EPA–600/3–77–075
June 1977

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EFFECTS OF TEMPERATURE ON HATCHING SUCCESS AND SURVIVAL OF LARVAE IN THE WHITE BASS

Environmental Research Laboratory
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EFFECTS OF TEMPERATURE ON HATCHING SUCCESS AND SURVIVAL OF LARVAE IN THE WHITE BASS

by

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FOREWORD

Our nation's fresh waters are vital for all animals and plants, yet our diverse uses of water---for recreation, food, energy, transportation, and industry---physically and chemically alter lakes, rivers, and streams. Such alterations threaten terrestrial organisms, as well as those living in water. The Environmental Research Laboratory in Duluth, Minnesota, develops methods, conducts laboratory and field studies, and extrapolates findings

—to determine how physical and chemical pollution affects aquatic life

--to assess the effects of ecosystems on pollutants

—to predict effects of pollutants on large lakes through use of models

--to measure bioaccumulation of pollutants in aquatic organisms that are consumed by other animals, including man.

This report provides data relative to the thermal limits and best temperatures for successful survival of embryos and larvae of an important freshwater sport and commercial fish, the white bass (Morone chrysops).

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ABSTRACT

To determine temperature effects on hatching success of white bass (Morone chrysops) embryos, sample lots of fertilized eggs were exposed to 10 constant temperatures, 6° through 30° C. Exposures were begun at two stages of embryonic development, before gastrulation and after closure of the blastopore. Embryos exposed before gastrulation were more sensitive to extreme temperatures than those exposed after closure of the blastopore. The percentage of normal larvae hatched from embryos exposed before gastrulation was not significantly impaired over the temperature range 18 - 26° C ($P>0.05$). When first exposed after blastopore closure the range of temperatures allowing unimpaired hatching was extended to 14° - 26° C ($P>0.05$). Normal larvae hatched at 14 - 28° C from embryos exposed before gastrulation and at 10 - 28° C when exposed after blastopore closure, but at the extremes in significantly reduced numbers ($P<0.05$). Hatching took place 4.5 days after fertilization when incubated at 14° C and 1 day after fertilization at 26° C.

The 24-hr TL50 for white bass larvae exposed within 24 hr of hatching and acclimated at temperatures from 14° to 26° C was between 30° and 32° C and was not altered by acclimation.

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ACKNOWLEDGMENTS

I thank Mr. D. D. Johnson and Miss C. A. Church for their assistance in conducting these tests, Mr. B. W. Hawkinson of the Minnesota Department of Natural Resources and his staff for their cooperation in providing the white bass brood stock, and Mr. J. N. Dryer for his assistance in calculating larval TL50 values.

SECTION 1

INTRODUCTION

Thermal requirements for fishes are dependent on a number of variables, among which are the species and the life stage being considered (Badenhuizen, 1968). This is a report of the response of three early life stages of the white bass (Morone chrysops) to a range of constant temperatures.

The life stages studied were embryos before gastrulation, embryos after closure of the blastopore, and newly hatched larvae (<24 hr old). These stages are of particular importance since their success is critical to the establishment of the year-class strength of the species and because once deposited they are unable to avoid areas of altered temperatures.

The determination of thermal requirements for the well-being of this species is of particular interest because of its importance both as a sport and commercial fish (Howell, 1945; Ruelle, 1971; Scott and Crossman, 1973; Great Lakes Commission, 1974), and because it inhabits large bodies of fresh water where, through the activities of steam electric generation, perturbation of natural thermal regimes is most likely to occur. Previous studies of thermal requirements of this species have been limited or too general to determine thermal limits for survival or degrees of success at various temperatures (Yellayi and Kilambi, 1969).

SECTION 2

CONCLUSIONS

Thermally unimpaired hatching of white bass embryos occurred when the embryos were exposed from before gastrulation until hatching to temperatures from 18° to 26° C. Hatching took place at 14° and 28° C, but with reduced success. After closure of the blastopore, embryos are more tolerant, and hatching was unimpaired from 14° to 26° C. At 28° C slightly reduced numbers of normal larvae were produced ($P<0.05$). Hatching took place 4.5 days after fertilization at 14° C and 1 day after fertilization at 26° C. The 24-hr TL50 of 1-day-old larval white bass lies between 30° and 32° C.

SECTION 3

RECOMMENDATIONS

If white bass reproduction is to be successful, temperatures in the spawning area during the spawning period and the week after spawning should be above 14° C and below 28° C. For greater potential year-class strength, temperatures between 18° and 26° C are preferable, particularly during the first day or two of incubation. Following hatching, temperatures below 30° C are required to prevent thermally-induced deaths of newly hatched larvae.

