BIOCHEMISTRY OF CHEMOSYNTHETIC LIFE: MICROORGANISM BEHAVIOR AT DIFFERENT PRESSURES

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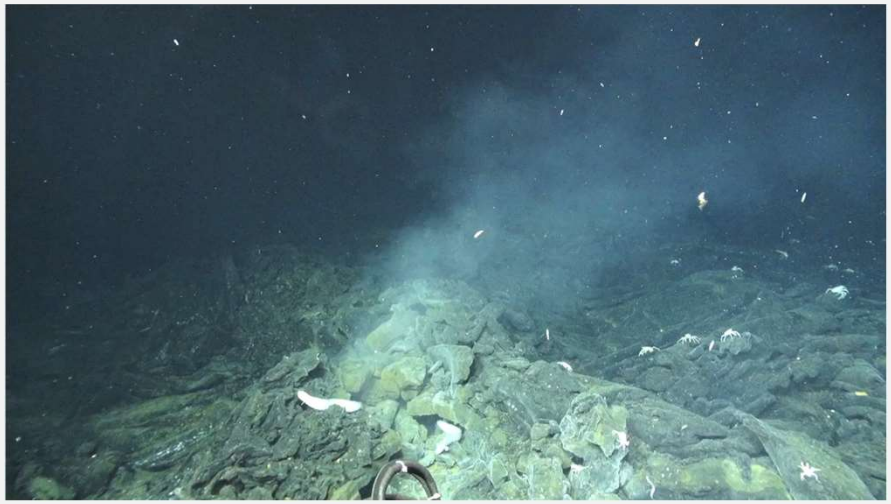
Summer 2022

CARNEGIE SCIENCE

MICROBES: EPSILON-PROTEOBACTERIA

Deep-sea hydrothermal systems

- Chemosynthesis as predominant source of energy

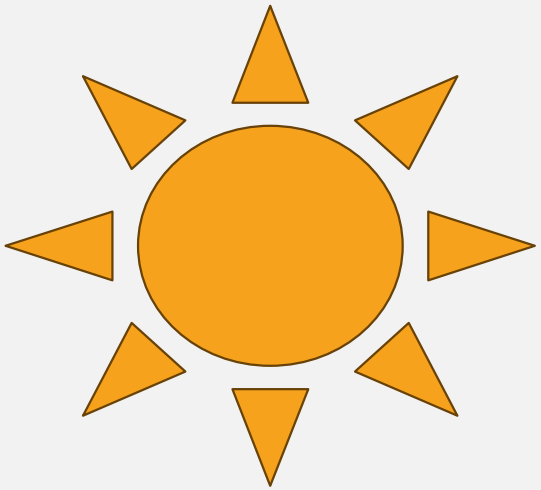


Foustoukos (2015), *BCO-DMO*

WHAT IS CHEMOSYNTHESIS?

Photosynthesis

Sunlight = energy source



Chemosynthesis

Inorganic elements = energy source

SO_4^{2-}

sulfate

CO_2

carbon dioxide

NO_3^-

nitrate

ANAEROBIC NITRATE REDUCTION

Pathways

- Denitrification
  - More common at lower temperatures

$$NO_3^- + H_2 \rightarrow N_2$$

(nitrate) (hydrogen) (nitrogen)

