



**3<sup>rd</sup> D-A-CH Geobiology  
Symposium**  
for early career researchers  
30.09.2024 - 01.10.2024 Bayreuth, Germany

**Organized by:** Felix Pfaff, Franziska Zahn, Xiaoqiao Tang,

Myrhe Detiger, Dimitri Meier

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# Program

**Sunday, 29.09.2024**

|       |  |
|-------|--|
| 15:00 | <b>Bus ride to Fichtelberg</b>                           |
| 16:00 | <b>Guided tour of the Gleissinger Fels historic mine</b> |
| 18:00 | <b>Bus ride back to Bayreuth</b>                         |

**Monday, 30.09.2024**

|       |   |
|-------|---|
| 09:00 | <b>Early-career-event: Geological city tour of Bayreuth</b><br><b>Guide:</b> Dr. Florian Heidelbach (Bavarian Research Institute of Experimental Geochemistry and Geophysics)<br><b>Meeting point:</b> 09:00 AM, front door of Iwalewahaush, Wölfelstraße 2 |
| 10:00 | <i>Registration and welcome coffee</i>  |
| 10:45 | <b>Welcome to Geobiology 2024 - welcoming words by the organizing committee</b>   |

## Session I: Geobiology - The Past, the Deep, the Rock

|       |   |
|-------|---|
| 11:15 | <b>Manuel Reinhardt (University of Göttingen)</b><br>Traces of microbial diversity 3.42 billion years ago (Buck Reef Chert, Barberton greenstone belt)  |
| 11:30 | <b>Alisha Sharma (University of Jena)</b><br>Exploring the colonization dynamics and genetic potential of rock-attached microbes of carbonate aquifers  |
| 11:45 | <b>Aurèle Vuillemin (GFZ Potsdam)</b><br>Metagenomics of Lake Towuti's subsurface biosphere reveals adaptive features to redox fluctuations in ferruginous anoxia   |
| 12:00 | <b>Gaziza Konyssova (Wadden Sea Station Sylt, AWI Bremerhaven)</b><br>The Role of Wind, Tides, and Phytoplankton Dynamics in Seasonal Variability of Suspended Particulate Matter in a Sandy Tidal Basin                |
| 12:15 | <b>Eleanor Georgiadis (ETH Zürich)</b><br>Temperature control of CO <sub>2</sub> fluxes from fossil organic carbon oxidation in sedimentary rocks is mediated by microbial activity—insight from incubation experiments |

## **Monday, 30.09.2024 (continued)**

|       |                    |
|-------|--------------------|
| 12:30 | <i>Lunch break</i> |
|-------|--------------------|

### **Session II: Ecosystems & environmental change**

|       |  |
|-------|--|
| 13:30 | <b>Angela Cukusic (University of Vienna)</b><br>Groundwater microbial communities of two large cities, Berlin and Vienna, in comparison to a rural region, the River Mur valley, Austria                           |
| 13:45 | <b>Armando Alibrandi (GFZ Potsdam)</b><br>Insights into the microbial diversity of a high-temperature North Sea oil reservoir and the effects of anthropogenic disturbance   |
| 14:00 | <b>Anita Sanchez (TU Bergakademie Freiberg)</b><br>Hotspots of metal(loid) mobilization and water quality dynamics in a historic mine drainage system  |
| 14:15 | <b>Xingyu Liu (University of Bayreuth)</b><br>The export of groundwater-borne geogenic phosphorus from a drained wetland into surface water is governed by P-Fe interactions                                       |
| 14:30 | <b>Juan Carlos Trejos-Espeleta (LMU Munich)</b><br>Principal role of fungi in soil carbon stabilization during early pedogenesis in the high Arctic  |
| 14:45 | <b>Jarin Jose (MPI for Biogeochemistry Jena)</b><br>Divergent Pathways of Necromass Accumulation: Variations in Microbial Physiological Traits in Histosols and Mineral Soils Across Topsoil and Subsurface Layers |
| 15:00 | <i>Coffee break</i>  |
| 15:15 | <b>Poster session</b><br>Odd poster numbers: 15:15 - 16:15, Even poster numbers: 16:15 - 17:15   |
| 17:15 | <i>Short coffee break</i>  |
| 17:30 | <b>Round Table Discussion: Career advice and hot topics in GeoBio-sciences</b><br>Prof. William Orsi (LMU Munich), Prof. Johanna Pausch (Uni Bayreuth), Dr. Emil Ruff (MBL Woods Hole)                             |
| 19:30 | <b>Dinner</b>  |

## Tuesday, 01.10.2024

|       |  |
|-------|--|
| 09:00 | <i>Morning coffee</i>  |
| 09:30 | <b>Keynote: S. Emil Ruff (MBL, Woods Hole)</b><br><b>Is anoxia a hoax? Evidence for the widespread occurrence and cycling of “dark oxygen” in apparently anoxic environments</b> |
| 10:30 | <i>Coffee break</i>  |

### Session III: Organisms & processes

|       |   |
|-------|---|
| 11:00 | <b>Beatrix Heinze (University of Jena)</b><br>Microbial methane oxidation in groundwater ecosystems   |
| 11:15 | <b>Felix Pfaff (University of Bayreuth)</b><br>Promoting autotrophic denitrification in shallow porous aquifers for drinking water production   |
| 11:30 | <b>Mitali Chitnis (LMU Munich)</b><br>Tripartite symbiosis in a magnetic ciliate  |
| 11:45 | <b>Patrycja Janina Petrasz (University of Applied Sciences and Arts Neuchâtel)</b><br>Iron corrosion transformation triggered by utilization of <i>Meyerozyma</i> spp. dead biomass.            |
| 12:00 | <b>Eric Runge (University of Göttingen)</b><br>Framboid-like pyrite is a signature of microbial iron-cycling in hydrothermal sulfide systems  |
| 12:15 | <b>Nicole Geerlings (University of Vienna)</b><br>Using stable isotope probing and Raman microspectroscopy to determine single-cell activity in microorganisms with a high cytochrome c content |
| 12:30 | <i>Lunch break</i>  |
| 13:30 | <b>How was Geobiology 2024 and the future of the symposium?</b>   |
| 14:30 | <i>Farewell</i>   |



# Venue



## Iwalewahaus

Wölfelstraße 2

95444 Bayreuth

## The way to the symposium

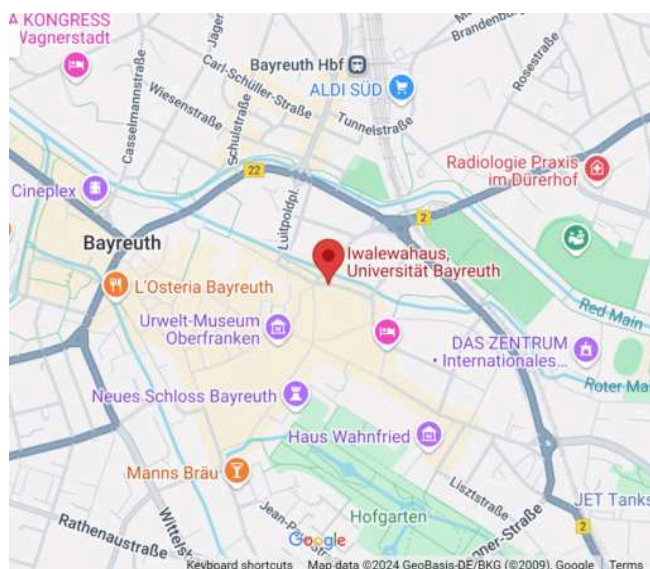
The best way to reach the Iwalewahaus is by bus or foot.

## From Bayreuth main station

VGN lines 302, 303, 305, 309 and 328 in the direction of ZOH, get off at the Luitpoldplatz stop.

## From ZOH

Line 304 in the direction of Birken, get off at the Opernhaus stop.



# Book of abstracts

## Keynote

01.10.2024 09:30 - 10:30

***Is anoxia a hoax? Evidence for the widespread occurrence and cycling of “dark oxygen” in apparently anoxic environments.***

*S. Emil Ruff*<sup>1</sup>,

<sup>1</sup> Marine Biological Laboratory, Woods Hole, USA

Emil Ruff will talk about the microbial production of molecular oxygen (O<sub>2</sub>) in the absence of light and its relevance in apparently anoxic environments. He and his collaborators found evidence of the in-situ production and consumption of O<sub>2</sub> in diverse ecosystems across large temporal and spatial scales. This “dark oxygen” could originate from the microbial dismutation of nitric oxide and/or chlorite, microbial processes that were so far not deemed to be important on a global scale. The discovery challenges our current understanding of the evolution, ecology, and biogeochemistry of the biogeosphere, suggesting aerobic niches in many environments that were thought to be anoxic.

# Short talk sessions

## *I Geobiology - The Past, the Deep, the Rock*

30.09.2024, 11:15-12:30

### **Traces of microbial diversity 3.42 billion years ago (Buck Reef Chert, Barberton greenstone belt)**

*Manuel Reinhardt*<sup>1</sup>, Volker Thiel<sup>1</sup>, Jan-Peter Duda<sup>1</sup>, Joachim Reitner<sup>1</sup>, Henrik Drake<sup>2</sup>

<sup>1</sup> Department of Geobiology, University of Göttingen, Göttingen, Germany

<sup>2</sup> Department of Biology and Environmental Science, Linnæus University, Kalmar, Sweden

Microbes inhabited Earth already in the Paleoproterozoic (3.6 – 3.2 Ga ago), but the structure and complexity of early ecosystems is still a matter of debate. Here we investigated carbonaceous cherts from the ca. 3.42-billion-year-old Buck Reef Chert (Barberton greenstone belt), deposited as primary silica precipitates in a back-arc basin. The carbonaceous matter (CM) is exceptionally well preserved, resulting from rapid silicification (Reinhardt et al., 2024). The bulk CM pool derives from phototrophic primary producers. Microscale stable carbon and multiple sulfur isotope analysis on the CM and associated pyrites, however, indicate the presence of additional microbial metabolisms in the ancient ecosystem, including sulfate reduction, methanotrophy or acetotrophy, and their metabolic counterparts (Reinhardt et al., 2024). Together, these signals underline the metabolic diversity in parts of the Paleoproterozoic Buck Reef Chert environment, and reflect an advanced biological carbon cycle during these early times.

#### References

Reinhardt, M., Thiel, V., Duda, J.-P., Hofmann, A., Bajnai, D., Goetz, W., Pack, A., Reitner, J., Schanofski, M., Schönig, J., Whitehouse, M.J., and Drake, H. (2024). Aspects of the biological carbon cycle in a ca. 3.42-billion-year-old marine ecosystem. *Precam. Res.*, 402, 107289. <https://doi.org/10.1016/j.precamres.2024.107289>

## Exploring the colonization dynamics and genetic potential of rock-attached microbes of carbonate aquifers

Alisha Sharma<sup>1</sup>, Martin Taubert<sup>1</sup>, Olga M Pérez Carrascal<sup>1</sup>, Robert Lehmann<sup>2</sup>, Thomas Ritschel<sup>2</sup>, Carl-Eric Wegner<sup>1</sup>, Kai Totsche<sup>2</sup>, Cassandre S Lazar<sup>3</sup>, Kirsten Kuesel<sup>1</sup>

<sup>1</sup> Department of Aquatic Geomicrobiology, Friedrich Schiller University Jena

<sup>2</sup> Department of Hydrogeology, Friedrich Schiller University Jena

<sup>3</sup> Research Chair in Aquatic Genomics, Tier II NSERC Canada

Most investigations in groundwater ecosystems targeted planktonic microbes which are easily obtained via groundwater sampling. In contrast, little is known about the diversity and functions of microbes adhering to solid rock surfaces, especially in limestone aquifers. To investigate microbial colonization dynamics to carbonate rock surfaces, we subjected rock chips from fractured limestone aquifers to a six-week incubation in flow-through bioreactors using groundwater from oxic and anoxic wells. In addition, crushed aquifer rock material was deployed for up to one year in groundwater wells in passive samplers. The attached microbiome was subjected to metagenomic analyses and compared to the planktonic community for taxonomic and functional differences. Groundwater bacteria rapidly colonized the rock chips within two days. Scanning electron microscopy analyses revealed extensive colonization on rock chips after 44 days, with chips from oxic bioreactors more densely colonized than from anoxic ones. Oxic bioreactors yielded higher attached-to-planktonic ratios ( $10^6:1$ ) than anoxic ( $10^3:1$ ) bioreactors. Metagenomics of rock-attached communities, yielding 605 metagenome-assembled genomes (MAGs), confirmed the presence of Proteobacterial taxa along with Bacteroidetes and Myxococcota. In contrast, the planktonic community mainly comprised *Patescibacteria* and *Nitrospirota*. Over 70% of attached MAGs exhibited genes for motility and biofilm formation, while these were found in only 15% of planktonic MAGs. Nutrient transporters, especially sulfur and iron transporters, were more prevalent in attached bacteria, facilitating efficient nutrient uptake from rocks and biofilms. The Calvin cycle was the preferred CO<sub>2</sub> fixation pathway for both attached and planktonic bacteria, with up to 10% of the aquifer microbiome harboring genes for complete CO<sub>2</sub> fixation. Moreover, nearly 90% of the attached community possessed functions for biofilm and organic matter degradation. These functions were only present in 28% of the planktonic community. In conclusion, attached and planktonic communities in carbonate rock aquifers featured high differences in both taxonomy and functions. The attached community is adapted to use potential electron donors from rocks for fueling CO<sub>2</sub> fixation, which likely contributes to biofilm formation, and lead to the high attached-to-planktonic ratios observed.



