Continuation of an Investigation into the Anaerobic Intrinsic Bioremediation of Whole Gasoline

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Title: Continuation of an Investigation into the Anaerobic Intrinsic Bio remediation of Whole Gasoline
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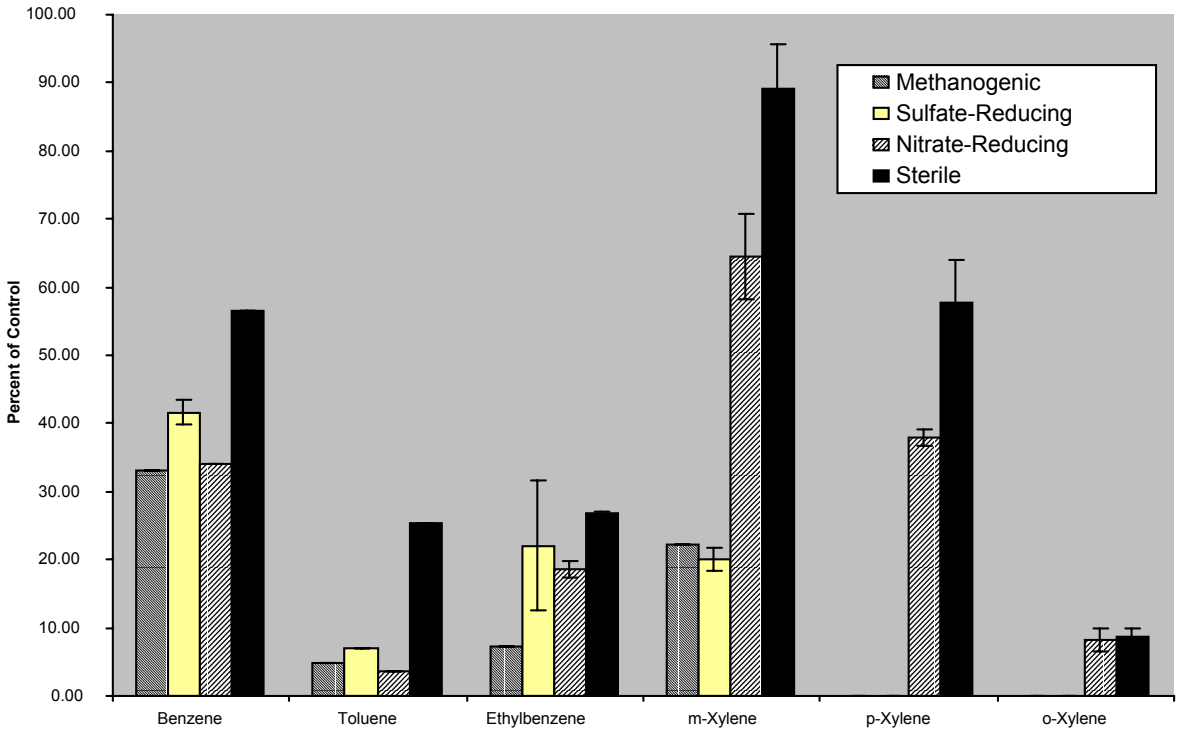
Description:

