

Physiological Response of the Mud Crab, *Eurypanopeus depressus* to Cadmium

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Although there is an increasing interest in heavy metals as pollutants in the marine environment, relatively little work has been done to determine the effect of such metals on marine organisms (KATZ et al. 1971, 1972; REISH 1970, 1971, 1972; SELLECK 1970, 1971). Most of these studies have dealt with lethal effects of copper and mercury on fouling organisms (BRYAN 1971). Several investigators have reported heavy-metal effects on adult decapod crustaceans (EISLER 1971; EISLER et al. 1972; RAYMONT and SHIELDS 1964; VERNBERG and O'HARA 1972). There is, however, a paucity of information on the effects of cadmium on this group. The present study was undertaken to determine the toxicity of acute exposure of cadmium to the mud crab, *Eurypanopeus depressus*, as evidenced by mortality and by differential oxygen consumption occurring at non-lethal levels of this contaminant. *E. depressus* was chosen for study because its estuarine habitat is likely to be polluted with heavy metals in the heavily industrialized areas of the Northeast. Cadmium was chosen as the test metal because of the growing awareness of its toxic properties, its persistent nature and its increasing occurrence in the environment. (U. S. COUNCIL ON ENVIRONMENTAL QUALITY 1971).

Experimental

The crabs were collected in rocky, intertidal areas of Milford Harbor, Connecticut, and held in the laboratory in tanks of recirculated, artificial seawater at $21 \pm 2^{\circ}\text{C}$, for at least one week before testing. They were fed minced hardshell clams, *Mercenaria mercenaria*, daily, but were unfed two days prior to and during the test exposure.

Crabs were exposed to cadmium in one-gallon glass jars, filled to three liters with a synthetic medium developed by ZAROOGIAN et al. (1969). This synthetic medium, suggested by LaROCHE et al. (1970) as a standard testing medium, was prepared using technical-grade chemicals dissolved in fresh well-water and adjusted to 25 ppt salinity. The pH of the seawater remained at 7.0 for the range of cadmium concentrations tested. Analysis of the synthetic medium by an independent testing firm indicated that cadmium was present at 0.0045 ppm and that other background metals were negligible. All jars were rinsed with dilute sulfuric and nitric acid between repetitive tests, to prevent any accumulation of

metal residue.

An aqueous solution of cadmium as $\text{CdCl}_2 \cdot 2\frac{1}{2} \text{H}_2\text{O}$ (50 g/l) was added to test jars in various amounts to produce 12 test concentrations. A wide range of concentrations (1.0–48.0 ppm) was used initially to determine final test concentrations, which were from 1.0 ppm through 12.0 ppm. All cadmium concentrations mentioned refer to ppm of the metal ion in solution.

Three crabs, averaging 1 g each, were placed in each test jar of seawater, which was aerated and maintained at room temperature ($21^\circ \pm 2^\circ\text{C}$). Each of ten tests consisted of four jars at each concentration plus four control jars. The crabs were inspected every 24 hrs and dead crabs were removed, weighed, and discarded. Exposure was terminated at 72 hrs with a final tally of mortality and removal of selected live animals for determination of oxygen consumption.

Oxygen consumption measurements were made on whole animals exposed to 0.0, 3.0, 4.0, 6.0, and 7.0 ppm Cd (10–15 animals per concentration) and on excised gill tissue of animals exposed to 0.0, 4.0, and 7.0 ppm Cd (12 animals per concentration). A high mortality rate precluded the use of animals at higher concentrations. The crabs used in these tests ranged in weight from 0.85 to 1.45 g.

Whole animals were held in 100 ml, wide-mouth, Warburg-type reaction vessels containing 50 ml seawater (1 animal per vessel). Excised gill tissues were held in 7 ml Warburg-type reaction vessels containing 2 ml seawater (tissue from 2 animals per vessel). The seawater in each vessel was taken from the corresponding exposure jar from which the test crab was removed. Oxygen consumption was monitored over a 4-hr period in a Gilson differential respirometer. The wet weight of each whole crab and the dry weight (oven-dried to a constant weight) of the gill tissue was recorded upon completion of every experiment. Oxygen consumption was calculated as μLO_2 consumed per hr per g (wet or dry weight).

Results & Discussion

The results from each of ten tests were averaged and the LC_0 , LC_{50} , and LC_{100} values were determined. Values for LC_0 and LC_{100} were determined by observation, whereas the LC_{50} value was derived by graphical probit analysis. The LC_0 , LC_{50} , and LC_{100} values were 1.0 ppm, 4.9 ppm, and 11.0 ppm, respectively, with 95% confidence limits for the LC_{50} being 3.9–5.4 ppm.

