The Influence of Microorganisms on Oil-Mineral Fine Interactions in Low Energy Coastal Environment: Preliminary Results.

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Abstract

When spilled in the environment, oil frequently interacts with fine mineral particles to form aggregates. This process can contribute to the natural shoreline restoration process as the residual oil remains mobile and becomes more accessible to oil-degrading biota. Studies have been undertaken to accelerate this natural oil dispersion process as an oil spill countermeasure (i.e., surf-washing). Mixing energy (waves) has been considered a key factor controlling the formation of oil-mineral fine aggregayes. While some laboratory studies required strong mixing energy to induce the formation of oil-mineral fine aggregates, they also have been proven to occur with moderate agitation in the environment. A possible explanation for this difference maybe the influence of biosurfactants generated by the biota.

To test this hypothesis, experiments were undertaken in 12 mesocosms, that simulated the conditions of a sandy shoreline environment subject to moderate wave action and tidal movement. Experiments were conducted in triplicate with the following test conditions: oil alone, oil with addition of clay with and without bacterial enhancement, and oil with addition of clay and mercuric chloride to suppress bacterial activity. During experiments running from 3 to 5 weeks in duration, oil in the sediment, tidal water, and suspended particulate fraction were analyzed. Preliminary results show a positive influence of microbial activity on the formation of oil-mineral fine aggregates that accelerate the rate oil removal from the sand.

1.0 Introduction

When spilled in the environment, especially in coastal systems, oil frequently interacts with natural fine mineral particles to form aggregates. This process has been observed and can contribute to natural shoreline restoration; oil dispersion is enhanced as the oil trapped on the fines remains mobile. The resulting lower oil

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concentrations are more suitable for to biodegradation, and the aggregates are believed to provide a favourable habitat for oil-degrading bacteria.

Different studies have explored the interaction of oil and mineral fines and their potential benefit in spill cleanup operations (e.g., surf-washing). Mixing energy is considered a key factor in generating oil-mineral aggregates. Some laboratory simulations have required strong mixing energy to generate the aggregates, however, the addition of surfactants (chemical dispersants) can enhance the formation of low-energy aggregates.

Oil-mineral fine interactions have been demonstrated to occur in situ with even the moderate agitation found within sheltered sites. A possible explanation of this phenomenon may be the influence of biosurfactants generated by natural biota. To test this hypothesis, a study was undertaken in mesocosms, simulating a sheltered sandy shoreline subjected to controlled tidal movement and wave action, to compare the behaviour of oil with and without the addition of fine minerals and/or biota.

2.0 Experimental Design

Mesocosm experiments were conducted at Cedre test facility to assess the influence of clay particles, with and without microflora, on the biodegradation rate of the oil and the degree of sediment restoration in a low-energy environment, such as a sheltered sandy beach.

Twelve replicate mesocosms, each consisting of an aquarium filled with a bed of sorted medium sand (300 to 1000 μm) and fresh Atlantic seawater, were fixed on an oscillating table (periodic movement : 35s) to generate an identical wave action in the tanks. To simulate tidal movement, each mesocosm was connected to another aquarium (renewal tank) by a small hose with a peristaltic pump to facilitate water exchange twice daily (Figure 1). The seawater in the renewal tank was replaced twice a week to simulate the dilution process which occurs in the natural environment.

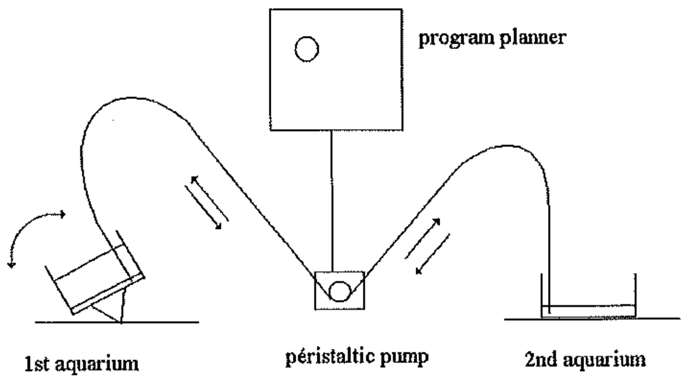

Figure 1 : Experimental Design of Mesocosm Table

An experimental treatment matrix was designed with the following controlled test criteria: clay particles absent [C0]; clay particles present [C1]; abiotic control (mercuric chloride treated) [B0]; natural microflora [B1]; natural microflora and

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nutrient addition [B2]; and the addition of oil-degrading bacteria and nutrients [B3].

The sandy sediments in all mesocosms were treated with identical amounts of oil (110°C topped Arabian crude oil) several days before the start of the experiment. To maintain specified test conditions, mercuric chloride, clay and nutrient were re-added to the appropriate mesocosms, as required. The clay was a mixture of quartz, muscovite, montmorillonite, and vermiculite. Each treatment condition was in triplicate.

