Growth of Dechlorinating Bacteria in Enrichment Cultures, Derived from Contaminated Soil and Sediment from Guadeloupe, that Dechlorinate Hexachlorocyclohexane Isomers

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Background/Objectives. Hexachlorocyclohexane (HCH) isomers are polychlorinated organic compounds of environmental concern. A mixture of different HCH isomers (α, β, δ and γ), known as technical-grade HCH and γ-HCH (lindane) were extensively used as pesticides in banana plantations in the French West Indies from 1950 to 1972. HCH molecules are chemically stable and are transferred from one compartment of the environment to another and persist for a long time in soil (several decades). HCH isomers are ubiquitous in all environmental compartments and in the trophic chain. In particular, beta-HCH, is the most difficult isomer to degrade in both water and soil, because of the equatorial position of the chlorine atoms (Doesburg et al., 2005; Phillips et al., 2005). Given its intensive use as pesticides, HCH contamination is a worldwide problem. HCH isomers can be subjected to microbial biodegradation. However, to date, the anaerobic biotransformation of HCH isomers is not well characterized. The growth of specific bacterial genera involved in the dechlorination of HCH isomers has yet to be demonstrated.

Approach/Activities. In this study, we aimed to identify the bacterial populations that grow as a result of the dechlorination of α-HCH and β-HCH after performing a 5% dilution transfer of two enrichment cultures that independently and steadily dechlorinate these isomers. Dechlorination products were measured periodically by GC-FID and liquid culture was sampled for DNA extraction as well. Previous sequencing studies had identified *Dehalobacter* and *Geobacter* as candidate dechlorinators in the α-HCH and β-HCH enrichment cultures. *Dehalobacter* is known to be a genus capable of dechlorinating β-HCH (Doesburg et al., 2005). Thus, we performed quantitative Polymerase Chain Reaction (qPCR) to track the growth of *Dehalobacter* during the dechlorination of β-HCH and α-HCH. We will perform qPCR for *Geobacter* as well.

Results/Lessons Learned. The final stable products of the dechlorination of α-HCH and β-HCH were benzene and monochlorobenzene (MCB). The ratios of benzene to mono-chlorobenzene are 0.58 ± 0.130 for α-HCH and.76 ± 0.16 for β-HCH. In accordance with the theoretical predictions based on the stereochemical structures of the HCH isomers, the alpha and beta isomers with even numbers of axial / equatorial chlorine atoms on the carbon skeleton produce more benzene. For the α-HCH cultures, the relative abundance of *Dehalobacter* increased from 2.18%±1.9% on day 0 to 34.66%±7.6% on day 63. For the β-HCH cultures, *Dehalobacter* increased from 1.21%±1.1% on day 0 to 13.52%±4.8% on day 63. So far, we can conclude that *Dehalobacter* spp. are involved in the dechlorination of these isomers. At the time of the Conference, we expect to have the qPCR results for *Geobacter.* It is conceivable to have both *Dehalobacter* and *Geobacter* involved in the dechlorination of α-HCH and β-HCH. Knowledge on the microorganisms that can dechlorinate HCH isomers is essential for the application of potential bioremediation approaches and/or implementation of monitored natural attenuation.