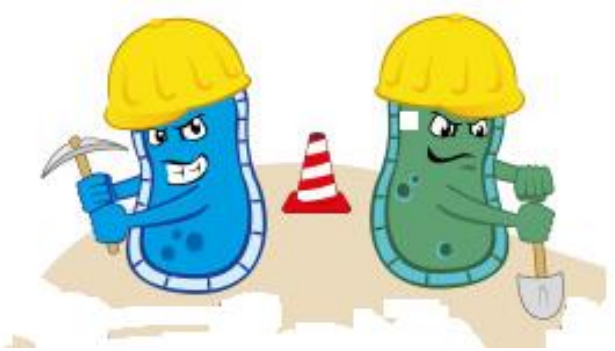
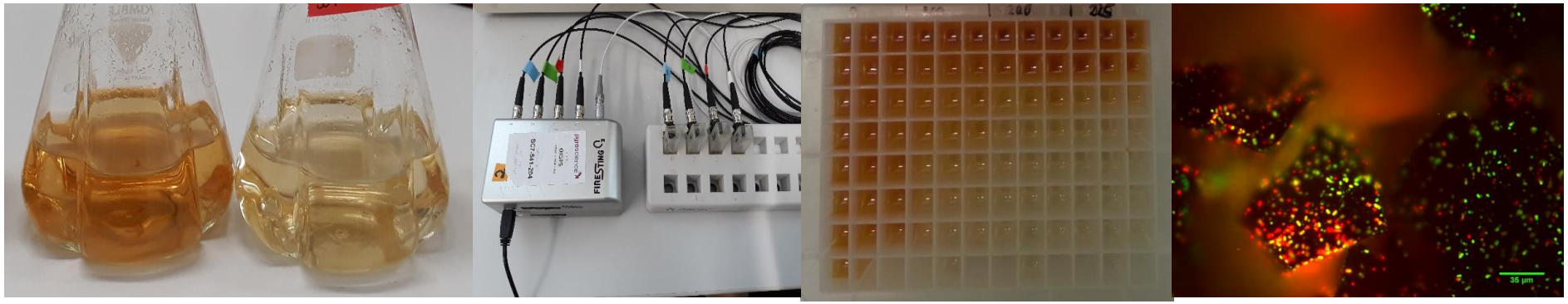


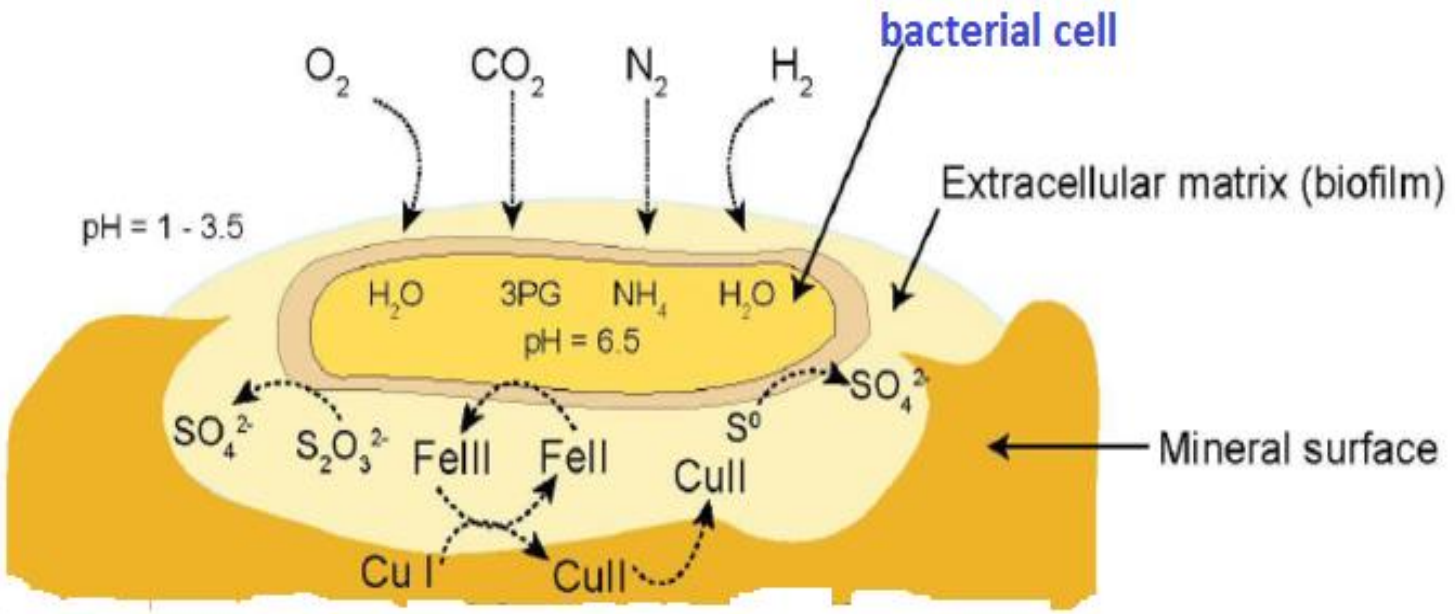
Effect of Sodium Chloride on Ferrous Iron Oxidation and Respiratory Rate by *Sulfobacillus thermosulfidooxidans*

