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The Effectiveness and Toxicity of the Oil Dispersant, Oilsperse 43, In Large Outdoor Tanks

Surveillance Report EPS 5-AR-75-8
Atlantic Region

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THE EFFECTIVENESS AND TOXICITY OF THE OIL DISPERSANT, OILSPERSE 43, IN LARGE OUTDOOR TANKS

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ATLANTIC REGION

APRIL, 1975

EPS-5-AR-75-8

ABSTRACT

Use of the dispersant, Oilsperse 43, increased the dispersion of Venezuelan Guanipa crude oil. The resulting mixture was more homogeneous and the oil slick less viscous than in the oil tank. The dispersant appeared to retard formation of the familiar "crust" on the surface.

A weathered crude oil plus dispersant mixture with an oil concentration of 250 µg/l was lethal to over 50% of the test organisms, green sea urchins, within 4 days. No mortalities occurred among urchins exposed to the crude oil treatment.

If similarly effective, "low-toxicity" dispersants are used on this crude oil spilled in an inshore marine area, concentrations of oil and dispersant lethal to the green sea urchin could conceivably be attained.

Résumé

L'emploi de l'agent de dispersion Oilsperse 43, a augmenté la dispersion du pétrole brut Venezuelien Granipa. Le mélange résultant était plus homogène et la nappe d'huile était moins visqueux qu'auparavant dans le réservoir. L'agent de dispersion a paru retarder la formation de la "croûte" normallement trouvé sur la surface.

Un mélange d'agent de dispersion et de pétrole usé ayant une concentration d'huile de 250 ug/l était fatal pour plus de 50% des oursins verts en 4 jours. Il n'y avait pas de fatalités chez les oursins qui avait été seulement exposés au pétrole brut.

Si un agent de dispersion pareillement efficace et de toxicité basse est utilisé sur une nappe de pétrole près de la côte, il est possible qu'un mélange fatal de pétrole et d'agent de dispersion pourrait se produire.

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1 INTRODUCTION

During oil spills in Canadian waters, chemical dispersants can be used under special circumstances. To control their use, the Environmental Emergency Branch of the Environmental Protection Service has prepared a set of guidelines describing preliminary laboratory procedures for determining the effectiveness, toxicity and degradability of dispersants (Anon., 1973). The tests used to evaluate the dispersants are restricted by the size of the experimental tanks, extent of chemical analysis and use of fresh water and rainbow trout as the testing system. It was felt that these restrictions might significantly affect or limit the evaluation of particular dispersants, especially in view of their frequent use in marine waters.

Two groups at the Bedford Institute of Oceanography (the Environmental Protection Service and the Marine Ecology Laboratory) conducted a 31 day experiment to determine the effectiveness and toxicity of an oil spill dispersant with crude oil in marine conditions, simulated using large-volume, outdoor tanks. The objective was to initiate a comparison between the fate and effects of dispersants under laboratory conditions, following the guidelines (Anon., 1973), and under simulated field conditions. The specific objectives were: 1) to determine the effectiveness of the dispersant by comparing concentrations of petroleum hydrocarbons in the water column of an oil and sea water tank and an oil plus dispersant and sea water tank over the 31 day test period; and 2) to determine the acute toxicity of the undiluted solution from the respective tanks by estimating the LT50's (median lethal times) of the water to the boreal green sea urchin Strongylocentrotus droebachiensis, in 4-day static tests.

