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STUDIECENTRUM VOOR KERNENERGIE
CENTRE D'ETUDE DE L'ENERGIE NUCLEAIRE

Geomicrobiology of waste repositories

Gas generation & removal
Radionuclide migration & retention

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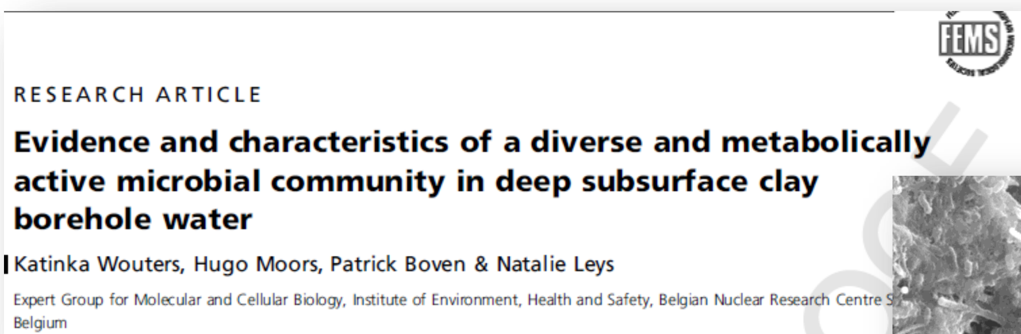
Geomicrobiology in waste repositories

- Geochemical experimental set-ups were, are and will be influenced by microbiological activity.
  - E.g. Microbial H_2 consumption & CH_4 production in 'gas diffusion tests'
  - E.g. Microbial gas production in 'nitrate tests'
  - E.g. Biocorrosion and biodegradation of equipment & sensors
  - E.g. Microbial colonisation & biofilm development 'hampering *in situ* monitoring' and analysis of piezometers
  - ....

- Repository and host rock were, are and will never be sterile.

(And for safety it is irrelevant whether the microbes are introduced or indigenous.)


FEMS

RESEARCH ARTICLE

Evidence and characteristics of a diverse and metabolically active microbial community in deep subsurface clay borehole water

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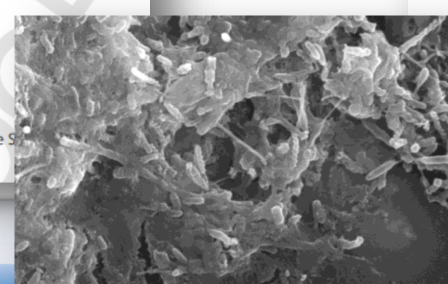
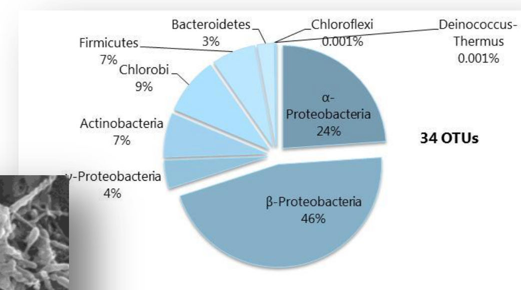
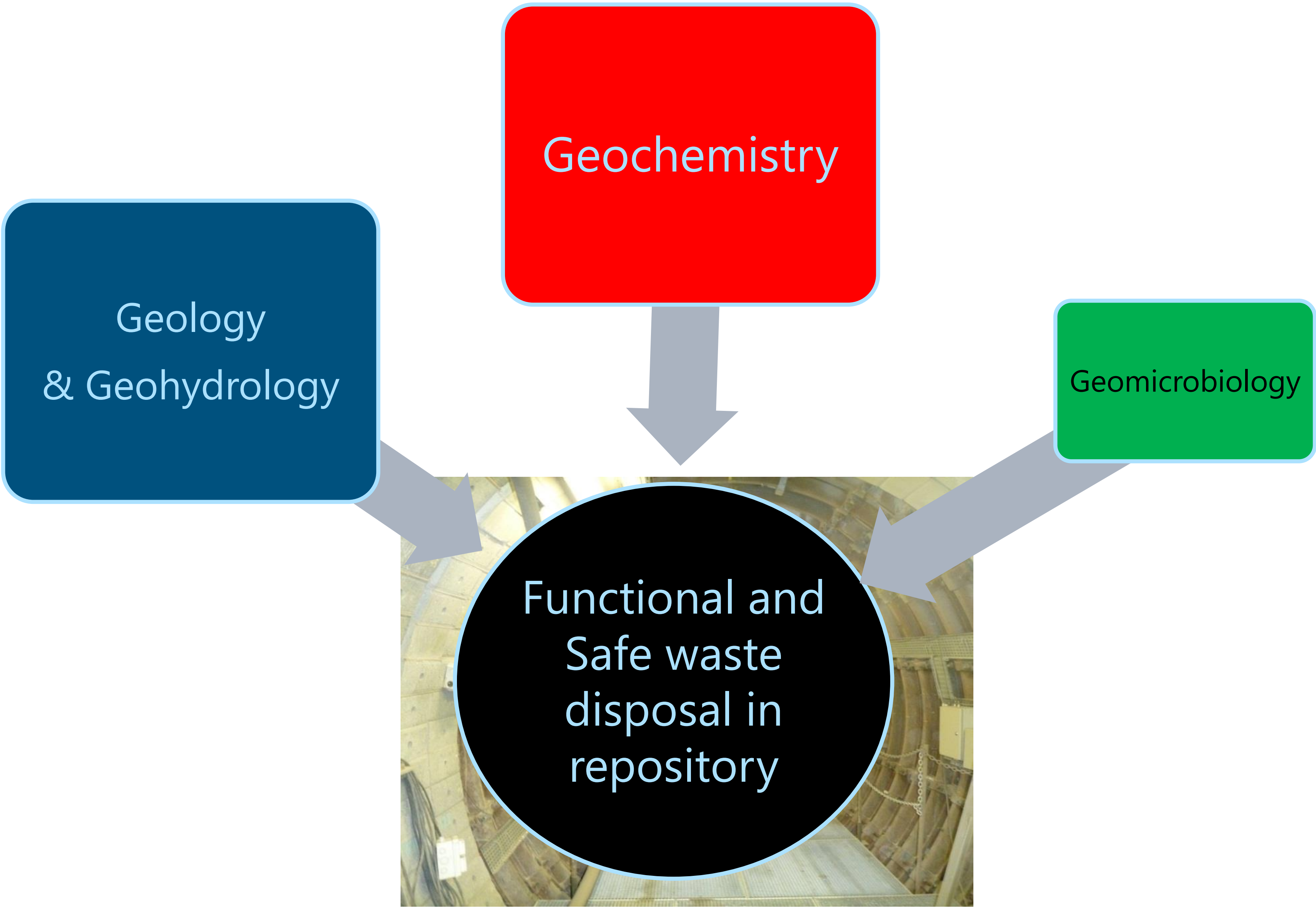
Belgium






Geochemistry
Geology
& Geohydrology
Geomicrobiology
Functional and
Safe waste
disposal in
repository

