

COMMUNITY STRUCTURE AND FUNCTION ANALYSES OF A HMW PAH-DEGRADING SOIL MICROBIAL CONSORTIUM



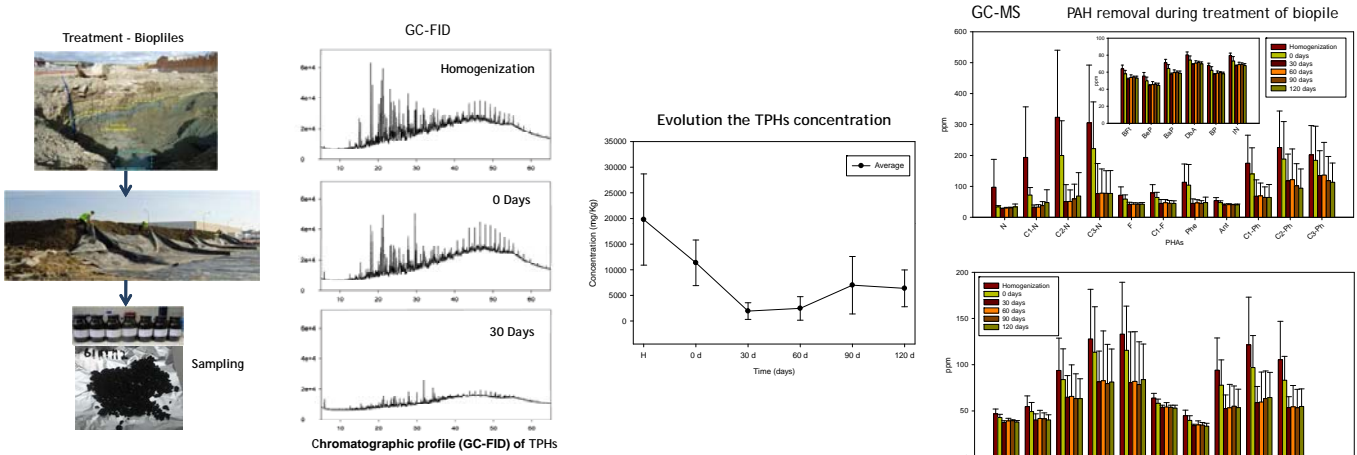
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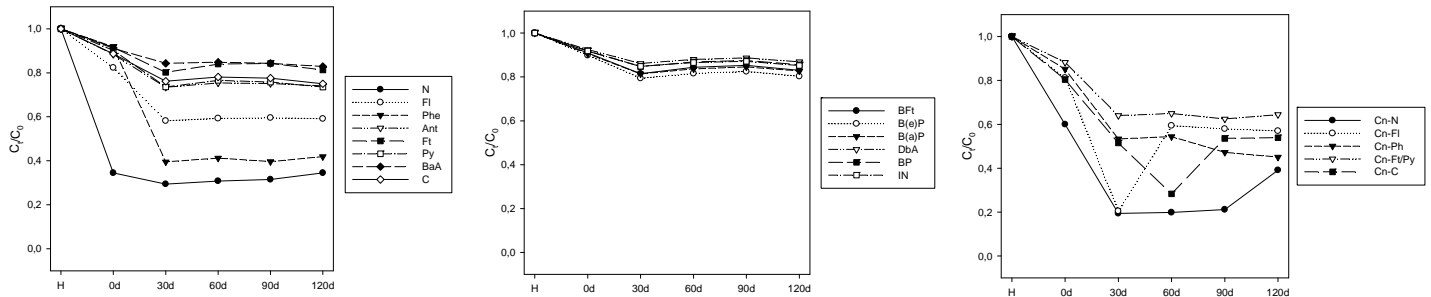
INTRODUCTION

The main interest of our group is to understand the microbial processes determining the fate of PAHs in soils, and their application in the optimization of bioremediation technologies and in risk assessment. We have a broad experience in the analysis of metabolic pathways involved in the degradation of single PAHs or PAH- environmental mixtures (creosote, crude oil) by single bacterial strains and enrichment cultures (consortia). Here we investigate the changes in hydrocarbon composition during the bioremediation of a fuel polluted soil and those in the microbial community structure in order to link the disappearance of specific substrates to key natural microbial populations. The industrial soil under study was polluted due to repetitive accidental leaks of fuel containing underground storage tanks during fifty years. The preliminary risk assessment analysis recommended an aerobic biopile treatment. The homogenated soil was amended with nitrogen and phosphorous and treated in actively aerated biopiles during 90 days. Monthly soil samples were analysed for hydrocarbon composition (GC-MS) and microbial community structure using both molecular (PCR amplification of 16S rRNA genes followed by DGGE and clone library analysis) and culture-dependent methods. Chemical characterization of the residues will be used to investigate possible strategies to reduce the end-point concentrations.

