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Effect of incubation temperature on green sturgeon embryos, *Acipenser medirostris*

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Synopsis

Regulation of river flow and the amount of winter rainfall are the major factors affecting the water temperature of the spawning grounds, for green sturgeon in the Klamath River. During the primary spawning period of green sturgeon, mid-April to June, the water temperature may vary from 8 to 21°C. To estimate the potential implications of this modified thermal regime, we examined the survival and development in three progeny groups of green sturgeon embryos from zygote to hatch, at constant incubation temperatures (11–26°C). Temperatures 23–26°C affected cleavage and gastrulation and all died before hatch. Temperatures 17.5–22°C were suboptimal as an increasing number of embryos developed abnormally and hatching success decreased at 20.5–22°C, although the tolerance to these temperatures varied between progenies. The lower temperature limit was not evident from this study, although hatching rate decreased at 11°C and hatched embryos were shorter, compared to 14°C. The mean total length of hatched embryos decreased with increasing temperature, although their wet and dry weight remained relatively constant. We concluded that temperatures 17–18°C may be the upper limit of the thermal optima for green sturgeon embryos, and that the river thermal regime during dry years may affect green sturgeon reproduction.

Introduction

The green sturgeon, *Acipenser medirostris*, is an anadromous species inhabiting the North American Pacific Ocean, from the Aleutian Islands to California. Despite its wide geographic distribution and some significant commercial landings (Houston 1988), the green sturgeon is considered a vulnerable species in the United States and Canada (Moyle et al. 1994, Campbell 1997). A closely related Asian green sturgeon (Sakhalin sturgeon, *A. mikadoi*) is similar in morphology (North et al. 2002) but was recently separated as a species based on the cell DNA content and cytochrome-b gene sequence (Birstein & DeSalle 1998). The Sakhalin sturgeon is an endangered species in Russia and Japan, but the 2001 petition to list the American green sturgeon under the U.S. Endangered Species Act was unsuccessful. After reviewing the best available scientific and commercial information for green sturgeon the National Marine Fisheries Service determined that listing the green sturgeon as threatened or endangered is not warranted, at this time.[1]

Unfortunately there has been a scarcity of research on green sturgeon life history, habitat, and stock abundance. Although some recent studies

[1] U.S. Federal Register Vol. 68, No. 19, pp. 4433–4441; 29 January 2003.

have provided new information on green sturgeon karyotype (Van Eenennaam et al. 1999), reproductive biology (Van Eenennaam et al. 2001, Deng et al. 2002), adult movement (Erikson et al. 2002), and physiology (Gisbert et al. 2001, Gisbert & Doroshov 2003, Lankford et al. 2003), many aspects of green sturgeon biology remain obscure. Knowing the temperature tolerance of green sturgeon embryos may assist in protection of this species.

Reproduction and stock recruitment are the most vulnerable phases of sturgeon life history (Dettlaff et al. 1993). Green sturgeon spawn in the Klamath, Rogue, and Sacramento Rivers, all of which have flow regimes affected by water projects (Moyle et al. 1994). The Klamath River is a major site of green sturgeon reproduction in the Pacific Northwest (Moyle et al. 1994, Van Eenennaam et al. 2001) and the spawning run starts in April and extends through June, with the peak and duration dependent on river flow and water temperature. Seven hydroelectric dams and numerous irrigation diversions built between 1906 and 1962 greatly affect the river flow and anadromous fish migration, particularly during the 'dry' years (Service 2003).

The variation in annual precipitation and flow regulation results in large fluctuations of river temperature (10–23°C) during the spawning run of green sturgeon (unpublished data for the Klamath River at Weitchpec, California). This has raised concerns regarding the survivability of green sturgeon embryos and the effect of controlled flow on natural recruitment.

In laboratory studies with sympatric white sturgeon, *A. transmontanus*, temperatures between 14 and 16°C were optimal and temperatures above 20°C were lethal for the embryos during development from fertilization to hatch (Wang et al. 1985, 1987). Studies with the Caspian beluga, *Huso huso*, ship, *A. nudiventris*, Russian sturgeon, *A. gueldenstaedtii*, and sevruga, *A. stellatus*, revealed generally similar temperature effects on the early life stages (Nikol'skaya & Sytina 1978, Igumnova & Dubinin 1987, Sytina & Shagaeva 1987). Observations on natural spawning, documented by the collection of eggs and larvae, indicate that the majority of sturgeons reproduce within the temperature range of 10–20°C, including Eurasian (Dettlaff et al. 1993) and North American (McCabe & Tracy 1994, Bruch & Binkowski 2002, Perrin et al. 2003) species.

