Bull. Environm. Contam. Toxicol. 29, 631–636 (1982)

A Comparison of Chlorine Toxicity to Three Life Stages of Striped Bass (*Morone saxatilis*)

Lenwood W. Hall, Jr.,[1] William C. Graves,[1] Dennis T. Burton,[1] Stuart L. Margrey,[1] Frank M. Hetrick,[2] and Bob S. Roberson[2]

[1]*The Johns Hopkins University, Applied Physics Laboratory, Aquatic Ecology Section, Shady Side, MD 20764, and* [2]*University of Maryland, Department of Microbiology, College Park, MD 20742*

Chlorine is a widely used biocide in municipal sewage treatment operations and power plant facilities in the United States. Due to the widespread use of chlorine, numerous reviews have resulted from concern about its effects on aquatic biota (BRUNGS 1976; MORGAN 1980; OPRESKO 1980; HALL et al. 1981). Responses of an aquatic species to stress conditions such as chlorine exposure can be influenced by such biological factors as age (size, developmental stage), population density, physiological state and preexposure conditioning (acclimation). One biological factor affecting chlorine toxicity that has not been adequately addressed is a comparison of various life stages of an estuarine fish species ranging from the late larval stage to one year old juveniles.

The objective of this study was to determine and compare chlorine toxicity among larvae, young juveniles and older juvenile striped bass Morone saxatilis. The striped bass was selected for this study for the following reasons: (1) it has been suggested that the population decline of this species is partially attributed to chlorine use (COULTER in press), (2) it is a recreationally and commercially important species on both the east and west coast of the United States and (3) it has national importance as evidenced by the Emergency Striped Bass Research Study (DEPARTMENT OF THE INTERIOR, U.S. FISH AND WILDLIFE SERVICE 1980).

MATERIALS AND METHODS

Striped bass larvae (22 d old, mean total length = 10.6 mm), young juveniles (60 d old, mean total length = 29.9 mm) and older juveniles (388 d old, mean total length = 94.6 mm) were obtained from the following hatcheries, respectively: Moncks Corner Striped Bass Hatchery, Moncks Corner, South Carolina; Delmarva Ecological Laboratory, Middletown, Delaware and Orangeburg State Hatchery, Orangeburg, South Carolina. All test organisms were maintained at densities of < 0.1 g of fish/l under continuous flow conditions during acclimation. Larvae, acclimated in 376 l rectangular fiberglass tanks, were fed brine shrimp, Artemia salina, twice daily supplemented by plankton present in the incoming Chesapeake Bay water. Juveniles, acclimated in 1893 l circular fiberglass tanks, were fed trout chow daily. The range of water quality parameters during acclimation and testing are

0007-4861/82/0029-0631 $01.20

shown in Table 1. A photoperiod simulating natural conditions was used during the acclimation period.

A continuous flow toxicant system, similar to the one described by VANDERHORST et al. (1977), was used to test total residual chlorine (TRC) toxicity to all striped bass life stages for a 96-h period. A TRC concentration of 10 mg/l was established in a chlorine stock tank using chlorine gas fed through a chlorine ejector system. The TRC stock solution was mixed with ambient water to achieve the desired concentrations in 95 l test aquaria. Replicates of 50 larvae were tested at the following concentrations: 0.000, 0.125, 0.150, 0.175, 0.200, 0.225 and 0.250 mg/l TRC. Replicates of 10 young juveniles were tested at TRC concentrations of 0.000, 0.025, 0.050, 0.100, 0.150, 0.200 and 0.250 mg/l. Both larvae and young juveniles were tested in 19.6 l mesh baskets placed in the 95 l test aquaria. Replicates of 10 older juvenile striped bass were tested in 95 l test aquaria at 0.000, 0.130, 0.160, 0.190, 0.220, 0.250 and 0.280 mg/l TRC.