The oxygen consumption rates of whole animals showed great individual variation, ranging from 35 to 75 μl oxygen/hr/g. This variability was true of controls and of all exposure groups. There was no difference in over-all oxygen consumption among the various test concentrations. Oxygen consumption rates of the gill tissue alone, however, decreased as the cadmium concentration increased. The mean oxygen consumption rates were as follows: controls, 0.785 $\mu\text{l/hr/mg}$ ($\text{SE}_M = 0.060$); 4 ppm Cd, 0.725 $\mu\text{l/hr/mg}$ ($\text{SE}_M = 0.064$); and 7 ppm Cd, 0.501 $\mu\text{l/hr/mg}$ ($\text{SE}_M = 0.080$). Figure 1 illustrates the relation of survival and gill tissue oxygen consumption to

increasing amounts of cadmium.

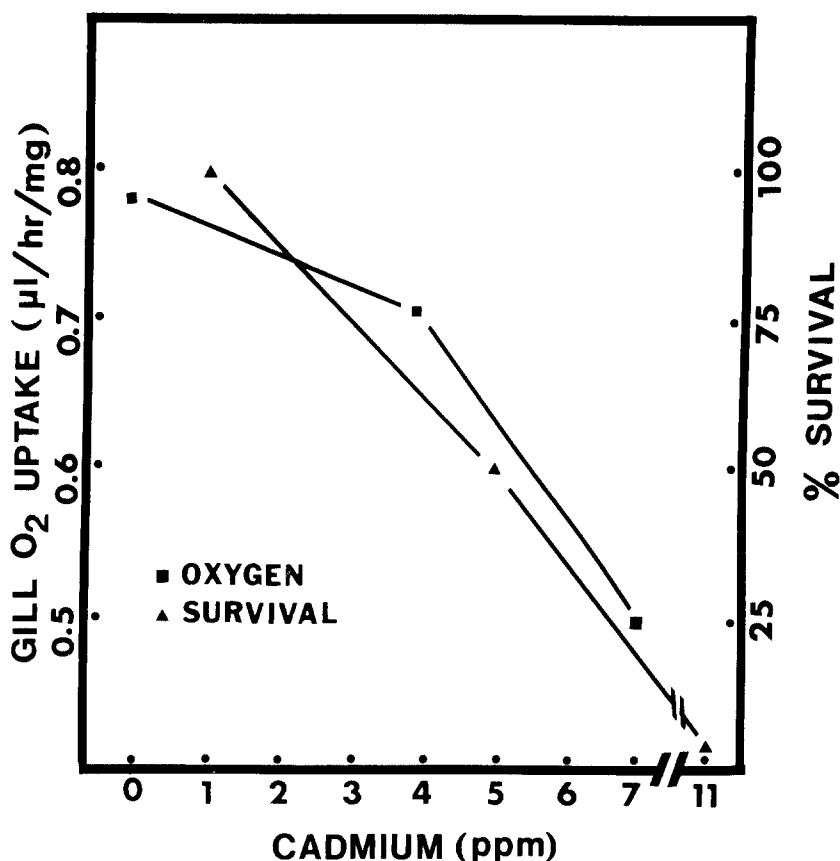


Figure 1. Response of Eurypanopeus depressus to cadmium.

There are certain disadvantages inherent in tests performed in static water. Among these are possible loss of toxicant via aeration, adsorption of the toxicant by the container, and uptake of the toxicant by the test animal (PORTMANN 1970 a & b).

These factors were not critical under the conditions of the present study. Cadmium tends to be very stable and is little affected by aeration. Under a similar experimental regime, EISLER (1971) found less than a 5% cadmium loss from concentrations of 0.1–400 ppm after 264 hrs. It is possible that glass containers adsorb some cadmium, and for that reason they were acid-washed after each experiment to prevent a buildup of the metal. Test animals may also have removed some cadmium, but if so, this uptake was not determined. In a similar experiment conducted at this laboratory with fish, the cunner, Tautoglabrus adspersus, was exposed to cadmium for 96 hours

and the total loss of cadmium from the water was determined to be less than 6%.

Dissolved organics, trace metals, and particulate matter found in natural seawater cause variation in toxicity studies. An artificial medium, whose composition can be precisely reproduced avoids such variation, and was therefore used in this study. Some caution must be exercised in relating the exposure of animals in this medium to the exposure of animals in nature, as the constituents of natural seawater vary so widely.

