

Temperature Tolerance of Humpback Chub (*Gila cypha*)  
and Colorado Squawfish (*Ptychocheilus lucius*),  
With a Description of Culture Methods for Humpback  
Chub

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**Abstract--**The closure of Glen Canyon Dam on the Colorado River above Grand Canyon, Arizona, in 1963 resulted in a depression of spring-summer downstream water temperatures. Post-dam temperatures likely have precluded successful mainstem reproduction of most native species due to mortality during incubation. In addition, movements of early life stage fishes from the Little Colorado River, a major undammed tributary used for spawning and rearing, to the Colorado River mainstem across a large thermal gradient have been surmised to negatively affect survival and growth. We exposed early life stages of endangered humpback chub (*Gila cypha*) and Colorado squawfish (*Ptychocheilus lucius*) to several temperatures to examine effects on survival and growth in the laboratory. All eggs were incubated at 18.6°C and moved to 20°C at swimup for a minimum of 24 h. When transferred from 20°C to 10°C for 4 h, 5-7 d old humpback chub larvae lost equilibrium and mobility for 90 min, but recovered. Same age humpback chub larvae became lethargic but did not lose equilibrium when transferred from 20°C to 12°C, and no behavioral effects were observed when moved to 14°C. Similar cold shock experiments with 11-13 d chub larvae affected only the 10°C group, which lost equilibrium for 15 min before recovering. No mortality was observed under any treatment during the 4 h observation period. Growth patterns of 6-8 d humpback chub larvae reared at 10°C, 14°C, and 20°C averaged 10%, 37%, and 83% length gain, respectively, and 28%, 195%, and 951% weight gain, over 30 d. Overall patterns of growth of 13-15 d humpback chub and Colorado squawfish larvae and 39-41 d chub post-larvae reared at these temperatures were similar. Results suggest that detrimental effects of reduced growth on

individuals and populations of native fishes in Grand Canyon can be ameliorated by reducing levels of daily discharge fluctuations from Glen Canyon Dam, which will provide greater stability and potential for warming of mainstem rearing habitats. We also recommend consideration of thermal modification of discharges from Glen Canyon Dam.

The Colorado River system in Grand Canyon, Arizona, supports the largest of the seven remaining populations of the endangered humpback chub, *Gila cypha* (USFWS 1990). Although the species historically occurred downstream in the Colorado River below present-day Hoover Dam (Miller 1955), the Grand Canyon population is the last remaining in the Lower Colorado River Basin. Closure of Glen Canyon Dam in 1963 effected profound physical changes to the Colorado River downstream (Carothers et al. 1981, Maddux et al. 1987). These alterations proved detrimental to the native fish fauna (Holden and Stalnaker 1975, Suttkus et al. 1976, Kaeding and Zimmerman 1983, Angradi et al. 1992, and others). Colorado squawfish (*Ptychocheilus lucius*), and bonytail (*G. elegans*) and roundtail chubs (*G. robusta*) were soon extirpated from the Colorado River in Grand Canyon, and the continued presence of razorback sucker (*Xyrauchen texanus*), humpback chub, and other native fishes is threatened (*op cit*, Nelson 1978, Minckley 1991, and others).

One of the most notable alterations of the Colorado River resulting from closure of Glen Canyon Dam was a depression of spring-summer water temperatures. This feature likely has precluded successful mainstem reproduction of most native fish species due to mortality during incubation (Hamman 1982, Kaeding and Zimmerman 1983, Marsh 1985). Presently the Little Colorado River (LCR), a perennial tributary to the Colorado River in Grand Canyon, serves as the only known site of reproduction and recruitment for humpback chub (Kaeding and Zimmerman 1983, Maddux et al. 1987, Valdez et al. 1992). Known and suspected movements of early life stage humpback chub from the LCR to the Colorado River mainstem across a large thermal gradient were reported by Kaeding and Zimmerman (1983), Angradi et al. (1992), AGFD (1993), and Robinson et al. (1994). Effects on the life histories of native fishes in the Colorado River Basin resulting from rearing in cold temperatures include increased early life mortality and decreased survival to sexual maturity (Kaeding and Osmundson 1988), reduced condition, lipid stores, and size that results in elevated overwinter mortality for young-of-year fishes (Thompson et al. 1981), and lowered egg production by adults (McAda and Wydoski 1983). Actual effects in Grand Canyon, however, have been poorly studied.

