CRUDE OIL BIOREMEDIATION FIELD EXPERIMENT IN THE SEA OF JAPAN

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Experimental bioremediation of crude oil was conducted for approximately 3 months in the intertidal zone of the Sea of Japan, Hyogo Prefecture. Artificial mixtures of weathered Arabian light crude oil and sand taken from the experimental site were wrapped in polyester net envelopes. The envelopes were placed in drum-shaped acrylic vessels with perforated sides to facilitate seawater exchange. The vessels were laid in the intertidal area. Slow release nitrogen and phosphorus synthetic fertilizer granules were added to the oil–sand mixtures in 3 different amounts. Some oil–sand mixtures were unfertilized controls. The oil–sand mixtures were periodically sampled and changes in the composition of the residual oils were monitored. Oil samples were subjected to gas chromatography coupled with mass spectrometry for analysis of a number of semi-volatile aliphatic and aromatic compounds. All values for each analysis were normalized against that of hopane to evaluate the extent of oil biodegradation. In addition, the quality of the seawater in each acrylic vessel was evaluated by conducting an algal growth potential (AGP) test with a diatom, *Skeletonema costatum*, and an acute toxicity test with a marine hyalid amphipod, *Hyale barbicornis*. Significant increases in the concentrations of both nitrogen and phosphorus were found in the fertilized sections in accordance with the amounts of added fertilizers. Although significant natural attenuation of oil was observed in the unfertilized sections, fertilization stimulated the degradation rate of the oil in the early stage of the experimental term. The extent of the oil biodegradation increased as the amount of added fertilizer increased. This was paralleled by changes in the microbial community structures; i.e., the structures of the microbial communities changed more greatly as the amount of added fertilizer was increased. However, the final degradation efficiencies for each oil component in the fertilized sections were not significantly different from those in the unfertilized sections, and the degradation of each oil component had almost ceased after 6 weeks. Alkanes degraded to a lesser extent than naphthalenes or fluorenes and to almost the same extent as dibenzothiophenes and phenanthrenes. We conclude that excessive amounts of macronutrients are required to accelerate oil biodegradation and that fertilization is only effective in the early stages. The acute toxicity testing of the seawater in the experimental acrylic vessels showed no adverse effects due to the seawater in either the unfertilized or fertilized sections. The AGP tests showed no significant inhibitory effects on algal growth in either the unfertilized or fertilized sections and showed that the algal growth stimulating effect of fertilization was much lower than expected from the actual nitrogen concentration. Since the fertilizer nitrogen was in the form of urea, it is likely that the released nitrogen did not straightforwardly stimulate algal growth.