

# Interaction of uranium with isolated microorganisms of the former uranium mine Königstein (Saxony, Germany)

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Underground before flooding

## Background: The former uranium mine Königstein

- Mining from 1976 - 1990, area of 6 km<sup>2</sup>
  - Leaching by sulfuric acid
- ➔ leads to mobilization of uranium and other heavy metals
- Since January 2013 controlled flooding of the underground mine
- Detection of high biodiversity (eukaryotic and prokaryotic) of microorganisms by Isabel Zirnstein in the flooding water [1]
- Expensive waste water treatment of the flooding water



Surface after flooding

## Methods and Materials

- Isolation of microorganisms by SDA medium (for eukaryotic organisms)
- Identification with 16S rDNA and 18S rDNA analysis → sequencing
- Biosorption experiments with uranium (initial concentration 1x10<sup>-4</sup>M, background tap water pH 5,0) → measurement ICP-MS/-OES
- Cell viability → live-dead staining, flow cytometry (in coop. with M. Merroun University Granada, Spain)

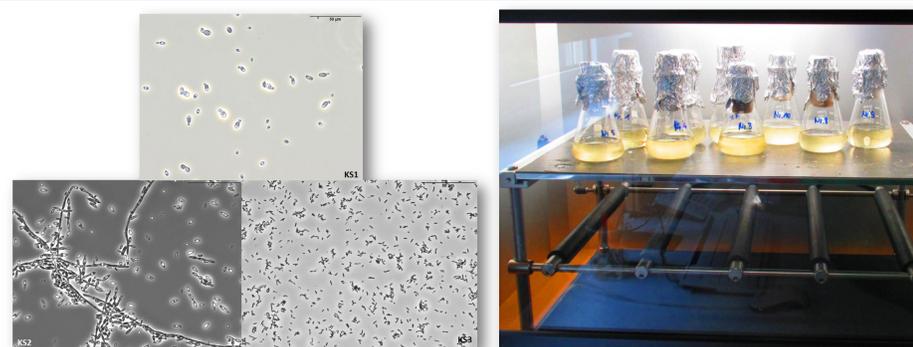


Figure: Left: Light microscopy of isolated microorganisms (KS1-3); Right: Cultivation of isolates

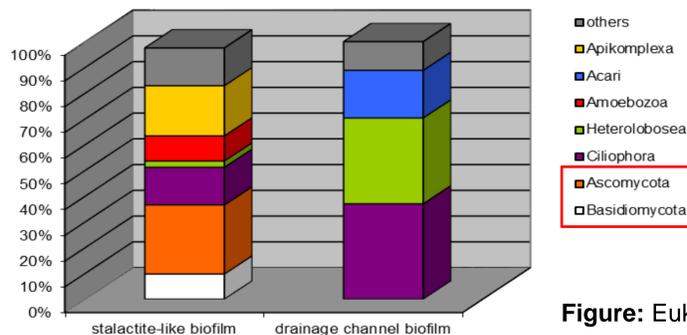
## Results

### Isolated microorganisms by culture dependent methods vs. detected microorganisms by culture independent methods

Table: Isolated microorganisms of the flooding water, on SDA medium, Eukaryotic organisms detected by 18S rDNA, Prokaryotic organisms detected by 16S rDNA

Phylum	Isolate	Closest phylogenetic relative	Accession no., similarity (%)
Basidiomycota	KS1	<i>Rhodospiridium toruloides</i>	DQ647614.2, 99
Ascomycota	KS2	<i>Microscypha cajaniensis</i>	EU940037.1, 95
Proteobacteria	KS2	<i>Acidocella sp.</i>	AF531447.1, 98
Firmicutes	KS3	<i>Bacillus sp.</i>	AB769179.1, 99

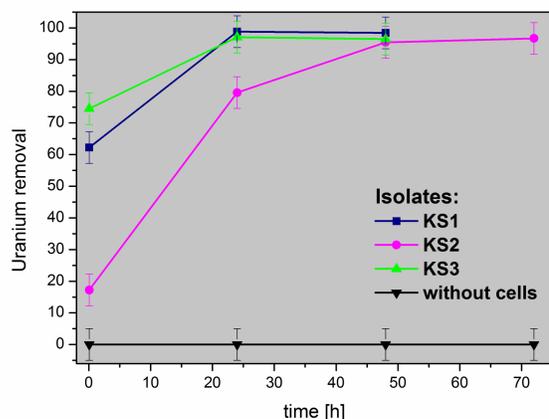
Eukaryote biodiversity by 18S rDNA analyses



- Isolates of the culture dependent method are dominant species which were also detected by culture independent method

Figure: Eukaryotic biodiversity by Isabel Zirnstein [1]

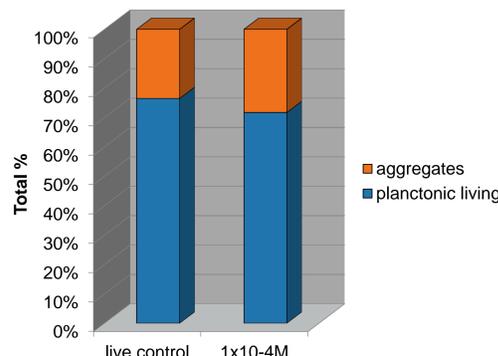
### Biosorption of uranium by isolates KS 1, KS2 and KS3



- Isolates KS1 and KS3: fast uranium removal → **passive biosorption**
- Isolate KS2: slower removal → **active bioaccumulation**

Figure: Initial uranium concentration: 1x10<sup>-4</sup>M, background: tap water, pH: 5,0

### Flow cytometry of isolate KS1 in coop. with M. Merroun (University Granada, Spain)



- Isolate KS1 shows a high tolerance to a uranium concentration of 1x10<sup>-4</sup>M

Figure: Cells were grown on LPM without uranium (live control) and with uranium (1x10<sup>-4</sup>M); for flow cytometry the cells were stained with FDA and PI

References: [1] Zirnstein et al., (2012), MicrobiologyOpen, 1: 83-94

## Conclusion

- The isolated eukaryotic microorganisms are the dominant species in the flooding water of the former uranium mine in Königstein
- The isolated microorganisms KS1 and KS3 show fast and high uranium removal within the first 24 h → **passive process** by biosorption
- The isolate KS2 shows a slower uranium removal, but also high amounts → **active process** by bioaccumulation
- The yeast cells of isolate KS1 (*Rhodospiridium toruloides*) show a high tolerance to uranium

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