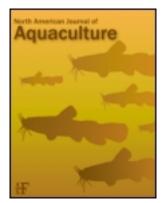
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Captive Breeding of Endangered Mohave Tui Chub

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Abstract.—The endangered Mohave tui chub Gila bicolor mohavensis, which occurs only in three populations in southern California, has not been previously spawned in captivity. Captive breeding of imperiled fishes can be important to conserve biodiversity and aid in native fish conservation efforts by reducing the collection of wild fish for translocations, providing individuals for experimental studies, and ensuring species survival. To spawn Mohave tui chub, we lowered water temperature to 10°C, held this temperature constant for 30 d, and then warmed the tank at a rate of 1°C per day to reach ambient air temperature (20–22°C). We used a photoperiod of 10 h light: 14 h dark and adjusted the photoperiod to 14 h light: 10 h dark when the tank reached 15°C. These photoperiod and water temperature regimes were designed to simulate Mojave Desert springtime conditions. Artificial plants were provided as spawning substrate. Three spawning events produced over 1,700 larval fish. Fish not subjected to this photoperiod and water temperature regime did not spawn. Simulation of a spring photoperiod and temperature regime was associated with successful spawning of Mohave tui chub in captivity and may be effective for spawning other endangered cyprinids with similar requirements.

Captive breeding can be important for conserving biodiversity and aiding in native fish conservation efforts by reducing the collection of wild fish for translocations or experimental studies, allowing accurate observations of early life history traits, and ensuring survival of species (Buyanak and Mohr 1981; Rakes et al. 1999; Sarkar et al. 2006). Many common and endangered fishes, including cyprinids, have been spawned in the laboratory (Buyanak and Mohr 1981; Hamman 1982a, 1982b; Kaya 1991; Brandt et al. 1993; Rakes et al. 1999).

Mohave tui chub *Gila bicolor mohavensis* (Snyder 1918) is the only native fish in the Mojave River basin, California. Populations declined after the 1930s, when competition occurred with arroyo chub *Gila orcuttii* (Hubbs and Miller 1943), which were believed to have been introduced into the headwaters by anglers. Mohave tui chub were eliminated from the Mojave River system by the late 1960s and existed only in one isolated pool in Mojave National Preserve at Zzyzx Mineral Springs, California (Miller 1968). Mohave tui chub were federally listed as endangered in 1970. Recovery efforts involved transplantations of fish to establish new populations, and despite many attempts, only three populations occur currently in springs in

Mohave tui chub are inactive during the coldest months (Vicker 1973), and the average minimum water temperature during January is about 8°C. In Lake Tuendae, Mohave tui chub spawn as early as February, with peak spawning occurring when the water warms to 18°C in mid-March (Vicker 1973). Tui chub spawn over vegetation, and Kimsey (1954) observed that Eagle Lake tui chub *Gila bicolor* eggs that fell into the substrate did not develop. Our objective was to determine whether Mohave tui chub could be spawned, without the use of hormones, by manipulating photoperiod, temperature, and substrate.

Methods

Fish collection and husbandry.—We used minnow traps to collect 25 adult Mohave tui chub from Lake Tuendae, located at Zzyzx, California (Mojave National Preserve), in August 2005 (U.S. Fish and Wildlife Service Recovery Permit TE086593–0). Fish total length ranged from 120 to 220 mm at the time of collection. We followed guidelines in Widmer et al. (2005) to transport fish to holding facilities in Tucson, Arizona, and to treat them for external parasites. In

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southern California: Lark Seep on the China Lake Naval Weapons Center, Camp Cady near Harvard Road in Barstow, and Zzyzx Mineral Springs in Mojave National Preserve (Hoover and St. Amant 1983; Hughson and Woo 2004). Captive breeding would contribute to efforts to maintain and propagate this highly endangered species. Vicker (1973) made several unsuccessful attempts to induce spawning in the laboratory, but no other captive breeding has been attempted.

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addition, we placed fish in a praziquantel (Prazi Pond, AquaScience Research Group, North Kansas City, Missouri) bath (6 mg/L for 24 h) to remove Asian tapeworms *Bothriocephalus acheilognathi* and other internal parasites.

Because of the highly endangered status of Mohave tui chub, we could not collect large numbers of broodstock for numerous tank replicates of spawning experiments. We housed 15 of the fish, after 2 months of acclimation to aquaria, in a 476-L acrylic tank (510 \times 510 \times 1,830 mm) fitted with a custom-built recirculating biofilter and filled with well water (pH = 8.0). We pumped water to the filter with an in-line pump (Model 5, maximum flow rate = 1,900 L/h; Aquatic Eco-Systems, Inc., Apopka, Florida). Water exited the biofilter by gravity, passed through an in-line chiller (Prime Chiller Model 2626; Current-USA, Vista, California) and returned to the tank through a 5-cm polyvinyl chloride T-fitting to diffuse the return flow. The water level in the tank was maintained at 450 mm, the same height at which water was returned to the tank from the chiller. Bloodworms and pellet food (ZP1; Aquatic Eco-Systems) were each fed to the fish once per day ad libitum. Waste was removed daily, and routine water changes and cleaning were performed as needed to maintain ammonia and nitrite levels at undetectable levels (0 mg/L). At least once per month, we replaced 10\% of the tank water with clean well water.