The objectives of this study were to obtain humpback chub reproductive products from the LCR in order to rear larvae and juveniles for use in laboratory experiments to examine effects of

temperature on aspects of survival and growth. In addition, we conducted similar experiments with Colorado squawfish larvae obtained from hatchery broodstock for comparisons with humpback chub.

Previously, adult humpback chub were collected from the LCR and from the Colorado River at Black Rocks, held in captivity for one year or more, spawned by hormone-induced gonadal maturation, and reproductive products used for culture and incubation experiments (Hamman 1982, Marsh 1985). Fertilized ova of humpback chub were also collected from a field spawning effort involving the Black Rocks, Colorado, population, transported to Dexter National Fish Hatchery, and successfully reared (Valdez and Valdez-Gonzales 1991). However, three field spawning attempts of the LCR population resulted in no live larvae (Minckley 1989, Raisanen et al. 1991, Angradi et al. 1992). Thus, an additional goal of the present study was to attempt to determine the cause(s) of previous failures.

## METHODS

### *Spawning and Incubation*

Humpback chub used in temperature experiments were obtained from manually stripped reproductive products of wild fish from the LCR. Spawning and incubation equipment was set up near Salt Trail Canyon, 10.5 km above the confluence with the Colorado River. Ten 6.4 mm mesh hoop nets were set in the immediate vicinity of Salt Trail Canyon, and were checked for presence of fish at least twice per day from April 26 to May 8, 1993. Humpback chub that exhibited secondary sexual characteristics were transported in 19 L buckets equipped with battery powered aerators to 1.2 m X 1.2 m X 1.2 m, 6.4 mm mesh live cars set in a backwater at the Salt Trail Canyon site. Fish were checked for presence of passive integrated transponders (PIT), and unmarked fish were injected intraperitoneally with PIT tags.

Female chub were tested for ripeness by applying pressure along their flanks (Ingram 1985). Females that did not readily express eggs were injected with 4 mg/kg body weight of acetone-dried carp pituitary (10 mg/cc) decanted from previously prepared solution in sterile water held in an ice chest. Males not expressing sperm were also injected in the same manner. Unripe females were injected daily for up to 3 d (Hamman 1982, Marsh 1985).

When ripe, ova from one female and milt from two or three males were stripped manually into a plastic bowl at a shaded spawning table. The area around the vent was dried with clean paper towels to prevent contamination by blood, skin mucus, or water (Piper et al. 1982). The egg/sperm mixture was stirred continuously with a clean feather for 10 min. Sperm diluent (pH stabilizer) and bentonite (egg clumping preventative) were not utilized, as they were suspected agents in previous field culture

failures. Eggs were then poured into an egg basket constructed of 500 :m mesh nylon screen attached to a flotation frame in a cooler of desilted LCR water for water hardening (1-2 h). After spawning, fish were returned to the LCR near the point of capture.

After water hardening, eggs (on their flotation screens) were transferred to 19 L buckets (containing desilted LCR water) floating in the LCR by means of a styrofoam collar. Battery powered aerators provided necessary turbulence as well as oxygen to the eggs. The incubation buckets were shaded with a styrofoam lid.

A small lot of eggs was incubated in an instream hatching box set up in an area of low velocity in the LCR. This apparatus was comprised of a PVC pipe frame, sealed for flotation, and surrounded with 500 :m mesh nylon screen and a styrofoam lid. A Heath tray was suspended inside to hold the eggs, inclined 30° into the current to provide aeration (Hamman 1982).

Temperature, conductivity, dissolved oxygen and pH in the LCR during the field spawning procedures were monitored hourly with a Hydrolab Surveyor 3 data logger and H20 sonde. Turbidity was measured periodically with a Baush and Lomb Spectronic Mini-20 nephelometer. Water quality within hatching buckets was also measured periodically with these instruments.

After 1-3 d of onsite incubation, eggs were airlifted by helicopter to Bubbling Ponds Hatchery. They were double-bagged in 4 mil plastic bags filled 1/3 with desilted LCR water and 2/3 pure oxygen, and crated in insulated styrofoam egg shipping containers.

Upon arrival at Bubbling Ponds, bags of eggs were transitioned in Bubbling Ponds water for temperature and chemical acclimation. Ten percent of the LCR water volume was exchanged with Bubbling Ponds water every ten minutes for 90 minutes. The eggs (still adhered to the nylon screens) were then transferred to Heath tray incubators. Water flow rates in the Heath tray apparatus was adjusted to preclude movement of eggs. Eggs were treated with 1,667 ppm formalin in a 15 min drip daily for control of aquatic fungi (*Saprolegniales*) (Schreck and Fitzpatrick, 1991).

