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Impacts of Microbial Heterogeneity on Degradation of Pesticides in Soil and Groundwater Aquifers

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oil biodegradation, it is clear that our understanding of crude oil alkane degradation in the absence of exogenous electron acceptors is far from complete.

**K8
Marine benthic archaea - the unseen majority:
a geochronist's perspective**

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Marine sediments play a central role for our planet's redox balance and climate but, constraining the importance of the deep biosphere in this context, remains a great challenge. Recent molecular work based on membrane lipids and DNA suggests that unique and diverse benthic archaea with no cultured representatives constitute a sizeable, if not dominant, fraction of the deep biosphere. However, the validity of various molecular markers for the detection and quantification of prokaryotic biomass remains controversial; potential problems and experimental strategies for addressing these will be addressed. Several lines of evidence suggest that benthic archaea in sub-seafloor sediments are largely heterotrophic, i.e., they are probably involved in the slow degradation of aged and recalcitrant organic matter. The details on how benthic archaea utilize the highly refractory organic matter and which fraction thereof need to be explored and are relevant to our understanding of the deep biosphere in the carbon cycle. The lecture will review recent evidence on the mass and distribution of benthic archaeal communities in marine sediments, discuss their impact on widely applied lipid proxies for the reconstruction of past sea-surface temperatures, and highlight the exciting open questions and avenues for future research.

**S9-11
Geomicrobiology of extremely acidic subsurface environments**

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Abandoned underground mines, when accessible, can provide significant insights into the diversity of life in subsurface environments. In the case of metal and/or sulfur (pyrite) mines, the oxidation of sulfidic minerals which is greatly accelerated by access to oxygen, can lead to extremely acidic conditions developing, with underground streams and pools having pH values of < 3, as well as containing significant mineral acidity in the form of soluble iron, aluminium and manganese. Acidophilic prokaryotic communities have been described within the Richmond mine at Iron Mountain, California, and the Frasassi cave system in Italy. Research centred at two contrasting mine sites in north Wales have revealed extensive and unexpected microbial diversity and dynamic geomicrobiology, mostly involving redox transformations of iron and sulfur. One site, a former pyrite mine, has been abandoned for almost 100 years and within this time has become populated with massive growths (>100 m³) of microbial slimes, streamers and stalactites. The other, a former copper mine, has become accessible since the underground water table was lowered in 2004. In both cases, novel genera and species of acidophilic bacteria and archaea have been encountered, and isolates obtained. Psychrotolerant chemolithotrophic acidophiles are particularly successful in exploiting these environments.

**S9-01
Diversity and distribution of Fe(II)-oxidizing and Fe(III)-reducing microorganisms in salt lake sediments of Southern Russia**

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The extreme hypersaline conditions prevailing in hypersaline environments result in decreasing metabolic diversity with increasing salinity. Nonetheless, various microbial metabolisms have been found to occur at high salinity. Currently, information about microbial Fe metabolism in hypersaline environments is scarce. We studied Fe(II)-oxidizing and Fe(III)-reducing bacteria and archaea in Russian salt lake sediments using culture-dependent and -independent techniques. Our goals were to identify and quantify anaerobic Fe(II)-oxidizers and Fe(III)-reducers in the sediments and to analyze their distribution in a heterogeneous sediment profile.

Enrichment experiments showed that Fe(III)-reducers were actively growing even at 5 M salinity, while Fe(II)-oxidizers only remained active for several transfers at 0.5 M NaCl. Most probable number counts and quantitative PCR revealed that culturable Fe(II)-reducers and anaerobic Fe(II)-oxidizers represent < 0.1% of all bacteria present in the sediments. A 16S rRNA gene clone library was constructed from one of the sediments to study the bacterial and archaeal diversity. Currently, we analyze the abundance and distribution of different Fe-metabolizers in the sediments by qPCR. In summary, this study demonstrates that Fe(II)-oxidizers and Fe(III)-reducers are abundant and active in hypersaline environments. Combined with geochemical data this suggests the presence of an active Fe cycle even at high salinity.

**S9-02
Diversity of deep branches in the Fungal Kingdom revealed in hydrothermal ecosystems**

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The metaphor of 'coses of life' is widely used when talking about deep-sea hydrothermal ecosystems. Indeed, living organisms in hydrothermal systems seem to be concentrated near the active vents comprising the surrounding abyssal plains, where the biomass is more scattered. (Most of the diversity surveys focused on Bacteria, Archaea and Animals; and revealed numerous evolutionary hypotheses, the fungal kingdom was investigated at hydrothermal vents (Le Galvez et al., Appl Environ Microbiol, 2008 ; Bergaud et al., Environ Microbiol, 2009). Consistent with these hypotheses, phylogenies within Basidiomycota, Ascomycota and Chytridiomycota phyla were found. Interestingly, an old evolutionary lineage within Chytridiomycota was suggested to novel understanding of the diversification was suggested to explain the fungal diversity.

Using a fungi dedicated database, PHMYCO-DB (Le Galvez et al., Appl Environ Microbiol 2009), new primers were designed to focus on Chytridiomycota and deep branches within Opisthokonta. Preliminary results were obtained revealing a new Chytridiomycota diversity. More surprisingly, we highlighted the presence of Apusozoa (Bikonts) forming an interesting old group of living Eukaryota likely connected to the Opisthokonta. Further analyses are in progress.