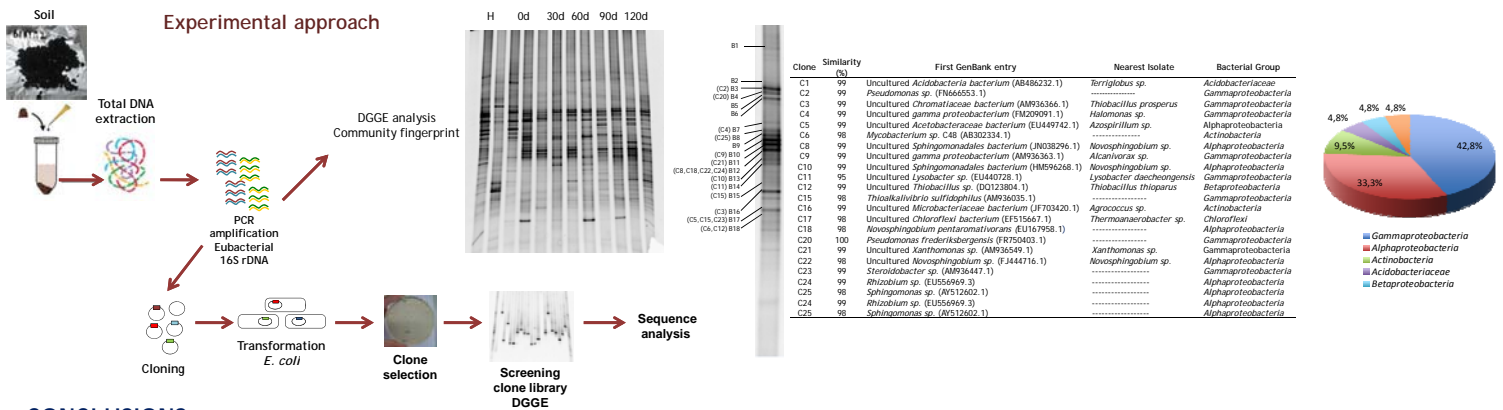
BIODEGRADATION OF TPHs AND PAHs IN THE AEROBIC BIOPILE



DEGRADATION OF PAHs AND ALKIL-PAHs



COMMUNITY STRUCTURE ANALYSIS AND THE DINAMICS



CONCLUSIONS

- The designed system that uses sand in order to improve the biodegradation of the residues of the creosote degradation is a good method to enrich a natural community with HMW PAHs degrading microbial populations.
- A microbial consortium with a stable community and a capacity to degrade HMW PAHs was obtained by this method. Although the results show that, for the purpose of retrieving recalcitrant compounds degrading bacteria, the incubation time in every transfer should be prolonged.
- The characterization of the chrysene degrading subpopulation was started and it was demonstrated that this community was able to degrade the 36% of the chrysene supplied as the only source of carbon in six weeks.
- The majority of the clone library members belong to Gammaproteobacteria (44.7%), like *Pseudoxanthomonas* (described as a pyrene degrading bacteria) and *Pseudomonas*, which have been related to the degradation of PAH. Alphaproteobacteria (29.8%) also constitute an important group, appearing species of Sphingobium (described as a pyrene and fluoranthene degrading bacteria), *Azospirillum* (related to the bioremediation of a creosote-contaminated soil) and *Bradyrhizobium* (isolated from contaminated soil). Furthermore, 19.1% of clones belong to CFB and the rest are Betaproteobacteria (6.4%) like *Achromobacter*.
- Obtaining a HMW PAH degrading community and its first characterization will allow knowing in depth the populations involved in the elimination of these products in contaminated soils. So, relations between structure and function of every population could be established.