Third Quarterly Report

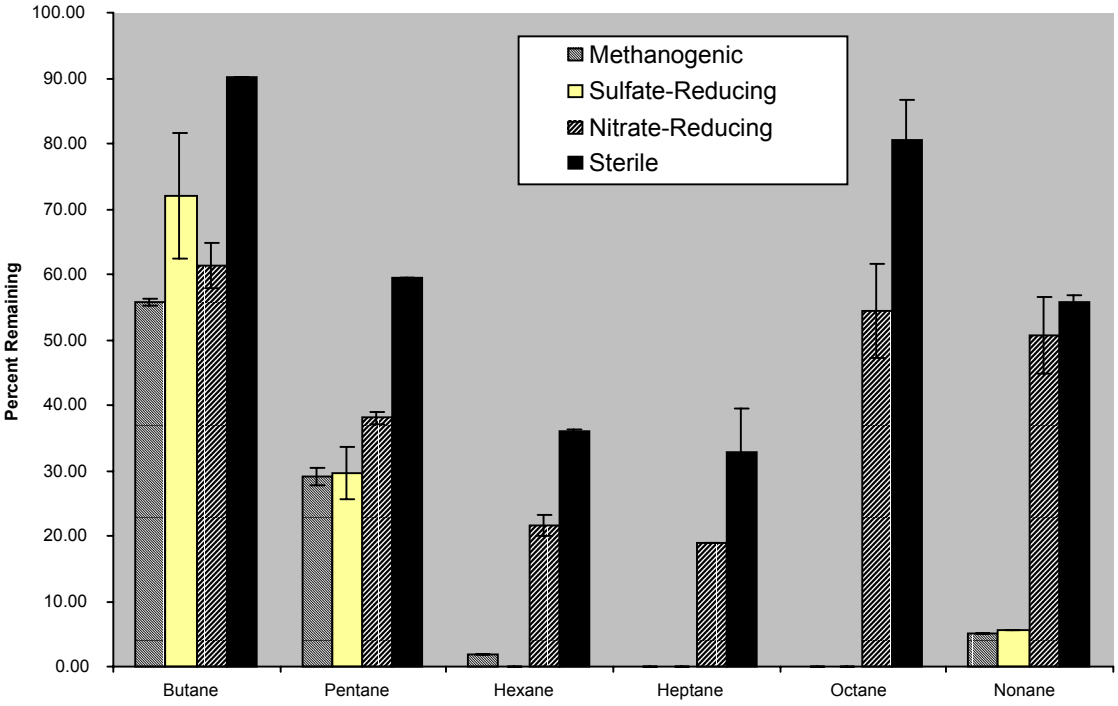
In the previous quarter, we developed a method for the simultaneous monitoring of over 50 volatile components of gasoline in anaerobic incubations by headspace analysis. This technique has afforded us the opportunity to determine the fate of the most volatile fractions of gasoline at extremely low concentrations as well as reexamine incubations assembled during the initial funding period. These incubations were amended with 10 µl of gasoline in July of 1999. They have been incubated anaerobically under methanogenic, sulfate-reducing, and nitrate-reducing conditions. The residual concentrations of individual gasoline components are compared in all treatments to freshly-made standards and reported as the percent remaining in comparison to a fresh 10 µl standard

Generally, we found a wide variety of hydrocarbons were biodegraded under both methanogenic and sulfate-reducing conditions. With one exception, toluene, hydrocarbons were not biodegraded by these aquifer slurries in the presence of nitrate. The following graphs show the extent of degradation of the individual components of gasoline.

