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IMPACT OF DISPERSED AND UNDISPERSED OIL ENTERING A GULF COAST SALT MARSH

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Abstract — The effect of dispersed and undispersed South Louisiana crude oil on the growth of *Spartina alterniflora* and associated meiofaunal communities was investigated in a replicated experiment conducted in a uniform Louisiana salt marsh. The oil and the oil plus dispersant were applied to open water adjacent to the marsh and forced onto the marsh, using a pump to create a "head" of water that simulated tidal conditions. Neither crude oil nor oil plus dispersant had any inhibitory or stimulatory effect on the growth of *S. alterniflora* or the meiofaunal communities, including the meiobenthos.

Keywords — Oil   Meiofauna   *Spartina alterniflora*   Salt marsh

INTRODUCTION

Accidental release of crude oil can cause extensive damage to marshlands and beaches. Currently, mechanical cleanup techniques are used to restore the oil-contaminated environments, but such methods can cause severe environmental damage [1]. Although the use of chemicals has been proposed for cleanup, there is limited documentation that chemicals cause less environmental damage. Chemicals used in restoration techniques are surface-active and have two functions. The surfactant must first remove the oil from the surface, and then it must prevent redeposition. Westfall [2] found that Gulf ryegrass and white clover were killed by oil, but application of a water-based surfactant on the plants reduced the injury. Damage occurred because the plant surface was wetted by oil, which prevented water uptake. However, the presence of water-soluble chemical surfactants prevented oil from wetting the plant tissue.

The purpose of this study was to determine the effects of oil and oil treated with dispersant in open water areas on the growth of *Spartina alterniflora* and the meiofaunal communities, including the meiobenthos, in a Louisiana salt marsh. Louisiana coastal wetlands support a rich assortment of fisheries and wildlife that is heavily dependent upon the annual production of estuarine biota. Estuarine-dependent

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organisms in turn rely on the production of the natural marsh.

MATERIALS AND METHODS

Experimental design

A study was initiated on May 28, 1982, to determine the impact of dispersed and undispersed South Louisiana crude oil on the growth of the marsh macrophyte *S. alterniflora* and associated meiofaunal communities. The treatments included (a) crude oil, (b) the application of a chemical dispersant to oil in open water before allowing the "dispersed" oil to enter the marsh and (c) control (no oil or dispersant). Each treatment was replicated four times and arranged in a completely randomized experimental design. The study was conducted in a remote area of the marsh bordering open water located in the Wisner Wildlife Refuge, LA. Approval was obtained from appropriate government agencies for applying oil to the marsh.

Replicate test plots were 2.45 × 4.90 m and extended 2.45 m from the marsh into the open water. South Louisiana crude was added at the rate of 2 L m^{-2} to the open water portion of plywood-enclosed plots. Dispersant was aerially applied to the oil at the rate of 1 liter per plot or 0.17 L m^{-2} to create a 1:12 ratio of dispersant to crude oil. The oil or oil plus dispersant was left in the open water portion of the plots for 1 h before being forced onto the marsh. During this time the oil or oil plus dispersant was physically mixed to obtain a uniform distribution. The oil or oil plus dispersant was forced onto the marsh by a pump that created a water "head" in the open water area bordering the marsh, thereby simulating tidal flow of water across the marsh. The plot retainers were removed after 24 h to allow normal tidal flushing.

Oil content of sediment

The petroleum hydrocarbon content of the sediment was determined by removing five cores (10 cm in diameter × 10 cm deep) from each plot 1 d after the application of the oil and/or oil plus dispersant. A composite sample was made for each plot and the hydrocarbon content determined by fluorescence analysis. Sediment samples were first homogenized. One hundred cubic centimeters of sediment suspension and 150 cm^3 of water were placed in a blender and thoroughly mixed. The sediment–water mixtures were extracted with 350 cm^3 of spectrograde hexane on a wrist shaker. The extracts were analyzed with a fluorescence spectrophotometer by scanning from 250 to 450 nm. Peak intensities measured at 365 nm were converted to $\mu g\ g^{-1}$ dry sediment using an oil sample as the standard, and reported as total aromatic hydrocarbons.

Response of S. alterniflora

Effect of crude oil and oil plus dispersant on photosynthetic carbon fixation was determined 6, 13 and 53 d after oil introduction. Portable light and dark chambers were placed over portions of the marsh for determination of the relative amount of CO_2 fixed by marsh macrophytes and epiphytic algae [3]. The method is a very sensitive indicator of plant stress. The aboveground biomass was determined 123 d after oil introduction, when plant biomass production was at a peak [4].