Dieu Huynh, Stefan Kaschabek, and Micheal Schlömann

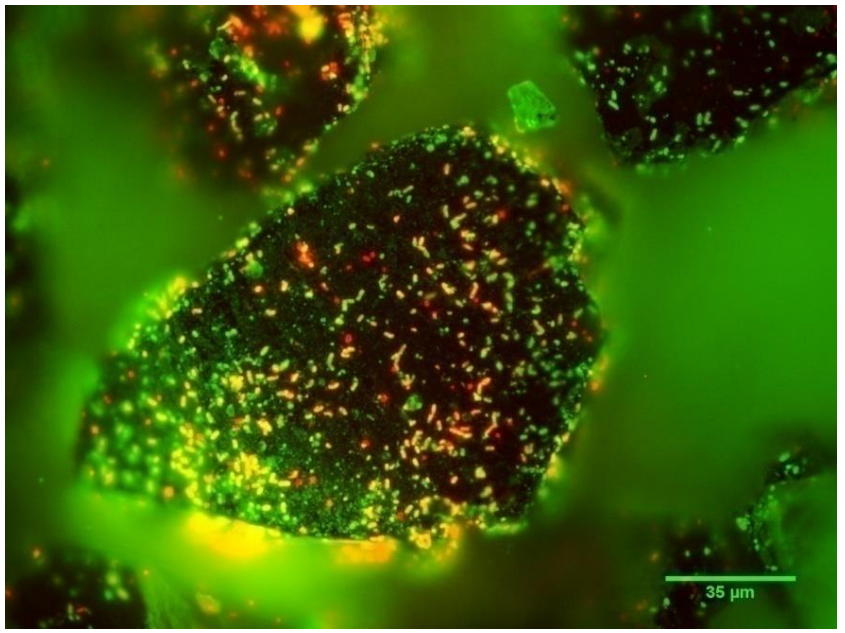
EGU, 09 April 2019 Vienna



An example of bioleaching



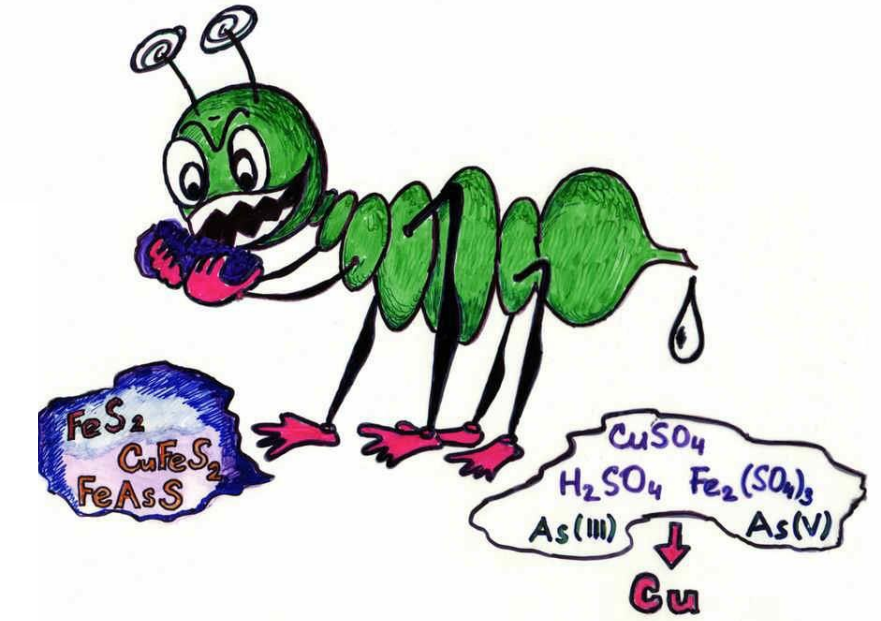
(*Modified from Valdés, J., et. al. 2008)*



(1) $CuFeS_2 + 4\,Fe^{3+} \longrightarrow Cu^{2+} + 5\,Fe^{2+} + 2\,S_0$ spontaneous

(2) $4\,Fe^{2+} + O_2 + 4\,H^+ \longrightarrow 4\,Fe^{3+} + 2\,H_2O$ (iron oxidizers)