The successful spawning and early development form the basis for sturgeon recruitment. Since the temperature range permitting normal development is critical in fish reproduction, the primary objective of this research was to determine the effects of egg incubation temperature on green sturgeon embryo survival to hatch. This study provides the first information on development and survival of green sturgeon embryos incubated, from zygote to hatch, at different constant temperatures under laboratory conditions.

Materials and methods

Broodfish and spawning

We obtained mature green sturgeon on 12 May 2000 (one female and two males, river temperature 12–13°C) and on 26 April 2002 (two females and two males, 12–14°C) from the Yurok Tribe gillnet fishery in the lower Klamath River (Van Eenennaam et al. 2001). After 1–5 days holding in river cages, we transported the fish to the Center for Aquatic Biology and Aquaculture (CABA, University of California, Davis) and placed them for spawning in 4 m diameter flow-through tanks. We also collected semen from one naturally milting male in 2002 and stored it in plastic bags with oxygen on wet ice. We weighed brood fish ($\pm$0.1 kg), measured their length ($\pm$0.5 cm), and collected small samples of ovarian follicles ($n = 15$–20) by catheter to determine oocyte diameter and polarization index, PI (Van Eenennaam et al. 2001), using a dissecting scope with camera lucida and a digital image-analyzing tablet with computer interface ($\pm$0.01 mm).

The hormonal induction of spawning followed the methods of Van Eenennaam et al. (2001), with slight modifications. Males received a single injection (10 $\mu g\ kg^{-1}$) of mammalian GnRHa [D-Ala^6, Des-Gly^{10}] – LH-RH Ethylamide (Peninsula Laboratories, Belmont, California). An additional male green sturgeon, captured in the Sacramento River as a yolk sac larva and reared to full sexual maturity at the CABA facilities, was induced to spermiate in 2002. The females received two GnRHa injections: priming (1 $\mu g\ kg^{-1}$) and

resolving (19 μg kg^{-1}), with a 12-h interval. In addition, the females received 1–3 mg kg^{-1} of the dopamine antagonist domperidone (Research Diagnostics, Flanders, New Jersey). All injections were intramuscular and given underwater to minimize handling stress.

Broodfish holding temperature during spawning ranged 13.1–14.6°C. We periodically examined the female holding tank for released eggs, starting 12 h after the second injection. We collected milt with a 60 ml plastic syringe and a 4 cm long vinyl catheter inserted into the urogenital pore (Conte et al. 1988) and sperm was evaluated for the percent and duration of motility (Van Eenennaam et al. 2001). We extracted the ovulated eggs surgically (Conte et al. 1988), from fish under anesthesia (MS-222).

Artificial fertilization followed techniques developed for white sturgeon (Conte et al. 1988). We fertilized ovulated eggs in bowls, using 4 l of diluted semen (1:100 or 200 with hatchery water) per 1 l of ova. After fertilization, we gently mixed the eggs in a suspension of Fuller's Earth (100–200 mesh size, Sigma Chemical Co., St. Louis, Missouri) for de-adhesion, and two subamples (ca. 350 eggs) of each progeny were incubated at 15.5°C (Deng et al. 2002) to determine 'control' fertility at the 4-cell stage (Dettlaff et al. 1993).

We incubated small batches of eggs in 15 l tanks (28 cm diameter, 35 cm deep, flow rate 1.5–2.0 l min^{-1}) housed in six temperature-controlled recirculation systems each with a YSI thermostat, chiller, heater, biological filter, and aeration. During the course of the experiments, dissolved oxygen was 95–100% air saturation, pH ranged 8.5–8.6, and ammonia nitrogen did not exceed 0.05 ppm. Temperature variation in each system, measured twice a day with a certified calibrated thermometer (National Institute of Standards and Technology), did not exceed ±0.1°C. We used artificial photoperiod 16L: 8D and protected the tanks with shade-cloth covers that blocked 80% of the light. Two experiments were conducted, one during 2000 (range-finding) and another in 2002 (upper temperature tolerance).