Response of meiofaunal communities

Four meiofauna samples were taken from each experimental plot on day 0 (a few hours before oil application on May 28, 1982), and on days 5, 36 and 77 after oil introduction. On day 0 all plots were sampled for meiofauna (four replicates from each plot), 1 to 3 h before oil introduction to establish population density estimates and natural faunal variability before the oil was introduced. Cores were taken with 2.54-cm diameter core tubes modified from 50-cm^3 syringes. All cores were fast-frozen in liquid nitrogen in the field.

In the laboratory, cores were sectioned to 4 cm with a coping saw, but only the 0 to 4 cm interval was retained. Preliminary studies showed that 90 to 95% of the nematodes and 100% of the copepods were found in the upper 4 cm of sediment. Sediments were fixed in a buffered 10% formalin–rose bengal solution, and later extracted in a LUDOX-AM with centrifugation [5]. Sediments were spun first to remove excess water (1,300 rpm for 2 min). LUDOX was then added and the sediment was centrifuged at 975 rpm for 3 min. The supernatant was rinsed through a 63-μm sieve and retained. LUDOX was reintroduced into the centrifuge tube and a second centrifugation was performed. This extraction procedure has been shown to be 92 to 95% efficient in removing nematodes and 98% efficient in removing copepods from the sediments. All animals from three replicate samples retained on a 63-μm sieve were identified to major taxon and counted.

Statistical analysis was conducted by GLM [6]. Analysis of variance was used to test for significant differences among dates, treatments (control, oil alone or oil with dispersant) and for treatment–date interactions for nematodes, copepods, polychaetes and total meiofauna. Duncan's multiple range (DMR) test was used to detect significant differences for each collection period for each taxon.

RESULTS AND DISCUSSION

Analysis of composite sediment samples taken from each plot 1 d after the application of crude oil or dispersed oil revealed sediment hydrocarbon content of 295 $\pm$ 46, 198 $\pm$ 65 and 18 $\pm$ 6 $\mu g\ g^{-1}$ for the oil, dispersed oil and control, respectively. Even though a significant amount of oil was recovered in the sediment, apparently the majority of the oil entering the marsh was removed by tidal flushing.

Statistical analysis (by DMR) of the data obtained from the 12 plots revealed no significant difference ($p \leq 0.05$) in the sediment hydrocarbon content of plots which received oil and oil plus dispersant. The reduction in hydrocarbon content achieved by applying dispersant to the oil before it entered the marsh was not significant due to the large variation between the replicate plots. This reflects the heterogeneous nature of the system and the problems in obtaining a representative sediment sample. Hydrocarbon content of the sediments treated with oil and with oil plus dispersant was statistically greater than the concentration in the sediment of the control plots.

The relative CO_2 fixation following the treatment of a uniform stand of *S. alterniflora* with South Louisiana crude oil and dispersed oil is shown in Table 1. Crude oil had significantly ($p \leq 0.1$) reduced the gross $CO_2 - C$ fixation 6 d after its introduction into the marsh. Oil treated with dispersant did not significantly reduce the gross $CO_2 - C$ fixation relative to that of the control. The reduction in photosynthetic activity in the oil-treated plots relative to the control plots was short-lived, and *S. alterniflora* had recovered within 13 d after treatment. The quick recovery of photosynthetic activity is consistent with the longer-term aboveground biomass data (Table 2). *S. alterniflora* aboveground biomass was not statistically decreased or increased when oil or oil plus dispersant entered the marsh. Total biomass values reported are similar to those reported by DeLaune et al. [4].

Apparently the amount of crude oil entering the marsh from the open water was not toxic to *S. alterniflora* root or stem tissue and did not inhibit macrophyte growth. Oil on the soil surface or water within the tidal zone apparently had little influence on mature *S. alterniflora*. In a previous study, the gross photosynthetic rate of the macrophyte *S. alterniflora* was decreased to 19 to 37% of that determined in untreated areas 7 and 14 d after the application of 2 and 8 liters of oil per square meter [3]. The plants in that study were coated to a height of 50 cm as a result of unusually high tides due to a tropical depression. Normal tidal amplitude in this area

Table 1. Effect of oil or dispersed oil on gross CO_2 fixation by *Spartina alterniflora*

Days after oiling	Treatment		
	Oil	Oil plus dispersant	Control
6	406 ± 101	694 ± 211	817 ± 184
13	618 ± 92	590 ± 108	683 ± 103
53	359 ± 43	357 ± 53	585 ± 125

Values are means of four replicates ± SD (mg C m^{-2} h^{-1}); least significant difference ($p \leq 0.1$) = 267.

is only 20 to 30 cm; consequently, oil on the surface of tidal water does not normally contact the leaf blades. Greenhouse and field studies in Louisiana have indicated that *S. alterniflora* can tolerate South Louisiana crude applied at rates equivalent to 32 L m^{-2} without a decrease in aboveground biomass [4].

