

HABITAT SUITABILITY AND PREDICTIVE ANALYTICS FOR INFORMING THE
TRANSLOCATION OF AN ENDANGERED DESERT FISH, GILA CHUB
(*GILA INTERMEDIA*)

BY

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ABSTRACT

Translocation is an important element in the recovery of the federally endangered desert fish, Gila Chub *Gila intermedia*. Replication of populations is critical to ensuring resiliency to environmental and biological pressures throughout its range in Arizona, New Mexico, and Sonora, Mexico. While the species is found throughout the Gila River basin in Arizona, it is largely extirpated in New Mexico. Our goal was to determine the suitability of translocation sites in the San Francisco River of the Gila River basin in New Mexico based on predictions from systematic habitat assessments of extant populations. First, we elicited specific habitat attributes for Gila Chub from a species expert panel and applied known biological variables for the species as prerequisite constraints to four potential translocation sites. We used a random forest model as a predictive analytical tool trained on habitat and fish community attributes from five extant populations. We then predicted Gila Chub abundance at randomly selected 100 m reaches within translocation sites to determine the level of suitability. In the top random forest model, predicted Gila Chub abundance was supported by the following variables (in order of importance): proportion of pool mesohabitat, discharge, Speckled Dace *Rhinichthys osculus* abundance, proportion of fine substrate, median stream temperature, dominant substrate type, conductivity, elevation, hardness, and the proportion of riffle mesohabitat. We then compared the mean predicted Gila Chub abundance across translocation sites. Three sites were deemed suitable with average predicted abundances of 11.6 – 29.0 fish per 100 m. We deemed one site unsuitable because it did not meet the prerequisite biological constraint of spawning temperature duration (15 – 26°C for ≥ 60 consecutive days). Our work furthers the ecological understanding of the

species while increasing the likelihood of successfully translocating Gila Chub to the San Francisco River, New Mexico.

PREFACE

I have written this thesis in manuscript format for publication in the Journal of Fish and Wildlife Management. A large portion of the results are supported by the appendices which are substantive but informative.

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I would like to acknowledge the evolving taxonomic understanding of Gila Chub and the *Gila robusta* species complex. The taxonomic status of *Gila* spp. in the Gila River drainage has changed several times in the last 50 years (Rinne 1976; Bestgen and Propst 1989; Paroz and Propst 2007). Most recently, the U.S. Fish and Wildlife Service (2022; USFWS) announced findings on the petition to add the Lower Colorado River basin distinct population segment of Roundtail Chub *Gila robusta* to the Endangered Species List (ESA 1973). The findings suggest that Lower Colorado River basin Roundtail Chub do not warrant listing under the Endangered Species Act and that the USFWS should consider removing Gila Chub from the Endangered Species List under the presumption that Gila Chub and Roundtail Chub are the same species. This consideration by the U.S. Fish and Wildlife Service comes after the 2016 reclassification of Gila Chub, Roundtail Chub, and Headwater Chub *Gila nigra* into a single species, *Gila robusta*,

by the Committee on Names of Fishes, a joint committee of the American Fisheries Society and the American Society of Ichthyologists and Herpetologists (Page et al. 2017). In the past 25 years, however, numerous genetic and morphological studies and reviews have refuted (Minckley and DeMarais 2000; Chafin et al. 2021) and supported (Dowling et al. 2015; Carter et al. 2018; Copus et al. 2018) a single species designation. Regardless of Gila Chub's taxonomic status, it is in the interest of managers to translocate Gila Chub to the San Francisco River drainage to ensure resiliency because the species belongs to the river's historical native fish community (Minckley 1973; Sublette et al. 1990). We refer to Gila Chub as their own nominal species throughout the document. All species names have been verified as valid by the Integrated Taxonomic Information System (www.ITIS.gov, accessed 15 February 2023).

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INTRODUCTION

Endemic desert fishes of the American Southwest rely on suitable perennial habitat to fulfill their life requisites. The ecological effects of both a warming climate (Jaegar et al. 2014) and nonnative fish establishment (Dudley and Matter 2000) negatively impact native desert fishes by decreasing the physical area of suitable habitat or reducing the level of suitability. Native fishes of the Lower Colorado River basin are especially at risk due to their small and fragmented populations (Miller 1961; Fagan et al. 2002) which reduces their resiliency to events such as drought, wildfire, nonnative species establishment, and disease (Clarkson et al. 2004). Ruhí et al. (2016) found that nearly three-quarters of native fishes throughout the Lower Colorado River basin had a greater than 50% probability of population level extirpation and several native fishes of the region are already extinct (Jelks et al. 2008). Clarkson et al. (2004) suggests that the entirety of the basin's endemic ichthyofauna is in jeopardy. As a result, fisheries managers often implement management strategies that focus on preserving and restoring current populations of conservation concern to prevent local extirpation and extinction of native desert fishes.

Translocation is one important conservation tool to expand the range of at-risk species (Cochran-Biederman et al. 2015). Managers use translocation to conserve endemic species within their historical range through human assisted reintroduction (Dodd and Seigel 1991). In the 1990s, as much as 70% of recovery programs for threatened and endangered species contained a translocation element (Tear et al. 1993). Managers continue to utilize translocation in conservation efforts (Berger-Tal et al. 2019; Novak et al. 2021). However, less than half of translocation efforts across taxa are successful (Cochran-Biederman et al. 2015). Differences in

habitat suitability across release sites often explain why translocations are successful in some locations and fail in others (Moorhouse et al. 2009; Jarchow et al. 2016; Albrecht and Long 2019). If the habitat at a translocation site is unsuitable for the target species, the translocation will fail (Armstrong and Seddon 2008; George et al. 2009). Thus, assessing habitat suitability at potential translocation sites is a critical step in preventing translocation failure and predicting translocation success at a release site. Aquatic translocation efforts use habitat suitability parameters to determine adequate translocation sites (Daugherty et al. 2008; Fisk et al. 2014; Hickerson and Walters 2019). Though critical for the success of a translocation, selecting a reintroduction site is one of the most complicated steps of a reintroduction effort (Minckley 1995).

Translocation is part of the recovery strategy and a critical element in the recovery of Gila Chub *Gila intermedia*, an endemic cyprinid of the Gila River basin (USFWS 2005, 2015). In 2005, the U.S. Fish and Wildlife Service (USFWS) listed Gila Chub as endangered with critical habitat under the U.S. Endangered Species Act (ESA 1973; USFWS 2005) and categorized the species as priority 2c (USFWS 2005). The designation describes a high degree of threat with high potential for recovery. Historically, Gila Chub inhabited streams throughout the Gila River basin in Arizona, New Mexico, and northern Sonora, Mexico (Rinne and Minckley 1970; Varela-Romero et al. 1992), but the USFWS (2005) estimates that Gila Chub have been eliminated from approximately 85% of their previously occupied habitat. Between 1981 and 1999, the distribution of the species declined by nearly 16% (Olden and Poff 2005), primarily due to competition and predation from nonnative fishes as well as habitat degradation from livestock grazing (USFWS 2005). As of 2005, Gila Chub populations were reported to inhabit

approximately 30 isolated waterbodies (USFWS 2005), but over half of these populations were described by Weedman et al. (1996) as “unstable” and all the populations were considered vulnerable to degradation or loss from wildfire, grazing practices, nonnative species establishment, development, recreation, or a combination of the aforementioned (Weedman et al. 1996; USFWS 2005).

Though several translocation events have occurred in Arizona and New Mexico, the New Mexico translocations experienced varying levels of success (USFWS 2015). In 2010 and 2011, Gila Chub were translocated from Dix Creek, Arizona to Red Rock Ciénega, New Mexico but subsequent surveys determined the fish was rarely observed (USFWS 2015). Gila Chub were also translocated into Mule Creek, New Mexico in 2012 where they continue to persist in small numbers (NMDGF et al. 2019). Despite the two attempts to translocate Gila Chub, the species is considered largely extirpated from New Mexico (W. J. Koster, University of New Mexico, 1948, unpublished field notes; Bestgen and Propst 1989; Paroz and Propst 2007).

The San Francisco River is the largest tributary of the upper Gila River and within the species’ historical range (Figure 1; USFWS 2005). Bestgen and Propst (1989) commented that *Gila* spp. with “...greater tendencies towards the Gila Chub phenotype” were relatively common and widespread throughout the San Francisco River prior to the prolonged drought of the 1950s. The last confirmed capture of Gila Chub in the San Francisco River, New Mexico, hereby Upper San Francisco River, was in 1948 (Koster, field notes, unpublished). A combination of habitat degradation (USFWS 2005), nonnative species establishment (Bestgen and Propst 1989; USFWS 2005), and drought leading to the decline of pool and ciénega habitat (Hendrickson and Minckley 1984; USFWS 2005) resulted in the presumed extirpation of Gila Chub from the

Upper San Francisco River. However, the drainage has undergone changes in livestock management which may contribute to the success of native desert fishes (Carman 2006). Additionally, while much of the Upper San Francisco River is continuous, small diversions on the river aid in keeping nonnative species abundances above the diversions relatively low (Carman 2006; Clarkson and Marsh 2013).

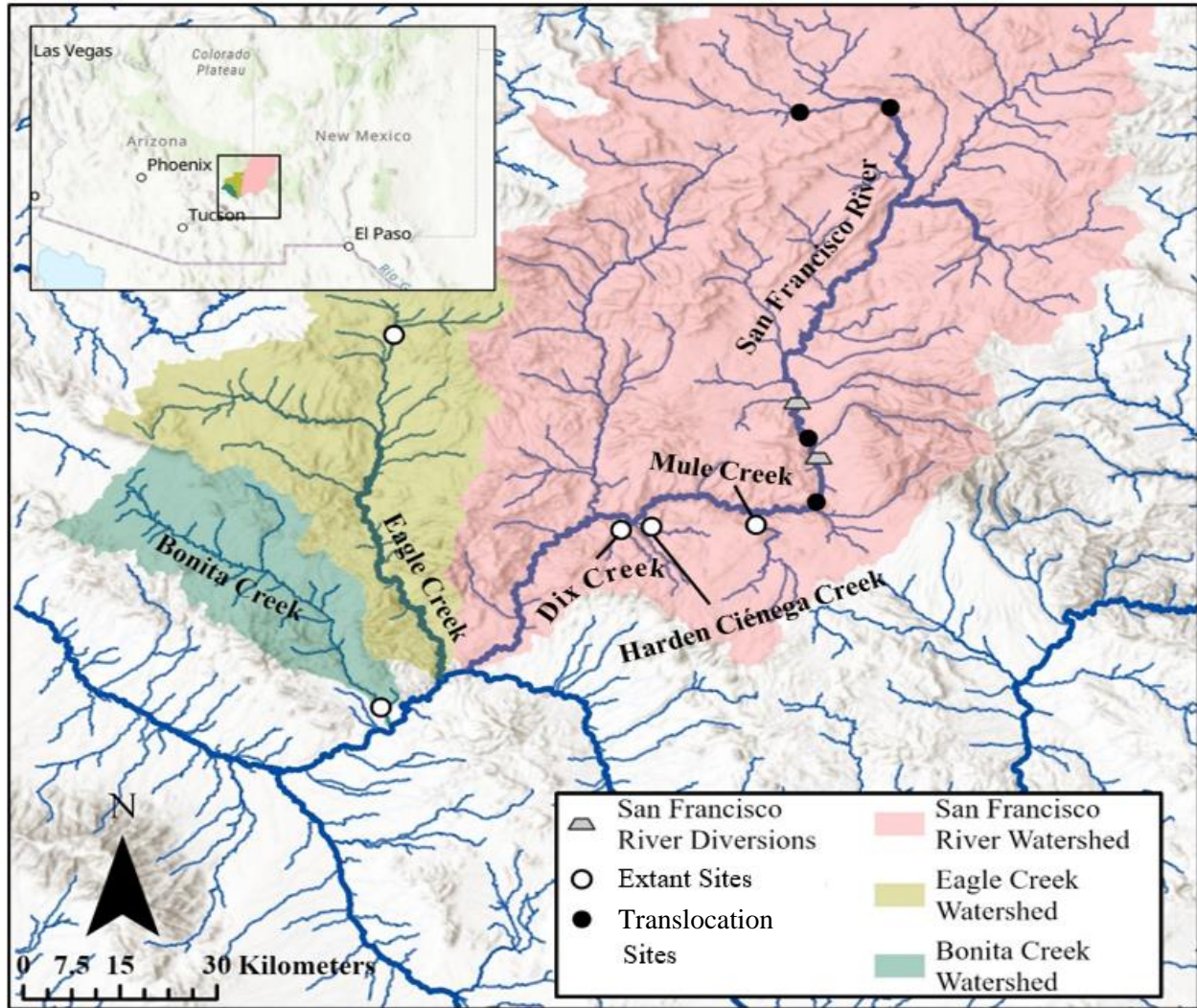


Figure 1. Map of the San Francisco River watershed and surrounding basins in Arizona and New Mexico in relation to the Gila Chub *Gila intermedia* extant sites, translocation sites, and San Francisco River diversions (Alma Diversion and Pleasanton Diversion).

Preliminary observations by species authorities suggest that portions of the Upper San Francisco River may provide suitable habitat to support a Gila Chub translocation effort (Dustin Myers, U.S. Forest Service-Carson National Forest, personal communication). Additionally, in 2020, Marsh and Associates captured an unidentified *Gila* spp. during a survey of the Upper San Francisco River downstream of the Pleasanton Diversion (Shollenberger et al. 2021). *Gila* spp. DNA was also detected in November 2020 using environmental DNA (eDNA; Dustin Myers, U.S. Forest Service-Carson National Forest, personal communication). Though these detections do not confirm the presence of Gila Chub, streams with characteristics similar to the Upper San Francisco River were envisioned by Clarkson et al. (2004) to be ideal for native fish recovery due to relatively limited numbers of nonnative fishes and the presence of remote and inaccessible refuge locations.

Translocation of Gila Chub to suitable reintroduction segments of the Upper San Francisco River may promote the resilience and continuance of the species. Thus, we aimed to quantitatively determine if suitable translocation sites for Gila Chub exist in the Upper San Francisco River. Our objectives were to (1) characterize habitat variables in streams with extant Gila Chub populations, (2) determine initial suitability of potential translocation sites in the Upper San Francisco River using a suite of prerequisite biological constraints, (3) apply a predictive analytic approach to identify the relationships between variables deemed necessary to promote Gila Chub abundance, and (4) offer recommendations for potential translocation sites.

METHODS

Study Areas

Extant Site Selection

We used extant and translocated Gila Chub populations to establish relationships between habitat variables, fish community variables, and Gila Chub abundance. We conducted a pilot study in May 2020 to gather information about the hydrology, substrate, and water quality at potential translocation sites in the Upper San Francisco River. We shared this information with species authorities, fisheries biologists, and land managers, who suggested extant populations with similar attributes to the potential translocation sites to best capture the ranges of habitat attributes at the extant sites that we would likely observe at the translocation sites. We selected extant populations in Bonita Creek, Eagle Creek, Harden Ciénega Creek, and Dix Creek Arizona, as well as a translocated population in Mule Creek, New Mexico, referred to collectively as the Gila Chub extant sites (Figure 1).

We selected a scale of 100 m to examine habitat variables at the extant sites. Other studies with similar objectives have used this scale to assess habitat variable importance as it relates to native fish abundance (Wilson and Belk 2001; Hickerson and Walters 2019). Since Gila Chub are minimally dispersive (Bestgen et al. 1987) the 100 m scale is appropriate. By generating random numbers corresponding to all available and accessible 100 m reaches within a given site, we randomly selected eight reaches to survey, representing between 5 and 10% of available reaches in most extant and translocation sites. High discharge and prolonged summer monsoons prohibited us from safely surveying all eight reaches at some sites. As a result, less reaches were surveyed in some extant and translocation sites.

Translocation Site Selection

The Upper San Francisco River is primarily located in Catron County, New Mexico flowing through both the Gila National Forest and private lands. Approximately 2,300 ha of surface water is withdrawn annually from the Upper San Francisco River and its tributaries for agriculture and livestock production (Longworth et al. 2005). The potential translocation sites span a large portion of the Upper San Francisco River (Figure 1) and were selected by species authorities (Dustin Myers, U.S. Forest Service-Carson National Forest, personal communication) due to the sites' perceived abundant pool habitat and limited numbers of nonnative fish. The downstream-most site (Hot Springs site) is located near the *Gila* spp. environmental eDNA and the 2020 *Gila* spp. collections. The site is downstream of two diversions, the Pleasanton Diversion and Alma Diversion (Figure 1). The Pleasanton Diversion represents a partial fish barrier, impeding nonnative fish passage upstream and promoting low abundance of nonnative fishes (Paroz et al. 2006; Clarkson and Marsh 2013; USFWS 2015). The Alma Diversion, located upstream of the Pleasanton Diversion (Figure 1), is also considered an effective fish barrier (Clarkson and Marsh 2013). While these diversions may impede connectivity among native populations, they also act to reduce or prevent the upstream migration of nonnative fishes and may increase the success of translocation efforts (Fausch et al. 2006; Clarkson and Marsh 2008). Two of the four potential translocation sites (the Luna and the Box sites) are located upstream of both the Alma Diversion and Pleasanton Diversion. The New Mexico Department of Game and Fish Permanent Monitoring (NMDGF Permanent) site is located between the two barriers. All the sites in the Upper San Francisco River experience a bimodal discharge pattern, with peak discharge occurring during spring snow runoff and summer monsoons (Mueller 1984).

Extant Site Descriptions: Bonita Creek (May and October 2021)

Bonita Creek, Arizona is an approximately 65 km long tributary of the Gila River, originating on the San Carlos Apache Indian Reservation in Arizona. The USFWS designated Bonita Creek as critical habitat for Gila Chub in 2005 (USFWS 2005). From its origin, Bonita Creek flows south through Graham County, Arizona before entering the Gila River (Arthun and Zaines 2020). Much of Bonita Creek resides within the U.S. Bureau of Land Management (USBLM) managed Gila Box Riparian National Conservation Area, though city, tribal, and private lands also comprise the Bonita Creek watershed (USBLM 2019). Bonita Creek is subject to dewatering from the city of Safford's infiltration gallery, located approximately 8 km upstream from the confluence of Bonita Creek and the Gila River (Heindl and McCullough 1961; USBLM 2019).

The infiltration gallery serves as one of two fish barriers on Bonita Creek. In 2008, the U.S. Bureau of Reclamation (Reclamation) installed a concrete fish barrier through the Gila River Basin Native Fishes Conservation Program 6 km downstream of the infiltration gallery. The barrier promotes extant native fish species that include Gila Chub, Speckled Dace *Rhinichthys osculus*, Longfin Dace *Agosia chrysogaster*, Sonora Sucker *Catostomus insignis*, and Desert Sucker *Pantosteus clarkii* (USBR and USBLM 2007; USBLM 2019; USBLM 2021). Other native desert fishes including Loach Minnow *Rhinichthys cobitis*, Spikedace *Meda fulgida*, Desert Pupfish *Cyprinodon macularius*, and Gila Topminnow *Poeciliopsis occidentalis*, were translocated to Bonita Creek in 2008 and 2009 after the completion of the Reclamation barrier and chemical renovation to remove nonnative fishes (Robinson et al. 2009). Nonnative Western Mosquitofish *Gambusia affinis*, Green Sunfish *Lepomis cyanellus*, Fathead Minnow *Pimephales*

promelas, and Yellow Bullhead *Ameiurus natalis* were redetected upstream of the Reclamation barrier soon after its construction (USBLM 2021). Shortly after the redetection, mechanical removal methods were initiated with the goal of complete eradication; mechanical removal of nonnative fishes in Bonita Creek are ongoing and eDNA suggests that Green Sunfish have since been eradicated above the barrier (USBLM 2021). Nonnative Northern Clearwater Crayfish *Faxonius propinquus* are also present in the stream and Anchor Worm *Lernaea cyprinacea* parasitizes fish in Bonita Creek (USBLM 2021).

Highly variable stream reaches from narrow canyon-bound stretches to broad beaver dam complexes characterize Bonita Creek. Between the Reclamation barrier (downstream) and the San Carlos Apache Indian Reservation (upstream), we surveyed five of the eight 100 m reaches in May and October of 2021 due to high discharge. All five of these reaches are located upstream of the Reclamation barrier. Upper Site 1 and the Gallery are located below the City of Safford's infiltration gallery, while Lee Trail, Red Knolls, and Midnight Canyon are located above (Figure 2A). We excluded Red Knolls from analysis because the habitat complexity and depth of the pools would not allow us to complete three-pass depletion needed to determine Gila Chub abundance.

