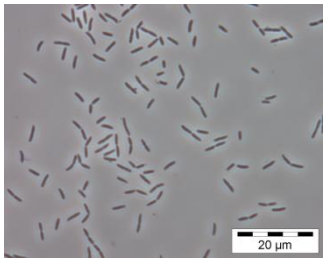
Microbial influences on radionuclide behavior – an example of less-understood problems and how to solve them

Henry Moll, Laura Lütke(1), Velina Bachvarova(2), Andrea Cherkouk, Sonja Selenska-Pobell, Gert Bernhard, Vinzenz Brendler



(1) Institut für Radioökologie und Strahlenschutz (IRS), Leibniz Universität Hannover, 30419 Hannover, Germany.
(2) Department of Developmental Biology / Center for Medical Biotechnology, University of Duisburg-Essen, 45117 Essen, Germany.

IGD-TP 4th Exchange Forum
WG 5 „Microbial Studies“
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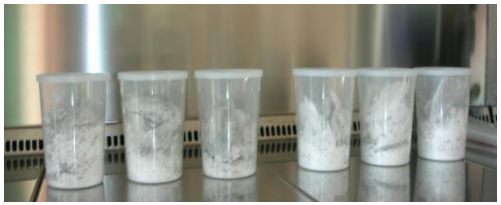
Outline

1. Expectations on the outcome of WG-5 “Microbial Studies”
2. Microbial research at HZDR/IRE – Selected examples
   - → Bacterial diversity in Mont Terri Opalinus Clay
   - → Plutonium interaction experiments with a typical isolate from Mont Terri Opalinus Clay
   - → Direct and indirect interactions of curium with the Äspö –groundwater bacterium *Pseudomonas fluorescens*
3. How can we contribute to a competent safety case?
4. Further research topics that need to be investigated

1. Expectations on the outcome of WG-5 “Microbial Studies”

- Agreement, that indigenous microbes can impact the speciation/migration of actinides in many ambivalent ways.
- Exchange of knowledge concerning microbial impact on radionuclide speciation and hence their migration in the environment after an accidental release on a European scale.
- Initiate cooperation's between laboratories/institutions.
- Major goal: implementing microbial processes in safety assessment.

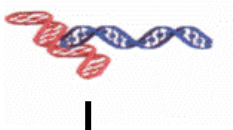
2. Microbial research at HZDR – Microbial Diversity



Enrichment and cultivation of microbial isolates

Clay/salt samples from potential nuclear waste repositories

DNA recovery via direct lysis

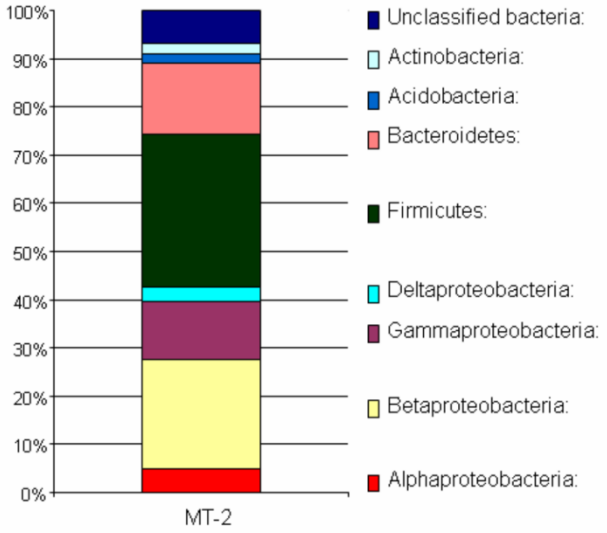


High molecular DNA fragments

Amplified 16S rRNA gene fragments

Cloning, sequencing and phylogenetic affiliation

Bacterial diversity of the unperturbed Opalinus Clay sample MT-2, Mont Terri, Switzerland



Dominance of *Beta-Proteobacteria*, *Firmicutes* and *Bacteriodetes*

2. Microbial research at HZDR – Microbial Diversity (2)

Clay/salt dissolved in different enrichment media incubated under aerobic and/or anaerobic conditions