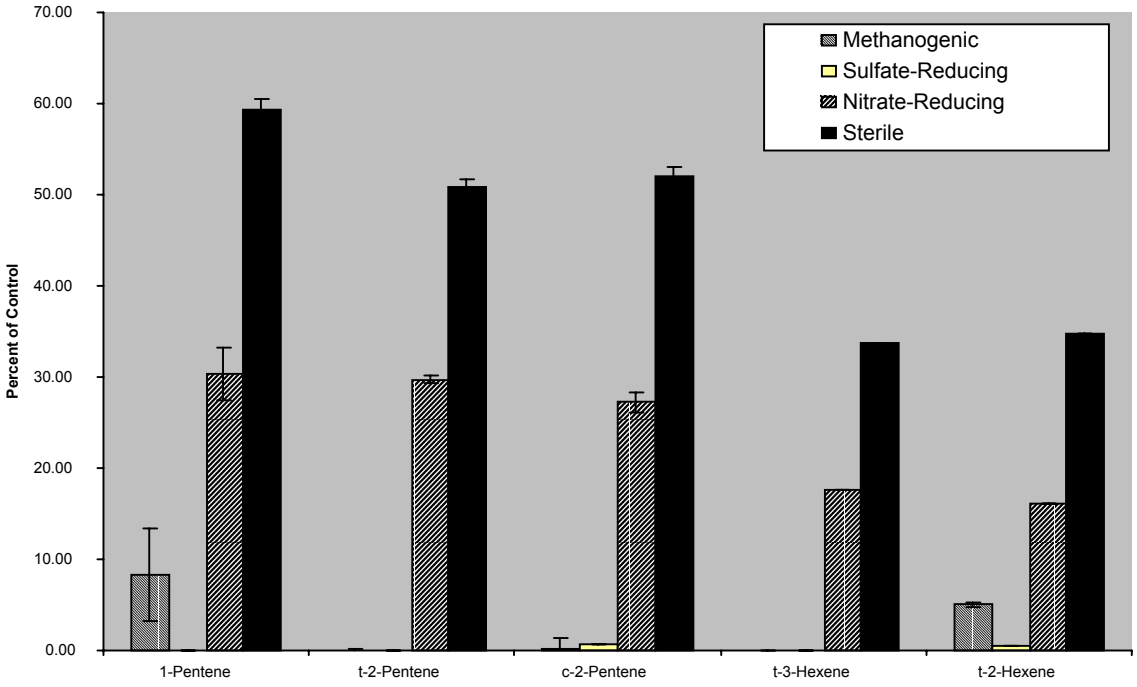
BTEX Compounds



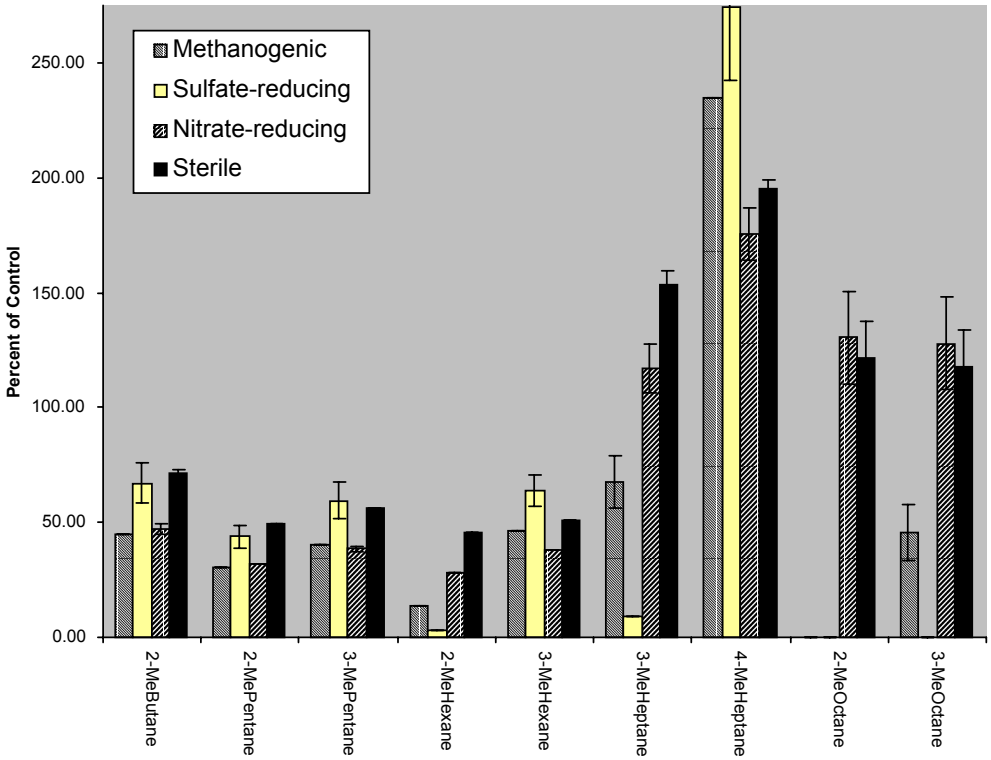
n-Alkanes



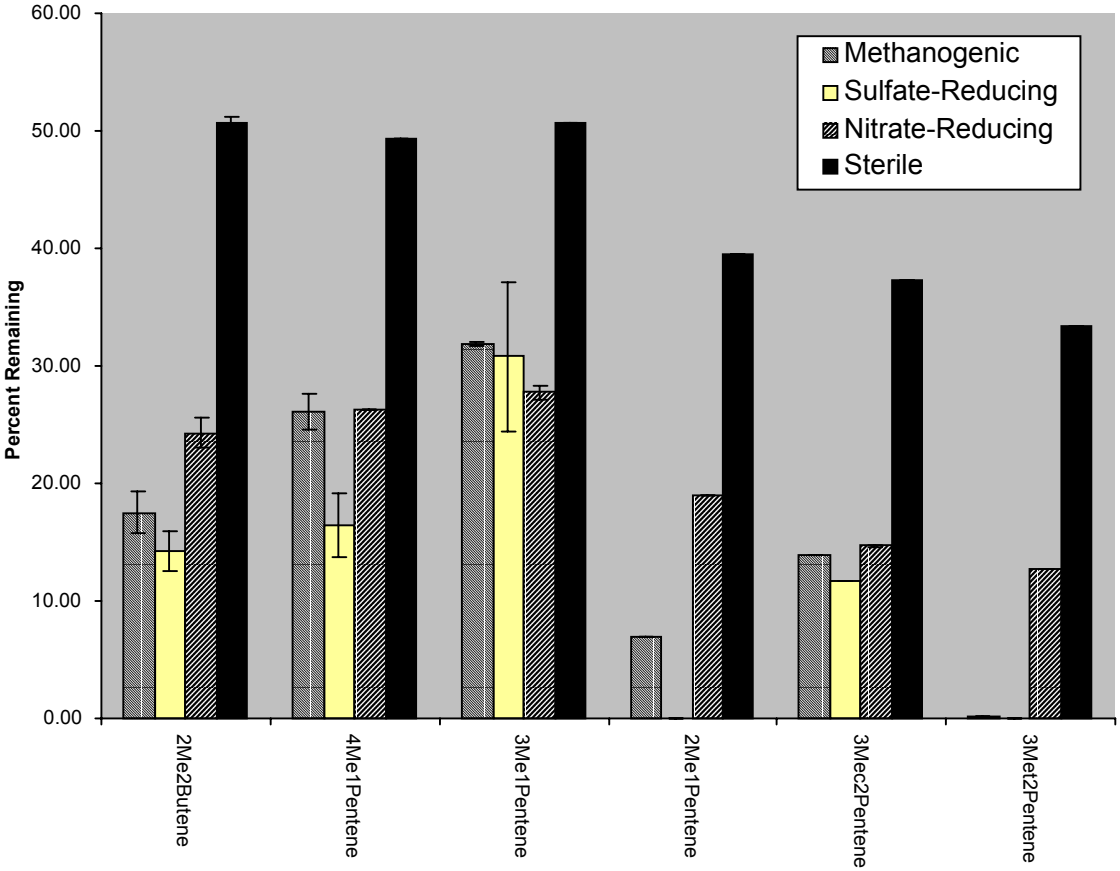
n-Alkenes



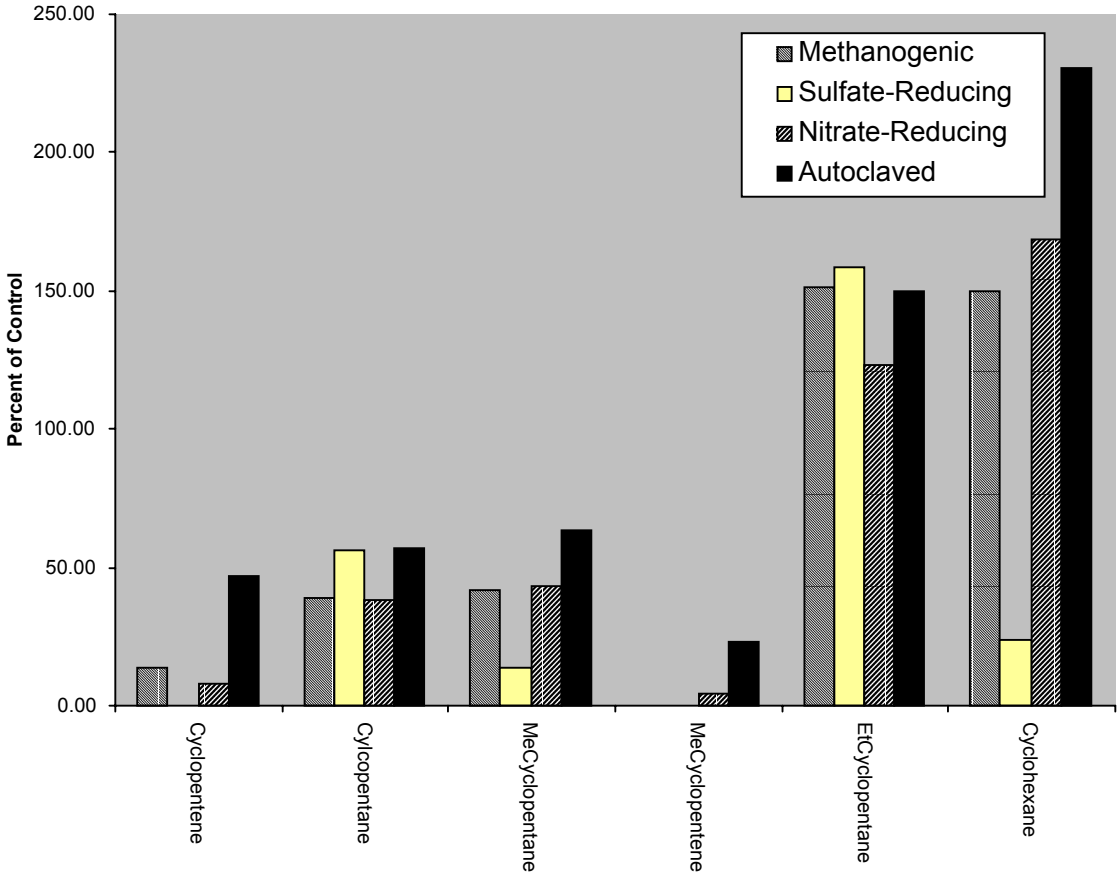
Branched Alkanes



Branched Alkenes



Alicyclic Compounds



Summary

The resident microbiota of the aquifer underlying the Ft. Lupton natural gas field harbor diverse abilities to metabolize the hydrocarbon constituents of gasoline in the absence of oxygen. Nitrate does not support hydrocarbon biodegradation at this site with one notable exception, that being the metabolism of toluene. Sulfate is the preferred electron acceptor though many of the hydrocarbon compounds studied were biodegradable in its absence, most likely with methanogenesis serving as the terminal electron-accepting process.