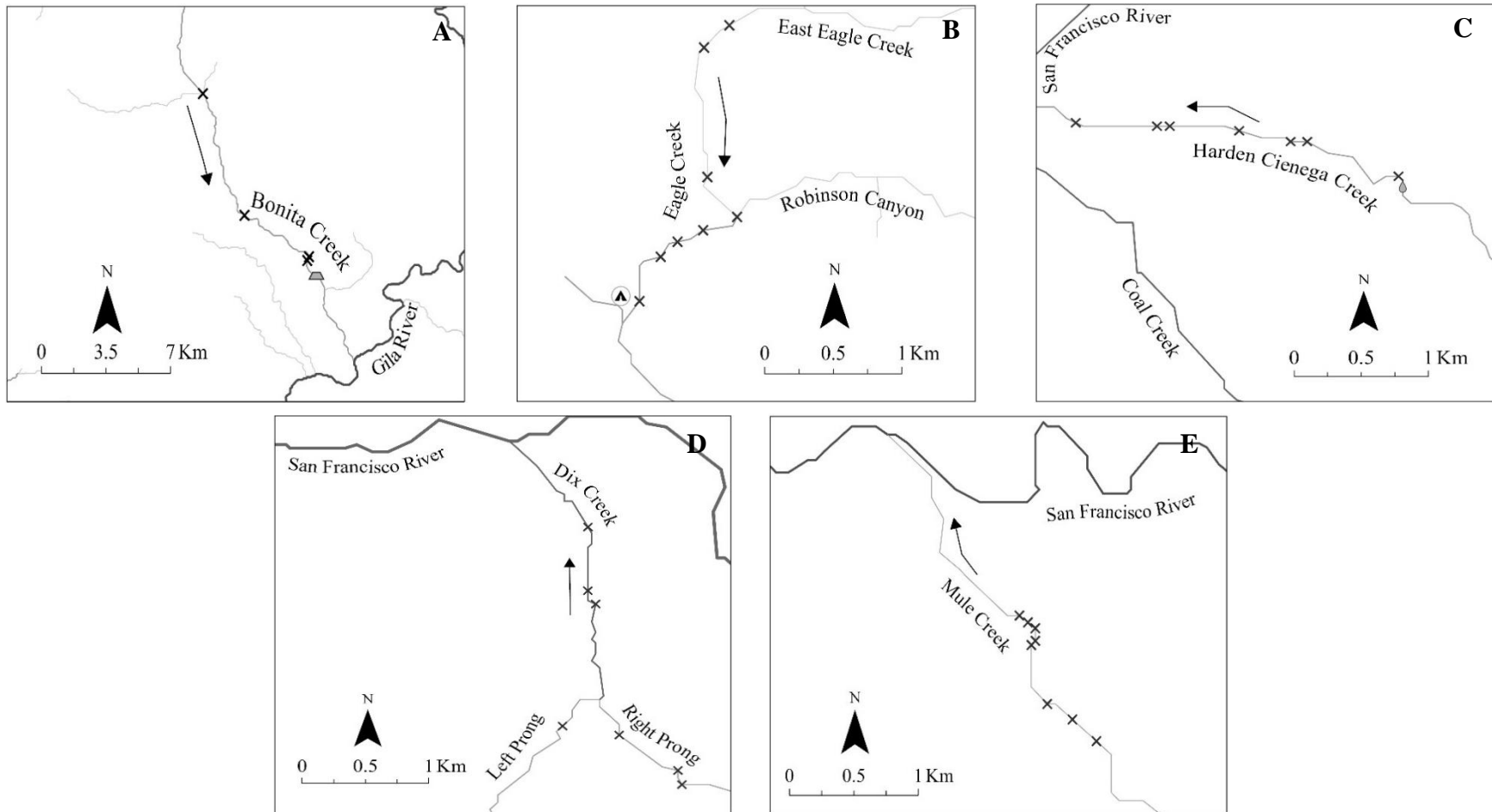


Figure 2. Maps of extant sites including the (A) Bonita Creek, Arizona study reaches and the U.S. Bureau of Reclamation barrier (trapezoid), (B) Eagle Creek, Arizona study reaches and Honeymoon Campground (tent symbol), (C) Harden Ciénega Creek study reaches and the waterfall barrier (raindrop symbol), (D) Dix Creek study reaches, and (E) Mule Creek, New Mexico study reaches. Small x represent study reach locations and arrows represent stream flow direction.

Extant Site Descriptions: Eagle Creek (November 2021)

Eagle Creek is an approximately 135 km second order tributary of the Gila River, primarily flowing through the Apache-Sitgreaves National Forest of eastern Arizona and private land (Marsh et al. 1991, 2003). From its headwaters in the White Mountains of Arizona, Eagle Creek meanders south through conifers and broad river valleys then enters a nearly 64 km canyon-bound stretch prior to joining the Gila River (Marsh et al. 1991). Groundwater withdrawal used for open-pit copper mining operations in Clifton and Morenci, Arizona dries portions of Eagle Creek near the confluence of the Gila River, threatening Gila Chub habitat (USFWS 2005). The USFWS designated 39 km of the upstream-most portion of Eagle Creek as critical habitat for Gila Chub (USFWS 2005).

The Eagle Creek critical habitat segment, hereby referred to as Upper Eagle Creek, is or has been home to Loach Minnow, Spikedace, Desert Sucker, Sonora Sucker, Roundtail Chub, Razorback Sucker *Xyrauchen texanus*, Gila Chub, and an unknown trout species *Oncorhynchus* sp. (Marsh et al. 1991; USFWS 2005). A small diversion dam, directly upstream of the critical habitat designation, limits nonnative fish establishment upstream (Marsh et al. 1991; USFWS 2005). However, limited numbers of nonnative Northern Clearwater Crayfish occupy Upper Eagle Creek (Clarkson et al. 2009). In 2021, the Bear wildfire burned over 10,000 ha of land in the headwaters of Eagle and East Eagle Creek, which may have impacted the fish community in Eagle Creek. We surveyed eight 100 m reaches in Upper Eagle Creek between the U.S. Forest Service (USFS) managed Honeymoon Campground (downstream) and the confluence of East Eagle Creek and Upper Eagle Creek (upstream) in November of 2021 (Figure 2B).

Extant Site Descriptions: Harden Ciénega Creek (May and June 2022)

Harden Ciénega Creek is a 23 km first order tributary of the San Francisco River. The stream is primarily located in the Apache-Sitgreaves National Forest in eastern Arizona, though its headwaters originate in the Gila National Forest, New Mexico. Near its origin, water from Harden Ciénega Creek feeds irrigation channels and livestock watering tanks in New Mexico and Arizona (Hickerson et al. 2021a). As it flows towards the San Francisco River, Harden Ciénega Creek passes through narrow canyons and slightly wider valley bottoms. As one of two tributaries of the San Francisco River with extant Gila Chub populations, the USFWS lists the entirety of Harden Ciénega Creek as critical habitat for the species (USFWS 2005).

Endemic desert fishes in Harden Ciénega Creek include Longfin Dace, Speckled Dace, Sonora Sucker, Desert Sucker, and Gila Chub (USFWS 2005; Hickerson et al. 2021a). Additionally, the Arizona Game and Fish Department (AZGFD) stocked 631 Gila Topminnow in Harden Ciénega Creek in 2019, but post-stocking surveys failed to detect the fish (Hickerson et al. 2021a). Prior to 2015, Gila Chub were limited to the downstream-most 4 km of Harden Ciénega Creek due to a large waterfall barrier, impeding fish passage (Hickerson et al. 2021a). However, in 2013, AZGFD concluded that additional habitat for Gila Chub existed above the barrier and, in 2015, managers captured 102 Gila Chub and placed them above the waterfall, hereby referred to as Upper Harden Ciénega Creek (Hickerson et al. 2021a). AZGFD augmented Upper Harden Ciénega Creek with five individuals in 2018 and 104 individuals in 2019 (Hickerson et al. 2021a). The Gila Chub population in Upper Harden Ciénega Creek shows signs of reproductive success and is considered established (Hickerson et al. 2021a). Nonnative fish also inhabit Harden Ciénega Creek and Upper Harden Ciénega Creek, including Red Shiner

Cyprinella lutrensis (K. Field, personal observation) and Green Sunfish (Hickerson et al. 2021a). Since 2020, the AZGFD has annually suppressed Green Sunfish; however, Green Sunfish detected within stock tanks in the Harden Ciénega Creek drainage may act as an upstream source population during heavy monsoon precipitation (Hickerson et al. 2021a). We surveyed seven reaches between the San Francisco River confluence (downstream) and the waterfall barrier (upstream) in May and June of 2022 (Figure 2C).

Extant Site Descriptions: Dix Creek (October 2021)

Dix Creek is a first-order stream comprised of a left and right prong, both of which reside entirely on the Apache-Sitgreaves National Forest in Arizona. A partial rock barrier, approximately 2 km from the San Francisco River confluence, marks the beginning of Gila Chub critical habitat (Weedman et al. 1996; USFWS 2005). The critical habitat designation continues upstream to the confluence of the left and right prongs of Dix Creek, 2 km up the left prong, and 4.8 km up the right prong (USFWS 2005). However, Turner and List (2006) state that only 6 km of Dix Creek are perennial. Dix Creek is one of only a few streams in Arizona that are devoid of nonnative fish species (Turner and List 2006), although populations of nonnative Northern Clearwater Crayfish have been observed in the stream (Kent Mosher, U.S. Bureau of Reclamation, personal communication). Native fishes of Dix Creek include populations of Longfin Dace, Speckled Dace, Sonora Sucker, Desert Sucker, and Gila Chub (Weedman et al. 1996). We surveyed seven reaches in the perennial portion of Dix Creek in October of 2021 (Figure 2D).

Extant Site Descriptions: Mule Creek (May 2022)

Mule Creek is a first order tributary of the Upper San Francisco River with headwaters near the town of Mule Creek, New Mexico. The downstream-most 5 km of Mule Creek reside on the Gila National Forest, New Mexico (NMDGF et al. 2012). Mule Creek is cooler and more shaded than the nearby Upper San Francisco River (R. Anderson and P. Turner, New Mexico State University, 1977, report to the New Mexico Department of Game and Fish on stream surveys of the San Francisco River). Narrow and steep canyon walls surround the USFS managed portion of the stream. Approximately 800 m upstream from the Upper San Francisco River confluence, Mule Creek becomes intermittent, flowing in the morning and drying by the afternoon (Dustin Myers, U.S. Forest Service-Carson National Forest, personal communication). The intermittent portion of Mule Creek resides on the Lower San Francisco Wilderness Study Area.

Mule Creek is the only stream surveyed in New Mexico with an extant Gila Chub population. The USFWS did not designate Mule Creek as critical habitat for Gila Chub in 2005 because Gila Chub were not documented at that time. Unlike the other Gila Chub extant sites, Mule Creek's Gila Chub population is a translocated one, although anecdotal reports indicate that there may have been an extant population of remnant Gila Chub in Mule Creek prior to the translocation (Propst 1999; NMDGF et al. 2019). From 2012–2014, a multi-agency effort translocated 299 Gila Chub from Harden Ciénega Creek, Arizona to Mule Creek, New Mexico (NMDGF et al. 2019).

In addition to Gila Chub, Mule Creek is home to native desert fishes including Sonora Sucker, Desert Sucker, Speckled Dace, and Longfin Dace. The intermittency of the downstream-

most portion of Mule Creek suppresses nonnative fishes to relatively low numbers. However, Black Bullhead *Ameiurus melas*, Smallmouth Bass *Micropterus dolomieu*, Green Sunfish (NMDGF et al. 2012, 2019), and Common Carp *Cyprinus carpio* (K. Field, personal observation) have been reported in Mule Creek. We surveyed eight 100 m reaches in Mule Creek, upstream of the intermittent portion, in May of 2022 (Figure 2E).

Translocation Site Descriptions: Hot Springs site (August and October 2021)

The Hot Springs site is located on the main stem of the Upper San Francisco River near the Frisco Hot Springs, which have been documented releasing 40.6°C water into the Upper San Francisco River (Anderson and Turner, report, unpublished). A significant amount of geothermal activity occurs in this portion of the Upper San Francisco River (Witcher and Hahman 1979). Native fish species documented near the Hot Springs site include Desert Sucker, Sonora Sucker, Loach Minnow, Speckled Dace, Longfin Dace, Spikedace (Anderson and Turner, report, unpublished; Schollenberger et al. 2021), and Gila Chub, prior to the 1950s (Bestgen and Propst 1989). Nonnative species documented near the Hot Springs site include Channel Catfish *Ictalurus punctatus*, Flathead Catfish *Pylodictis olivaris*, Smallmouth Bass, Largemouth Bass *Micropterus salmoides*, Common Carp, Red Shiner, Western Mosquitofish, Yellow Bullhead, Fathead Minnow, and Green Sunfish (Anderson and Turner, report, unpublished; Bestgen 1985; Schollenberger et al. 2021). We surveyed six 100 m reaches between the confluence of Big Dry Creek (downstream) and the Pleasanton Diversion (upstream; Figure 3A) in August and October of 2022.

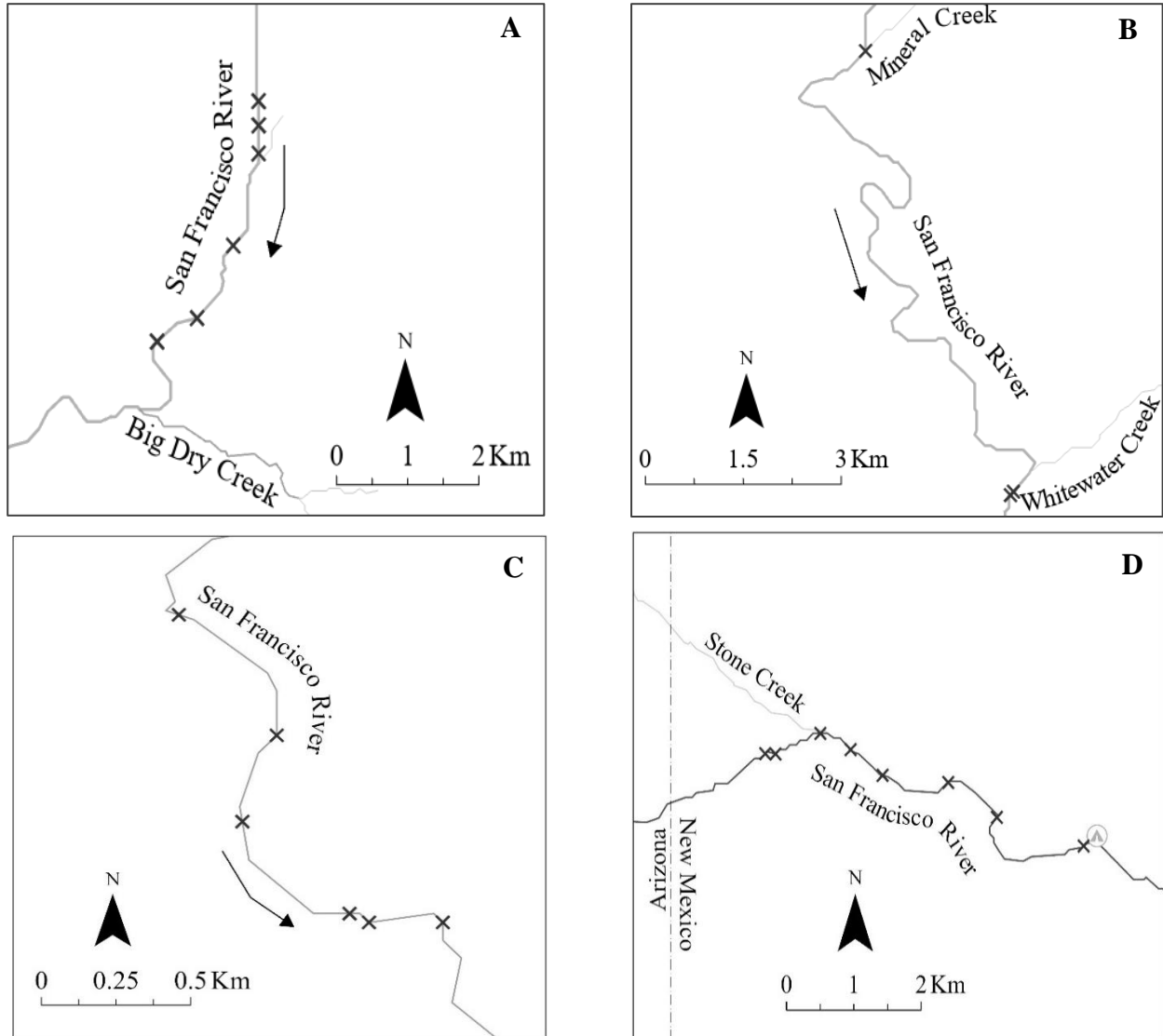


Figure 3. Maps of potential translocation sites on the San Francisco River, New Mexico, including the (A) Hot Springs site study reaches, (B) New Mexico Department of Game and Fish Permanent Monitoring site study reaches, (C) the Box study reaches, and (D) and the Luna site study reaches in relation to the Arizona/New Mexico state boundary (dashed line), as well as the Head of the Ditch Campground (tent symbol). Small x represent study reach locations and arrows represent stream flow direction.

Translocation Site Descriptions: New Mexico Department of Game and Fish Permanent Monitoring site (August and November 2022)

The New Mexico Department of Game and Fish Permanent Monitoring (NMDGF Permanent) site is located near the town of Glenwood, New Mexico. Directly upstream of the site, the Upper San Francisco River flows through pastures, irrigated fields, and towns (Paroz et al. 2006). A tributary, Whitewater Creek, flows into the Upper San Francisco River near the downstream-most study reaches (Figure 3B). Between 1988 and 2005, surveys indicated native Longfin Dace, Sonora Sucker, Desert Sucker, Speckled Dace, Spikedace, and Loach Minnow were present at the site (Paroz et al. 2006). Surveys also detected low densities of nonnative Western Mosquitofish, Rainbow Trout *Oncorhynchus mykiss*, Largemouth Bass, and Fathead Minnow (Paroz et al. 2006). However, surveys in 2021 captured only Desert Sucker, Sonora Sucker, Longfin Dace, Speckled Dace, and Spikedace (NMDGF 2022). We surveyed three reaches between the Pleasanton Diversion (downstream) and the Alma Diversion (upstream; Figure 3B) in August and November of 2022. Hazardous weather and stream flow conditions repeatedly prevented us from surveying more reaches at this site.

Translocation Site Descriptions: The Box site (October 2022)

The Upper San Francisco River flows through a steep and tall canyon known as the Box, upstream of Reserve, New Mexico, approximately 85 river km upstream of the Alma Diversion. Species experts were particularly optimistic about this location's suitability for Gila Chub due to its steep canyons, abundant pools, and remote location (Dustin Myers, U.S. Forest Service-Carson National Forest, personal communication). The site has a steep slope, is heavily shaded, and possesses numerous high-quality pools (Gary L. Helbing, Gila National Forest, 1982,

memorandum to the Gila National Forest range, forest, and wildlife staff). However, due to the inaccessibility of this portion of the river, no published or official fish community surveys exist for the site (Paroz and Propst 2007). Access to the site is challenging because the site is flanked, both upstream and downstream, by private property. However, in 1948, W. J. Koster detected a single *Gila* sp. in the Upper San Francisco River near Reserve, New Mexico (Koster, field notes, unpublished). We surveyed six 100 m reaches between the two Gila National Forest boundaries in October 2022 (Figure 3C).

Translocation Site Descriptions: The Luna site (August and September 2022)

The Luna site is the upstream-most translocation site, so named for its proximity to the town of Luna, New Mexico. The site is downstream approximately 10 km from Luna Lake, Arizona. The Luna site extends from the Arizona-New Mexico border (upstream) to the USFS managed Head of the Ditch Campground (downstream). A small tributary, Stone Creek, flows into the Upper San Francisco River within the site (Figure 3D). Field notes indicate that Desert Sucker, Speckled Dace, Red Shiner, Rainbow Trout, Fathead Minnow, and possibly Rio Grande Sucker *Pantosteus plebeius* were captured at the Luna site prior to our surveys (E. Jaquez and M. Martinez, NMDGF, and P. Morrison, USFS Biologist 1993 field notes). The surveyors were uncertain about the Rio Grande Sucker identification during the 1993 surveys, but research by Turner et al. (2019) suggests that these captures are plausible. We surveyed eight 100 m reaches between the USFS managed Head of the Ditch Campground (downstream) and the Arizona-New Mexico state line (upstream) in August and September of 2022 (Figure 3D).