Experiment 1 (2000)

We incubated the eggs of one female, fertilized at 14°C with pooled semen of two Klamath males, in six treatments (11, 14, 17, 20, 23, 26°C) with four replications. The 60 ml egg batches (ca. 700 eggs) were placed in 1.5 l half-filled crystallizing dishes floating in the 14°C treatment tanks. We transferred dishes stepwise to warmer or cooler treatments, at a rate of temperature change of 4°C h^{-1}, and submerged them on the tank bottoms. Each tank received flowing water (1.5 l min^{-1}) via tygon tubing positioned at the inner edge of the crystallizing dish, ensuring continuous exposure of the embryos to fresh water. We removed and counted the unfertilized (not cleaving) and dead (opaque) eggs at stages 5–6 (4–8 cells), 22–23 (closure of neural tube), and 36 (hatch, stages as in Dettlaff et al. 1993). We recorded the time of 50% hatch, counted the hatched embryos, and sampled 10 embryos from each tank for length (±0.01 mm), live weight (±0.1 mg), and dry weight (±0.01 mg, dessicated 15 h at 100°C). The length of bent larvae (prevalent disorder at higher temperatures) was measured by tracing specimens along the curved notochord with the light cursor of the digital image-analyzer. Due to a technician error, the embryos hatched in replicate tanks were pooled within each treatment, thus we recorded the proportions of abnormal embryos in pooled samples.

Experiment 2 (2002)

We incubated the eggs of two females in six treatments (16, 17.5, 19, 20.5, 22, 23.5°C) with five replications for each progeny. The eggs of Female 1 were fertilized with semen of one Klamath male with the best sperm motility, and the eggs of Female 2 with pooled semen of three males (from Klamath and Sacramento rivers). The egg transfer, incubation, and sampling were similar with Experiment 1, except for lower egg densities in crystallizing dishes (20 ml egg volume, ca. 240 eggs per tank) and a fertilization temperature of 16°C. We recorded percent abnormalities in each tank and photographed representative hatched embryos under a dissecting scope.

Data analyses

We used JMP statistical software (Version 4, SAS Institute, Cary, North Carolina) for data analysis. The arcsine transformation of percent survival data was necessary to normalize distributions. We

transformed zero percent survival data as an arcsine $(1\ 4n^{-1})^{0.5}$, where 'n' was the number of stocked eggs (Zar 1984). The normality of transformed data was confirmed by the Shapiro–Wilk test. We used two-way ANOVA with repeated measure (tanks within the treatment) and Tukey HSD test to test the effect of temperature on survival to cleavage, neurulation and hatching. The egg source (female) effect was included for Experiment 2. One-way ANOVA and Tukey test were used for all other data. Pooled proportions of the abnormal embryos in Experiment 1 were compared by contingency tables. The accepted significance level was $p < 0.05$. Data in figures and text are untransformed means and standard deviations.

Results

Spawning

Mature females were significantly larger than males (student *t*-test, for both years of observations combined) (Table 1). The fully grown oocytes of three females ranged 4.17–4.45 mm in mean diameter and 0.03–0.04 in PI (distance from the germinal vesicle to the animal pole is 3–4% of egg diameter). These low PI values indicate the advanced stage of oocyte development and maturational competence in acipenserids (Dettlaff et al. 1993). All broodfish responded to hormonal treatment, with a latent period from the resolving injection to ovulation of 14–17 h. The number of ova collected from each female ranged 88–133 thousand. All males had milt with a high percentage of motile sperm (>80%) and extended duration of motility (>4.5 min), although the milt stored for 24 h on ice and the fresh milt collected from the domestic male had about 10% less motile sperm and 1–2 min shorter motility. Mean fertilization rates in controls were 41% in 2000, and 56% (Female 1) and 62% (Female 2) in 2002.

Experiment 1 (2000)

Due to the difficulty of sorting through the large quantity of eggs in each tank, only the obvious unfertilized eggs (with no cleavage at all) were counted as 'mortalities' at the 4-cell stage, and the eggs with mosaic (part of the egg does not divide) and parthenogenetic (activated unfertilized egg with irregular furrows) cleavage (Dettlaff et al. 1993) were kept in each tank until neurulation. As a result, cleavage survival was not estimated properly (ranged 64–69% in the experiment, compared to 41% in the fertilization control) and was excluded from data analysis.

ANOVA revealed a significant ($p < 0.0001$) effect of incubation temperature on survival to neurulation and hatching; 36–40% of embryos (a majority of fertilized eggs) survived to neurulation in temperatures 11–20°C, but only 16% survived in 23°C, and none of the embryos survived in the 26°C treatment (Figure 1), where the cleavage was abnormal and ceased before gastrulation. Survival to hatch was the highest in the 14 and 17°C treatments (39 and 36%) and lowest in the 11 and

Table 1. Reproductive characteristics of green sturgeon broodstock used for spawning.