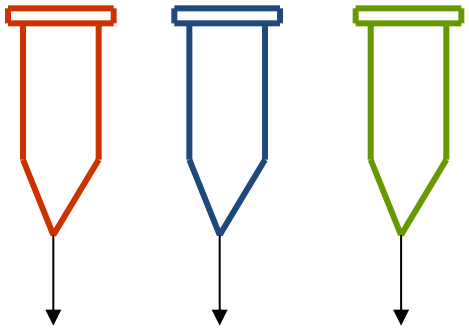
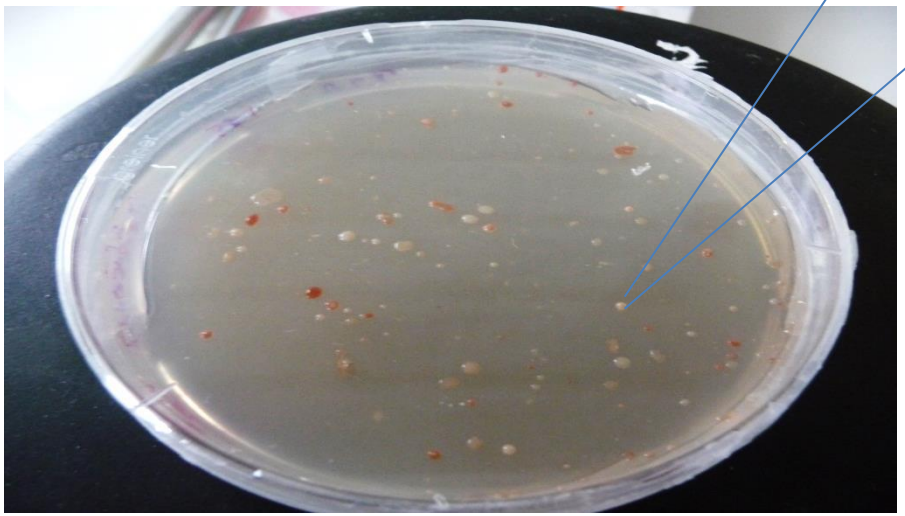
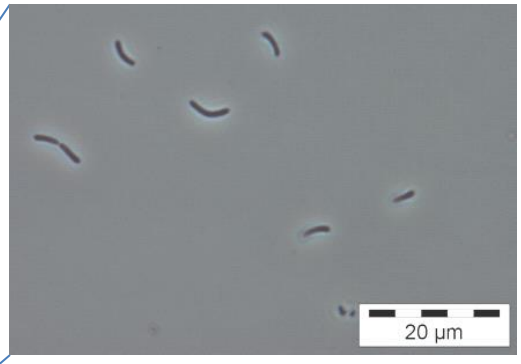


Plate liquid media of each approach on agar plates containing the media and incubate



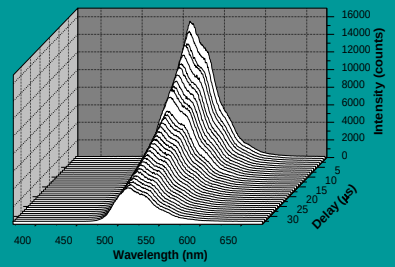


Sporomusa sp. MT-2.99, isolated from the Mont Terri Opalinus clay sample MT-2

Interaction studies of *Sporomusa* sp. MT-2.99 cells with Pu

Techniques at IRE to Study Radionuclide Binding Form

Time-resolved Laser-induced Fluorescence Spectroscopy

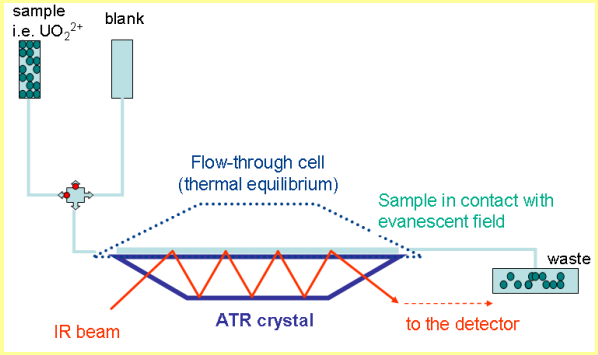


fs-TRLFS technique:
measurement of fluorescence of organic ligand

ns-TRLFS technique:
measurement of RN's luminescence (U(VI), Am(III), Cm(III))