Geomicrobiology in waste repositories

- Problem

  Microbes will be present and active,

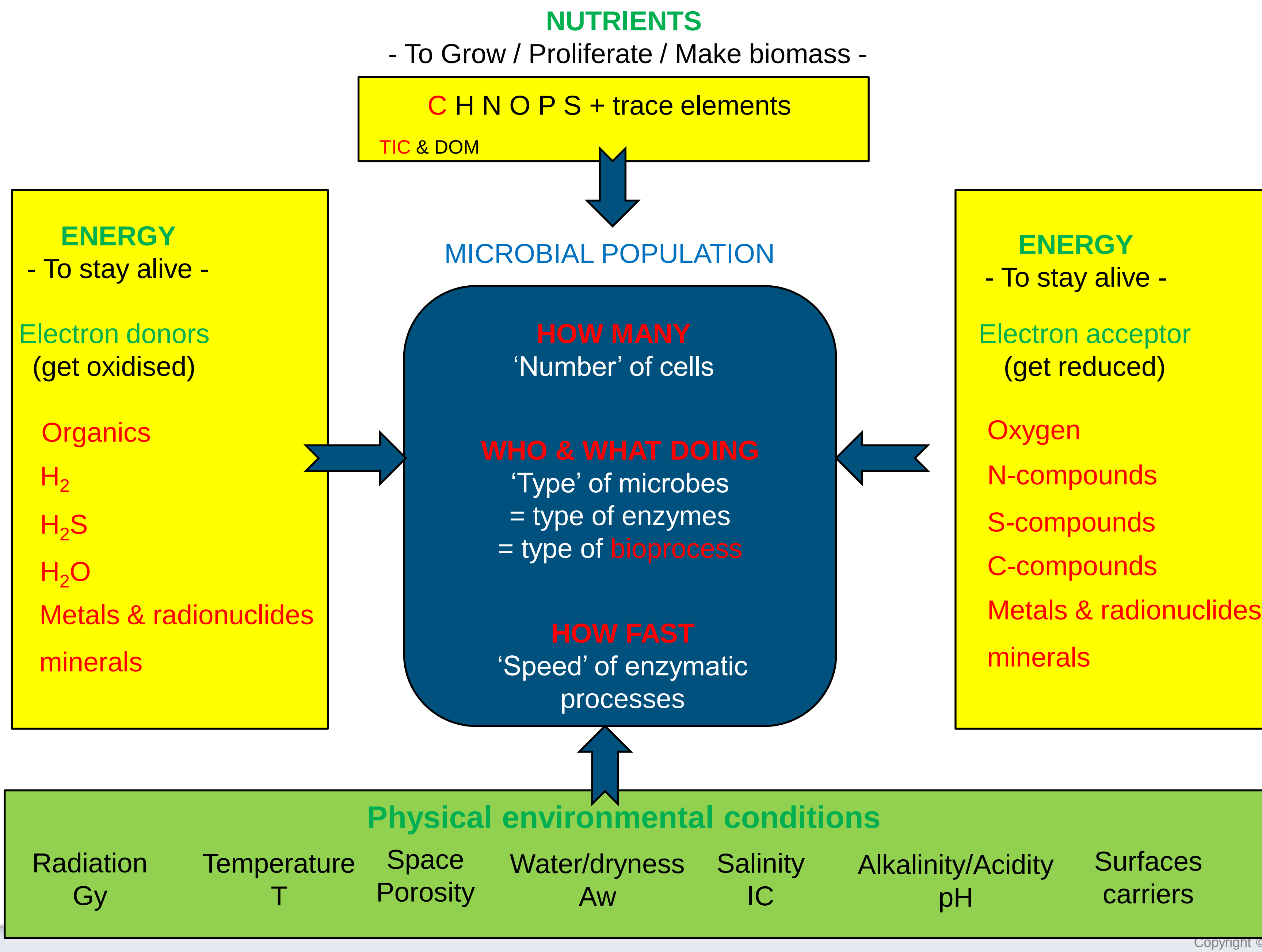
  and will interact with waste, container, repository and host rock,

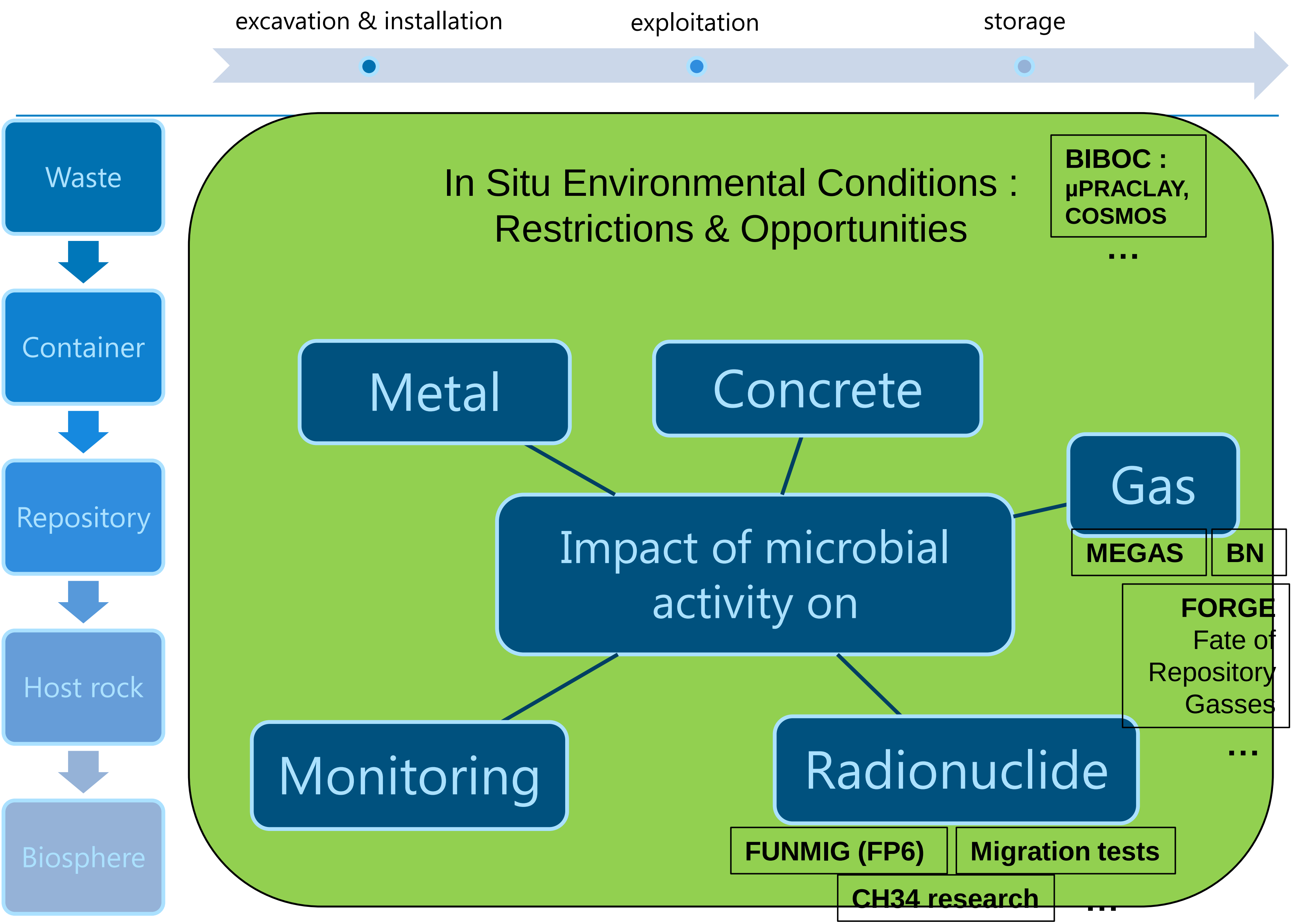
  during excavation, exploitation, and storage.