Total residual chlorine concentrations were measured amperometrically (AMERICAN PUBLIC HEALTH ASSOCIATION, AMERICAN WATER WORKS ASSOCIATION AND WATER POLLUTION CONTROL FEDERATION 1981) using a Fischer and Porter amperometric titrator every 2-4 h during the 96-h exposure period. Mortality observations were conducted for all life stages at the following time intervals: 1, 2, 4, 8, 16, 24, 36, 48, 72 and 96 h. Death was defined as cessation of opercular rhythm and/or no visible movement of the organism after probing.

LC50 values and 95% confidence limits at 24-, 48- and 96-h intervals were calculated for each life stage by using the logistic function as described by ASHTON (1972). The failure of 95% confidence limits to overlap was used to determine significant differences among the LC50 values for each life stage.

RESULTS AND DISCUSSION

The LC50 values and associated 95% confidence limits for all life stages are presented in Table 2. A comparison of the 96-h LC50 values shows that larvae (0.14 mg/l TRC) are the most sensitive stage followed by young juveniles (0.19 mg/l TRC). The older juveniles (0.23 mg/l TRC) were found to be the most resistent life stage. A similar trend exists for the 24- and 48-h LC50 values.

The toxicity of TRC was generally found to increase with exposure time for each life stage, although the influence of this factor varied with each life stage. There was a significant difference in TRC toxicity at 24, 48 and 96 h for larvae as evidenced by non-overlaping 95% confidence limits. The 24-, 48- and 96-h confidence limits for the young juveniles overlaped thus indicating that the influence of exposure time was minimal after 24 h. A comparison of the 24-, 48- and 96-h LC50 values for the

TABLE 1. Range of water quality parameters during acclimation and testing of various striped bass life stages

Life stage	Temperature (^{o}C)	Salinity ($^{o}/oo$)	D.O. (mg/1)	pH	Ammonia-Nitrogen (mg/1)
22 d larvae	18.5–19.0	5.5	8.0–9.4	8.9–9.2	<0.10
60 d juveniles	24.0–25.0	10.0	5.1–6.5	7.1–7.6	<0.15
388 d juveniles	20.0–20.5	10.0–10.5	5.5–6.8	7.7–8.0	<0.10

TABLE 2. LC50 values for various striped bass life stages

	LC50s (mg/1 TRC) and 95% Confidence Limits		
Life stage	24 h	48 h	96 h
22 d larvae	0.19 (0.184–0.190)	0.16 (0.151–0.160)	0.14 (0.141–0.147)
60 d juveniles	0.21 (0.196–0.225)	0.20 (0.180–0.215)	0.19 (0.178–0.209)
388 d juveniles	0.29[a]	0.25[a]	0.23 (0.226–0.240)

[a]Confidence limits could not be calculated.

older juveniles suggest that 24- and 48-h values may be different; however, it appears that 48- and 96-h LC50's are similar. Exact comparisons of TRC toxicity over time with this life stage are not possible as 95% confidence limits could not be calculated at 24 and 48 h.

The literature indicates that age-related trends in chlorine tolerances of fish are not consistent for all species studied. Chlorine toxicity studies conducted with various freshwater fish species have demonstrated that chlorine tolerance may increase with age (EREN & LANGER 1973; WOLF et al. 1975; ROSEBOOM & RICHEY 1976, 1977; BROOKS & SEEGERT 1977) decrease with age (HUGHES 1975; FANDREI & COLLINS 1979; BROOKS & SEEGERT 1977) and may not be affected by age (WOLF et al. 1975; DICKSON et al. 1977). The specific ages of fish that were compared for chlorine toxicity was likely the reason for these inconsistent trends. OPRESKO (1980) has reported that most of the present data with freshwater fish indicate that the larger or older organisms are more tolerant to chlorine than younger organisms.

