishing at the Blue River and Spring Creek.

Taxa	Location	Date	Gear Type	Sample Size	Statistics	2024
Gila Topminnow	Aravaipa Creek	10/1/2024	Minnow Trap	10	#Ind	254
					%YOY	17.70
					Mean CPUE	11.09
					SE	9.15
Gila Topminnow	Aravaipa Creek	10/1/2024	Dip Net	15	#Ind	31
					%YOY	83.90
					Mean CPUE	4.77
					SE	3.13
Gila Topminnow	Aravaipa Creek	10/1/2024	Seine	16	#Ind	28
					%YOY	67.90
					Mean CPUE	7.00
					SE	6.67
Gila Topminnow	Arnett Creek	10/23/2024	Minnow Trap	2	#Ind	0
					%YOY	0
					Mean CPUE	0
					SE	0
Gila Topminnow	Arnett Creek	10/23/2024	Seine	1	#Ind	710
					%YOY	37.60
					Mean CPUE	NA
					SE	NA
Gila Topminnow	Maternity Wildlife Pond	8/26/2024	Minnow Trap	10	#Ind	2078
					%YOY	44.94
					Mean CPUE	75.14

					SE	19.04
Gila Topminnow	Sharp Spring	8/27/2024	Minnow Trap	24	#Ind	1
					%YOY	0
					Mean CPUE	0.02
					SE	NA
Gila Topminnow	Telegraph Canyon	10/23/2024	Minnow Trap	10	#Ind	452
					%YOY	26.76
					Mean CPUE	15.03
					SE	8.48
Gila Topminnow	Telegraph Canyon	10/23/2024	Dip Net	1	#Ind	1
					%YOY	100
					Mean CPUE	2.703
					SE	0
Gila Topminnow	Unnamed Drainage 68b	10/21/2024	Minnow Trap	15	#Ind	2,477
					%YOY	61.84
					Mean CPUE	51.95
					SE	12.29
Loach Minnow	upper Blue River	9/16/2024	Backpack Electrofisher	15	#Ind	23
					%YOY	4.34
					Mean CPUE	4.05
					SE	1.89
Loach Minnow	middle Blue River	9/23/2024	Backpack Electrofisher	12	#Ind	13
					%YOY	0
					Mean CPUE	3.01
					SE	1.14
Roundtail Chub	upper Blue River	9/16/2024	Backpack Electrofisher	15	#Ind	0
					%YOY	0
					Mean CPUE	0
					SE	0
Roundtail Chub	middle Blue River	9/23/2024	Backpack Electrofisher	12	#Ind	76

					%YOY	0
					Mean CPUE	16.59
					SE	2.86
Roundtail Chub	Rarick Canyon	10/16/2024	Angling	2	#Ind	5
					%YOY	0
					Mean CPUE	7.76
					SE	2.95
Roundtail Chub	Rarick Canyon	10/16/2024	Minnow Trap	8	#Ind	45
					%YOY	2.22
					Mean CPUE	0.236
					SE	0.07
Roundtail Chub	Rarick Canyon	10/16/2024	Mini-Hoop Net	11	#Ind	107
					%YOY	0
					Mean CPUE	0.41
					SE	0.09
Roundtail Chub	Rarick Canyon	10/16/2024	Dip Net	1	#Ind	61
					%YOY	100
					Mean CPUE	54.95
					SE	54.95
Roundtail Chub	Sabino Canyon	5/7/2024	Mini-Hoop Net	10	#Ind	8
					%YOY	
					Mean CPUE	0.39
					SE	0.09
Roundtail Chub	Sabino Canyon	5/7/2024	Snorkeling	1	#Ind	152
					%YOY	NA
					Mean CPUE	1302.85
					SE	0
Roundtail Chub	Sabino Canyon	5/7/2024	Dip Net	2	#Ind	4
					%YOY	100
					Mean CPUE	4.05

					SE	1.35
Spikedace	upper Blue River	9/16/2024	Backpack Electrofisher	15	#Ind	0
					%YOY	0
					Mean CPUE	0
					SE	0
Spikedace	middle Blue River	9/23/2024	Backpack Electrofisher	12	#Ind	471
					%YOY	0.84
					Mean CPUE	94.00
					SE	29.64
Spikedace	Spring Creek	9/10/2024	Backpack Electrofisher	3	#Ind	25
					%YOY	0
					Mean CPUE	26.88
					SE	23.20