  ↓

  Microbiology cannot be neglected but has to be considered and taken into account in the experimentation and modelling, and the final design and operational procedures, to assure functional and safe waste storage.

- Need
  - A better understanding of
    - the microbial populations present in waste, repository and host rock
    - the microbial bioprocesses that can occur at in situ conditions
    - the impact of those processes on the water & surface geochemistry
    - the impact of those geochemical changes on the waste, repository and host rock barrier function


NUTRIENTS
- To Grow / Proliferate / Make biomass -
C H N O P S + trace elements
TIC & DOM
ENERGY
- To stay alive -
Electron donors
(get oxidised)
Organics
H_2
H_2S
H_2O
Metals & radionuclides
minerals
MICROBIAL POPULATION
HOW MANY
'Number' of cells
WHO & WHAT DOING
'Type' of microbes
= type of enzymes
= type of bioprocess
HOW FAST
'Speed' of enzymatic processes
ENERGY
- To stay alive -
Electron acceptor
(get reduced)
Oxygen
N-compounds
S-compounds
C-compounds
Metals & radionuclides
minerals
Physical environmental conditions
Radiation Gy
Temperature T
Space Porosity
Water/dryness Aw
Salinity IC
Alkalinity/Acidity pH
Surfaces carriers



Elie Valcke, Christophe Bruggeman, Norbert Maes,

Microbial Production of Gas

- Nitrogen cyclus :
  - nitrate from waste → N_2O, NO, N_2 (NRB, Nitrate E-acceptor, only when anoxic)
- Sulphur cyclus :
  - clay → pyrite → sulphate (gips) (during excavation, when oxic)
  - sulphate (gips) → H_2S (SRB, sulphate E-acceptor, only when anoxic)
- Carbon cyclus :
  - organic acids → CO_2 (Hetrotrophs & organic E-donor, oxic & anoxic)
  - CO_2 → methane CH_4 (methanogens, CO_2 E-acceptor, anoxic)

→ **Production of reduced gas species with impact on**
- → **water geochemistry**
- → **structural materials (corrosion)**
- → **dissolution chemistry and mobility of radionuclides**

Microbial Consumption of Gas

- Oxygen
  - Most preferred E-acceptor for most microbes, can be consumed very fast

- Hydrogen from anoxic corrosion & radiolysis
  - very good ‘fuel for microbes’ (E-donor)
  - oxic (with O_2 as E-acceptor)
  - anoxic (with nitrate or sulphate as E-acceptor)

- Carbon dioxide
  - As carbon source for autotrophic proliferation

e.g. BN-Experiment, Mont Terri

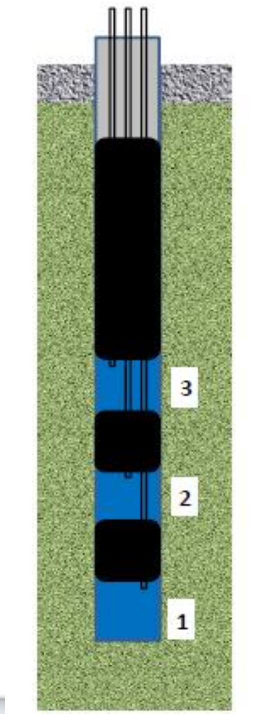
Elie Valcke et al.