## Metagenomics of Lake Towuti's subsurface biosphere reveals adaptive features to redox fluctuations in ferruginous anoxia

Aurèle Vuillemin<sup>1</sup>, Fatima Ruiz-Blas<sup>1</sup>, Alexander Bartholomäus<sup>1</sup>, Sizhong Yang<sup>1</sup>, Dirk Wagner<sup>1</sup>, Cynthia Henny<sup>2</sup>, James M. Russell<sup>3</sup>, Jens Kallmeyer<sup>1</sup>

<sup>1</sup> Section Geomicrobiology, GFZ German Research Centre for Geosciences, Potsdam, Germany

<sup>2</sup> Research Center for Limnology and Water Resources, National Research and Innovation Agency (BRIN), Republic of Indonesia

<sup>3</sup> Department of Earth, Environmental, and Planetary Sciences, Brown University, United States

Ferruginous conditions prevailed through much of Earth's Ocean history. Yet biogeochemical cycles interpreted from the rock record remain elusive in terms of redox processes and organic matter remineralization, due to a scarcity of modern analogues in which to study an active subsurface biosphere. In Lake Towuti, Indonesia, ferruginous sediments are deposited under stratified conditions that mimic Earth's early oceans. Dynamic conditions at the lake floor supply microbial life with variable ferric and organic substrates, hence lacustrine sediments offer an ecological niche for investigating microbial metabolisms inherent to ferruginous anoxia.

Combining pore water geochemistry, microbial proxies with metagenome-assembled genomes (MAGs), we show that geochemical evolution of the sediment selects specific microorganisms during burial, mainly *Chloroflexota* and *Bathyarchaeia* upon depletion of reactive ferric iron and labile organic matter. Anaerolineae (*Chloroflexota*) hold metabolic potential to use unconventional electron acceptors, several cytochromes and redox metalloproteins to cope with oxygen fluctuations, thereby effectively colonizing the sediment-water interface. In sediments, Dehalococcoidia evolved to be acetogens, scavenging fatty acids, haloacids and aromatic acids as electron donors, bypassing specific steps in carbon assimilation pathways to perform secondary fermentations combined with CO<sub>2</sub> fixation via the Wood-Ljungdahl pathway. *Bathyarchaeia* prevailed below the sulfate reduction zone. Their MAGs encode sulfhydrogenase (*hydB*), with *dsrE* and rubredoxin as sulfur and electron carriers, and bifurcating-ferredoxin sulfide dehydrogenase (*sudA*) as electron acceptor. Along with heterodisulfide reductase (*hdr*) and electron bifurcation complexes (*Ech*, *Nuo*), these enzymes enable *Bathyarchaeia* to harness reactive sulfur species from hydrogen sulfide via polysulfides. Alongside carbohydrate and protein degradation, *Bathyarchaeia* exhibit a partial carbonyl- and complete methyl-branch Wood-Ljungdahl pathway used in homoacetogenic fermentation. Their capacity to fix CO<sub>2</sub> with molecular hydrogen and convert C<sub>1</sub>-C<sub>2</sub> compounds suggest close interactions with methylotrophs.

Such redox and fermentative processes constitute interwoven features, prominent in Lake Towuti's subsurface, that present analogies to primeval microbial life in ancient ferruginous systems.

## **The role of wind, tides, and phytoplankton dynamics in seasonal variability of suspended particulate matter in a sandy tidal basin**

*Gaziza Konyssova*<sup>1</sup>, Vera Sidorenko<sup>1</sup>, Sara Rubinetti<sup>2</sup>, Karen Wiltshire<sup>3</sup>, Justus van Beusekom<sup>4</sup>

<sup>1</sup> Wadden Sea Station Sylt, Alfred Wegener Institute Helmholtz Centre for Polar and Marine Research

<sup>2</sup> Wadden Sea Station Sylt, Alfred Wegener Institute Helmholtz Centre for Polar and Marine Research; Institute of Atmospheric Sciences and Climate CNR-ISAC

<sup>3</sup> Wadden Sea Station Sylt, Alfred Wegener Institute Helmholtz Centre for Polar and Marine Research; Climate Science Trinity College Dublin

<sup>4</sup> Institute for Carbon Cycles, Helmholtz Centre Hereon

This study investigates the seasonal and interannual dynamics of suspended particulate matter (SPM) in a sandy tidal basin, focusing on the influences of wind, tides, and biological processes. Using the Sylt Roads LTER data collected in 2000-2019, the research explores how these factors drive SPM variability on time scales ranging from hours to months. Preliminary findings indicate a pronounced seasonal cycle in SPM concentrations, with peaks during winter and lows in summer. Wind speed plays a significant role in SPM resuspension, particularly at shallow stations where changes are observed almost immediately. In contrast, at deeper stations, SPM dynamics exhibit a delayed response, which appears to be influenced by tidal propagation. Additionally, the spring phytoplankton bloom leads to increased chlorophyll-a levels, which cause flocculation of particulate matter. Such biological processes result in the sinking of flocculated material, contributing to the observed decrease in SPM starting in spring. The study highlights the complex interplay between physical and biological processes in coastal sediment dynamics, providing insights into the mechanisms driving SPM distribution in tidal inlets. Ongoing modeling will help clarify these processes and improve our understanding of the coastal physical-biological coupling.

# Temperature control of CO<sub>2</sub> fluxes from fossil organic carbon oxidation in sedimentary rocks is mediated by microbial activity — insight from incubation experiments

Eleanor Georgiadis<sup>1</sup>, Lena Bakker<sup>1</sup>, Jordon Hemingway<sup>1</sup>, Tim Eglinton<sup>1</sup>, Cara Magnabosco<sup>1</sup>, Robert Hilton<sup>2</sup>

<sup>1</sup> Department of Earth and Planetary Sciences, ETH Zurich

<sup>2</sup> Department of Earth Sciences, University of Oxford

In the geological carbon cycle, carbon can be stored in sedimentary rocks as petrogenic organic carbon (OC<sub>petro</sub>) on timescales of millions of years, before re-emerging to the surface via orogenesis and erosion. As the rock enters the critical zone of Earth's surface, a suite of physical, chemical and biotic processes occur during oxidative weathering and can result in CO<sub>2</sub> fluxes towards the atmosphere at a rate significant enough to influence Earth's climate on millennia timescales<sup>1,2</sup>. Whereas the chemically-mature OC<sub>petro</sub> contained in sedimentary rocks was classically perceived as a largely inert pool of carbon in the textbook view of the carbon cycle, the emerging theory is that the positive correlation between temperature and CO<sub>2</sub> fluxes from oxidative weathering of OC<sub>petro</sub> in sedimentary rocks constitutes a positive feedback to global warming<sup>1,2</sup>. To better understand the mechanisms involved in this CO<sub>2</sub> release, we incubated marly limestone and shale material from the Terre Noire region of the Prealps, France—a badland landscape known to display temperature-sensitive *in situ* CO<sub>2</sub> emissions<sup>1,3</sup>. Surface regolith (ca. 0–5 cm depth) and subsurface rock (ca. 5–10 cm) was retrieved from two catchments with different OC<sub>petro</sub> (0.45–0.78% wt.) and carbonate (30–45% wt.) contents, transferred to airtight bottles with a saline media and CO<sub>2</sub>-free headspace, and incubated at 4, 10, 16, 30 and 40 degrees Celsius. Half of the bottles were sterilised with mercuric chloride (HgCl<sub>2</sub>) prior to incubation. The gas phase was monitored weekly for four weeks and analysed for CO<sub>2</sub>(g) concentrations and stable isotopic (<sup>13</sup>C) composition. Early terminations of triplicate bottles allowed us to monitor changes in microbial biomass and community composition during the incubation experiment, using phospholipid fatty acids, and amplicon sequencing, respectively. Our data reveal that microorganisms accelerated the oxidation of OC<sub>petro</sub> at higher temperatures, particularly in the higher OC<sub>petro</sub>-containing material. This work demonstrates that the temperature sensitivity of CO<sub>2</sub> fluxes from sedimentary rocks likely operates predominantly via temperature control on microorganisms, bringing us one step closer to understanding the mechanics behind oxidative weathering in, and CO<sub>2</sub> fluxes from sedimentary rocks.

## References:

1. Soulet, G. et al. Temperature control on CO<sub>2</sub> emissions from the weathering of sedimentary rocks. *Nat. Geosci.* 14, 665–671 (2021).
2. Zondervan, J. R. et al. Rock organic carbon oxidation CO<sub>2</sub> release offsets silicate weathering sink. *Nature* 1–5 (2023) <https://doi.org/10.1038/s41586-023-06581-9>
3. Roylands, T. et al. Probing the exchange of CO<sub>2</sub> and O<sub>2</sub> in the shallow critical zone during weathering of marl and black shale. *Earth Surf. Dyn.* 12, 271–299 (2024)

## ***II Ecosystems & environmental change***

30.09.2024, 13:30-15:00

### **Groundwater microbial communities of two large cities, Berlin and Vienna, in comparison to a rural region, the River Mur valley, Austria**

*Angela Cukusic*<sup>1</sup>, Clemens Karwautz<sup>1</sup>, Constanze Englisch<sup>1</sup>, Eva Kaminsky<sup>2</sup>, Marielle Geppert<sup>3</sup>, Cornelia Steiner<sup>4</sup>, Christine Stumpp<sup>2</sup>, Jens Bölscher<sup>3</sup>, Christian Griebler<sup>1</sup>

<sup>1</sup> Department of Functional and Evolutionary Ecology, University of Vienna

<sup>2</sup> Department of Water, Atmosphere and Environment, Institute of Soil Physics and Rural Water Management, University of Natural Resources and Life Sciences Vienna

<sup>3</sup> Senate Department for Urban Mobility, Transport, Climate Action and the Environment, Berlin, Germany

<sup>4</sup> Department for Mineral Resources and Geoenergy, GeoSphere Austria

Increasing urbanization leads to groundwater warming and puts pressure on groundwater quality. However, the consequences for groundwater ecosystems, particularly from phenomena like the urban heat island effect and urban pollution, have been largely overlooked. The gap in urban groundwater ecology is particularly apparent when it comes to microorganisms, which are omnipresent in groundwater and intimately involved in essential ecosystem services such as cycling of carbon and nutrients, the (im)mobilization of metals, and the natural attenuation of organic contaminants.

Within the framework of the research projects 'Heat below the City' and 'CHARMANT', over 150 groundwater wells in Berlin and Vienna were sampled, respectively. Subsequent analyses encompassed a range of physical, chemical, and biological variables, including the characterization of microbial communities using 16S rDNA amplicon sequencing. Groundwater warming, along with other stressors, such as organic and inorganic pollution and low levels of dissolved oxygen, was hypothesized to prominently influence the composition, diversity, abundance and activity of the microbial communities. We hypothesized that factors unique to urban groundwater are important drivers of microbial communities, leading to the prevalence of similar dominant taxa in both cities, which would differentiate them from rural groundwater samples, i.e. groundwater from the River Mur valley in Styria, Austria. Moreover, urban groundwaters were expected to exhibit elevated diversity in comparison to the Mur Valley groundwater.

The results confirmed our hypotheses, revealing higher alpha diversity, activity, and abundance of prokaryotes in urban sampling sites. A complex interplay of hydrogeological and physico-chemical conditions with microbial community variables was recognized, i.e. the

hydrogeological aquifer type, as well as the individual combination of stressors in each city, played a crucial role, giving non-linear temperature-dependent trends in diversity between Berlin and Vienna. Furthermore, both urban and rural sites were strongly dominated by ultra-small Archaea and Bacteria. The community compositions based on phylum classification were similar at the different sites, regardless of temperature or other urban-specific factors. A closer look into predominant microbial functions and their dependence on the urban-rural gradient is planned in the future.