SECTION 4

MATERIALS AND METHODS

On June 7, 1974, sexually mature white bass were brought to the Environmental Research Laboratory at Duluth, Minnesota. The fish were collected during the preceding 1 or 2 days from spawning populations in Sturgeon Lake, a backwater area of the Mississippi River near Red Wing, Minnesota. Water temperatures in the collection area during and immediately preceding netting were about 16 - 20° C. The fish were held at 16° C until June 8, when the temperature was raised to 19° C. At this time the females used as the egg source were each injected intraperitoneally with 1 mg of dried triturated carp pituitary, in 1 ml of Cortland physiological saline solution (Sneed and Clemens, 1960). The next day eggs were obtainable from only one female. These were fertilized with the milt from two males by using the dry method (Davis, 1956), and the resulting embryos were used as the test specimens. Duplicate lots of 50 embryos each were exposed to test temperatures. Exposures were initiated at two stages to determine whether embryonic stage at first exposure to test temperatures had any effect on thermal tolerance (Kelly, 1968; Frank, 1974). Initial exposures to test temperatures (6 - 28° C) were completed in exposure group I by the 32-cell stage, Oppenheimer stage 7 (Rugh, 1962), before gastrulation. Embryos in exposure group II were held at 18° C for exposure after closure of the blastopore between Oppenheimer stages 17 and 18. Exposure group II was "screened" to remove infertile and dead eggs before sampling and distribution to test temperatures (6 - 30° C).

The range of tested temperatures was expanded for exposure group II to insure sufficiently high temperatures to exceed the upper lethal limit, and to allow for possible higher tolerance of more advanced embryos. Increased range was indicated by the failure to achieve 100% mortality at the upper limit in group I exposures.

Periodically the eggs were inspected to determine stage of development and to remove and record dead eggs and embryos for fungus control. As hatching approached the eggs were inspected daily, and the dead embryos and infertile eggs were removed and counted. The larvae were segregated and counted as dead or deformed, or both and "normal" larvae categories. All live larvae not obviously deformed when examined under 30x magnification were considered normal larvae. Time to hatching was assessed to the nearest one-half day in which 50% or more of the larvae hatched.

The embryos were exposed to test temperatures in 4- x 4-cm glass "jars" whose bottoms were replaced with 0.23-mm opening stainless steel screen. The "jars" were supported on 13-mm glass legs that held the screens off the

bottoms of the water baths to allow free movement of water past the embryos. The water baths were 10 x 40 x 10 cm deep and received a continuous mean flow of 215 (200-250) ml/min of Lake Superior water at constant temperatures. At the drain end of each water bath a self-starting siphon provided an oscillating water depth. These depth changes provided a flow-through condition about the embryos of 4.5 - 5.9 cm/min velocity during the draw-down portion of the cycle and a 0.4 - 0.5 cm/min velocity during the up-welling portion. The ratio of draw-down time to up-welling time was approximately 1 to 9.

Water temperatures were controlled by a thermostatically controlled immersion heater system and monitored by a Honeywell Multipoint recording telethermometer. Recorder values were daily verified by thermometer readings in each water bath. Thermometers used in verification were standardized within $\pm$0.1° C of a calibration thermometer certified by the American Society for Testing and Materials. Mean test temperatures and ranges are reported in Table 1. Lake Superior water drawn directly from the lake had the following chemical characteristics: total hardness, 43 - 47 mg/l.; total alkalinity, 42 - 43 mg/l.; and pH 7.1 - 7.3. Dissolved oxygen concentrations within the exposure chambers were 7 ppm or greater and the upper limit was controlled to 104% saturation or lower by air-bubble stripping before delivery to the exposure water baths. Other chemical characteristics of the test water were essentially unchanged from those reported by Biesinger and Christensen (1972). Lighting was by Duro-Test Vita-Lite fluorescent lamps with the photoperiod normal for the time of year at Duluth, Minnesota.

The results are reported as the percentage of eggs producing normal larvae. Dead or deformed larvae or both were not considered for analysis as they were not believed likely contributors to year-class recruitment (Volodin, 1960; Kokurewicz, 1969). The percentage normal hatch data were converted to arcsin percentage and analyzed by one-way analysis of variance. Tukey's multiple range tests were used to identify treatment effects statistically significant at the 95% level (Steel and Torrie, 1960).

The 24-hr high temperature TL50 values of newly hatched larvae were determined for larvae from embryos incubated at 14°, 18°, 20°, and 26° C. The upper tolerance limits were determined in the same water-bath system used for the embryo studies. Lots of 10 larvae each were tested at 26°, 28°, 30°, 32°, and 34° C. The data were analyzed by probit analysis (Finney, 1964) to derive the TL50 values and the 95% confidence limits.

SECTION 5

RESULTS

Among group I embryos the maximum mean percentage normal larvae produced (59%) was from embryos incubated at 26° C. Those exposed at 18 - 26° C produced normal larvae at percentages not significantly less than those at 26° C ($P>0.05$) (Table 1). Normal larvae were hatched at 14° and 28° C, but in significantly reduced numbers ($P<0.05$).