Sea urchins were chosen as test organisms because

of availability, presence in inshore areas, suspected sensitivity to changes in water quality and ease of maintenance in the laboratory. In addition, little is known about effects of naturally or chemically dispersed crude oil on sea urchins. North (1967) reported many dead sea urchins, Strongylocentrotus spp. after the spill of diesel oil from the TAMPICO MARU off California in 1957. According to Nelson-Smith (1970), laboratory tests by North and his associates showed that a 0.1% emulsion of the oil inactivated tube feet of the urchins; urchins died when exposed to emulsions for more than an hour. Urchins (S. droebachiensis), in the vicinity of the ARROW spill in Chedabucto Bay, were contaminated with levels of 21-372 µg of Bunker C per gram of thin tissue, compared to levels of 22 µg/g in urchins from control areas (Scarratt et al, 1970). No apparent adverse effects on urchins were noted in this study. Allen (1971) observed no effects of various crude and refined oils on the fertilization of eggs of S. purpuratus but found that water extracts of the oils were highly toxic to cleaving eggs over short exposure periods. In the most recent study, Phinney (pers. comm.) noted that feeding in S. droebachiensis was affected at 200-700 ppb dispersed No. 2 fuel oil, while attachment was affected at 30,000 ppb but not affected at 18,500 ppb in 8 hr tests.

From the above, there was an apparent need to investigate the fate and toxicity of oil plus dispersant and oil mixtures at concentrations likely to be encountered in the area of a spill. In addition, none of the studies with sea urchins investigated the acute toxicity of oil plus dispersant mixtures but suggested the use of urchins as sensitive test organisms.

2 METHODS AND MATERIALS

2.1 Experimental Design

The experimental tanks were polyvinylchloride-lined, metal-walled swimming pools, 12 ft in diameter by 3 ft deep, situated on the eastern shore of Bedford Basin. Surface water from Bedford Basin was pumped into the tanks and was allowed to settle for 24 hr prior to the experiment. The water level was 15 cm below the top edge, resulting in approximately 8000 l of water in each tank. The temperature of the water was occasionally measured and dissolved oxygen was determined at the end of the experiment using a Radiometer Model PHM 71 acid-base analyzer with a Model PHA 930 oxygen module. Both tanks were fitted with siphons made from 1/2" i.d. Tygon tubing before oil was added to allow sampling without encountering the surface film. One tank was fitted with an electric stirrer which was used to agitate the oil plus dispersant mixture.

The oil used in the experiment was Venezuelan Guanipa crude which had been stored at 0 C in sealed metal cans. The dispersant, Oilsperse 43, was supplied by the manufacturer, Diachem of B.C., Ltd. It had been determined effective and of acceptable toxicity in tests by E.P.S., Atlantic Region, as set forth in the guidelines (Anon, 1973).

The experiment began at 1000 hr on 1 October, 1974. Surface water temperature was 12 C. One liter (± 100 ml) of oil was poured into one tank and one liter of oil plus one liter of dispersant were simultaneously added to the other tank. The dispersant was applied directly to the oil in two ways. Approximately two-thirds was sprayed onto the oil using a common garden spray gun. The rest was poured onto the oil in the wash of the stirrer.

At the end of the experiment, remaining oil was mechanically recovered from the sides and surfaces of the

tanks and weighed so that an approximate oil budget could be determined for each tank.

2.2 Chemical Analysis

The water column in both outdoor tanks was sampled in duplicate on seven occasions at mid-depth with a one liter glass sample bottle. The open bottle, clamped to a metal rod, was lowered quickly through the surface film. It was retrieved through the area cleared by bubbles formed as the bottle filled. The concentration of petroleum hydrocarbons in the samples was estimated by fluorescence analysis (Gordon and Keizer, 1974).

Samples of water were obtained from each bioassay tank by dipping a one liter glass bottle into the tank at 0-2 hr and again at 96-98 hr. Samples were analysed as above.

The features and shortcomings of fluorescence spectroscopy for measuring the concentration of "oil" extracted from seawater have been discussed previously (Keizer & Gordon, 1973). This experiment was an ideal situation for using fluorescence analysis. The type of oil was known and sampling was conducted under conditions minimizing problems due to sampling error and contamination. Fluorescence interference from the dispersant was negligible. Dispersant added to seawater in the ratio used in the outdoor tanks resulted in fluorescence equivalent to approximately 6 µg of oil/l. This was less than 3% of the lowest concentration of oil found in the oil plus dispersant tank.