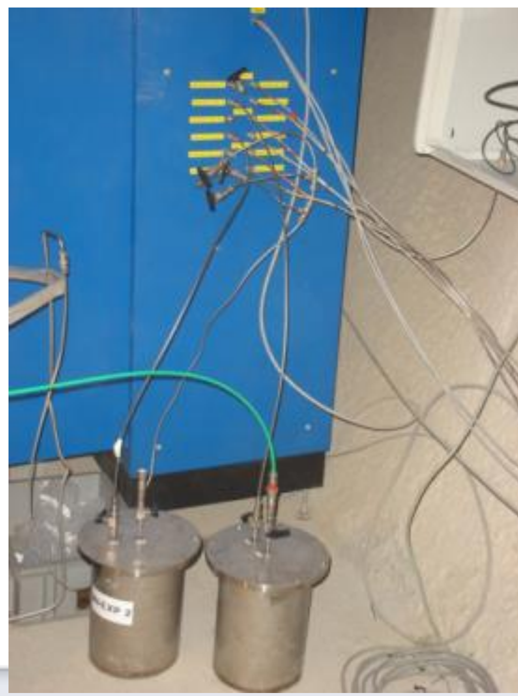
Interaction clay – bitumen waste

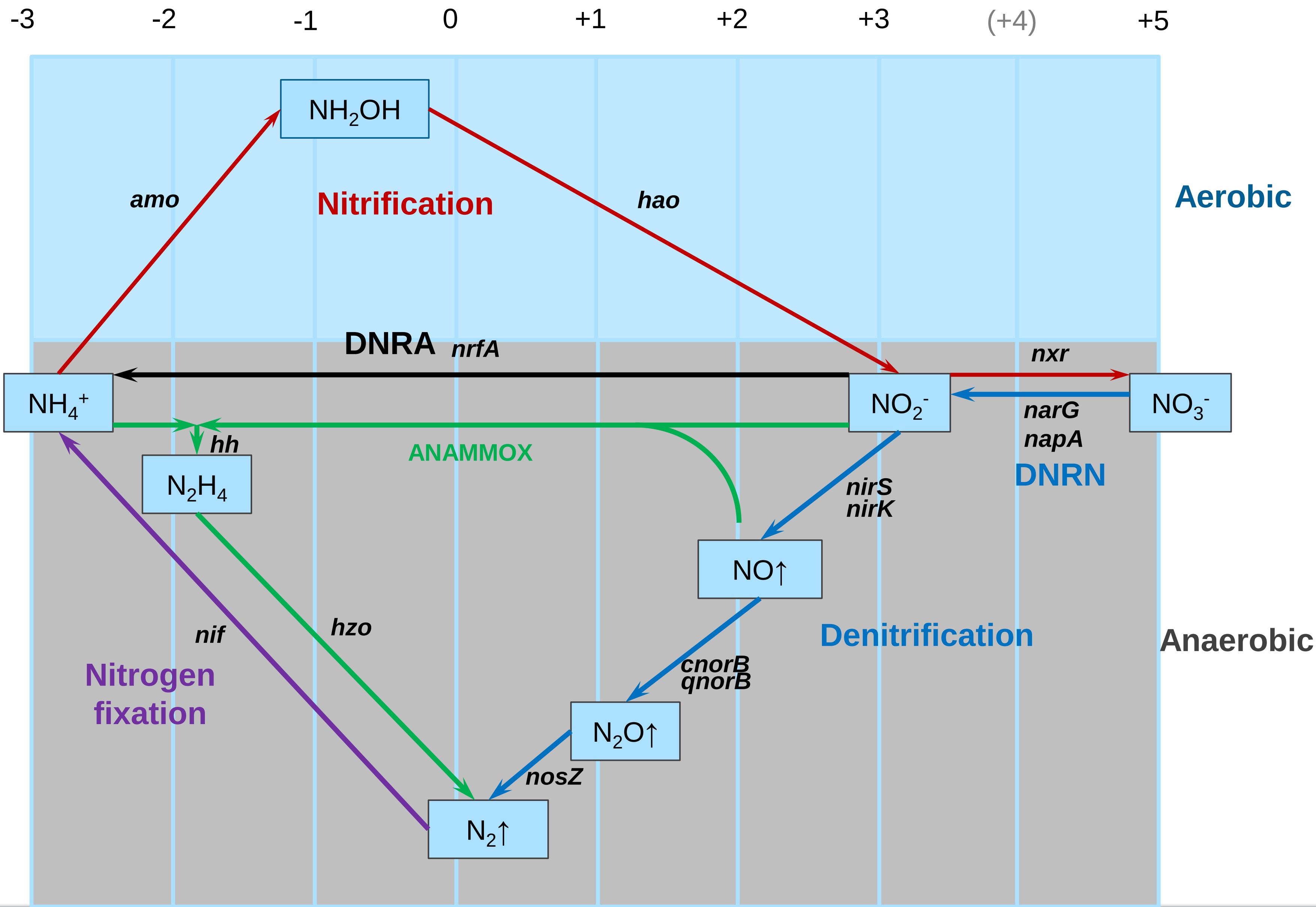
Nitrate & Acetate injection as products

- 3 packed-off intervals
- Sintered stainless steel filter screen surrounding a central tube
- Water lines connecting intervals with recirculation cabinet in gallery

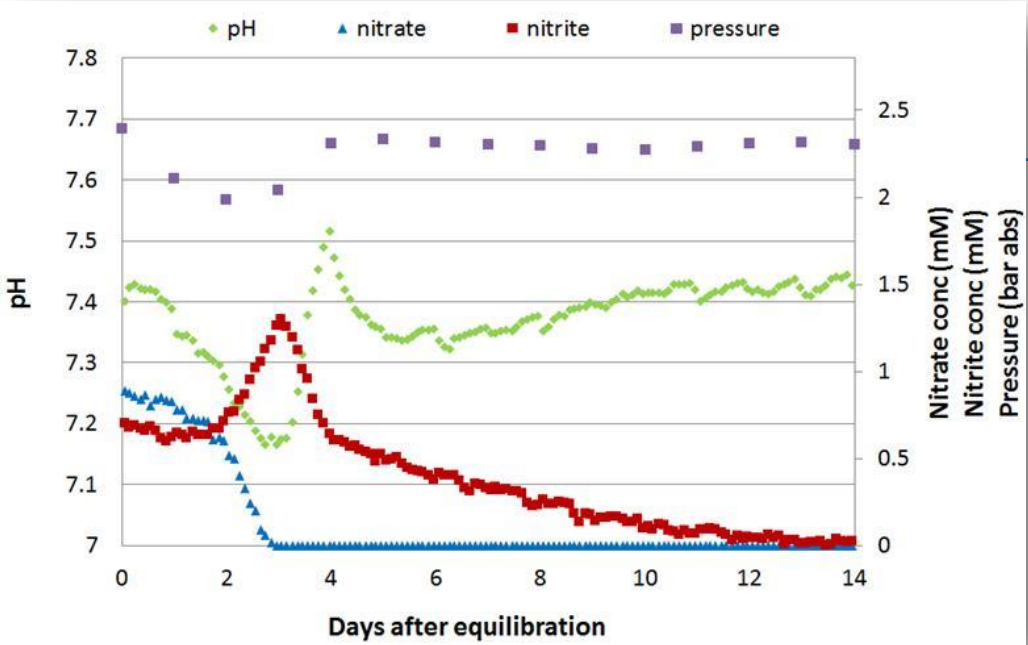
→ Providing 'space' and 'water' & 'rock contact'