Of the BTEX compounds, benzene appears to remain recalcitrant under all reducing conditions, while toluene and the xylene isomers were biodegraded under both methanogenic and sulfate reducing conditions. Ethylbenzene was one of the few gasoline components in which greater biodegradation was observed under methanogenic than sulfate-reducing conditions.

The C6 to C9 alkanes were biodegraded under both methanogenic and sulfate-reducing conditions. The shorter chain pentane and butane were not metabolized to an appreciable extent, demonstrating a limit of biodegradative abilities by these sediments. n-Alkenes appeared to be even more labile than the n-alkanes, and these substrates were consumed regardless of the position or conformation of the double bond. Although branched alkanes were more recalcitrant, they are not found to be completely resistant to anaerobic decay. Notably, 2-methylhexane, 3-methylheptane, and 2- and3-methyloctane were found to be biodegraded. The relatively high levels of 4-methylheptane demonstrate this compound's persistence within the contaminated

sediments and continued recalcitrance during the laboratory incubation. Our results from the alicyclic fraction of the gasoline reveal striking differences in biodegradation based upon both structure and presence of electron acceptor and novel activities. Most striking was the complete recalcitrance of cyclopentane contrasted with the biodegradation of both methylcyclopentane and cyclohexane only in the presence of sulfate.

As we continue to monitor this and other studies and compile data, definitive patterns emerge regarding the rate and extent of anaerobic biodegradation of the individual components of gasoline. These studies document, for the first time, the order of anaerobic degradation of a complex hydrocarbon mixture when it is present at an environmentally-relevant concentration.