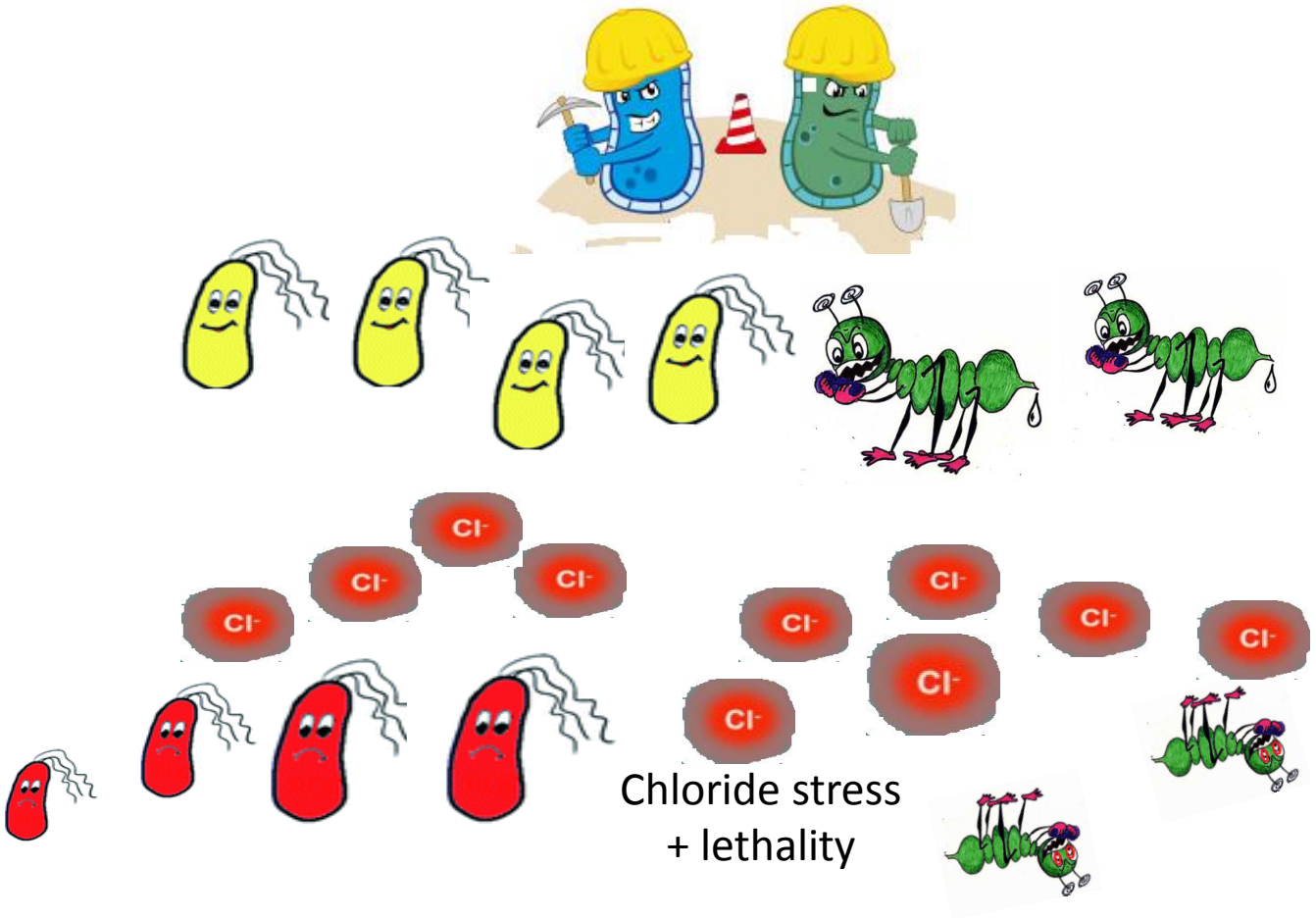
(3) $2\,S^0 + 3\,O_2 + 2\,H_2O \longrightarrow 2\,SO_4^{2-} + 4\,H^+$ (sulfur oxidizers)

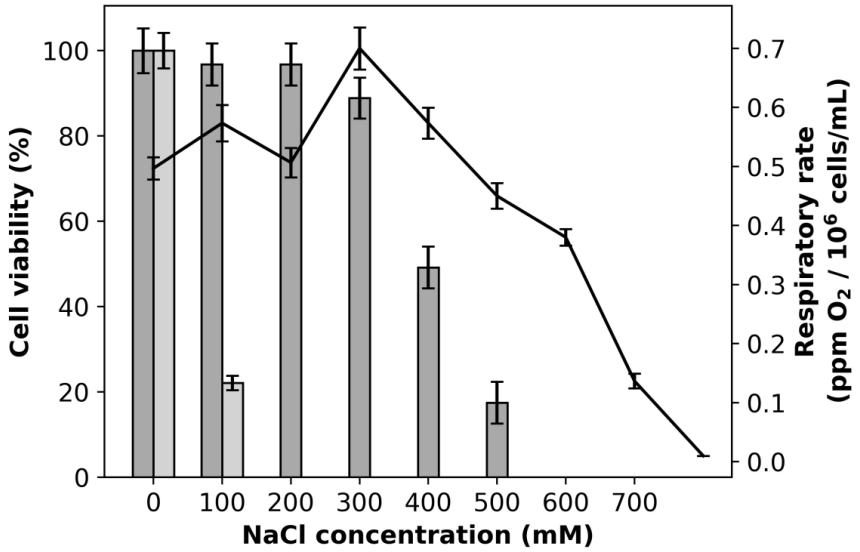


Chloride in bioleaching processes

→ Chloride ions enhance leaching of copper by increasing porosity of formed sulfur layer during chalcopyrite leaching

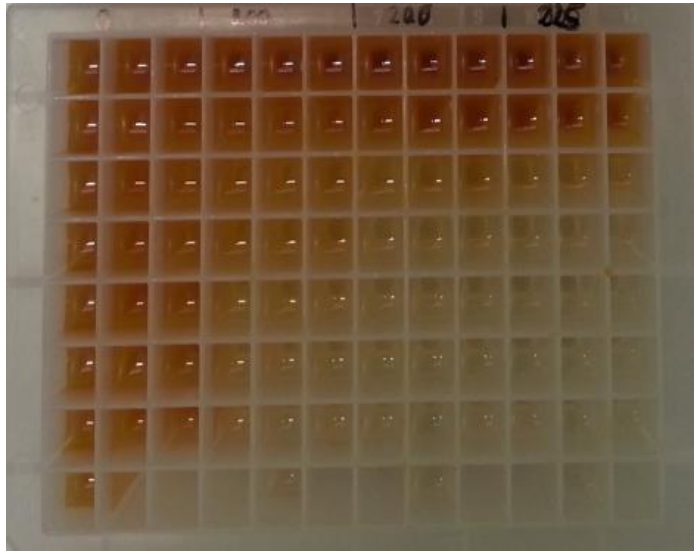
→ **Chloride ions are toxic to bacteria, inhibit growth and iron oxidation activity**