Heath trays were checked 4 times daily for larvae. Larvae were removed with a pipette and placed in shallow troughs of through-flowing 18.6°C water until swimup. Larval baskets were placed under each Heath rack at the outflow to catch any larvae otherwise missed (Dupree and Huner 1984).

Zooplankton collected from hatchery ponds, *Artemia salina* larvae hatched from cysts, and Bio-Kyowa B-250 were fed to larvae approaching first-feeding stage. Later (in 2-4 weeks), larvae were fed a diet of commercial trout feed of progressively larger sizes as needed.

#### *Temperature Experiments*

Eighteen 10 gal aquaria were individually supplied with recirculating Bubbling Ponds water inside a temperature-controlled room. Two Aquanetics 1/4 hp chiller system packs (Model No. CSP-3) were individually connected to each of 2 sets of six aquaria. The third set of aquaria was supplied with ambient 20°C recirculated water. The chiller system packs were equipped with mechanical, biological, and chemical filters, and 1,900 lph pumps. A separate, identical pump and filter system handled the 20°C set of tanks. A 14:10 h light-dark photoperiod, approximating the springtime day length, was provided with illumination from wide spectrum fluorescent lights.

In an attempt to simulate conditions experienced by fishes entering the Colorado River mainstem from the LCR, thermal shock experiments were conducted on 5-7 d old (swimup stage) and 11-13 d humpback chub larvae. Water temperature was first gradually increased from 18.6°C (incubation temperature) to 20°C, and larvae were allowed to acclimate for a minimum of 24 h. Three replicates of 20 fish each were then transferred separately to tanks at 10°C, 12°C, and 14°C  $\pm$  0.5°C. Three replicate control groups were handled in like manner but transferred to 20  $\pm$  0.5°C tanks. Fish were observed for 4 h under each treatment.

Tolerance of humpback chub and Colorado squawfish to temperatures approximating those of the Colorado River in Grand Canyon was examined by comparing growth rates among treatment and control temperatures. Following incubation at 18.6°C and acclimation to 20°C after swimup, fishes were transferred to aquaria maintained at 10.0°C, 14.0°C, and 20.0°C  $\pm$  0.5°C. Experimental groups consisted of: 1) 6-8 d humpback chub reared for 30 d; 2) 13-15 d humpback chub reared for 24 d; 3) 39-41 d humpback chub reared for 93 d, and; 4) 13-15 d Colorado squawfish reared for 92 d. Two aquaria of 56 individuals each were maintained for each temperature for groups 1 and 2, three aquaria of 10 individuals were maintained for each temperature for group 3, and three aquaria of 50 individuals were maintained for each temperature for group 4.

All larval fish were fed Bio-kyowa B-250, *Artemia salina*, and zooplankton *ad libitum* at 0600, 0900, 1200, 1500, and 1700 hrs. Older fish were fed commercial trout feed. Waste feed and excrement were siphoned from all tank bottoms every other day. Dissolved oxygen was monitored daily with a YSI meter and maintained at 90-100% saturation. Ammonia (NH<sub>3</sub>-N), nitrite (NO<sub>2</sub>-N) and nitrate (NO<sub>3</sub>-N) nitrogen were monitored with Chemetrics titration kits to assess the effectiveness of the chemical (zeolite) and biological filters. Adjustments to filtration were made to maintain ammonia at <0.1 ppm, nitrite <0.1 ppm, and nitrate <4 ppm. pH was maintained at 8-9.

Approximately every 7 d, study individuals from groups 1 and 2 were anesthetized in a solution of 125 ppm MS-222, placed on blotter paper for 5 s, and weighed to the nearest 0.0001 g on a Mettler H80 balance. Total lengths were determined to the nearest 0.1 mm with a micro grid. Group 3 fish lengths and

weights were measured at 14 d intervals with a micro grid and an Ohaus CT200 electronic balance (to  $\pm 0.01$  g). Five fish were sacrificed from each aquarium from group 4 fish every 15 d for length (as above) and weight ( $\pm 0.0001$  g) measurements.