Among group II embryos the maximum mean percentage of normal larvae (87%) was produced at 18° C. The percentages of normal larvae produced at temperatures from 14° to 26° C were not significantly less than those at 18° C ($P>0.05$) (Table 1). Normal larvae were hatched at 10° and 28° C, but in significantly reduced numbers ($P<0.05$).

At 14° C approximately 4.5 days were required after fertilization for hatching, but only 1 day was required at 26° C. Rates of development increased directly with temperature. Tolerance of newly hatched larvae did not change with acclimation. Larvae acclimated at 14°, 18°, 20°, and 26° C all had 24-hr TL50's near 31° C; overlapping 95% confidence limits ranged from 28.9° C to nearly 33.6° C (Table 2).

TABLE 1

PERCENTAGE NORMAL HATCH OF WHITE BASS EMBRYOS EXPOSED TO CONSTANT TEMPERATURES.

MEAN PERCENTAGE HATCHES UNDERSCORED BY CONTINUOUS DOTTED LINES ARE NOT SIGNIFICANTLY

DIFFERENT FROM THE MAXIMUM MEAN PERCENTAGE HATCH ($P<0.05$)[a]

	Nominal Temperature (°C)									
Exposure Group (embryonic stage)	6	10	14	18	20	22	24	26	28	30
Group I (before gastrulation)										
Mean temperature (°C)	5.8	9.8	14.2	18.0	20.1	21.8	23.7	25.9	28.0	
Range (°C)	5.4-6.5	9.0-11.0	13.3-14.8	17.8-18.2	19.8-20.5	21.4-22.0	23.3-23.8	25.7-26.1	27.7-28.1	
Replicate A	0	0[b]	28	34	38	66	30	56	28	-
Replicate B	0	0[b]	24	30	48	50	32	62	18	-
Mean A and B	0	0	26	32	43	58	31	59	23	-
Group II (after closure of the blastopore)										
Mean temperature (°C)	6.0	10.0	14.2	18.0	20.1	21.8	23.6	26.1	27.9	30.2
Range (°C)	5.4-6.5	9.2-11.0	13.3-14.8	17.8-18.2	19.8-20.5	21.4-22.0	23.3-23.8	25.7-26.3	27.5-28.2	30.0-30.5
Replicate A	0	34	76	84	78	66	78	58[c]	50	0
Replicate B	0	22	72	90	74	78	84	74	58	0
Mean A and B	0	28	74	87	76	72	81	66	54	0

[a] When in the transformed form for statistical testing. Tukey's honestly significant difference for the before-gastrulation results was 16.4% ($P=0.05$), for the after-closure-of-the-blastopore data it was 20.2% ($P=0.05$). All data here are presented as before normalization and transformation.

[b] Exposed first 14 hr to mean of 9.2° C, thence to 10.0° C until hatching.

[c] Probably was higher as container holding embryos was found tipped during part of the day of hatching allowing possible escape of swimming larvae without recapture for counting.

TABLE 2

TWENTY-FOUR-HR TL50'S OF NEWLY HATCHED LARVAL WHITE BASS.[a]

(ACCLIMATION TEMPERATURES ARE INCUBATION TEMPERATURES).

Acclimation temperature (°C)									
14		18		20		26		14-26 Combined	
Test temperature	Dead (%)	Test temperature	Dead (%)	Test temperature	Dead (%)	Test temperature	Dead (%)	Test temperature	Dead (%)
34.2	100	34.4	100	34.4	100	34.2	100	34.4	100
32.3	40	32.3	100	32.3	40	32.3	70	32.3	62
30.1	100[b]	30.3	10	30.3	20	30.3	20	30.3	17
28.2	10	28.0	0	28.0	0	28.0	30	28.0	10
26.2	0	26.4	0	26.4	0	26.4	90[b]	26.3	0
TL50	31.7		30.8[c]		32.0		30.6		31.3
95% C.L.	30.1-33.0		-----		31.1-33.0		29.0-31.9		28.9-33.6

[a] N = 10, all test groups.

[b] Points arbitrarily considered aberrant, listed for completeness of data but not used in analysis.

[c] Graphic method with logarithmic probability paper; two values between 0 and 100% effect required for Finney's (1964) method not available.

SECTION 6

DISCUSSION

Hatching success in group I was uninhibited from 18° to 26° C. These limits extend well beyond the 15.6 - 16.7° C optimum limits previously reported by Yellayi and Kilambi (1969). Their suggested upper lethal limit, 20.0° C, was also found too restricted; normal larvae in this study were produced through 28° C, though with reduced success at the upper temperature. Yellayi and Kilambi's (1969) lower lethal limit, 12.8, seems a reasonable estimate of expected 100% mortality based on the downward trend between 18° and 10° C in the data reported here.

