

The Feeding Behavior of *Mytilus edulis* in the Presence of Methylmercury Acetate

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Mercury in the marine environment was first recognized as a serious problem, with the outbreak of Minamata disease in Japan (ABELSON, 1970). Toxicity has been found to be greatest in the organomercury compounds (PEAKALL & LOVETT, 1972). JENSEN and JERNELOV (1969) found that microorganisms in bottom sediment are able to convert less toxic inorganic mercury to highly toxic methyl- and dimethyl mercury.

The principal site of biological action of mercury is the central nervous system. The toxic action is due to the affinity of mercury for sulfhydryl (SH) groups in proteins (WEBB, 1966); this chemical binding to proteins in cell membranes alters ionic distribution and osmoregulatory activity (PASSOW et al., 1961).

There is also evidence that marine animals accumulate and store mercury in high concentrations (JOHNELS & WESTERMARK, 1969); filter feeding bivalves are also vulnerable to mercury accumulation (MAYER, 1970).

The subject of this investigation is a filter feeding bivalve, Mytilus edulis. Mytilus is found on rocky coasts and in estuaries on the eastern coast of the United States and on the western coasts of Europe (HARGER, 1970). FIELD (1922) has shown that Mytilus can survive great salinity fluctuation (15‰-40‰) and temperatures ranging from -1°C to 28°C, while actively feeding between -1°C and 27°C (LOOSANOFF, 1942). Mytilus utilizes a wide variety of food: dinoflagellates, diatoms, bacteria and organic detritus (COE & FOX, 1942; 1944; FOX & COE, 1943; COE, 1945).

The purpose of this investigation is to determine the effects of sub-lethal levels of methyl mercury upon the feeding behavior of Mytilus edulis.

METHODS AND MATERIALS

The experimental animals, Mytilus edulis, were collected from the littoral zone of an estuary located in Reynolds Channel, Far Rockaway, New York. The animals were maintained in holding tanks at 12°C containing filtering synthetic sea water (Triton Synthetic Sea Salts) at 29‰, and were fed diatoms once a week.

Nitzschia acicularis was chosen because it has been shown to be a preferred food by Mytilus (JORGENSEN, 1949). Although GLOOSCHENKO (1969) showed that small concentrations of mercury (50 parts per billion) are lethal to diatoms, there is no feeding preference by Mytilus whether or not the diatoms are alive (FOX & COX, 1944). This mercury effect on the diatoms would therefore have no effect upon the feeding mussels.

Nitzschia acicularis, was cultured in Erd-Schreiber media (AARONSON, 1970); maintained at pH 8.4 in synthetic sea water 29°/oo and a 12:12 photoperiod.

$\text{NaH}^{14}\text{CO}_3$ (0.25 μCi) was used to label Nitzschia for the feeding experiment. Maximum $^{14}\text{CO}_2$ uptake by the diatoms was determined during continuous light conditions. Activity was measured by withdrawing aliquots every four hours. Diatoms were withdrawn from the culture, serially washed in synthetic sea water, Millipore filtered, and counted on a Coulter Counter. A cocktail mixture of 0.8 g 1,4-bis-2-(4-methyl-5-phenyloxazolyl)-benzene, ("POPOP"); 5.0 g 2,5-diphenyloxazole, ("PPO"); 1000 ml scintillation grade toluene, was prepared and 10 ml added to each vial containing labelled diatoms. To each vial, Cab-O-Sil, a thixotropic gelling agent was added. Activity was measured in a Nuclear-Chicago Mark I liquid scintillation counter. Activity was recorded as counts per minute per diatom (CPM/diatom).

Mytilus was fed at 12°C in polyvinyl chloride (PVC) beakers, five mussels to a beaker. Diatoms were fed to Mytilus in concentrations of 0.4, 0.8, 1.2, 1.6, 2.0, and 2.8 mg/L (parts per million-ppm) methyl mercury acetate, including a control. After Mytilus was exposed to methyl mercury for 24 hours, labelled diatoms were introduced after their activity was determined. A 24 hour period was allowed for feeding; solutions were under constant mixing by magnetic stirring rods to assure that diatoms did not settle out and become inaccessible to the feeding mussels.