Treatment of the protozoan parasite *Ichthyobodo necatrix* (costiasis) was with formalin at 25 ppm for 24 h, followed by Chloramine T the next day at 9 ppm for 1 h. Prophylactic treatments to older fishes (groups 3 and 4) were made every 7 d by adding 2 ppm 5% methylene blue solution. Nine ppm Chloramine T was added every 30 d, and flushed after 1 h.

## RESULTS

### *Spawning and Incubation*

A total of 75 humpback chub >200 mm TL (32 females, 43 males) was captured and held in live cars for spawning and propagation efforts on the LCR from 26 April-7 May, 1993. Twenty-eight females >250 mm were injected intraperitoneally with carp pituitary on 1-3 consecutive days, and 11 males were similarly treated. One female that was ripe at the time of capture was stripped of approximately 200 eggs and released. Three additional females 250 mm or less in length were released without injection. Thirty-two males were ripe at the time of capture. No mortalities were observed resulting from the propagation procedures.

Approximately 2,800 eggs were manually stripped from 10 hormone-injected females; four females expressed eggs on two consecutive days. Since bentonite was not added to the egg/sperm mixture, eggs were adhesive to each other and to screens of the hatching trays, preventing precise determination of numbers. Siltation on eggs in the instream hatching box (-200 eggs) was high and the apparatus was discontinued after the first trial. Egg development within aerated 19 L buckets with desilted LCR water appeared normal.

Turbidity of the LCR on 26 April was 282 NTU, and slowly declined to 89 NTU by 7 May. Turbidity within hatching buckets of desilted LCR water was 12 NTU. Maximum temperature of the LCR during this period was 20.4°C and the minimum was 17.0°C. Temperatures in hatching buckets fluctuated approximately 1°C greater in both directions than those in the LCR. Dissolved oxygen in the LCR ranged between 6.3 and 8.2 mg/l and 76.5-97.6% saturation; full saturation was maintained within aerated hatching buckets. Conductivity in the river increased from near 2800 :S on April 26 to 3800 :S on May 7, with similar levels maintained in buckets. pH fluctuated between 7.7-8.2 in the LCR, with hatching bucket values similar.

At Bubbling Ponds Hatchery following transport from the LCR, approximately 500 eggs appeared damaged or broken. The approximately 200 eggs that were covered with silt succumbed, 1,000 others were killed by fungus, and 1,100 hatched. Hatching

took place on day 6 (25%), day 7 (50%), and was completed 8 d post-fertilization. Temperature, pH, and conductivity of incubation water were nearly constant at 18.6°C, 7.6, and 365 :S, respectively. Dissolved oxygen levels fluctuated between 4.3 and 7.3 mg/l.

### *Temperature Experiments*

All 5-7 d humpback chub larvae transferred from 20°C to 10°C immediately entered "cold coma", a condition characterized by an inability to maintain equilibrium and position in the water column. Fish drifted in the current until settling to the bottom of the tank. They remained immobile for approximately 90 min. Over the next 60 min they regained their ability to swim in the water column but appeared lethargic compared to the control groups at 20°C. Observations of heart rate in these fish indicated that rates slowed from 100-120 beats/min at 20°C to 30-40 beats/min at 10°C. Larvae transferred from 20°C to 12°C became lethargic but did not lose their ability to remain in the water column. No behavioral effects were noted for 5-7 d larvae transferred from 20°C to 14°C.

Eleven to thirteen day old humpback chub larvae transferred from 20°C to 10°C also entered cold coma, but regained normal behavior after 15 min. All other temperature treatment groups at this age exhibited no obvious adverse behavioral effects. No mortalities for either age group occurred during the 4 h observation periods.

Replicate groups of 6-8 d humpback chub larvae transferred from 20°C to 10°C for determination of growth effects gained 10% in length and 28% in weight over 30 d (Table 1). These values compared to 37% and 195% length and weight gains over 30 d at 14°C, and 83% and 951%, respectively, at 20°C. Weight gains failed to keep pace with increases in length at 10°C, and fish appeared emaciated. Based on nonoverlap of 95% confidence intervals (Figure 1), the differences in length and weight after 30 d were significantly different among all combinations of the three temperatures.

Similar experiments with 13-15 d humpback chub larvae produced like results, although differences in length and weight at 10°C and 14°C were not significant (Table 1). These results suggest that growth rates of older fish were less affected than those of younger fish by the coldest temperature.

