

Life on the edge: Modeling thermophile habitability in deep-sea hydrothermal vents



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Acknowledgments



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David Butterfield (Univ. of Washington/NOAA)

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Julie Huber (Marine Biological Laboratory)

Darby Dyar (Mount Holyoke College)

John Jamieson (Univ. of Ottawa)

Mark Hannington (Univ. of Ottawa)

Pilots and crew of DSV *Alvin* and R/V *Atlantis*



Biological Oceanography
RIDGE 2000 Program



Pacific Marine Environmental Laboratory
NOAA Vents Program



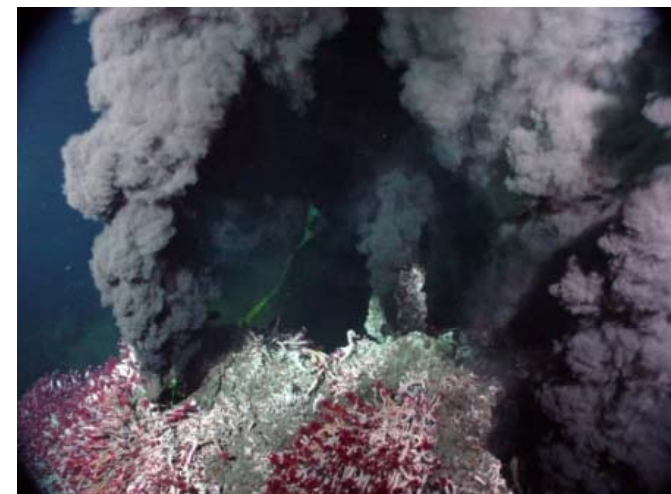
NASA Earth and Space Science
Graduate Fellowship (NESSF)

Strategy for assessing habitability and biogeochemical impact

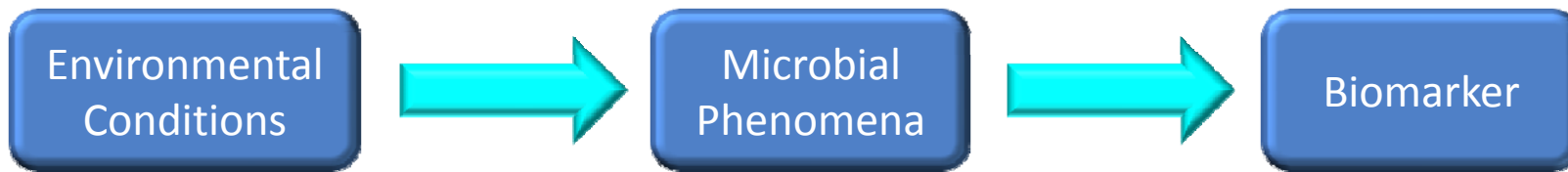
Astrobiology mantra:

1. Follow the water
2. Follow the energy
3. Follow the nutrients

****Also true for understanding past and present life in terrestrial environments**



How is this actually done?