2.3 Bioassays

At 48, 168, 336 and 648 hr from the start of the experiment, samples for 96 hr acute toxicity bioassays were withdrawn through siphons from midwater of both tanks. The bioassays are referred to as numbers 1-4. On each sampling date, 40-50 l of water were siphoned from the outdoor tanks

and transferred to bioassay test tanks in pre-rinsed plastic buckets. There were two test tanks for each of three treatments - 1) oil plus dispersant; 2) oil; and 3) seawater control. The water for the seawater control was taken from Bedford Basin.

Five urchins were placed in each tank. They were collected from Paddy's Head, near Peggy's Cove, Nova Scotia, in late September, 1974, and were maintained at ambient temperatures in tanks with well aerated, flowing seawater. They were not fed in the laboratory before or during the toxicity tests, but they occasionally cannibalized a weaker urchin in the holding tanks. The urchins were transferred in seawater to the test tanks no longer than 3 hr after the test solutions had been withdrawn from the outdoor tanks. Solutions were then slowly brought to test temperatures (Table 1).

Observations were made on the sea urchins every 24 hr throughout the test. The position of each urchin was observed; those clinging to the sides of a tank were considered alive and healthy. Urchins on the bottom of tanks were examined individually, gently prodded to determine extent of attachment by their tube feet, and then examined to determine condition and responsiveness of their spines and appearance of their peristomeal membrane. All urchins were removed from each tank to minimize bias due to handling. They were then quickly returned to the bottom of their respective tanks. At the end of each bioassay, diameters of urchin tests were measured using vernier calipers.

Temperature, oxygen concentration and pH were measured throughout each test. Salinity of the control seawater was monitored occasionally and was 30.9 - 31.1^{o}/oo for the entire test period. The light regime approximated a 12 hr on - 12 hr off cycle, at 1-5 ft-c.

3 RESULTS

3.1 Outdoor Tanks

3.11 Visual Observations

As the oil was poured into the oil tank, it spread out to cover the entire water surface. In some places the oil film was several millimeters thick while in other areas there was only an iridescent film. After three days much of the oil had formed long strings and lumps of tar-like material and there was a viscous coating of oil on the sides of the tank, 5-10 cm above the water line. By the end of the experiment, all of the oil remaining on the surface was of tar-like consistency, either in the form of lumps or a crusty coating on the sides of the tank.

The behavior of the oil plus dispersant mixture was much different. The oil plus dispersant mixed with the sea water and formed an opaque, milky white solution which persisted for the duration of the experiment. The oil was broken into small drops which were mixed throughout the tank. An oil slick did not form in this tank until approximately one hour after the oil was added. This slick was much less viscous than in the oil tank and was several millimeters thick. Oil on the sides of this tank was also less viscous than in the oil tank. During the experiment, the oil in this tank did not form tar lumps nor did a "crust" appear on the surface of the exposed oil.

3.12 Physico-Chemical Measurements

The temperature in both outdoor tanks was initially 12 C, rose to 13.5 C on day 2 and by day 20 had dropped to 2.5 C. There was a corresponding drop in air temperature during this period.

From past experiments it was known that the concentration of oxygen, in tanks to which oil had been added, remained close to saturation for up to four months. At

the end of this experiment, duplicate samples from mid-depth in both tanks were taken for oxygen determination. The oxygen concentration was 9.38 mg/l (82.5% saturation) and 11.38 mg/l (100% saturation) in the oil plus dispersant and oil tanks, respectively.

3.13 Chemical Analysis

The results of fluorescence analysis of all samples taken from outdoor tanks are given in Table 2. In the oil tank, as observed in previous experiments (Gordon et al, 1975), concentrations reached a "steady state" of 25 to 30 µg/l after two weeks. Prior to this, the concentrations were higher and duplicates varied greatly indicating a non-homogeneous mixture.

In the oil plus dispersant tank initial concentrations were much higher but dropped off from 16,700 to 3,600 µg/l in 4.5 hr. There was much better agreement between duplicate samples, indicating a more homogeneous mixture. After two weeks the concentration of oil had dropped to 253 µg/l and remained relatively constant for the duration of the experiment.

Synchronous excitation of emission spectra (Lloyd, 1971) were obtained for selected samples throughout the experiment. There was no distinguishable change in the spectra for the samples during the 31 day experiment from either tank which indicated no major changes in the composition of the high molecular weight polycyclic aromatic hydrocarbons.