Total meiofauna (Fig. 1) ranged from 243 to 661 organisms per 10 cm^2, and was comprised largely of nematodes (~80% of all fauna) and copepods (~10% of all fauna). The day 0 heterogeneity was greater than that found on any other collection date. Total meiofauna densities in control plots were approximately twice those in plots that subsequently received oil or oil plus dispersant. Copepod densities (Fig. 2) were heterogeneous, with much higher densities on day 0 on control plots. Higher densities of nematodes (Fig. 3) and polychaetes (Fig. 4) were found on day 0 in the control plots prior to the application of oil or oil plus dispersant. The cause of this increased heterogeneity is not known, but may be related to natural year-to-year variation or to spatial variation patterns throughout the marsh system.

Statistical analyses were influenced by the day 0 variability for total meiofauna and copepods. Statistical analysis for total meiofauna indicated no treatment-related effects due to oil or oil plus dispersant. A day-by-day test for differences among means showed that the treatments varied for total meiofauna only on day 0. On day 0, control plot densities were significantly greater than those in plots that subsequently received oil. Similarly, copepods showed a significant overall treatment effect, but this effect may also have been due to natural variation associated with the day 0 collection. Only day 0 data (measured before the introduction of oil or oil plus dispersant) showed differences among treatments (Fig. 2) because control densities were greater than at any other treatment time. No unequivocal oil-induced effect can be observed because of the background variation on day 0 for total meiofauna and copepods. Oil-induced initial mortality or stimulation of meiofauna or copepods would have been observable under the conditions of this *in situ* study.

Nematodes and polychaetes did not show statistically significant heterogenetiy on day 0, and oil effects can be examined without complication. Nematodes did not show a significant treatment effect, but did show a treatment–date interaction ($p \leq 0.05$) on day 5 (Fig. 3). However, it was questionable that there was any significant biological difference, since there was no significant change in densities between treatments for the other dates. Copepod species were dominated by *Enhydrosoma woodini*, which comprised >60% of all individuals. Also abundant were *Nitocra lacustris* and *Nannopus palustris*. Each of these species is commonly found in salt marsh sediments on the coast of the Gulf of Mexico.

Of the three major copepod species, only *E. woodini* showed significant differences due to the application of oil and oil plus dispersant. *E. woodini* density was initially variable on day 0, with control plots having highest densities. Density in plots receiving oil and oil plus dispersant was significantly lower than in the control plot on day 5. However, no long-term effect was observed

Table 2. Effect of oil or dispersed oil on aboveground biomass of *Spartina alterniflora* 123 d after oil application

Treatment	Aboveground biomass		
	Live	Dead	Total
Oil	712 ± 28	350 ± 73	1,062
Dispersed oil	720 ± 152	375 ± 61	1,095
Control	792 ± 208	324 ± 41	1,116
Least significant difference ($p \le 0.1$)	188	196	

Values are means of four replicates ± SD (g m^{-2}).