## **The export of groundwater-borne geogenic Phosphorus from a drained wetland into surface water is governed by P-Fe interactions**

*Xingyu Liu*<sup>1</sup>, Stefan Peiffer<sup>1</sup>

<sup>1</sup>Hydrology, University of Bayreuth

Groundwater-derived phosphorus (P) loading into surface waters stands as a prominent contributor to eutrophication. While numerous studies have addressed the spatial distribution of geogenic P, there have been few attempts to investigate its temporal dynamics and the transfer mechanisms from groundwater to drainage ditches, particularly in dynamic environmental settings. The present study, conducted in a drained riparian wetland in southeastern Germany, integrates hydrogeochemical monitoring of groundwater and drainage water over two years and vertical profile sampling within the monitoring wells. Elevated soluble reactive phosphorus (SRP) concentrations, reaching up to 16  $\mu\text{mol/L}$  or 0.5 mg/L, were observed in two of four groundwater wells, exhibiting no distinct seasonal pattern. Drainage water SRP varied from 6 to 15  $\mu\text{mol/L}$ , with some interannual variability due to dilution during wet periods. The spatial variability was suspected to be controlled by site-specific geochemical settings. The weathering of P-bearing minerals serves as the primary store of geogenic P in groundwater. Redox conditions and associated iron-cycling emerge as key factors controlling the release and mitigation of P. Under reducing conditions, microbially mediated reductive dissolution of iron oxides releases P, which was previously adsorbed, complexed, or coprecipitated in oxic groundwater. Upon export to the ditch, ferrous iron-rich groundwater undergoes rapid oxidation, leading to a significant decrease in SRP concentrations. Consequently, total reactive phosphorus (TRP), less readily available than its dissolved counterpart, becomes the dominant P species. This study provides novel insights into geogenic P enrichment in groundwater and its export through drainage systems. Eutrophication mitigation measures should increasingly focus on reducing geogenic P. Although concentrations may be relatively low compared to surface runoff or agricultural drainage, riparian groundwater containing geogenic P persists over extended periods, continuously contributing to surface water eutrophication. Our study offers valuable insights into the significance of geogenic-driven P transport into surface waters and underscores the influence of drainage systems in reducing P load into surface waters.



## **Principal role of fungi in soil carbon stabilization during early pedogenesis in the high Arctic**

*Juan Carlos Trejos-Espeleta*<sup>1</sup>

<sup>1</sup> Geomicrobiology, Ludwig-Maximilian-University Munich

Climate warming is causing widespread deglaciation and pioneer soil formation over glacial deposits. Melting glaciers expose rocky terrain and glacial till sediment that is relatively low in biomass, oligotrophic, and depleted in nutrients. Following initial colonization by microorganisms, glacial till sediments accumulate organic carbon and nutrients over time. However, the mechanisms driving soil nutrient stabilization during early pedogenesis after glacial retreat remain unclear. Here, we traced amino acid uptake by microorganisms in recently deglaciated high-Arctic soils and show that fungi play a critical role in the initial stabilization of the assimilated carbon. Pioneer basidiomycete yeasts were among the predominant taxa responsible for carbon assimilation, which were associated with overall high amino acid use efficiency and reduced respiration. In intermediate- and late-stage soils, lichenized ascomycete fungi were prevalent, but bacteria increasingly dominated amino acid assimilation, with substantially decreased fungal:bacterial amino acid assimilation ratios and increased respiration. Together, these findings demonstrate that fungi are important drivers of pedogenesis in high-Arctic ecosystems that are currently subject to widespread deglaciation from global warming.

## Hotspots of metal(loid) mobilization and water quality dynamics in a historic mine drainage system

Anita Sanchez<sup>1</sup>, Jackisch Conrad<sup>2</sup>, Hedrich Sabrina<sup>3</sup>, Lau Maximilian<sup>4</sup>

<sup>1</sup> Institute of Mineralogy, TU Bergakademie Freiberg

<sup>2</sup> Institute of Drilling Technology and Fluid Mining, TU Bergakademie Freiberg

<sup>3</sup> Institute of Biosciences, TU Bergakademie Freiberg

<sup>4</sup> Interdisciplinary Environmental Research Centre, TU Bergakademie Freiberg

Water quality degradation by decommissioned mining sites is an environmental issue recognized globally. In the Ore Mountains (Erzgebirge), a wide array of contaminants is released by old subsurface mining sites threatening the quality of receiving surface water bodies. While remediation has focused on treating contaminated mine water once it surfaces, the potential for controlling pollutant release within the mines have been largely overlooked. Here, we study temporal and spatial pollutant release dynamics in an abandoned but accessible mine (Reiche Zeche) draining directly to Elbe river tributaries. Using in-situ sensors and performing extensive biogeochemical monitoring over a 2-year period across and below former extraction horizons, we observe a prominent bulk hydrological response that varies across each sampling site and level as flow paths increase. Our results show dynamics in dissolved organic matter composition (DOM) and metal(loid) loads during different flow phases and across different depths. We can further conclude that there are hotspots of increased metal(loid) dissolution and hot moments of intensified metal(loid) discharge that can further complex metal(loid) mobilization mechanisms influenced by changes in DOM quality and flow path connectivity within the mine drainage system. The insights gained from this study offer valuable guidance for developing effective remediation and water management strategies for highly modified subsurface-surface water interfaces like mine outlets.

## **Insights into the microbial diversity of a high-temperature North Sea oil reservoir and the effects of anthropogenic disturbance**

*Armando Alibrandi*<sup>1</sup>, Plewka Julia<sup>2</sup>, DiPrimio Rolando<sup>3</sup>, Bartholomäus Alexander<sup>1</sup>, Probst Alexander J.<sup>2</sup>, Kallmeyer Jens<sup>1</sup>

<sup>1</sup> Geomicrobiology, GeoForschungsZentrum Potsdam

<sup>2</sup> University of Duisburg-Essen

<sup>3</sup> AkerBP

Microorganisms in petroleum reservoirs have to endure extreme conditions like high pressure, temperature and salinity as well as isolation from other ecosystems. Still, these communities are metabolically active and play a key role in the biodegradation and souring of the reservoirs and hence have a great effect on the quality of the oil. During exploitation, these reservoirs face significant anthropogenic changes due to the introduction of electron acceptors, donors, and introduction of foreign microorganisms. Most studies on the microbial ecology of oil reservoirs were carried out on samples from reservoirs that have already been exposed to anthropogenic disturbances for long periods of time, especially flooding i.e., the injection of fluids like seawater and production waters to maintain reservoir pressure. This study explores microbial community structure in the North Sea's Edvard Grieg oil reservoir using 16S rRNA sequencing and metagenomics. Fluid injection followed by water breakthrough, i.e. when the injected fluid reaches the extraction wells, had only started recently, allowing for a detailed look at its effects on the microbial communities of the reservoir. The reservoir's microbial community composition displays regional variations, low taxonomic diversity, and an abundance of thermophilic, anaerobic, and halotolerant taxa. After the fluid breakthrough, the community structure changed, marked by a substantial increase in specific groups like sulfate-reducing bacteria. Communities within the different parts of the reservoir homogenized due to fluid injection but remained distinctly different from the fluid's community

## **Divergent pathways of necromass accumulation: variations in microbial physiological traits in histosols and mineral soils across topsoil and subsurface layers**

Jarin Jose<sup>1</sup>, Qing-Fang Bi<sup>1</sup>, Marion Schrumpf<sup>1</sup>

<sup>1</sup> Biogeochemical Processes, Max Planck Institute for Biogeochemistry

Recent research highlights the pivotal role of microorganisms in carbon stabilization through the formation of microbial necromass, necessitating a deeper understanding of necromass dynamics within biogeochemical cycles. The BioNeCS project seeks to elucidate the processes underlying necromass accumulation by analyzing variations in microbial physiological traits, including carbon use efficiency (CUE), microbial growth, specific growth rate (qGrowth), and turnover rates. Grasslands with varying soil types—high organic matter soils (Histosols) and mineral soils—were studied, focusing on topsoils (0-10 cm) and subsurface layers (20-30 cm). The research employed stable isotope oxygen-18 (<sup>18</sup>O) to measure these traits in a controlled laboratory setting. Additionally, the impact of different management practices and key drivers of microbial activity were assessed. Initial results reveal that Histosols exhibit higher CUE compared to mineral soils, with topsoils showing higher CUE than subsurface soils. This increased CUE is associated with greater microbial growth and lower biomass specific respiration, suggesting a dominance of anabolic over catabolic processes. In contrast, mineral soils display lower CUE and reduced microbial growth. However, despite the reduced overall growth, the qGrowth, representing the specific growth rate relative to existing microbial biomass, was higher in mineral soils, indicating a more rapid turnover of microbial biomass. This rapid turnover, driven by the energy demands of accelerated growth, contributes to higher metabolic rates, thereby reducing CUE. Interestingly, necromass accumulation was found to be higher in mineral soils than in Histosols, largely due to their higher growth efficiency. A deeper look into these processes indicates that microorganisms may follow different pathways in these environments, with a possible selective preservation of necromass in mineral soils. These preliminary findings are small steps towards understanding the intricate relationships between soil type, microbial physiology, and necromass dynamics, enhancing our understanding of stable organic carbon.

## **III Organisms & processes**

### **Microbial methane oxidation in groundwater ecosystems**

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Groundwater holds most of the world's terrestrial freshwater and a substantial amount of its prokaryotic biomass. Trace gasses such as methane can threaten groundwater quality and, if released into the atmosphere, contribute to climate change. Many studies showed the genetic potential of groundwater microbes to oxidize methane, but little is known about the amounts removed by microbial activity. Using an ultra-low level <sup>14</sup>C-labeling approach, we quantified in situ microbial methane oxidation rates in carbonate and sand aquifers from central and northern Germany. While rates in the carbonate system ranged from  $0.002 \pm 0.0004$  to  $0.02 \pm 0.0007 \mu\text{g CH}_4 \text{ L}^{-1} \cdot \text{d}^{-1}$ , rates in the sand aquifers reached as high as  $116.7 \pm 69.6 \mu\text{g CH}_4 \text{ L}^{-1} \cdot \text{d}^{-1}$ . With abundant electron acceptors present, groundwater methane concentration appeared to be the main rate limiting factor. Independent from the rates, most samples showed fast methane turnover times of <2 years, indicating rapid methane consumption by groundwater microbes. During the 24 h incubation, <10% of the labeled methane was incorporated into microbial biomass and most was converted to CO<sub>2</sub>, suggesting its use as an energy rather than carbon source for the groundwater microbiome. Quantitative PCR and 16S sequencing revealed high abundances of genes for methane oxidation (*pmoA*, *mmoX*, *mcrA*) at high-rate sites with *Crenothrix*, *Candidatus Methanoperedens* and *Candidatus Methylomirabilis* as key methane oxidizers. Our results indicate microbial methane removal in groundwater to be as efficient as in some marine systems or lakes, where microbial oxidation is estimated to remove >90% of all methane before it reaches the atmosphere.

## Promoting autotrophic denitrification in shallow porous aquifers for drinking water production

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Nitrate pollution in groundwater is a global challenge since groundwater serves as a vital drinking water source across Europe. Avoiding a nitrate overload requires sustainable management schemes for shallow aquifers with high groundwater recharge. However, their attenuation potential against pollutants like nitrate is often limited. High nitrate levels persist in many aquifers even long after reducing nitrogen inputs. Especially for otherwise oligotrophic shallow groundwater systems, innovative and cost-effective technologies to eliminate nitrate loading and to ensure a sustainable supply of drinking and utility water are urgently needed to meet rising water demands.

In this project we will implement a novel approach for nitrate removal from shallow groundwater. Via the injection of reduced gaseous electron donors ( $H_2$  &  $CH_4$ ), we aim to stimulate autotrophic denitrification directly upstream of burdened drinking water wells. This new project goes beyond existing nitrate reduction methods for drinking and utility water, which mostly involve post-treatment of pumped groundwater with reverse osmosis, anion exchange, electro dialysis, or water treatment in bioreactors. Furthermore, stimulating denitrification with inorganic electron donors has the advantage of avoiding an enrichment of heterotrophic, potentially pathogenic microorganisms.

In lab scale microcosms, we have simulated a range of *in-situ* conditions and aquifer materials. We identified temperature as a key driver for denitrification rates. A temperature rise of 10 °C induced a significant increase in denitrification kinetics, with complete denitrification being observable at 12 °C and above. Microbial communities shifted towards *Rhodocyclaceae* and *Hydrogenophaga*.

Enrichment cultures of autotrophic denitrifying microorganisms for a potential augmentation and stimulation of denitrification inside natural aquifers were successfully introduced to a sediment matrix in a bioreactor. We inoculated non-denitrifying aquifer material with enriched stable denitrifying communities and successfully reestablished denitrification in the presence of hydrogen and methane. In a currently running mesoscale pilot plant experiment we are testing the principles of gas injection into porous media and monitor the stimulation



effects over time. In a final stage, we hope to successfully implement our approach at the field scale to eliminate nitrate in an actual drinking water production system.