Non-invasive direct speciation technique, advantage of very low radionuclide concentrations

ATR FT-IR Spectroscopy

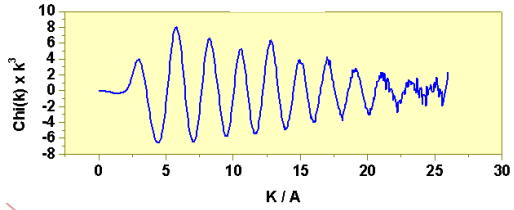


Information about the molecular structures of RNs bound to minerals, ligands or bacterial surfaces

$X \quad Y$
$\diagdown \; \diagup$
Rn^{m+}
$|$
Z

X-ray Absorption Spectroscopy (XAS) @ ROBL

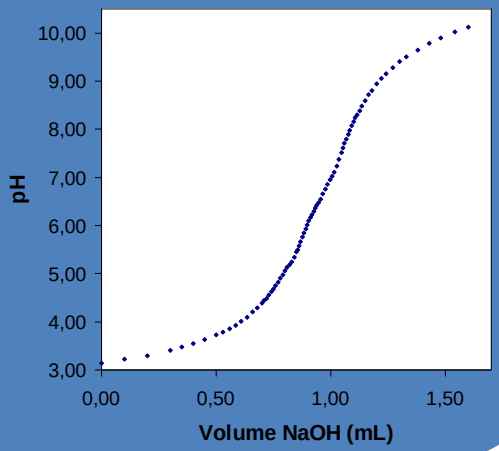
Oxidation state of RN, bond distances, type and number of near neighboring atoms



Sources:
www.hzdr.de
www.esrf.fr

Potentiometric Titration

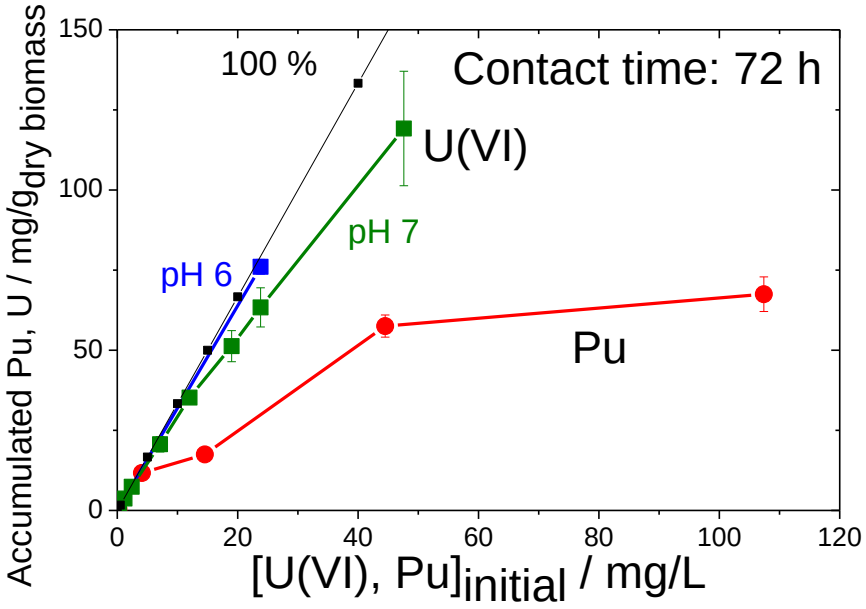
Detection of complex species simultaneously occurring in aqueous system