Date of capture	12 May 2000		25–26 April 2002		
Sex (F/M)	F	M (n = 2)	F1	F2	M (n = 3)
Fork length, cm	166	156 ± 2	185	179	148 ± 7
Weight, kg	38	24 ± 1	50	52	24 ± 4
Oocyte PI, ratio	0.03	–	0.04	0.03	–
Oocyte D, mm	4.17	–	4.34	4.45	–
Latent period, h	17	–	14	14	–
Ova collected, $\times 10^3$	88	–	113	109	–
Sperm motility,%	–	95 ± 3	–	–	89 ± 7
Motility duration, s	–	387 ± 9	–	–	320 ± 60
Control fertility,%	41 ± 3	–	56 ± 1	62 ± 1	–

Data for males are means and SD. The oocyte polarization index (PI) and diameter (D) are means of 15–20 eggs. Latent period is time from the resolving GnRHa injection to ovulation. Control fertility was based on two subsamples at the 4–8 cell stage.

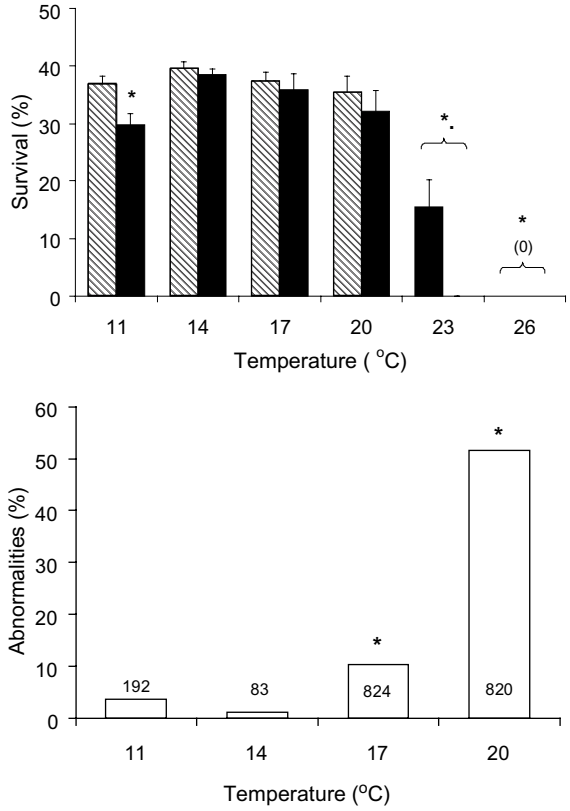

Figure 1. Top: survival (mean, SD) of green sturgeon embryos at neurulation (shaded bars, n = 4) and hatch (solid bars, n = 4) in Experiment 1. Asterisks indicate significantly different sample means, for stages among the treatments ($p < 0.05$). Bottom: proportions of hatched abnormal embryos in pooled samples (asterisk denote different proportions, $p < 0.05$, numbers are total counts).

20°C treatments (30 and 32%), with detected significant difference in hatch rate between the 11 and 14°C treatments. No embryos survived to hatch at 23°C. The proportions of deformed hatched embryos were low at 11 and 14°C (3.7 and 1.2%) but increased at 17 and 20°C (10.3 and 51.6%, Figure 1). The deformities were similar to those observed in the 2002 experiment.

Embryos hatched in the 11–20°C treatments did not differ in dry weight (15.0–15.1 mg), but were significantly longer at 17 and 14°C (12.5 ± 0.29 and 11.8 ± 0.12 mm) compared to the 11 and 20°C treatments (10.9 ± 0.08 and 11.0 ± 0.07 mm). The percent dry weight was highest at 14°C (46.8% ± 0.46) and lowest at 20°C (44.1% ± 0.47, significantly different).

Experiment 2 (2002)

Lower egg densities in this experiment facilitated identification and accurate counts of unfertilized eggs. The percent of cleavage in treatments 16–22°C were similar to control fertilization rates (52–56% vs. 56% control for progeny 1, and 60–62% vs. 62% control for progeny 2). ANOVA revealed a significant interaction between the treatment and egg source ($p < 0.0001$), with a higher survival to neurulation and hatch at 20.5 and 22°C in the progeny of Female 2. The effect of treatment was analyzed separately for each progeny (Figure 2).