**S9-03
Diversity, abundance, and potential activity of nitrifying and nitrate-reducing microbial assemblages in a subglacial ecosystem**

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Ice currently covers 11% of the terrestrial landmass and has covered significantly greater portions of the planet during Earth history. Subglacial microbial populations may impact global biogeochemical cycles on glacial-marginal tundra, however, nitrogen cycling in subglacial systems is poorly understood. Subglacial sediments sampled from beneath Robertson Glacier, Alberta, Canada harbor a diverse assemblage of potential nitrifiers, nitrate reducers, and diazotrophs, as assessed by *amoA*, *nifH*, and *nifH* gene biomarker diversity. Archaeal *amoA* genes were less abundant and less diverse than bacterial *amoA*. Nitrification and nitrate reduction were measured in microcosms using subglacial sediments incubated at 4°C, indicating the potential for these processes to occur *in situ*, beneath the glacier. Subglacial sediment porewaters and bulk meltwaters have low concentrations of dissolved inorganic and organic nitrogen compounds and a high C/N ratio of dissolved organic matter in sediment porewaters, indicating that the sediment communities are N limited. This may reflect the combined biological activities of organic N mineralization, nitrification, and nitrate reduction. Despite evidence for N limitation and detection of *nifH*, biological nitrogen fixation was not detected in subglacial sediment microcosm experiments. Collectively, our results suggest a role for nitrification and nitrate reduction in sustaining microbial communities in subglacial environments.

Oral Abstracts

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**S9-04
Existence and Expression, from a study on denitrification and methanogenesis in deep subsurface**

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We revealed a very active methanogenesis at 500 m deep ground water of sedimentary rock area in Horonobe, Hokkaido and candidate clones were confirmed by 16S rDNA. In addition, a sort of substrate shift was found from methanol to H₂/CO₂, which could be carried out by diversified archaeal populations. However, any active methanogenesis was observed for the ground water taken from 150 m in the same area, while molecular signature showed existence of candidate archaeal clones. Inactivation of related functional genes of archaea can be ascribable to the given environmental condition with relatively high ORP. Denitrification activity, on the other hand, measured by using stable isotope tracer experiment suggested related functional genes expression occurred under the given *in situ* condition. Thus, retrievable of methanogenesis archaeal clones may suggest their existence with active state in the past as genes remained, or their large migration from 500 m deep.

**S9-05
Microbial sulphur isotope fractionation in a Mars analogue environment at Rio Tinto, SW Spain**

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Sulphur isotopes may be a key tool for the detection of possible past life on Mars where abundant sulphate minerals are present. To investigate the link between sulphate reducing microorganisms and sulphur isotope fractionation, we incubated sediments from a modern hyper-acidic, Fe-rich environment at Rio Tinto, as a geochemical analogue of Mars, where iron-sulphate minerals, such as jarosite, may record the activity of sulphur metabolizing microorganisms.

Sediments were sampled from the upper part of Rio Tinto (Marismilla) and the estuary (Moguer). Laboratory incubations

were carried out at 30°C, using an artificial input solution with sulphate in excess [1]. Electron donors were provided by the natural substrate. Initial data indicate moderate biological sulphate reduction rates of between 5 and 90 mmol-m⁻²-h⁻¹ both in Marismilla and in Moguer, independent of pH of the input solution. Sulphur isotope fractionation was extreme in the Moguer estuary, extending beyond the maximum of 47‰ as predicted by the standard Rees model [2]. These data indicate that sulphur isotopes have a potential to be sensitive indicators of biotic activity on Martian sulphate minerals.

**S10-11
Impacts of spatial microbial heterogeneity on the fate of pesticides in soil and groundwater aquifers**

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Millions of tonnes of pesticides are used each year worldwide in agricultural production resulting in pollution of groundwater aquifers. There is, however, a striking contrast between the input levels (up to several kg per hectare) and the contaminant concentrations detected in groundwaters, which are normally in the microgram to nanogram per litre range. Recent research has revealed a large spatial variation in pesticide mineralisation potentials, but little is known about the scale at which these variations/heterogeneities affect the fate of contaminants. We analysed how mineralisation potentials of phenoxo acid herbicides (MCPA, 2,4-D) were spatially distributed in soil, subsoil, and groundwater aquifers using a 96-well microplate mineralisation assay. In the top soil, all samples showed rapid mineralisation following Monod mineralisation kinetics. In the subsoil sediments, a more heterogeneous distribution of mineralisation potentials was observed with fewer samples showing rapid mineralisation and more samples showing either slow 0-order mineralisation kinetics or no degradation. A heterogeneous distribution of herbicide mineralisation potentials was also observed in the groundwater sediment showing the most rapid mineralization close to the water table. The impacts of microbial heterogeneity on degradation and leaching of MCPA through the upper meter of subsurface sediment is evaluated applying a numerical model.

**S10-01
Isolation, characterisation and application of a 1,2-DCA degrading community from a solvent contaminated and acidic sandy aquifer**

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A 1,2-Dichloroethane (DCA) degrading mixed bacteria culture was enriched from the DCA contaminated Botany Sands aquifer in Sydney, Australia. Low pH 5.5 and chloroform limit natural attenuation in the aquifer. Here we isolated, characterised a DCA degrading culture and present evidence of enhanced DCA activity in a pilot scale bioaugmentation field trial. The mixed culture was cultivated anaerobically at pH 5.5 with 2.5 mM DCA and 2.2 mM ethanol. The culture can degrade up to 6 mM DCA at a rate of 370 μM/day at pH 5.5 and is not inhibited by 15 μM Chloroform. The pH/day at pH 5.5 and is not inhibited by the dehalogenating bacteria *Desulfotomaculum* was detected based on pyrosequencing of the SSU rRNA gene. Bioaugmentation of DCA in situ after two days and could reduce the concentration of DCA in situ after two days and remained viable at the site of inoculation for six months without addition of a carbon or energy source or nutrients. In conclusion, we have developed and demonstrated an active DCA degrading culture for application to acidic sandy aquifers. This is the first culture available in Australia for this purpose.