3.14 Oil Budget

More oil was lost over the one month period from the oil plus dispersant tank than the oil tank (Table 3). The value most likely to be inaccurate is that for the "tar" collected after 31 days (Table 3). It was impossible to collect only "tar"; some water as well as foreign objects

which had been coated with oil were also collected. Likewise, it was not possible to remove all the oil from the sides of the tanks, particularly in the oil plus dispersant tank where the oil was less viscous. Therefore the estimate for the amount of oil lost due to weathering, which is the difference between the oil added and the oil recovered, may be low.

From past experience (Gordon et al, 1975) the amount of oil settling to the bottom of the oil tank in this time period is very small. However, no data were collected on this aspect of the fate of the oil in the oil plus dispersant tank.

3.2 Bioassays

3.21 Visual Observations

During the 96 hr of each bioassay conducted, a visible sheen of oil appeared on the surface of the oil treatments. This did not occur in the oil plus dispersant treatments. The latter were highly turbid throughout each bioassay.

3.22 Physico-Chemical Measurements

The temperature at which the four individual bioassays were conducted varied from 13.3 C to 16.0 C (Table 1). However, all test solutions during a single bioassay were at the same temperature. The pH and oxygen concentrations in the oil plus dispersant tanks were consistently lower than in the oil and control tanks.

The sea urchins were approximately the same size for each of the trials (Table 4).

3.23 Chemical Analysis

The initial concentrations in the bioassay tanks (Table 5) were in general agreement with concentrations found at the same time in the outdoor tanks. No changes in the

synchronous spectra for any tank were observed except in one case of apparent contamination of unknown origin.

In the first bioassay there was a marked decrease, from 1428 to 423 µg/l, in the concentration of oil in the oil plus dispersant test tanks. This may be attributed to the movement of some of the dispersed oil to the surface as was observed in the outdoor tanks during this period. Initial and final oil concentrations in the oil plus dispersant tanks were similar during the other three bioassays. Final concentrations of oil were lower than initial concentrations in oil and control treatments of 3 of 4 bioassays.

The small sample sizes precluded statistical analysis of the data.

3.24 Toxicity

In each bioassay, 50 to 100% of the sea urchins exposed to the aged oil plus dispersant treatment died between 48 hr and 96 hr (Table 6). There was no mortality in either the aged oil or control treatments in any bioassay. In two bioassays, moribund urchins from the oil plus dispersant treatment were returned to clean water at 96 hr; none survived longer than 1-2 days.

The behaviour (i.e. position) of the sea urchins in each tank was observed at 24 hr and 48 hr (Table 7). In the oil plus dispersant treatment, the urchins remained stationary and unattached until death. In the control and oil treatments, with one exception, 50% or more of the animals were found climbing on the sides of the tanks.

4 DISCUSSION

A maximum of 16% of the oil was dispersed in the oil plus dispersant tank compared to only 0.4% in the oil tank. Several factors affected these values. The oil was spilled and the dispersant was added under ideal conditions; the oil was confined over calm water and the dispersant was applied as the oil was spilled. However, the agitation supplied by the stirrer and the wave action may have been less than encountered in field conditions. How these factors balance one another is not known. Therefore the values obtained for dispersed oil could be either high, low or similar to those obtained in a field situation.

Similar problems exist with the values obtained for the oil tank. Field conditions are more severe with regard to wave action and dispersion of the oil over a greater area would result in dilution of oil concentrations. It is known, however, that peak concentrations which occurred in this and other tank experiments (Gordon et al, 1975) were similar to those encountered under spills on open waters.

The use of the dispersant also resulted in a higher concentration of oil persisting in the water column after 2 weeks, i.e. 200 to 300 µg/l for the oil plus dispersant tank versus 20 to 30 µg/l for the oil tank. While this dispersed oil would be further dispersed by wave and tidal action under natural conditions, these high concentrations could persist in confined areas. In a moderately polluted harbour such as Halifax Harbour, concentrations of oil seldom exceed 20 µg/l except in the vicinity of recent spills (Michalik and Gordon, 1971).