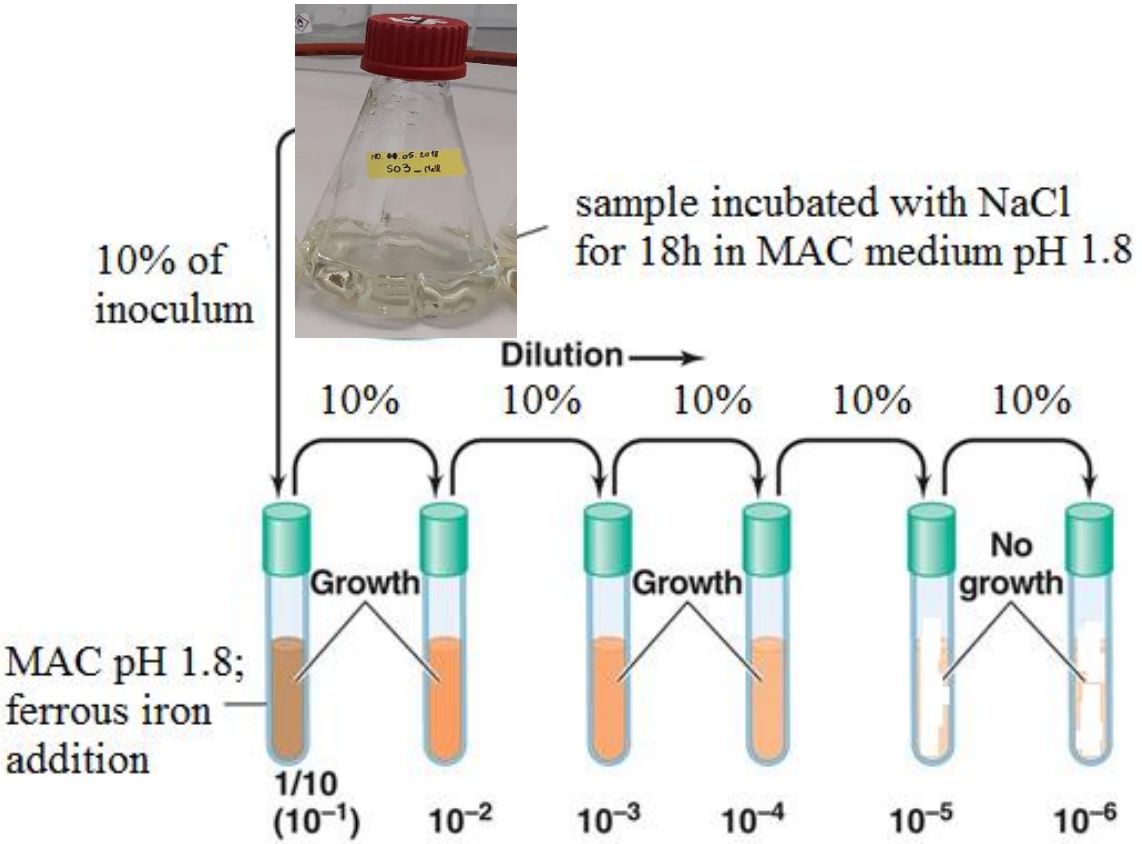




Materials and Methods

1. Determining the chloride effect on viability by means of the most probable number (MPN) analysis





10^{-7} 10^{-8} ……

Following 18 h exposure of $3*10^6$ cells/mL of *Sb. thermosulfidooxidans* to different NaCl levels, cells were grown in NaCl-free Mac medium.

MPN was determined with the *MPN calculation program*, v. 5 (Jarvis *et al.* 2010)

Materials and Methods

2. Determining the chloride effect bacterial growth, respiration rates and iron oxidation activity

NaCl (mM)	0	100	200	300	400	500	600	1000
Fe^{2+} (mM)	50							
Yeast extract (%)	0.02							
Inoculum (%)	10							
Temperature (°C)	45							