Tolerance experiments with 39-41 d humpback chub demonstrated that temperature had significant effects on growth of juvenile life stages (Table 1, Figure 1). After 93 d, humpback chub gained 24% in length and 71% in weight at 10°C, compared to 148% and 1,236% length and weight gains, respectively, at 20°C. Gains in length and weight after 93 d for chub held at 14°C were 67% and 286%, respectively. These growth trends were also evident with Colorado squawfish based on 92 d temperature treatments on 14 d larvae (Table 2, Figure 2). Final

length at 20°C was 263% greater than initial length, 100% greater at 14°C, and 17% greater at 10°C. Weight gains over 92 d at 20°C were 7,281%, 880% at 14°C, and 71% at 10°C. The relatively static growth between day 42 and day 59 (Figure 2) was a result of an outbreak of costiasis. Black and Bulkley (1985) reported that length and weight gains of yearling Colorado squawfish over 84 d were approximately three times greater at 20°C than at 15°C.

## DISCUSSION

We previously failed in attempts to propagate eggs from humpback chub in the field. We are unable to definitively determine reasons for previous failures based on comparisons with successful procedures utilized in 1993, but we suspect that use of commercial bentonite clay (to prevent clumping of eggs following fertilization) in earlier efforts was a possible factor. Complex adsorption interactions between differently charged clay particle types (i.e. bentonite and LCR clays) on the surfaces of eggs may have interfered with the fertilization process or oxygen uptake by eggs. Low suspended sediment loads (i.e. clay) typical of hatchery waters presumably allows successful use of bentonite in fish culture facilities. Because of the low number of eggs collected in 1993, we were unable to compare propagation results with and without bentonite treatments. This potential avenue of failure requires additional investigation. We consider other differences in methods among trials relatively minor, and insufficient to account for total failures of early attempts.

Although similar numbers of fish were handled, approximately an order of magnitude fewer eggs were obtained in 1993 compared with 1992 field propagation efforts (Hines 1993, Clarkson and Robinson 1993). This difference illustrates the tremendous logistical difficulties involved with field propagation efforts at remote sites such as the LCR, especially when using experimental procedures on endangered fishes. Annual variability of water discharge, water temperature, fish catchability, and reproductive condition of fishes combine to render such operations unreliable and expensive. We recommend that further attempts at field propagation of Grand Canyon native fishes generally be abandoned in favor of hatchery culture. A genetic breeding program could then be generated (Kapusinski and Philipp 1988), culture methods further refined, and prophylactic treatments and schedules developed. Progeny from hatchery brood stock can then be dependably available for experimental purposes or for reintroductions, and adults can serve as refugia individuals in the event of catastrophe in the wild.

Our experimental study design attempted to simulate temperature conditions experienced by native fishes within the Grand Canyon riverine system. Spawning and early development of native fishes occurs in the LCR and other warmwater Grand Canyon tributaries at temperatures near 20°C. At various stages within

their life histories, fish may enter the mainstem Colorado River, where temperatures typically are 10-12°C. A large number of most native species enter the mainstem as larvae through entrainment or active drift (Robinson et al. 1994), while others are flushed into the mainstem during summer floods as early juveniles (AGFD 1993, Hendrickson 1993). Mixing between warm tributary water and cold mainstem water may occur for some distance downstream. We attempted to approximate this mixing zone with our experimental 14°C temperature.

Assuming no temperature amelioration through mixing of tributary and mainstem waters, our results indicate that 5-7 d and 13-15 d larval humpback chub that enter the mainstem Colorado River at temperatures near 10°C from the LCR will immediately enter cold coma for 15-90 min. Similar cold shock experiments conducted on 14 d Colorado squawfish by Berry (1986) indicated that mortality and cold coma occurred with temperature changes from 22°C to 7°C over 5 min, and that some cold coma occurred, activity levels were reduced, and other behavioral modifications noted with changes from 22°C to 12°C. Our experiments on 5-7 d humpback chub larvae moved from 20°C to 12°C also noted behavioral differences (lethargy) compared to controls, but no effects were noted with older larvae at 12°C, nor with any group at 14°C. Several researchers determined that temperature effects were lessened with slower rates of temperature change (Speakman and Kenkel 1972, Griffith 1978, Burton et al. 1979, Berry 1986) and with older fish (Pitkow 1960, Nickum 1966, Berry 1986).

The effects of entering cold coma in the Colorado River are potentially severe. Rates of predation may be increased (Coutant et al. 1974), physical damage and death may occur from abrasion against the substrate or entrainment in extreme current velocities and turbulence, or fish may be buried if they settle on the substrate. Muscle performance is likely reduced (Webb 1993).