- **DRNA**
  - Dissimilatory reduction of nitrate to ammonium

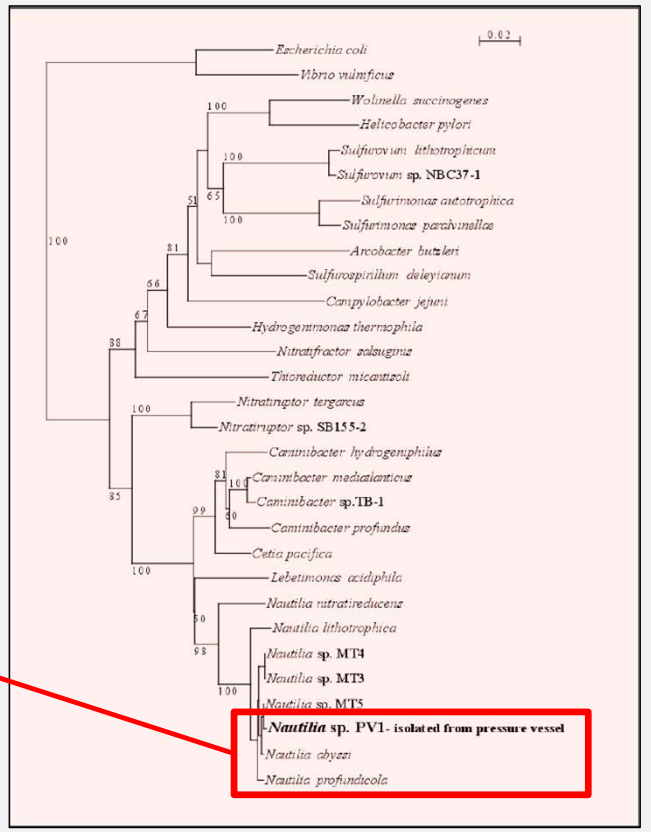
$$NO_3^- + H_2 \rightarrow NH_4^+$$

(nitrate) (hydrogen) (ammonium)

RESEARCH GOAL

I. If there is a physiological difference among genetically similar microbes
   - *Nautilia* species ***PV1*** and ***Abyssi***

Physiological difference: behavior at different **pressures**



PV1, abyssi, profundicola

Vetriani Lab (2020)

EXPERIMENT: STUDYING *N. ABYSSI*

Nautilia PV1: piezophilic

Piezophilic: grow optimally at high pressures

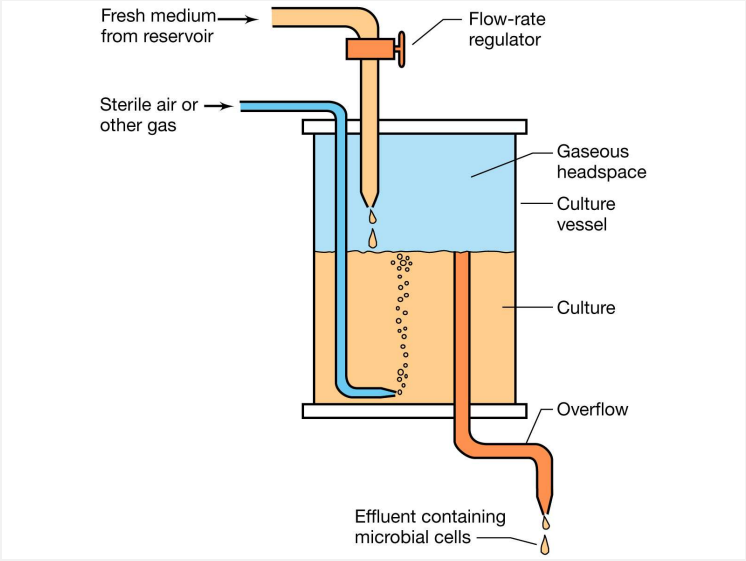
Nautilia Abyssi: ??

Must test at different pressures

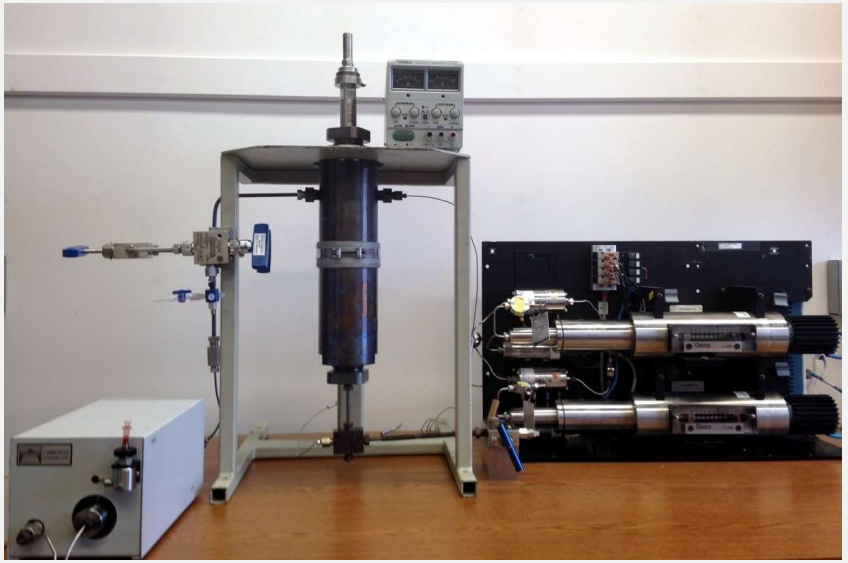
- Ambient: 5 bars
- High: 200 bars

GROWING *N. ABYSSI* SAMPLES

Rutgers University--New Brunswick



Sievert (WHOI)