We aim to offer proof that injecting  $H_2/CH_4$  can effectively eliminate nitrate from groundwater aquifers without adversely impacting water quality or hydraulic characteristics through secondary reactions, like mineral dissolution and precipitation. Harnessing the intrinsic potential of autotrophic microorganisms native to groundwater systems and using gasses that can be produced with green biotechnology offers a new and sustainable solution to a notorious problem.

## Tripartite symbiosis in a magnetic ciliate

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Magnetotactic bacteria (MTB) use intracellular structures called magnetosomes as built-in compasses to navigate along Earth's magnetic field. Magnetosomes are composed of biomineralized magnetic iron minerals and guide MTB toward optimal redox conditions along dissolved O<sub>2</sub> gradients<sup>1</sup>. While magneto-sensing has been well studied in MTB, recent research indicates that this ability extends to microbial eukaryotes (protists), as well. Indeed, certain protists exhibit magnetoreception, potentially through mechanisms like predation upon MTB, symbiosis with MTB, and even the direct biomineralization of magnetosomes<sup>2</sup>. Here we report the discovery of a novel magnetic protist with endosymbiotic MTB and methanogens. Through a combination of light and electron microscopy, we observed that this holobiont is responsive to magnetic fields and possesses bullet-shaped magnetosome crystals. 18S rRNA gene analysis revealed that the protist is a ciliate from the novel family Tropicotractidae and transcriptomes show that it expressed an Fe-Fe hydrogenosomal hydrogenase gene - indicating an anaerobic lifestyle and H<sub>2</sub> production through fermentative hydrogenosomes. Transcriptomes of the magnetic protist revealed expression of genes involved in magnetosome formation (*mamA*), sulfate reduction (*dsrB*), which are closely associated with magnetotactic sulfate-reducing bacteria (*Desulfovibrionaceae*). Expression of the methyl-coenzyme M gene (*mcrA*) involved in methanogenesis was detected from the magnetic protists, that were derived from a group of endosymbiotic H<sub>2</sub>-oxidizing methanogenic archaea (*Methanoregula*). These results indicate that a bacterial endosymbiont likely produces the magnetosomes inside of the ciliate, and reduces sulfate using molecular hydrogen produced by hydrogenosome-like organelles of the eukaryote host. A second archaeal symbiont also appears to use the host derived hydrogenosomal H<sub>2</sub> for methanogenesis. Overall, these findings expand the diversity of magnetotactic microbes to include anaerobic ciliates, suggesting that magnetotaxis in eukaryotes may be wider distributed than previously thought.

### References:

<sup>1</sup>Uebe & Schüler 2016 *Nature Reviews Microbiology*

<sup>2</sup>Monteil & Lefevre 2020 *Trends in Microbiology*

## **Iron corrosion transformation triggered by utilization of *Meyerozyma* spp. dead biomass.**

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Archeological iron artifacts retrieved from salt-contaminated sites frequently undergo active corrosion after exposure to the atmospheric environment. This process is mainly driven by low humidity, high oxygen, and presence of chloride ions (Salwyn 2004; Reuger *et al.* 2009) that triggers formation of unstable corrosion compounds. A particularly challenging corrosion product is akaganeite, its tunnel-like structure prevents complete removal of chloride ions leading to loss of iron structural integrity and causing further degradation.

To address this challenge, physicochemical abilities of extremophilic yeast *Meyerozyma guillermondii* HEK2 are investigated in the framework of the Horizon Europe project GoGreen “Green strategies to conserve the Past Present, and Future of Cultural heritage”. Preliminary studies performed on dead biomass demonstrated unique properties of this extremophilic yeast to interact with corroded iron under aerobic conditions in two distinct ways. First mechanism involves biosorption of iron and chloride ions onto the cell wall, while the second triggers a redox reaction that results in conversion of active corrosion products into more thermodynamically stable compounds. The utilization of X-ray diffraction (XRD) analysis carried out before and after treatment, confirmed this latter.

This peculiar redox reaction observed between dead biomass *Meyerozyma* spp. and corroded iron may be associated with active agents present on the outer layer of the cell wall. Therefore, to investigate the mechanism involved in this transformation, ongoing studies focus on the dynamics of the electrochemical system as well as characteristics of the biomass surface, with emphasis on functional groups activity. The point of zero charge measurements of *Myeyerozyma* spp. provided insights into the surface reactivity across different pH and by that allowed a better understanding of potential electrostatic interactions with corrosion products on artificially aged iron mock-ups. Furthermore, these results combined with Fourier Transform Infrared spectroscopy (FTIR) analyses helped identification of functional groups present on the cell wall surface and their activity during corrosion conversion shedding light on the mechanism involved in it.

## **Framboid-like pyrite is a signature of microbial iron-cycling in hydrothermal sulfide systems**

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Pyrite [FeS<sub>2</sub>] is ubiquitous in sulfidic marine sediments and sedimentary rocks, including hydrothermal sulfide deposits. Below ~120°C, its formation is controlled by interactions of sulfur- and iron-cycling microorganisms, which are abundant and active in hydrothermal sulfide systems, making pyrite a promising target for microbial biosignatures in these environments. The most recognized examples of pyrite-encoded biosignatures are pyrite framboids, spherical aggregates of unidimensional pyrite microcrystals, which are chiefly attributed to the (past) activity of sulfate-reducing bacteria. However, experimental evidence has demonstrated abiotic pathways for their formation. Moreover, pyrite formation in pure and enrichment cultures including sulfate-reducing, sulfur disproportionating, and Fe(III)-reducing microorganisms repeatedly yielded “framboid-like” spheroids, which differ from framboids in their non-aggregational texture. This demonstrates that the role of microorganisms in controlling pyrite morphology is poorly understood. Here, we demonstrate the formation of pyrite with various distinct morphologies, including framboid-like spheroids, via hydrothermal sulfidation of abiogenic and biogenic (i.e., organic matter-associated) magnetite nanoparticles, derived from Fe(III)-reducing bacteria (*Geobacter sulfurreducens*). The products from our incubation experiments (sulfide/Fe=4, 80°C, pH 7.5) in the presence of different sulfur species (sulfide, elemental sulfur, polysulfides) were characterized with analytical imaging techniques, mineralogical methods, and geochemical approaches (SEM-EDS, FIB,  $\mu$ -XRD, Raman spectroscopy, sequential Fe extraction). We suggest the variability in pyrite morphologies resulted from the modulation of pyritization rates by the interrelated effects between organic matter and elemental sulfur (crystalline or colloidal). Notably, framboid-like pyrite was exclusively produced from the sulfidation of biogenic magnetite. Our results demonstrate that framboid-like pyrite can result from the abiotic sulfidation of biogenic iron minerals, supplied by iron-cycling microorganisms. Thus, organic matter associated with precursor iron minerals may be a more important driver of framboid-like pyrite formation than microbial sulfide supply.

## Using stable isotope probing and Raman microspectroscopy to determine single-cell activity in microorganisms with a high cytochrome content

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Measurements of metabolic activities at the single-cell level help us to understand how microorganisms in complex microbial communities interact with and shape their environment. Only at a single-cell resolution, key questions on the distribution of cellular activities in spatially structured communities (e.g., in biofilms) and on metabolic heterogeneity within microbial populations can be properly addressed. Additionally, single-cell metabolic analyses greatly facilitate physiological studies of uncultured microbes and of organisms that are difficult to culture. To this end, stable isotope probing combined with nanoscale secondary ion mass spectrometry (nanoSIMS) or with Raman microspectroscopy has proven to be highly useful.

Here, we present a novel application of stable isotope probing combined with Raman microspectroscopy for assessing single-cell activities of prokaryotes containing elevated amounts of cytochrome c. This protein is an essential part of the electron transport chain in many organisms and shows particularly high expression levels in microbes such as diazotrophs, nitrifiers, anaerobic ammonium oxidizers, and sulfide oxidizers. Raman spectra of cytochrome c exhibit a rich pattern of bands from wavenumbers 600 to 1700  $\text{cm}^{-1}$ , arising from vibrational modes of the heme chromophores. So far, these distinct vibrational peaks in Raman spectra have not been used to assess single cell assimilation rates. Here, we show that the incorporation of isotopically labeled substrates ( $^{13}\text{C}$ ,  $^{15}\text{N}$ ) in the hemes can be readily observed via Raman microspectroscopy. We investigate the potential to quantify single-cell growth rates and compare this novel method to other Raman-based approaches to determine single-cell activity.

# Poster session

## 1. The Rise of Algae promoted eukaryote predation in the Neoproterozoic benthos

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In modern ecosystems, predatory single-celled eukaryotes (protists) accelerate organic carbon remineralization, nutrient cycling, and fixed carbon transfer to higher trophic levels. Geochemical and paleontological evidence suggest that the ecology of predatory eukaryotes was stimulated by the Rise of Algae in the Neoproterozoic Era (1000-539 million years ago), when the relative abundance of eukaryote primary producers increased and anoxic bottom waters overlaid most of the global seafloor. The relationships, however, between increasing algal matter and the activity of anaerobic benthic protozoa in the Neoproterozoic remain unconstrained by modern observations. Here, we demonstrate that the introduction of algal particulate matter (APM) to marine sediments underlying low-oxygen bottom waters significantly stimulates protistan – and decreases archaeal and bacterial – gene expression under anoxia. Under these conditions, protists significantly overexpressed key genes involved in anaerobic energy metabolism and phagocytosis while remineralizing APM to CO<sub>2</sub>. Taxonomically, APM significantly increased the relative abundance of 18S rRNA from predatory protists with known Proterozoic origins, including the Amoebozoa, Cercozoa, and Foraminifera. Moreover, APM stimulated the growth of benthic foraminifera under anoxia, and resulted in net growth efficiencies higher than those predicted by existing models, potentially explained by the over expression of a eukaryotic dissimilatory nitrite reductase. Overall, our findings suggest that the export of marine algae to the seafloor during the Rise of Algae would have stimulated the growth and activity of benthic predatory protists under anoxia, thereby creating more modern food webs by enhancing the transfer of fixed carbon and energy to higher trophic levels, including the earliest animals.



## 2. Potential formation of marine snow driven by primary production under Precambrian conditions

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Drivers behind Earth's oxygenation pattern and climatic perturbations over the Precambrian, 4-0.5 billion years ago, are still poorly constrained. Crucial factors were likely primary production, as well as the ancient biological carbon pump (BCP). Nowadays, the BCP is important for climate due to the ocean's uptake and storage of CO<sub>2</sub>. Thus, it also likely regulated major transitions through Earth's history. However, despite its importance, a comprehensive model detailing the evolution of the BCP and the factors influencing its efficiency remains elusive. Here, my aim is to constrain conditions driving marine snow formation, a crucial aspect of the BCP, under Precambrian conditions. I simulated primary production by oxygenic and anoxygenic phototrophs under ferruginous Archean vs. sulfidic Proterozoic conditions. Specifically, *Rhodovulum iodolum*, capable of both Fe(II)- and H<sub>2</sub>S-driven photosynthesis, and the cyanobacterium *Synechococcus* sp. PCC7367 were grown in seawater medium supplemented with various dissolved silica concentrations, and with either H<sub>2</sub>S or FeCl<sub>2</sub> as electron donors over eight weeks. To monitor oxidation rates of the electron donors, I used colorimetric H<sub>2</sub>S and iron assays. Using pH sensors and O<sub>2</sub> optodes, I determined O<sub>2</sub> production and pH modulation. Additionally, the growth rates were analyzed via (bacterio-) chlorophyll signals. In parallel, I monitored the formation of aggregates, and assessed morphology (size and sinking velocity). The analysis showed that aggregates analogous to marine snow formed in all incubations with high silicate concentration indicating that silica polymers drive initial coagulation. Degree and type of aggregation was dependent on the addition of H<sub>2</sub>S or Fe(II). The lower density and different morphology of aggregates forming in abiotic controls suggest that biotic processes including biofilm formation, played a crucial role. Hence, with the changing biogeochemistry of the Precambrian oceans, marine snow formation would have also changed and, thus, these alterations may be related to the major climate shifts in Earth's history.