The dispersant appears to increase the "weathering" of the oil. "Weathering" includes all of those natural mechanisms such as biodegradation, evaporation and oxidation

which result in the physical disappearance or chemical change of the oil. While there are some uncertainties in the values given in Table 3, there is sufficient evidence to indicate that the dispersed oil weathers more rapidly than the oil alone. After 31 days, 50% of the oil could not be accounted for in the oil plus dispersant tank, while only 24% had disappeared from the oil tank. The absence of the familiar crusty coating from the floating dispersant treated oil could be the reason for an increased rate of weathering of the oil. The stranded oil (i.e. oil on the sides of the tank) in the oil tank was covered with a crusty coating as were the tar lumps collected from the water.

The oxygen concentration and pH of samples from the oil plus dispersant tank were lower than in samples from the oil tank. The dispersant was directly or indirectly responsible for this difference. The lower concentration of oxygen may have been due to a high biological oxygen demand of the dispersant or the opaqueness of the solution may have resulted in decreased respiration by photosynthetic organisms. The pH change may have been due to the slight acidity of the dispersant as reported by the manufacturer.

Concentrations of oil and dispersant over the duration of the experiment were lethal to 50% or more of the urchins over 96 hr. In the oil plus dispersant treatment, there was 187-1427 µg/l oil at 0 hr in the exposure period (Table 5). Hence, the observed toxicity is in contrast to Phinney (1975) who found that, using a No. 2 fuel oil in eight hour tests, attachment of sea urchins was affected at 30,000 µg/l but not at 18,000 µg/l. No mortalities were observed at these concentrations. Hence, the results suggest that the dispersant in this study caused most of the acute toxicity and that there may be sublethal effects at concentrations of oil and dispersant lower than observed in the tests (i.e. < 187-1427 µg/l).

Only part, if any, of the observed acute toxicity in this study may have been due to the chemically dispersed oil. This is suggested by recent studies on the toxicity of oil dispersions, prepared by agitation alone, to sensitive planktonic crustacean larvae. Four day LC50's based on measured concentrations were 3.2 µg/ml No. 2 fuel oil for rock crab larvae (Vaughan, 1973), > 19.8 ppm WSF (water soluble fraction) of crude oil for Penaeus post-larvae (Anderson et al, in press) and 2.3 (95% C.L. 1.4-3.9) mg/l Venezuelan crude oil for Homarus larvae (Wells, 1975). Only North's experiments (Nelson and Smith, 1970) have demonstrated lethality of a 0.1% emulsion of diesel oil to urchins after short exposure times. It is considered unlikely that urchins in this study died due to a 4-day exposure to 187-1427 µg/l dispersed crude oil. Consequently, most of the toxicity of the oil plus dispersant treatment is attributed to the dispersant. Similar future experiments should include a dispersant tank to verify this conclusion.

There were no toxic effects of initial concentrations of 26-35 µg/l of crude oil. These levels are generally above background levels measured along the northeastern U.S. and eastern Canadian coasts (Anon., 1975).

As observed by both North (Nelson-Smith, 1970) and Phinney (1975) where dispersions of diesel oil and No. 2 fuel oil in high concentrations inactivated the tube feet of urchins, tube feet were rapidly affected by immersion into the oil plus dispersant treatment in this study. No such effect was observed in the oil treatments. These observations collectively suggest a rapid, perhaps irreversible, malfunction of the water vascular system, attributable to either oil or the oil dispersant under different conditions.