Foustoukos and Perez-Rodriguez (2015), AEM
Foustoukos and Houghton (2022), in press

S. Fong

CHARACTERIZATION OF *N. ABYSSI*

Measurements

Nitrate Reduction Rates
Most efficient nitrate reduction
$[NO_3]$ and $[NH_4]$

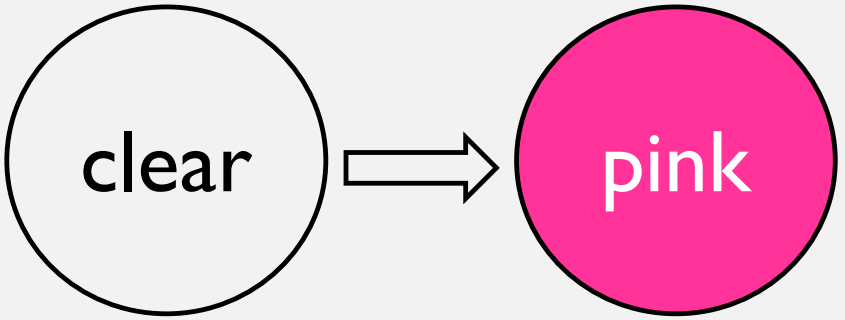
Kinetic Rate Constant
Speed of the nitrate reduction reaction
Integrated rate laws