Juveniles that enter the mainstem, and larvae that survive the immediate effects of cold coma or do not experience cold coma, are subject to greatly reduced growth rates. It is likely that growth rates in the wild (Colorado River) are even poorer than our results suggest when competition and other environmental stresses are considered. Effects of reduced growth rates include increased early-life mortality and decreased survival to sexual maturity (Kaeding and Osmundson 1988), reduced condition, lipid stores, and size that result in elevated overwinter mortality for young-of-year fishes (Thompson et al. 1991), lowered egg production by adults (McAda and Wydoski 1983), and other less studied effects (Kaeding et al. 1986).

These findings indicate that benefits are accrued to individual fish and populations by remaining in Colorado River warmwater tributaries as long as possible. We recommend that the historic pattern of high springtime discharges in the Colorado River be simulated through releases from Glen Canyon Dam. This flow pattern will impound tributary mouths and form slow

velocity, warm, and productive refugia for rearing of early life stage, tributary-spawned native fishes, and potentially reduce losses to the mainstem. In addition, we advise a reduction in fluctuations of daily flows in the Colorado River to allow greater warming of backwaters and potentially other low velocity mainstem native fish rearing areas. Finally, we advocate that thermal modification of dam releases be considered to reduce detrimental effects of low temperatures on native fishes in Grand Canyon.

TABLE 1. Initial (day 0) and final length-weight statistics of pooled replicate data for temperature tolerance experiments with variable aged humpback chub reared at three different temperatures.

Initial age: 6-8 d post-hatching

Day	°C	n	Length			Weight		
			Mean	SD	Range	Mean	SD	Range
0	20	20	9.5	0.3	9.3-9.8	3.9	0.5	3.5-4.5
30	10	10	10.5	0.4	10.0-11.0	5.0	0.7	3.6-6.0
30	14	21	13.0	0.6	12.0-14.0	11.5	3.1	8.0-16.0
30	20	13	17.4	1.8	15.0-21.0	41.0	14.0	23.0-73.8

Initial age: 13-15 d post-hatching

0	20	20	11.2	0.6	11.0-12.0	5.0	0.7	4.5-6.0
24	10	60	14.1	0.8	11.0-15.0	18.4	4.2	6.5-24.9
24	14	30	14.3	3.7	13.5-17.6	20.9	4.6	13.5-27.4
24	20	30	19.7	1.1	18.0-22.0	57.2	13.8	36.0-84.4

Initial age: 39-41 d post-hatching

0	20	15	20.9	1.4	19-23	84.8	22.5	54-126
93	10	24	26.0	1.7	24-29	145.4	27.8	100-200
93	14	28	34.8	1.7	32-38	327.5	44.8	260-410
93	20	25	51.9	4.3	46-60	1136.0	216.5	850-1510

TABLE 2. Initial (day 0) and final length-weight statistics of pooled replicate data for temperature tolerance experiments with 13-15 d post-hatching Colorado squawfish reared at three different temperatures.

Day	°C	n	Length			Weight		
			Mean	SD	Range	Mean	SD	Range
0	20	10	10.0	0.5	9.0-11.0	4.5	0.4	4.1-4.9
92	10	21	11.7	0.6	11.0-13.0	7.7	1.0	5.0-8.9
92	14	30	20.0	0.4	19.5-21.0	44.1	4.3	36.6-53.5
92	20	25	36.3	2.4	32.0-40.0	329.3	69.4	170.0-440.0