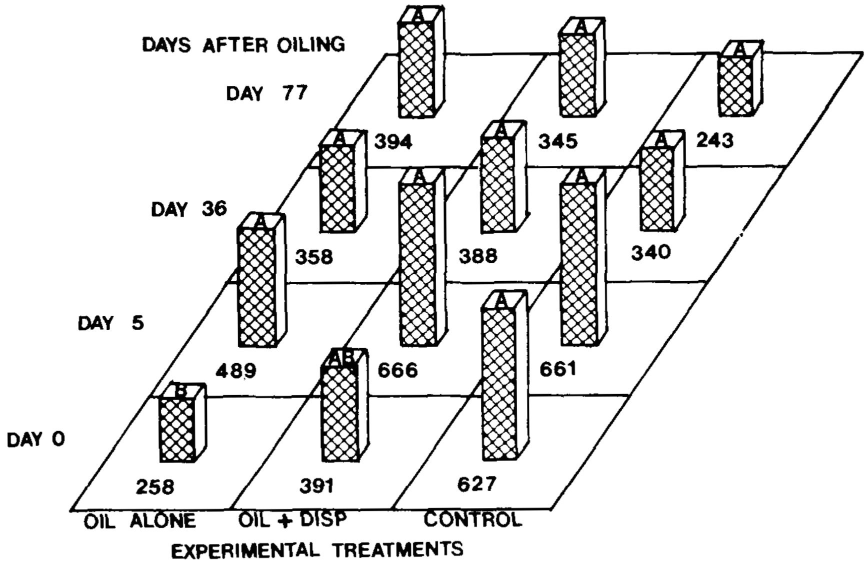

Fig. 1. Densities (per 10 cm^2) of total meiofauna on each collection date. Means with same letters are not significantly different ($p \le 0.05$).

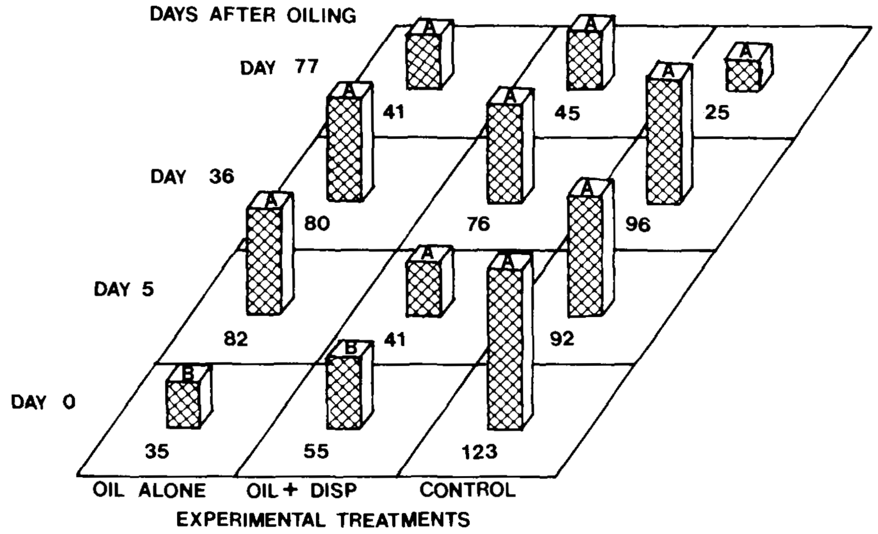

Fig. 2. Densities (per 10 cm^2) of copepods on each collection date. Means with same letters are not significantly different ($p \le 0.05$).

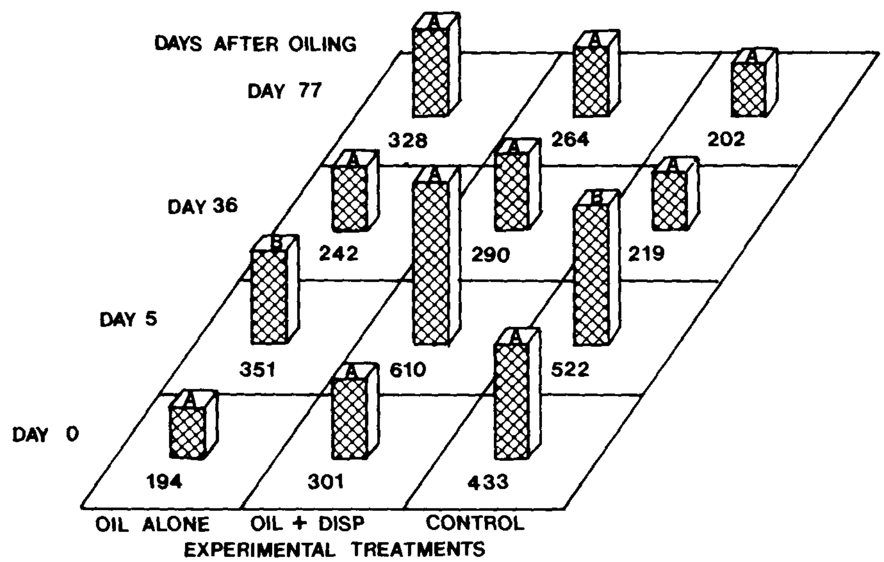

Fig. 3. Densities (per 10 cm^2) of nematodes on each collection date. Means with same letters are not significantly different ($p \leq 0.05$).

because no statistically significant effects could be shown on days 36 or 77.