Site Characterization

Initial Suitability Determination

Identifying impediments to native fish establishment is critical for assessing suitability of translocation sites (Hickerson et al. 2021b). In this investigation, determining if potential translocation sites were initially suitable for Gila Chub was critical for gauging potential translocation success. The extant Gila Chub sites could not provide information on locations unsuitable for Gila Chub due to a biological constraint since Gila Chub are found in all the extant sites. Thus, investigations of potential translocation sites began with an initial suitability determination. We based initial suitability on the known prerequisite constraints for Gila Chub of maximum lethal temperature ($< 37^{\circ}\text{C}$), spawning temperature duration ($15 - 26^{\circ}\text{C}$ for ≥ 60 days), and minimum dissolved oxygen concentration (≥ 4 mg/L). Translocation locations with nonnative fish species present also required special consideration. Piscivorous nonnative fish species such as Largemouth Bass, Smallmouth Bass, Channel Catfish, Flathead Catfish, Green Sunfish, and others, are the greatest threat to Gila Chub populations in the Gila River basin (USFWS 2015). Thus, the presence of nonnative fishes at a translocation site is a serious concern and may be an impediment to Gila Chub establishment. However, the vast majority of Gila Chub occupied streams also have nonnative fish populations (USFWS 2005). Pool et al. (2013) suggests that investigating possible restoration sites with high abundances of nonnative fish species is an important action in expanding Gila Chub populations. If the potential translocation site did not meet the initial suitability requirements for the maximum lethal temperature, spawning temperature duration, or the minimum dissolved oxygen concentration, we considered

the site to be unsuitable for translocation. Translocation sites with nonnative fish species present, were reviewed with caution.

The lethal maximum temperature for Gila Chub is approximately 38°C (Carveth et al. 2006); thus, sites with stream temperatures $\geq 37^\circ\text{C}$ are unsuitable for Gila Chub (USFWS 2015). Additionally, while the Gila Chub spawning period typically reaches its peak in spring and summer, temperature elicits spawning in Gila Chub and thus, multiple spawning attempts in one reproductive season can occur if spawning temperatures are maintained (Bestgen 1985; Schultz 2009). The USFWS (2015) suggests that stream temperatures should remain between 15 and 26°C for a minimum of 60 consecutive days to allow for Gila Chub to spawn and for larval development to occur (Bestgen 1985; Schultz and Bonar 2016). We launched one ProV2 (Model U22-002; $\pm 0.21^\circ\text{C}$ accuracy, Onset Computer Corporation) in a pool and one Tidbit (Model UTBi-001; $\pm 0.21^\circ\text{C}$ accuracy) attached to a boulder or canyon wall using methods described by Isaak et al. (2013), at each site to record hourly water temperatures throughout one year. The loggers were launched in the spring and summer of 2021 and retrieved in the summer and fall of 2022 to capture the seasonal range of stream temperatures. From the temperature data, we determined if each translocation site met the prerequisite constraints for spawning temperature duration and maximum lethal temperature by calculating the longest number of consecutive days the stream temperature remained between 15 and 26°C and the maximum water temperature for the entire launch duration. We could not locate data loggers at the Hot Springs site. Thus, we utilized temperature data from the nearest data logger located at the NMDGF Permanent site, approximately 7 km upstream. Potential translocation sites that did not maintain spawning

temperatures for a minimum of 60 consecutive days or had recorded stream temperatures $\geq 37^{\circ}\text{C}$ were considered unsuitable for Gila Chub.

We could not identify specific investigations describing minimum dissolved oxygen concentrations for Gila Chub. However, it is reasonable to assume that Gila Chub experience similar physio-chemical constraints of other endemic desert fishes. To that end, the USFWS (2015) describes the appropriate dissolved oxygen concentrations for Gila Chub as ≥ 4 mg/L (USFWS 2015). We used instantaneous dissolved oxygen measurements averaged across reaches to determine if potential translocation sites met the minimum requirements of dissolved oxygen. Potential translocation sites with average dissolved oxygen concentrations of < 4 mg/L were considered unsuitable for Gila Chub.

Researchers and managers have thoroughly documented the deleterious impacts of nonnative fishes on endemic desert fishes (Minkley 1973; Dudley and Matter 2000; Olden and Poff 2005; USFWS 2005; Hedden et al. 2016). Similar to much of the endemic ichthyofauna of the Lower Colorado River basin, Gila Chub evolved with no aquatic predators and few competitors (Miller 1961). Piscivorous nonnative fishes such as Flathead Catfish, Channel Catfish, Largemouth Bass, and Smallmouth Bass pose substantial predation risk to juvenile and adult Gila Chub (Minkley 1973; Bestgen and Propst 1989; USFWS 2015) and additional research suggests nonnative Green Sunfish negatively impact smaller size classes of Gila Chub and can cause recruitment failure (Dudley and Matter 2000). There is also evidence that Rainbow Trout will consume native desert fishes when introduced to desert streams (Marsh and Douglas 1997; Yard et al. 2011). Even small-bodied Western Mosquitofish will consume Gila Chub eggs and larval fish (USFWS 2005).

Nonnative fish at translocation sites can also compete with Gila Chub for food and habitat. For example, Common Carp prefer low velocity pool habitat with fine substrate (Swee and McCrimmon 1966). Gila Chub are also associated with pool habitat (Barber and Minckley 1966; Griffith and Tiersch 1989). Therefore, Common Carp and Gila Chub may compete for optimal pool habitat. Similarly, Crowl et al. (1992) suggests that Rainbow Trout will compete with native fishes for optimal cover locations. In addition to habitat, competition for food resources is also a concern. Fathead Minnow, while not native to the San Francisco River, are native to other waterbodies in New Mexico, such as the Rio Grande (Hoagstrom et al. 2010). Both Gila Chub and Fathead Minnow are dietary generalists, feeding on a variety of food items including aquatic invertebrates, detritus, diatoms, and terrestrial vegetation (Seegert et al. 2014), resulting in Fathead Minnow as possible food competitors. Minckley (1973) also suggests Red Shiner competes with native southwestern cyprinids and may be partially responsible for their decline.

The presence of nonnative piscivorous fishes negatively impacts native fish translocation efforts (Al-Chokhachy et al. 2011). The USFWS (2005) describes critical habitat for Gila Chub, as it pertains to nonnatives, as locations that are either devoid of nonnative fish, or locations with low enough nonnative fish abundance for Gila Chub to reproduce and survive. The agency further emphasizes that recovery efforts should only take place in locations free of nonnatives or where nonnatives can be controlled, but acceptable numbers of nonnatives are not well-defined and difficult to quantify (USFWS 2015). Therefore, we did not exclude sites with nonnatives present from further investigation or consideration.

Habitat Variable Data Collection

We conducted a literature review to determine habitat and fish community variables known to be important to Gila Chub. We searched using terms “Gila Chub” and “*Gila intermedia*”, in both the Google Scholar and all databases within the New Mexico State University Library search engine between June 2020 and January 2021. Due to the classification status and naming of members of the *Gila* genus in the Gila River basin, we also searched using terms “Roundtail Chub” and “*Gila Robusta*”. We presented the selected variables to a Gila Chub Species Authority Panel on 26 February 2021. Panel participants included species authorities and fisheries biologists from AZGFD, NMDGF, USBLM, Reclamation, USFS, U.S. Geological Survey, and the Jicarilla Game and Fish Department. Species authorities and collaborators agreed upon the following variables and their presumed importance to Gila Chub abundance. We collected the variable measurements at each selected 100 m reach within all sites.

We obtained the elevation of each reach at the downstream most portion of the 100 m segment by positioning ourselves at one half of the wetted width and capturing the elevation using a Garmin GPSmap 62s GPS. Water quality measurements included instantaneous temperature (°C), dissolved oxygen (mg/L), and conductivity (µS/cm; Hach HQ40d). We measured visual clarity (cm) using a Secchi disk (Tyler 1968; Davies-Colley and Smith 2007). At each reach, we measured water hardness (mg/L as total CaCO₃) and alkalinity (mg/L as total CaCO₃) using APHA (1975a–c) methodology. We obtained pH using a calibrated meter (Oakton pH model 150).

We collected benthic macroinvertebrates using a 250 µm mesh kicknet with modified methodology described by Barbour et al. (1999). We disturbed substrate and stream banks in all

mesohabitat types (pools, riffles, runs, and cascades) within each reach and immediately fixed the samples in 95% ethyl alcohol until sorting at the New Mexico State University laboratory. During sorting, we identified all individual macroinvertebrates collected to the lowest taxonomic group possible, typically family, using textbooks and dichotomous keys (Merritt et al. 2008). Taxon standardization was necessary in samples when we could not identify the family of a rare macroinvertebrate. We used the *Distribute-Parents-Among-Children* method described by Cuffney et al. (2007) to assign samples with known orders, but unknown families. When parent taxa were present, but children taxa were not, we used the *Replace-Parent-Keep-Children-Grouped-Knowledge* method to assign samples to families (Cuffney et al. 2007). We standardized the sample size to 100 individuals using techniques by Wilson et al. (2015), which allows rare or infrequently captured macroinvertebrates the opportunity for representation in a standardized sample. This was necessary as our surveyors and effort varied among reaches. From our standardized samples, we calculated the Shannon-Weaver diversity index (Shannon and Weaver 1949; H) and Simpson diversity index (Simpson 1949; D) for each reach using the “vegan” package in R (Oksanen et al. 2020). Only the Shannon-Weaver diversity index was used in analysis.

We measured velocity (m/s) along perpendicular cross sections at the 20, 40, 60, 80, and 100 m mark of each reach for a total of five cross sections. At each mark we obtained the wetted width perpendicular to flow and divided it equally to obtain 10 velocities at 60% of depth across the stream. We measured velocity using a standard top-loading rod (metric) and either a Marsh McBirney model 2000 flow meter or Hach FH950 flow meter. From the velocity measurements, we calculated the proportion of refuge flow, described as velocity measurements < 0.1 m/s, by

dividing the number of velocity measurements classified as refuge flow by the total number of velocity measurements. We calculated the proportion of refuge flow because there is evidence that Gila Chub and Roundtail Chub select habitats with relatively low velocity (Barrett and Maughan 1995; Schultz 2009). We calculated fast flow (> 0.4 m/s) in the same manner since there is evidence that other members of the *Gila* genus, namely Roundtail Chub, cannot maintain neutral swimming posture at velocities > 0.4 m/s (Moran et al. 2018a) which may indicate that higher velocities may impact Gila Chub habitat preference. Moreover, Gila Chub prefer habitats with velocities < 0.5 m/s (Schultz 2009). From our velocity measurements, we calculated discharge (m^3/s) using the velocity-area method (Herschey 1993).

We collected information on both the aquatic and terrestrial vegetation at each reach. At each of the 20, 40, 60, 80, and 100 m cross sections, we used an adaptation of line-point intercept methodology described by Madsen (1999) for use in streams. We determined presence or absence of vegetation (aquatic or terrestrial) at 10, equally spaced points across the wetted width of the stream. Presence was marked as a “hit” if any part of the top-loading rod, used to measure velocity on the same points, touched any part of the plant. We then calculated the proportion of the stream covered by vegetation by dividing the number of “hits” by the total number of points. We also took one canopy cover measurement for each cross-section, facing upstream, with a model-A spherical densiometer positioned at one-half of the wetted width. We then obtained our final canopy cover measurement for the reach by averaging the percentage of canopy cover at each of the 20, 40, 60, 80, and 100 m cross sections. We counted large woody debris (LWD), defined as woody debris ≥ 1 m in length and ≥ 10 cm in diameter (Platts et al. 1987), within the

wetted width of each 100 m reach by walking the entirety of the reach and tallying the number of LWD observed.

We collected the bank angle on river right and river left at each of the 20, 40, 60, 80, and 100 m cross sections with a Nikon Forestry Pro range finder using protocols established by Heitke et al. (2008). If the bank was undercut (had an angle measuring $< 90^\circ$), we also measured the undercut depth of the bank (Heitke et al. 2008). We calculated the proportion of undercut banks by dividing the number of undercut bank angles by the 10 total bank angle measurements. By averaging the undercut depth measurements, we obtained the mean undercut depth for the reach. Some reaches did not have any undercut banks.

We defined and measured the length of the predominant in-stream habitat type or mesohabitat (pools, runs, riffles, cascades, or dry channel) to the nearest tenth of a meter for the entirety of each 100 m reach using visual methodology outlined by Kaufmann et al. (2006). When two or more mesohabitat types spanned the wetted width of the channel, we recorded the mesohabitat type that included the thalweg. In addition to length, we measured pool tail depth and pool maximum depth to calculate the pools' residual depths independently of discharge (Lisle 1987).

We measured the diameter of 100 substrate pieces while conducting a Wolman Pebble Count (Wolman 1954). We selected a random sample of substrate using zig-zag methodology (Bevenger and King 1995) and measured the diameter of the substrate according to Kaufmann et al. (2006). We measured diameter (mm) to classify substrate pieces into categories (silt, sand, gravel, cobble, boulder, and bedrock) based on size buckets described by Lane (1947). We determined the dominant substrate type by calculating the relative abundance of each category.

We examined each substrate using an underwater viewer and assigned an embeddedness rating between one and five. More embedded substrate had lower embeddedness ratings. When possible, the same observer for all reaches determined embeddedness visually using guidance from Platts et al. (1983). We did not assign embeddedness ratings to substrate classified as fines (sand and silt, ≤ 2 mm). The average embeddedness rating was determined by calculating the mean embeddedness ratings for all substrates > 2 mm. We also determined the proportion of substrate classified as fines by adding the relative abundance of sand and silt together.

Fish Community Variable Data Collection

To obtain estimates of fish abundance, we closed each 100 m reach using blocknets with 0.635 cm mesh positioned at the top and bottom of each reach. Using a Smith and Root LR-24 electrofishing backpack unit, we completed at least a three-pass electrofishing effort, sampling in an upstream direction. We used similar voltage, pulse, and frequency on each sampling occasion within each reach (voltage = 150 – 250V pulsed DC, frequency = 40Hz, pulse width = 20%). After each pass, we placed fish into live wells several meters above the top of the reach. We identified each fish to species and measured their total length to the nearest millimeter. In addition to length, we collected wet weight (± 1.0 g) for Gila Chub. Using the “removal” function in the Fisheries Stock Analysis package in R (Ogle et al. 2021), we obtained abundance estimates for every fish species in each reach using the Carle and Strub (1978) method. Upon completing our measurements, we released all fish alive into the reaches where they originated.

High discharge and shifting substrate prevented us from closing systems at one reach in the NMDGF Permanent site and three reaches within the Hot Springs site, violating the assumption of a closed system for our abundance estimates. Hedger et al. (2013) suggested that

if prior information about capture probability does not exist for sites that violate the no immigration and/or no emigration assumption(s) of removal abundance estimates, one should use the Carle and Shrub (1978) method. The method produces plausible abundances and fewer outliers with the highest correlation between actual total capture and the estimated abundance when compared to other popular abundance estimation methods (Hedger et al. 2013).

Analysis

Determining Important Habitat Variables

We conducted all analyses using R 4.2.0 software. We used a random forest nonparametric regression framework (Liaw and Wiener 2002) to predict Gila Chub abundance per 100 m at the translocation sites using the data collected from the extant Gila Chub populations. The random forest algorithm, as described by Breiman (2001), has been used to examine habitat suitability, habitat preference, and abundance for other native fish species such as Hornyhead Chub *Nocomis biguttatus* (Hickerson and Walters 2019) and Roundtail Chub (Bottcher 2009; Walsworth and Budy 2015). Random forest models are a type of ensemble learning technique that creates multiple decision trees, each trained on a different bootstrapped dataset with a random subset of predictor variables (Breiman 2001; Biau and Scornet 2016). Aggregation of the predictions from all individual trees occurs by calculating the mean. Various sources have demonstrated that random forest models can produce more accurate (Forkuor et al. 2017) and less accurate (Smith et al. 2013) predictions than multilinear regression techniques, but bagging (bootstrap-aggregation) allows random forest models to be relatively accurate when dealing with small sample sizes and datasets where the number of predictor variables outweighs

the number of observations (Biau and Scornet 2016), which were both major considerations for our dataset.

Using the “rf.modelSel” function in the “rfUtilities” package (Liaw and Wiener 2002), which utilizes methodologies from Murphy et al. (2010), we retained ten variables in our top model. We then tuned the parameters of the model (number of trees and splits tried at each node) to lower the Out-Of-Bag Error Rate and maximize the amount of variation in the response variable explained by the predictor variables. Next, we calculated variable importance for all variables based on a mean decrease in accuracy by testing how poorly the prediction would perform if the data for that predictor was randomly permuted, and others were left unaltered (Prasad et al. 2006). The closer the variable importance value was to 1.0, the more important the variable was in predicting Gila Chub abundance. Next, we calculated the predicted Gila Chub abundance per 100 m using the “predict” function in the “randomForest” package (Liaw and Wiener 2002) for all reaches in each of the suitable translocation sites.

Detecting Differences Between Sites

We used a one-way analysis of variance (ANOVA; Ott and Longnecker 2016) to determine if detectable differences existed between the mean predicted Gila Chub abundances per 100 m across translocation sites. We defined the level of significance as $\alpha \leq 0.1$. We deployed the outlier strategy by defining outliers as data points with studentized residuals larger than two (Ramsey and Schafer 2002). Based on observational and statistical considerations, we removed the Box 4 reach because we determined that this reach was not representative of the Box site, and we reported the analyses with and without this outlier. We then used Fisher’s Least

Squared Difference (Fisher's LSD; Fisher 1937) post-hoc analysis ($\alpha \leq 0.1$) to examine relationships between the specific translocation sites to rank the locations.

RESULTS

Initial Suitability Determination

One of the four potential translocation sites did not meet the minimum biological prerequisite constraints for initial suitability (Table 1). The Luna site did not meet the prerequisite biological constraint requirements for spawning temperature duration. The longest duration the stream temperature at the Luna site remained between the spawning temperature range of 15 and 26°C was 7.3 days (27 July 2022 – 3 August 2022). Thus, the Luna site was deemed unsuitable for a Gila Chub translocation, and we excluded the Luna site from further investigation. Though not exclusionary, nonnative fishes were present at both the Hot Springs site and the Box site. We captured Flathead Catfish, Channel Catfish, Common Carp, and/or Red Shiner in all but one of the Hot Springs site study reaches and Rainbow Trout were captured in all Box site study reaches. Our findings were included as a cautionary note in our translocation recommendation, as the presence of nonnative fishes at these potential translocation sites, may be an impediment to Gila Chub establishment.

Model Variables and Gila Chub Abundance Predictions

Model Variables

The ten variables retained in our top random forest model were (in order from most important to least important) proportion of mesohabitat classified as pools, discharge, Speckled

Dace abundance, proportion of substrate classified as fines, median stream temperature, dominant substrate type, conductivity, elevation, hardness, and the proportion of mesohabitat classified as riffles (Table 2). The proportion of mesohabitat classified as pools (1.00) was the most important variable in our top model, followed by discharge (0.44). All other variables retained in the top model had importance values less than 0.28 (Table 2). The percent of variance explained by our model was 58.71 % and the mean squared error was 515.97.

Table 1. Gila Chub *Gila intermedia* translocation sites in the Upper San Francisco River, New Mexico, including the Hot Springs, New Mexico Department of Game and Fish Permanent Monitoring (NMDGF Permanent), the Box, and the Luna sites initial suitability measurements. Biological prerequisites of maximum lethal temperature ($\geq 37^{\circ}\text{C}$; Carveth et al. 2006; USFWS 2015), spawning temperature duration (15 – 26°C for ≥ 60 consecutive days; USFWS 2015), and minimum dissolved oxygen concentration (≥ 4 mg/L; USFWS 2015) are displayed. Nonnative fish presence or absence is presented as a possible impediment to native fish establishment.

Translocation Site	Maximum Temperature (°C)	Longest Spawning Temperature Duration (days)	Dissolved Oxygen (mg/L)	Nonnative Fish Presence
Hot Springs	26.2	82.5	6.9	Present
NMDGF Permanent	26.2	82.5	8.2	Absent
The Box	24.2	89.9	9.8	Present
Luna	27.5	7.3*	7.2	Present

* denotes initial suitability requirements that were not met

Table 2. Importance of variables in the random forest model with Gila Chub *Gila intermedia* abundance per 100 m as the response variable. Bolded variables were retained in the top model.