and others…

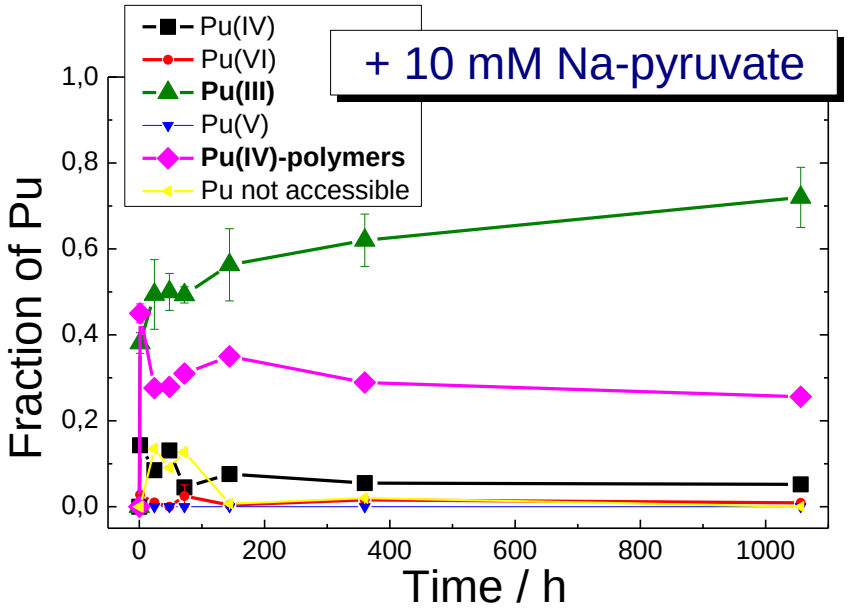
Pu interactions with *Sporomusa* sp. MT-2.99 from Mont Terri Opalinus Clay

An accumulation studies



→ Good U(VI) binding properties
→ Less Pu accumulated[1]

Pu oxidation state distribution on biomass



→ The cells create a reducing environment
→ Enrichment of Pu(III) on biomass

[1] Moll, H. et al. Migration 2013, 08.-13.09.2013, Brighton, UK, poster.

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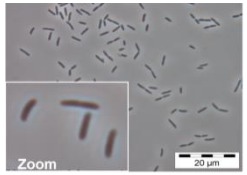
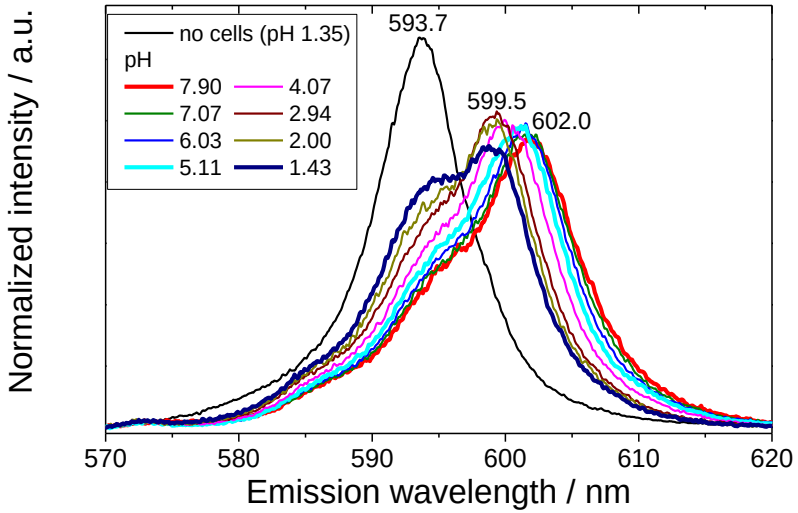
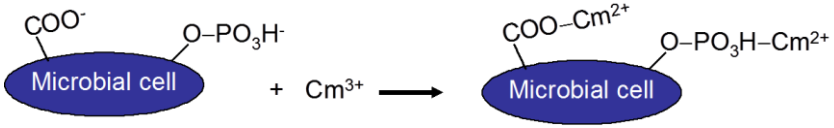
Cm interactions with the Äspö–strain *Pseudomonas fluorescens*



Direct interactions with planktonic cells[1]