At the end of the feeding period, the mussels were removed, washed in ethyl alcohol to remove adhering diatoms, sacrificed and again washed in alcohol. Mussels were weighed and then dissolved in HNO_3 . The tissue was submitted to liquid scintillation counting above. Results were recorded as diatoms/mussel, using data from diatom $^{14}\text{CO}_2$ activity.

$$\frac{\text{CPM/mussel}}{\text{CPM/diatom}} = \text{diatoms/mussel}$$

Additional controls were performed using mussels sacrificed in alcohol and sterilized in synthetic sea water. This was done to account for possible absorption of $^{14}\text{CO}_2$ by bacteria and background effects.

"Feeding in this experiment was defined as labelled diatoms found within either the digestive system, incorporated into the body tissue or on the gill area. All other adhering cells were removed in alcohol washings.

RESULTS

The preliminary experiment to determine maximum $^{14}\text{CO}_2$ uptake in Nitzschia, showed greatest activity after 48 hours of exposure, reaching 21.78 CPM/diatom. Results are shown below in Table 1.

TABLE 1 - $^{14}\text{CO}_2$ Uptake by Nitzschia acicularis

<u>TIME (HOURS)</u>	<u>CPM/DIATOM</u>	<u>TIME (HOURS)</u>	<u>CPM/DIATOM</u>
4	0.63	36	---
8	0.91	40	13.29
12	0.99	44	15.25
16	2.29	48	21.78
20	3.16	52	21.40
24	4.98	56	18.24
28	10.08	72	6.70
32	12.55		

The results of feeding Mytilus edulis in control conditions, showed that the mussels were capable of ingesting and retaining large amounts of diatoms ranging from 145 to 34,045 cells per day. All concentrations of methyl mercury resulted in decreased feeding. The data is compiled in (Table 2) and mean feeding rates are illustrated in (Figure 1).

Statistical treatment of data showed significant differences at all concentrations except between 8 and 12 ppm, and 24 and 28 ppm. Data was analyzed by a student's "t" test for unpaired comparisons (Table 3).

TABLE 3 - Student's 't' Test for Unpaired Comparisons

<u>Concentration</u>	<u>t value</u>
0-0.4 ppm	t ₅₆ = 4.406*
0.4-0.8	t ₄₈ = 4.440*
0.8-1.2	t ₄₈ = 0.012 ^{ns}
1.2-1.6	t ₄₈ = 2.235*
1.6-2.0	t ₅₅ = 4.677*
2.0-2.4	t ₆₂ = 2.889*
2.4-2.8	t ₅₃ = 0.066 ^{ns}