### 3. Branched GDGT lipid based soil fertility proxy: Development and implication

Anirban Kumar Mandal<sup>1</sup>, Franz Kerschhofer<sup>1</sup>, Cindy de Jonge<sup>1</sup>

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Branched glycerol dialkyl glycerol tetraethers (brGDGTs) are ubiquitous cell membrane-spanning lipid molecules, predominantly synthesized by Acidobacteria. In soils, these bacteria adjust the composition of their core membrane lipids in response to temperature by altering the relative abundance of methyl branches in the central alkyl chain. On a global scale, a relative increase in brGDGT Ia and a decrease in brGDGT IIa and IIIa form the basis of the paleotemperature proxy MBT<sup>5</sup>Me. However, recent findings suggest that this soil paleotemperature proxy can be influenced by soil pH and chemistry. Specifically, studies have shown that increases in exchangeable calcium and soil pH increase the concentration of 6-methyl brGDGT compounds and other cyclopentane-containing brGDGT compounds (De Jonge et al., 2021). Initial research along different elevation gradients (De Jonge et al., 2024) has proposed the development of separate proxies for exchangeable bases in soils, such as exchangeable Ca<sup>2+</sup> and Σbases, based on concentrations of specific cyclopentane-containing brGDGTs (Ib, IIa', IIb'). The same study found that the distribution of isoprenoid GDGTs (specifically, the crenarchaeol isomer), a second group of GDGT compounds produced by archaea, correlates with exchangeable iron. The development of a proxy to reconstruct past soil calcium and iron availability, fills a gap in our knowledge on past soils. On a Holocene timescale, soil chemistry parameters (exchangeable cations, calcium, free acidity), or so-called soil mineral fertilities, are expected to vary over time due to changes in land use and climatic conditions. These variations in soil mineral fertility significantly impact soil nutrient status, influencing shifts in vegetation and soil organic carbon (von Fromm et al., 2021). However, reconstructing historical changes in soil fertility directly from soil archives is challenging due to erosion or burial effects, which alter the chemical signature of soil particles over time. Based on the influences of soil chemistry parameters on brGDGTs, we propose the development of a new brGDGT-based proxy to trace soil mineral fertility in modern and past soils, and reconstruct changes in soil organic carbon over time. We present novel data from surface soils from central Africa, and discuss the apparent dependencies on soil chemistry.

#### **4. Orbitrap HRMS lipidomics to develop a biomarker proxy for diazotrophs (nitrogen fixers) in soils**

Franz Kerschhofer<sup>1</sup>, Anirban Mandal<sup>1</sup>, Cindy De Jonge<sup>1</sup>

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In soils, a large fraction of atmospheric CO<sub>2</sub> is stored as fixed carbon (organic compounds). However, the future development of this carbon sink is unclear, as soil fertility potentially limits the amount of primary production (carbon fixation) and belowground carbon storage. Nitrogen clearly has the potential to influence soil carbon storage by limiting primary productivity (N deficiency in an infertile soil) and the amount of mineral-associated organic matter (MAOM), but a better understanding of its variability on longer timescales is needed. However, there is currently no method to reconstruct soil nitrogen availability through time.

As a first step, a biomarker proxy for the presence of nitrogen fixers in soils will be developed. Therefore, the lipidome of cultures of nitrogen fixing heterocyst cyanobacteria (*Anabaena* sp., *Nostoc* sp., *Stigonema ocellatum*, *Fischerella* sp.) and environmental samples with these and non-heterocyst cyanobacteria will be determined. Using high-resolution mass spectrometry (orbitrap), the bacteria's cell membrane lipids will be targeted in respective of the environmental gradients of nitrogen availability. This lipidome will be compared with the results from traditional methods, i.e. GC-FID, GC-MS and HPLC-MS. Eventually, we aim to find these lipids in downcore soil profiles, to reconstruct their presence through time.

At this event, I will be presenting the general research idea and the lipid distribution of selected cultures based on GC-FID and GC-MS.

## **5. Alpine microbiome under pressure: Investigating soil community dynamics in a warming world**

*Laureen Ahlers<sup>1</sup>, Ianina Altshuler*

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The alpine ecosystem is highly vulnerable to climate change, yet the effects of warming on high-altitude soils remain largely unexplored. Particularly, it is unknown how soil microbial communities will adapt to these rapid environmental changes. This project aims to investigate the impact of warming on the microbial communities of European alpine soils and their role in greenhouse gas fluxes, specifically methane (CH<sub>4</sub>), carbon dioxide (CO<sub>2</sub>), and nitrous oxide (N<sub>2</sub>O). To achieve this, we use warming chambers (open top chambers, OTC) at 3 high-altitude sites across the Alps in permafrost-affected soils. Our study will provide crucial insights into microbial adaptation and its implications for greenhouse gas emissions in alpine ecosystems.

## 6. The Drivers of High Arctic Tundra Soil Microbial Communities: A Biogeography Study in Light of Arctic Greening

Lena Bakker<sup>1</sup>, Moritz Mainka<sup>1</sup>, Annina Meier<sup>1</sup>, Jana Rütters<sup>1</sup>, Dario Barilla<sup>1</sup>, Jamila Gisler<sup>1</sup>, Elias Meier<sup>1</sup>, Simone Fior<sup>1</sup>, Aline Frossard<sup>2</sup>, Marijn Van de Broek<sup>2</sup>, Kristine Bakke Westergaard<sup>3</sup>, Jake Alexander<sup>3</sup>, Sebastian Doetterl<sup>3</sup>, Cara Magnabosco<sup>3</sup>

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The Arctic has been warming at an unprecedented rate. In response, Arctic plants are experiencing prolonged growth seasons and higher productivity. On the global scale, the plant response to these changes is commonly referred to as “Arctic Greening” but above ground changes are not homogeneous. While soil microorganisms are known to play an important role in many ecosystems, their connections to the above ground vegetation in the greening Arctic is largely unknown. To evaluate the role that microorganisms play in Arctic Greening, we sampled and analyzed different sites on Svalbard. Thirty-one topsoil samples from the three site types were sequenced for 16S rRNA and a quantitative PCR of 16S rRNA and ITS was performed. This study shows that high Arctic tundra ecosystems have specific impacts on the soil microbial communities. Anthropogenic disturbance leads to a microbial community with more cells, that is lower in diversity and that has a very steep distance decay of community similarity. This is due to a few dominant groups such as *Clostridia* and *Bacteroidia* and the phylum of *Desulfobacterota* profiting from high nutrient input and outcompeting other community members. Soils surrounding bird cliffs have a higher abundance of fungi over prokaryotes, a significantly more diverse soil community and very slow distance decay. This is likely due to less selective pressures and more dispersing effects caused by the spread of bird droppings, the large water flow in the beginning of the growing season and potentially by fungi. Important biological above ground associations with community composition are the graminoids that are strong drivers of greening observed in sites (ii) and (iii) and the dwarf shrubs that were dominant in (i). Below ground parameters affected by climate change such as the soil moisture and deep soil temperature are also associated with the communities, while signs of exogenous input such as total nitrogen, total organic carbon and manganese correlated as well. Preliminary data on microbe abundances in a geological sequence will also be presented. A more comprehensive understanding of the biogeographical processes that influence these microbes is crucial for clarifying their role in Arctic greening and predicting how this will evolve with climate change.

## 7. Seasonal drought reduces microbial diversity and functional richness in the Amazon

Jessica Finck<sup>1</sup>, Dan Frederik Lange<sup>1</sup>, Beto Quesada<sup>2</sup>, Bruno Takeshi<sup>2</sup>, Savio Ferreira<sup>2</sup>, Fernando Dini Andreote<sup>3</sup>, Erika Kothe<sup>4</sup>, Gerd Gleixner<sup>1</sup>

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Tropical rainforests such as the Amazon are of high importance as a global carbon sink. Due to its well-known nutrient limitation, the Amazon rainforest relies heavily on rapid microbial decomposition of biomass to release freshly available nutrients for plant growth. Despite the fundamental importance of decomposers for this ecosystem, little is known about the biodiversity of such microbiomes, their functional activity, and spatial and seasonal variability. We used 16S rDNA and ITS rDNA sequencing to analyze the microbial communities of the Amazon's terra firme and the much drier white-sand ecosystems during the dry and wet seasons in 2022. Bacterial microbiomes differed significantly between seasons, displaying lower bacterial species richness and diversity in response to seasonal drought. In contrast, fungal richness and diversity differed strongly between sites, but were less affected by seasonal variation, suggesting their hyphae network and associations with plants as potential protectors against drought effects. Fungal and bacterial communities alike showed lower abundance of taxa involved in organic matter decomposition following seasonal drought. These changes were also reflected at the functional level, with samples collected during the dry season and at white-sand sites featuring lower abundances of decomposition and denitrification pathways. Soil hydro-chemical data also emphasizes how prolonged drought may limit soil nutrient supply via local microbiomes. Our results suggest that the reduced nutrient availability and soil connectivity during drought and within the white-sand ecosystem lower microbial activity and functional redundancy, henceforth demonstrating a strong impact of ecosystem type and drought on tropical microbiomes and their functional capacities. Our results further highlight that the observed increase in droughts in the Amazon rainforest may additionally limit nutrient supply through the microbial community, limiting carbon sequestration in the ecosystem with negative consequences for the global climate system.

## **8. Physico-chemical analysis of a Small Mountainous Tropical River in Costa Rica: the Reventazon-Parismina Basin**

*Ana Vasquez<sup>1</sup>, Nur Sakinah Abdul Razak<sup>2</sup>, Shouye Yang, Zhiwei He*

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Chemical weathering plays a key role in the global geochemical cycle by the alteration of the mineral/rock structure via hydrolysis, oxidation, or carbonation reactions. Here we present the trace elemental and water parameters in the dissolved pool in the Reventazon-Parismina basin sampled from Tapanti to the estuary. This study aims to decipher the natural and anthropogenic sources in the catchment's geochemical pool, its controlling factors, and discuss the implications for chemical weathering and anthropogenic impact in one of the major basins in Costa Rica.

## **9. Emerging contaminants, Perfluoroalkyl and polyfluoroalkyl — PFAS under changing climatic variability: Insights from a preliminary survey in the Niger Delta Region, Nigeria.**

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Emerging contaminants, including poly-fluoroalkyl substances — PFAS, remain among the greatest threats to freshwater ecosystems worldwide. Despite the increasing recognition of their effects on freshwater integrity, monitoring these contaminants in Africa, including Nigeria, remains scarce. We provide a preliminary insight into the presence and distribution of PFAS under varying climatic conditions in Ogba River, Edo State, Nigeria. We sampled 10 Perfluorinated Carboxylic Acids (PFCAs), 5 Perfluorosulfonic Acids (PFSA), 5 Fluorotelomers, 5 Perfluoroalkyl Sulfonamides (FASAs), and 3 Perfluoroalkyl Sulfonyl Amide Acetates in eight sites across the dry, wet and harmattan seasons between 2019 and 2023. Sites were also selected to reflect varying land uses, including agriculture, informal settlements and industry. Results showed that all analyzed PFAs were detected in all sites. The concentrations of PFAS detected ranged from 0.05–2.21 µg/L across the sites and seasons. Seasons were more influential in shaping the distribution of PFAS in the study area than sites. The informal settlement has higher concentrations (2.22 µg/L) of PFAS than agriculture. The highest concentration of PFAS was Perfluorohexanesulfonic Acid (PFCAs—1.99 µg/L), followed by Perfluorohexane Sulfonic Acid (PFHxA) (0.30 µg/L), and Perfluorooctanesulfonic Acid (PFOS) (0.22 µg/L). The findings of this study provided baseline data and can contribute towards developing an effective regional strategy for managing PFAS influence in freshwater ecosystems in Nigeria.



## **10. Assessment of urban contaminants degradation in soil-plant systems by multi-element compound-specific stable isotope analysis (ME-CSIA)**

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Benzothiazole (BT) and mercaptobenzothiazole (MBT) are common organic pollutants due to extensive application in industrial and consumer products, resulting in widespread contamination in the environment. However, knowledge on the putative degradation and degradation pathways of these pollutants in soil-plant systems is limited. In this study, we investigate the biodegradation of BT and MBT in soil-plant systems using three different plants during hydroponic and classical soil cultivation, applying multi-element compound-specific isotope analysis (ME-CSIA) for biodegradation verification. ME-CSIA can provide qualitative and quantitative information on biodegradation patterns while avoiding the limitations of traditional purely concentration-based methods. The hydroponic experiment using wheat has been completed. Wheat exposed to MBT (MBT-ST) of 50 mg/L produced lower biomass of the aboveground parts than non-treated control wheat (PC); in addition, MBT-ST roots turned red and dark. The results indicate that the applied MBT concentrations are toxic to wheat. In contrast, the biomass of the aboveground parts of wheat exposed to BT (BT-ST) of 50 mg/L was slightly higher than the biomass of the PC, but the plants exhibit certain stress responses, such as a lush main root system with shorter, darker secondary roots. These findings suggest that while BT induces some level of stress in plants, it may not be as toxic as MBT. Additionally, evapotranspiration data indirectly support these observations.

