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A Morphological Defect in Shiner Perch Resulting from Chronic Exposure to Chlorinated Sea Water

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The use of chlorine as a bleaching and biocidal agent has become common in numerous municipal and industrial processes. The most obvious examples include pulp and paper mills, chlorinated potable water, sewage effluent treatment, and cooling water systems which are subject to biofouling (e.g., steam electrical generating stations). Concern for the environmental impact of the chlorine-bearing effluents released at these sites has resulted in many laboratory and field studies in recent years, and these have been adequately reviewed previously (BECKER and THATCHER 1973, BRUNGS 1973, 1976, PAGE 1975; and MATTICE and ZITTEL 1976).

Information on concentrations of chlorinated fresh and sea water acutely lethal to fishes and invertebrates far surpasses that available on sublethal or lethal effects resulting from chronic exposures. It is likely that the most important impact of chlorinated effluents to the environment will result from chronic exposures to low levels of the total residual oxidant (TRO) fraction[2] or to persistent byproducts such as halogenated organics. Acutely lethal levels of the halogen components of chlorinated effluents are, in general, rapidly reduced by chemical or physical processes and dilution; and lower levels of chlorine treatment are resulting from economic considerations, recent toxicity information, and associated regulatory agency limitations.

Several studies have reported upon malformed fishes resulting from exposures to chlorinated water or to specific chlorinated organics known to occur from the chlorination of water. EYMAN et al. (1975) exposed carp eggs to 5-chlorouracil, a byproduct which occurs in chlorinated sewage plant effluents (JOLLEY 1974). They determined that some of the newly hatched larvae were affected by severely coiled body form. ARTHUR et al. (1975); BELLANCA and BAILEY (1976), have reported that exposure

[2] The TRO fraction includes free available "chlorine" as HOCl, OCL^-, HOBr or OBr^-, and combined available "chlorine" chiefly as the various chloramines in fresh water, and similar halogen forms which appear rapidly upon the chlorination of sea water.

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to chlorinated sewage effluents caused deformed backs in fishes in fresh water and sea water, respectively. More recently, ongoing studies at this laboratory (Battelle M. R. L.) have resulted in hatching of deformed flatfish from eggs exposed for 30 minutes to 1.5 mg/ℓ TRO.

To increase knowledge of the sublethal effects of chlorinated sea water, an experiment to determine effects of low levels of chlorination upon the growth of shiner perch (Cymatogaster aggregata) was initiated. As this study progressed, a morphological defect to the head of these fish was detected. That defect and its consequences are the subject of this report.

METHODS

Adult shiner perch (live bearers) were collected by beach seine from Sequim Bay and held at the laboratory in a 350 liter (ℓ) continuous-flow chamber for approximately 60 days. At this time, females carrying young could easily be identified by their distended abdomens. Fish were fed Oregon Moist Pellet® during laboratory acclimation.

Throughout the experiment, the temperature was maintained at 15 C ± 0.5, dissolved oxygen was always above 7.5 mg/ℓ, pH was 8.0 ± 0.2, and the salinity was 29.5 ‰ ± 1.5. Sea water is pumped (through an epoxy lined pump) from the main channel at the mouth of Sequim Bay, from a depth of about 12 meters, to a 57,000ℓ holding chamber, then flows by gravity to the experimental areas. Large debris was removed by filtration. All water lines throughout the systems are PVC, glass or tygon.

The exposure system consisted of glass-fronted, 45ℓ fiberglass aquaria, which received chlorinated sea water from a proportional diluter (MOUNT and BRUNGS 1967). Water passed through the system only once, and each aquaria received approximately 700 ml/minute for a 95% replacement time of slightly over 3 hours (SPRAGUE 1969).

Total residual oxidant concentrations were measured by an amperometric titration system which differs from the procedure described by APHA et al. (1976) in three ways: 2 ml each of the KI and the Acetate Buffer (pH:4) are mixed together before the 200 ml sample is added; both electrodes are platinum; and a polargraph with printout is used for endpoint detection. The detection limit of this method is approximately 1 µg/ℓ (~1 ppb). TRO concentrations (standard deviations in parentheses) over the entire 29 weeks experimental period in the respective exposure chambers were: control[3], 0.02 (0.01), 0.05 (0.02), 0.07 (0.03), 0.09 (0.03), and 0.11 (0.04) mg/ℓ.

Four adult female shiner perch containing young were placed into each of six exposure aquaria in late August. They were fed approximately 5% of their body weight daily. The young shiner

[3] TRO concentrations shown for the exposure chambers have been corrected for the 0.01 mg/ℓ found in the control. TRO was measured daily.

perch began appearing the next day after the adults were placed in the exposure chambers, and they continued to be released for approximately 2 weeks. After 6 weeks in the exposure chambers (mid-October), the adult females and all but 5 of the young in each chamber were removed. At this age, the young remaining in the exposure chambers weighed from 1.00 to 1.75 g each.