Isotope analysis
Isotope signature of ^{15}N
Isotope-Ratio Mass Spectrometry

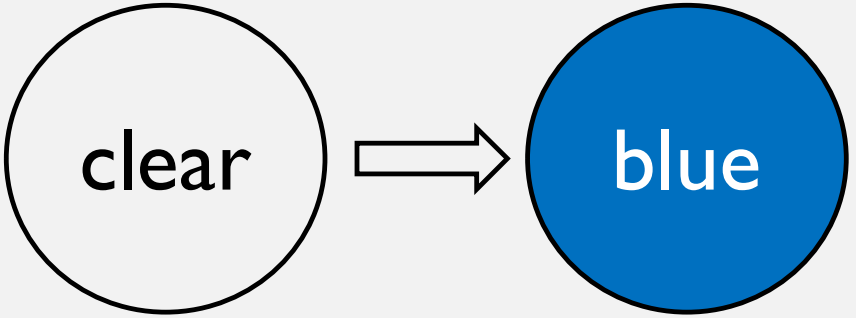
MEASURING REDUCTION RATE

Culture Assays

I. Cadmium reduction method: [NO_3]
   - Cadmium metal catalyst



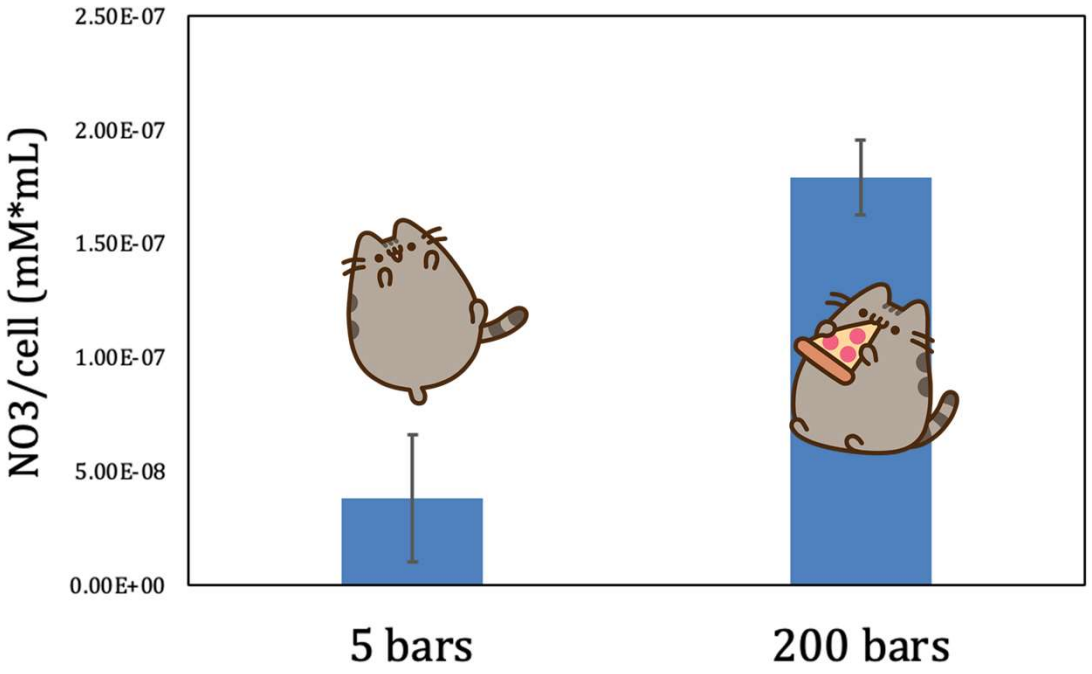
II. Berthelot reaction method: [NH_4]
   - Indophenol-blue indicator



CONSUMPTION OF NITRATE

Nitrate concentration per cell

High pressure requires more nitrate for cell growth



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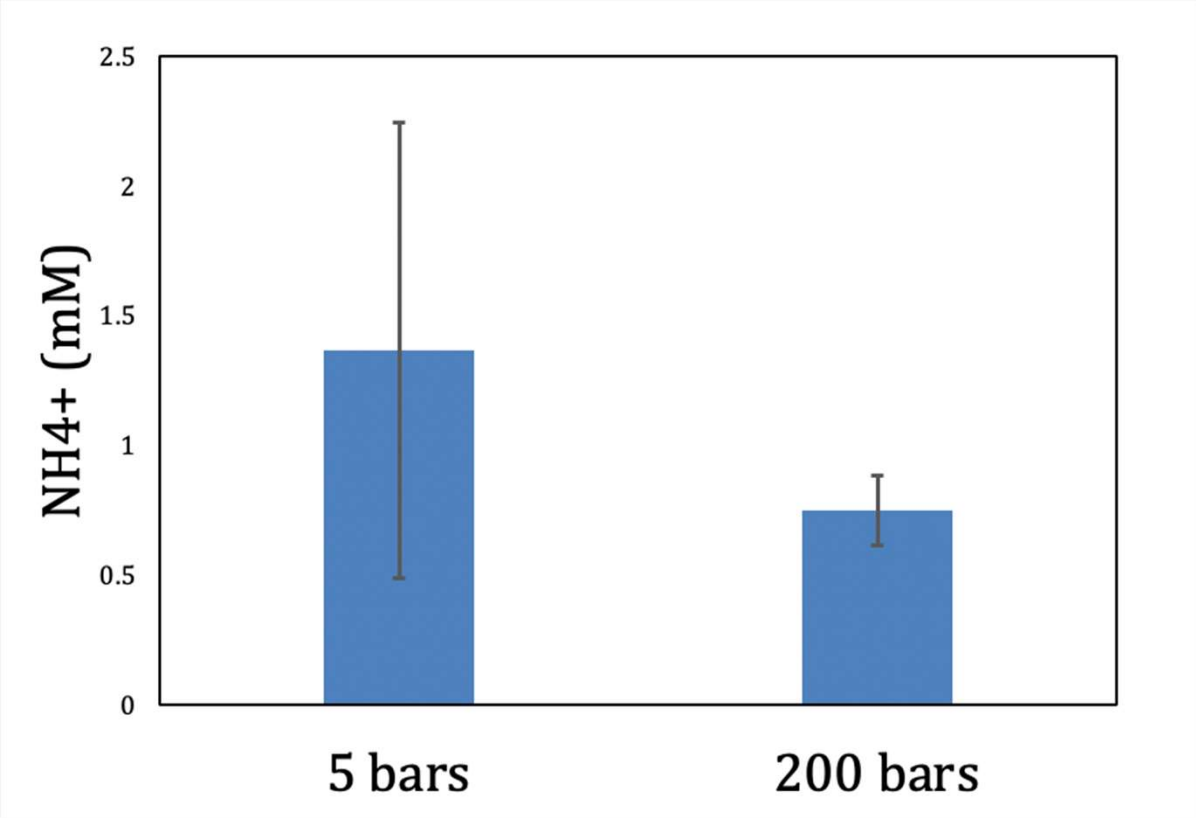
PRODUCTION OF AMMONIUM