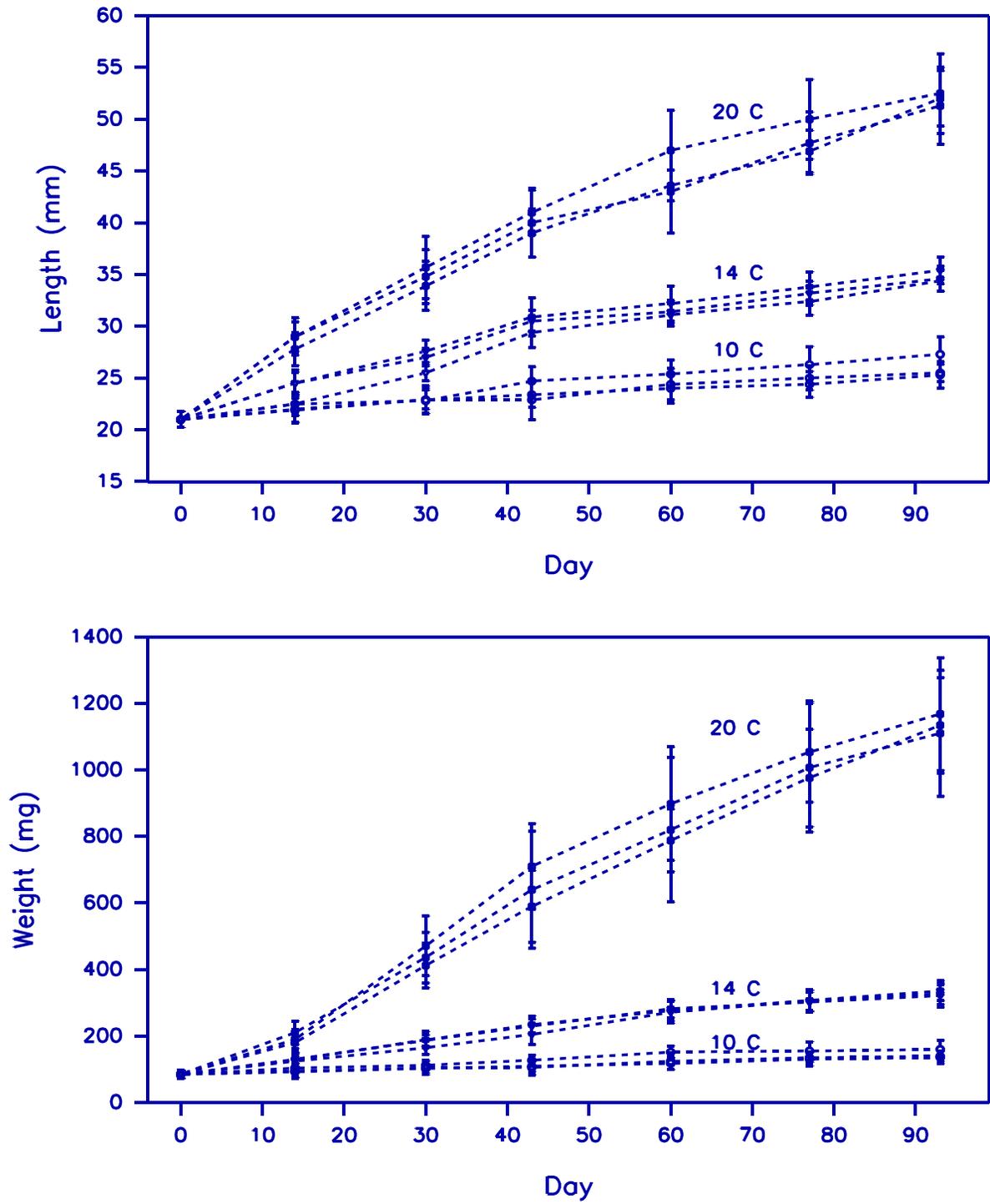


FIGURE 1. Mean lengths and weights of replicate groups of humpback chub transferred from 20°C tanks as 39-41 d old juveniles to 10°C (circles), 14°C (triangles), and 20°C (squares; control) tanks over 93 d. Vertical bars represent 95% confidence limits.

