

## Growth and Survival of Larval and Juvenile Gila Chub at Different Temperatures

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**Abstract.**—The information needed to effectively culture imperiled native fishes for recovery efforts is lacking for certain species, yet it is critical for proper management and conservation. Culture techniques and requirements are virtually unknown for Gila chub *Gila intermedia*, a species federally listed as endangered. We tested the effect of four different water temperatures on the growth, survival, and overt health and appearance of larval (20, 24, 28, and 32°C) and two sizes of juvenile Gila chub (20, 23, 26, and 29°C). Growth of larvae was highest at 28°C and lowest at 32°C, whereas survival of larvae was highest at 24°C and lowest at 20°C. Spinal deformities were common for larvae reared at 32°C but generally uncommon for those reared at lower temperatures. Although growth of small (32–49 mm total length) and large (52–72 mm) juveniles generally increased with temperature, the differences were not statistically significant. In any experiment testing small and large juveniles, survival was 100% and no external abnormalities were noted. Water temperatures from 20°C to 28°C appear suitable for rearing larval Gila chub, temperatures from 24°C to 28°C being optimal. Water temperatures from 20°C to 29°C appear suitable for rearing juvenile Gila chub.

Gila chub *Gila intermedia* is a moderate-sized cyprinid endemic to the Gila River basin of central and southeastern Arizona, southwestern New Mexico, and northern Sonora, Mexico (Rinne 1976; Minckley and DeMarais 2000) whose range has been reduced by the loss and modification of aquatic habitats (Hendrickson and Minckley 1984; Rinne et al. 2005) and the introduction of nonnative species (Minckley and Deacon 1991; Dudley and Matter 2000). This species is currently limited to about 29 isolated streams, cienegas (marshy areas), and springs (USFWS 2005), only one of which contains a population that is considered stable and secure (Weedman et al. 1996). The Gila chub is listed as endangered with critical habitat under the U.S. Endangered Species Act (USFWS 2005).

The ability to culture threatened native fishes of the Southwest for recovery efforts is lacking for certain species, yet it is critical for proper management and conservation. The limited information available on culture techniques and the general life history of Gila chub hampers recovery of this species. The future of Gila chub may someday depend in part on hatchery propagation to provide specimens for restocking formerly occupied habitats and for establishing refuge populations.

Although growth is affected by many factors, none may be as important as water temperature (Dwyer et al. 1983). Higher growth as temperatures approach an

optimum is well known and is probably related to an increase in food intake, metabolism, and nutrient absorption, as well as other factors (Brett 1964; Harrelson et al. 1988; Kroll et al. 1992; Jobling and Baardvik 1994; Koskela et al. 1997; Deng et al. 2002). Although maximizing production is probably not a main goal in the culture of many imperiled native fishes at this time, providing for an efficient grow-out phase when producing fish for stocking and other efforts has distinct benefits. Faster grow-out to a certain size allows stocking for a greater part of the year, may lower feed and labor costs, and may increase available rearing space. Where piscivores are present, stocking of larger-size individuals may be necessary to lower loss of stock from predation (Marsh and Brooks 1989).

Our objective was to identify the effect of different water temperatures on the growth, survival, and overt health and appearance of Gila chub larvae and juveniles under laboratory conditions.

### Methods

We randomly assigned Gila chub to each of four different treatment levels (test temperatures) with three replications (tanks) per treatment level for each size-class tested. Each 38-L rectangular glass tank was fitted with a recirculating filter system with a stocking density of 40 larval chub (6.0–7.5 mm total length [TL]), 7 small juveniles (32–49 mm), or 5 large juveniles (52–72 mm), yielding a mean initial biomasses of 0.004, 0.19, and 0.49 g/L, respectively. Gila chub were acclimated by increasing water temperature in equally divided intervals over a 5-d period until the desired test temperature was reached. Larval Gila chub

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Received November 16, 2006; accepted November 2, 2007  
Published online January 15, 2009