The incubation at 23.5°C decreased cleavage survival and resulted in 100% mortality before neurulation in both progenies. Treatment 22°C decreased neurulation survival in progeny 1 and survival to hatch in both progenies, with the only 0.3% hatch in progeny 1 (all deformed embryos) and 32% in progeny 2 (78.4% deformed embryos).

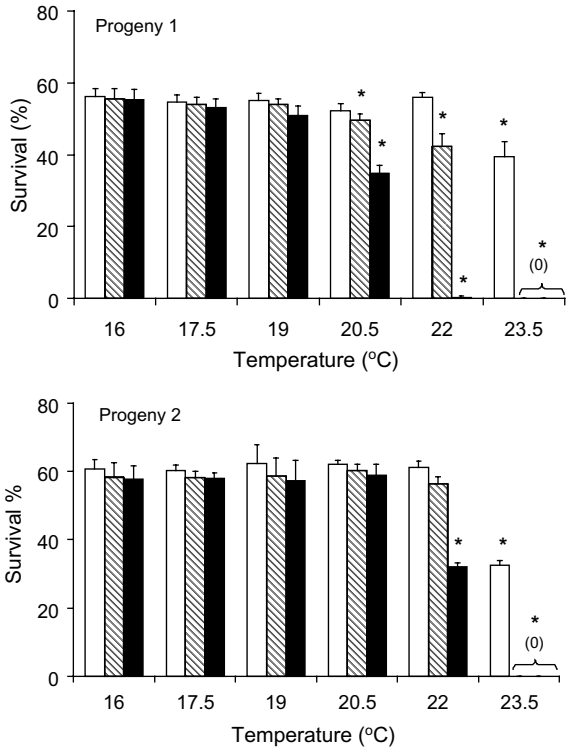

Figure 2. Survival (mean, SD) of green sturgeon embryos at cleavage (open bars, n = 5), neurulation (shaded bars, n = 5), and hatch (solid bars, n = 5) in Experiment 2. Asterisks denote significantly different treatment means ($p < 0.05$).

Treatment 20.5°C decreased survival to neurulation and hatch in progeny 1, but not in progeny 2. The proportions of deformed hatched embryos increased with temperature in both progenies, and were higher in progeny 1 (Figure 3). In both progenies, the highest hatching success (56.2 and 60.6%) and the lowest proportions of deformed embryos (1.0 and 0.3%) were in 16°C treatment. The abnormalities at the elevated temperatures were predominantly (>80%) body deformations, such as the bent and shortened posterior trunk and tail regions, including lordosis and kyphosis; edema and the more severe developmental defects such as an underdeveloped head, trunk, and tail regions were less common (Figure 4).

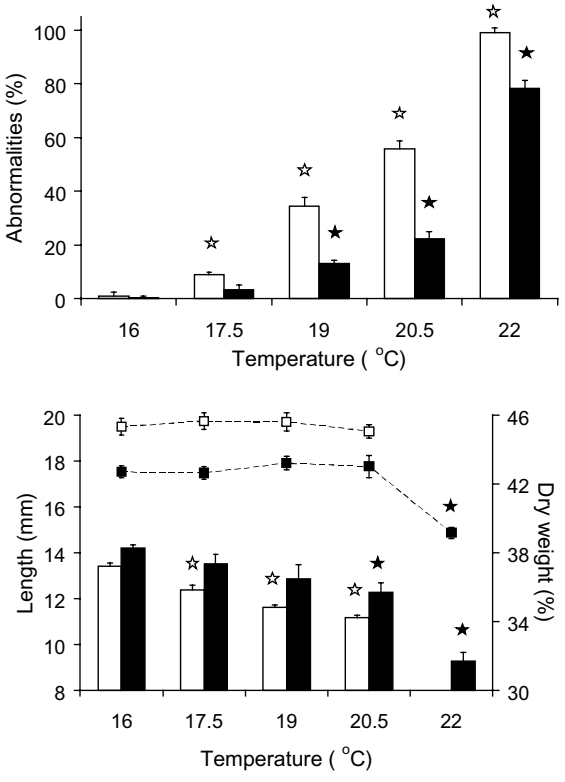

Figure 3. Top: proportions (mean, SD) of hatched abnormal green sturgeon larvae in Experiment 2, for progenies 1 (open bars, n = 5) and 2 (solid bars, n = 5). Stars indicate significantly different ($p < 0.05$) means among the treatments within each progeny. Bottom: hatched larvae length (mean and SD, open and solid bars for progenies 1 and 2, n = 5) and dry weight (open and solid squares for progenies 1 and 2, n = 5). Stars denote different treatment means within each progeny ($p < 0.05$).