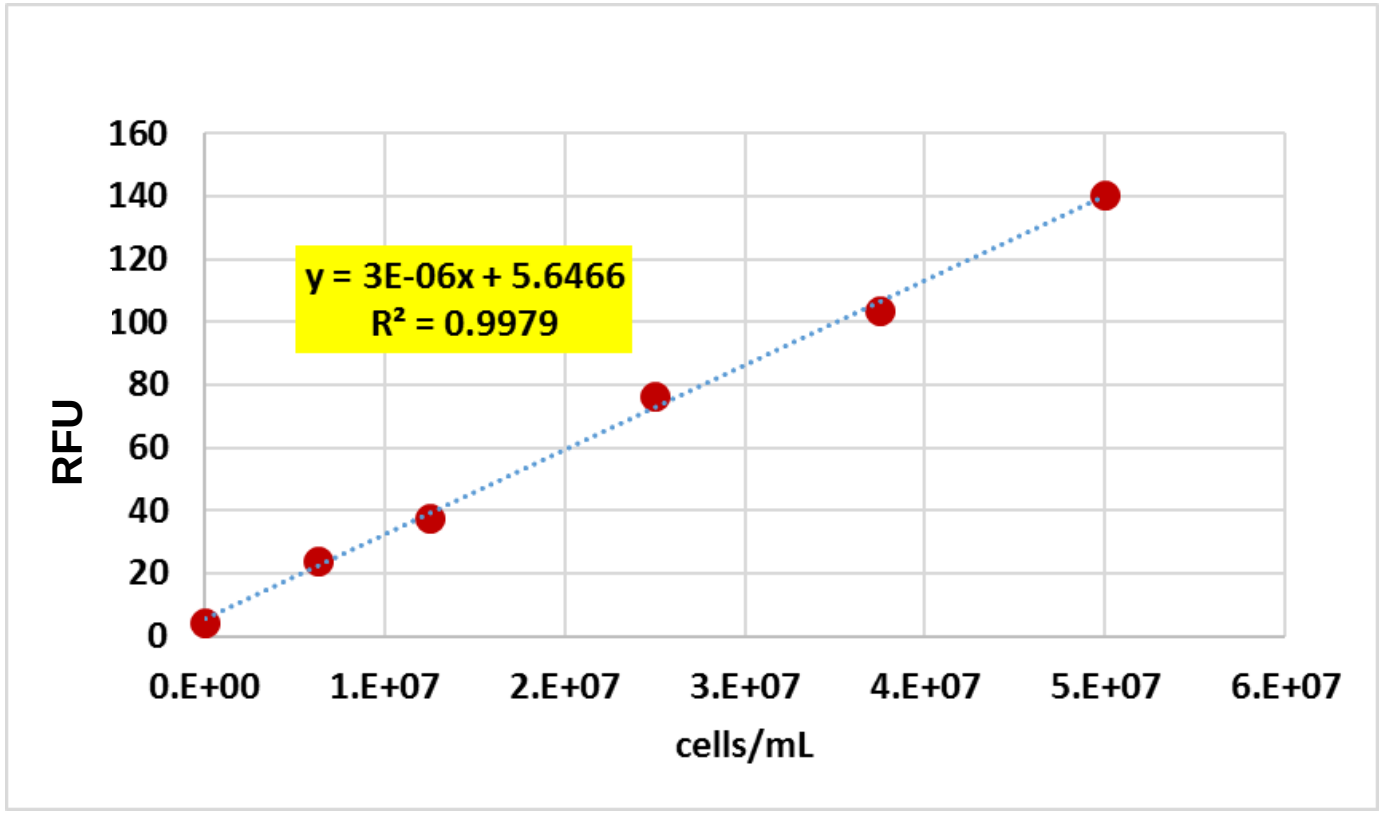
- Determination of cell growth
- Determination of microbial respiration rates
- Determination of iron oxidation

Materials and Methods

2.1. Determining the chloride effect on total cell number of *Sb. thermosulfidooxidans* using the PicoGreen assay *(Giebner et al. 2015)*

Principle: Staining a bacterial suspension (e.g. *Sb. thermosulfidooxidans*) with PicoGreen leads to a linear response between cell numbers and relative fluorescense units (RFU)

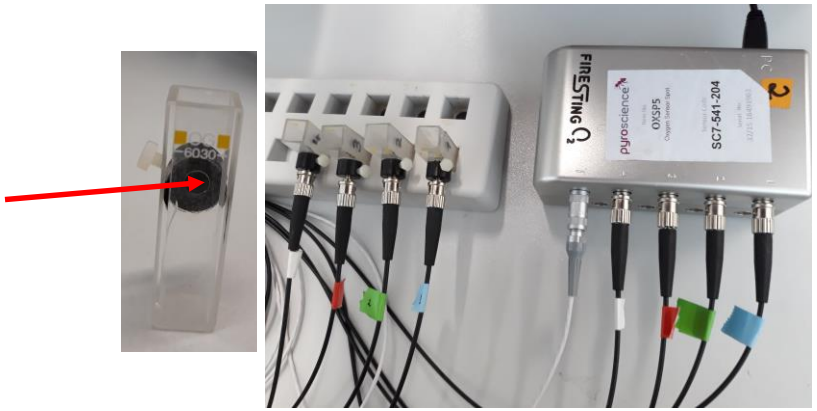
Correlation RFU and cell number of *Sb. thermosulfidooxidans*