## 11. Elevated nitrate concentrations in soil solution under pure Douglas fir stands can be lowered by mixing with European beech and by site selection

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Forest management aims for productive and stable forests that continuously provide ecosystem services, including balancing nutrient fluxes. Increasing heat and frequent droughts in temperate European forests make the introduction of non-native Douglas fir (*Pseudotsuga menziesii* (Mirb.) Franco) an increasingly relevant climate change adaptation strategy, particularly as an admixture to native tree species, such as European beech (*Fagus sylvatica* [L.]). Douglas fir can alter biogeochemical processes in forest soils, potentially leading to an excess of nitrogen in the ecosystem, but the biotic and abiotic controls of this effect need further examination. Here we studied nitrate leaching on plots of two contrasting textures (loamy and sandy site) planted with either pure Douglas fir, pure Norway spruce (*Picea abies* [L.] Karst.), pure European beech or a mixture of beech with either of the conifers. We collected soil solution from two depths over two years to estimate nitrate leaching risks. We found highest concentrations of nitrate in soil solution in lower soil layers under Douglas fir (29.14 mg/L). Comparisons of concentrations below the litter layer and below the main rooting zone indicate an accumulation and microbial production of nitrate under Douglas fir. In Douglas fir-beech mixed stands, however, we found significantly lower nitrate concentrations below the main rooting zone, implying a mixture effect. Furthermore, this effect was pronounced on sites with finer soil texture and a lower C:N ratio. Elevated nitrate concentrations were not the result of differential throughfall or litter inputs and were associated with a P-limitation found in the tree foliage, likely causing reduced nitrate uptake. Further ongoing experiments show that nitrate leaching is associated with 1) a lower tree nitrate uptake and 2) distinct microbial nitrifier community presence and activity. We conclude that increased nitrate concentrations under Douglas fir stands may pose a relatively higher nitrate leaching risk than Norway spruce and a considerably higher risk compared to beech stands. However, the low susceptibility to leaching under beech stands seems to be a strong effect trait in mixtures, diminishing the high leaching potential Douglas fir induces on some sites. Low N leaching potential is key for adequate forest nutrition and groundwater protection.

## **12. Intrinsic potential and activity of nitrate turnover examined for different hydrogeological aquifer settings**

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Nitrate is one of the most serious contaminants in groundwater, frequently deteriorating water quality and its use as drinking water. However, nitrate can, at specific environmental conditions, be removed within the subsurface environment via natural biogeochemical processes, such as denitrification and dissimilatory nitrate reduction to ammonium (DNRA). Nitrate reduction mostly depends on two key factors, i.e. the prevalence of hypoxic or anoxic conditions and the availability of a suitable electron donor, be it organic or inorganic. For a long time, research concentrated mainly on the nitrate attenuation potential and activity in groundwater, ignoring the aquifers' sediment matrix. We hypothesize that the sedimentary deposits host the major potential for nitrate reduction, carrying the majority of microorganisms as well as different sources of electron donors. It is further assumed that the diverse hydrogeological settings harbor different sources and of varying amounts of electron donors. Microbial communities harboring the metabolic potential for nitrate reduction are believed to be ubiquitously present. At locations where physicochemical conditions favor nitrate reduction, we may expect microbes involved in these processes to be more abundant and highly active, leading to a fast nitrate removal from groundwater. In repeated field campaigns, fresh sediments and groundwater were collected by drilling from shallow aquifers different in geological background and physico-chemical conditions. Subsequently, flow-through sediment column and batch experiments were carried out in a temperature-controlled climate room at 12°C, to mimic ambient groundwater temperature. Our goal is to understand the overall potential and in situ activity of nitrate removal in different shallow aquifers related to their nitrate load, redox conditions, hydrogeology, and microbial community characteristics.

### **13. Can Microbes Inform about the Vulnerability of Spring Waters — Assessment of Microbial Communities and Water Quality in Austrian Springs**

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Springs are vital water sources, and their vulnerability to environmental changes, particularly climate change, is a growing concern. The PhD project focuses on the assessment of microbial communities and water quality patterns in Austrian springs, with an emphasis on understanding the vulnerability of spring waters.

The first aim of the study is to characterize springs based on their spatial and temporal dynamics in terms of physical and chemical characteristics. This involves a detailed analysis of various parameters such as temperature, pH, conductivity, nutrients, and the concentration of major ions. These characteristics provide valuable insights into their geological background and hydrological conditions, as well as their catchment areas.

A key aspect of the project is the phylogenetic and functional composition of microbial communities in spring waters. Microbes are integral components of aquatic ecosystems and can serve as sensitive indicators of environmental conditions including changes. By comparing the physical-chemical characteristics with microbial communities' members, we aim to explore the potential of individual microbes as environmental tracers.

The research comprises two parts: a local to regional study in the province of Styria, where three springs are sampled monthly, and a national study sampling 86 springs across Austria in two campaigns, one in the spring season and one in autumn, the hydrological extremes.

In summary, this research seeks to enhance our understanding of the vulnerability of spring waters to anthropogenic pressures such as climate change. The findings will provide a most needed knowledge base for future water resources management and contribute to the sustainable use of these vital resources.

## 14. Mineralization of <sup>13</sup>C-labeled microbial biomass by thermophilic microorganisms from aquifer sediments

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High-Temperature Aquifer Thermal Energy Storage (HT-ATES) is a low-carbon energy system with high energy storage capacity based on injection and extraction of heated and cooled water in shallow aquifers. However, the heat injection ( $\geq 45^{\circ}\text{C}$ ) alters aquifer microbial community composition potentially affecting important microbial-mediated ecosystem services. Previous studies have shown that thermophilic endospore-forming bacteria rapidly develop and dominate the microbial community under high temperature. Therefore, understanding the structure, biodiversity and physiology of thermophilic endospore-forming microorganisms in aquifer sediments, especially their capacity to purify groundwater is crucial to evaluate potential environmental risks associated with HT-ATES. In this study, we tested whether accumulated <sup>13</sup>C-labeled microbial biomass from natural groundwater can be a carbon source for thermophiles naturally occurring in an aquifer sediment; both groundwater and sediment were sampled from a shallow sandy aquifer in Wittstock/Dosse (Brandenburg, Germany). We investigated the effects of three temperatures (60, 70, and 80°C) on the capacity of the aquifer community to mineralize the <sup>13</sup>C-labeled biomass in anaerobically incubated sediments. Increased biomass mineralization rates were only observed in two replicates at 60°C and one replicate at 70°C among 6 replicates. At 80°C, no mineralization was observed within 72 days. Sequencing of 16S rRNA genes revealed that the bacterial diversity and richness were significantly higher at 60°C than at other incubation temperatures. Original sediment samples shared relatively similar microbial communities as those in the microcosms incubated at 80°C, with most samples being dominated by Proteobacteria. However, in some cultures at 60°C and 70°C, phylotypes affiliated with spore-forming Firmicutes further developed. Members of *Thermaerobacteria* and *Symbiobacteriia* were dominant in the two active replicates at 60°C, whereas the members of *Thermoanaerobacteria* were significantly abundant in the only active culture at 70°C. Our results suggest that some thermophilic endospore-forming bacteria grew at 60°C and 70°C, using microbial biomass generated from the natural environment as carbon source, while 80°C may be too high for the thermophilic microbial community to grow, which indicates fatal conditions for the microorganisms in aquifer sediments at this temperature.

## 15. Microbial diversity in a saline siliciclastic aquifer at the ATES exploration site Berlin-Adlershof

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Microbial processes such as biofilm formation (clogging) and mineral precipitation (scaling) can affect the effectiveness of aquifer thermal energy storages (ATES). They can reduce the permeability of potential reservoirs and compromise the efficiency of ATES facilities. In addition, microbial processes can release toxic trace elements such as arsenic through iron mineral dissolution in the subsurface. To evaluate the microbial impact on the performance of ATES, it is crucial to identify in situ metabolic processes and microbial key players.

At the ATES exploration site Berlin-Adlershof, we monitored the microbial abundance, community composition and metabolic functions for 2 years after drilling in a Jurassic sandstone aquifer at ~225 m depth. We applied culture-dependent and -independent approaches such as enrichment cultures, amplicon sequencing, metagenomics and -transcriptomics in the context of the groundwater hydrochemical conditions.

The aquifer was characterized by an in-situ temperature of 17 °C, Na and Cl dominated fluid (TDS ~20 g L<sup>-1</sup>) and organic substrates including acetate. The microbial community was adapted to saline and alkaline conditions. Over time, the community shifted from mainly fermenting bacteria, capable of hydrogen and organic acid production, to a syntrophic community of fermenting and sulfate reducing bacteria, with the latter consuming the fermentation products. These processes involve the risk of corrosion, but also offer the possibility to dissolve mineral scales.

Results of this study in combination with a percolation experiment analyzing biotic versus abiotic processes at different temperature conditions, will help to develop prediction tools for potential system operational failures and appropriate countermeasures in ATES.

## 16. Unraveling the temporal variability and functional resilience of groundwater microbiomes

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Understanding microbial community responses to natural disturbances is crucial for maintaining healthy groundwater ecosystems, particularly with increasing weather extremes. This 10-year study investigates groundwater microbiomes across various permeable aquifers, focusing on their stability and metabolic potential. We analyzed groundwater bacterial 16S rRNA gene sequences in relation to hydrological changes, seepage microbiomes, groundwater metabolomes, and DOM compositions. Our findings reveal sinusoidal oscillations in groundwater microbiomes in response to hydrological changes, with distinct temporal variations linked to aquifer permeability and assembly processes. Microbiomes in highly permeable aquifers, dominated by stochastic processes, exhibited pronounced short-term variability and sensitivity to environmental changes. In contrast, microbiomes in low-permeability aquifers, governed by deterministic processes, showed greater resistance and temporal stability, albeit with ongoing changes due to environmental shifts. These patterns serve as representative models for karst aquifers and hydrologically isolated groundwater. Despite low compositional stability, the metabolic potential of groundwater microbiomes was highly resilient, indicating adaptability to disturbances. However, seepage-derived inputs into shallow or recharge area groundwaters could alter microbiome compositions and impact groundwater quality. These insights highlight the need for continuous monitoring and inform strategies for sustainable groundwater management in changing environments.

## 17. Comparative analysis of two eutrophic lake sediments treated with a CaO<sub>2</sub>-based lake restoration agent.

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Eutrophication has become a global problem, mainly caused by anthropogenic influences and climate change. Eutrophication describes the drastic shift of an aquatic ecosystem from a healthy and diverse macrophyte-dominated to a phytoplankton-dominated state characterized by algal blooms and decreasing biodiversity. Consequently, elevated water turbidity and oxygen depletion cause aquatic plants and animals to be extinct. Therefore, the water body loses its ecological function and becomes unusable for recreational and commercial purposes. Especially small and shallow lakes are highly vulnerable to eutrophication, making it a contemporary problem for many cities and towns.

Lake restoration approaches are numerous but, in many cases, expensive and ecologically destructive. A relatively cheap and gentle method is the application of calcium peroxide (CaO<sub>2</sub>), which slowly releases oxygen for eight weeks when dissociating in water. In theory, this should promote aerobic microbial degradation of organic matter. Additionally, released calcium ions bind bio-available phosphates, eliminating them from the system. However, the mechanisms of CaO<sub>2</sub> as a lake restoration agent are still poorly understood.

In this experiment, the impact of CaO<sub>2</sub> on two eutrophic lake sediments is analyzed via microcosm incubation under both aerobic and anaerobic conditions. The experiment was performed with a commercially available CaO<sub>2</sub>-containing lake restoration agent over an incubation time of approx. 13 weeks. The release of CO<sub>2</sub> and CH<sub>4</sub>, as well as O<sub>2</sub> consumption in the microcosms, were measured using gas chromatography. The concentration of relevant nutrients (NH<sub>4</sub><sup>+</sup>, NO<sub>x</sub><sup>-</sup>, SO<sub>4</sub><sup>2-</sup>, Fe(II) and PO<sub>4</sub><sup>2-</sup>) was determined with colorimetric assays. Surprisingly, the two sediments responded distinctly to the CaO<sub>2</sub>-containing lake restoration agent. While one sediment responded with increased CO<sub>2</sub> emission, the other had lower CO<sub>2</sub> production rates, indicating changes in microbial activity. Furthermore, 16S rRNA gene amplicon sequencing data is currently being analyzed to identify enriched members of the prokaryotic community driven by CaO<sub>2</sub> application.