HUGHES (1975) conducted a freshwater chlorine toxicity study with striped bass larvae (7 d old) and young juveniles (≈ 30 d old). This investigator used a single dose static exposure of 70% calcium hypochlorite with each life stage. A 24-, 48- and 96-h LC50 of 0.5 mg/l chlorine (species unknown; 70% $Ca(ClO)_2$ used) was reported for striped bass larvae. The 24-, 48- and 96-h LC50s for juveniles were 0.30, 0.25 and 0.25 mg/l chlorine, respectively. Exact comparison between the HUGHES (1975) study and the present study are difficult due to different age fish (7 and 30 d fish versus 22, 60 and 388 d fish), exposure conditions (single static dose versus continuous-flow dose), water type (freshwater versus estuarine), chlorine species used (species unknown versus total residual chlorine) and method for analytical measurement (unknown versus amperometric). However, it would seem plausible that age specific toxicity trends would be similar between the two studies. HUGHES (1975) found the younger larval stage to be less sensitive to chlorine than the older fingerling (juvenile) stage. Our results demonstrate that striped bass become more tolerant to chlorine as they increase in age from 22 d to approximately one year. The contrasting trends found in our study and the HUGHES (1975) study can be explained by examining the age of fish being tested. HUGHES (1975) reported the larval stage (7-30 d) of striped bass may become more sensitive to chlorine until 30 d. MIDDAUGH et al. (1977) verified a similar trend comparing 12 and 30 d old striped bass larvae in a continuous flow chlorine toxicity study. When comparing the chlorine tolerance of striped bass ($\leq$ 30 d) as reported by HUGHES (1975) and MIDDAUGH et al. (1977) with our data, it appears that after larvae complete metamorphosis (30-40 d of age depending on temperature) into young juveniles their resistence to chlorine begins to increase with age.

Age specific responses of young developmental stages of striped bass (eggs, 12-20 h old; prolarvae, 24 h old and larvae, 9-16 d old) exposed to simultaneous chlorine, ΔT and exposure duration conditions have been evaluated in our laboratory (HALL et al. in press). The egg or embryo stage was generally found to be more resistent to simultaneous chlorine, ΔT and exposure duration conditions followed by larvae and prolarvae, respectively. Other investigators have also shown in chlorine toxicity studies that larvae of striped bass and other estuarine and marine species are more sensitive than eggs under constant exposure conditions (ALDERSON 1972, 1974; MORGAN & PRINCE 1977; MIDDAUGH et al. 1977; BURTON et al. 1979). When summarizing data from the present study with information collected during other investigations it appears that the egg stage of the striped bass is relatively tolerant to chlorine followed by a sensitive prolarval or yolk-sac stage that continues until approximately 7 d of age and a less sensitive larval stage that increases in sensitivity until metamorphosis. Beyond the late larval or early juvenile stage, striped bass become more tolerant to chlorine until an age of approximately one year. Additional studies would be necessary to determine the tolerance of older life stages of this species to chlorine.

ACKNOWLEDGEMENTS

We wish to thank Elgin S. Perry for statistical analysis of the data, Michael Jepson for his technical assistance and Beverly Knee for typing the manuscript. We acknowledge the Maryland Power Plant Siting Program (Contract No. P84-81-04) for supporting this research. Special consideration is extended to Dr. Paul Miller for his technical review.

REFERENCES

ALDERSON, R.: Pages 312-315 in M. Ruivo (ed.) Marine pollution and sea life. Fishing News (Books) Ltd., Surrey, England (1972).

ALDERSON, R.: Aquaculture 4, 41 (1974).

AMERICAN PUBLIC HEALTH ASSOCIATION, AMERICAN WATER WORKS ASSOCIATION AND WATER POLLUTION CONTROL FEDERATION: Standard methods for the examination of water and wastewater. 15th ed. Amer. Public Health Assoc., Washington, DC. (1981).

ASHTON, W. D.: The logit transformation with special reference to its uses in bioassay. Charles Griffin & Co. Ltd. London, England (1972).