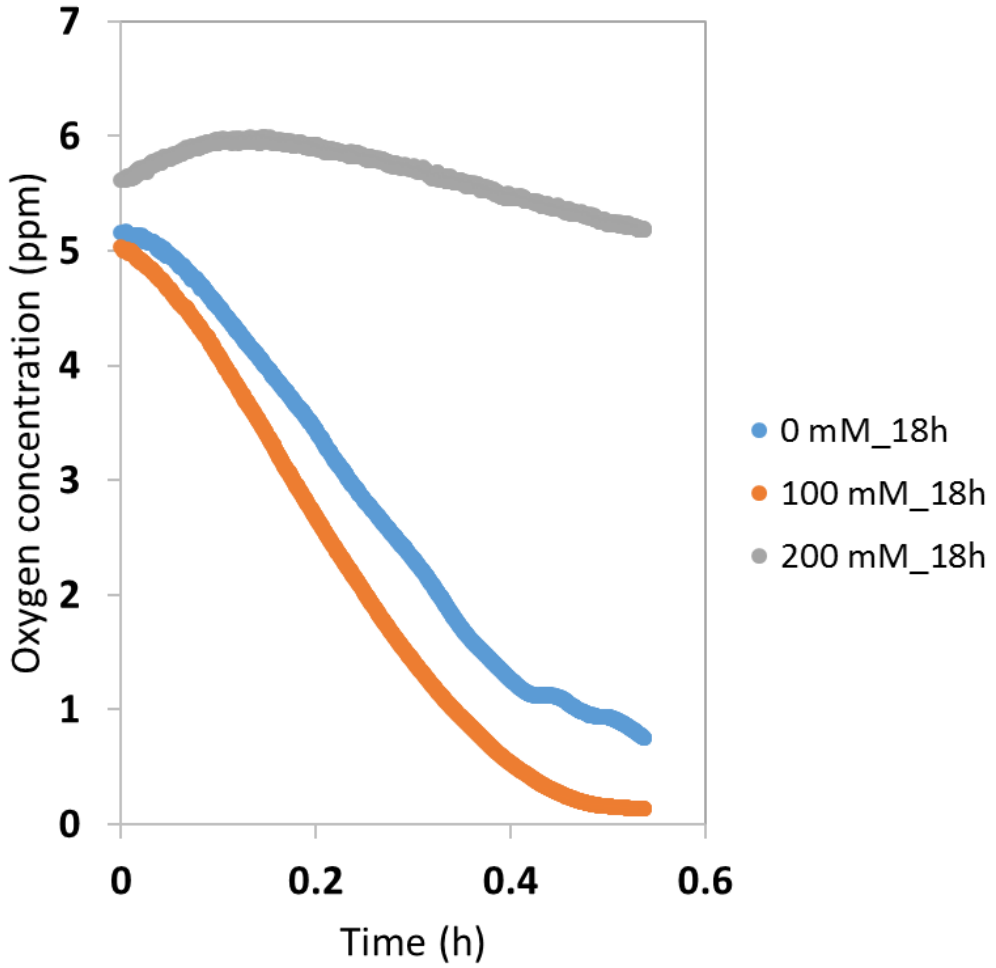


Materials and Methods

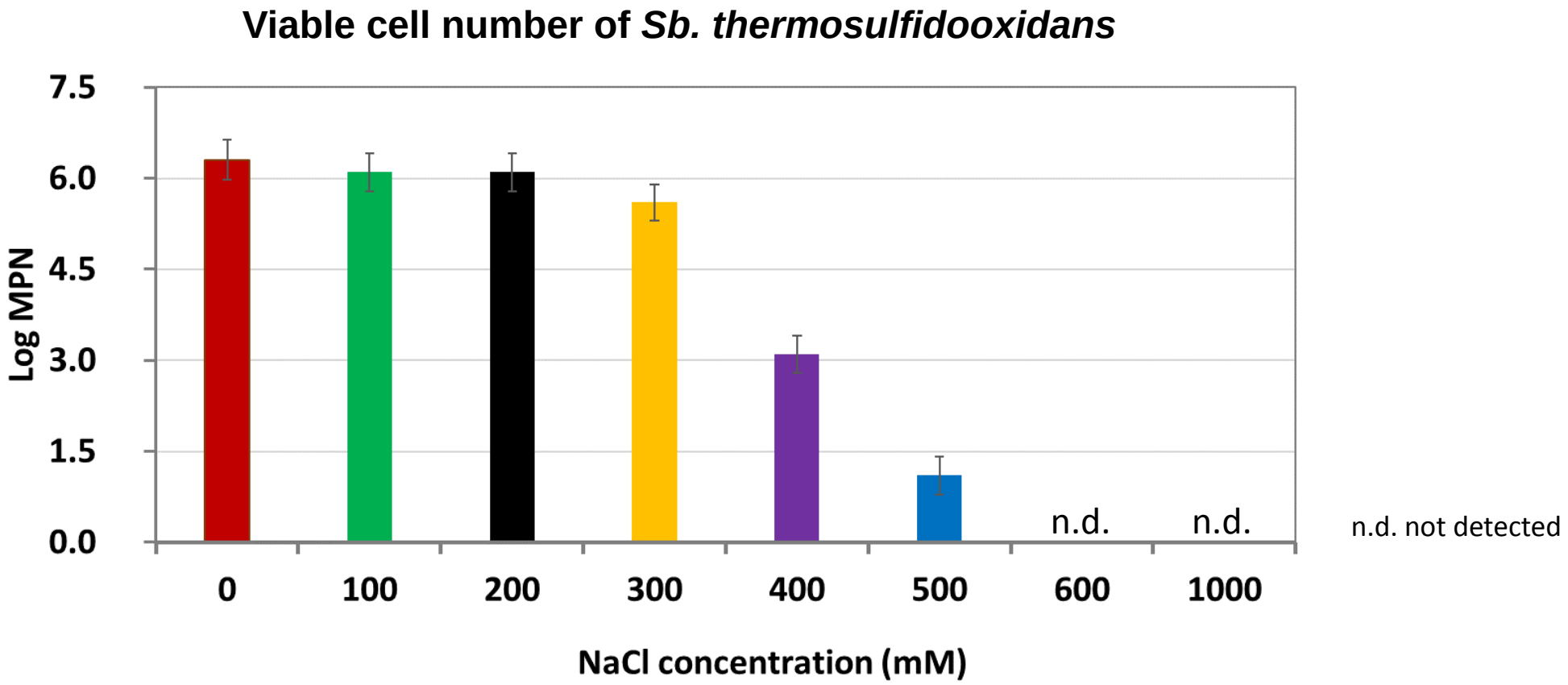
2.2. Determining the chloride effect on respiration rates of *Sb. thermosulfidooxidans* using Optode system *(Giebner et al. 2015)*