Variable Name	Importance
Pool habitat (%)	1.0000
Discharge (m³/s)	0.4379
Speckled Dace abundance	0.2731
Fines (%)	0.2637
Median temperature (°C)	0.2238
Dominant substrate type (silt, sand, gravel, cobble, boulder, bedrock)	0.1994
Conductivity (µS/cm)	0.1555
Elevation(m)	0.1515
Hardness (mg/L as total CaCO₃)	0.1368
Riffle habitat (%)	0.1168
Average substrate embeddedness	0.1059
Average wetted width (m)	0.0679
Visual clarity (cm)	0.0596
Instant temperature (°C)	0.0523
Total nonnative fish abundance	0.0516
pH	0.0414
Aquatic vegetation presence (%)	0.0370
Minimum temperature (°C)	0.0344
Average pool residual depth (m)	0.0339
Average canopy cover (%)	0.0223
Average bank angle (°)	0.0217
Longfin Dace abundance	0.0208
Dissolved oxygen (mg/L)	0.0091
Shannon Diversity Index for macroinvertebrates (H')	0.0069
Desert Sucker abundance	0.0051
Western mosquitofish abundance	0.0033
Refuge flow (%)	0.0027
Total native fish abundance (excludes Gila Chub)	0.0006
Gila Topminnow abundance	0
Loach Minnow abundance	0
Fathead Minnow abundance	0
Fathead Minnow abundance	0
Common Carp abundance	0
Yellow Bullhead abundance	-4.6425 ×10 ⁻⁵
Large woody debris	-9.0721 ×10 ⁻⁴
Red Shiner abundance	-0.0037
Sonora Sucker abundance	-0.0055
Fast flow (%)	-0.0131

Partial dependency plots for each of the ten retained variables in our top model display the marginal effect and relationship between the individual predictor variables and the response variable, Gila Chub abundance per 100 m (Figure 4), while accounting for the average effect of the other model variables. The values of the response variable, Gila Chub Abundance Partial Dependence, vary due to each predictor variable's presumed importance to Gila Chub abundance. The general relationship between the proportion of mesohabitat classified as pools and Gila Chub abundance was positive with the number of Gila Chub increasing as the pool proportion increased above 40% (Figure 4A). We observed a similar pattern with discharge. Gila Chub abundance increased slightly with increasing discharge and then markedly increased when discharge measured above $0.63 \text{ m}^3/\text{s}$ (Figure 4B). Speckled Dace abundance and Gila Chub abundance had a negative relationship (Figure 4C), but as the proportion of substrate classified as fines increased, so did the Gila Chub abundance (Figure 4D). Median stream temperatures between 12 and 16°C differed little in Gila Chub abundance. However, once the median temperature increased above 16°C , the relationship was positive (Figure 4E). Additionally, Gila Chub abundance was greater if the dominant substrate type was silt compared to sand, gravel, and cobble substrate types (Figure 4F). In general, as conductivity increased, so did the Gila Chub abundance (Figure 4G). However, the relationship between elevation and Gila Chub abundance was negative (Figure 4H). Gila Chub abundance remained relatively stable as hardness increased $60 - 150 \text{ mg/L}$ as total CaCO_3 , but abundance increased as hardness increased above 150 mg/L as total CaCO_3 (Figure 4I). Finally, as the proportion of mesohabitat classified as riffles increased from 0% to 5% , Gila Chub abundance decreased before plateauing at higher riffle proportions (Figure 4J).

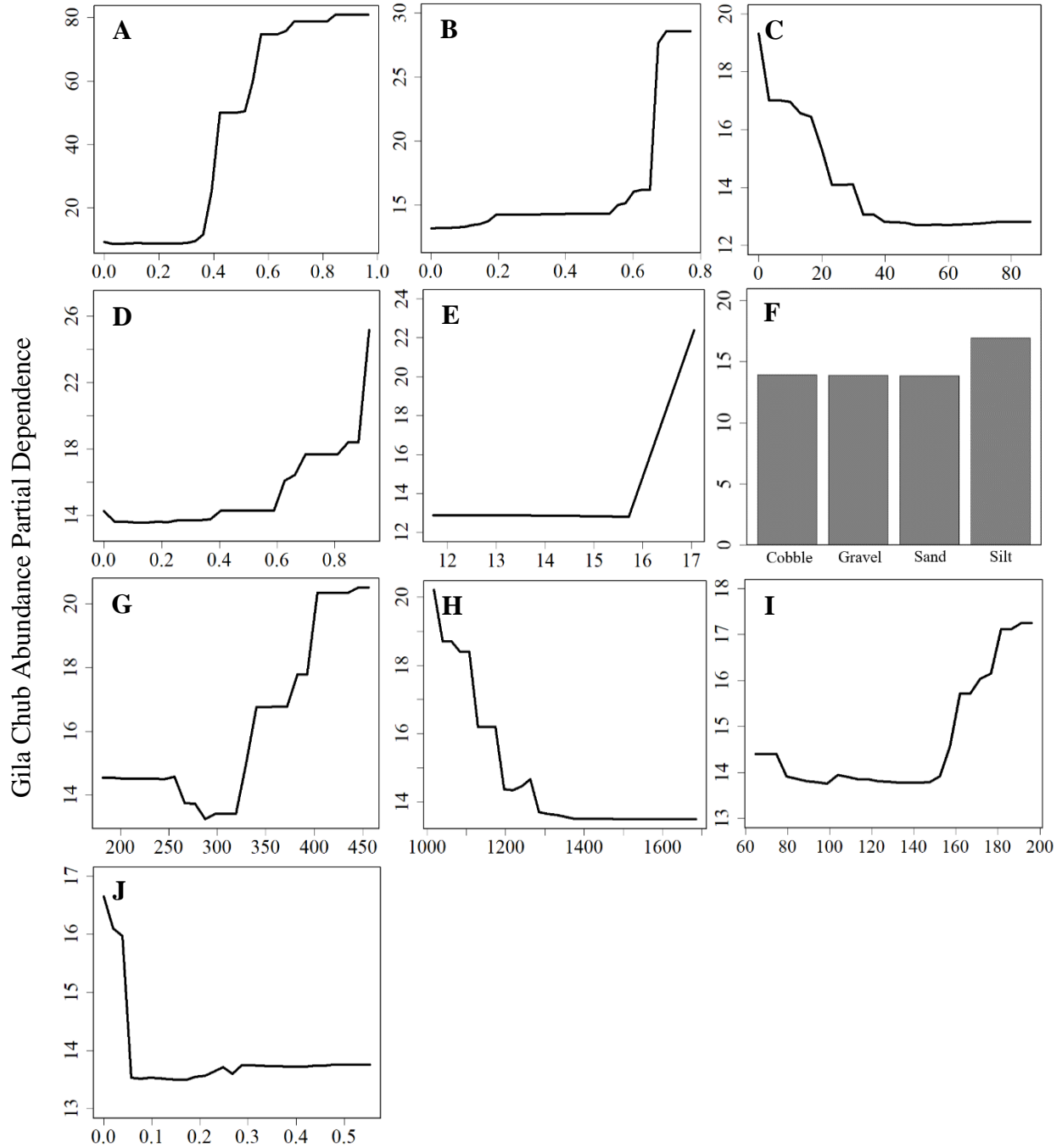


Figure 4. Gila Chub *Gila intermedia* abundance per 100 m partial dependency for the ten variables retained in the top random forest model: (A) proportion of mesohabitat classified as pools, (B) discharge (m^3/s), (C) Speckled Dace *Rhinichthys osculus* abundance, (D) proportion of substrate classified as fines (≤ 2 mm), (E) median stream temperature ($^{\circ}\text{C}$), (F) dominant substrate type, (G) conductivity ($\mu\text{S}/\text{cm}$), (H) elevation (m), (I) hardness (mg/L as total CaCO_3), and (J) proportion of mesohabitat classified as riffles.

Translocation Site Predictions

The average predicted Gila Chub abundance per 100 m for the NMDGF Permanent site and the Box site was 26.4 (standard error, 1.6) and 18.6 (7.0), respectively (Table 3). After deploying the outlier strategy, the average predicted abundance declined for the Box site to 11.6 (0.8; Table 3). The average predicted abundance of Gila Chub at the Hot Springs site was 29.0 (3.5; Table 3). Using a one-way ANOVA, we compared the mean predicted Gila Chub abundance of the Hot Springs site, NMDGF Permanent site, and the Box site with all data points, including the outlier. We found no detectable differences among the translocation sites ($F_{2, 12} = 1.10, p = 0.37$). After deploying the outlier strategy, we found that predicted Gila Chub abundance for the Box 4 reach (53.5 fish per 100 m) was an outlier as its studentized residual was 6.3 and, thus, larger than 2. This reach occurred within a section of river where the gradient changed dramatically. Waterfalls and deep plunge pools spanned nearly the entire reach. Because of this, the proportion of mesohabitat classified as pools was nearly double that of the other surveyed Box site reaches (42.6%). Since the proportion of mesohabitat classified as pools is the most influential variable in our top random forest model, the model's prediction for Gila Chub abundance per 100 m at Box 4 was nearly four times larger than the site's next highest predicted abundance for a reach. With this outlier removed, we detected differences between the mean predicted Gila Chub abundances among the potential translocation sites ($F_{2, 11} = 12.16, p = 0.0016$). Using post-hoc analysis (Fisher 1937), we detected differences in mean predicted Gila Chub abundance between the Box site 11.6 (0.8) and the Hot Springs site 29.0 (3.5; $p = 0.0006$), and between the NMDGF Permanent site 26.4 (1.6) and the Box site ($p = 0.0066$; Figure 5). We did not detect differences in predicted Gila Chub abundance between the Hot Springs site and the NMDGF Permanent site ($p = 0.55$; Figure 5).

Table 3. Site name includes both translocation and extant sites. Average predicted (translocation sites) and estimated (extant sites) Gila Chub *Gila intermedia* abundance per 100 m, standard error, and range for the translocation and extant sites. Data for translocation sites include the Hot Springs, New Mexico Department of Game and Fish Permanent Monitoring (NMDGF Permanent), and the Box sites, including and excluding the outlier reach (Box 4). Data for extant sites include Bonita Creek, Dix Creek, Harden Ciénega Creek, Mule Creek, and Eagle Creek. Sites are ordered from the greatest estimated or predicted abundance per 100 m to the least.

Site Name	Average Gila Chub Abundance (Standard Error, Range)
Bonita Creek, Arizona	82.3 (34.2, 33 – 178)
Hot Springs, New Mexico↓*	29.0 (3.5, 14.2 – 37.7)
NMDGF Permanent, New Mexico↓*	26.4 (1.6, 24.8 – 29.7)
The Box, New Mexico (with outlier)↓*	18.6 (7.0, 9.9 – 53.5)
Dix Creek, Arizona↓	15.1 (12.1, 0 – 87)
The Box, New Mexico (without outlier)↓*	11.6 (0.8, 9.9 – 14.0)
Harden Ciénega Creek, Arizona↓	8.4 (3.9, 0 – 27)
Mule Creek, New Mexico↓	1.3 (0.5, 0 – 4)
Eagle Creek, Arizona	0.1 (0.1, 0 – 1)

↓ indicates sites in the San Francisco River drainage

* indicates translocation sites

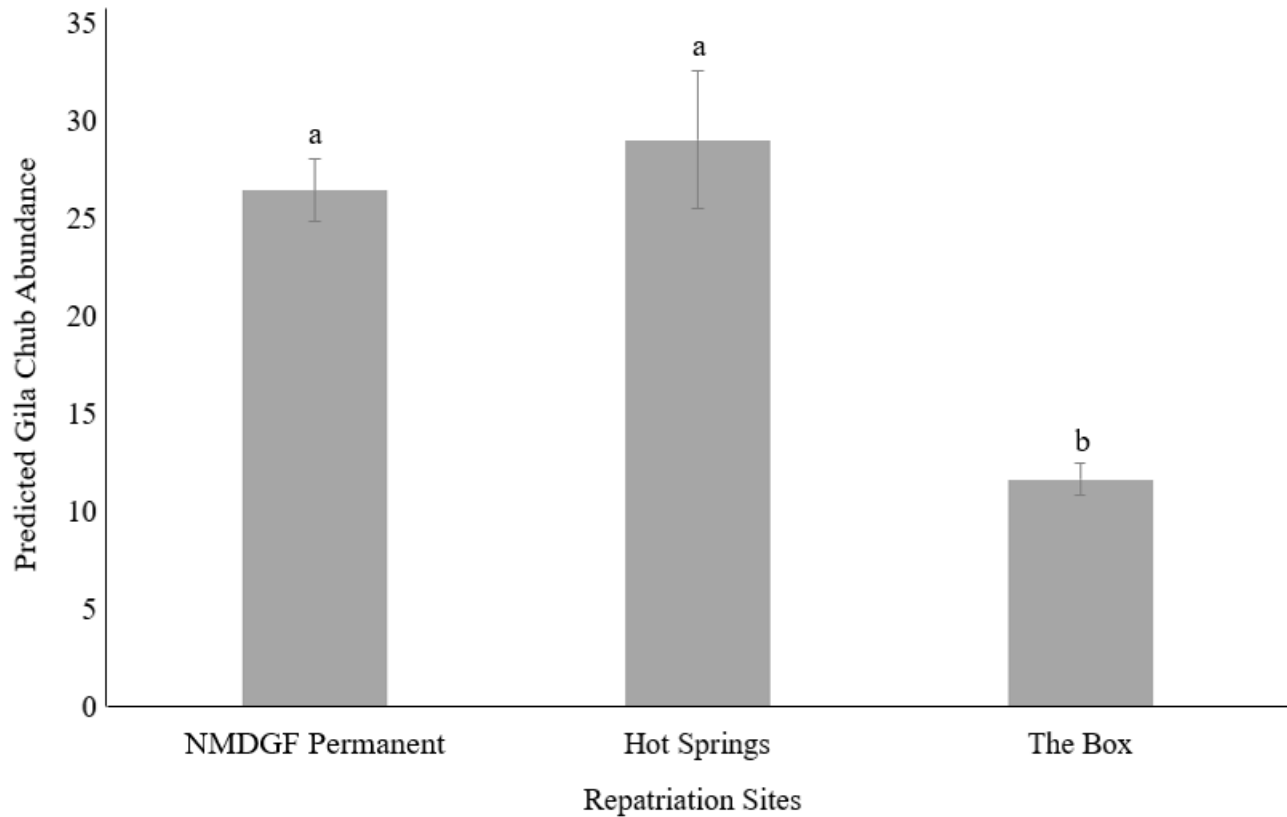


Figure 5. Mean Gila Chub *Gila intermedia* predicted abundance per 100 m by potential translocation sites with standard error bars for the Hot Springs, New Mexico Department of Game and Fish Permanent Monitoring (NMDGF Permanent), and the Box sites with the single outlier removed. Sites with the same mean separation letter do not significantly differ from one another ($\alpha \leq 0.1$, Fisher's LSD).

Average abundance estimates for the extant sites was greatest in Bonita Creek, Arizona, which had an average estimated abundance per 100 m of 82.3 (34.2, Table 3). The extant sites within the San Francisco River drainage, Mule Creek, New Mexico, Harden Ciénega Creek, Arizona, and Dix Creek, Arizona, had average estimated abundances per 100 m of 1.3 (0.5), 8.4 (3.9), and 15.1 (12.1), respectfully (Table 3). The translocation sites had predicted abundances that were generally similar to those of the extant sites within the San Francisco River drainage, with all translocation sites having lower average predicted Gila Chub abundance than the average estimated Gila Chub abundance at Bonita Creek, Arizona (Table 3). However, all potential translocation sites had greater predicted Gila Chub abundance per 100 m, with the Box 4 outlier removed, than average abundance estimates of Harden Ciénega Creek, Arizona, Mule Creek, New Mexico, and Eagle Creek, Arizona (Table 3).

DISCUSSION

Characterization of Habitat Variables of Extant and Potential Translocation Sites

The selection of a translocation site can be the most complicated step in a reintroduction effort (Minckley 1995). In this investigation, the absence of the target species from the potential translocation sites for nearly 80 years (Koster, field notes, unpublished) added an additional level of complexity. We overcame this initial hurdle by utilizing the expert opinions and observations of species authorities. We characterized the habitat attributes of sites with extant populations and used the attributes to inform a model that predicted the magnitude of suitability of unoccupied potential translocation sites. This investigation identifies important ecological considerations and may allow for a systematic review of other translocation locations for Gila Chub across its historical range.

Initial Suitability Determination

Identifying suitable locations for a target species and possible impediments to establishment is critical to ensure a successful translocation attempt (Lamothe and Drake 2019; Hickerson et al. 2021b). Moreover, complete habitat characterization of translocation sites can open doors to remediation and restoration opportunities (Lamothe and Drake 2019), as is the case with the Hot Springs and Box sites, where the elimination of nonnative fishes may create translocation sites more suitable for Gila Chub. We found that an initial suitability determination using prerequisite biological variables is not only insightful, but critical for successfully ascertaining information about translocation success. Random forest models that are trained on data collected from extant populations are limited. They cannot be used to understand potential translocation sites that are completely unsuitable for the target species and the Luna site is an excellent example of this. Had we not used an initial suitability determination for the Luna site, the random forest model would have predicted Gila Chub abundances for the reaches at the site, despite stream temperatures measuring too cold to support spawning. Considering a location where the target species is unlikely to have reproductive success could be viewed as an inefficient use of time and resources. Understanding the limits of our model allowed us to make informed translocation recommendations.

Most nonnative species found in the translocation sites were never captured in the extant sites. Thus, the limited number of nonnative abundance estimates at the extant sites prohibited the random forest model from accurately establishing relationships between Gila Chub abundance and nonnative fish species' abundances. Though the USFWS (2015) acknowledges the need to control and/or eradicate nonnative fishes from Gila Chub occupied habitat and potential translocation sites, the precise and quantitative questions surrounding acceptable

abundances is largely unknown. For example, Rainbow Trout and warmwater nonnative fishes are present in portions of Turkey Creek, New Mexico, a Gila Chub occupied stream (USFWS 2015). Despite the presence of these nonnative fishes, the Gila Chub population is considered large and stable (USFWS 2015). However, in Bonita Creek, Arizona, Gila Chub were absent from reaches surveyed by the USBLM (2021) downstream of the Reclamation barrier containing only nonnative fish species, namely Western Mosquitofish, Common Carp, Green Sunfish and Yellow Bullhead. Gila Chub were found in all other reaches upstream of the Reclamation barrier, some of which contained relatively low numbers of nonnative fishes compared to those captured in the reach downstream of the barrier (USBLM 2021). While we were unable to quantify the relationships between nonnative fish species, their abundance, and the effect on Gila Chub abundance in our investigation, we recognize the importance in acknowledging this factor when identifying translocation locations for Gila Chub.

Important Model Variables and Gila Chub Abundance

While the initial suitability determination and the identification of impediments to Gila Chub establishment allowed us to work within the limits of our predictive model, the top random forest model provided an understanding of the level of suitability across potential translocation sites. Additionally, the ten variables retained in the top model provided some insight into the ecology of Gila Chub. The high importance of the proportion of mesohabitat classified as pools was unsurprising because the literature reflects Gila Chub's affinity for pool environments (Barber and Minckley 1966; Griffith and Tiersch 1989).

Our second most important variable of discharge is not as well studied. Our model shows that Gila Chub abundance and discharge are positively related. Since discharge is a product of both velocity and area, deeper stream reaches have higher discharge compared to shallower

reaches with the same velocity measurements. Gila Chub evolved in flashy flood-prone systems where discharge is highly variable, while many nonnative predators and competitors did not (Miller 1961). Elevated discharge decreases both the density and abundance of piscivorous nonnative fishes in desert streams (Propst and Gido 2004) favoring native fish (Moran et al. 2018a). USGS (2016) recorded varied discharge of the Upper San Francisco River, near Glenwood, New Mexico between $0.12 \text{ m}^3/\text{s}$ (1956) and $150.36 \text{ m}^3/\text{s}$ (2013). At this location, Propst et al. (2008) found that native fish densities were greater in years of higher discharge. In the nearby Verde River, Arizona, Ruhí et al. (2014) found that Roundtail Chub abundance was positively associated with high discharge and flood conditions. High discharge resulting in the decrease in piscivorous nonnative fish and more available habitat provides a possible explanation for the discharge-Gila Chub relationship.