Ammonium production

High pressure produces less waste as more energy goes towards survival



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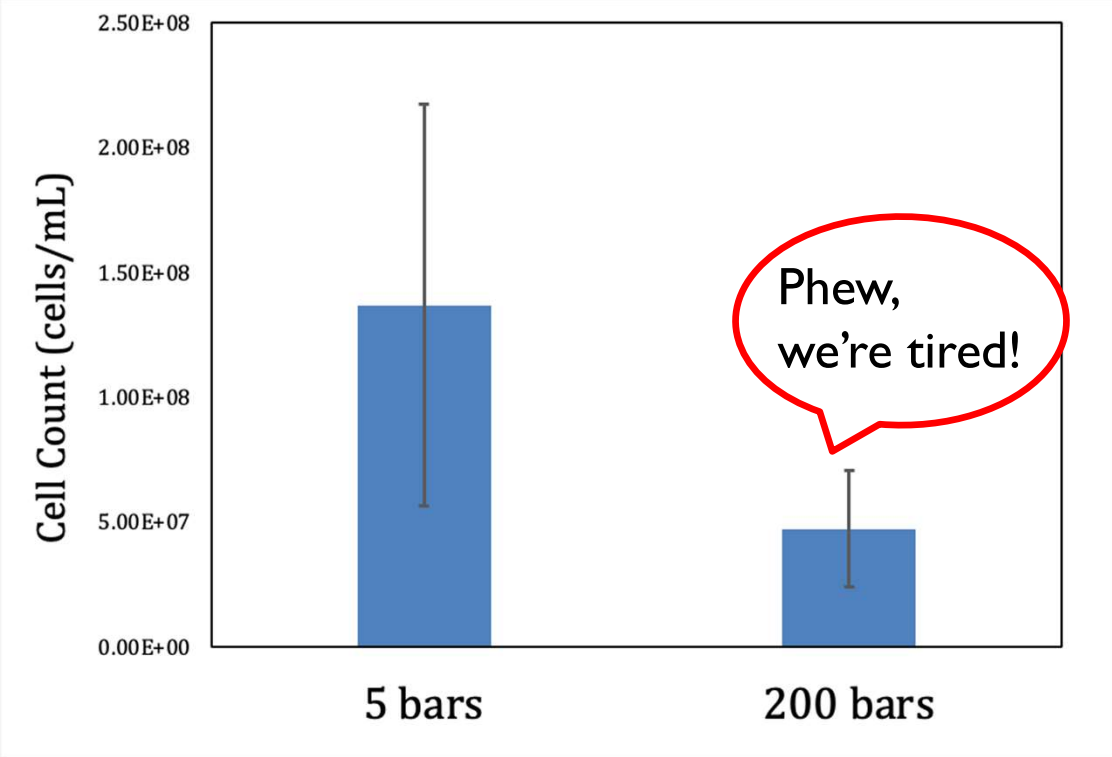
CULTURE CELL DENSITY

Total cell activity

More cells are active in nitrate reduction at ambient pressure



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OPTIMAL PRESSURE FOR *N. ABYSSI*

Ambient pressure!