The first experiment involved the test matrix:

C0B1
C1B0
C1B1
C1B2

and the second one:

C0B0
C1B0
C0B3
C1B3

2.1 Measurements

The following parameters were monitored and/or quantified for each of the experiments over a 6 week period :

- temperature;
- oxygen content in the water column;
- bacterial numbers;
- hydrocarbon content in the water column (IR measurement of extracts);
- oil content in the sediment (by weighing after soxlet extraction);
- oil chemistry in the sediment (GC); and
- aggregates in the water column (sampled after decanting the discarded water at each water change).

3.0 Preliminary Results

3.1 Oxygen Consumption in the Water

The oxygen depletion was mainly attributed to bacterial activity and not to the presence of clay particles, except at the end of the second experiment (Figure 2).

3.2 Bacterial Numbers

During the first experiment, with the exception of tanks treated with mercuric chloride, there was no significant difference between the number of bacteria in the different treatments (C0B1~C1B1~C1B2>C1B0). In the second experiment, the results were similar (B3>B0). Bacterial numbers were not dependent on the presence of clay particles. (Figure 3)

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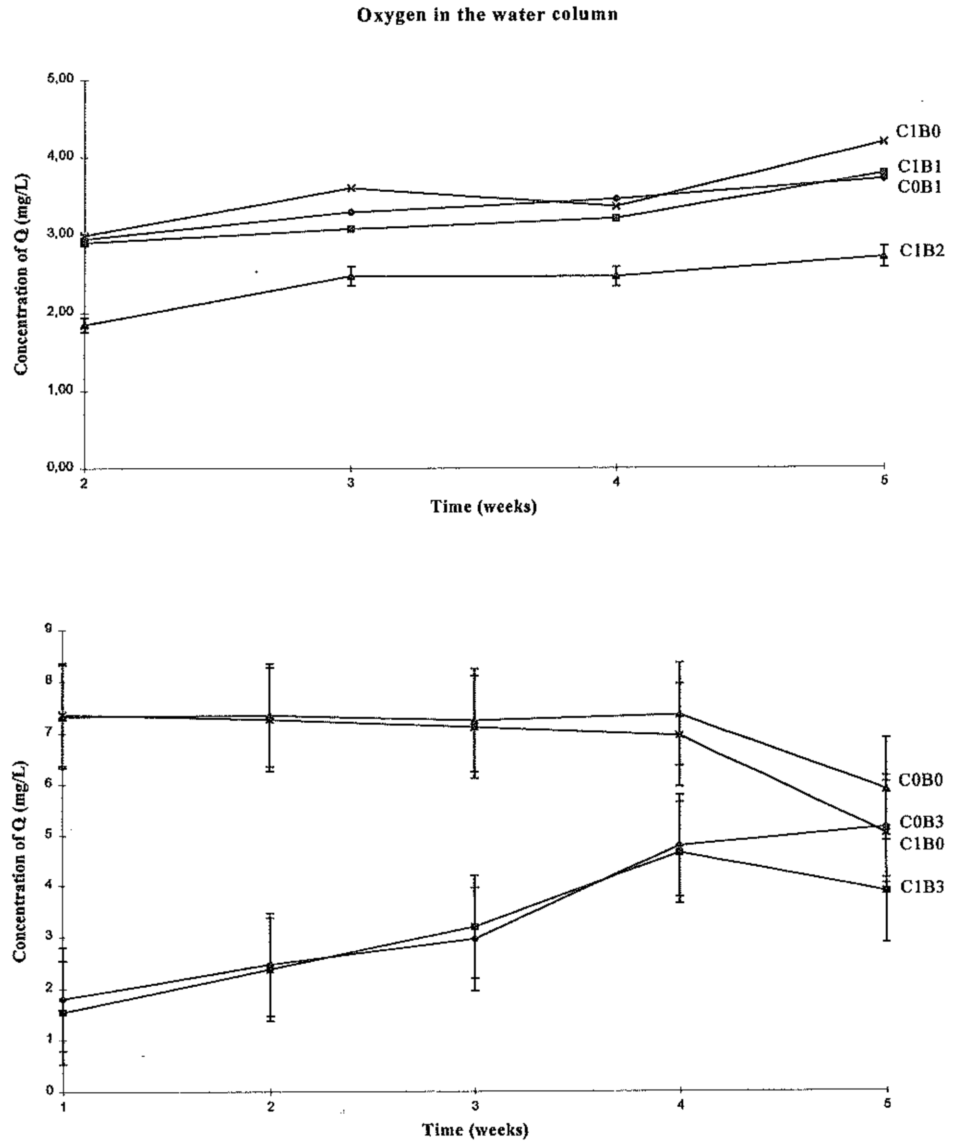