In progeny 1, the length of hatched embryos decreased at higher temperatures, from 13.4 mm at 16°C to 11.2 mm at 20.5°C. The live weight (40.0–40.5 mg) and percent dry weight (45.1–45.8%) were not affected by temperature. A similar relationship was observed in progeny 2 within the range of 11–20.5°C, but the embryos hatched at 22°C were shorter (9.3 mm) and had a lower (39.2%) dry weight (Figure 3). The hatchery control larvae (pooled progenies) were similar in length (13.9 ± 0.12 mm), live weight (40.4 ± 0.28 mg) and dry weight (45.1 ± 0.28%), to the 16 and 17.5°C treatment larvae. The longer larvae in the female 2 progeny were likely due to the larger egg size in this female (Table 1).

The relationship between median time of hatch and incubation temperatures 11–22°C (observations from both experiments) was best represented by an exponential equation (Figure 5) and was similar to the relationship for stage 35 (beginning of hatch) of white sturgeon reported by Wang et al. (1985).

Discussion

We provide the first information on the effect of incubation temperature on green sturgeon embryos. Temperatures 23°C and above resulted in total mortality before hatch. Suboptimal temperatures (17.5–22°C) reduced the number of normal embryos and decreased hatching success, but the responses to suboptimal temperature varied between progenies. This study did not reveal the low temperature limit for green sturgeon embryos, but the lowest experimental temperature of 11°C decreased hatching success and produced smaller embryos at hatch. Our study suggests that green and white sturgeon embryos have similar thermal optima, upper temperature limits and temperature-dependent rates of development (Wang et al. 1985). Given the same geographic range and spawning season of these two Pacific Coast species, these similarities are not surprising. Although white and green sturgeons may reproduce in the same river (e.g. Sacramento River), they appear to maintain reproductive isolation by selecting different spawning sites within one watershed (Deng et al. 2002). White sturgeon dominate the spawning run of the Sacramento river while the opposite

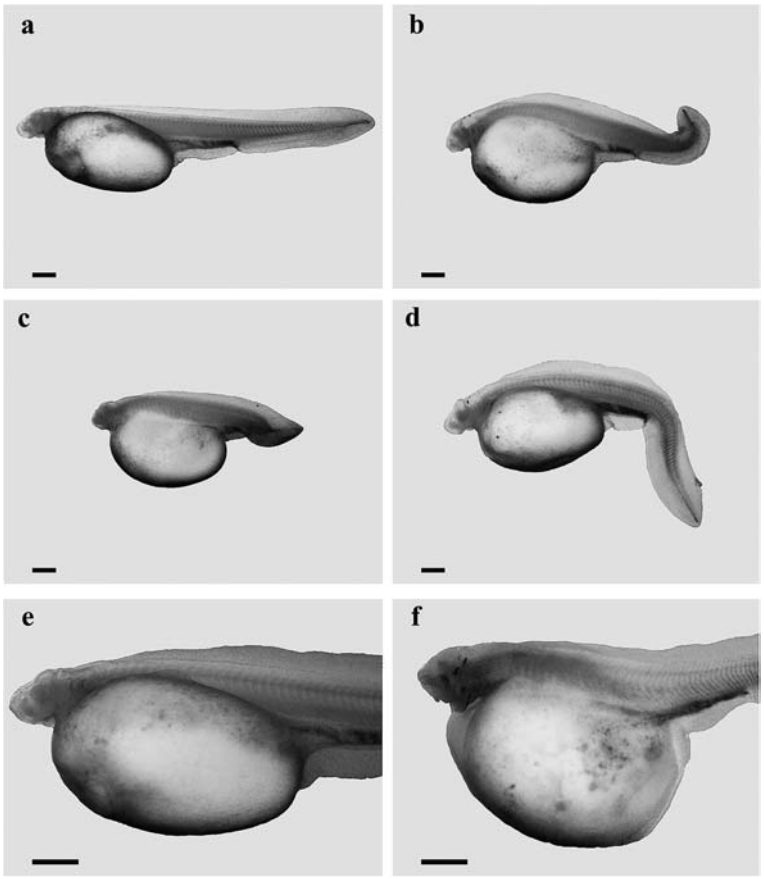

Figure 4. Representative abnormalities observed at hatch included kyphosis (b), lordosis (d), the more severe underdeveloped head, trunk, and tail regions (c), and edema (f). A representative normal embryo is included for comparison (a, e). Scale bars are 1 mm.

is true for smaller fast-flowing Klamath and Rogue rivers. There is no published information on the occurrence of hybrids between these two species, and the natural hybridization appears to be either rare or not adequately investigated (T. Rien & J. North, Oregon Department of Fish & Wildlife, D. Erickson, Wildlife Conservation Society, Oregon, personal communication). We have not investigated artificial hybridization between green and white sturgeon. However, Kolman et al. (1999) described hybrids between the Asian green sturgeon and Siberian sturgeon (*A. baerii*) produced in the hatchery.

