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**OSEI CORPORATION'S SUMMARY**  
of  
**Texas A&M's**  
**Microbial Petroleum Degradation Enhancement**  
**By Oil Spill Bioremediation Products**

The General Land Office for the state of Texas (USA) asked the University of Texas A&M to perform a study on 13 bioremediation products listed in the EPA National Contingency Plan for oil spills.

The efficacy tests were to be performed using the EPA / NETAC guidelines in their test protocol for bioremediation agents.

The test covered the total oil and grease (O&G), the aliphatic fraction of oil, the aromatic fraction of oil, and the plate counts on the numbers of hydrocarbon degraders grown or colonized during this test.

***OIL SPILL EATER II*** IS PRODUCT 10.

***Oil Spill Eater II*** was one of the best products at reducing the oil and grease. ***Oil Spill Eater II*** was the most effective product at reducing the aliphatic fraction of the oil.

***Oil Spill Eater II*** was the most effective product at reducing the Polar-aromatic (PAH, more toxic) fraction of the oil.

***Oil Spill Eater II*** grew the most hydrocarbon degraders, an acceptable product grew  $10^5$  numbers of hydrocarbon degraders while ***OSE II*** outperformed them all at enhancing hydrocarbon degraders at  $10^{7.5}$ .

***Oil Spill Eater II*** proved it was the most efficient product at biodegrading Alaskan North Slope crude oil out of the 13 EPA / NCP Listed products tested.

Steven R. Pedigo  
Chairman

**Microbial Petroleum Degradation Enhancement By  
Oil Spill Bioremediation Products**

**A Report Submitted to the Texas General Land Office**

**October 12, 1995**

**Principal Investigators:**

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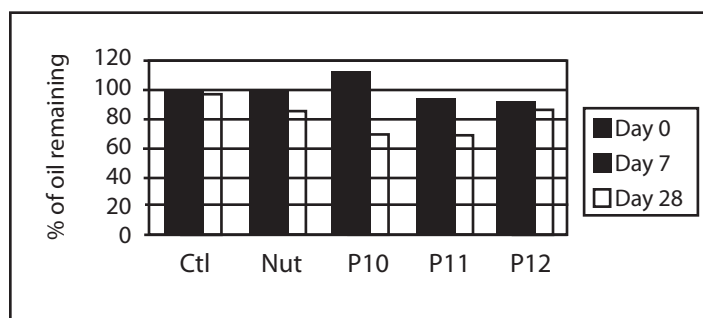
**Robin L. Autenrieth**

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**Salvador Aldrett**

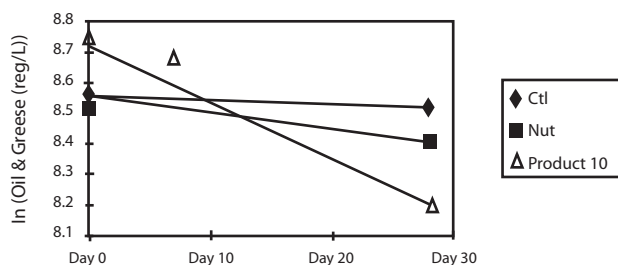
**Marc A. Mills**

**Frank Stephens**



**Figure 4 – Oil and Grease results (Batch D)  
P10 is OSE II**

High O&G numbers can be a result of a high production of extractable materials such as biomass or metabolites. As shown in Batch D, Product 10 is causing an increase in the O&G values at day 0 and 7, with an average value of 11% more of the initial weight. However, microbial counts indicate a high aliphatic degrader population through this period, as will be shown later Figure 16. After 28 days the oil was degraded more extensively by Product 10 than by the nutrient control. This suggests that the polar fraction is possibly being increased by the product’s contents, on days 0 and 7, but does not imply that the oil is remaining undegraded. Microbial degradation of Product 10 could be producing metabolites that are being completely oxidized between day 7 and day 28.



Treatment	Slope	R square
Control	-0.0013	0.9505
Nutrient	-0.00563	0.8041
Product 10	-0.01859	0.9228

**Figure 10 – Ln concentration change with time for product 10 (P10)  
as compared with the nutrient and non-nutrient control**

Figure 10 suggests a lag phase for Product 10 between day 0 and 7, after this period the microbial population shows a high degradation rate, achieving a final degradation extent higher than that of the nutrient and non-nutrient control.

The rate of oil removal is an important factor to consider when comparing the performance of each product. Table 7 presents a summary with the different rates of oil removal as well as the average.

Product	Rate	Non-nutrient control	Nutrient control
Product	0.007	0.00013	0.004
Product	0.012	0.00013	0.004
Product	0.014	0.002	0.005
Product	0.017	0.0003	0.014
<b>OSE II</b> → Product	0.018	0.00013	0.005
Product	0.011	0.00013	0.005
Average	0.013	0.0005	0.005

Table 7 - Rates of oil removal for the products passing the O7G criteria (mg of oil/L-Day)

**OSE II** had the highest rate of oil removal of the 13 EPA NCP Listed Products tested.

According to these results the average half-life of the petroleum mixture for this specific experiment is approximately 40 days. Prior studies suggest a half-life for petroleum mixtures of approximately 2 months (Stewart et. al., 1993).

