BACKGROUND: The northern Gulf Coast of the United States is a region of intense oil exploration, production, transmission, and refining. Consequently, coastal states, such as Louisiana, are subject to oil spills resulting from shipping accidents, production-related incidents, and pipeline ruptures. Since these incidents often occur in the nearshore environment, coastal salt marshes are frequently the first wetland habitat to be subjected to the oil. As a result, a large number of investigations have documented the effect of petroleum hydrocarbon on the dominant salt marsh species, especially *Spartina alterniflora*. In addition, some investigators have evaluated the impact of oil cleanup procedures in salt marshes. Not only can petroleum hydrocarbons have a detrimental impact on coastal marshes, but, additionally, the cleanup of the oil from these highly sensitive environments is often more damaging than the oil itself. Hence, it is important to develop less intrusive oil spill cleanup procedures that exert little to no impact to wetland ecosystems.
OBJECTIVES: (1) To determine the toxicity and ecological safety of some common biodegradation agents. (2) To determine the effect of these bioremediation agents on crude oil degradation under the most common marsh inundation environments. (3) To determine the effect of biostimulants on crude oil degradation as a function of soil texture. (4) To determine the comparative efficacy of bioremediation and phytoremediation of oil.

DESCRIPTION: Although bioremediation of oil spill cleanup has received considerable attention in recent years, its satisfactory use in the cleanup of oil spills in the wetland environment is still questionable and generally untested. We have conducted a multi-disciplinary experimental program to evaluate the use of various bioremediation products, including microbial seeding, inorganic fertilizer, and soil oxidant, as a means of enhancing oil biodegradation in coastal salt marshes. The program addresses the question: Is bioremediation, via fertilization, microbial seeding, or soil oxidant, an effective and ecologically safe means of oil spill cleanup in coastal wetlands?

The effectiveness of bioremediation and its ecological safety were assessed through a series of four experiments. The first experiment was designed to determine the toxicity and ecological safety of common biodegradation agents. In a greenhouse, the following bioremediation treatments were applied to salt marsh sods with intact S. alterniflora: (1) fertilizer, (2) microbial seeding, and (3) no bioremediation agent addition (control). The experiment used a randomized block arrangement. The second experiment was designed to determine the effect of bioremediation agents on oil degradation and biotic response under drained and flooded conditions in marsh mesocosms. In the greenhouse, the following bioremediation treatments were applied to salt marsh sods with intact S. alterniflora: (1) fertilizer, (2) microbial seeding, (3) oxidant plus fertilizer, and (4) no bioremediation agent addition (control). Two inundation regimes were used, flooded with 3 centimeters of standing water and drained with the water table 10 centimeters below the soil surface. The third experiment compared the three major categories of bioremediation agents, fertilizer, microbial seeding, and soil oxidant, as a means of enhancing oil biodegradation in coastal mineral and sandy marsh substrates under controlled greenhouse conditions. The fourth experiment compared the effects of phytoremediation and bioremediation on oil degradation. In the greenhouse, the following treatments were applied to the experimental units: (1) phytoremediation with S. alterniflora or without, (2) three nitrogen levels, and (3) two phosphorus levels.

SIGNIFICANT CONCLUSIONS: Bioremediation by adding non-bacterial agents to enhance natural oil degradation has considerable potential in wetland environments. Application of fertilizer, either in the form of slow release or water soluble, significantly enhanced plant growth with significantly higher photosynthetic rates, stem growth rates, and above- and below-ground biomass of S. alterniflora. In addition, application of fertilizer significantly reduced the concentrations of both the reduced crude oil and the artificially weathered crude oil. In the absence of S. alterniflora, however, bioremediation by nitrogen and/or phosphorus addition did not significantly affect...
residual oil concentration. These results suggest that oil biodegradation in marsh systems may be enhanced by fertilization (biostimulation).

**STUDY RESULTS:** Efficacy and success of bioremediation of oil to accelerate the biodegradation process depend on the extent of contaminant removal and on the ecological safety of the bioremediation agents. The bioremediation agents used in this research, including a microbial product and inorganic fertilizers, had no negative impacts to the plant, animal, and microbial responses evaluated in this study. Application of the bioremediation agents at the specified application rates to the salt marsh sods did not adversely affect the marsh macrophyte, *S. alterniflora*, various microbial populations and soil respiration, and various infaunal animals including macrofauna and meiofauna. Application of fertilizer, either in the form of slow release or water soluble, significantly enhanced plant growth with significantly higher photosynthetic rates, stem growth rates, and above- and below-ground biomass of *S. alterniflora*. The fertilizer-enhanced oil degradation occurred only in the presence of the marsh macrophyte, *S. alterniflora*. The effect of fertilization on oil degradation by directly enhancing soil microbial activity was separated from the phytoremediation effect by comparing bioremediation by added nitrogen and phosphorus in the absence of plans with that in the presence of *S. alterniflora*. Application of the soil oxidant did not significantly affect oil degradation in the marsh sods. The microbial product did not significantly affect oil degradation. This study suggests that the high cost of microbial amendments and soil oxidant application for oil bioremediation may not be warranted in coastal marshes.