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## **Extended Abstracts**

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title: **DNA-microarrays for monitoring natural attenuation of emissions from abandoned landfill sites in contaminated groundwater plumes**

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Degradation of contaminants by microbial communities in soil and groundwater is the main activity in natural attenuation. Due to the complexity of the emitted organic components, natural attenuation of contaminated groundwater plumes from abandoned landfill sites can be evaluated only to a very limited extent by instrumental analysis.

The influence of pollutants on growth characteristics as well as composition of microbial communities in groundwater has been confirmed by denaturing gradient gel electrophoresis (DGGE) of PCR-amplified marker-gene fragments such as the 16S rRNA gene (Röling et al., 2001). Until today, PCR-DGGE is the method of choice to generate genetic fingerprints for comparison of microbial communities involved in degradation of a variety of contaminants. PCR-DGGE has some major disadvantages: It is strictly site-dependent and does not facilitate identification of microbial communities involved in degradation of contaminants and, therefore, the precise monitoring of natural attenuation.

Over the past years developments in the field of microbial ecology improved considerably. The application of DNA-microarrays in the area of marine biology as well as soil- and water ecology facilitated the analysis of microbial communities on species-level by specific detection of microbial rRNA-genes (Loy, 2005; Peplies, 2004). These findings led to the assumption that DNA-microarray-technology is applicable for monitoring natural attenuation of contaminated groundwater. Compared to other methods, which are state of the art in molecular biology, the potential to analyze a large number of parameters in parallel is a major advantage of DNA-microarrays. Based on known probe-sequences the composition of microbial communities can be analyzed on any taxonomic level.

To overcome the limitations of PCR-DGGE and to apply the benefits of DNA-microarray technology to environmental analytics, a 16S rRNA directed DNA-microarray for the analysis of microorganisms, which have been detected in soil and groundwater, was designed (Kühn et al., 2009). The main objective in microarray-design was to detect as many microorganisms as possible with a minimal number of probes. Therefore a higher taxonomic level was selected to detect bacterial families instead of single species. Since more than 90% of the existing prokaryotes are still unknown, a microarray for the detection of microorganisms on species level is not applicable for routine monitoring of natural attenuation.

The DNA-microarray facilitated the simultaneous detection of Bacteria and Archeae in groundwater for the first time ever and confirmed results obtained with PCR-DGGE. Furthermore the composition of microbial communities in tested groundwater-samples could be assessed. It could be shown that the Bacteria to Archeae ratio is a function of the groundwater contamination. DNA of sulfate-reducing bacteria from the families *Desulfobacteraceae*, *Desulfobulbaceae*, *Desulfovibrionaceae* and others could be detected in every analyzed groundwater sample. The fraction of sulfate-reducing bacteria is significantly increased in contaminated groundwater samples and therefore a marker for contamination. In contrast to the ubiquitously prevailing sulfate-reducing bacteria, DNA of methane-producing Archeae from the families *Methanobacteriaceae*, *Methanomicrobiaceae* and *Methanosarcinaceae* can be detected in contaminated groundwater exclusively. Hence, a detailed analysis of methane-producing Archeae provides a marker for contamination of ground water.

Due to the selection of defined microorganism families and the PCR-parameters chosen for the amplification of 16S rRNA Genes the analytical significance of the developed DNA-microarray is

somewhat limited. In spite of the limitations mentioned above, combined with determination of microbial concentration, the family-specific DNA-microarray facilitates quantitative and functional statements regarding the composition of microbial communities in groundwater and therefore the evaluation of natural attenuation of contaminations along the downstream flow from abandoned landfill sites

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