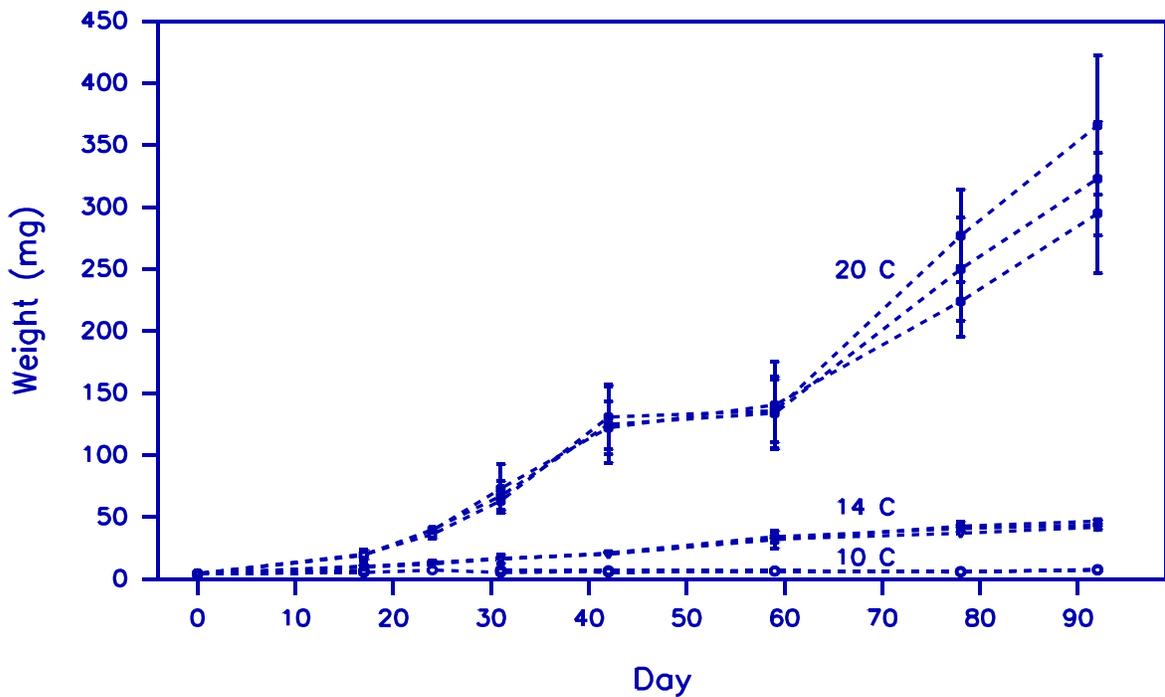
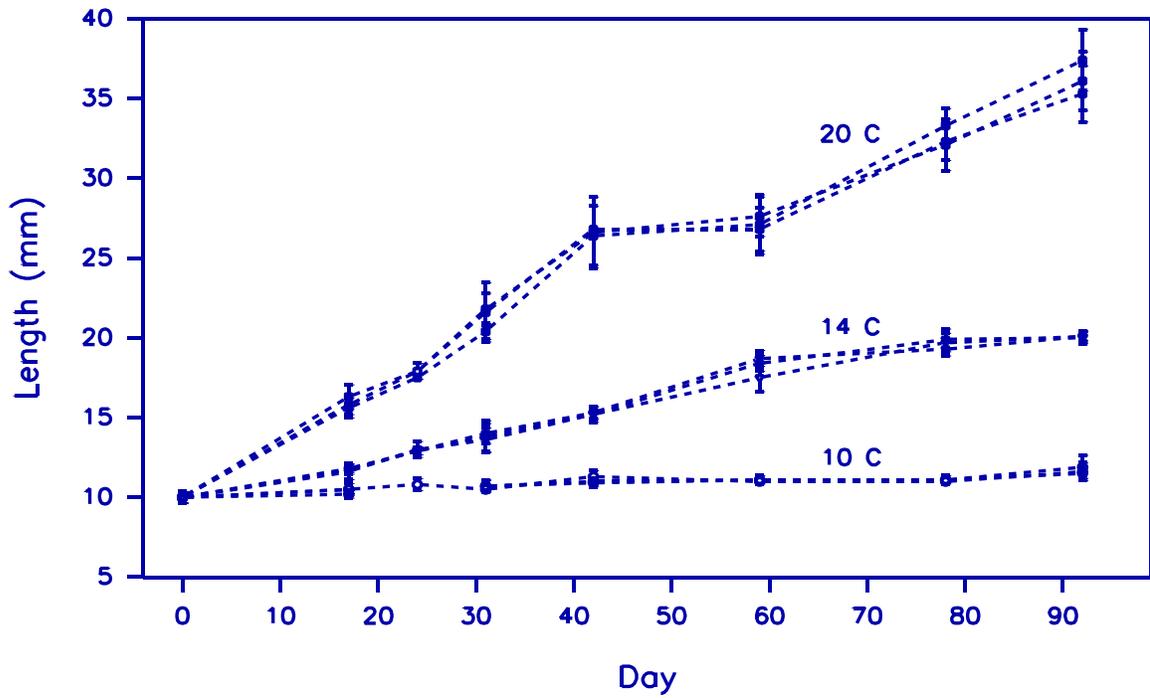


FIGURE 2. Mean lengths and weights of replicate groups of Colorado squawfish transferred from 20°C tanks as 14 d old larvae to 10°C (circles), 14°C (triangles), and 20°C (squares; control) tanks over 92 d. Vertical bars represent 95% confidence limits.

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