It is useful to compare the results obtained in the outdoor tank tests with those obtained when rating the

same dispersant using procedures described in the "Guidelines" (Anon., 1973). The outdoor tank experiment attempted to define the absolute toxicity and effectiveness of a dispersant applied to a crude oil in simulated marine field conditions. The "Guidelines" are used to rate dispersants in terms of their relative toxicities and effectivenesses in fresh water. Using its procedures, 77% of a no. 2 fuel oil was dispersed after 10 min and a 96 hr LC50 of 130 mg/l (1:1 v/v no. 2 fuel oil - Oilsperse 43 dispersant mixture) was obtained with rainbow trout (K. Doe, pers. comm.). Although results of the two types of studies are not directly comparable, they are sufficiently different to re-emphasize the caution, as expressed in the "Guidelines", that no laboratory method of estimating field effectiveness and toxicity of dispersants is completely satisfactory. Outdoor tank experiments as described in this paper could be considered as a logical next step in evaluating oil spill dispersants tentatively accepted by Environment Canada based on the "Guidelines" procedures.

5 SUMMARY AND CONCLUSIONS

5.1 Use of the dispersant, Oilsperse 43, increased the dispersion of Venezuelan Guanipa crude oil. The resulting mixture was more homogeneous and the oil slick less viscous than in the oil tank. The dispersant appeared to retard formation of the familiar "crust" on the surface.

5.2 A weathered crude oil plus dispersant mixture with an oil concentration of 250 µg/l was lethal to over 50% of the test organisms, green sea urchins, within 4 days. No mortalities occurred among urchins exposed to the crude oil treatment.

5.3 If similarly effective, "low-toxicity" dispersants are used on this crude oil spilled in an inshore marine area, concentrations of oil and dispersant lethal to the green sea urchin could conceivably be attained.

6 RECOMMENDATIONS

6.1 The outdoor tank experiments could be usefully extended to investigate the effectiveness and toxicity of a wider range of dispersants. Such experiments would greatly assist an evaluation of the toxicity and effectiveness of dispersants tentatively designated as acceptable for combating oil spills in the marine environment, and would serve as a link to actual field trials with spilled oils and dispersants.

6.2 The bioassay media and organisms in dispersant ranking tests and simulated field tests should be appropriate for the area where the dispersants may be employed. Sensitive commercially and/or ecologically important species in a given area should be tested. The green sea urchin, Strongylocentrotus droebachiensis, is an excellent test animal for salt-water bioassays, but a wide range of organisms, including a marine fish, would be preferable.

ACKNOWLEDGEMENTS

The authors wish to acknowledge the assistance of Messrs. K. Doe and W. R. Hardstaff in conducting these experiments. Mr. C. C. Cunningham, of Chemical Oceanography, A.O.L., conducted the salinities. E.P.S., Atlantic Region, and Mr. P. Phinney (Dalhousie Univ.) graciously provided unpublished data for the discussion. We thank E. Pessah and D. C. Gordon for their continual interest in this project and K. Doe, D. C. Gordon, E. Pessah and G. J. Farmer for critically reading the manuscript. We especially thank Marie Sweet for typing it.

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TABLE 1 Temperature, pH and oxygen levels of test solutions during bioassays in which sea urchins were exposed to dispersions of oil plus dispersant and oil alone. Each mean represents 10 observations. Bioassays 1 to 4 started at 48, 168, 336 and 648 hr respectively, during the one month test

Parameter	Treatment	Bioassay			
		1	2	3	4
Temp., C	Oil + Disp	13.6±0.2*	14.8±0.1	16.0±0.2	13.3±0.8
	Oil	13.7±0.1	14.7±0.1	16.0±0.2	13.3±0.8
	Control	13.7±0.1	14.7±0.1	15.9±0.2	13.8±0.6
pH	Oil + Disp	7.6±0.05	7.6±0.05	7.6±0.07	7.6±0.06
	Oil	7.8±0.03	8.0±0.09	7.8±0.03	7.8±0.07
	Control	7.8±0.03	7.9±0.08	7.8±0.06	7.8±0.06
O_2,mg/l	Oil + Disp	6.4±0.4	6.6±0.4	6.8±1.1	7.3±0.4
	Oil	7.8±0.2	8.9±0.2	8.4±0.6	8.0±0.5
	Control	7.8±0.2	8.6±0.2	7.7±0.7	7.8±0.4

*Standard error of the mean

TABLE 2 Fluorescence analysis of duplicate water samples from mid-depth of the outdoor tanks, over 31 day period. Oil concentrations are in µg extractable organics per liter of water

