## BIOREMEDIATION: RESULTS OF THE FIELD TRIALS OF LANDEVENNEC (FRANCE)

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**ABSTRACT:** Results of studies performed all around the world on bioremediation techniques are not often comparable because they have been performed through different methodologies. To address this problem, several organizations have combined their efforts to define a common methodology to assess bioremediation techniques' efficiency through field experiments.

To confirm the validity of this protocol, a 9 month experiment was performed on oiled plots on a sheltered estuarine beach in France. In this experiment, biodegradation efficiency was assessed though multiple analyses (chemical and microbiogical) including changes in chemical composition of the residual oil.

During the past decade, bioremediation techniques for cleaning up oiled shorelines have been of great interest and many studies have been performed all over the world. However, as these studies have been performed using different methodologies, it is difficult to compare their results. To address this problem, the authors' organizations have combined their efforts to define a common methodology to assess the efficiency of bioremediation products to treat oil polluted shorelines.

To confirm the validity of this protocol, a 9 month experiment was performed on a coarse sand sheltered estuarine beach on the west coast of France using 5 replicate blocs of 3 plots, (oil, oil and nutrient, and unoiled plots). The Arabian Light crude oil used was preweathered  $(C_9 +)$ , and preemulsified (20% sea water), and applied at 3.8 L/m<sup>2</sup>. The fertilized oil plots, were treated with large additions of slow release fertilizer (monthly applications up to 60 g nitrogen/m<sup>2</sup>), to ensure that nutrients were not a limiting factor despite dilution by tidal immersion.

The biodegradation process was assessed by documenting the changes in chemical composition of the residual oil through multiple analyses (GC-FID/FPD, MS-fisher, and GC/MS). In addition, microbiological analyses were performed (bacterial counts, activity measurements, and respirometry), as well as a followup of the nutrient concentrations in the interstitial water.

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## Results

During the first two months following the initial application of fertilizers, the interstitial water nitrogen concentrations in all the plots treated with oil were much lower than those in the surrounding sediment. Following this period the nutrient concentrations increased unevenly in the fertilized plots, (between 1 and 5 times the coastal sea water concentration).

During the first 6 to 8 weeks, the oil concentrations in the sediments decreased rather quickly to 3,000 to 1,000 ppm (total hydrocarbons). However, until the end of the experiment, at 35 weeks, the loss of residual oil slowed considerably to final concentration between 900 and 100 ppm. Moreover, in a given plot, the decrease in oil concentration was not regular with time, due to uneven distribution of the oil in the plots; for example, higher concentrations were generally found at 16 weeks.

In this trial the straight-chain alkanes and their corresponding isoprenoids degraded at the same speed (Figure 1); this result excludes the use of  $c_{17}$ /pristane and  $c_{18}$ /phytane ratios to quantify biodegradation. Biodegradation rates were thus assessed by chemical analyses of the oiled sediment samples, to determine the concentration of oil in sediment relative to a natural biomarker (7a(H), 21b(H)-30norhopane) present in the oil (Figure 2). Biodegradation was faster during the first 8 weeks, and the oil seems significantly more degraded in the treated plots than in the untreated ones. However, considering the low nutrient concentration during the first weeks, these results may be attributed to the variability in each plot rather than to fertilizer effect. Beyond this period there was no significant difference.

Individual compounds within the oil degraded at different rates during the experiment. Initially, the oil contained 37% of saturates (9% aliphatics and 28% cyclo-alkanes), 33% aromatics, and 30% resins-asphaltenes; after 35 weeks, the residue was composed of 24% saturates, 28% aromatics, and 48% resins-asphaltenes; the overall biodegradation of the oil can be assessed at 34%, (77% for the alipha-



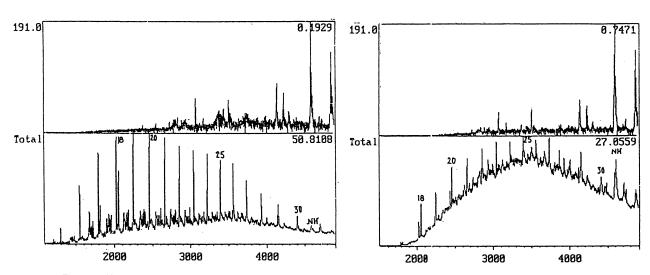


Figure 1 (A & B). Evolution of the alkane fraction during the experiment; and identification of the C<sub>30</sub> norhopane

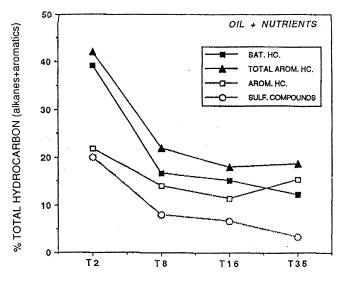


Figure 2. Evolution of the oil composition (example of the MS-fisher analysis performed on the alkanes + aromatic fraction)

tics, 48% for the cycloalkanes, 40% for the aromatics, and 0% for the resins and asphaltenes).

The importance of statistical analysis for the experimental design and data analysis has been illustrated during this experiment. Clear evidence of enhanced oil biodegradation was not observed in the chemical and microbiological results because of the high variability in the data results both within and between the different plots.

These results were taken into consideration in finalizing the experimental protocol during the meeting of the working group held on the experiment site in September 1994.

## Author

Graduated in chemistry and experienced in oil drilling, François-Xavier Merlin joined CEDRE in 1979. He is in charge of the studies and development dealing with treatment products to be used on oil spills. He has published various publications and handbooks on this subject.