The tolerance of *E. depressus* to cadmium is similar to that reported by other workers using different species of marine invertebrates. EISLER (1971) found the 96 hr TL50 value for green crabs, *Carcinus maenas*, exposed to cadmium to be 4.1 ppm. CALABRESE *et al.* (in press) found 48 hr LC₀, LC₅₀, and LC₁₀₀ values for embryos of the American oyster, *Crassostrea virginica*, exposed to cadmium to be 1.0, 3.8, and 6.0 ppm, respectively.

Because the whole animals exhibited great variations in oxygen consumption rates, no cadmium-induced changes could be detected. These variations were possibly due to differential activity between individual crabs, because they were held unrestrained and free to move about in the reaction vessels. Gill tissue oxygen uptake, however, showed a graded decrease with increasing cadmium, and may reflect tissue pathology. The killifish, *Fundulus heteroclitus*, was found to suffer hypertrophy of the gill filaments and hyperplasia of the epithelial surface of respiratory lamellar and interlamellar filament epithelium within 20 hrs of exposure to 50 ppm cadmium, less than half the 24 hr TL50 for the species (GARDNER and YEVICH 1970; EISLER 1971). THURBERG (unpublished data) has found gill tissue oxygen consumption rates for the green crab, *Carcinus maenas*, and the rock crab, *Cancer irroratus*, to be reduced by 20-25% after a 48 hr exposure to 2 ppm cadmium.

The results of this work suggest other approaches to the study of heavy metals as pollutants. One logical avenue to pursue is the study of possible synergism or antagonism of several metals in combination. Another is the exposure of embryonic and larval stages of selected marine organisms to metals, because the different life stages often have different tolerances to pollutants. For example, larvae of the green crab, *C. maenas*, when subjected to solvent emulsifiers, yielded LC₅₀ values 3-10 times lower than those for adult crabs (PORTMANN and CONNOR 1968). Another important approach is to determine the effect of long-term exposure of one or more generations to a metal. These suggested studies are certainly necessary for a fuller interpretation of the effects of heavy metals in the marine environment.

References

- CALABRESE, A., R. S. COLLIER, D. A. NELSON, and J. R. MacINNES:
Mar. Biol. (In Press).
- BRYAN, G. W.: Proc. Roy. Soc. Lond. B177, 389 (1971).
- EISLER, R.: J. Fish. Res. Bd. of Canada 28, 1225 (1971).

- EISLER, R., G. E. ZARROGIAN and R. J. HENNEKEY: J. Fish. Res. Bd. Canada 29, 1367 (1972).
- GARDNER, G. R., and P. P. YEVICH: J. Fish. Res. Bd. Canada 27, 2185 (1970).
- KATZ, M., R. S. LEGORE, D. WETCAMP, J. M. CUMMINS, D. ANDERSON, and D. R. MAY: J. Wat. Poll. Cont. Fed. 44, 901 (1972).
- KATZ, M., C. H. WAHTOLA, R. S. LEGORE, D. ANDERSON, and S. McCONNEL: J. Wat. Poll. Cont. Fed. 43, 933 (1971).
- LaROCHE, G., R. EISLER, and C. M. TARZWELL: J. Wat. Poll. Cont. Fed. 42, 1982 (1970).
- PORTMANN, J. E.: FAO Tech. Conf. on Mar. Poll. and its Effects on Living Resources and Fishing FIR: mp/70/E-31 (1970 a).
- PORTMANN, J. E.: FAO Tech. Conf. on Mar. Poll. and its Effects on Living Resources and Fishing FIR: mp/70/G-33 (1970 b).
- PORTMANN, J. E. and P. M. CONNOR: Mar. Biol. 1, 322 (1968).
- RAYMONT, J. E. G. and J. SHIELDS: Int. Conf. on Wat. Poll. Rev., Lond. Oxford: Pergamon Press, (1964).
- REISH, D. J.: J. Wat. Poll. Cont. Fed. 42, 861 (1970).
- REISH, D. J.: J. Wat. Poll. Cont. Fed. 43, 933 (1971).
- REISH, D. J.: J. Wat. Poll. Cont. Fed. 44, 901 (1972).
- SELLECK, R. E.: J. Wat. Poll. Cont. Fed. 42, 861 (1970).
- SELLECK, R. E.: J. Wat. Poll. Cont. Fed. 43, 933 (1971).
- U. S. COUNCIL ON ENVIRONMENTAL QUALITY: Toxic substances. Washington, D. C., U. S. Government Printing Office, (1971).
- VERNBERG, W. B. and J. O'HARA: J. Fish. Res. Bd. Canada 29, 1491 (1972).
- ZARROGIAN, G. E., G. PESCH, and G. MORRISON: Amer. Zool. 9, 1144 (1969).