Figure 2 Oxygen Concentrations (mg/L) in the Water Column

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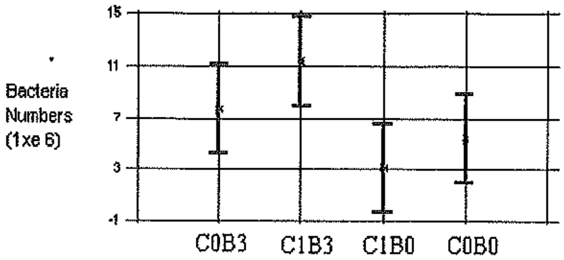

Figure 3 Bacterial Numbers (number/g sediment; error bars represent 95% confidence limits)

3.3 Hydrocarbon Concentration in the Water Column and Sediment

No significant difference was observed between the mesocosms according to the oil quantity within the water column and sediment.

3.4 Hydrocarbon Chemistry

The extent of oil biodegradation in the sediment was assessed by GC analysis using the ratios of nC_{17}/pristane and nC_{18}/phytane. At the time this paper is written, only the GC analyses of the first experiment have been completed. Preliminary results show a greater degree of biodegradation in mesocosms containing both biota and particles (nC_{17}/pristane: C1B2 $\leq$ C1B1 $\leq$ C0B1 $\leq$ C1B0). Analyses will be replicated to determine the statistical significance of these preliminary observations (Figure 4).

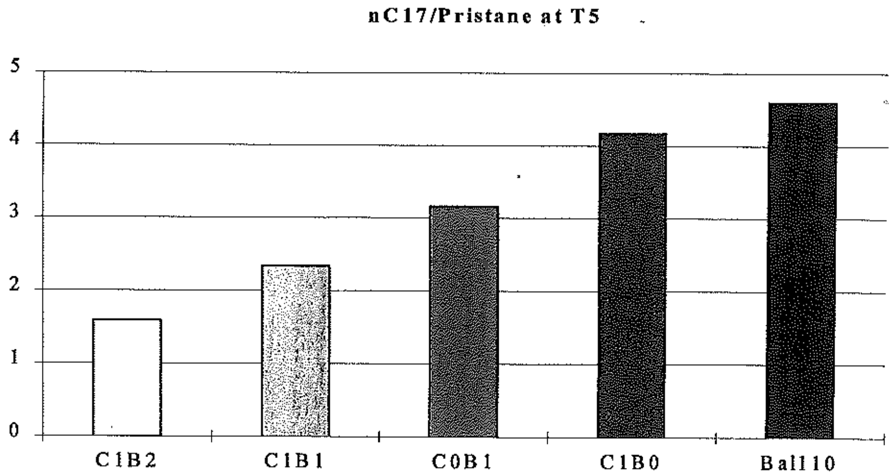

Figure 4 Oil Degradation in the Sediment Relative to Test Oil

3.5 Aggregate Analyses

Qualitative and quantitative analyses will be performed on the archived samples of aggregates recovered during the water changes.

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4.0 Discussion

These 6 week experiments were designed to determine whether the presence of clay particles associated with the presence of active micro-organisms could help in the natural restoration of a coastal site .

Preliminary data obtained on oxygen consumption and microbiological analyses clearly showed that biological activity was sustained within the mesocosms, and further enhanced with treatment by the addition of nutrients and/or an oil degrading consortium. Biological activity was greatly reduced, but not totally stopped with the experimental addition of mercuric chloride (abiotic control mesocosms, B0).

The activity of micro-organisms in the biotic mesocosms (B1, B2, B3) led to a visible presence of emulsified oil in the water column. This evidence of biosurfactant production was verified by direct observation as much more oil coalesced and adhered to the walls of the abiotic (B0) mesocosm tanks. Unfortunately, these observations could not be confirmed by analyses of hydrocarbon concentrations in the water column. The accidental transfer of some free oil by the pump at low tide during the water exchange may have masked some observable differences between the biotic (B1, B2, B3) and abiotic mesocosms (B0).

Concerning the effect of the clay particles, we did not see significantly more oil transferred with the tidal water of the mesocosms treated with the addition of (C1). This is likely to be attributed to the release of fine material associated with the apparently insufficiently sorted sand used in the experiment, and from the bacterial consortium added. The analyses of the aggregates in the mesocosms should resolve this issue.

The GC analyses performed on the first experiment showed that most degraded oil was correlated to the mesocosms simultaneously amended with clay and biota (C1B2 and C1B1). This preliminary result will be confirmed by additional GC analyses of triplicate samples in the second experiment to provide statistical evidence.

Lastly, taking into account these findings, especially those concerning the experimental methodology, a third experiment is being carried out. This time, special measures are being taken to avoid interferences from any external fine particles other than the experimental source clay added.

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