

TOXICITY TESTS OF OIL DISPERSANTS ON SOME MARINE ANIMALS

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ABSTRACT

Toxicity of four oil dispersants, manufactured in India, were tested on some marine animals, using static bioassay technique. Experiments have shown that, inspite of major differences in physico-chemical properties, all the four dispersants are toxic, and hence their use should be restricted. Effects of furnace oil mixed with each of the four dispersants indicated that oil-dispersant mixture reduced the toxicity markedly.

INTRODUCTION

The magnitude of oil pollution in the marine environment, especially in the coastal zone, is increasing rapidly. Ecological effects of oil contamination on marine biota are varied and are well documented (Corner, Southward and Southward, 1968; Nelson-Smith, 1968 & 1972; McCaul, 1969; Za Roche-Eisler and Tarzwell, 1970; Blumer, 1973 and Dow, Hurst Jr., Mayo, Gogger, Donovan, Gunbarde, Barry and Zevich, 1975).

As reported earlier (Dwivedi and Parulekar, 1974), oil pollution in the coastal waters of India, is fairly widespread. Various mechanical and chemical methods are commonly used for controlling the oil pollution at sea. Solvent emulsifiers or dispersants are widely used for this purpose. Generally a dispersant consists of a solvent for dissolving the oil; a surfactant which acts as an emulsifier and a stabilizer for making the emulsion stable. The aromatic component in the solvent is the main cause for the toxicity of a dispersant (Nelson-Smith, 1972). Toxicity studies on different dispersants have shown that the disper-

sant, at times, is more toxic than the oil itself (Portman and Connor, 1968; Eisler, 1975). So far, no studies on the effect of oil dispersant on marine life have been made in India.

Recently, some Indian companies have started manufacturing dispersants indigenously. This gave an opportunity to study the toxic effects of four different oil dispersants on marine organisms. As seen in Table I, all the 4 dispersants tried by us are quite different from each other and except for the general appearance, they differ in their physical properties. Similarly, they also differ in their active chemical components which has been determined by some of our colleagues in NIO (Solimabi and Topgi, personal communication).

MATERIAL AND METHODS

Gift samples of four different dispersants were received from the manufacturers. As the commercial names are covered by registered trade mark, the dispersants were coded by us as 1, 2, 3 and 4. Furnace oil was used to prepare oil dispersant mixture for undertaking the toxicity tests.

Table 1. Showing some of the physical properties of different oil dispersants used in the experiments.

Characteristic	Disp. 1	Disp. 2	Disp. 3	Disp. 4
Appearance	Light yellow	Yellow and slightly turbid	Light yellow clear liquid	Light yellow clear liquid
Viscosity	Non-viscous	Non-viscous	Highly viscous	Viscous but less than disp. 3
Solubility	Insoluble in water; partly soluble in alcohol but completely soluble in acetone	Insoluble in water; partly soluble in alcohol but completely soluble in acetone	Soluble in water; alcohol and acetone	Soluble in water, alcohol and acetone
pH	6.5 - 7.5	7.95	5.3	5.3

The test animals used were the green mussel (*Mytilus viridis*), mole crab (*Emerita holthuisi*) and prawn (*Macrobrachium idella*). The animals after their collections, were kept in the laboratory aquarium tanks for about 72 hours. Before the experiments, the length and weight of all the test animals were recorded.

Experimental work was carried out from February to May 1977. Static bioassays with green mussels were conducted in rectangular glass tanks of 2 litre capacity for a period of 168 hours. Experiments on the other two species were performed in glass jars of 1 litre capacity for a maximum period of 8 hours. Continuous slow aeration was maintained throughout the experiments for keeping the dissolved oxygen content above 4 ml/l. Salinity of the sea water used was about 35‰ and all the experiments were conducted at room temperature of 28-30°C ($\pm 1^\circ\text{C}$). No feeding of test animals was done 48 hours before the experiments or during the experiments.

Standard solutions of dispersants and oil dispersant mixtures were prepared in filtered sea water. Preparation of oil dispersant mixture was done by mixing the furnace oil and dispersant in the proportion of 1 : 1 in 500 ml of filtered sea water. The mixture was well agitated in a separating funnel and then it was allowed to settle for about 2-3 hours. The mousse-like material formed at the surface was removed and the remaining liquid was used for preparing the different concentrations.

The test containers were inspected at fixed intervals, and the dead animals were removed. Records were also maintained for any abnormal behaviour of the test animals. In the case of green mussels, symptoms of ill health, such as, failure to adhere to the walls by new byssal threads, slowness in closing of valves after any mechanical stimulus and profuse secretion of mucous in comparison to controls, were recorded. Complete inactivation of adductor

muscles and mantle cavity muscles as used as a criterion for mortality of the mussels. In the case of mole crabs and prawns inactivation of appendages was used as an indicator of death and then such animals were transferred to clean sea water to test their recovery.