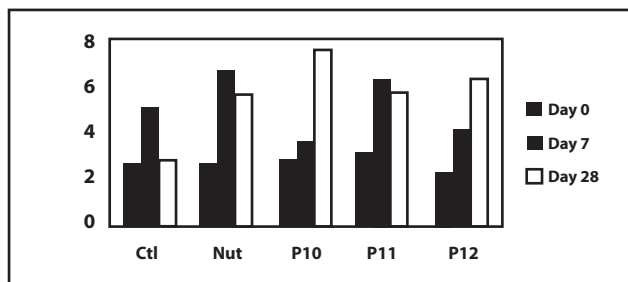


Figure 16 - MSN aliphatic degraders results (Batch D)

**OSE II** grew the highest number of oil degrading bacteria at  $10^{7.5}$ .

Products with a significant extent of oil removal show microbial counts in the order of  $10^5$  for the aliphatic degraders as presented in Figure 14, Figure 15, Figure 16, and Figure 17. Treatments with higher microbial populations, but similar degradation extents a compared with the control suggest the addition of an alternative carbon source other than the petroleum hydrocarbons.

Figures 32-34 show the composition of aliphatics, aromatics, and polars for batch D. As presented earlier for batches A and B, the aliphatic fraction is being degraded more severely than the aromatic fraction. The same results are found in the next two figures. Microbial counts for aliphatic degraders (Figure 16) show a higher number for Product 10, with a value of  $4.06 \times 10^7$  at day 28, as compared with the rest of the treatments in this batch, with values in the order of  $10^6$  at the most. This is reflected as a decrease in the aliphatic fraction composition from a 100% to 46% after 28 days.

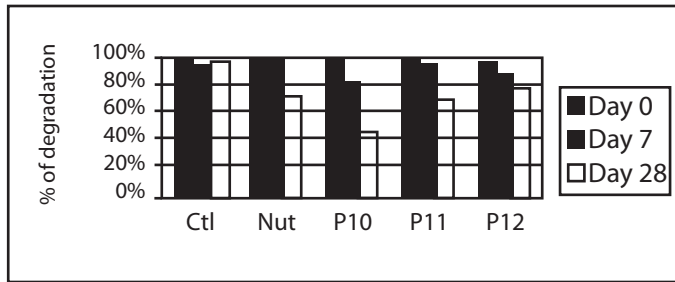


Figure 32 – Aliphatic fraction composition through time (% of degradation (Batch 1))

**OSE II** had the highest rate of degradation.

Products 10, 11, and 12 are decreasing in aliphatic and aromatic composition up to 50% for the aliphatic fraction and 25% for the aromatic. It is clear from these results that the oil is being degraded, and therefore, changing its composition. However, the aliphatic fraction is being degraded at a greater extent than the aromatic fraction, as mentioned before. Product 10 is showing a significant extent of hydrocarbons removal as presented in Figure 33 and Figure 34 for Product 10.

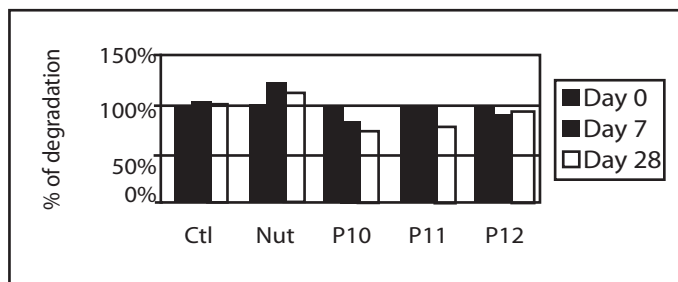


Figure 33 – Aromatic fraction composition through time (% of degradation (Batch D))

**OSE II** had the most (highest rate of) degradation of the aromatic fraction of the oil.

As presented in Figures 23 and 33 show the average of aliphatic fraction biodegraded was 34% (54% decrease for OSE II), while only 21% of the aromatic fraction showed to be biodegraded. The most degradation was by OSE II.

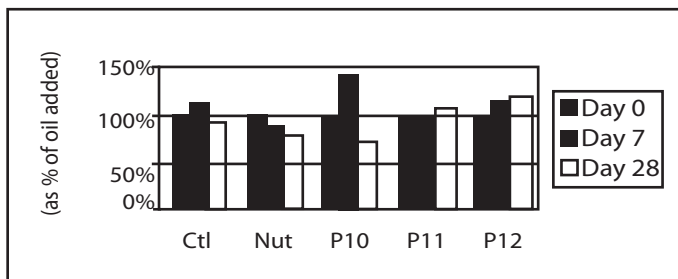


Figure 34 – Polar fraction composition through time as a percentage of the amount initially present (Batch D)

**OSE II** had the most or highest rate of (Polar) aromatic hydrocarbon degradation.