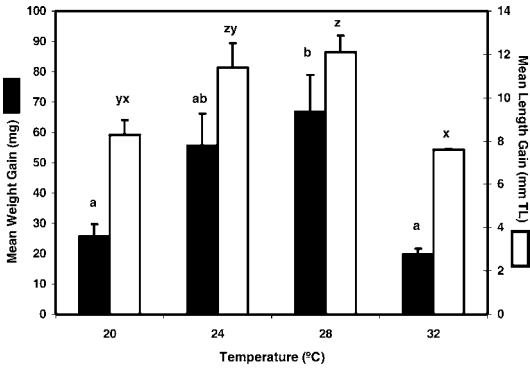


FIGURE 1.—Mean weight and length gains (with standard errors of the means) per test temperature for larval Gila chub after about 30 d. Values with different lowercase letters (a and b for weight; x, y, and z for length) are significantly different ( $P \leq 0.05$ ).

were tested at 20, 24, 28, and 32°C. Juvenile Gila chub were tested at 20, 23, 26, and 29°C. Test temperatures were monitored daily for accuracy and adjusted when necessary. Experiments ran for 29–30 d.

Larval Gila chub were euthanized with MS-222 (3-aminobenzoic acid ethyl ester) prior to measurement. Initial larval measurements were derived from a random subsample ( $n = 18$ ) acquired within 24 h of hatching. Final larval measurements were derived from a random subsample ( $n = 10$ ) of survivors from each treatment group. We measured wet weight (to the nearest microgram for larval fish) by using an electronic scale. Particular care was taken to systematically remove excess water from all larval Gila chub before measurement. We used an ocular micrometer to measure initial TL (to the nearest 0.1 mm) of larvae and calipers to measure final TL (to the nearest 0.1 mm) of

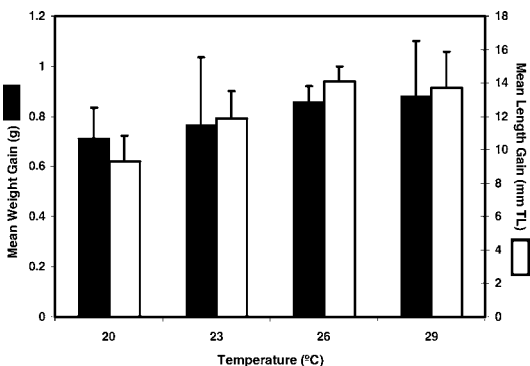


FIGURE 2.—Mean weight and length gains (with standard errors of the means) per test temperature for small juvenile Gila chub (32–49 mm) after about 29 d.

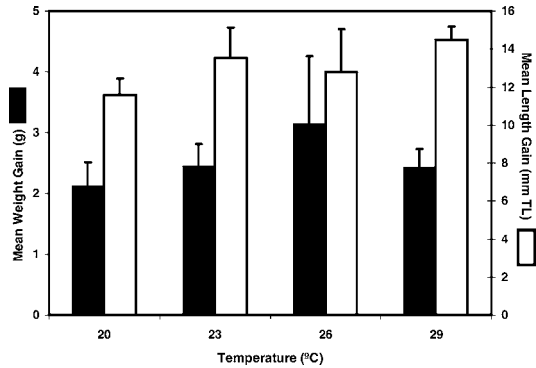


FIGURE 3.—Mean weight and length gains (with standard errors of the means) per test temperature for large juvenile Gila chub (52–72 mm) after about 29 d.

larvae. To measure TL (to the nearest millimeter) of juveniles, we used a measuring board.

Each replicate group of larval Gila chub was fed to excess four times daily with a combination of thawed nauplii of brine shrimp *Artemia* spp. (Hikari Bio-Pure; Hikari, Inc.) and Hikari First-bites (Hikari, Inc.). Each replicate group of juvenile chub was fed to excess three times daily with a combination of unfrozen chironomid larvae and Hikari Micro-pellets for small juveniles or Silver Cup (Nelson and Sons, Inc.) for large juveniles.

We used one-way analysis of variance (ANOVA) or Welch's ANOVA test (when group variances were significantly different) to test for significant differences in mean weight and length gain and the percent survival of larvae and juveniles among test temperatures. If a statistically significant ( $P \leq 0.05$ ) difference was detected in ANOVA tests, we used a Tukey–Kramer honestly significant difference multiple comparison procedure to identify which means differed. We used Pearson's chi-square test to determine whether the incidence of spinal deformity of larvae was different among different test temperatures.