RESULTS AND DISCUSSION

Results, presented here, are based on the mean values of at least three experiments. LC50 values for dispersant 1, 2, 3 and 4 were calculated and these are shown in Tables II and III. Mortality occurred within 48 hours of the exposure. Observations during the rest of the period, upto 168 hours, did not show any further mortality. Thus the maximum effect of dispersant on the test animals, occurred within 48 hours and,

Table II. LC50 values in green mussels (*Mytilus viridis*) after 24, 48 and 96 hours exposure of different dispersants.

Dispersant	24 hr LC 50 ppm	48 hr LC 50 ppm	96 hr LC 50 ppm
1	> 10	> 10	> 10
2	0.16	0.09	0.09
3	0.085	0.079	0.079
4	0.09	0.08	0.08

Table III. Concentration of dispersants at which 0%, 50% and 100% mortality was observed in *Mytilus viridis* after 48 hours of exposure.

Dispersant	LC 0% ppm	LC 50% ppm	LC 100% ppm
1	> 10	> 10	> 10
2	0.025	0.09	0.16
3	0.04	0.079	0.1
4	0.04	0.08	0.1

with the passage of time they could adapt themselves to the changed environment.

Mytilus viridis, when subjected to the maximum concentration of 10 ppm of dispersant 1, did not show any mortality upto 96 hours. Mussels reacted quickly to the mechanical stimulus by closing the valves and about 70% of the animals showed the ability to adhere to the glass walls by secreting new byssal threads. Secretion of copious mucous was observed at 10 ppm concentration of dispersant 1, whereas below this concentration, animals behaved as normally as the controls.

Ten different concentrations of dispersant 2, ranging between 0.01 and 0.32 ppm V/V, showed no secretion of mucous and all the test animals showed an attachment with byssus in the concentration of 0.025 ppm upto 168 hours, i. e., till the last day of the experiments. Above 0.06 ppm concentration, secretion of byssal threads got retarded and response to mechanical stimulus also became weak (Tables II and III).

Effects of dispersant 3 and 4 were almost similar, with LC50 values at 0.079 and 0.08 ppm, in an exposure of 48 hours. Even at 0.08 ppm concentration, mussels could secrete byssal threads. Response to closing of valves was quick at concentrations lower than 0.08 ppm, indicating that the damage to adductor muscles was less than that caused by dispersant 2.

Experiments with *Emerita holthuisi* and *Macrobrachium idella* were slightly modified to suit the conditions such as are likely to occur in the field. The application of dispersant in the field is generally done at higher concentration for their maximum dispersal under varying

wave actions. Hence, the mole crab (*E. holthuisi*) and the prawn (*M. idella*) were subjected to higher concentrations of dispersants and oil-dispersant mixtures and accordingly, the mortality was recorded at short intervals of time. From these observations (Table IV), time for 50% death (Tm 50) in each of the different concentrations were determined and these are shown in Figs. 1 and 2.

Table IV. Showing time at which 50% and 100% mortality was observed in the mole crab (*Emerita holthuisi*) at various concentrations of dispersants and oil-dispersant mixtures.

Substance	Conc in ppm	Tm 50 in minutes	Tm 100 in minutes
Dispersant 2	0.5	150	>420
	1.0	120	360
	2.5	96	300
	5.0	54	180
Disp. 2 + oil mixture	0.5	1260	>1440
	1.0	420	>1440
	2.5	192	420
	5.0	168	420
Dispersant 3	1.0	>420	>420
	2.5	>420	>420
	5.0	>420	>420
(No mortality upto 420 min. of exposure)			
Disp. 3 + furnace oil mixture	1.0	No mortality upto 420 min. of exposure	
	2.5	420 min. of exposure	
	5.0	exposure	
Dispersant 4	1.0		
	2.5	- do -	
	5.0	- do -	
Disp. 4 + furnace oil mixture	1.0		
	2.5	- do -	
	5.0	- do -	

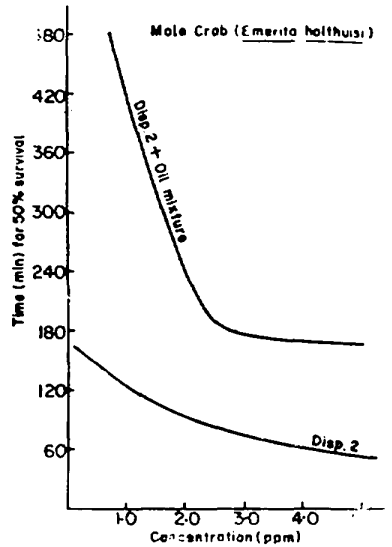


Fig. 1. Effect of dispersant and oil dispersant mixture on mole crab (*E. holthuisi*).