Aerobic respiration: $4Fe^{2+} + 4H^{+} + O_2 \rightarrow 4Fe^{3+} + 2H_2O$





Results



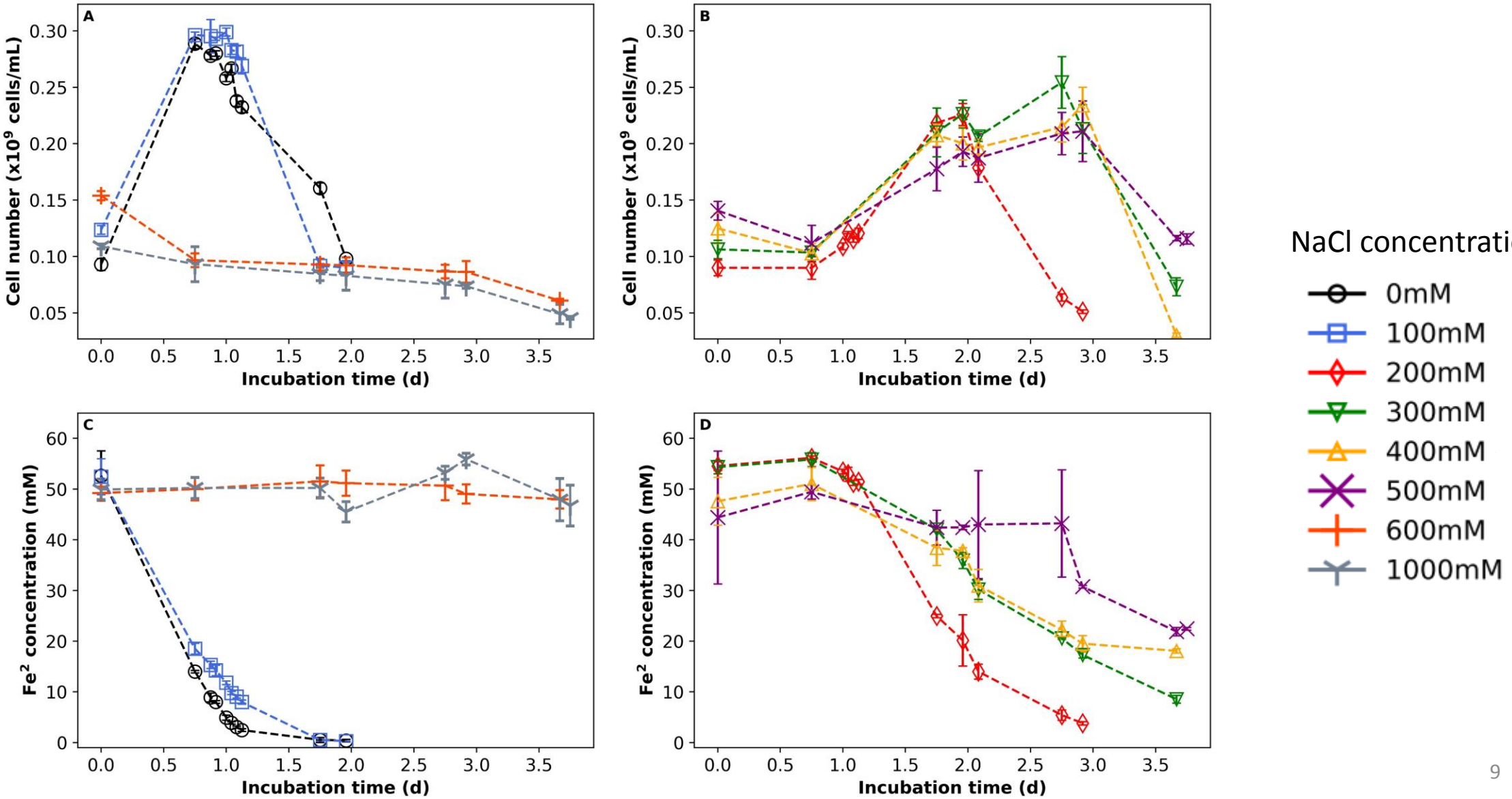
n.d. not detected

18 h exposure to up to 200 mM NaCl cause no significant change in viable cell number

Similar exposure to 400 mM NaCl cause a decline of viable cell number by more than 3 orders of magnitude

