Microbiology In Nuclear waste Disposal

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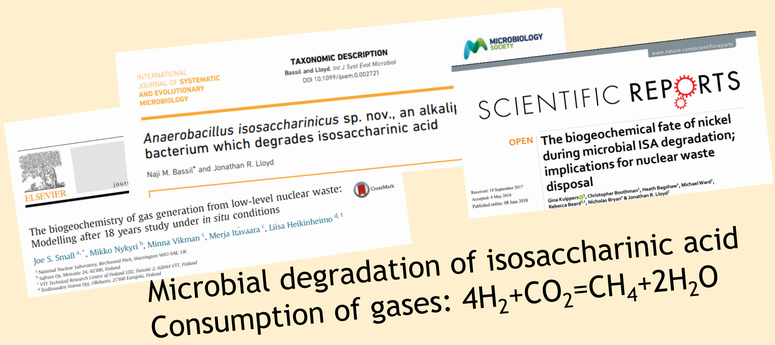
OBJECTIVES

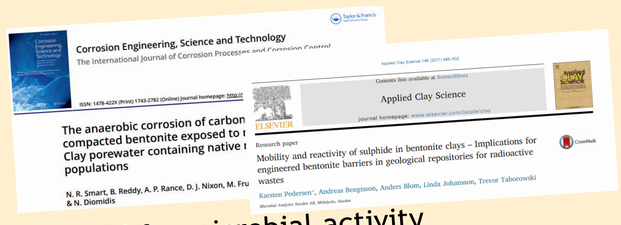
The objectives of the project target key technical issues such as **Waste forms and their behaviour** and **Technical feasibility and long-term performance of repository components.** This project will increase the understanding of how microbial processes will influence the safety and performance of future repositories.

HIGHLIGHTS

Microbial impact on the repository has both positive and negative aspects eg. degradation of complexants(+) and formation of sulfide(-).

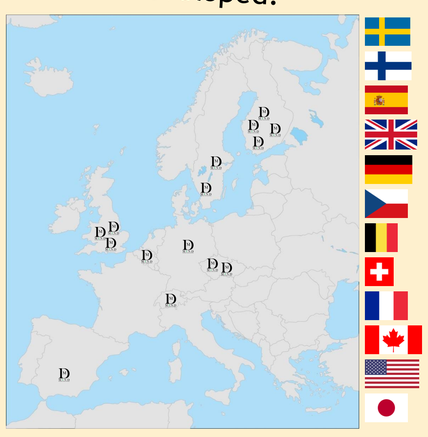


Microbial degradation of isosaccharinic acid
Consumption of gases: $4H_2+CO_2=CH_4+2H_2O$



Threshold for microbial activity
Sorption of sulfide in the buffer
Iron in the rock matrix acts as a sink for sulfide

An advanced training course has been developed.



A network of experts from 12 countries. MIND member

MIND PROJECT WORKPACKAGES #1, #2 and #3

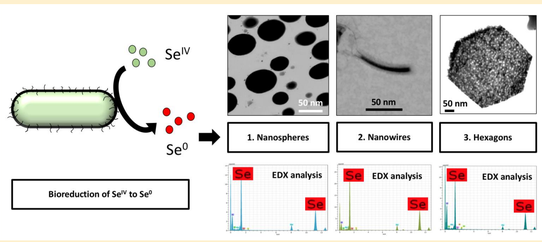
#1 Behaviour of organic containing long-lived ILW

This WP addresses remaining uncertainties concerning the release of chemical, gases and radionuclides that are important to the safety case for ILW disposal. Research has focussed on microbiological processes occurring under alkaline conditions representative of the concrete based engineered barrier system. Outputs from laboratory and *in situ* experimental systems (Deliverables 1.1-1.7).

The biodegradation of organic polymers such as cellulose, PVC, ion exchange resins and bitumen have been studied after ^{60}Co γ irradiation and under alkaline (pH 10-12) conditions

- A novel bacteria capable of degrading isosaccharinic acid (ISA) has been characterised and sequenced [1][2] Cellulose degradation is enhanced after irradiation but subject to fermentation, which neutralises pH.
- Plasticisers present in PVC are biodegradable at pH 10, but the PVC polymer is resistant to biodegradation and thus would not contribute to gas generation [3].

PVC materials before and after irradiation.



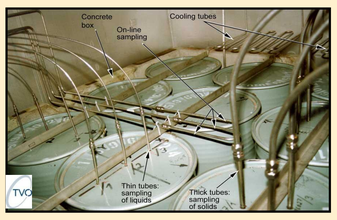
Selenium nanoparticles associated with a bacterium.

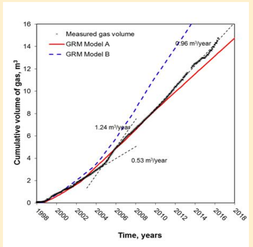
Radionuclide interactions have been examined by spectroscopy and high resolution electron microscopy studies of U, Eu, Se, Ni interactions with cellulose degradation products and microbial cells.

- Characterisation of *Stenotrophomonas bentonitica* that is able to immobilise Se(IV) by bioreduction[4].
- The fate of Ni initially complexed with ISA that is utilised by sulfate reduction processes was examined [5].