## Results

The mean weight and length gains of larval Gila chub were significantly different ( $F = 6.87$  and  $11.05$ ;  $df = 3, 8$ ;  $P = 0.05$  and  $0.03$ , respectively) among test temperatures. Growth of larvae increased as temperature increased up to 28°C but decreased markedly at 32°C (Figure 1). The mean weight gain of larvae was significantly greater at 28°C than at 20°C and 32°C. There was no statistical evidence of differences in mean weight and length gain for small ( $F = 0.17$  and  $1.80$ ;  $df = 3, 8$ ;  $P = 0.91$  and  $0.22$ , respectively) or large ( $F = 0.47$  and  $0.67$ ;  $df = 3, 8$ ;  $P = 0.70$  and  $0.59$ , respectively) juveniles among test temperatures (Figures 2, 3).

Mean percent survival was highest for larvae reared at 24°C, but we found no statistical evidence (ANOVA = 2.76; *df* = 3, 8; *P* = 0.11) of a difference in survival among test temperatures (Figure 4). Mortalities were all but nonexistent (one accidental) for either juvenile size-class. There was strong evidence ( $\chi^2 = 31.11$ ; *P* < 0.001) that spinal deformities in larvae differed among test temperatures; such deformities were present in almost half (47%) of the larval chub reared at 32°C but were less common (23%) for those reared at 24°C and nonexistent for those reared at 20°C and 28°C. No other overt abnormalities were noted for larvae. All of the juveniles tested appeared overtly healthy throughout the experiment.

### Discussion

Of the temperatures we tested, the optimal temperature for the growth of larval Gila chub was 28°C; moreover, there probably was a temperature threshold (28–32°C) at which the growth rate markedly decreased. The optimal temperature for the survival and health of larvae remains unclear, but survival was on average better at 24°C than at the other temperatures tested. Although a positive relationship between growth and temperature was sometimes apparent and juveniles seemed to grow best between 26°C and 29°C, these results were not statistically significant. A statistically significant difference in growth among test temperatures for juveniles may have been revealed if we had conducted the experiment for a longer period or used a wider range of test temperatures or more replicates to produce a more powerful test.

The temperature at which the highest growth rate occurred is probably optimal for most physiological processes (Harrelson et al. 1988). However, further insight as to the relationship between optimal growth and factors independent of growth can shape criteria when determining optimal culture temperature. Diseases, and resistance to certain diseases, can vary with temperature (Harrelson et al. 1988) and are always a concern. In addition, temperature is one of many factors that appear to play a role in the development of deformations (Abdel et al. 2005; Fitzsimmons and Perutz 2006). In general, a higher incidence of malformations has been found in cultured rather than wild fishes (Komada 1980 and citations therein) and such malformations are still considered an important problem in intensive aquaculture (Aritaki et al. 1996; Fraser et al. 2004). Although we found the incidence of spinal deformities for larval Gila chub to be much higher at 32°C, there was no clear trend across temperatures: deformities occurred at 24°C and 32°C but not at 20°C and 28°C. It is generally considered prudent for culturists to produce fish that are similar in

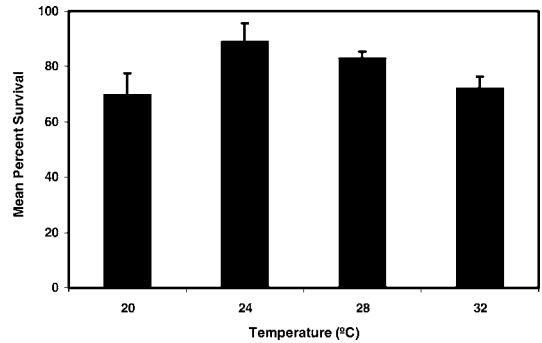


FIGURE 4.—Mean percent survival (with standard errors of the means) per test temperature for larval Gila chub after about 30 d.

morphological, physiological, behavioral, and biochemical characteristics to their wild counterparts. Because our observations were made on strictly overt signs of deformation, further investigation into unseen effects of various culture conditions on Gila chub may be warranted. Matsouka (2003) found that reared fish with abnormalities often showed no obvious external signs of deformation.