## 18. Arsenic exposure renders oxygenic phototrophs altered in growth physiology

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Arsenic is known for its detrimental toxic effects in all domains of life. Since being concentrated to appreciable levels billions of years ago, almost every organism seems to possess processes and genes for detoxification. However, the notion of a potentially beneficial semi-metal has arisen in the past decades, whether it be fueling anoxygenic photosynthesis or as a terminal electron acceptor in microbial respiration. Rather recently, mechanisms for the production of complex lipid-soluble arsenic analogs have been described that are solely produced by phototrophic microbes. Their functions remain elusive and biosynthetic mechanisms have been barely explored, though a potential physiological purpose might be suggested. In a quantitative approach to growth physiology, differential proteomics, lipidomics, and *in-situ* physicochemistry, we are seeking to address the multi-functional implications of arsenic incorporation into complex biomass to enlighten a potential purpose that enriches an arsenic-dependent microbial evolution and sheds light on the fate of arsenic in geochemical cycles. Prerequisite physiological results indicate an improved growth behavior of the model cyanobacterium *Synechocystis* sp. PCC6803 supplied with considerable arsenate (As(V)) concentrations (3 mM).

## 19. *Methanosphaerula subterraneus* sp. nov., a hydrogenotrophic methanogen isolated from lacustrine subsurface sediment exposed to high CO<sub>2</sub> concentrations

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In the Eger Rift area in the Czech Republic, constant mantle CO<sub>2</sub> degassing since roughly one million years has led to adaptation of subsurface microbial communities to high CO<sub>2</sub> conditions. The Hartoušov Mofette Field (HMF) in the Eger Rift is characterized by an extremely high flux of geogenic CO<sub>2</sub>, reaching up to 50 kg/m<sup>2</sup> per day. A drilling campaign by the International Continental Scientific Drilling Program (ICDP) recovered a drill core reaching 230 m below the surface from the HMF. Using drill core samples from 50 m below the surface, we enriched a novel strain, first using minimum mineral medium, followed by isolation in strain-specific medium. Cells were regular cocci with 0.5-0.8 µm diameter. Optimal growth was observed at 20~33 °C and slightly acidic pH (5.9) with a doubling time about 3 days. We detected optimal growth at sodium concentrations between 11 mM and 150 mM. H<sub>2</sub>/CO<sub>2</sub> and formate can be used as substrates but not acetate, methanol, ethanol and trimethylamine. However, acetate, methanol and ethanol can boost growth with H<sub>2</sub>/CO<sub>2</sub>. Growth was hampered under formate concentration above 40 mM. Phylogenetic analysis of the 16S rRNA gene and conserved archaeal genes showed that the new strain fulfills the criteria of being a novel species of the genus *Methanosphaerula*. The name *Methanosphaerula subterraneus* sp. nov. is proposed. Strain EG is the type strain of *Methanosphaerula subterraneus*.

## **20. Tracing the influence of minor hydrocarbon seepage on sulfur cycling in marine subsurface sediments**

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All hydrocarbon (HC) reservoirs tend to leak to some extent, releasing small quantities of HCs that migrate upwards into the overlying sediments, e.g. seeps. Through microbial activity, these HCs can be completely metabolized before reaching the seafloor, thus not creating any surface manifestations. Despite their inconspicuous nature, these HC fluxes can potentially influence the geochemistry of surrounding sediments and the composition and activity of microbial populations therein, as they add electron donors into the system.

We analyzed 50 gravity cores from the South Western Barents Sea. The sampling sites were located in three areas overlying known HC reservoirs (HC sites) and two reference areas (reference sites) of pristine seabed not affected by HC leakage. Despite the very similar nature of their sediment composition (clay-rich, organic-poor), the 50 gravity cores revealed considerable variability in pore water concentration gradients of various dissolved ions. Nearly linear profiles of pore water sulfate and alkalinity were observed, indicating that there is minimal to no net production or consumption of these ions within the sediment. Still, low rates of sulfate reduction ( $\text{pmol} \times \text{cm}^3 \times \text{d}^{-1}$ ) were measured and modeled, primarily at HC sites. Transcriptomic analysis of functional marker genes provided further evidence of enhanced metabolic activity by sulfate-reducing bacteria and methanogenic/methanotrophic archaea at HC sites.

Our findings demonstrate that inconspicuous HC seepage plays a significant role in sedimentary biogeochemical cycles by shaping pore water concentration gradients, influencing sulfate reduction rates, and altering the microbial community composition and activity in marine subsurface sediments. Therefore, sediment geochemistry combined with omics potentially constitutes a non-invasive tool for HC exploration.

## 21. Microbiology and geochemistry of the deep-sea Kairei plume: 16S rRNA taxonomy and functional prediction of microbial communities

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Hydrothermal activity is crucial for ocean biogeochemistry and global heat fluxes<sup>1,2</sup>. The Kairei Hydrothermal Field (KHF) represents a unique site along the Central Indian Ocean Ridge (CIR), globally ranked as the third most hydrogen-rich field<sup>3,4</sup>. In addition, previous studies highlighted high concentrations of Fe in end-member fluids<sup>4-6</sup>. Despite the extensive studies made on end-member fluid composition and sediments as well as the general interest in the particular chemistry of the site, the microbial activity in the plumes of the KHF are as yet poorly studied. Only a single diversity study has been published<sup>7</sup>, limited to few samples and a specific area around KHF. These authors highlight a difference among putative vent structures generating northern and southern plume at Kairei, without a clear explanation for this observation. In our study we analyze the plume microbial community systematically along different transects. 16S rRNA gene tag sequencing of the V3-V4 hypervariable region has been performed. Moreover, we carried out enrichment studies on-board, in order to understand the effect of iron on the microbial communities of this particular vent field. Molecular data will be coupled with geo-chemical and oceanographic data in a multidisciplinary analysis framework, to better understand the role of interactions between trace metals and microbes along the hydrothermal plume for their fate and distribution.

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## 22. Bioavailability of the organic carbon associated to biogenic iron (oxyhydr)oxides (BIOS) for transformation by fermentative microbes

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Biogenic iron (oxyhydr)oxides (BIOS) are iron–organic matter (OM) coprecipitates that can be found in diverse terrestrial and aquatic environments, such as acid mine drainage, wetlands, and river systems<sup>1,2</sup>. These iron minerals may store organic carbon (OC) effectively and act as a “rusty sink” through sorption and coprecipitation of OC, e.g. in sediments, where  $21.5 \pm 8.6$  % of the total OC are associated with iron<sup>2,3</sup>. The morphology of BIOS is determined by the microorganisms that play a role in their formation and affects its properties<sup>2</sup>. Thus, also the bioavailability of the OC associated with BIOS is influenced and determines its properties as a carbon sink. The complexity of the OM associated with BIOS prevents it from being used as electron donor by Fe(III)-reducers, which can effectively utilize the BIOS-Fe(III) as electron acceptor. However, in suboxic and anoxic environments, fermenters are known to play an important role in the degradation of OM and could potentially use the BIOS-associated OM. Given the importance of BIOS as a carbon sink, we will test the ability of a consortium of fermenting microorganisms enriched from Lake Constance to degrade BIOS-associated OC. Preliminary tests show that the Fe(III)-reducing bacteria *Shewanella oneidensis* MR-1 and *Geobacter sulfurreducens* are not able to use the BIOS associated OC as sole electron donor to fully reduce the Fe(III), while they can reduce the Fe(III) to an extent of 100 % in BIOS from *Rhodopseudomonas palustris* TIE-1, strain BoFeN1, and enrichment culture KS with an additional carbon source. Therefore, we are enriching a consortium of fermenting microorganisms from sediments of Lake Constance that afterwards will be amended with BIOS from different sources to test the bioavailability of the BIOS-associated OC.

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### 23. Magnetite facilitates interspecies electron transfer in tidal sediments

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Magnetite has been shown to act as a recyclable electron donor and acceptor (*i.e.* biogeochemical battery) for Fe-metabolizing bacteria capable of direct interspecies electron transfer (DIET), depending upon ambient environmental conditions. However, the environmental prevalence and biogeochemical implications of this phenomenon have not been explored yet.

Here, we present an environmental example of DIET in magnetite particles present in intertidal sediments from the coast of Normandy (France). Microbial Fe(III) reduction was demonstrated via environmental magnetite incubations, where Fe(II) was immediately released in anoxic conditions. Spiking anodes of sediment microbial fuel cells with colonized magnetite boosted the performance demonstrating the microbial electrogenic activity. 16S rRNA gene amplicon sequencing and fluorescence *in-situ* hybridization showed substantially more abundant populations of anaerobic, exoelectrogenic *Desulfuromonadaceae* and *Desulfobulbales* and aerobic, electro-autotrophic *Tenderiales* on isolated magnetite vs. sand particles. The genomic repertoire of metagenome-assembled genomes confirmed the electroactive potential of both functional groups. Further, candidate electrotrophy including *Ca. Tenderia* sp. were enriched from coastal sediments on synthetic magnetite spiked on cathodes, only in a closed electric circuit.

The coexistence of syntrophic bacterial populations with contrasting oxygen demands on single magnetite particles is most plausibly explained by tidal oxygen fluctuations. Here, magnetite may transiently accumulate reductive power during low tide (anoxia) fueling oxygen respiration and coupled CO<sub>2</sub> fixation during high tide (oxia).

Marine magnetite occurs in virtually all marine coastal sediments. Based on our findings, magnetite particles have the potential to be a widespread substrate and evolutionary hotspot for DIET, and thus play a larger environmental role than previously considered.

## **24. Insights from a four year in-situ incubation experiment at the Indian Ridge: How Microbes Impact Seafloor Massive Sulfide Weathering**

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Seafloor massive sulfide (SMS) deposits form on the modern ocean seafloor at active hydrothermal vent systems through mixing of mineral-rich, hydrothermal fluids with ambient oxygenated seawater. SMS deposits are primarily composed of pyrite and chalcopyrite, and serve as a source for valuable metals such as copper. Once hydrothermal activity ceases these SMS deposits undergo abiotic oxidative weathering when penetrated by oxygenated seawater.

Microbial activity considerably accelerates this process transforming sulfide minerals, enhancing metal transport and mineral dissolution. Under certain circumstances the formation of iron-rich silica caps can create low oxygen conditions below the caprock, shielding SMS deposits from both abiotic and biotic oxidative weathering, likely extending the lifetime of these deposits.

Here we will examine the impact of microbial activity for SMS transformation and dissolution of minerals under oxic and low oxygen conditions. For this we incubated sulfide minerals for four years on the seafloor at an active and an inactive venting site on the Indian Ridge and used hydrothermal vent material for laboratory experiments. We employed a combination of molecular biological techniques, microbial enrichment experiments, physiological studies, microscopy, and geochemistry, to identify key microbial actors responsible for mineral transformation and metal mobilization. SEM images reveal diverse mineral structures which are indicative of the presence of various microorganisms involved in Fe-cycling, including twisted stalks and nanowires. By determining turnover rates and mineral modifications, we aim to predict how microbial activity influences the lifetime of SMS deposits under different oxygen conditions.

## 25. Lithium bioextraction from geothermal brines

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To achieve the European objective of climate neutrality by 2050, the evolution of the energy and transport sectors is key. This will, however, lead to changes in the current demand for raw materials. Europe has created a list of critical raw materials (CRM) whose demand is set to rise significantly, far exceeding current resources (Carrara et al. 2023). Among them lithium (Li) and rare earths will soon become more important than oil and gas. Some geothermal brines contain significant quantities of these CRM, and the CRM-geothermal project has been funded to explore their extraction coupled to the exploitation of geothermal heat. Within the project, we are exploring a biological extraction method using a naturally occurring mechanism: the oxalate-carbonate pathway (Cailleau et al. 2011). In the OCP, two microbial metabolisms, fungal oxalogenesis and bacterial oxalotrophy, allow to mobilize and immobilize minerals, enabling the separation and specific precipitation of Li.

The aim of this study is to adapt this natural mechanism to an industrial biotechnological process for Li bioextraction. Besides Li geothermal brines contain many ion species in higher concentrations, so an initial purification step is necessary. For this, the properties of oxalate produced by fungi can be used to precipitate most of these impurities, without precipitating Li. In a second step, oxalate can be converted to carbonate by oxalotrophic bacteria, and this carbonate combined to a pH increase can be used to precipitate Li-carbonates.

The key to an economically viable bioextraction process is to optimise the production and consumption of oxalate by the microorganisms. To select the best combination of oxalic acid producers and production conditions, we compared fungal species against different carbon sources and pH. To set production objectives, the quantities required for a complete precipitation are expected to be  $3.57 \text{ g}_{\text{C}_2\text{O}_4}/\text{g}_{\text{impurities}}$  and  $4.29 \text{ g}_{\text{CO}_3}/\text{g}_{\text{Li}}$ .