Methanogenesis has been studied under *in situ* conditions at the Mont Terri rock laboratory and through the long term LLW gas generation experiment (GGE) at Olkiluoto.

- Results from the Mont Terri MA experiment indicate that CH_4 generation from hydrogen is outcompeted by sulfate reduction in Opalinus Clay.
- Modelled data from the GGE indicate an increase in rate of CH_4 generation after 8 years as a result of neutralisation of initially high pH (11) regions of the experiment by microbial degradation of cellulose [6]. The model results are consistent with DNA studies.





The gas generation experiment has provided further insights into microbial gas generation.

#2 Microbial processes on HLW in geological disposal

Near-field safety issues related to the engineered barriers canisters, bentonite clays and concrete are addressed here. Three main safety issues can be postulated due to bacterial activity.

1. Bacterial sulfide production in buffer and backfill and the concomitant risk for sulfide corrosion of canisters.
2. Bacterial acetate production in buffer and backfill and the concomitant risk for acetate induced stress corrosion cracking of canisters.
3. Bacterial degradation of buffers and backfill, and of concrete constructions such as tunnel plugs.

A laboratory scale MX-80 bentonite storage experiment simulated bentonite behaviour in interfaces of bentonite, host rock fractures. The bentonite was studied as a slurry in which water, gases, nutrients and microorganisms were able to move freely at the temperature hospitable for microorganisms. After one year of storage under aerobic and anaerobic conditions, no essential changes in bentonite mineralogy was found but there was clear microbial activity in terms of sulfide that affected the bentonite water-phase chemistry and bentonite cation exchange capacity. These effects were not detected in sterile controls, demonstrating their microbial origin (Deliverable 2.8).

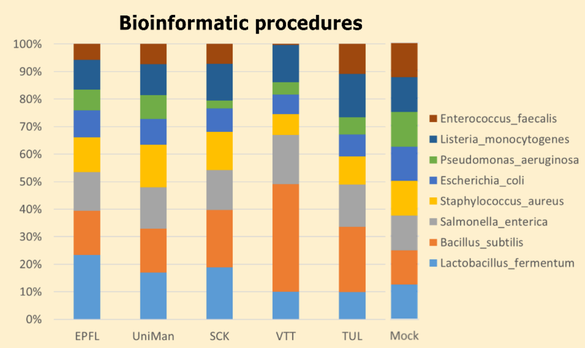
With microbes Without microbes

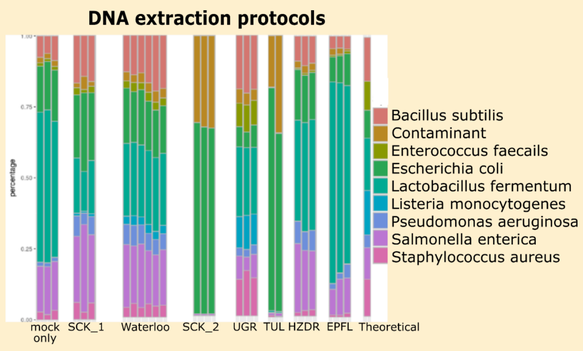
Sulfide reacts with bentonite clays
Transport of two different amounts of sulfide added to bentonite clays after 90 days of water saturation to 2000 kg m^{-3} wet density. There were significant amounts of S^0 in the black layers. These processes will cause an S immobilisation effect that can reduce the mass of sulfide that reacts with metal canisters over repository life times which may influence the longevity of metal canisters. However, the concomitant reduction of structural ferric iron may be problematic due to the destabilizing effect of ferrous iron on dioctahedral smectites (Deliverable 2.4).

#3 Integrate, communicate and disseminate results and conclusions from #1 and #2

Comparison of methods

The **bioinformatics analysis procedure** that is used to analyse microbial communities is highly diverse between different MIND partners. To compare the outcome of these pipelines, **DNA of a mock community** was **analysed by 5 partners. The different procedures scored quite well**, however, there were some differences in the results which should be taken into account when data are being compared. In a next step, an experiment was initiated to compare different **DNA extraction protocols to recover DNA from a clay-rich environment**. To this end, Opalinus Clay spiked with a cell mock community was distributed to different partners. Extracted DNA samples were collected, amplified and the V4 region of the 16S rRNA gene was sequenced. Seven distinct DNA extraction protocols were compared and **diverse results** were obtained. One method had a strikingly higher yield compared to other methods, while another method was outstanding in reassembling the mock community.





Education

Throughout the project, **2 courses** related to microbiology in nuclear waste disposal were organized.

An **exchange program** was set-up by the MIND partners with different internships among different partners

Communication

To study the impact of the inclusion of microbiology on expert conceptualization and public perception of geological disposal, **group discussions, interviews and an interactive workshop** were organized with implementers, project participants and civil society.

References

[1] Bassil N, Lloyd J 2018 DOI:10.1128/genomeA.01493-16
[2] Bassil N Lloyd J 2018 DOI 10.1099/ijsem.0.002721
[3] Nixon S et al 2018 DOI: 10.3389/fenvs.2018.00097
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[5] Kuipers et al 2018 (DOI: 10.1038/s41598-018-26963-8)
[6] Small et al (DOI: 10.1016/j.apgeochem.2017.07.012

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