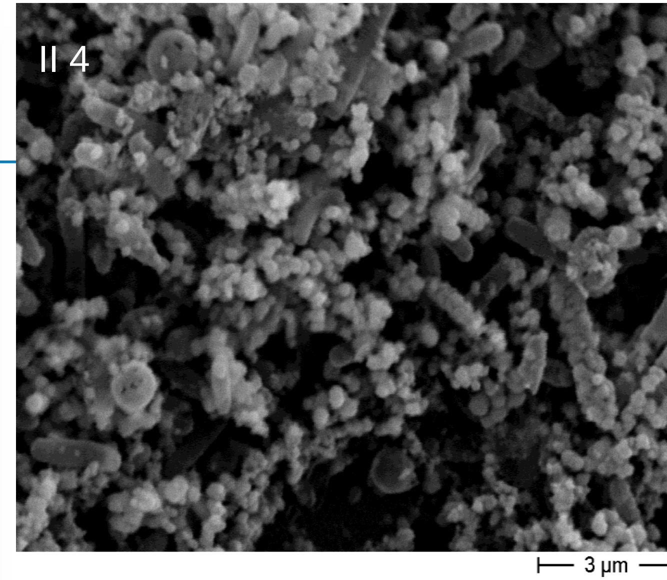


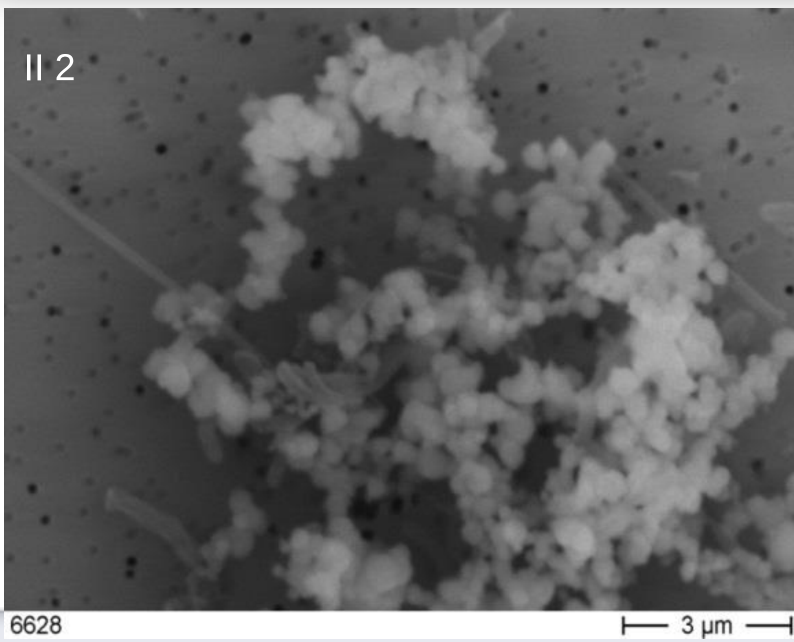




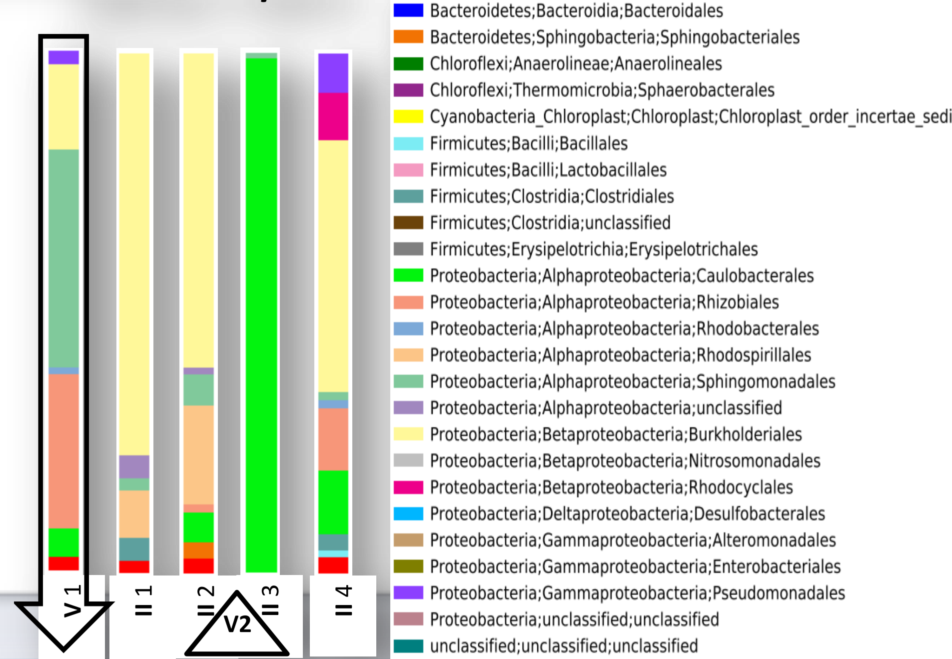
-3
-2
-1
0
+1
+2
+3
(+4)
+5
NH₂OH
amo
Nitrification
hao
Aerobic
DNRA
nrfA
nxr
NH₄⁺
NO₂⁻
NO₃⁻
narG
napA
DNRN
hh
ANAMMOX
N₂H₄
nirS
nirK
NO↑
Denitrification
Anaerobic
nif
hzo
cnorB
qnorB
Nitrogen fixation
N₂O↑
nosZ
N₂↑







Interval II N/C=1



Radionuclide migration

- RN : selenium (Se), uranium (U), neptunium (Np), plutonium (Pu), curium (Cm)

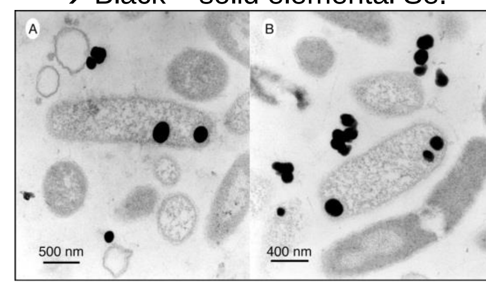
alkalinity (pH),
redox potential (Eh),

- Microbial impact on RN chemical form (speciation)
  - Microbial RN reduction *(RN as E-acceptor, anoxic)*
    → e.g. metalic nanoparticles

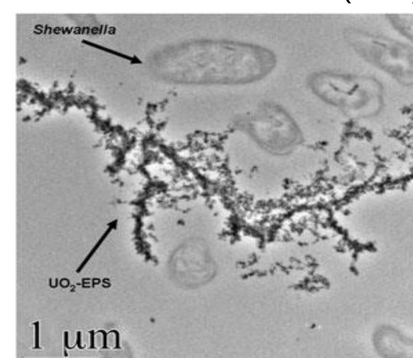
- Microbial impact on RN ligands
  - Degradation of organic ligands *(as C-source)*
  - Production of complexion compounds
    - biomineralisation with phosphate, carbonate, carboxylate …
    - Extracellular polymeric substances (EPS)
    - metallophores (e.g. organic acids, peptides, …)

- Microbial impact on RN sorption
  - RN sorption on microbial biofilm on wetted surfaces
  - RN sorption on microbial colloids

Selenihalanaerobacter shriftii & selenate (A) and selenite (B)
→ Black = solid elemental Se.



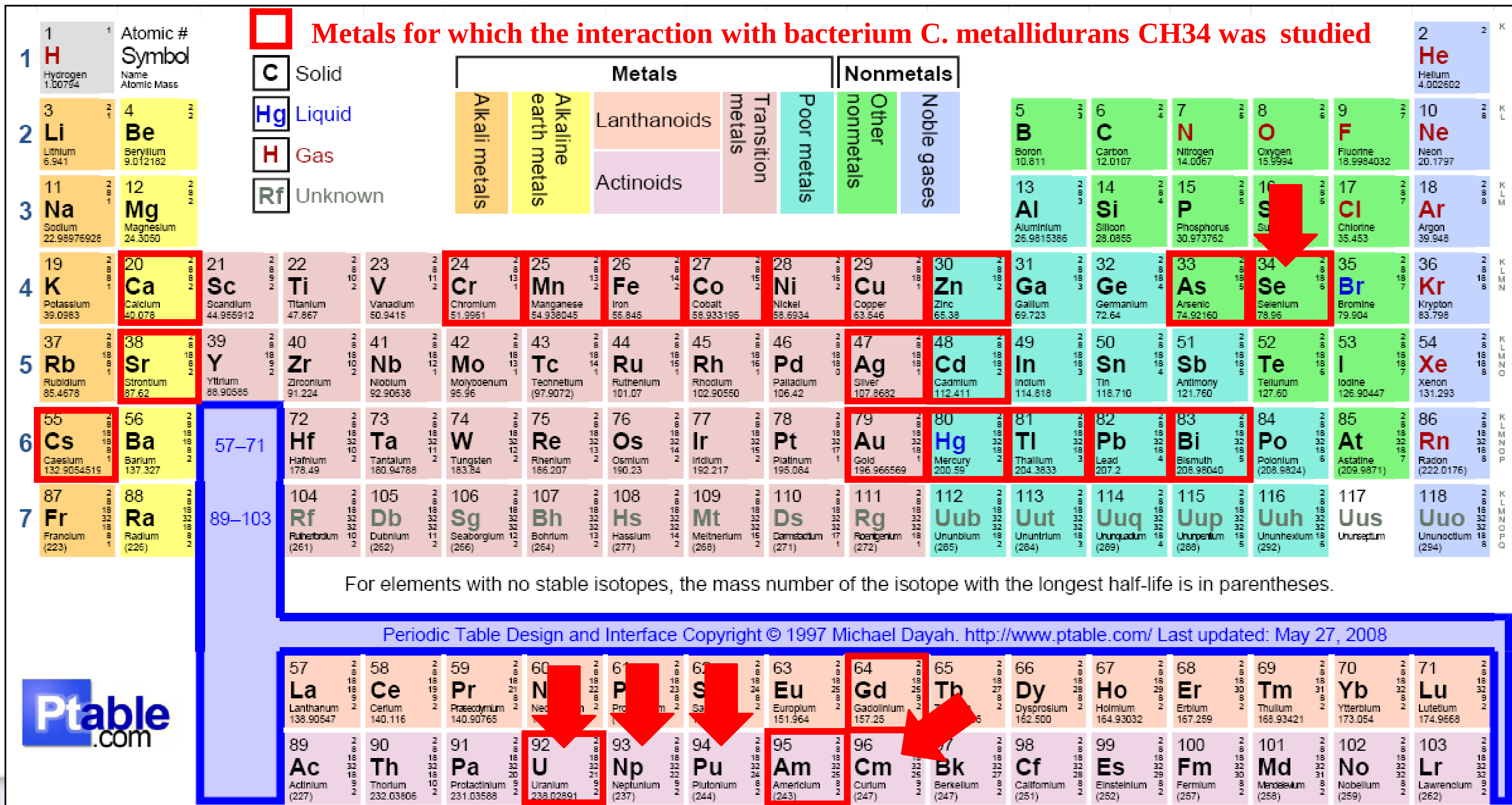
Shewanella & uranyl
→ Black = solid uraninite (UO2).