Several studies have shown that petroleum hydrocarbons or dispersants can harm meiofauna. In laboratory studies, Oviatt et al. [7] and Grassle et al. [8] showed that chronic additions of fuel oil depressed densities. Oviatt et al. [7] reported that benthic fauna were reduced by 90 ppb No. 2 fuel oil. In 1 year after additions ceased, the benthic fauna did not recover from the 90-ppb treatment. Giere [9] showed that an oligochaete species was not severely affected when exposed to crude oil concentrations up to 1:1,000, and when oil and dispersants were mixed together. In field studies, a general

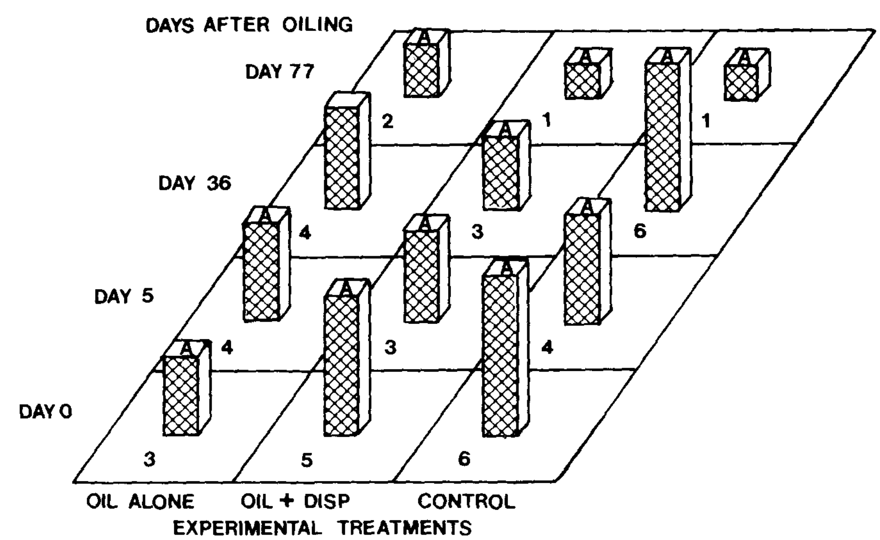

Fig. 4. Densities (per 10 cm^2) of polychaetes on each collection date. Means with same letters are not significantly different ($p \leq 0.05$).

decrease in nematode abundance was observed in intertidal sand 7 months after an oil spill [10]. The mean aliphatic hydrocarbon concentrations were about 100 mg kg^{-1} dry weight. McLachlan and Harty [11] conducted an experimental spill that showed negative effects of oil. Such field studies have shown that recovery for meiofauna varies from 5 months to 1 year, and that some groups are more sensitive than others to hydrocarbons. Boucher [10] reported that nematode diversity significantly decreased, mainly due to the increase of some dominant species. Copepods and oligochaetes appear to be highly sensitive, while nematodes are resistant to all but the highest concentrations of hydrocarbons.

In contrast to the studies cited above, no mortality that could be attributed to hydrocarbons or oil plus dispersant was observed in this study. The lack of strong effects due to the application of crude oil and dispersant may be related to the mechanism of application. Oil or oil plus dispersant was introduced onto the marsh with the tide, and may never have reached the benthos in concentrations equal to those in the previous studies, in which oil was sprayed directly onto the intertidal sand surface at low tide. The warm summer temperatures may have contributed to the volatilization and dissipation of oil and/or dispersant. Additionally, meiofauna may either be adapted to this concentration of oil by prior exposure to the high concentrations of hydrocarbons found in Louisiana marshes [12], or generally may be adapted to high stress and thus be biochemically or physiologically equipped to withstand novel stresses.

With regard to restoration of the marsh after oil enters via tidal action in Gulf Coast wetlands, the findings of this study suggest that, for macrophytes and meiobenthos, the best approach may be no cleanup at all when the system is impacted by the concentrations of crude oil within the sediment reported here. Oil caused no direct mortality. This does not imply, however, that oil is necessarily neutral or beneficial to the marsh system. Kalke et al. [13] and Harty and McLachlan [14] have indicated that processes associated with the benthos may be affected even though densities do not change. Such processes include nitrogen or other nutrient regeneration by microflora, and secondary production associated with the benthos.

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