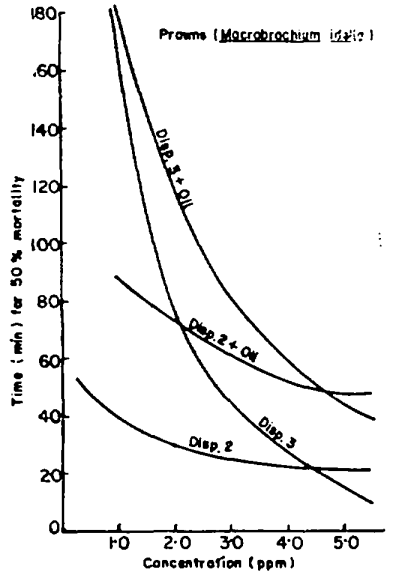


Fig. 2. Effect of dispersant and oil dispersant mixture on prawn (*M. idella*).

The results indicate that the mole crab is more resistant than the prawn. No mortality of mole crab occurred within 8 hours of exposure even at the highest concentration of 5 ppm of dispersants 3 and 4 as well as in the oil-dispersant mixture. It was also noted that the animals subjected to dispersant 4 were more active than those in dispersant 3. The Tm 50 of mole crab in dispersant 2 plus oil emulsion at a concentration of 5 ppm was found to be 54 and 168 minutes respectively. For the prawn, the dispersant 2 was found to be more toxic than the dispersant 3. From Figs. 1 and 2, it is evident that the oil-dispersant mixture reduces the toxicity of the dispersant.

Observations on the recovery of partially affected test animals, on their transfer to clean sea water showed that the prawns subjected to dispersant 2, did not recover, whereas about 90% of the prawns exposed to dispersant 3 and to its oil emulsion mixture showed a capacity to recover by attaining free swimming activity within 2-3 hours. In the mole crab, nearly 40% of the animals exposed to different concentrations of dispersant 2, showed less capacity to recover whereas animals exposed to dispersants 3 and 4 remained active throughout the duration of experiment.

DISCUSSION

Toxicity experiments, reported here, indicate that the dispersants 2, 3 and 4 are more toxic than the dispersant 1. Swedmark, Granmo and Kolberg (1973) while comparing the toxic effects of old and new oil-dispersants, have shown that the new dispersants are less toxic than the old ones. This difference in toxicity is due to their decrease to a minimum level in the solvent compo-

nent in new dispersants (Nelson-Smith, 1972). The dispersants 2, 3 and 4 appear to be of the old type.

While reporting the various effects of dispersants on marine organisms, Bardach, Fujiya and Holl (1965) observed that at a concentration of 0.05-10.0 ppm, the senses of fish to detect chemicals are greatly affected. Schmid and Mann (1961), Scheir and Cairns (1966) and Kunhold (1970) noted the erosion of gill epithelium and destruction of mucous cells when fishes were exposed to sublethal doses of dispersants. In the present study the excessive secretion of mucous and their failure to attach with new byssal threads, even in low concentrations of some of the dispersants, was observed in *M. viridis*. This could have occurred due to irritation resulting in the damage of epithelium of the green mussel.

Van Ouerbeek and Blondean (1954) have demonstrated that the hydrocarbons and surfactants can disrupt the protein layer and the general cell structure of the plasma cell membrane. During short exposures this effect was found to be reversible. In the present series of tests such a revival was clearly shown by prawns when they were transferred to clean sea water subjecting to high concentrations of dispersant 3.

As can be seen from Figs. 1 and 2, the toxicity of dispersant is far greater than when it is used as oil emulsion mixture. Tm 50 of dispersant 2 at 3 ppm concentration was 75 minutes for *E. holthuisi*, whereas at the same concentration, Tm 50 value of dispersant 3 for *M. idella* was 46 minutes. When used as oil-emulsions, Tm 50 values for *E. holthuisi* and *M. idella* were 60 and 81 minutes respectively. This has also been

observed by Portman and Connor (1968) and Eisler (1975). The decrease in toxicity of oil-emulsion mixtures is due to the inactivation of the toxic component of dispersant by oil which is a solvent fraction. Moreover, when dispersants are sprayed on spilled oil or in oil films, a considerable amount of dispersant mixes with oil, thus reducing the concentrations of free dispersants. A quantitative study of these aspects are in progress (Solimabi and Topgi, personal communication).

The results of our experiments, indicate that all the four dispersants tested are toxic. The relative toxicity varies between the dispersants. It was maximum in dispersant 2 and minimum in dispersant 1. This was further substantiated by the experiments on recovery of tests animals in dispersant free water. The rate of recovery was minimum with dispersant 2. As reported by Nelson-Smith (1972), the toxic effects of a dis-

persants are mainly due to the aromatic component in the solvent part. From the observations on the reduction of toxicity of dispersants when mixed with oil, it is encouraging to note that the use of dispersant for emulsifying the spilled oil is not likely to be very toxic to marine biota. Improved formulation can further reduce the toxicity of dispersants (Sweden, Granmo and Kollberg, 1973). It is, therefore, evident that although dispersants provide one of the most effective ways for fighting oil pollution at sea, the dispersants manufactured in India will need further refinements to make them safer to marine life.

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