e.g. Characterising and ‘engineering’ bacteria that bioaccumulate or bioprecipitate metals & RN

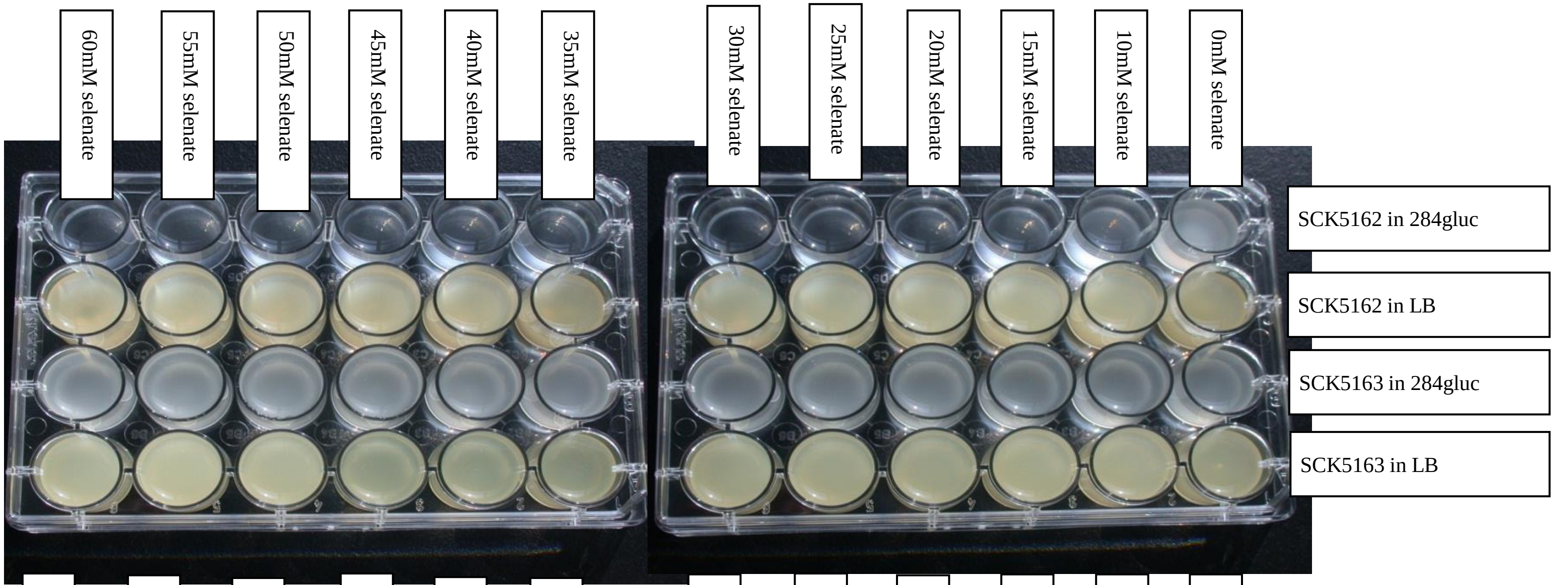
Our model bacterium: *Cupriavidus metallidurans* CH34

Isolated in 1976 from polluted sludge in metallurgical plant (Prayon-Liege, Belgium), by Dr. C. Houba (Ulg) & Dr. M. Mergeay (SCK•CEN), > 30 years of research in SCK, studied > 30 labs all over the world