The effect of temperature on different stages of embryo development was not fully revealed by this study, since the abnormalities at cleavage and neurulation were not characterized. Significant proportions of embryos survived to cleavage at 23 and 23.5°C, but abnormal cleavage could lead to gastrulation arrest and, consequently, mortality before neurulation (Sytina & Shagaeva 1987). Dettlaff et al. (1993) reported abnormal, mosaic cleavage in beluga (*H. huso*) eggs obtained from females maturing at the upper temperature limit for spawning. The mosaic cleavage pattern and the resulted deformed embryos were explained by the damaging effect of holding temperature on the ooplasm and the inhibition of the cortical reaction at fertilization in the areas associated with undivided portions of the egg (Dettlaff et al. 1993). The complete block of egg cleavage in acipenserids was reported at temperatures outside the spawning range, e.g. at ≥28°C in stellate sturgeon, *A. stellatus* (Sytina & Shagaeva 1987).

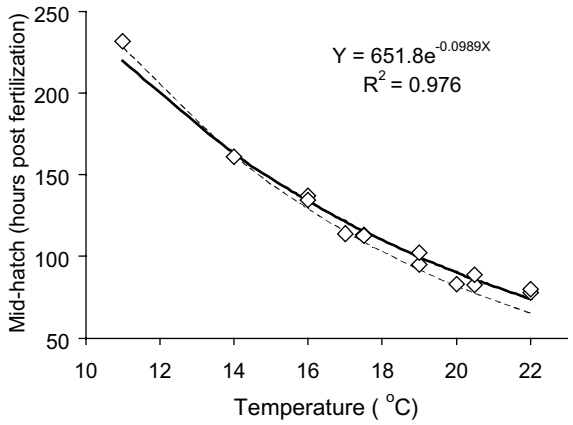

Figure 5. Rate of development to mid-hatch (hours post fertilization) of green sturgeon embryos at different temperatures. The data are from observations in both experiments, and the best fit relationship is shown by the solid line and an exponential equation. For comparison the exponential equation for white sturgeon (dashed line, Wang et al. 1985) is included.

The deformities of green sturgeon embryos hatched at elevated temperatures were similar to those described and illustrated for Russian sturgeon *A. gueldenstaedti* by Dettlaff et al. (1993). These investigators also noted the relationship between the rate of deformities and egg quality (up to 25–30% in poor quality eggs and few in high quality eggs) and the effect of high incubation temperature ('at times, higher than 26°C', Dettlaff et al. 1993, p. 138) on the increase, up to 90%, of distinct structural defects. The prevailing abnormality in our study was a curved notochord, downwards (lordosis) or upwards (kyphosis), which affected mobility of hatched embryos. The notochord development and the osmotic mechanism of its elongation, straightening and stiffening (as in the amphibian embryo, Adams et al. 1990) might be affected by high incubation temperatures. The shortening of the embryo body, decrease in percent dry weight, and edema were observed in green sturgeon embryos hatched at 22°C (Figure 3). The reduced mean length of embryos hatched at 17.5–20.5°C is more likely to reflect the effect of temperature on somitogenesis (Brooks & Johnston 1994).

Factors affecting temperature tolerance of fish embryos may include egg quality (Dettlaff et al. 1993), as well as thermal experience of the parents (Hubbs & Bryan, 1974). Embryos of *A. stellatus*, obtained from spawners during different periods of migration into the Volga River exhibited varying tolerance to upper temperature, with the differences of 2–3°C in lethal, sublethal, and suboptimal temperatures between the early and late spawning run (Sytina & Shagaeva 1987). In our study, broodfish were obtained from the same area of the Klamath River and at similar river temperatures. Egg quality was likely the main factor affecting the different responses of two progenies in the 2002 experiment, with lower egg fertility and consistently higher proportions of abnormal embryos in all temperature treatments in the female 1 offspring.