Another important consideration to the presumed importance of discharge to Gila Chub abundance is the limits that our model poses on the values of the variable. Since this model is trained on data from extant sites, predicted abundance will be limited by bounds placed on the upper and lower limits of our training (extant site) data. The greatest discharge measurement we obtained from a Gila Chub extant site was $0.77 \text{ m}^3/\text{s}$. The greatest discharge measurement obtained from the Gila Chub translocation sites was $1.44 \text{ m}^3/\text{s}$. We speculate the relationship between Gila Chub abundance and discharge is nonlinear resulting in a negative association with discharge when discharge measurements surpass an unknown threshold. At discharge measurements above this threshold, Gila Chub may not be able to maintain position in the stream. This phenomenon has been demonstrated under laboratory conditions for Roundtail Chub (Moran et al. 2018a). Due to the limits of the training data obtained from the extant sites, we were unable to observe this relationship for Gila Chub.

While the presence of nonnative fishes is understood to negatively impact Gila Chub populations (Bestgen and Propst 1989; Dudley and Matter 2000; Olden and Poff 2005), we found that Gila Chub abundance was negatively related with the abundance of Speckled Dace, a native cyprinid. Competition for food sources seems an unlikely explanation for this relationship as Speckled Dace are bottom-dwelling insectivores and Gila Chub are generalists that feed throughout the water column. Despite there being no comparative study between Gila Chub and Speckled Dace diets, Schrieber and Minckley (1981) compared stomach contents of Roundtail Chub and Speckled Dace from Aravaipa Creek, Arizona and found little overlap. A more likely explanation is that Speckled Dace and Gila Chub do not utilize the same mesohabitat types and are, thus, not commonly found together. Rinne (1992) and Zaines et al. (2019) found that Speckled Dace were positively associated with riffle and run habitat. In our investigation, the partial dependency for Gila Chub abundance demonstrated a negative relationship between both Speckled Dace abundance and the proportion of mesohabitat classified as riffles, while the proportion of mesohabitat classified as pools was positively related and overwhelmingly important to increased predicted Gila Chub abundance.

Gila Chub prefer pool habitat associated with fine sediment, particularly in large deep pools (Lisle and Hilton 1992). Our investigation demonstrated a positive relationship between predicted abundance of Gila Chub and the proportion of fine sediment. We also observed increases in predicted abundance of Gila Chub when the dominant substrate type was silt compared to sand, gravel, and cobble. Although no previous study exists relating Gila Chub abundance to substrate, Bottcher (2009) found that Roundtail Chub preferred habitats with relatively fine substrate. Increases in sedimentation, sometimes referred to as siltation, is commonly associated with a decrease in fish abundance, presumably due to its negative impact

on macroinvertebrate communities (Ramezani et al. 2014). However, the declining trends described by Ramezani et al. (2014) and others, typically refer to more specialized feeding guilds and salmonids (Kemp et al. 2011). Berkman and Rabeni (1987) demonstrated that fish with generalist feeding strategies undergo negligible change in abundance when fine sediments increased. Gila Chub are dietary generalists, feeding on fish, aquatic macroinvertebrates, terrestrial insects, terrestrial plants, algae, and diatoms (Griffith and Tiersch 1989). There is also evidence terrestrial insects make up a large portion of Gila Chubs' diet (Griffith and Tiersch 1989). This is true of other members of the *Gila* genus in the Lower Colorado River basin, notably Humpback Chub *Gila cypha*, especially during times of high turbidity which are associated with sedimentation events (Behn and Baxter 2019). Fine sediments are an important part of southwestern riverine systems. Clay, silt, and sand provide food, cover (turbidity), and nutrients to lotic systems (Kemp et al. 2011; Kondolf et al. 2014). In the Lower Colorado River basin, decreases in fine sediment have negatively impacted native fish communities. A notable example is the construction of the Glenn Canyon Dam on the Colorado River and the resulting decrease in sedimentation downstream of the dam (Schmidt et al. 1998). The decrease in sedimentation creates low turbidity waters which favor piscivorous nonnative fishes with better visual acuity (Moran et al. 2018b).

Another relationship that may be explained by Gila Chub's adaptation to turbid and flood-prone systems is the positive relationship we observed between Gila Chub abundance and conductivity. Although we could not locate published studies that investigated the specific relationship between conductivity and Gila Chub, Stephani et al. (2015) investigated the effect of total suspended solids on Yaqui Chub *Gila purpurea* and found the lethal threshold for total suspended solids was much higher than that of salmonids. The same authors suggested that this

was due to the fish's adaptation to naturally turbid waters. Total suspended solids and conductivity exhibit a colinear relationship (Tan et al. 2017). Increases in total suspended solids and, hence, conductivity and turbidity, reduced predation risk in other *Gila* spp. in the Lower Colorado River basin (Ward et al. 2016). Water hardness was another water quality variable retained in the top model. As hardness increased, abundance of Gila Chub increased. The divalent cations calcium (Ca^{2+}) and magnesium (Mg^{2+}) are vital to water hardening of newly fertilized eggs and skeletal development of larval fish (Wurts and Durborow 1992; Swain et al. 2020). For freshwater fish, elevated hardness in their environment can reduce the loss of vital monovalent sodium (Na^+) and potassium (K^+) ions from the fish's blood (Wurts and Durborow 1992) and thereby ameliorate stressful events.

Stream temperature represents an important environmental variable necessary for successful Gila Chub reproduction because the primary cue for spawning initiation is water temperature. Schultz and Bonar (2016) found that spawning events for Gila Chub will occur in stream temperatures that range from 15 to 26°C, but optimum spawning temperatures occur between 18 and 24°C. Median stream temperature was among the variables retained in our top random forest model. We observed a positive relationship between Gila Chub abundance and median stream temperatures above 16°C. Thus, as the median temperature approached the optimum spawning temperature described by Schultz and Bonar (2016), the Gila Chub abundance increased, revealing a possible increase in reproductive success associated with optimal spawning temperatures.

Although an elevational limit for Gila Chub has not been established, the USFWS (2015) service states that Gila Chub are found in suitable locations in the Gila River basin with elevations between 609 and 1,676 m. Our study also suggests that there may be an elevation limit

to Gila Chub distribution we observed a negative relationship between elevation and Gila Chub abundance, with minimum Gila Chub abundance occurring at approximately 1300 m. Research describes similar patterns in other native fishes (Wilson and Belk 2001). For example, in an investigation of habitat variables on the abundance of Leatherside Chub *Lepidomeda copei*, Wilson and Belk (2001) observed that elevation was negatively associated with Leatherside Chub occurrence. The authors noted that Leatherside Chub did not occur at elevations greater than 2195 m.

Translocation Site Selection and Management Recommendations

The extant populations have persisted through time, indicating that their populations are robust enough to withstand stochastic events such as wildfires and floods. Except for the extant Bonita Creek site in Arizona, the translocation sites, Hot Springs, NMDGF Permanent, and the Box (including the outlier) have exhibited greater average predicted Gila Chub abundances than all average Gila Chub abundance estimates of the extant sites. With the outlier removed, the Box site had a greater average predicted abundance than the Harden Ciénega Creek, Arizona, Mule Creek, New Mexico, and the Eagle Creek, Arizona extant sites. Abundance estimates are vital for assessing species population status and describing population sizes (Stewart et al. 2017). Small population size is a risk criterion understood to increase fish species' likelihood of local extirpation or extinction (Williams et al. 1989). Thus, establishing whether the Upper San Francisco River translocation sites would be able to support robust Gila Chub populations more resilient to threats was important in our work. Based on qualitative comparisons between the predicted Gila Chub abundances of the translocation sites and the average estimated abundances of the extant sites, we concluded that the predicted abundance estimates calculated for the Gila Chub translocation sites are likely great enough to persist through time.

The NMDGF Permanent site provides a potential translocation site that meets the minimum prerequisite biological constraints, was devoid of nonnative fish species, and may represent the most suitable site for Gila Chub compared to the Hot Springs and the Box sites. Our investigation suggests that a combination of water quality, biological, and hydrological parameters best predict the success of a potential translocation site for Gila Chub. Our research supports previous studies that emphasized the importance of pool habitat (Barber and Minckley 1966; Griffith and Tiersch 1989). While our variable importance corroborated the value of plentiful pool habitat for Gila Chub, our investigation also suggests that managers consider a more robust characterization to select translocation locations. For example, the Box site had the highest mean pool proportion of any translocation site with and without the outlier removed. However, we found that the average predicted Gila Chub abundance per 100 m was the lowest of any of the three final potential translocation sites. The Box site's high elevation, low median temperature, and low discharge ultimately reflected the site less suitable for Gila Chub than translocation locations further downstream. Moreover, abundance estimates for nonnative Rainbow Trout were 4 – 28 per 100 m (excludes outlier reach) and 4 – 38 per 100 m (includes outlier reach), indicating possible impediments to Gila Chub establishment. The Hot Springs site was determined by our post-hoc analysis of the potential translocation sites to be as suitable for Gila Chub as the NMDGF Permanent site based on Gila Chub predicted abundance. However, the presence of nonnative Flathead Catfish, Channel Catfish, Red Shiner, and Common Carp at the Hot Springs site was an important consideration in our cautionary recommendation.

Although nonnative fishes are present in both the Hot Springs and the Box sites, these locations represent opportunities for remediation to increase suitable recovery locations for Gila Chub. Successful translocation of native desert fishes following remediation efforts to remove

nonnative fishes in the San Francisco River basin have occurred. Between 2012 and 2019, in conjuncture with mechanical removal of nonnative fishes, native Roundtail Chub, Loach Minnow, and Spikedace were translocated to the Blue River, Arizona, a major tributary of the San Francisco River (Hickerson et al. 2021a). Fish assemblage changed dramatically over the course of the project from predominantly nonnative fishes to almost exclusively native species (Hickerson et al. 2021a). Hickerson et al. (2021a) stresses the challenges in achieving these results in a large river system like the Blue River, Arizona, but demonstrates that successful translocation is possible in systems with nonnative species with remediation to remove nonnative fishes. Pool et al. (2013) also emphasizes the benefits of including locations with nonnative fishes present when considering recovery opportunities for Gila Chub and other native species. Both the Hot Springs site and the Box site represent secondary opportunities for potential translocation sites for Gila Chub, particularly if remediation to remove or control nonnative fishes was conducted.

The methods used in this investigation are directly applicable to the translocations of other species of conservation need. For example, as of 2003, Desert Pupfish were reintroduced to more than twelve locations, but only two (16.7%) of these translocated populations were successful in establishing the species (DFT 2003). Similarly, the Desert Fishes Team (2003) stated that of 175 Gila Topminnow translocated populations, approximately two dozen (13.7%) persisted. These Desert Pupfish and Gila Topminnow translocations reflect overall trends of varying success across freshwater fish translocation events (Cochran-Biederman et al. 2015). While there are numerous reasons why translocation attempts fail, we demonstrated that it is possible to evaluate potential translocation sites in the absence of the target species, and thereby lessen the uncertainty of translocating extirpated populations. We accomplished this by utilizing

expert opinion, identifying biological prerequisites, and systematically characterizing extant and targeted translocation sites. While the threat of local extirpation is an unfortunate reality for many endemic Lower Colorado River basin fishes (Fagan et al. 2002), successful translocations are possible (Hickerson et al. 2021a) and can be used to promote the resiliency, redundancy, and continuance of endemic desert fishes.

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APPENDIX A

SUMMARY DATA FOR GILA CHUB EXTANT SITES

Appendix Table A. 1. Summary habitat data for Bonita Creek, Arizona, collected in May and October 2021. NA denotes measurements not applicable to a reach.

Suitability Variable	Upper Site 1	The Gallery	Lee Trail	Midnight Canyon
Elevation (m)	1018	1020	1064	1150
LWD	8	8	7	5
Minimum Temperature (°C)	5.2	5.2	5.2	5.2
Maximum Temperature (°C)	30.6	30.6	30.6	30.6
Median Temperature (°C)	17.1	17.1	17.1	17.1
Instantaneous Temperature (°C)	21.9	21.6	19.8	23.3
Dissolved Oxygen (mg/L)	6.3	6.3	7.6	9.0
Visual Clarity (cm)	50	50	30	34
Hardness (mg/L as total CaCO ₃)	186	196	194	170
Alkalinity (mg/L as total CaCO ₃)	201	214	215	189
Conductivity (µS/cm)	433	456	443	356
pH	7.2	7.3	7.8	7.4
Average Wetted Width (m)	6.6	5.2	9.6	2.6
Pool Proportion (%)	40.2	31.5	96.7	11.4
Average Pool Residual Depth (m)	0.6	0.2	1.4	0.2
Riffle Proportion (%)	0	0	0	40
Run Proportion (%)	55.6	50.5	0	52
Cascade Proportion (%)	4.3	0	3.3	0
Average Bank Angle (°)	100.9	150.9	109.5	115.7
Undercut Proportion (%)	30	0	10	30
Average Undercut Depth (m)	0.46	NA	0.74	0.13
Discharge (m ³ /s)	0.77	0.62	0.72	0.09
Proportion of Refuge Flow (% of velocities <0.1 m/s)	72	50	76.7	47.6
Proportion of Fast Flow (% of velocities >0.4 m/s)	8	30	23.3	31
Canopy Cover (%)	47.0	63.2	24.9	59.5
Aquatic Vegetation Cover (%)	10	10	10	42
Shannon-Weaver Diversity Index (H)	2.1	1.9	1.6	2.3
Simpson's Diversity Index (D)	0.8	0.8	0.7	0.9
Dominate Substrate Type	Silt	Silt	Silt	Gravel
Proportion of Fine Substrate (%)	74	88	92	44
Average Embeddedness (1–5)	2.3	1.8	1.8	3.9

Appendix Table A. 2. Abundance estimates and 95% confidence intervals in parentheses for the four Bonita Creek, Arizona study reaches, collected in October 2021. NA denotes reaches where we did not capture a particular fish species. * denotes fish species where reasonable confidence intervals could not be obtained due to either 1) incidental capture of a single fish of a species or 2) not achieving depletion for the species. Species observed in Bonita Creek, Arizona include Speckled Dace *Rhinichthys osculus*, Gila Topminnow *Poecilopsis occidentalis*, Loach Minnow *Rhinichthys cobitis*, Sonora Sucker *Catostomus insignis*, Desert Sucker *Pantosteus clarkii*, Gila Chub *Gila intermedia*, Fathead Minnow *Pimephales promelas*, Western Mosquitofish *Gambusia affinis*, and Yellow Bullhead *Ameiurus natalis*.

Study Reach	Speckled Dace	Gila Topminnow	Loach Minnow	Sonora Sucker	Desert Sucker	Gila Chub	Fathead Minnow	Western Mosquitofish	Yellow Bullhead
Upper Site 1	NA	NA	NA	5 (3, 7)	NA	85 (69, 101)	9 (9, 9)	6 (5, 7)	1(*)
Gallery	NA	NA	NA	6 (5, 7)	NA	33 (29, 37)	NA	25 (3, 47)	1(*)
Lee Trail	NA	24 (13, 35)	NA	83 (79, 87)	26 (24, 28)	178 (137, 218)	NA	NA	NA
Midnight Canyon	6 (*)	NA	1(*)	21 (20, 22)	44 (39, 49)	33 (26, 40)	NA	NA	NA

Appendix Table A. 3. Standardized taxa and counts for the Bonita Creek, Arizona macroinvertebrate samples, collected in May 2021.

Study Reach	Orders and Other Names	Family	Counts	
Upper Site 1	Araneae- spiders		1	
	Coleoptera-larval beetles	Elmidae	1	
		Haliplidae	1	
		Hydrophilidae	3	
	Diptera- true flies	Ceratopogonidae	1	
		Chironomidae	39	
		Simuliidae	1	
	Ephemeroptera-mayflies	Caenidae	4	
		Leptophlebiidae	1	
		Siphonuridae	15	
	Odonata- dragonflies and damselflies	Coenagrionidae	14	
		Gomphidae	1	
		Lestidae	3	
		Libellulidae	4	
		Leptophlebiidae	1	
		Planorbidae	9	
		Sphaeriidae-freshwater mussels	1	
	Gallery	Coleoptera-larval beetles	Ptilodactylidae	1
		Diptera- true flies	Chironomidae	36
Ephemeroptera-mayflies		Siphonuridae	11	
Hemiptera- true bugs		Belostomatidae	3	
		Vellidae	12	
Odonata- dragonflies and damselflies		Coenagrionidae	7	
Basommatophora-snails		Planorbidae	19	
Sphaeriidae-freshwater mussels			11	
Lee Trail		Diptera- true flies	Chironomidae	53
		Ephemeroptera-mayflies	Boetidae	10
	Hemiptera- true bugs	Belostomatidae	5	
		Veliidae	2	
	Odonata- dragonflies and damselflies	Coenagrionidae	15	
		Corduliidae	6	
	Plecoptera- stoneflies		2	
	Pulmonata-snails		7	
	Midnight Canyon	Diptera- true flies	Chironomidae	2
		Ephemeroptera-mayflies	Baetidae	2
		Leptohephidae	19	
		Siphonuridae	11	
Hemiptera- true bugs		Belostomatidae	2	
		Veliidae	17	
Odonata- dragonflies and damselflies		Calopterygidae	2	
		Coenagrionidae	2	
Basommatophora-snails		Planorbidae	8	
Trichoptera-caddisflies		Hydropsychidae	11	
	Leptoceridae	15		
	Philoptamidae	8		

Appendix Table A. 4. Summary habitat data for Eagle Creek, Arizona, collected in November 2021. NA denotes measurements not applicable to a reach.

Suitability Variable	Eagle Creek 1	Eagle Creek 2	Eagle Creek 3	Eagle Creek 4	Eagle Creek 5	Eagle Creek 6	Eagle Creek 7	Eagle Creek 8
Elevation (m)	1653	1655	1657	1665	1666	1666	1683	1685
LWD	4	9	6	6	5	6	8	3
Minimum Temperature (°C)	7.3	7.3	7.3	7.3	7.3	7.3	7.3	7.3
Maximum Temperature (°C)	23.2	23.2	23.2	23.2	23.2	23.2	23.2	23.2
Median Temperature (°C)	15.5	15.5	15.5	15.5	15.5	15.5	15.5	15.5
Instantaneous Temperature (°C)	17.9	11.8	17.1	12.8	12.9	20.1	15.3	19.8
Dissolved Oxygen (mg/L)	6.1	8.6	7.6	7.9	8.3	7.2	7.4	7.2
Visual Clarity (cm)	60	60	60	60	60	60	60	60
Hardness (mg/L as total CaCO ₃)	138	134	133	139	133	129	127	128
Alkalinity (mg/L as total CaCO ₃)	135	135	134	139	134	132	129	127
Conductivity (µS/cm)	301	294	290	292	290	284	285	282
pH	8.2	8.4	8.4	8	8.1	8.4	8.1	8.3
Average Wetted Width (m)	6.3	4.6	5.9	5.2	5.8	5.6	7.8	6.4
Pool Proportion (%)	26.5	15.6	15.9	21.6	8	23.8	21.9	13.9
Average Pool Residual Depth (m)	0.3	0.5	0.2	0.3	0.2	0.1	0.7	0.4
Riffle Proportion (%)	45.6	49	52.5	35.3	55.4	40.5	18.7	2.4
Run Proportion (%)	27.9	34.4	29.6	43.1	35.8	24.9	59.4	83.7
Cascade Proportion (%)	0	1	2	0	0.8	10.8	0	0
Average Bank Angle (°)	126.3	145.6	116	134.4	133.8	141.3	143.7	137.8
Undercut Proportion (%)	30	0	20	0	10	20	0	10
Average Undercut Depth (m)	0.17	NA	0.16	NA	0.07	0.05	NA	0.17
Discharge (m ³ /s)	0.14	0.35	0.18	0.21	0.26	0.1	0.12	0.12
Proportion of Refuge Flow (% of velocities <0.1 m/s)	54.3	25	28.2	22.5	36.6	47.6	61.9	40.9
Proportion of Fast Flow (% of velocities >0.4 m/s)	22.9	25	5.1	30	29.3	11.9	4.8	6.8
Canopy Cover (%)	48.1	60.5	67	62.7	36.8	64.3	42.7	50.8
Aquatic Vegetation Cover (%)	12	0	4	2	6	0	6	2
Shannon-Weaver Diversity Index (H)	1.3	2.5	1.9	1.9	1.2	0.7	1.4	1.5
Simpson's Diversity Index (D)	0.6	0.9	0.8	0.8	0.6	0.4	0.6	0.6
Dominate Substrate Type	Gravel	Gravel	Cobble	Cobble	Cobble	Cobble	Sand	Sand
Proportion of Fine Substrate (%)	23	22	19	23	30	27	36	46
Average Embeddedness (1–5)	3.9	3.6	3.6	3.9	3.8	3.3	3.6	3.6

Appendix Table A. 5. Abundance estimates and 95% confidence intervals in parentheses for the eight Eagle Creek, Arizona study reaches, collected in November 2021. NA denotes reaches where we did not capture a particular fish species. * denotes fish species where reasonable confidence intervals could not be obtained due to the incidental capture of a single fish of a species. Species observed in Eagle Creek, Arizona include Speckled Dace *Rhinichthys osculus*, Longfin Dace *Agosia chrysogaster*, Sonora Sucker *Catostomus insignis*, Desert Sucker *Pantosteus clarkii*, and Gila Chub *Gila intermedia*.

