Made in United States of America
Reprinted from COPEIA 1967, No. 2 June 5
pp. 467–468

drin levels recovered from the gut, low residue content of the gills after exposure, and upon observations of Iyatomi *et al.* (1958) that young fish were resistant to endrin, presumably because non-functional mouths prevented swallowing of water. Large amounts of water would have to be swallowed to account for the concentration of residues in the tissues of exposed fish, and this is contrary to statements of Black (1957) that most water enters the body of freshwater fishes via the gill and oral surfaces, some through the body surface, and the remainder in and with food.

Ferguson *et al.* (1966) reported similar mortality rates in fed and starved groups of mosquitofish (*Gambusia affinis*) exposed to high endrin concentrations and suggested that the amount of endrin ingested with food was inconsequential. When heads or bodies of mosquitofish were exposed to an endrin solution while the remainder of the fish was immersed in tapwater, much high mortality occurred in specimens whose heads were exposed to the toxicant. Although these experiments failed positively to exclude entry in swallowing water, the rate of endrin concentration in the body was too rapid to be attributed to swallowing only.

The present study was initiated to obtain additional evidence concerning the pathway of endrin entry into exposed fish. The procedure involved the comparison of mortality rates of normal black bullheads (*Ictalurus melas*) and others that were rendered incapable of swallowing in an endrin solution.

Materials and methods: In August, black bullheads ranging from 50 to 112 mm standard length were trapped from isolated pools of a local stream. All specimens were taken to the laboratory and held in aged tapwater for 48 hr prior to being used in tests.

One group of 40 fish was subjected to an operation in which the gut was tied-off in the region of the upper esophagus. A specimen was placed on its back in a U-shaped trough of hardware cloth, secured with rubber tubing, and partially immersed in water. The restraining action of the trough and tubing was supplemented by projecting the dorsal spine through the mesh of the hardware cloth. A longitudinal incision was made along the mid-line of the abdomen, extending posteriorly from the region of the pectoral girdle approximately two-thirds the distance to the bases of the pelvic fins. The

THE PATHWAY OF ENDRIN ENTRY IN BLACK BULLHEADS, *ICTALURUS MELAS*.—Several reports dealing with the effects of endrin on fish have contained conflicting interpretations concerning the mode of endrin entry into the body. Mount (1962) concluded that most endrin entered carp (*Cyprinus carpio*) with swallowed water. This conclusion was based on relatively high en-

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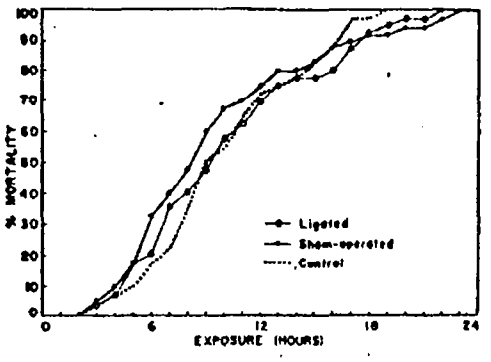

Fig. 1. Mortality in 40 black bullheads, *Ictalurus melas*, with ligated esophagi, in sham-operated fish, and in normal fish (controls) during a 24-hr exposure to a 50 ppb endrin solution.

liver was reflected and a ligature tied near the junction of the esophagus and pharynx, taking care to exclude all major blood vessels from the ligature. A ligature was prepared with sufficient tension to obliterate the lumen of the esophagus. The incision was closed with 3–6 sutures, depending on the length of the incision. About 15 min were required to perform the operation, after which, all specimens were observed for 3–9 hr prior to use in experiments. One group of seven fish was not tested but was held a week in tapwater to determine the effects of the operation.

A second group of 40 bullheads was subjected to the same operation including putting the thread in place for the ligature, but omitting the constriction and tying steps in the procedure. These specimens constituted a sham-operated control. A third group of 40 fish served as unimpaired controls and were spared the operation.

Initially, 11 fish from each group were tested. Later the procedure was repeated using 14 fish in each group. Finally, 15 in each group were employed in another series of tests, bringing the totals to 40 specimens per group.

An acetone stock solution of endrin was diluted with tapwater (pH 7.8, hardness 28 ppm) to prepare 30 l of 50 ppb concentration in 15-gal aquaria (glass with slate bottoms). A total of 6–8 fish (depending on size), representing two or three individuals from each group, was tested in each aquarium. Exposure was at room temperature (72° ± 4° F) and continued until all fish were dead. Mortality was recorded hourly, at which time dead fish were removed and examined internally to identify them as sham-operated controls or ligated individuals. All fish were denied food during exposure to endrin.

Results and conclusions: In every test, all fish from each of the three groups were dead after a 23-hr exposure and the mortality rates were nearly identical (Fig. 1). The fact that operated and normal fish survived equally well is a testimonial to the hardiness of bullheads and indicates that deaths were due to causes other than the operation. This was confirmed when seven specimens survived a week after having their esophagi ligated.

These results showed that fish unable to swallow the endrin solution died as rapidly as those free to swallow. If any endrin was obtained from swallowed water by the fish with functional esophagi, the amount was too small in proportion to that entering by other avenues to alter the mortality rate. In view of these findings and those reported by Ferguson *et al.* (1966), we conclude that the principal mode of endrin entry into the body of fish is by way of the gill surfaces.

Acknowledgments: Supported by grant ES 00086-02 to D. E. Ferguson from the Office of Resource Development, U. S. Public Health Service.

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STLCOPCB4016844