During the initial 6 week period (through mid-October), before numbers of fish were reduced to 5 young per exposure, some mortality of the young occurred in all chambers. These losses did not appear to be related to chlorine exposure[4] but were rather randomly distributed among the chambers, perhaps resulting from such factors as defective birth, and crowding with its resultant competition. During the next 9 weeks of exposure (to mid-December) there were no mortalities. Then, in the following one-week period (December 12-18), all the fish at 0.09 mg/ℓ died. During the first week of January, (after 18 weeks of exposure) those at 0.07 mg/ℓ began dying, and the last one in that chamber died two weeks later (January 20). Dead individuals were frozen.

The original purpose of this experiment was to determine the effects of chlorinated sea water on growth of the shiner perch. To accomplish this, the fish were weighed monthly. During the third monthly weighing (January), some of the exposed fish were observed to have one eye "turned in", when viewed from the dorsal aspect (Figure 1). This observation led to the examination of the fish at monthly intervals up to the termination of the experiment in March.

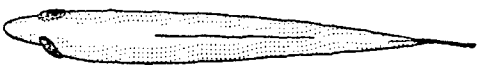

Figure 1. Shiner perch with turned in eye, dorsal view.

[4] In the highest concentration chamber (0.11 mg/ℓ TRO) 14 young were released the first day and all were dead the second; no more appeared in that chamber.

Since the original observation of the eye condition occurred by chance several months after the initiation of the growth experiment, data are not available to determine how soon the problem developed after the exposures began. The earliest it could be determined from these data was on December 12th (13 to 15 weeks after the young were released by the females) as detected from the analysis of frozen fish. The young fish which died prior to mid-October were preserved in formalin, and the effect of the preservative on the eyes of these small fish was such that the presence or absence of the defect could not be determined.

RESULTS AND DISCUSSION

The data for the morphological defect to the eyes of young shiner perch are presented in Table 1. There appeared to be a concentration-response relationship between the TRO exposure level and the defective eye condition. There was no effect in the control fish, while at the higher levels of TRO, the frequency of occurrence of the eye problem was greater and developed earlier. At the lower exposures, the numbers of afflicted fish increased as the experiment progressed. Over the experimental period, 17 of the 20 (85%) fish exposed to chlorinated water developed the defect. It should be noted that death was not correlated with the apparent severity of the morphological aspects of the defect. Several of the fish which died during the experiment (0.07 and 0.09 mg/ℓ) did not exhibit severely defective eyes.

TABLE 1.

Numbers of shiner perch observed with the "turned-in" eye condition. Five fish were initially exposed at each concentration.

TRO Conc. (mg/ℓ)		0.00	0.02	0.05	0.07	0.09
Date	(weeks of exposure)					
January 1	(19)	0	3	2	(4)[a]	(5)[a]
February 5	(23)	0	3	3	(5)	(5)
March 4	(27)	0	4	3	(5)	(5)
March 19	(29)	0	4	3	(5)	(5)

[a] The defect was first detected on January 8 when only 1 fish was still alive at 0.07 mg/ℓ, the other 4 having died since January 2. At 0.09 mg/ℓ, all 5 fish died December 12-18. Reexamination of the fish which died prior to January 8 indicated that the onset of the defect was prior to December 12, the 15th week of exposure.

None of the affected fish exhibited the defect in both eyes. In some, the right eye was turned in, while in others it was the left. In the most severely deformed cases, the entire side of the head was affected, including the adjacent side of the mouth and the opercular area.

The defect did not appear to affect swimming ability; however, feeding ability appeared to be altered. During attempts to observe the behavior of exposed fish, those with the most advanced state of the defect could be singled out for observation. It was characteristic for both severely affected and apparently unaffected perch to go through the same feeding motions. They would cruise around at several cm above the bottom, occasionally swimming "nose down" to the bottom to pick up a pellet, then "back pedalling" to their horizontal position to resume cruising. The most obvious difference between those severely affected and the rest, was that there, typically, were no food pellets at the point where the former touched the aquarium bottom, while the latter nearly always picked up pellets. Additionally, fish with the most severe cases were not able to avoid the dip-net as effectively as others, presumably because they could not see it or determine its actual location.

The extent to which the defect contributed to the death of the fish is not known. However, the observations of vision impairment and reduced feeding certainly suggest detrimental effects. Impaired feeding can reduce growth and stress other physiological functions. Those effects, plus others such as the reduced ability to avoid predators and perhaps defend territories, may result in an impacted population under field conditions.

CONCLUSIONS

1) A morphological defect to one eye was observed in 17 of 20 young shiner perch exposed to chlorinated sea water, and it was absent in the 5 controls. The occurrence of the defect was apparently related to the concentration of total residual oxidants.
2) Behavioral aberrations appeared to be associated with the morphological defect in exposed fish. Fish with severe cases were observed to be unsuccessful at feeding and at avoiding capture by dip-net.
3) Fish which died during the experiment varied in the degree of severity of the observable morphological aspects of the defect.

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