Study Reach	Speckled Dace	Longfin Dace	Sonora Sucker	Desert Sucker	Gila Chub
1	60 (49, 71)	4 (1, 7)	6 (6, 6)	76 (67, 85)	NA
2	69 (57, 81)	34 (18, 50)	11 (11, 11)	194 (173, 215)	NA
3	53 (44, 62)	9 (3, 15)	2 (1, 3)	127 (80, 173)	NA
4	61 (59, 63)	8 (6, 10)	18 (11, 25)	157 (143, 171)	NA
5	86 (53, 119)	67 (46, 88)	1 (*)	169 (157, 181)	NA
6	50 (32, 68)	12 (10, 14)	5 (3, 7)	123 (49, 197)	NA
7	60 (42, 78)	NA	6 (6, 6)	70 (53, 86)	NA
8	5 (5, 5)	NA	10 (4, 16)	24 (20, 28)	1 (*)

Appendix Table A. 6. Standardized taxa and counts for the Eagle Creek, Arizona macroinvertebrate samples, collected in November 2021.

Study Reach	Order	Family	Counts	
Eagle Creek 1	Coleoptera-larval beetles	Carabidae	2	
		Diptera- true flies	56	
	Ephemeroptera-mayflies	Dixidae	5	
		Simuliidae	18	
	Hemiptera-true bugs	Siphonuridae	16	
		Veliidae	1	
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	1	
		Odonata- dragonflies and damselflies	Coenagrionidae	1
	Eagle Creek 2	Coleoptera-larval beetles	Dytiscidae	4
			Elmidae	4
Diptera- true flies		Certopogonidae	3	
		Chironomidae	28	
		Dixidae	5	
Ephemeroptera-mayflies		Simuliidae	11	
		Baetidae	4	
		Siphonuridae	7	
Hemiptera-true bugs		Gerridae	4	
Hymenoptera- sawflies, wasps, bees and ants		Cephidae	3	
Megaloptera-alder flies, Dobson flies, and fish flies		Corydalidae	3	
		Odonata- dragonflies and damselflies	Coenagrionidae	4
Trichoptera-caddisflies		Branchycentridae	4	
		Hydropsychidae	3	
		Limnephilidae	3	
	Chironomidae	29		
	Dixidae	3		
Eagle Creek 3	Diptera- true flies	Simuliidae	6	
		Tipulidae	1	
		Baetidae	8	
	Ephemeroptera-mayflies	Siphonuridae	22	
		Corixidae	1	
		Gerridae	3	
	Hemiptera-true bugs	Veliidae	23	
		Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	3
		Trichoptera-caddisflies	Hydroptilidae	1
	Eagle Creek 4	Diptera- true flies	Chironomidae	36
Dixidae			4	
Simuliidae			34	
Ephemeroptera-mayflies		Tabanidae	2	
		Baetidae	4	
		Siphonuridae	5	
Hemiptera-true bugs		Veliidae	5	
		Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	4
Trichoptera-caddisflies		Hydropsychidae	3	
		Hydroptilidae	3	
		Chironomidae	45	
Eagle Creek 5		Diptera- true flies	Dixidae	1
	Simuliidae		42	

Appendix Table A. 6-continued. Standardized taxa and counts for the Eagle Creek, Arizona macroinvertebrate samples, collected in November 2021.

Study Reach	Order	Family	Counts
		Tabanidae	1
	Ephemeroptera-mayflies	Caenidae	2
		Siphonuridae	5
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	2
Eagle Creek 6	Diptera- true flies	Chironomidae	72
	Hemiptera-true bugs	Veliidae	2
	Diptera-true flies	Dixidae	1
		Simuliidae	26
	Ephemeroptera-mayflies	Siphonuridae	1
Eagle Creek 7	Diptera- true flies	Chironomidae	62
		Simuliidae	7
		Tipulidae	3
	Ephemeroptera-mayflies	Baetidae	7
		Siphonuridae	10
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	4
	Odonata- dragonflies and damselflies	Calopterygidae	2
		Coenagrionidae	1
	Trichoptera-caddisflies	Hydropsychidae	4
Eagle Creek 8	Coleoptera-larval beetles	Hydrophilidae	1
	Diptera- true flies	Athericidae	2
		Chironomidae	56
		Tipulidae	3
	Ephemeroptera-mayflies	Siphonuridae	20
	Hemiptera-true bugs	Belostomatidae	1
		Corixidae	6
	Odonata- dragonflies and damselflies	Coenagrionidae	3
	Plecoptera-stoneflies	Leuctridae	3
	Trichoptera-caddisflies	Hydropsychidae	5

Appendix Table A. 7. Summary habitat data for Harden Ciénega Creek, Arizona, collected in May and June 2022. NA denotes measurements not applicable to a reach.

Suitability Variable	Backup 1	Harden Ciénega 1	Harden Ciénega 2	Harden Ciénega 3	Harden Ciénega 4	Harden Ciénega 5	Harden Ciénega 6
Elevation (m)	1221	1230	1230	1248	1254	1272	1306
LWD	1	17	13	2	6	10	4
Minimum Temperature (°C)	7.3	7.3	7.3	7.3	7.3	7.3	7.3
Maximum Temperature (°C)	27.9	27.9	27.9	27.9	27.9	27.9	27.9
Median Temperature (°C)	17.0	17.0	17.0	17.0	17.0	17.0	17.0
Instantaneous Temperature (°C)	16.5	21.3	16.4	21.3	18.3	21.5	21.5
Dissolved Oxygen (mg/L)	7.8	7.9	7.9	7.3	7.2	7.3	7.3
Visual Clarity (cm)	46	29	60	60	54	44	60
Hardness (mg/L as total CaCO ₃)	101	100	102.5	109	103	112	118
Alkalinity (mg/L as total CaCO ₃)	85	111	113	110	112	118	111
Conductivity (µS/cm)	261	258	261	254	260	253	254
pH	8.3	8.5	8.1	8.2	8.3	8.1	8.2
Average Wetted Width (m)	2.7	2.1	3.0	2.8	3.3	2.1	3.1
Pool Proportion (%)	5.2	0	6.1	7.9	17.4	26	21.7
Average Pool Residual Depth (m)	0.2	NA	0.1	0.2	0.2	0.3	0.3
Riffle Proportion (%)	15.1	17.4	36.9	24.7	28.9	48.2	31.4
Run Proportion (%)	74.9	76.5	57	61.7	50.7	24	38.6
Cascade Proportion (%)	4.8	6.1	0	5.7	3	1.8	9.0
Average Bank Angle (°)	120.5	145.1	113.7	148.2	152.9	131.5	127.1
Undercut Proportion (%)	20	0	40	10	0	10	20
Average Undercut Depth (m)	0.13	NA	0.14	0.04	NA	0.04	0.11
Discharge (m ³ /s)	0.01	0.01	0.01	0.02	0.02	0.02	0.44
Proportion of Refuge Flow (% of velocities <0.1 m/s)	82.9	56.8	55	66.7	70.2	48.5	65.9
Proportion of Fast Flow (% of velocities >0.4 m/s)	5.7	0	5	0	6.4	3	6.8
Canopy Cover (%)	52.4	79.5	91.9	89.2	77.8	71.9	64.3
Aquatic Vegetation Cover (%)	12	2	24	56	36	14	18
Shannon-Weaver Diversity Index (H)	2.1	2	1.7	2.3	2.1	1.5	1.5
Simpson's Diversity Index (D)	0.9	0.8	0.7	0.9	0.8	0.7	0.7
Dominate Substrate Type	Gravel	Gravel	Gravel	Gravel	Gravel	Gravel	Gravel
Proportion of Fine Substrate (%)	33	25	28	27	24	22	13
Average Embeddedness (1–5)	3.6	3.3	3.7	3.4	3.6	3.6	3.9

Appendix Table A. 8. Abundance estimates and 95% confidence intervals in parentheses for the seven Harden Ciénega Creek, Arizona study reaches, collected in May and June 2022. NA denotes reaches where we did not capture a particular fish species. * denotes fish species where reasonable confidence intervals could not be obtained due to the incidental capture of a single fish of a species. Species observed in Harden Ciénega Creek, Arizona include Speckled Dace *Rhinichthys osculus*, Longfin Dace *Agosia chrysogaster*, Sonora Sucker *Catostomus insignis*, Desert Sucker *Pantosteus clarkii*, Gila Chub *Gila intermedia*, and Red Shiner *Cyprinella lutrensis*.

Study Reach	Speckled Dace	Longfin Dace	Sonora Sucker	Desert Sucker	Gila Chub	Red Shiner
Backup 1	66 (61, 71)	91 (65, 117)	3 (3,3)	32 (31, 33)	NA	14 (12, 16)
1	86 (62, 110)	116 (94, 138)	NA	85 (71, 99)	NA	NA
2	23 (17, 29)	39 (36, 42)	NA	15 (9, 20)	2 (0, 4)	26 (25, 27)
3	17 (15, 19)	8 (6, 10)	2 (2, 2)	17 (10, 24)	18 (11, 25)	6 (5, 7)
4	32 (27, 37)	5 (4, 6)	8 (6, 10)	32 (29, 35)	27 (25, 29)	NA
5	16 (14, 18)	4 (4, 4)	2 (0, 4)	23 (5, 41)	7 (4, 10)	NA
6	17 (13, 21)	1 (*)	3 (2, 4)	38 (26, 50)	5 (4, 6)	NA

Appendix Table A. 9. Standardized taxa and counts for the Harden Ciénega Creek, Arizona macroinvertebrate samples, collected in May and June 2022.

Study Reach	Order	Family	Counts	
Backup 1	Amphipoda-scuds or side swimmers	Gammaridae	16	
	Coleoptera-larval beetles	Elmidae	4	
	Diptera- true flies	Chironomidae	7	
	Ephemeroptera-mayflies	Caebidae	4	
		Siphonuridae	22	
	Hemiptera-true bugs	Veliidae	10	
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	4	
	Odonata- dragonflies and damselflies	Gomphidae	4	
		Coenagrionidae	22	
	Trichoptera-caddisflies	Hydropsychidae	7	
	Harden Ciénega 1	Coleoptera- larval beetles	Psephenidae	8
		Diptera-true flies	Chironomidae	6
			Tabnidae	8
Haplotaxida-worms		Lumbriculidae	35	
Megaloptera-alder flies, Dobson flies, and fish flies		Corydalidae	8	
Odonata-dragonflies and damselflies		Coenagrionidae	8	
		Gomphidae	13	
Trichoptera-caddisflies		Hydropsychidae	6	
Harden Ciénega 2		Basommatophora-snails	Planorbidae	6
		Coleoptera-larval beetles	Dytiscidae	4
	Diptera-true flies	Chironomidae	6	
		Tabmidae	4	
	Ephemeroptera-mayflies	Siphonuridae	6	
	Hemiptera-true bugs	Veliidae	18	
	Haplotaxida -worms	Lumbriculidae	48	
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	2	
	Odonata	Coenagrionidae	6	
	Harden Ciénega 3	Amphipoda-scuds or side swimmers	Gammaridae	16
Basommatophora-snails		Planorbidae	7	
Coleoptera-larval beetles		Elmidae	10	
Diptera- true flies		Chironomidae	5	
		Simuliidae	4	
		Tipulidae	1	
Ephemeroptera-mayflies		Siphonuridae	14	
Hemiptera-true bugs		Veliidae	4	
Odonata- dragonflies and damselflies		Calopterygidae	8	
		Coenagrionidae	14	
		Gomphidae	1	
Trichoptera-caddisflies		Hydropsychidae	16	
Harden Ciénega 4		Amphipoda-scuds or side swimmers	Gammaridae	8
	Basommatophora-snails	Planorbidae	29	
	Coleoptera-larval beetles	Dytiscidae	2	
		Elmidae	4	
	Diptera- true flies	Chironomidae	7	

Appendix Table A. 9-continued. Standardized taxa and counts for the Harden Ciénega Creek, Arizona macroinvertebrate samples, collected in May and June 2022.

Study Reach	Order	Family	Counts
Harden Ciénega 5		Stratiomyidae	2
	Hemiptera-true bugs	Notonectidae	2
		Veliidae	7
	Haplotaaxida -worms	Lumbriculidae	5
	Odonata- dragonflies and damselflies	Gomphidae	2
		Coenagrionidae	20
	Trichoptera-caddisflies	Hydropsychidae	12
	Coleoptera-larval beetles	Elmidae	10
	Hemiptera-true bugs	Gerridae	10
		Veliidae	21
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	3
Odonata- dragonflies and damselflies	Calopterygidae	10	
	Coenagrionidae	46	
Harden Ciénega 6	Basommatophora-snails	Planorbidae	10
Coleoptera-larval beetles	Elmidae	6	
Diptera- true flies	Chironomidae	32	
Emphemeroptera-mayflies	Siphonuridae	18	
Hemiptera	Veliidae	1	
Trichoptera-caddisflies	Hydropsychidae	33	

Appendix Table A. 10. Summary habitat data for Dix Creek, Arizona, collected in October 2021. NA denotes measurements not applicable to a reach.

Suitability Variable	Dix 1	Dix 2	Dix 3	Dix 4	Dix 5	Dix 6	Dix 7
Elevation (m)	1215	1216	1242	1270	1281	1302	1362
LWD	7	10	18	18	14	18	11
Minimum Temperature (°C)	4.1	4.1	4.1	4.1	4.1	4.1	4.1
Maximum Temperature (°C)	31.3	31.3	31.3	31.3	31.3	31.3	31.3
Median Temperature (°C)	14.7	14.7	14.7	14.7	14.7	14.7	14.7
Instantaneous Temperature (°C)	20.5	15.7	17.9	16	19.2	19.6	19.6
Dissolved Oxygen (mg/L)	7.6	7.7	8.3	6.3	6.5	6.9	6.9
Visual Clarity (cm)	60	60	60	60	60	60	60
Hardness (mg/L as total CaCO₃)	154	91	86	65	65	65	65
Alkalinity (mg/L as total CaCO₃)	169	86	75	60	60	60	60
Conductivity (µS/cm)	220	248	182	272	284	237	237
pH	7.4	8.2	8.2	8.4	8.4	8.4	8.4
Average Wetted Width (m)	4.7	3	2.8	2.5	2.8	1.8	2.8
Pool Proportion (%)	8.8	34.2	17.9	34.3	21	38.7	70.2
Average Pool Residual Depth (m)	0.3	0.3	0.3	0.2	0.1	0.1	0.3
Riffle Proportion (%)	7.8	23.3	5.3	12.8	26.7	4.8	4.2
Run Proportion (%)	83.4	42.5	76.5	49.6	47	35.1	19.6
Cascade Proportion (%)	0	0	0	3.3	5.3	21.4	6
Average Bank Angle (°)	152.8	151.2	162.4	152.9	150.3	141.4	128.2
Undercut Proportion (%)	0	0	0	0	0	10	0
Average Undercut Depth (m)	NA	NA	NA	NA	NA	0.03	NA
Discharge (m³/s)	0.02	0.11	0.03	0.07	0.07	0.05	0.14
Proportion of Refuge Flow (% of velocities <0.1 m/s)	70.4	45.2	60	27.6	56.3	58.1	16.7
Proportion of Fast Flow (% of velocities >0.4 m/s)	7.4	9.5	8.6	51.7	0	6.5	83.3
Canopy Cover (%)	94.6	94.6	88.1	95	91.9	98.4	91.4
Aquatic Vegetation Cover (%)	2	0	6	14	28	0	2
Shannon-Weaver Diversity Index (H)	1.2	1.1	0.7	1.7	2	1.4	1.7
Simpson's Diversity Index (D)	0.5	0.7	0.5	0.8	0.8	0.6	0.8
Dominate Substrate Type	Cobble	Cobble	Cobble	Cobble	Cobble	Cobble	Cobble
Proportion of Fine Substrate (%)	14	5	16	16	16	0	26
Average Embeddedness (1–5)	3.3	3.9	3.5	3.2	3.6	3.7	3.7

Appendix Table A. 11. Abundance estimates and 95% confidence intervals in parentheses for the seven Dix Creek, Arizona study reaches, collected in October 2021. NA denotes reaches where we did not capture a particular fish species. * denotes fish species where reasonable confidence intervals could not be obtained due to either 1) incidental capture of a single fish of a species or 2) not achieving depletion for the species. Species observed in Dix Creek, Arizona include Speckled Dace *Rhinichthys osculus*, Longfin Dace *Agosia chrysogaster*, Sonora Sucker *Catostomus insignis*, Desert Sucker *Pantosteus clarkii*, and Gila Chub *Gila intermedia*.

Study Reach	Speckled Dace	Longfin Dace	Sonora Sucker	Desert Sucker	Gila Chub
1	85 (71, 99)	46 (44, 48)	NA	10 (10, 10)	NA
2	55 (52, 58)	15 (14, 16)	2 (2, 2)	6 (4, 8)	1(*)
3	34 (30, 38)	34 (31, 37)	NA	17 (15, 19)	2 (2, 2)
4	28 (25, 31)	20 (16, 24)	NA	NA	12 (11, 13)
5	26 (22, 30)	1(*)	NA	NA	NA
6	76 (71, 81)	NA	NA	NA	4 (3, 5)
7	19 (*)	10 (9, 11)	3 (3, 3)	11 (9, 13)	87 (81, 93)

Appendix Table A. 12. Standardized taxa and counts for the Dix Creek, Arizona macroinvertebrate samples, collected in October 2021.

Study Reach	Order	Family	Counts
Dix 1	Diptera-true flies	Chironomidae	69
		Simuliidae	8
		Tipulidae	4
	Ephemeroptera-mayflies	Caenidae	11
		Ameletidae	4
Dix 2	Odonata- dragonflies and damselflies	Coenagrionidae	4
	Diptera-true flies	Tipulidae	33
	Isopoda- crustaceans	Armadiillidiidae	34
Dix 3	Odonata- dragonflies and damselflies	Coenagrionidae	33
	Diptera-true flies	Chironomidae	65
Dix 4	Ephemeroptera-mayflies	Simuliidae	34
		Caenidae	1
	Diptera-true flies	Ceratopogonidae	3
		Chironomidae	9
		Simuliidae	8
	Ephemeroptera-mayflies	Ameletidae	9
		Caenidae	9
Dix 5	Hemiptera-true bugs	Veliidae	27
	Odonata- dragonflies and damselflies	Coenagrionidae	35
		Diptera-true flies	Ceratopogonidae
	Ephemeroptera-mayflies	Chironomidae	15
		Simuliidae	17
		Tipulidae	1
		Ameletidae	12
		Caenidae	1
	Odonata- dragonflies and damselflies	Siphonuridae	28
		Calopterygidae	8
		Coenagrionidae	14
Corduliidae		1	
Hydropsychidae		1	
Dix 6	Trichoptera-caddisflies	Philopotamidae	1
		Planorbidae	2
	Basommatophora-snails	Dytiscidae	7
	Coleoptera-larval beetles	Chironomidae	53
		Diptera-true flies	Ameletidae
	Ephemeroptera-mayflies	Caenidae	2
		Hemiptera-true bugs	Gerridae
Odonata- dragonflies and damselflies	Coenagrionidae	2	
	Corduliidae	2	
	Elmidae	19	
Dix 7	Coleoptera-larval beetles	Chironomidae	29
		Simuliidae	2
	Diptera-true flies	Caenidae	6
		Ephemeroptera-mayflies	Corydalidae
	Megaloptera-alder flies, Dobson flies, and fish flies	Calopterygidae	2
		Coenagrionidae	29
	Odonata- dragonflies and damselflies	Hydropsychidae	7
		Trichoptera-caddisflies	

Appendix Table A. 13. Summary habitat data for Mule Creek, New Mexico, collected in May 2022. NA denotes measurements not applicable to a reach.