### References:

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## 26. Anti-phage defense supports persistence of an anoxygenic phototrophic *Chlorobi* bloom

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Green sulfur bacteria (class Chlorobia) are anaerobic, anoxygenic phototrophs that are known to form extremely high cell density and persistent blooms in sulfidic, anoxic aquatic ecosystems. In Trunk River, a brackish coastal lagoon in Falmouth, MA, blooms of GSB occur seasonally and persist for weeks to months. These blooms are greatly dominated by a near-clonal population of a new species-level lineage within the genus *Prosthecochloris*. Despite the persistence of this population over long periods of time phage infection seems to be absent or greatly attenuated. In this ongoing work, we use meta 'omics approaches to investigate the viral ecology of these phototrophic blooms. Though we find abundant viral contigs linked to a diversity of hosts, the paucity of *Chlorobia* host-linked viral contigs indicates that few phages infect *Chlorobia*-affiliated hosts. An enrichment of viral defense related genes in *Chlorobia* population genomes suggests that these organisms invest heavily in countering phage infection. We also find putative *Prosthecochloris* sp. affiliated plasmids associated with viral defense, perhaps indicating the horizontal transfer of defense genes among the population. Future work will investigate the abundance of viral particles in the bloom using epifluorescence microscopy and use proteomic data to quantify infection and defense activity.

## 28. Microbial communities and functions are structured by vertical geochemical zones in a northern peatland

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Northern peatlands are important carbon pools; however, differences in the structure and function of microbiomes inhabiting contrasting geochemical zones within these peatlands have rarely been emphasized. Using 16S rRNA gene sequencing, metagenomic profiling, and detailed geochemical analyses, we investigated the taxonomic composition and genetic potential across various geochemical zones of a typical northern peatland profile in the Changbai Mountains region (Northeastern China). Specifically, we focused on elucidating the turnover of organic carbon, sulfur (S), nitrogen (N), and methane (CH<sub>4</sub>). Three geochemical zones were identified and characterized according to porewater and solid-phase analyses: the redox interface (<10 cm), shallow peat (10–100 cm), and deep peat (>100 cm). The redox interface and upper shallow peat demonstrated a high availability of labile carbon, which decreased toward deeper peat. In deep peat, anaerobic respiration and methanogenesis were likely constrained by thermodynamics, rather than solely driven by available carbon, as the acetate concentrations reached 90 μmol·L<sup>-1</sup>. Both the microbial community composition and metabolic potentials were significantly different ( $p < 0.05$ ) among the redox interface, shallow peat, and deep peat. The redox interface demonstrated a close interaction between N, S, and CH<sub>4</sub> cycling, mainly driven by *Thermodesulfovibrionia*, *Bradyrhizobium*, and *Syntrophorhabdia* metagenome-assembled genomes (MAGs). The archaeal Bathyarchaeia were indicated to play a significant role in the organic carbon, N, and S cycling in shallow peat. Although constrained by anaerobic respiration and methanogenesis, deep peat exhibited a higher metabolic potential for organic carbon degradation, primarily mediated by Acidobacteriota. In terms of CH<sub>4</sub> turnover, subsurface peat (10-20 cm) was a CH<sub>4</sub> production hotspot, with a net turnover rate of ~2.9 nmol·cm<sup>-3</sup>·d<sup>-1</sup>, while the acetoclastic, hydrogenotrophic, and methylotrophic methanogenic pathways all potentially contributed to CH<sub>4</sub> production. The results of this study improve our understanding of biogeochemical cycles and CH<sub>4</sub> turnover along peatland profiles.

## 29. Is the impact of groundwater on lake greenhouse gas dynamics underestimated? A comparative analysis of subsurface and ecological factors

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Lakes are recognized as important sources of greenhouse gasses (GHGs) emissions to the atmosphere, influenced by various ecological processes. Groundwater discharge into lakes, despite its small volume, has high concentrations of dissolved carbon and nitrogen that significantly affect the production and emission of GHGs from lakes. Therefore, a comprehensive investigation of the mechanisms behind lake GHGs emissions under the influence of groundwater discharge and ecological processes, holds substantial scientific importance. However, due to uncertainties and quantification challenges associated with groundwater discharge, related research is currently limited. Here, we conducted year-round field observations on Honghu Lake, a large shallow eutrophic lake in Hubei Province, China. We analyzed the seasonal variation of GHGs emissions and quantified the groundwater discharge flux using radon ( $^{222}\text{Rn}$ ). The fluxes of methane ( $\text{CH}_4$ ), carbon dioxide ( $\text{CO}_2$ ), and nitrous oxide ( $\text{N}_2\text{O}$ ) at the water–air interface were estimated to be  $31.1 \pm 4.88 \text{ mg m}^{-2} \text{ d}^{-1}$ ,  $386 \pm 90.7 \text{ mg m}^{-2} \text{ d}^{-1}$ , and  $0.327 \pm 0.072 \text{ mg m}^{-2} \text{ d}^{-1}$ , respectively.  $\text{CH}_4$  is the primary greenhouse gas in Honghu Lake, contributing to 82.2 % and 62.2 % of the lake's total emissions over 20-year and 100-year frames. The average rate of groundwater discharge was  $8.19 \pm 0.471 \text{ mm d}^{-1}$ , with the highest discharge rate in winter and the lowest in spring. The daily groundwater discharge volume accounts for 0.649 % of the lake's total water volume. The daily contributions of groundwater discharge to the lake's total emissions of  $\text{CH}_4$ ,  $\text{CO}_2$ , and  $\text{N}_2\text{O}$  were 0.318 %, 12.1 %, and 2.59 %, respectively. The facilitative role of groundwater discharge in  $\text{CO}_2$  emissions primarily manifests through the transport of dissolved organic carbon (DOC) and high concentrations of  $\text{CO}_2$  into the lake. Meanwhile,  $\text{CH}_4$  emissions depend on the activity of methanogenic bacteria, substrate availability, and anaerobic conditions, whereas  $\text{N}_2\text{O}$  emissions are influenced by temperature and nutrient levels. Our study reveals that in the short term, the effect of

groundwater on lakes is relatively minimal. Yet, its long-term role as a steady supplier of carbon and nutrients to lakes should not be overlooked. This study reveals the ways in which groundwater discharge and internal lake dynamics collectively fuel GHGs emissions.

### 30. High intraspecific diversity of *Candidatus Bathyarchaeia* across an 8,000-year lacustrine sedimentary sequence

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*Candidatus Bathyarchaeia* is one of the most abundant microbial groups on Earth (~2.0 to 3.9 × 10<sup>28</sup> cells globally) and constitutes approximately 10% of the microbial biomass found in marine sediments. However, research on diversity, biogeography, and the ecological role of the group is in its early stages. Here, we studied the intraspecific taxonomic diversity, functional diversity, and genomic potential of *Bathyarchaeia* within the local geographical context of an 8000-year Holocene sedimentary succession from Lake Cadagno, a sulfidic, Alpine lake with a complex depositional history. We used shotgun, genome-resolved, and Nanopore sequencing on 13 sediment samples that reflect the depositional history, lithology, and geochemical gradients of the sediments. The taxonomic profiling based on the ribosomal protein RpS3 clustered at 90% identity revealed the presence of 39 subgroups of *Candidatus Bathyarchaeia*, each distributed across one or more sediment samples. Furthermore, we found that in organic matter-depleted sediments below 40 cm (> 220 years), the *Bathyarchaeia*-related populations represent 18.8-39.34% of the microbial community and are potentially involved in carbon fixation and organic carbon cycling. To study the genomic potential and functional niche partitioning of the subgroups, we reconstructed 31 high-quality metagenome-assembled genomes (MAGs) from the different sediment samples. The MAGs belong to the orders *Wuzhiqibiales*, *Xuanwuarculales*, *Jinwuousiales*, *Bifargarchaeales*, and *Baizomonadales* and are distributed in eleven 97% nucleotide sequence identity clusters. Further analyses include performing genome-to-genome comparisons within the MAG clusters using Pangenomics to determine whether the phylogenetic composition and gene content of *Ca. Bathyarchaeia* representatives from Lake Cadagno are correlated to environmental factors.

### 31. The dynamics of methane ebullition in heterogeneous stream landscapes

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Methane (CH<sub>4</sub>) is a potent greenhouse gas with 25 times higher global warming potential than carbon-dioxide (CO<sub>2</sub>). Dramatic rise of methane concentration in the atmosphere since the industrial revolution has, therefore, mostly been due to the negative anthropogenic influence on the environment, even though it is produced in natural environments (e.g. wetlands, ruminants, paddy soils) as well. Although inland waters have only recently been recognised as a significant source of methane, it has since been found that they are responsible for up to roughly half of the global methane emissions. Due to the low solubility of methane in the water, streams are frequently oversaturated in CH<sub>4</sub>. Methane emission in the freshwater systems can occur via diffusion (*i.e.* concentration gradient) or through ebullition where CH<sub>4</sub> is formed and trapped in the sediments until it is rapidly transported in gas bubbles to the atmosphere. Production of methane in the sediments is attributed to the methanogenic archaea, while consumption in the sediment-water interface and water column to the methane-oxidizing bacteria (MOB). There are many knowledge gaps regarding methane ebullition, like uncertainties in the estimation of methane ebullition related to the temporal dynamics due to short-term measurements and limited knowledge on the distribution patterns in the heterogeneous stream landscape. Furthermore, a comparable and standardized sampling and measurement methodology is needed. The aim of this study is to elucidate temporal methane dynamics by performing sampling campaigns across seasons, which will establish the magnitude and drivers of methane ebullition and to investigate the microbial networks involved in processes of production and consumption of methane. Also, comparison and testing of different sampling approaches will be done in effort to improve and standardize ebullition measurement across different streams and scales. Better understanding of methane emissions from freshwater systems is urgent especially in the context of climate change where elevated methane emissions from inland waters are predicted.

## 32. Effect of temperature and liquid exchange on the structure of an aquifer microbial community and its capacity for dissimilatory nitrate reduction

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High temperature-aquifer thermal energy storage (HT-ATES) aims at the seasonal storage and extraction of large quantities of heat in the subsurface. However, the impacts of temperature fluctuations caused by HT-ATES towards biodiversity and ecosystem services in the subsurface environment remain unclear. Hence, studying possible temperature adaptation mechanisms of aquifer microbial communities is crucial to assess potential environmental risks associated with ATES. In this study, we investigated the effects of temperatures between 12 and 80 °C on a pristine aquifer microbial community and its capacity to reduce nitrate, a common global groundwater contaminant. <sup>13</sup>C-labeled acetate was used as an easily consumable carbon and energy source for nitrate reduction, allowing precise activity measurement by analysis of released <sup>13</sup>CO<sub>2</sub>. We observed nitrate reduction coupled to acetate mineralization at 12°C, 25°C, 38°C, 45°C and 60°C but not at 80°C. The rates of acetate-coupled nitrate reduction at 12 - 38°C were significantly higher than rates at 45°C and 60°C. Temperature significantly affected the composition of the acetate-mineralizing, nitrate-reducing microbial communities. Members of the family *Pseudomonadaceae* dominated in the enrichments incubated between 12°C, 25°C and 38°C, accompanied by phylotypes affiliated to the taxa *Comamonadaceae*, *Oxalobacteraceae*, *Caulobacteraceae* and *Rhodocyclaceae*, with varying abundances dependent on the temperature. At 45°C and 60°C, *Pseudomonadaceae*, *Symbiobacteriaceae*, *Paenibacillaceae* and *Planococcaceae* were the dominant family. These findings indicate that the indigenous aquifer microbiome can maintain the ability to reduce nitrate over a wide temperature range, with *Pseudomonadaceae* as key nitrate-reducing family. These results provide support that HT-ATES (temperatures up to 60°C) may allow thermal energy storage whilst simultaneously attenuating nitrate pollution.

### **33. Early Research on the Diversity and Distribution of Candidate Phyla Radiation (CPR) Bacteria in the Bedretto Tunnel**

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The Candidate Phyla Radiation (CPR) superphylum encompasses an extensive group of ultra-small, often symbiotic bacteria with limited metabolic capacities. Despite their broad distribution and potential ecological importance, CPR bacteria remain poorly understood, particularly in subsurface environments. This research focuses on the diversity and distribution of CPR bacteria in the Bedretto Tunnel, a deep subsurface environment located in the Swiss Alps. Using 16S rRNA gene sequencing, we analyzed groundwater samples collected from multiple tunnel fractures to identify the presence and variation of CPR bacteria. Preliminary results reveal a significant phylogenetic diversity of CPR lineages, with novel operational taxonomic units (OTUs) detected across different fractures. Comparative analysis of these results with data from other groundwater sites suggests that the unique geological and hydrochemical properties of the Bedretto Tunnel influence the structure and composition of CPR communities. This study represents a crucial step towards understanding the ecological roles of CPR bacteria in deep subsurface environments and provides a foundation for future investigations into their interactions with environmental factors and potential hosts.