

# Friend or Foe? Microbial impact of Calcigel bentonite on metal materials used for nuclear waste repository

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## Abstract

Multi-barrier concept is a favorable option to store high-level nuclear waste (HLW) in a deep geological repository. Bentonites are processed clay materials that are considered as a geotechnical barrier for metal containers storing HLW. To understand the impact of indigenous microorganisms from bentonites on these metal materials, anaerobic microcosms incubating Calcigel bentonite, synthetic Opalinus clay (OPA) porewater, lactate (one of the organic acids in natural OPA porewater) or H<sub>2</sub> gas (product from anaerobic metal corrosion) with or without cast iron metal plates were conducted for up to 9 months in triplicates for each condition and time point (sampling every 3 months).

The amplicon sequencing targeting V4 region of 16S rRNA genes showed that microbial communities of raw Calcigel bentonites mainly comprised phyla Acidobacteria, Actinobacteria, Chloroflexi, Firmicutes, Proteobacteria and Methylophilota. In the microcosms with lactate, enrichment of *Bacillaceae* (Firmicutes) and uncultured MB-A2-108 (Actinobacteriota) were observed; whereas in the presence of both lactate and cast iron, genera of Firmicutes, namely *Desulfotomaculum*, *Desulfitobacterium* and *Desulfallas-Sporotomaculum*, were highly enriched (relative abundance ranged from 60% to 95%) associating with large decrease in sulfate and lactate concentration. These bacteria appeared to be driven by H<sub>2</sub> gas generated from metal corrosion. Moreover, SEM-EDX analyses showed that the metal surface was corroded and covered by a carbonate passivation layer. In this layer, FeS appeared to be formed, further suggesting the influence on cast iron corrosion and formation of secondary minerals induced by sulfate-reducing bacteria.

On the other hand, we supplied N<sub>2</sub> gas mixed with H<sub>2</sub> and CO<sub>2</sub> (80:10:10) to stimulate growth of H<sub>2</sub>-oxidizing sulfate reducers. GC analyses showed that in the microcosms without cast iron, the content of H<sub>2</sub> gas in the headspace decreased accompanying with decrease in sulfate concentration (measured via IC). However, in the microcosms with cast iron we noted large accumulation of H<sub>2</sub> gas (~ 5 times more than

initial value) and greater decrease in sulfate concentration. Similarly, surface corrosion was visible by SEM-EDX, and a carbonate passivation layer with possible FeS precipitates was formed on the metal surface but in a shorter timeframe (3 months). Hence, we speculated that certain autotrophic H<sub>2</sub>-oxidizing sulfate reducers also corroded cast iron metal, and their taxonomy and mechanisms will be identified using metagenomic approaches.

Altogether we concluded that microbial communities in Calcigel bentonites lead to microbially induced corrosion for cast iron under certain conditions, yet interestingly, the formation of passivation layer enhances the resistance for further metal corrosion. The actual impact of indigenous microorganisms in different bentonites, either disadvantageous or beneficial, on metal containers for HLW requires comprehensive investigations.

## **Keywords**

Bentonite, MIC, corrosion, nuclear waste repository, sulfate-reducing bacteria, cast iron

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The authors have declared that no competing interests exist.