Suitability Variable	Mule 1	Mule 2	Mule 3	Mule 4	Mule 5	Mule 6	Mule 7	Mule 8
Elevation (m)	1291	1350	1351	1352	1371	1372	1373	1373
LWD	1	0	1	5	4	9	3	11
Minimum Temperature (°C)	7.3	7.3	7.3	7.3	7.3	7.3	7.3	7.3
Maximum Temperature (°C)	27.9	27.9	27.9	27.9	27.9	27.9	27.9	27.9
Median Temperature (°C)	11.7	11.7	11.7	11.7	11.7	11.7	11.7	11.7
Instantaneous Temperature (°C)	12.4	16.5	20.2	12.8	19.4	19.4	17.1	11.8
Dissolved Oxygen (mg/L)	8.3	8.2	7.4	7.7	7.4	7.4	7.7	8.3
Visual Clarity (cm)	60	60	60	60	60	60	60	60
Hardness (mg/L as total CaCO ₃)	106	110	102	105	103	108	99	99
Alkalinity (mg/L as total CaCO ₃)	120	116	116	119	121	118	113	117
Conductivity (µS/cm)	305	298	297	302	298	298	293	296
pH	8.5	8.5	8.6	8.6	8.5	8.2	8.5	8.4
Average Wetted Width (m)	4.9	3.9	3.2	5.1	4	4.8	3.5	6.4
Pool Proportion (%)	3.6	8.3	4.8	24.9	13.5	33.67	2.7	7.9
Average Pool Residual Depth (m)	0.1	0.2	0.2	0.3	0.3	0.1	0.1	0.2
Riffle Proportion (%)	5.9	10.6	7.8	10.7	23.7	4.5	28.4	0
Run Proportion (%)	90.5	81.1	87.4	64.4	60.7	61.9	68.9	92.1
Cascade Proportion (%)	0	0	0	0	2.1	0	0	0
Average Bank Angle (°)	146.7	148.2	147.4	156.3	122.3	162.3	153.8	122.4
Undercut Proportion (%)	10	0	0	0	20	0	0	30
Average Undercut Depth (m)	0.06	NA	NA	NA	0.13	NA	NA	0.12
Discharge (m ³ /s)	0.01	0.01	2.0 *10 ⁻³	0.04	0.02	0.02	0.02	0.06
Proportion of Refuge Flow (% of velocities <0.1 m/s)	59.4	63.9	62.2	73.8	81	73.3	82.1	51.3
Proportion of Fast Flow (% of velocities >0.4 m/s)	3.1	13.9	0	4.8	0	0	2.6	0
Canopy Cover (%)	71.4	62.2	83.8	88.1	89.2	77.3	97.3	76.3
Aquatic Vegetation Cover (%)	4	18	0	0	6	4	0	0
Shannon-Weaver Diversity Index (H)	1.1	2	2.1	2.6	2.3	2	2	2.2
Simpson's Diversity Index (D)	0.5	0.8	0.8	0.9	0.8	0.8	0.7	0.8
Dominate Substrate Type	Gravel	Gravel	Gravel	Gravel	Gravel	Gravel	Gravel	Gravel
Proportion of Fine Substrate (%)	25	20	17	25	18	12	11	17
Average Embeddedness (1-5)	4.1	4.3	4.3	4	4	4.3	4.3	4.4

Appendix Table A. 14. Abundance estimates and 95% confidence intervals in parentheses for the eight Mule Creek, New Mexico study reaches, collected in May 2022. NA denotes reaches where we did not capture a particular fish species. * denotes fish species where reasonable confidence intervals could not be obtained due to either 1) incidental capture of a single fish of a species or 2) not achieving depletion for the species. Species observed in Mule Creek, New Mexico include Speckled Dace *Rhinichthys osculus*, Longfin Dace *Agosia chrysogaster*, Sonora Sucker *Catostomus insignis*, Desert Sucker *Pantosteus clarkii*, Gila Chub *Gila intermedia*, and Common Carp *Cyprinus carpio*.

Study Reach	Speckled Dace	Longfin Dace	Sonora Sucker	Desert Sucker	Gila Chub	Common Carp
1	43 (40, 47)	32 (26, 38)	1 (0, 2)	101 (92, 110)	NA	NA
2	77 (66, 88)	27 (14, 40)	31 (38, 34)	151 (140, 162)	3 (1, 5)	NA
3	22 (20, 24)	17 (15, 19)	3 (2, 4)	71 (*)	NA	NA
4	30 (29, 31)	32 (15, 49)	71 (68, 74)	133 (126, 140)	1 (*)	2 (2, 2)
5	30 (23, 37)	18 (16, 20)	28 (27, 29)	108 (87, 129)	4 (3, 5)	NA
6	44 (29, 59)	18 (11, 25)	3 (2,4)	113 (108, 118)	NA	NA
7	33 (28, 38)	13 (13, 13)	NA	102 (91, 113)	NA	NA
8	26 (25, 27)	13 (11, 15)	22 (20, 24)	50 (45, 55)	2 (0, 4)	NA

Appendix Table A. 15. Standardized taxa and counts for the Mule Creek, New Mexico macroinvertebrate samples, collected in May 2022.

Study Reach	Order	Family	Counts	
Mule 1	Diptera-true flies	Tipulidae	73	
	Emphemeroptera-mayflies	Caenidae	3	
	Hemiptera-true bugs	Veliidae	5	
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	8	
	Odonata-dragonflies and damselflies	Coenagrionidae	1	
		Gomphidae	2	
Mule 2	Plecoptera- stoneflies	Perlodidae	5	
	Basommatophora-snails	Planorbidae	2	
	Coleoptera-larval beetles	Elmidae	3	
		Diptera- true flies	Chironomidae	16
	Emphemeroptera-mayflies	Simuliidae	2	
		Tipulidae	42	
		Caenidae	3	
		Siphionyridae	10	
	Haplotaxida-worms	Lumbriculidae	3	
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	5	
		Odonata	Calopterygidae	1
Mule 3	Basommatophora-snails	Coenagrionidae	3	
		Gomphidae	4	
	Coleoptera-larval beetles	Planorbidae	1	
		Dytiscidae	1	
	Diptera- true flies	Elmidae	4	
		Chironomidae	39	
		Empididae	1	
		Perloidae	1	
		Simuliidae	4	
		Stratiomyidae	18	
		Tabanidae	1	
	Emphemeroptera-mayflies	Tipulidae	3	
		Caenidae	12	
		Siphonuridae	4	
		Hemiptera-true bugs	Belostomatidae	1
		Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	2
	Odonata- dragonflies and damselflies		Calopterygidae	2
Trichoptera-caddisflies	Coenagrionidae	1		
	Hydropsychidae	3		
	Limnephilidae	1		
	Planorbidae	2		
Mule 4	Basommatophora-snails	Dytiscidae	6	
		Elmidae	5	
	Coleoptera-larval beetles	Haliplidae	2	
		Diptera- true flies	Calicidae	2
		Ceratopogonidae	2	
		Chironomidae	8	
		Muscidae	2	
		Pelecorhynchidae	30	
		Simuliidae	2	

Appendix Table A. 15-continued. Standardized taxa and counts for the Mule Creek, New Mexico macroinvertebrate samples, collected in May 2022.

Study Reach	Order	Family	
Ephemeroptera-mayflies	Stratiomyidae	8	
	Caenidae	2	
	Ephemerellidae	2	
Hemiptera-true bugs	Siphonuridae	11	
	Gerridae	2	
	Veliidae	2	
Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	2	
	Calopterygidae	2	
	Gomphidae	2	
Odonata- dragonflies and damselflies	Lestidae	2	
	Trichoptera-caddisflies	Helicopsychidae	2
	Limnephilidae	2	
Basommatophora-snails	Planorbidae	2	
Coleoptera-larval beetles	Dytiscidae	2	
	Elmidae	2	
	Diptera- true flies	Ceratopogonidae	3
Diptera- true flies	Simuliidae	7	
	Stratiomyidae	40	
	Tipulidae	3	
	Caenidae	3	
	Leptophlebiidae	1	
Ephemeroptera-mayflies	Siphonuridae	9	
	Belostomatidae	2	
	Gerridae	1	
Hemiptera-true bugs	Veliidae	1	
	Corydalidae	7	
	Calopterygidae	3	
Megaloptera-alder flies, Dobson flies, and fish flies	Coenagrionidae	2	
	Perlodidae	2	
	Plecoptera- stoneflies	Hydropsychidae	7
Trichoptera-caddisflies	Limnephilidae	1	
	Phlopteramidae	1	
	Polycentropodidae	1	
Basommatophora-snails	Planorbidae	2	
Coleoptera-larval beetles	Dytiscidae	7	
	Elmidae	6	
	Diptera- true flies	Chironomidae	47
Diptera- true flies	Dixidae	2	
	Simuliidae	6	
	Tipulidae	3	
	Caenidae	2	
	Siphonuridae	3	
Ephemeroptera-mayflies	Gerridae	2	
	Calopterygidae	2	
	Coenagrionidae	3	
Hemiptera-true bugs	Gomphidae	2	
	Odonata- dragonflies and damselflies	Helicopsychidae	3
	Hydropsychidae	2	
Trichoptera-caddisflies	Limnephilidae	8	
	Basommatophora-snails	Planorbidae	5
	Coleoptera-larval beetles	Dryopodidae	3

Appendix Table A. 15-continued. Standardized taxa and counts for the Mule Creek, New Mexico macroinvertebrate samples, collected in May 2022.

Study Reach	Order	Family
	Dytiscidae	3
	Elmidae	1
	Hydrophilidae	1
Diptera- true flies	Ceratopogonidae	1
	Chironomidae	9
	Dixidae	1
	Stratiomyidae	48
	Tipulidae	1
Emphemeroptera-mayflies	Caenidae	1
	Siphonuridae	3
Odonata- dragonflies and damselflies	Calopterygidae	3
	Coenagrionidae	1
	Gomphidae	1
Trichoptera-caddisflies	Helicopsychidae	5
	Hydropsychidae	3
	Limnephilidae	10
Basommatophora-snails	Planorbidae	3
Coleoptera-larval beetles	Dytiscidae	2
Diptera- true flies	Culicidae	2
	Chironomidae	30
	Dixidae	3
	Simuliidae	3
	Stratiomyidae	18
	Tabanidae	2
Emphemeroptera-mayflies	Caenidae	3
	Siphonuridae	18
Odonata- dragonflies and damselflies	Calopterygidae	3
	Coenagrionidae	3
	Gomphidae	2
Trichoptera-caddisflies	Hydropsychidae	2
	Limnephilidae	4

APPENDIX B

SUMMARY DATA FOR GILA CHUB TRANSLOCATION SITES

Appendix Table B. 1. Summary habitat data for the Hot Springs site, San Francisco River, New Mexico, collected in August and October 2022. NA denotes measurements not applicable to a reach.

Elevation (m)	Hot Springs 1	Hot Springs 2	Hot Springs 5	Hot Springs 6	Hot Springs 7	Hot Springs 8
Elevation (m)	1385	1389	1394	1401	1409	1415
LWD	10	6	7	20	2	2
Minimum Temperature (°C)	NA	NA	NA	NA	NA	NA
Maximum Temperature (°C)	NA	NA	NA	NA	NA	NA
Median Temperature (°C)	NA	NA	NA	NA	NA	NA
Instantaneous Temperature (°C)	20.2	24.7	21	24.8	23.7	19.4
Dissolved Oxygen (mg/L)	7.1	6.6	6.9	6.7	6.8	7.1
Visual Clarity (cm)	21	20	4	8	4	4
Hardness (mg/L as total CaCO₃)	123	125	117	115	132	128
Alkalinity (mg/L as total CaCO₃)	129	127	116	137	135	129
Conductivity (µS/cm)	491	489	398	311	312	299
pH	8.5	8.5	8.2	8.4	8.3	8.3
Average Wetted Width (m)	8.7	7.2	8.6	10.2	9.8	7.5
Pool Proportion (%)	16.8	0	0	0	11.2	16.9
Average Pool Residual Depth (m)	0.4	NA	NA	NA	0.9	0.4
Riffle Proportion (%)	1.7	0	3.9	63.4	13.1	24.1
Run Proportion (%)	81.5	100	96.1	36.6	75.7	59
Cascade Proportion (%)	0	0	0	0	0	0
Average Bank Angle (°)	117.7	100.8	98	122	122	143.1
Undercut Proportion (%)	30	40	40	30	30	10
Average Undercut Depth (m)	0.38	0.34	0.14	0.09	0.17	0.07
Discharge (m³/s)	1.25	1.25	1.20	0.67	0.80	0.68
Proportion of Refuge Flow (% of velocities <0.1 m/s)	0	4.4	6.7	13.3	16.7	19.2
Proportion of Fast Flow (% of velocities >0.4 m/s)	88.9	80	62.2	28.9	39.6	38.3
Canopy Cover (%)	18.4	64.3	31.9	43.8	41.1	67
Aquatic Vegetation Cover (%)	8	2	4	0	8	10
Shannon-Weaver Diversity Index (H)	1.5	1.4	1.4	2.2	2.1	1.9

Appendix Table B. 1-continued. Summary habitat data for the Hot Springs site, San Francisco River, New Mexico, collected in August and October 2022. NA denotes measurements not applicable to a reach.

	Hot Springs 1	Hot Springs 2	Hot Springs 5	Hot Springs 6	Hot Springs 7	Hot Springs 8
Simpson's Diversity Index (D)	0.7	0.8	0.7	0.8	0.8	0.8
Dominate Substrate Type	Sand	Gravel	Gravel	Sand	Sand	Gravel
Proportion of Fine Substrate (%)	66	45	34	39	60	36
Average Embeddedness (1–5)	3.1	3.8	3.3	3.4	3.6	3.5

Appendix Table B. 2. Abundance estimates and 95% confidence intervals in parentheses for the six Hot Springs site, San Francisco River, New Mexico study reaches, collected in August and October 2022. NA denotes reaches where we did not capture a particular fish species. * denotes fish species where reasonable confidence intervals could not be obtained due to either 1) incidental capture of a single fish of a species or 2) not achieving depletion for the species due to poor visibility and high stream flows. Species observed in the Hot Springs site include Channel Catfish *Ictalurus punctatus*, Flathead Catfish *Pylodictis olivaris*, Common Carp *Cyprinus carpio*, and Red Shiner *Cyprinella lutrensis*.

Study Reach	Channel Catfish	Flathead Catfish	Common Carp	Red Shiner
1	2 (1, 3)	2 (1, 3)	1(*)	NA
2	NA	NA	NA	NA
5	1 (*)	3 (2, 4)	2 (2, 2)	NA
6	1 (*)	NA	NA	NA
7	2 (*)	1 (*)	1 (*)	1 (*)
8	2 (*)	NA	15 (13, 17)	5 (*)

Appendix Table B. 3. Standardized taxa and counts for the Hot Springs site, San Francisco River, New Mexico macroinvertebrate samples, collected in August and October 2022.

Study Reach	Order	Family	Counts
Hot Springs 1	Amphipoda- scuds or side swimmers	Gammaridae	1
	Coleoptera-larval beetles	Cantharidae	1
		Elmidae	1
	Diptera-true flies	Chironomidae	6
		Simuliidae	44
	Emphemeroptera-mayflies	Heptageniidae	1
		Leptohyphidae	1
		Siphonuridae	6
	Hemiptera-true bugs	Belostomatidae	1
		Veliidae	1
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	1
	Odonata-dragonflies and damselflies	Calopterygidae	33
		Coenagrionidae	1
		Gomphidae	1
		Hydropsychidae	1
	Hot Springs 2	Ephemeroptera-mayflies	Leptohyphidae
Haplotaaxida-worms		Lumbriculidae	39
Hemiptera-true bugs		Belostomatidae	3
		Dytiscidae	3
		Veliidae	10
Hymenoptera- sawflies, wasps, bees and ants		Formicidae	3
Lepidoptera-moths		Erebidae	3
Megaloptera-alder flies, Dobson flies, and fish flies		Corydalidae	25
Odonata-dragonflies and damselflies		Calopterygidae	3
		Gomphidae	4
Hot Springs 5	Trichoptera-caddisflies	Hydropsychidae	4
	Coleoptera-larval beetles	Elmidae	5
	Diptera-true flies	Chironomidae	14
		Simuliidae	3
	Ephemeroptera-mayflies	Heptageniidae	1
		Leptohyphidae	37
		Siphonuridae	39
	Hemiptera-true bugs	Veliidae	1
Hot Springs 6	Amphipoda- scuds or side swimmers	Gammaridae	7
	Coleoptera-larval beetles	Carabidae	1
		Elmidae	4
	Diptera-true flies	Chironomidae	2
		Simuliidae	15
	Ephemeroptera-mayflies	Baetidae	4
		Leptohyphidae	32
	Haplotaaxida-worms	Lumbriculidae	9
	Hemiptera-true bugs	Belostomatidae	4
		Corixidae	1
		Veliidae	13
	Isopoda- crustaceans	Armadillidiidae	1
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	1
	Odonata-dragonflies and damselflies	Calopterygidae	3
		Gomphidae	2

Appendix Table B. 3-continued. Standardized taxa and counts for the Hot Springs site, San Francisco River, New Mexico macroinvertebrate samples, collected in August and October 2022.

Study Reach	Order	Family	Standardized Counts
Hot Springs 7	Trichoptera-caddisflies	Hydropsychidae	1
	Amphipoda- scuds or side swimmers	Gammaridae	4
	Coleoptera-larval beetles	Curculionidae	2
	Diptera-true flies	Baetidae	6
		Simuliidae	28
	Ephemeroptera-mayflies	Leptohyphidae	25
	Hemiptera-true bugs	Belostomatidae	2
		Notonectidae	2
		Veliidae	10
	Odonata-dragonflies and damselflies	Calopterygidae	11
Hot Springs 8	Trichoptera-caddisflies	Hydropsychidae	3
	Coleoptera-larval beetles	Chrysomelidae	1
		Elmidae	10
	Diptera-true flies	Gyrinidae	3
		Simuliidae	13
		Tabanidae	1
	Ephemeroptera-mayflies	Heptageniidae	2
		Leptohyphidae	43
		Siphonuridae	2
	Haplotaxida-worms	Lumbriculidae	10
	Hemiptera-true bugs	Belostomatidae	1
		Veliidae	10
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	1
Odonata-dragonflies and damselflies	Calopterygidae	3	

Appendix Table B. 4. Summary habitat data for the New Mexico Department of Game and Fish Permanent Monitoring (NMDGF Permanent) site, San Francisco River, New Mexico, collected in August and November 2022. NA denotes measurements not applicable to a reach.

Suitability Variable	NMDGF Permanent 2	NMDGF Permanent 3	NMDGF Permanent 8
Elevation (m)	1422	1429	1475
LWD	7	1	6
Minimum Temperature (°C)	6.5	6.5	6.5
Maximum Temperature (°C)	26.2	26.2	26.2
Median Temperature (°C)	16.7	16.7	16.7
Instantaneous Temperature (°C)	18.7	13	7.6
Dissolved Oxygen (mg/L)	7.3	8.2	9.3
Visual Clarity (cm)	7	60	36
Hardness (mg/L as total CaCO ₃)	101	120	143
Alkalinity (mg/L as total CaCO ₃)	102	131	154
Conductivity (µS/cm)	241	291	337
pH	8.3	8.5	8.6
Average Wetted Width (m)	9.3	9.1	5.6
Pool Proportion (%)	6.4	7.5	4.4
Average Pool Residual Depth (m)	0.5	0.8	0.2
Riffle Proportion (%)	12.5	17.6	7.8
Run Proportion (%)	81.1	74.9	87.8
Cascade Proportion (%)	0	0	0
Average Bank Angle (°)	152.4	147.5	124.8
Undercut Proportion (%)	10	0	20
Average Undercut Depth (m)	0.27	NA	0.03
Discharge (m ³ /s)	1.02	1.44	0.76
Proportion of Refuge Flow (% of velocities <0.1 m/s)	17	14.9	22.9
Proportion of Fast Flow (% of velocities >0.4 m/s)	53.2	57.4	35.4
Canopy Cover (%)	40.5	61.6	33
Aquatic Vegetation Cover (%)	4	0	0
Shannon-Weaver Diversity Index (H)	1.7	2.2	1.5
Simpson's Diversity Index (D)	0.8	0.8	0.7
Dominate Substrate Type	Gravel	Gravel	Sand
Proportion of Fine Substrate (%)	39	44	53
Average Embeddedness (1-5)	3.7	3.4	3.9

Appendix Table B. 5. Abundance estimates and 95% confidence intervals in parentheses for the three New Mexico Department of Game and Fish Permanent Monitoring (NMDGF Permanent) site, San Francisco River, New Mexico study reaches, collected in August and November 2022. NA denotes reaches where we did not capture a particular fish species. * denotes fish species where reasonable confidence intervals could not be obtained due to either 1) incidental capture of a single fish of a species or 2) not achieving depletion for the species due to poor visibility and high stream flows. Species observed in the NMDGF Permanent site include Speckled Dace *Rhinichthys osculus*, Longfin Dace *Agosia chrysogaster*, Sonora Sucker *Catostomus insignis*, Desert Sucker *Pantosteus clarkii*, and Spikedace *Meda fulgida*.