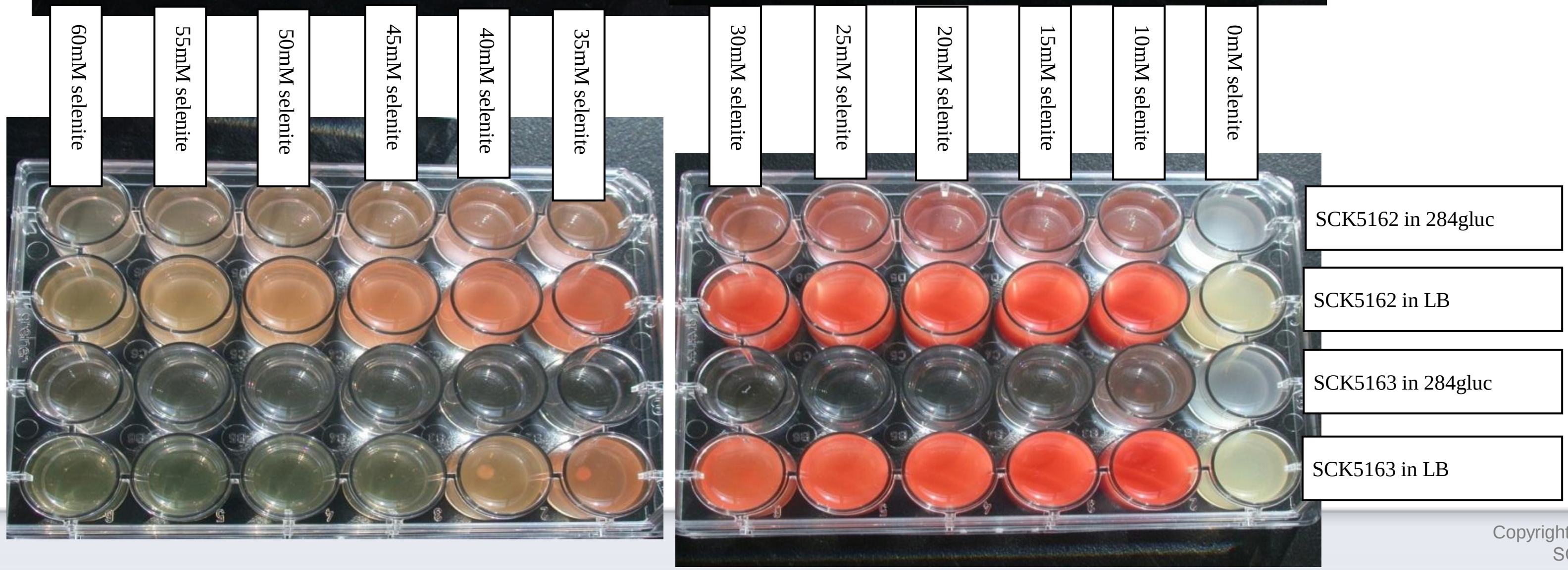


e.g. Toxicity and Reduction testing

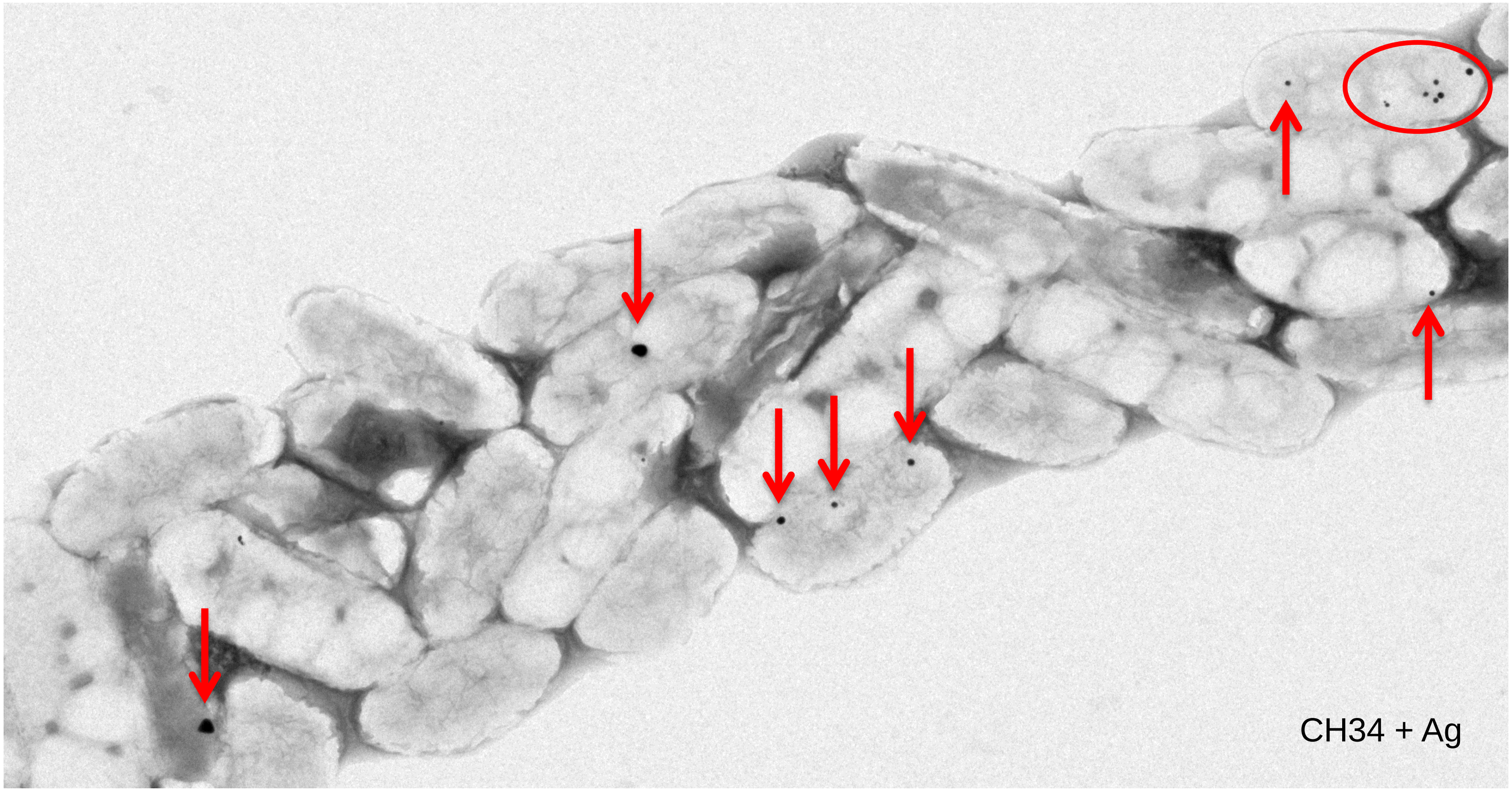
Selenate



Selenite



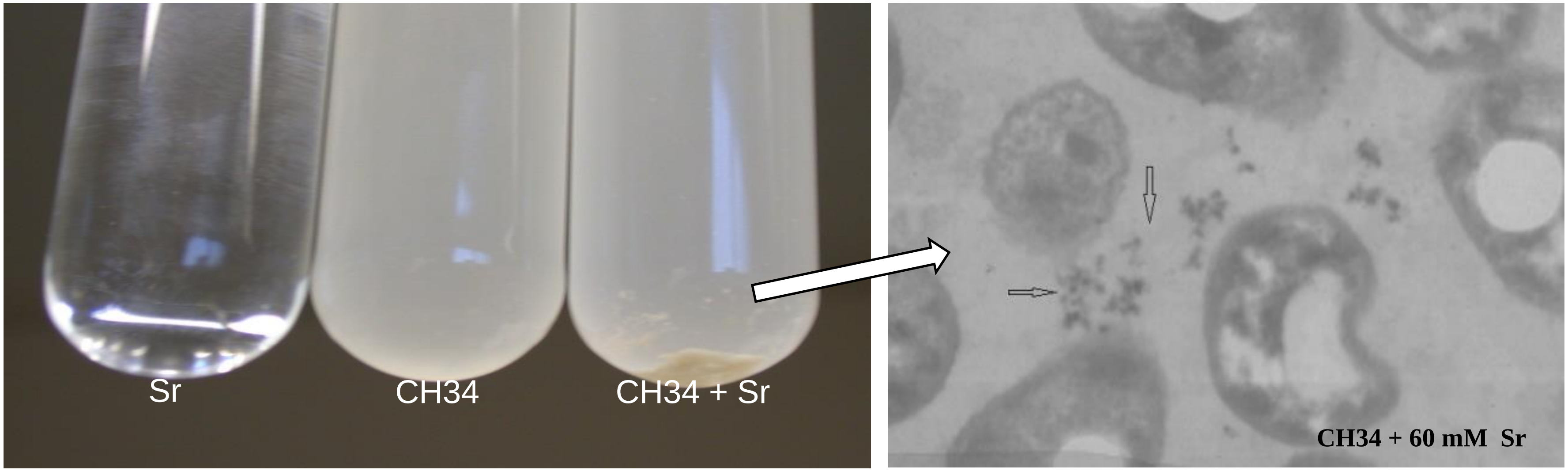
e.g. Biogenic metal nanoparticles



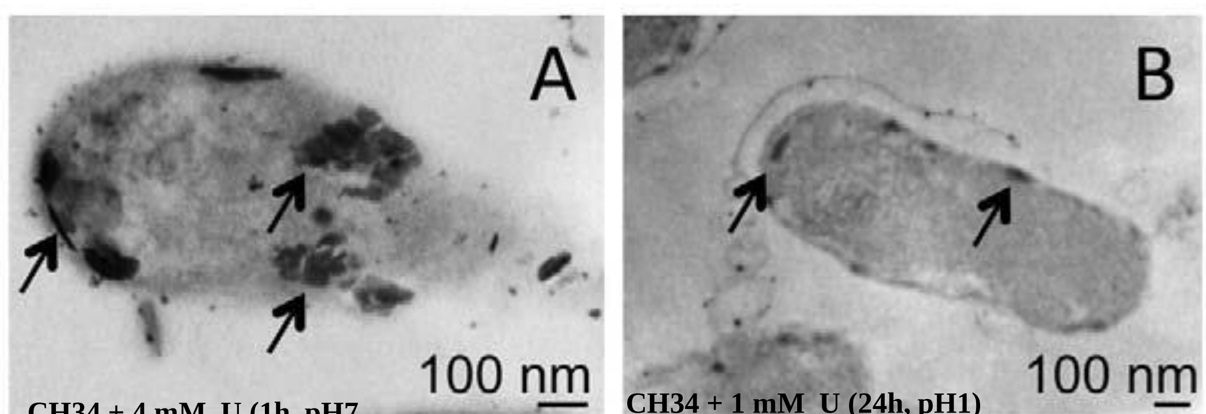
→ silver, paladium, gold, ….

e.g. Biogenic metal carbonates and phosphates

e.g. strontium carbonate

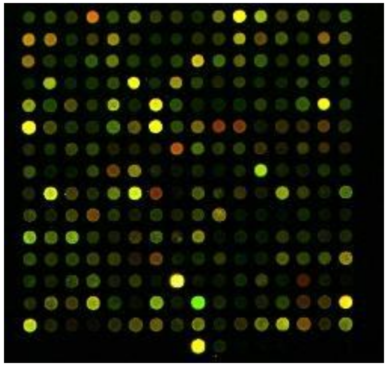


e.g. uranium phosphate



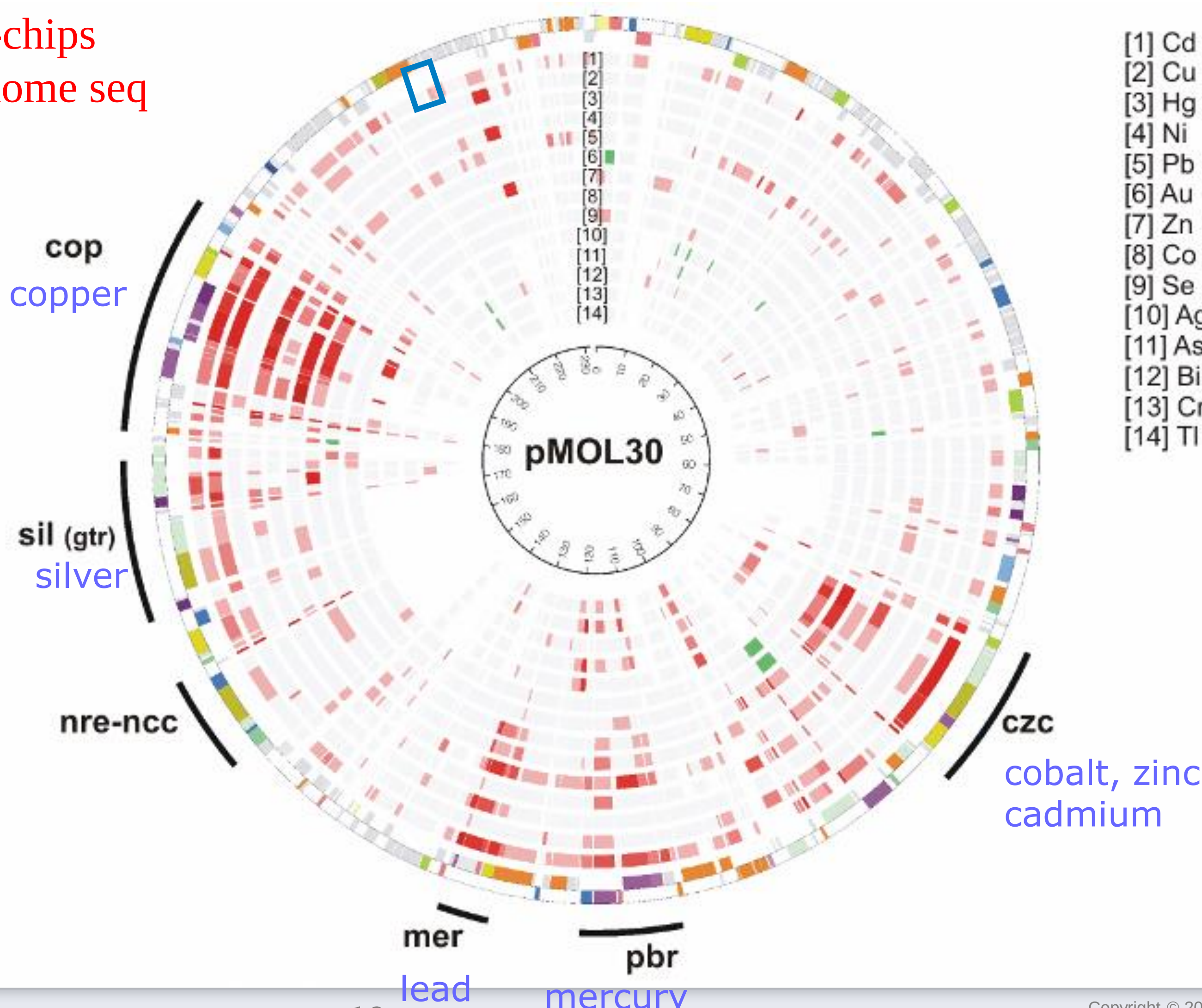
Llorens et al. 2012

e.g. Characterising genes and proteins in bacteria involved in the metal processing



DNA-chips
& genome seq

- Novel genes & proteins involved : mechanisms - efflux pumps, reduction, metallophores …
- Regulation : transcription, regulatory RNA, …
- Mobility : MGE - genomic islands, transposons plasmids, IS, …
- Bioreporters : makers, specificity, sensitivity, …



excavation & installation — exploitation — storage

- Waste
- Container
- Repository
- Host rock
- Biosphere

Gas

contribute to geology & geochemistry & material investigations, to address microbiology aspects in those

ex situ **laboratory batch and** ***in situ*** **field tests, specific for geomicrobiology**

- *Exploitation of existing In situ facilities & their 'diversity' (e.g. investigate same bioprocess but in different environment)*
- *Multidisciplinary (materials, molecular bio, biochemistry, modeling, …)*
- *Involvement of young researchers (e.g. PHDs, mobility, training)*
- *Data comparison*

…

In Situ Environmental Conditions : Restrictions & Opportunities


We are ready to jump on the new scientific challenges ….