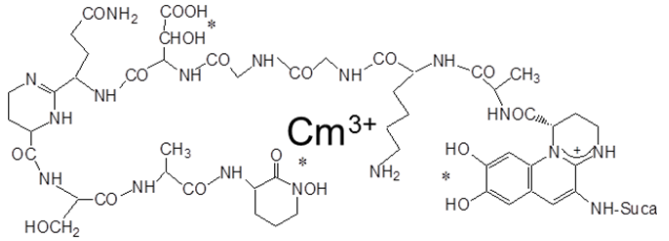
- [Cells] 0.2 $g_{dry\ weight}$/L + [Cm^{3+}] 0.3 µM, I = 0.1 M





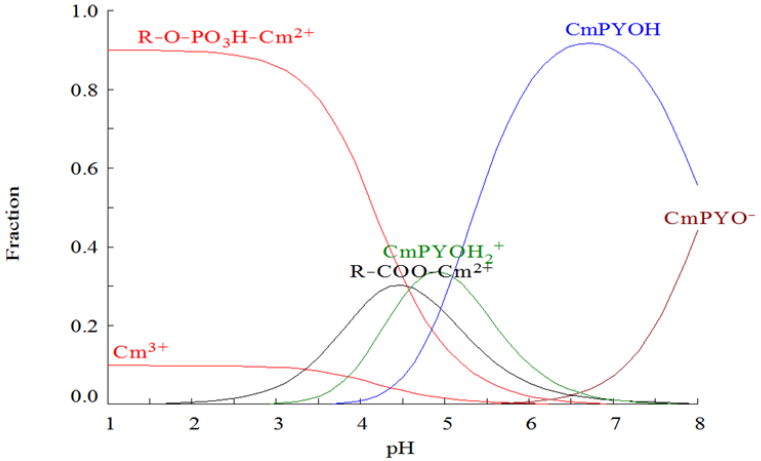
Indirect interactions with secreted pyoverdins[2]

- [PYO] 3 – 10 µM; [Cm^{3+}] 0.3 µM, I = 0.1 M



[Cells] 20 $mg_{dry\ weight}$/L

$[PYO^{4-}]_{TOT}$ = 0.30 µM
$[Cm^{3+}]_{TOT}$ = 0.30 µM
$[R\text{-}O\text{-}PO_3^{2-}]_{TOT}$ = 7.20 µM
$[R\text{-}COO^{-}]_{TOT}$ = 16.40 µM



Two pH-domains:
pH 1 to 5: Cm(III) binding on the cell envelope (biosorption)
pH 5 to 8: Cm(III) binding on pyoverdins (mobilization)

[1] Moll, H. et al. Geomicrobiol. J. 30, 337-346 (2013). [2] Moll, H. et al. Biometals 21, 219-228 (2008).

3. How can we contribute to a competent safety case?

- Study the indigenous microbial community in host rocks from potential nuclear waste repositories.

- Realistic consideration of microbial processes impacting the safety of a repository in the near and far field:
  - Change of geochemical conditions
  - Degradation of organic waste components (gas generation)
  - Degradation of back fill materials
  - Container corrosion induced by microbial action
    → These processes influence the radionuclide speciation after an accidental release.
  - Interactions of the microorganisms with radionuclides

⇨ Sensitization of stakeholders in safety assessment for a possible microbial impact on the safety of a repository especially in the far field.

4. Further research topics that need to be investigated

- Knowledge should be generated concerning the **characterization/identification of indigenous microbial communities** in host rocks (e.g. salt, clay) foreseen for a safe storage of nuclear waste including pore and ground waters: We need to know which microbes occur in order to judge their impact in case of a hazardous incident.

- **Metabolic activity of indigenous microbes** at the specific location of the repository.
  - Change of geochemical conditions; gas generation; corrosion of container

- Knowledge should be available concerning **main interaction processes/paths between radionuclides and microbes** (planktonic cells and biofilms):
  - How much is bound?
  - Radionuclide speciation?
  - Change of geochemical conditions by microbes
  - Immobilization/mobilization

- What parameters do we need to model microbial processes in a repository?

➡ Cooperation between experimenters and safety assessment people.

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Thank you for your kind attention!!

DRESDEN concept

HZDR

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