Our data show that temperatures above 20°C are most likely detrimental for green sturgeon reproduction. While the temperatures between 17.5 and 19°C may result in satisfactory hatching success, the percent of abnormal hatched embryos increases within this range and it can be hypothesized that the resulting offspring are subject to higher mortality rates. Although there are undoubtedly some females that produce embryos that can tolerate elevated river temperatures better than others, based on this study it seems that temperatures 17–18°C may be the upper limit of the optimal thermal range for green sturgeon development, wherein neither hatching success nor normal development are affected by temperature.

Klamath River temperature profiles were provided by the Yurok Tribe fishery biologists using data loggers located at approximately river kilometers (rk) 56 and 66 (Figure 6). The river temperature on the spawning grounds is affected by flow releases from the Iron Gate dam (rk 306), the amount of heating and cooling in transit to the spawning areas (below Ishi Pishi Falls, rk 107), thermal loading from other streams and rivers, and the amount of precipitation during the winter and spring. The predominant spawning period, from mid-April to the end of May (Figure 6), was based on the time when the tribal fishers collect approximately 80% of their annual catch (J.Van Eenennaam, unpublished data). In the past, the Iron Gate Dam on the Klamath River had Federal Energy Regulatory Commission (FERC) stipulated minimum flows that decreased during the time of sturgeon spawning (1300 cfs during April,

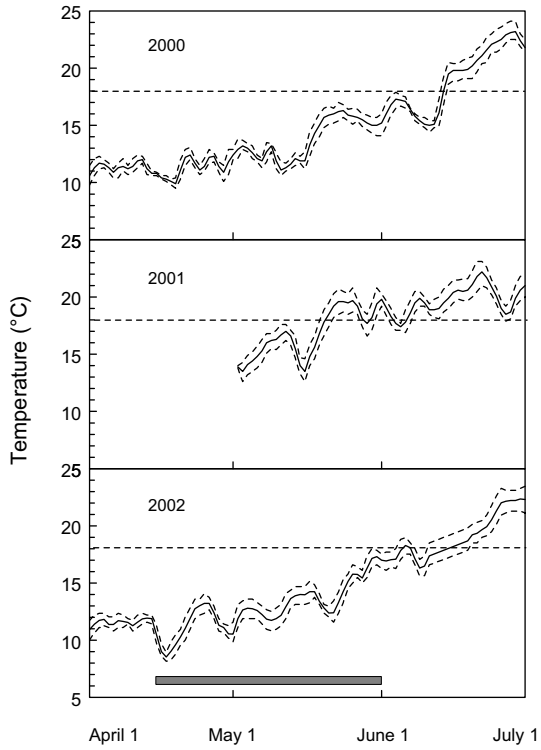

Figure 6. Klamath river temperature profiles for years 2000–2002 (daily means, minimum and maximum recorded temperatures at approximately river kilometer 56 and 66). The horizontal slashed bar indicates the time of the predominant spawning migration over a 5-year period (1999–2003).

1000 cfs during May, and 710 cfs during June).[2] However, since 1996, the United States Bureau of Reclamation annual Klamath Project Operations Plans has dictated flow releases that have generally exceeded the required FERC instream flows. During relatively normal rainfall years, such as 2000 and 2002, the potential for embryos being exposed to high water temperatures was relatively low. However, during dry years, such as 2001, green sturgeon may be exposed to water temperatures that would have a detrimental effect on developing embryos. In reality the elevated temperatures during 2001 truncated the spawning run and we were unable to obtain broodfish for spawning that year. We cannot conclude if the low water temperature during mid-April, 2002 had any effect on sturgeon spawning or embryo survival. Indeed, it may have shifted spawning to slightly later in the season.

The effect of regulated water flow on parameters other than temperature, such as decreases in turbidity and river level, could also affect the spawning run and the reproductive success. Sturgeons require specific current velocity, bottom substrate, riparian habitat, water temperature and turbidity for spawning, and terrestrial nutrients for survival of larvae and juveniles (Dettlaff et al. 1993, Bemis & Kynard 1997, Coutant 2004). Changes in the hydraulic regimes of rivers effected by hydroelectric construction and diversions have made them either unsuitable or inadequate for sturgeon natural recruitment. Investigation and protection of sturgeon spawning habitat seems to be the most important task for fishery management programs, to ensure the long-term survival of the few remaining species, including green sturgeon.

[2] Pacificorp Klamath hydroelectric project. FERC Project No. 2082. Chapter 10. Klamath River below iron gate dam. Available from the Internet URL http://www.pacificorp.com/File/File16145.pdf.

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