We covered the outside bottom, back, and sides of the tank with 5-mm foam insulation. Pea-sized gravel substrate was placed on one-half of the tank bottom; the other half was left bare, and three pottery shards were put in the tank to provide cover. We used two 40-W, 1,220-mm fluorescent lights placed 30 cm above the water level and connected to electronic timers to provide specific photoperiods.

Ten of the fish were housed under similar conditions including the same feeding regimes and access to spawning plants, cover, and pea gravel substrate. However, they were kept at ambient room temperature (approximately 20–23°C) and were not subjected to photoperiod manipulation (i.e., a photoperiod of 12 h light: 12 h dark was maintained at all times).

Temperature and photoperiod manipulations.—To induce spawning in the treatment tank, we simulated Lake Tuendae winter conditions and then springtime conditions. We first lowered the water temperature in the tank at a rate of 1°C per day to about 9°C and used a photoperiod of 10 h light: 14 h dark. When the water temperature reached 10°C, it was held constant for 30 d. After 30 d, we warmed the tank at a rate of 1°C per day to reach ambient air temperature (20–22°C) and adjusted the photoperiod to 14 h light: 10 h dark when

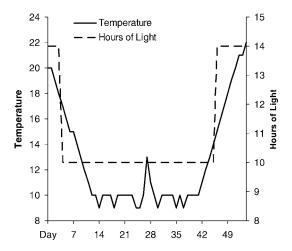


FIGURE 1.—Photoperiod (h of light) and temperature (°C) regimen for laboratory spawning of Mohave tui chub. Temperature was recorded daily at 0800 hours. Control fish, which did not spawn, were kept under similar conditions except that temperature and photoperiod were not fluctuated and remained at ambient laboratory conditions.

the tank reached 15°C (Figure 1). Tanks were checked daily for presence of eggs.

Spawning substrate.—We attached two artificial plants (Fancy Plants Giants asparagus fern; Aquarium Systems, Mentor, Ohio) to a plastic grate and placed them in the tank with a large rock to prevent the grate from floating. Plants provided a substrate for egg attachment, as well as additional cover, and could easily be removed. For the second and third spawns, unglazed ceramic tiles were placed under the grate to capture eggs that did not adhere to plants. We transferred the artificial plants and tiles containing eggs to 76-L rearing tanks after spawning occurred. Eggs were incubated at ambient temperature (20–23°C). Larval fish and later stages were fed appropriately sized commercial fish food (LD100, LD150, LD250, and ZP3; Aquatic Eco-Systems).

Results and Discussion

As water was warmed to 15°C, we noticed increased activity among fish. Many fish developed a red tinge on the bases of paired fins, and fish were often seen close to the artificial plants. These observations are in agreement with previous studies of tui chub spawning in the wild (Kimsey 1954; Vicker 1973). On 6 February 2006, 1 month after the tanks reached 20–23°C, we found eggs in the tanks. We suspect spawning occurs at night because we checked tanks daily and never witnessed spawning; eggs were always first noticed in the morning. Three spawns occurred within 2 weeks, but the total number of eggs was

difficult to estimate because they were hard to see in the artificial plants. The first spawn yielded 166 larval fish, and the latter two spawns yielded more than 800 larval fish each. At 20–23°C, eggs hatched in about 4 d, and prolarvae reached the swim-up stage less than 24 h after hatching.

Fish appeared agitated when first brought into captivity. Adding substrate and artificial plants calmed the fish. Temperature cycling and photoperiod may be important cues for spawning. The 10 fish that were kept under similar conditions with access to spawning plants but not subjected to temperature cycling or photoperiod manipulation did not spawn during 1 year of captivity. Also, no new eggs appeared in either of the tanks after artificial plants were removed. Whether the plants served as a cue for spawning or simply provided cover to prevent eggs from being eaten immediately is unknown.

About 400 offspring were used in subsequent laboratory growth and survival studies to assess the effects of parasites. The remaining fish were transported back to Mojave National Preserve and used to assess habitat suitability for future populations. Manipulating temperature and photoperiod to simulate spawning conditions in the wild was associated with successful spawning of this endangered cyprinid. These techniques might be useful for propagating other imperiled cyprinids as well.

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