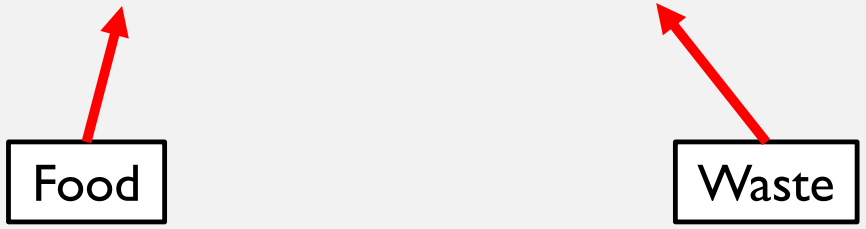
- More efficient nitrate reduction
- Higher cell density

Nautilia Abyssi prefers ambient pressure

DRNA Reaction

$$NO_3^- + H_2 \rightarrow NH_4^+$$

(nitrate) (hydrogen) (ammonium)



CALCULATING KINETIC RATE CONSTANT

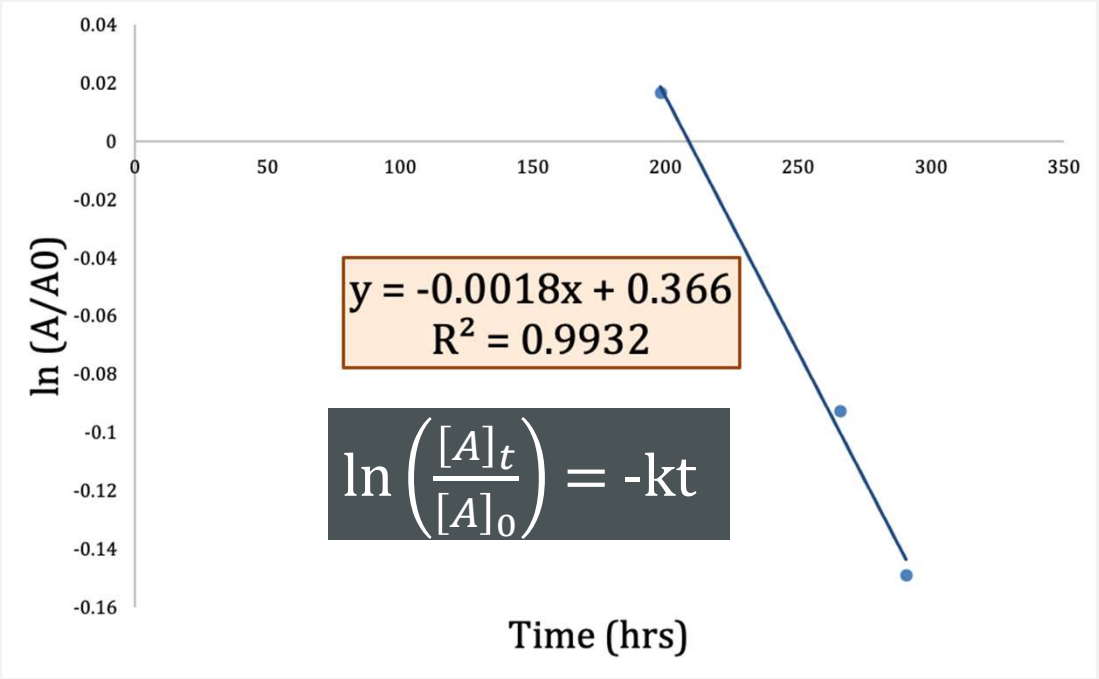
May 20-May 24

Kinetic rate constants

- Pseudo-first order reaction
- Integrated rate laws

Average rate constant of *N. Abyssi* = 0.0018

Ambient Pressure (5 bars)



Fong (2022)

COMPARING TO OTHER SPECIES

N. Abyssi rate constant = 0.0018

	Kinetic rate constant range
ε-proteobacteria (DRNA)	0.004-0.22
ε-proteobacteria (Denitrification)	0.01-0.44

Rate constant is within the range of similar species

Perez-Rodriguez, Foustoukos, et al. (2017), *GCA*

ISOTOPE ANALYSIS

Pending!

Potential contamination sources:

- At the source
- In the lab

WHY DOES THIS MATTER?

Microbes: insight to the evolution of life

Contribution to other fields

- Oceanography, genetics, microbiology, biogeochemistry

Climate policy

- Nitrogen cycyling

CONCLUSIONS AND FUTURE WORK

I. *Nautilia Abyssi* is piezotolerant
   - Physiological difference from *PV1* (piezophilic)

Next steps

- Complete isotope analysis
- Characterization of *Nautilia Profundicola*