The overall higher percentages of successful hatching at temperatures between 18° and 26° C among group II embryos than among group I embryos are not considered a function of the stage of development when first exposed. They are rather the result of the "screening" for live embryos that was carried out before sampling the test lots of the more advanced embryos. Thus more fertilized eggs are represented in group II samples, and higher percentage hatching would be expected.

The 24-hr TL50 values determined for 1-day-old white bass were not changed over the range of acclimations from 14° to 26° C. Acclimation has generally been accepted as a mechanism by which fish are able to adjust their thermal tolerance. However, the early larval stage is apparently unable to accomplish the necessary physiological changes. This result is consistent with previously reported responses among 1-day-old brook trout (McCormick *et al.*, 1972). McCauley (1963) suggests that the capacity for an organism to adjust to temperature changes probably requires functional organ systems not present in early embryonic forms.

The life stage of the organism is important in determining its responses to environmental temperatures, as illustrated by the lack of reduction in hatching success with increase in temperature range when embryos are first exposed after closure of the blastopore rather than before. These results also corroborate Badenhuizen's (1968) finding with largemouth bass that cold tolerance is gained more with increased age of the embryo than is heat tolerance. Thermal requirements determined in the laboratory correlated well with naturally occurring environmental requirements. In this case, the lower limits for successful incubation, $>10°$ and $\leq 14°$ C determined in this laboratory study, are close to the lower thermal threshold initiating spawning, 11.7° - 14.4° C (Webb and Moss, 1967), suggesting that natural selection sets the temperature at which the spawning act occurs.

Webb and Moss (1967) report observations of spawning in natural waters up to 23.9° C, somewhat cooler than the 26° C found in this study to permit uninhibited hatching success, but allowing for seasonal rising temperature trends during the time required before hatching, not an unreasonable value. Recognizing that field observations may misinterpret ripe-fish activity for spawning and fail to observe the remaining few late spawners after the preceding masses have departed the spawning area, field and laboratory values are probably even closer.

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TECHNICAL REPORT DATA (Please read Instructions on the reverse before completing)		
1. REPORT NO. EPA-600/3-77-075	2.	3. RECIPIENT'S ACCESSION NO.
4. TITLE AND SUBTITLE EFFECTS OF TEMPERATURE ON HATCHING SUCCESS AND SURVIVAL OF LARVAE IN THE WHITE BASS		5. REPORT DATE June 1977 issuing date 6. PERFORMING ORGANIZATION CODE
7. AUTHOR(S) J. Howard McCormick		8. PERFORMING ORGANIZATION REPORT NO.
9. PERFORMING ORGANIZATION NAME AND ADDRESS Environmental Research Laboratory - Duluth, MN Office of Research and Development U.S. Environmental Protection Agency Duluth, Minnesota 55804		10. PROGRAM ELEMENT NO. 1BA608 11. CONTRACT/GRANT NO. None (in-house)
12. SPONSORING AGENCY NAME AND ADDRESS Same as above		13. TYPE OF REPORT AND PERIOD COVERED 14. SPONSORING AGENCY CODE EPA/600/03
15. SUPPLEMENTARY NOTES		

16. ABSTRACT

To determine temperature effects on hatching success of white bass (Morone chrysops) embryos, sample lots of fertilized eggs were exposed to 10 constant temperatures, 6° through 30° C. Exposures were begun at two stages of embryonic development, before gastrulation and after closure of the blastopore. Embryos exposed before gastrulation were more sensitive to extreme temperatures than those exposed after closure of the blastopore. The percentage of normal larvae hatched from embryos exposed before gastrulation was not significantly impaired over the temperature range 18 - 26° C ($P>0.05$). When first exposed after blastopore closure the range of temperatures allowing unimpaired hatching was extended to 14° - 26° C ($P>0.05$). Normal larvae hatched at 14 - 28° C from embryos exposed before gastrulation and at 10 - 28° C when exposed after blastopore closure, but at the extremes in significantly reduced numbers ($P<0.05$). Hatching took place 4.5 days after fertilization when incubation at 14° C and 1 day after fertilization at 26° C.

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17. KEY WORDS AND DOCUMENT ANALYSIS		
a. DESCRIPTORS	b. IDENTIFIERS/OPEN ENDED TERMS	c. COSATI Field/Group
Fresh water fishes Bass Temperature Larvae Embryos Survival Tolerances (physiology)	White Bass Morone chrysops Time to hatching	06F
18. DISTRIBUTION STATEMENT RELEASE TO PUBLIC	19. SECURITY CLASS (This Report) UNCLASSIFIED 20. SECURITY CLASS (This page) UNCLASSIFIED	21. NO. OF PAGES 19 22. PRICE

☆ U.S. GOVERNMENT PRINTING OFFICE: 1977—757-056/6466