Our tests were conducted under relatively well-controlled laboratory conditions. Study of growth and other factors under more variable conditions, such as outdoor ponds, is needed for Gila chub. Growth rates can be greater in a cyclic temperature regime than in a static one (Harrelson et al. 1988).

Based on the parameters and results of our study, water temperatures from 20°C to 28°C appear suitable for rearing larval Gila chub; temperatures from 24°C to 28°C are recommended for faster growth. Water temperatures from 20°C to 29°C appear suitable for rearing juvenile Gila chub. The increasing prevalence of imperiled fish species and the importance of culturing them a conservation and management strategy (Johnson and Jensen 1991; Modde et al. 1995) are regrettable realities. Nonetheless, culturing can be a powerful tool when stock are needed to repatriate extirpated populations or establish refuge populations.

### Acknowledgments

This project was funded and supported by the Arizona Game and Fish Department (AGFD) Heritage Fund, the Central Arizona Project Transfer Fund, the University of Arizona, and the Arizona Cooperative Fish and Wildlife Research Unit and its cooperators. We thank all the staff and professionals at the AGFD, especially those at the Region V office. We thank Andrew Schultz's Ph. D. committee members—Courtney Conway, Kevin Fitzsimmons, Peter Reinthal,

and Cecil Schwalbe—and William Matter for design support. We thank Paul Barrett, Roger Hamman, and Manuel Ulibarri of the U.S. Fish and Wildlife Service for design and logistical support. We thank Joshua Taiz and associated staff of the U.S. Forest Service, Santa Catalina Ranger District. We thank all the staff and faculty at the University of Arizona who assisted in any respect during this study, especially Anne Hartley, Linda Lee, Dee Simons, Cecily Westphal, and Carol Yde. Special thanks go to Mark Borgstrom and Robert Steidl for statistical advice. We thank Craig Ivanyi and Ken Wintin of the Arizona Sonora Desert Museum. Thanks to Andrew Honaman and Gina Schultz for technical support. Special thanks go to Alison Iles of the Arizona Cooperative Fish and Wildlife Research Unit and to Mitch McClaran, Patrick Reid, and Malcolm Zwolinski of the University of Arizona's School of Natural Resources. Thanks to Daniel Arevalo, Sachiko Aso, Sheldon Caldwell-Meeks, Andrea Francis, Shawna Hehr, Michelle Martin, Michelle Riley, and Sebastian Zeltzer for their assistance in the laboratory. Thanks to all the other individuals, government agencies, nongovernmental groups, and businesses not mentioned here that assisted with the project. This manuscript benefited greatly from the comments of the editors and two anonymous reviewers.

