

Preliminary study regarding microbial diversity in landfarm soil by PCR-DGGE and prospection of surfactant and dibenzothiophene degradation activities

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Soil microorganisms have a high metabolic diversity, which allows them to inhabit different environments and interact with all kinds of substrates present in the middle part of the soil profile. The study of microbial communities that play a role in the biodegradation of the soil has been a challenge for microbiologists since less than 1% of these microorganisms are capable of cultivation. In view of this bias, the metagenomic tool has been used to understand the profile of the soil microorganisms and has enabled us to assess their biotechnological potential. The goal of this study was to evaluate the effects of oil-enrichment on diversity and functional structures of microorganisms from landfarm soil (Lf). For this purpose, metagenomic DNA was extracted from Lf and Lf enriched with petroleum (Lf⁺). The PCR/DGGE technique was used to compare the electrophoretic profiles of 16S rDNA amplicons for bacterial and archaeal communities, and 28S rDNA amplicons for yeast. Two metagenomic libraries of high molecular weight DNA were built using PCC2 fosmid vectors to perform the search for genes involved in the synthesis of biosurfactants and the degradation of dibenzothiophene (DBT). The clones were cultivated on standard medium, and screening of biosurfactant-producing colonies was performed based on the qualitative drop-collapse test, using mineral oil, diesel and petroleum as substrates. DBT biodegradation was verified by growing the clones for 72 h on enriched mineral medium with DBT as a sole sulfur source. The electrophoretic patterns have shown that the oil has a great influence on the diversity of archaeal communities, but only a minor influence on the bacteria and yeast in the Lf soil. The two libraries totalized 8700 fosmid clones. None of the clones could mineralize the DBT. However, five clones demonstrated surfactant activity, one from the Lf

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sample and four from the Lf^+ soil. Two clones from the Lf^+ sample were only biosurfactantpositive after cell-lyses by sonication. The results suggest that the oil-enrichment may not greatly interfere in the microbial diversity of the Lf soil, but it is very important for prospection of molecules with biotechnological applications, like biosurfactants. These results also show that the secondary compounds need to be correctly added to the extracellular medium to demonstrate surfactant activity.

Key words: Metagenomic; Landfarm; PCR/DGGE; Fosmid; Surfactants

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