Time after addition of oil	Oil Concentration, µg/l			
	Oil plus Dispersant Tank		Oil Tank	
Pre-oil	7.6	5.8	8.2	8.9
1 hr	16510,	16904	75.1	450
4.5 hr	2140,	5030	9.5,	23.4
1 day	1085,	847	47.2,	28.7
14 "	253,	217	21.8,	26.6
27 "	211,	218	28.0,	29.5
31 "	290,	359	30.0,	25,0

TABLE 3 Oil budget for outdoor tanks. Oil remaining in water column was calculated from "steady state" concentrations. Oil lost from system was determined by difference

	Oil Plus Dispersant Tank		Oil Tank	
	Total	% of oil added	Total	% of oil added
Oil added	843 g	100	843 g	100
"Tar" collected on day 32	420 g	49.8	640 g	76
Oil remaining in water column	2064 mg	0.245	216 g	.026
Oil lost from system (weathering, losses in spray, etc.)	421 g	50	203 g	24

TABLE 4 Diameters of sea urchins, Strongylocentrotus droebachiensis, used throughout the study on toxicity of dispersions of crude oil plus dispersant and crude oil. Four bioassays were conducted, starting at different times during the study

	Diameter, cm			
Bioassay	$\bar{x}$	S.E.	Range	n
1	3.9	0.1	3.4 - 4.5	15
2	3.7	0.1	3.1 - 4.3	15
3	4.0	0.1	2.8 - 5.1	30
4	3.9	0.1	3.3 - 4.9	29

TABLE 5 Fluorescence analysis of water samples from bioassay tanks. Concentrations of oil in µg extractable organics per liter, are given for both tanks for each treatment

Bioassay	Treatment	Concentration of oil, µg/l			
		Initial		Final	
			$\bar{x}$		$\bar{x}$
1	Oil plus dispersant	1255, 1600	1427.5	339, 507	423
	Oil	24, 46	35	11, 12	11.5
	Control	15, 24	19.5	6, 10	8
2	Oil plus dispersant	284, 238	261	271, 316	293.5
	Oil	24, 28	26	18, 20	19
	Control	8, 6	7	5, 6	5.5
3	Oil plus dispersant	243, 131	187	235, 196	215.5
	Oil	36, 33	34.5	17, 14	15.5
	Control	5, 21	13	5, 4	4.5
4	Oil plus dispersant	382, 321	351.5	296, 262	279
	Oil	31, 28	29.5	60, 181*	-
	Control	4, 5	4.5	26, 26	26

*Contaminated sample

TABLE 6 Percent of moribund and dead sea urchins at 48 and 96 hr when exposed to dispersions of oil plus dispersant and oil over 96 hr. Ten urchins were tested in each treatment in each bioassay

Bioassay	Time, hr	Treatment: Oil plus dispersant	Oil	Control
Response-Moribund				
1	48	90	0	0
	96	20*	0	0
2	48	100	0	0
	96	40*	0	0
3	48	100	0	0
	96	0	0	0
4	48	80	0	0
	96	50**	0	0
Response-Dead				
1	48	10	0	0
	96	80	0	0
2	48	0	0	0
	96	60	0	0
3	48	0	0	0
	96	100	0	0
4	48	0	0	0
	96	50	0	0

*Did not survive when placed back into clean sea water

**No data on survival when placed back into clean sea water

TABLE 7 Percent of sea urchins climbing on tank walls or staying on bottom of test tanks at 24 hr and 48 hr when exposed to dispersions of crude oil plus dispersant and crude oil

Bioassay	Time, hr	Treatment: Oil plus dispersant	Oil	Control
Response-% Climb				
1	24	0	100	80
	48	0	-	-
2	24	0	60	90
	48	0	70	50
3	24	0	90	50
	48	0	100	100
4	24	0	100	20
	48		-	-
Response-% on Bottom				
1	24	100	0	20
	48	90*	-	-
2	24	100	40	10
	48	100	30	50
3	24	100	10	50
	48	100	0	0
4	24	-	0	80
	48		-	-

*One urchin dead at 48 hr.