Study Reach	Speckled Dace	Longfin Dace	Sonora Sucker	Desert Sucker	Spikedace
2	17 (2, 31)	40 (*)	17 (*)	70 (*)	5 (3, 7)
3	7 (*)	163 (147, 179)	3 (2, 4)	8 (5, 10)	1 (*)
8	1(*)	16 (15, 17)	33 (*)	3 (0, 6)	NA

Appendix Table B. 6. Standardized taxa and counts for the New Mexico Department of Game and Fish Permanent Monitoring (NMDGF Permanent) site, San Francisco River, New Mexico macroinvertebrate samples, collected in August and November 2022.

Study Reach	Order	Family	Counts
NMDGF Permanent 2	Amphipoda- scuds or side swimmers	Gammaridae	1
	Basommatophora-snails	Planorbidae	1
	Coleoptera-larval beetles	Elmidae	26
	Diptera-true flies	Simuliidae	37
	Emphemeroptera-mayflies	Baetidae	1
		Leptohyphidae	1
	Hemiptera-true bugs	Belostomatidae	1
		Herbidae	9
		Veliidae	15
	Odonata-dragonflies and damselflies	Calopterygidae	1
	NMDGF Permanent 3	Amphipoda- scuds or side swimmers	Gammaridae
Diptera-true flies		Dixidae	2
		Tabanidae	2
Emphemeroptera-mayflies		Baetidae	5
		Leptohyphidae	13
Haplotaxida-worms		Lumbriculidae	5
Hemiptera-true bugs		Belostomatidae	10
		Corixidae	2
		Mesoveliidae	5
		Veliidae	28
Odonata-dragonflies and damselflies		Calopterygidae	13
NMDGF Permanent 8	Trichoptera-caddisflies	Hydropsychidae	5
	Diptera-true flies	Chironomidae	3
	Emphemeroptera-mayflies	Heptageniidae	5
		Leptohyphidae	6
		Siphonuridae	23
	Hemiptera-true bugs	Veliidae	5
	Odonata-dragonflies and damselflies	Gomphidae	2
	Plecoptera-stoneflies	Leutridae	51

Appendix Table B. 7. Summary habitat data for the Box site, San Francisco River, New Mexico, collected in October 2022. NA denotes measurements not applicable to a reach.

Suitability Variable	Box 1	Box 2	Backup 1	Backup 2	Box 4	Box 5
Elevation (m)	1861	1872	1881	1904	1917	1918
LWD	4	3	6	4	1	0
Minimum Temperature (°C)	1.6	1.6	1.6	1.6	1.6	1.6
Maximum Temperature (°C)	24.2	24.2	24.2	24.2	24.2	24.2
Median Temperature (°C)	13.4	13.4	13.4	13.4	13.4	13.4
Instantaneous Temperature (°C)	8	8.4	4.2	5	6.7	5.8
Dissolved Oxygen (mg/L)	9.2	9.4	10.2	10.3	9.9	10
Visual Clarity (cm)	31	37	37	38	60	48
Hardness (mg/L as total CaCO₃)	144	150	155	155	162	164
Alkalinity (mg/L as total CaCO₃)	177	182	183	185	192	190
Conductivity (µS/cm)	376	371	381	388	387	390
pH	9	9	8.9	8.9	9	8.9
Average Wetted Width (m)	5.7	4.3	6.1	5.4	6	3.9
Pool Proportion (%)	16	14.3	24.9	21.1	42.6	18.4
Average Pool Residual Depth (m)	0.4	0.4	0.5	0.7	0.6	0.4
Riffle Proportion (%)	2.9	17	13.4	9	14.9	62.3
Run Proportion (%)	80.1	68.7	49.7	42.5	27.2	17
Cascade Proportion (%)	1	0	12	27.4	15.3	2.3
Average Bank Angle (°)	154.1	157.7	107.2	104.9	141.7	121.4
Undercut Proportion (%)	0	0	40	50	10	10
Average Undercut Depth (m)	NA	NA	0.2	0.22	0.5	0.16
Discharge (m³/s)	0.1	0.15	0.15	0.16	0.20	0.12
Proportion of Refuge Flow (% of velocities <0.1 m/s)	77.1	57.5	55.6	56.8	53.2	41.3
Proportion of Fast Flow (% of velocities >0.4 m/s)	10.4	8.5	2.2	13.6	23.4	17.4
Canopy Cover (%)	70.8	88.7	89.7	16.2	7.6	7.6
Aquatic Vegetation Cover (%)	2	0	2	12	2	18
Shannon-Weaver Diversity Index (H)	2	1.2	2	1.6	2.3	2.3
Simpson's Diversity Index (D)	0.8	0.5	0.8	0.7	0.9	0.9
Dominate Substrate Type	Gravel	Sand/Cobble	Sand/ Gravel	Sand/Gravel	Sand	Sand
Proportion of Fine Substrate (%)	31	33	33	28	49	32
Average Embeddedness (1–5)	3.5	3.8	3.7	4.3	3.5	3.5

Appendix Table B. 8. Abundance estimates and 95% confidence intervals in parentheses for the six Box site, San Francisco River, New Mexico study reaches, collected in October 2022. NA denotes reaches where we did not capture a particular fish species. * denotes fish species where reasonable confidence intervals could not be obtained due to either 1) incidental capture of a single fish of a species or 2) not achieving depletion for the species. Species observed in the Box site include Speckled Dace *Rhinichthys osculus*, Longfin Dace *Agosia chrysogaster*, Sonora Sucker *Catostomus insignis*, Desert Sucker *Pantosteus clarkii*, Rio Grande Sucker *Pantosteus plebeius*, Loach Minnow *Rhinichthys cobitis*, Fathead Minnow *Pimephales promelas*, and Rainbow Trout *Oncorhynchus mykiss*.

Study Reach	Speckled Dace	Longfin Dace	Sonora Sucker	Desert Sucker	Rio Grande Sucker	Loach Minnow	Fathead Minnow	Rainbow Trout
1	20 (14, 26)	35 (*)	25 (8, 42)	24 (15, 33)	NA	1(*)	1 (0, 2)	4 (2, 6)
2	NA	13 (11, 15)	16 (14, 18)	37 (3, 71)	NA	NA	NA	27 (23, 31)
Backup 1	4 (4, 4)	110 (89,131)	36 (15, 57)	25 (8, 42)	19 (12, 6)	NA	NA	25 (19, 31)
Backup 2	NA	29 (24, 34)	25 (23, 27)	43 (32, 54)	41 (18, 64)	NA	NA	28 (26, 30)
4	NA	NA	23 (15, 31)	46 (35, 57)	10 (4, 16)	NA	NA	38 (30, 46)
5	2 (1, 3)	46 (43, 49)	24 (13, 35)	29 (19, 39)	76 (61, 91)	NA	NA	5 (3, 7)

Appendix Table B. 9. Standardized taxa and counts for the Box site, San Francisco River, New Mexico macroinvertebrate samples, collected in October 2022.

Study Reach	Order	Family	Counts	
Box 1	Amphipoda- scuds or side swimmers	Gammaridae	5	
	Coleoptera-larval beetles	Dytiscidae	5	
		Psephenidae	3	
	Diptera-true flies	Simulidae	3	
		Tabanidae	3	
		Tipulidae	3	
	Emphemeroptera-mayflies	Heptageniidae	7	
		Leptohyphidae	22	
		Siphonuridae	38	
	Hemiptera-true bugs	Acanthosomatidae	3	
	Odonata-dragonflies and damselflies	Coenagrionidae	5	
	Plecoptera-stoneflies	Leutridae	3	
	Box 2	Amphipoda- scuds or side swimmers	Gammaridae	66
Emphemeroptera-mayflies		Heptageniidae	5	
		Leptohyphidae	16	
		Siphonuridae	4	
Hemiptera-true bugs		Cicadellidae	3	
Odonata-dragonflies and damselflies		Coenagrionidae	3	
Plecoptera-stoneflies		Leutridae	3	
Backup 1		Amphipoda- scuds or side swimmers	Gammaridae	3
		Coleoptera-larval beetles	Psephenidae	4
		Diptera-true flies	Simulidae	4
	Emphemeroptera-mayflies	Heptageniidae	12	
		Leptohyphidae	24	
		Siphonuridae	10	
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	4	
	Odonata-dragonflies and damselflies	Coenagrionidae	3	
	Orthoptera-grasshoppers, locust and crickets	Gryllidae	2	
	Plecoptera-stoneflies	Leutridae	5	
	Trichoptera-caddisflies	Hydropsychidae	29	
	Backup 2	Amphipoda- scuds or side swimmers	Gammaridae	1
Diptera-true flies		Simulidae	3	
Emphemeroptera-mayflies		Heptageniidae	14	
		Leptohyphidae	27	
		Siphonuridae	40	
Haplotaxida-worms		Lumbriculidae	1	
Odonata-dragonflies and damselflies		Coenagrionidae	2	
Plecoptera-stoneflies		Leutridae	9	
Trichoptera-caddisflies		Hydropsychidae	3	
Box 4		Amphipoda- scuds or side swimmers	Gammaridae	3
	Coleoptera-larval beetles	Dytiscidae	1	
	Diptera-true flies	Certopognidae	1	
		Chironomidae	3	
		Simulidae	8	
		Tabanidae	3	
		Tipulidae	2	
	Emphemeroptera-mayflies	Heptageniidae	8	
		Leptohyphidae	22	
		Siphonuridae	26	
	Haplotaxida-worms	Lumbriculidae	4	

Appendix Table B. 9. -continued. Standardized taxa and counts for the Box site, San Francisco River, New Mexico macroinvertebrate samples, collected in October 2022.

Study Reach	Order	Family	Counts
Box 5	Hemiptera-true bugs	Cicadellidae	1
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	1
	Odonata-dragonflies and damselflies	Coenagrionidae	3
	Plecoptera-stoneflies	Leutridae	5
	Trichoptera-caddisflies	Hydropsychidae	8
	Amphipoda- scuds or side swimmers	Gammaridae	1
	Diptera-true flies	Chironomidae	4
	Emphemeroptera-mayflies	Heptageniidae	11
		Leptohyphidae	18
		Siphonuridae	19
	Hemiptera-true bugs	Cicadellidae	2
		Herbidae	2
		Mesoveliidae	2
		Veliidae	1
	Megaloptera-alder flies, Dobson flies, and fish flies	Corydalidae	1
	Odonata-dragonflies and damselflies	Coenagrionidae	5
Plecoptera-stoneflies	Leutridae	6	
	Perlodidae	2	
Odonata-dragonflies and damselflies	Coenagrionidae	5	

Appendix Table B. 10. Summary habitat data for the Luna site, San Francisco River, New Mexico, collected in August and September 2022. NA denotes measurements not applicable to a reach.

Suitability Variable	Luna 1	Luna 2	Luna 3	Luna 4	Luna 5	Luna 6	Luna 7	Luna 8
Elevation (m)	2188	2220	2232	2250	2261	2278	2284	2275
LWD	1	1	3	0	2	1	1	0
Minimum Temperature (°C)	0	0	0	0	0	0	0	0
Maximum Temperature (°C)	27.5	27.5	27.5	27.5	27.5	27.5	27.5	27.5
Median Temperature (°C)	9.1	9.1	9.1	9.1	9.1	9.1	9.1	9.1
Instantaneous Temperature (°C)	15.7	15.6	15.3	14.8	22	12.2	19.6	15.7
Dissolved Oxygen (mg/L)	7	7.6	7.4	7.4	6.8	8	6.6	7
Visual Clarity (cm)	34	9	19	13	60	60	60	60
Hardness (mg/L as total CaCO₃)	153	144	162	290	134	140	108	113
Alkalinity (mg/L as total CaCO₃)	158	152	176	142	150	153	100	108
Conductivity (µS/cm)	354	382	390	313	319	317	230	251
pH	8.6	8.6	8.7	8.6	8.7	8.4	8.0	7.8
Average Wetted Width (m)	2.2	2.3	2	2.8	3.5	2.8	2.2	2.3
Pool Proportion (%)	2.7	6.5	16.8	3.1	10.2	2.7	7	13.7
Average Pool Residual Depth (m)	0.2	0.1	0.2	0.2	0.3	0.1	0.2	0.2
Riffle Proportion (%)	1.4	5.8	3.6	25.6	63.4	42.6	15.1	18.2
Run Proportion (%)	95.9	87.7	79.3	70.4	24.2	54.7	77.9	68.1
Cascade Proportion (%)	0	0	0.3	0.9	2.2	0	0	0
Average Bank Angle (°)	138.7	127.6	125.3	117.7	163.1	143.5	135.6	132.7
Undercut Proportion (%)	0	20	10	20	0	10	0	10
Average Undercut Depth (m)	NA	0.05	0.12	0.05	NA	0.12	0.07	0.24
Discharge (m³/s)	0.01	0.01	0.01	0.01	0.13	0.1	0.01	0.02
Proportion of Refuge Flow (% of velocities <0.1 m/s)	91.1	76.5	84.9	75.6	38.3	7	61.4	81.4
Proportion of Fast Flow (% of velocities >0.4 m/s)	0	0	0	0	29.8	34.9	2.27	2.3
Canopy Cover (%)	35.7	36.8	46.5	40.5	31.9	24.3	45.9	18.4
Aquatic Vegetation Cover (%)	4	24	24	16	18	28	28	46
Shannon-Weaver Diversity Index (H)	1.3	1.1	2.2	2.1	2.4	1.2	1.2	1
Simpson's Diversity Index (D)	0.7	0.4	0.8	0.8	0.9	0.6	0.6	0.5
Dominate Substrate Type	Silt/Cobble	Gravel	Cobble	Sand	Gravel	Gravel	Cobble	Gravel
Proportion of Fine Substrate (%)	40	28	22	38	33	28	6	7
Average Embeddedness (1–5)	3.3	3.5	3.1	3.1	4	4	3.6	4

Appendix Table B. 11. Abundance estimates and 95% confidence intervals in parentheses for the eight Luna site, San Francisco River, New Mexico study reaches, collected in August and September 2022. NA denotes reaches where we did not capture a particular fish species. * denotes fish species where reasonable confidence intervals could not be obtained due to either 1) incidental capture of a single fish of a species or 2) not achieving depletion for the species. Species observed in the Luna site include Speckled Dace *Rhinichthys osculus*, Longfin Dace *Agosia chrysogaster*, Sonora Sucker *Catostomus insignis*, Desert Sucker *Pantosteus clarkii*, Rio Grande Sucker *Pantosteus plebeius*, and Fathead Minnow *Pimephales promelas*.

Study Reach	Speckled Dace	Longfin Dace	Desert Sucker	Rio Grande Sucker	Fathead Minnow
1	6 (3, 9)	149 (29, 259)	NA	NA	NA
2	11 (10, 12)	109 (95, 123)	NA	NA	NA
3	89 (72, 106)	261 (223, 299)	1 (*)	1 (0, 2)	NA
4	279 (198, 360)	525 (406, 643)	35 (19, 50)	11 (9, 13)	NA
5	79 (30, 128)	150 (110, 190)	2 (1, 3)	9 (*)	NA
6	153 (139, 167)	285 (265, 305)	NA	21 (19, 23)	NA
7	36 (31, 41)	20 (14, 26)	NA	5 (2, 8)	1 (*)
8	305 (238, 372)	198 (175, 221)	14 (10, 18)	42 (34, 50)	8 (6, 10)

Appendix Table B. 12. Standardized taxa and counts for the Luna site, San Francisco River, New Mexico macroinvertebrate samples, collected in August and September 2022.

Study Reach	Order	Family	Counts	
Luna 1	Coleoptera-larval beetles	Elmidae	46	
	Diptera-true flies	Chironomidae	8	
	Emphemeroptera-mayflies	Baetidae	8	
	Hemiptera-true bugs	Aphididae	3	
Luna 2	Hymenoptera- sawflies, wasps, bees and ants	Gerridae	32	
		Thynnidae	3	
	Amphipoda- scuds or side swimmers	Gammaridae	1	
	Basommatophora-snails	Planorbidae	8	
	Coleoptera-larval beetles	Dytiscidae	1	
		Elmidae	3	
	Diptera-true flies	Agromyzidae	1	
		Chironomidae	1	
	Emphemeroptera-mayflies	Dixidae	1	
		Simulidae	1	
	Haplotaaxida-worms	Baetidae	75	
	Hemiptera-true bugs	Lumbriculidae	1	
Luna 3	Odonata-dragonflies and damselflies	Corixidae	5	
		Gerridae	1	
	Coleoptera-larval beetles	Lestidae	1	
		Dytiscidae	2	
	Emphemeroptera-mayflies	Elmidae	8	
		Scarabaeidae	1	
	Diptera-true flies	Baetidae	33	
		Heptageniidae	8	
	Hemiptera-true bugs	Leptohypjidae	3	
		Chironomidae	14	
	Luna 4	Odonata-dragonflies and damselflies	Simulidae	5
			Tabanidae	2
		Amphipoda- scuds or side swimmers	Tipulidae	5
			Cicadellidae	3
Coleoptera-larval beetles		Corixidae	9	
		Gerridae	4	
Diptera-true flies		Gomphidae	2	
		Gammaridae	5	
Emphemeroptera-mayflies		Dytiscidae	5	
		Elmidae	5	
Hemiptera-true bugs	Chironomidae	17		
	Baetidae	30		
	Heptageniidae	13		
	Leptohyphidae	2		
	Corixidae	7		
	Gerridae	2		
Luna 5	Trichoptera-caddisflies	Veliidae	2	
		Hydropsychidae	10	
	Amphipoda- scuds or side swimmers	Gammaridae	4	
	Basommatophora-snails	Planorbidae	3	
	Coleoptera-larval beetles	Dytiscidae	5	
		Chironomidae	7	
	Diptera-true flies	Simulidae	27	
Stratiomyidae		4		
	Tabanidae	3		

Appendix Table B. 12-continued. Standardized taxa and counts for the Luna site, San Francisco River, New Mexico macroinvertebrate samples, collected in August and September 2022.

Study Reach	Order	Family	Counts
Luna 6	Ephemeroptera-mayflies	Tipulidae	3
		Baetidae	19
	Haplotaaxida-worms	Heptageniidae	6
		Leptohyphidae	6
	Hemiptera-true bugs	Lumbriculidae	3
	Lepidoptera-moths	Veliidae	3
	Trichoptera-caddisflies	Noctuidae	3
	Coleoptera-larval beetles	Hydropsychidae	4
	Diptera-true flies	Dyopidae	1
		Hydrophilidae	1
	Ephemeroptera-mayflies	Chironomidae	4
		Simulidae	55
Luna 7	Ephemeroptera-mayflies	Baetidae	31
		Caenidae	4
	Coleoptera-larval beetles	Leptohyphidae	4
		Curculionidae	2
	Diptera-true flies	Dytiscidae	2
		Chironomidae	30
Luna 8	Ephemeroptera-mayflies	Simulidae	6
		Baetidae	56
	Hemiptera-true bugs	Corixidae	2
		Gerridae	2
	Amphipoda- scuds or side swimmers	Crangonyetidae	20
	Coleoptera-larval beetles	Hydrophilidae	1
Diptera-true flies	Simulidae	1	
	Siphonuridae	71	
Ephemeroptera-mayflies	Baetidae	2	
	Heptageniidae	1	
Hemiptera-true bugs	Corixidae	1	
Odonata- dragonflies and damselflies	Calopterygidae	1	
	Lestidae	2	