### References

- Abdel, I., E. Abellan, O. Lopez-Albors, P. Valdes, M. J. Nortes, and A. Garcia-Alcazar. 2005. Abnormalities in the juvenile stage of sea bass (*Dicentrarchus labrax*) reared at different temperatures: types, prevalence, and effect on growth. *Aquaculture International* 12:523–538.
- Aritaki, M., T. Seikai, and M. Kobayasi. 1996. Reduction of morphological abnormalities in brown sole by larval rearing with higher temperature and early feeding of *Artemia* nauplii. *Bulletin of the Japanese Society of Scientific Fisheries* 62:857–864.
- Brett, J. R. 1964. The respiratory metabolism and swimming performance of young sockeye salmon. *Journal of the Fisheries Research Board of Canada* 21:1183–1226.
- Deng, D. E., S. J. Teh, F. C. Teh, and S. S. O. Hung. 2002. Effect of diets and water temperatures on growth performance of splittail larvae. *North American Journal of Aquaculture* 64:242–247.
- Dudley, R. K., and W. J. Matter. 2000. Effects of small green sunfish (*Lepomis cyanellus*) on recruitment of Gila chub (*Gila intermedia*) in Sabino Creek, Arizona. *Southwestern Naturalist* 45:24–29.
- Dwyer, W. P., R. G. Piper, and C. E. Simth. 1983. Brook trout growth efficiency as affected by temperature. *Progressive Fish-Culturist* 45:161–163.
- Fitzsimmons, S. D., and M. Perutz. 2006. Effects of egg incubation temperature on survival, prevalence, and types of malformations in vertebral column of Atlantic cod (*Gadus morhua*) larvae. *Bulletin of the European Association of Fish Pathologists* 26:80–86.
- Fraser, M. R., T. A. Anderson, and R. de Nys. 2004. Ontogenic development of the spine and spinal deformities in larval barramundi (*Lates calcarifer*) culture. *Aquaculture* 242:697–711.
- Harrelson, M. E., J. Hudson, and J. B. Cravens. 1988. Thermal effects. *Journal of the Water Pollution Control Federation* 60:978–983.
- Hendrickson, D. A., and W. L. Minckley. 1984. Cienegas—vanishing climax communities of the American Southwest. *Desert Plants* 6:141–175.
- Jobling, M., and B. M. Baardvik. 1994. The influence of environmental manipulations on interindividual and intraindividual variation in food acquisition and growth performance of arctic charr, *Salvelinus alpinus*. *Journal of Fish Biology* 44:1069–1087.
- Johnson, J. E., and B. L. Jensen. 1991. Hatcheries for endangered freshwater fishes. Pages 199–217 in W. L. Minckley and J. E. Deacon, editors. *Battle against extinction: native fish management in the American West*. University of Arizona Press, Tucson.
- Komada, N. 1980. Incidence of gross malformations and vertebral anomalies of natural and hatchery *Plecoglossus altivelis*. *Copeia* 1980:29–35.
- Koskela, J., J. Pirhonen, and M. Jobling. 1997. Effect of low temperature on feed intake, growth rate, and body composition of juvenile Baltic salmon. *Aquaculture* 5:479–488.
- Kroll, K. J., J. P. Vaneennaam, S. I. Doroshov, J. E. Hamilton, and T. R. Russel. 1992. Effect of water temperature and formulated diets on growth and survival of larval paddlefish. *Transactions of the American Fisheries Society* 121:538–543.
- Marsh, P. C., and J. E. Brooks. 1989. Predation by ictalurid catfishes as a deterrent to reestablishment of hatchery-reared razorback suckers. *Southwestern Naturalist* 34:188–195.
- Matsouka, M. 2003. Comparison of meristic variations and bone abnormalities between wild and laboratory-reared red seabream. *Japan Agricultural Research Quarterly* 37:21–30.
- Minckley, W. L., and J. E. Deacon, editors. 1991. *Battle against extinction: native fish management in the American West*. University of Arizona Press, Tucson.
- Minckley, W. L., and B. D. DeMarias. 2000. Taxonomy of chubs (Teleostei, Cyprinidae, genus *Gila*) in the American Southwest with comments on conservation. *Copeia* 2000:251–256.
- Modde, T., A. T. Scholz, J. H. Williamson, G. B. Haines, B. D. Burdick, and F. K. Pfeifer. 1995. Augmentation plan for razorback sucker in the upper Colorado River basin. Pages 102–111 in H. Schramm and R. Piper, editors. *Uses and effects of cultured fishes in aquatic ecosystems*. American Fisheries Society, Symposium 15, Bethesda, Maryland.
- Rinne, J. N. 1976. Cyprinid fishes of the genus *Gila* from the lower Colorado River basin. *Wasmann Journal of Biology* 34:65–107.
- Rinne, J. N., J. R. Simms, and H. Blasius. 2005. Changes in hydrology and fish fauna in the Gila River, Arizona—New Mexico: epitaph for a native fish fauna? Pages 127–137

- in J. N. Rinne, R. M. Hughes, and B. Calamusso, editors. Historical changes in fish assemblages of large rivers in the Americas. American Fisheries Society, Symposium 45, Bethesda, Maryland.
- USFWS (U.S. Fish and Wildlife Service). 2005. Endangered and threatened wildlife and plants; listing the Gila chub as endangered with critical habitat, Final Rule. Federal Register 70:211(2 November 2005):66664–66721.
- Weedman, D. A., A. L. Girmendonk, and K. L. Young. 1996. Status review of Gila chub, *Gila intermedia*, in the United States and Mexico. Arizona Game and Fish Department, Technical Report 91, Phoenix.