BROOKS, A. S. and G. L. SEEGERT: The effects of intermittent chlorination on the biota of Lake Michigan. University of Wisconsin Center for Great Lakes Studies Rept. No. 31, Milwaukee, WI (1977).

BRUNGS, W. A.; Effects of wastewater and cooling water on aquatic life. U.S. Environ. Prot. Agency Rep. No. EPA-600/3-76-036 (1976).

BURTON, D. T., L. W. HALL, JR., S. L. MARGREY and R. D. SMALL: J. Fish. Res. Board Can. 36, 1108 (1979).

COULTER, J. B.: Proceedings second national symposium on wastewater disinfection. U.S. Environ. Prot. Agency (in press).
DEPARTMENT OF INTERIOR, U.S. FISH AND WILDLIFE SERVICE: 1980 Annual report emergency striped bass research study. Department of the Interior, U.S. Fish and Wildlife Service Rept. (1980).
DICKSON, K. L., J. CAIRNS, JR., B. C. GREGG, D. I. MESSENGER, J. L. PLAFKIN and W. H. VAN DER SCHALIE: J. Water Pollut. Control Fed. 45, 35 (1977).
EREN, Y. and Y. LANGER: Bamidgeh 25, 56 (1973).
FANDREI, G. and H. L. COLLINS: Bull. Environ. Contam. Toxicol. 23, 262 (1979).
HALL, L. W., JR., G. R. HELZ and D. T. BURTON: Power plant chlorination A biological and chemical assessment. Ann Arbor Sci. Publ. Inc., Ann Arbor, MI (1981).
HALL, L. W., JR., D. T. BURTON, S. L. MARGREY and W. C. GRAVES: in R. L. Jolley, W. A. Brungs, J. A. Contruvo, R. B. Cumming and J. S. Mattice (eds.) Water chlorination environmental impacts and health effects, Vol. 4. Ann Arbor Sci. Publ. Inc., Ann Arbor, MI (in press).
HUGHES, J. S.: Striped bass, Morone saxatilis (Walbaum), culture investigations in Louisiana with notes of sensitivity of fry and fingerlings to various chemicals. Louisiana Wildlife and Fisheries Commission Fish Bull. No. 15, Baton Rouge, LA (1975).
MIDDAUGH, D. P., J. A. COUCH and A. M. CRANE: Chesapeake Sci. 18, 141 (1977).
MORGAN, R. P. II: Pages 75-102 in C. H. Hocutt, J. R. Stauffer, Jr., J. E. Edinger, L. W. Hall, Jr. and R. P. Morgan II (eds.) Power plants effects on fish and shellfish behavior. Academic Press Inc., New York, NY. (1980).
MORGAN, R. P. II and R. D. PRINCE: Trans. Am. Fish Soc. 106, 380 (1977).
OPRESKO, D. M.: Review of open literature on effects of chlorine on aquatic organisms. Electric Power Res. Inst. Rept. No. EPRI EA-1491, Palo Alto, CA (1980).
ROSEBOOM, D. P. and D. L. RICHEY: Trans. Ill. State Acad. Sci. 69, 230 (1976).
ROSEBOOM, D. P. and D. L. RICHEY: Acute toxicity of residual chlorine and ammonia to some native Illinois fishes. Illinois State Water Survey Rept. No. 85, Urbana, IL (1977).
VANDERHORST, J. R., C. I. GIBSON, L. J. MOORE and P. WILKINSON: Bull. Environ. Contam. Toxicol. 17, 577 (1977).
WOLF, E. G., M. J. SCHNEIDER, K. O. SCHWARZMILLER and T. O. THATCHER: Toxicity tests on the combined effects of chlorine and temperature on rainbow (Salmo gairdneri) and brook (Salvelinus fontinalis) trout. Battelle Northwest Laboratories Rept. No. BNWL-SA-5349, Richland, WA (1975).

Accepted October 17, 1982