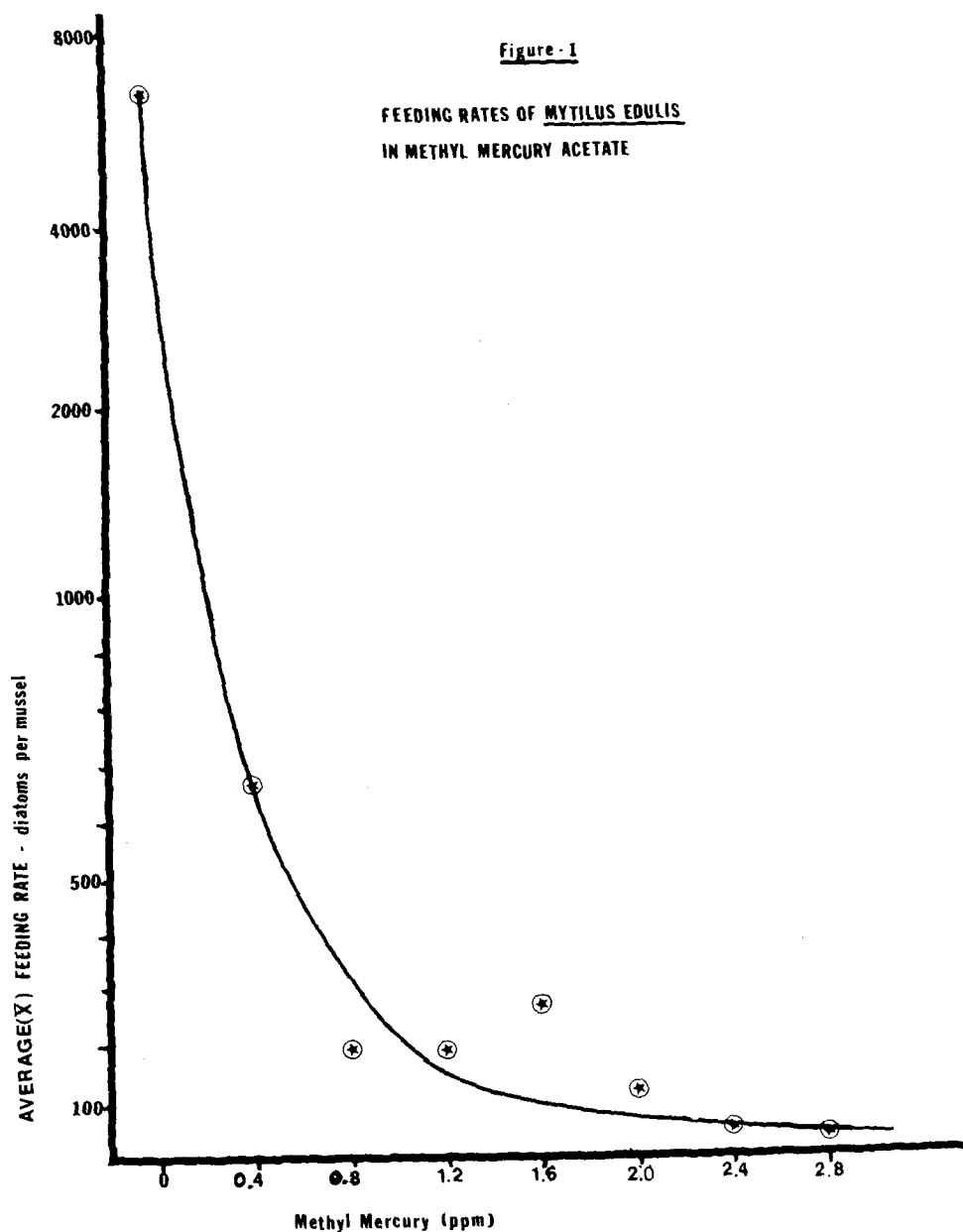
Legend: t degrees of freedom

ns - not significant
* - significant (P<0.05)

TABLE 2 - Analysis of Feeding Behavior Data

STATISTIC	Methyl Mercury (ppm)							Sterile Control
	0	0.4	0.8	1.2	1.6	2.0	2.4	
ΣX	221243	16436	4545	4657	6684	3933	1819	1111 92
\bar{X}	6704.33	659.96	181.80	186.20	267.36	122.90	53.5	52.9 23
S	5254.32	532.42	111.52	141.91	316.17	114.44	37.56	36.30 2.94
N	33	25	25	25	25	32	34	21 4

Σx = total diatoms; \bar{X} = mean; S = standard deviation; N = sample



During the entire experiment, the mussels remained open more than 99% of the time and showed active filtering. There was no significant mortality in any concentration, although some deaths occurred at 2.8 ppm methyl mercury.

DISCUSSION

Mercury was shown to have a harmful effect upon feeding in the mussels tested. A decline in feeding was showed as the mercury concentrations were increased.

Respiration was found to increase with increasing mercury concentration in Conger (DORN, 1974). The decrease in feeding of Mytilus with increasing mercury concentration can be related to stress on the animal's physiological state.

TAKAHASHI AND MURAKAMI (1968) found that the nervous system controls ciliary activity in Mytilus. It is possible that since mercury inhibits nervous activity (WEBB, 1966), decreased feeding may be caused by disruption of ciliary activity.

The results of this study have shown the severe effect of methyl mercury upon one aspect necessary for life functions in living organisms. Decreased feeding rates can be an indication of toxicity of an environment to its inhabitants.

ACKNOWLEDGMENTS

I wish to thank DR. H.D. Freudenthal, DR. G.W. Bane and DR. R.J. Valenti for their technical assistance.

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