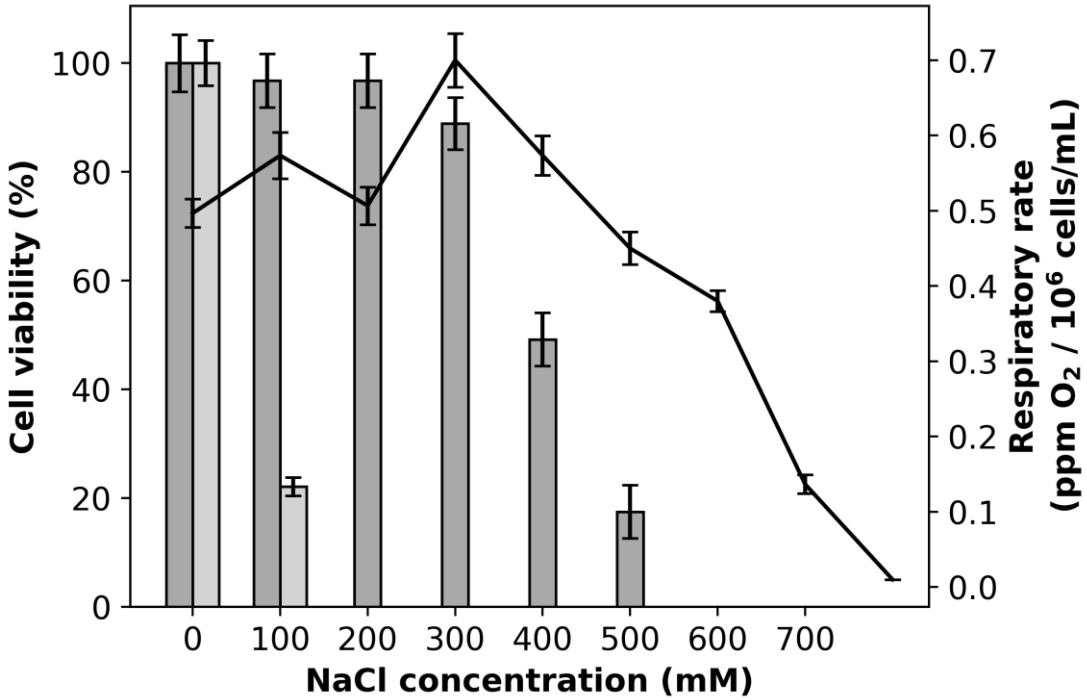
Results

Cell growth and iron oxidation of *Sb. thermosulfidooxidans* in the presence of different NaCl concentrations

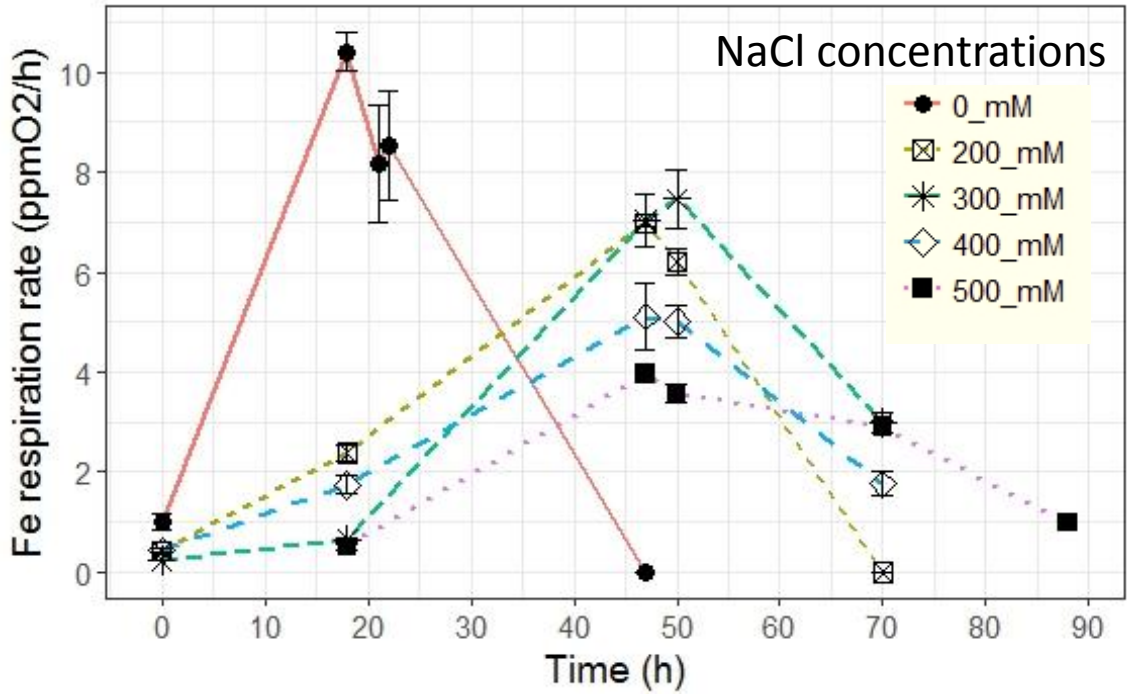


Results

Cell viability in comparison to *L. ferriphilum* and Fe respiration within 15 min exposure to NaCl



Fe respiration change during bacteria growth



Conclusion

- Exposure of cells to NaCl ≥ 200 mM caused an extension of lag-phase
- NaCl ≥ 300 mM strongly affected cell viability, growth and iron oxidation of *Sb. thermosulfidooxidans*

Outlook

- Effect of NaCl on sporulation of *Sb. thermosulfidooxidans*
- Effect of NaCl on *Sb. thermosulfidooxidans* in dependence of other substrates (sulfur, pyrite)
- Biofilm formation and Bioleaching of metal sulfides by *Sb. thermosulfidooxidans* in the presence of tolerated conc. of NaCl
- Metabolomics response of NaCl stress in *Sb. thermosulfidooxidans*