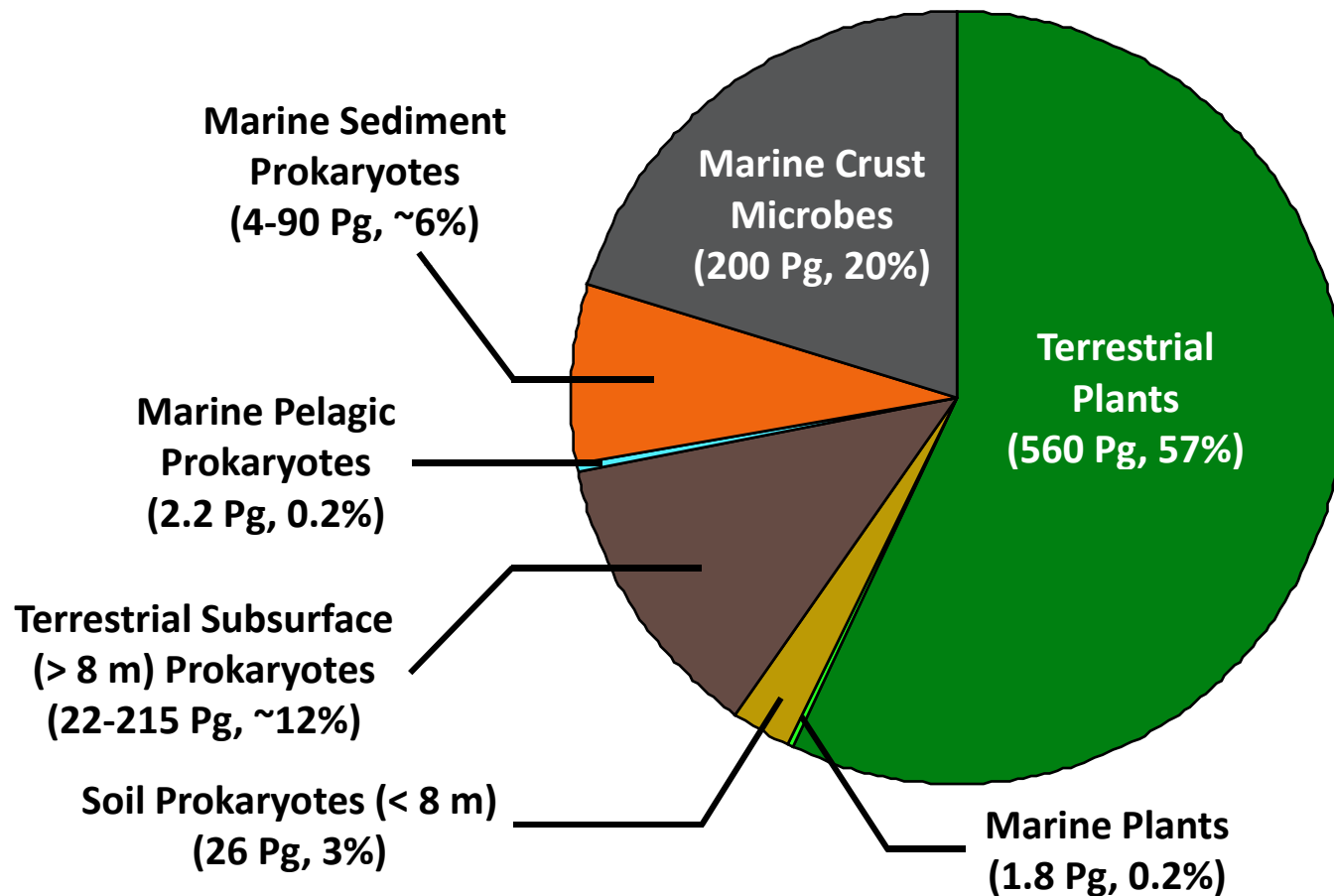


- physical and chemical constraints on life
- rates of biogeochemical transformations
- microbial bioenergetics
- microbe-mineral interactions
- population structure and dynamics
- functional diversity

- mineral, organic and isotopic biosignatures
- differentiate biotic and abiotic markers
- depositional characteristics of markers
- effect of material properties on mineral bioalteration

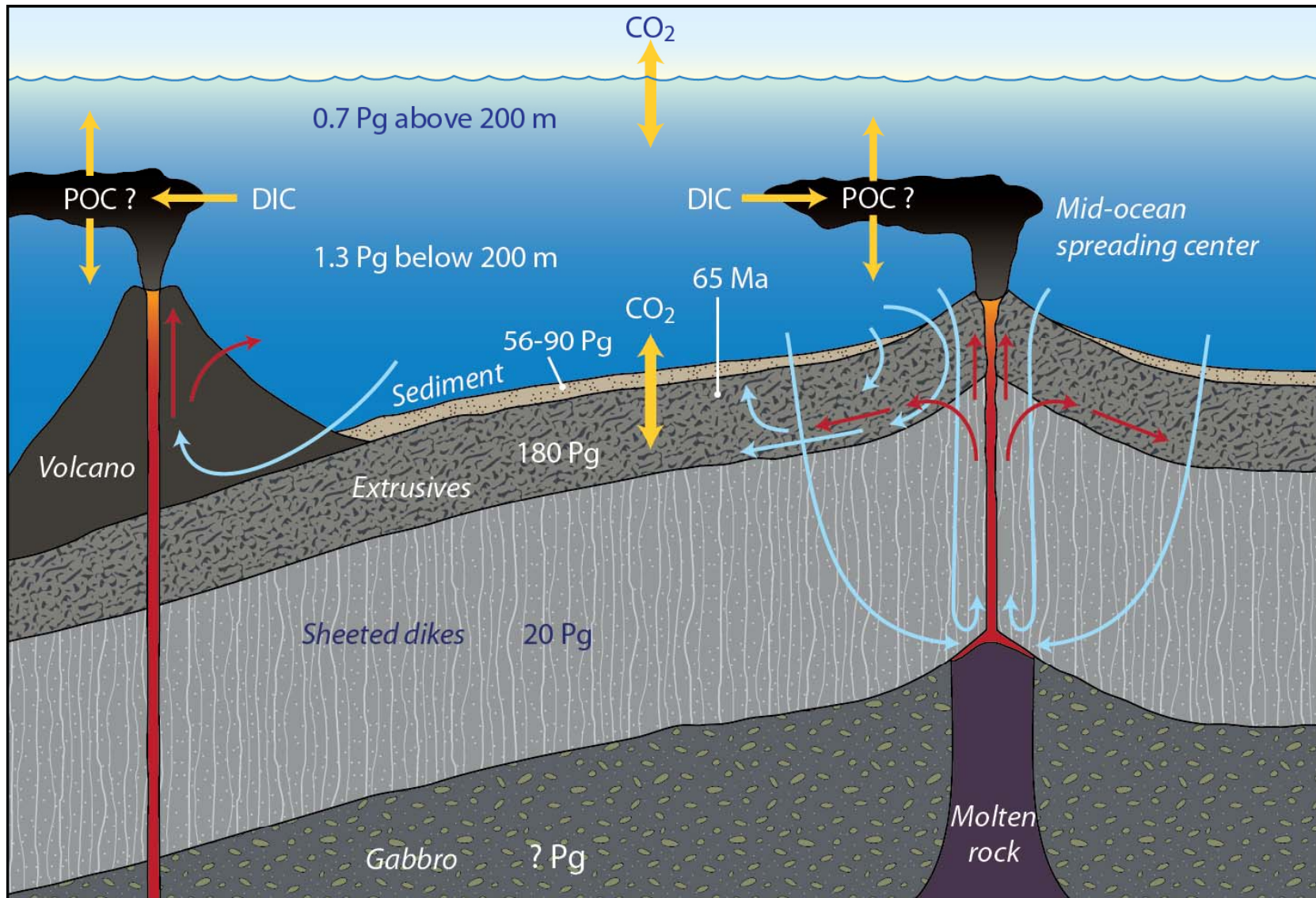
Global plant and prokaryotic biomass

Total global carbon is estimated to be 965 Pg (or 965×10^{15} g)



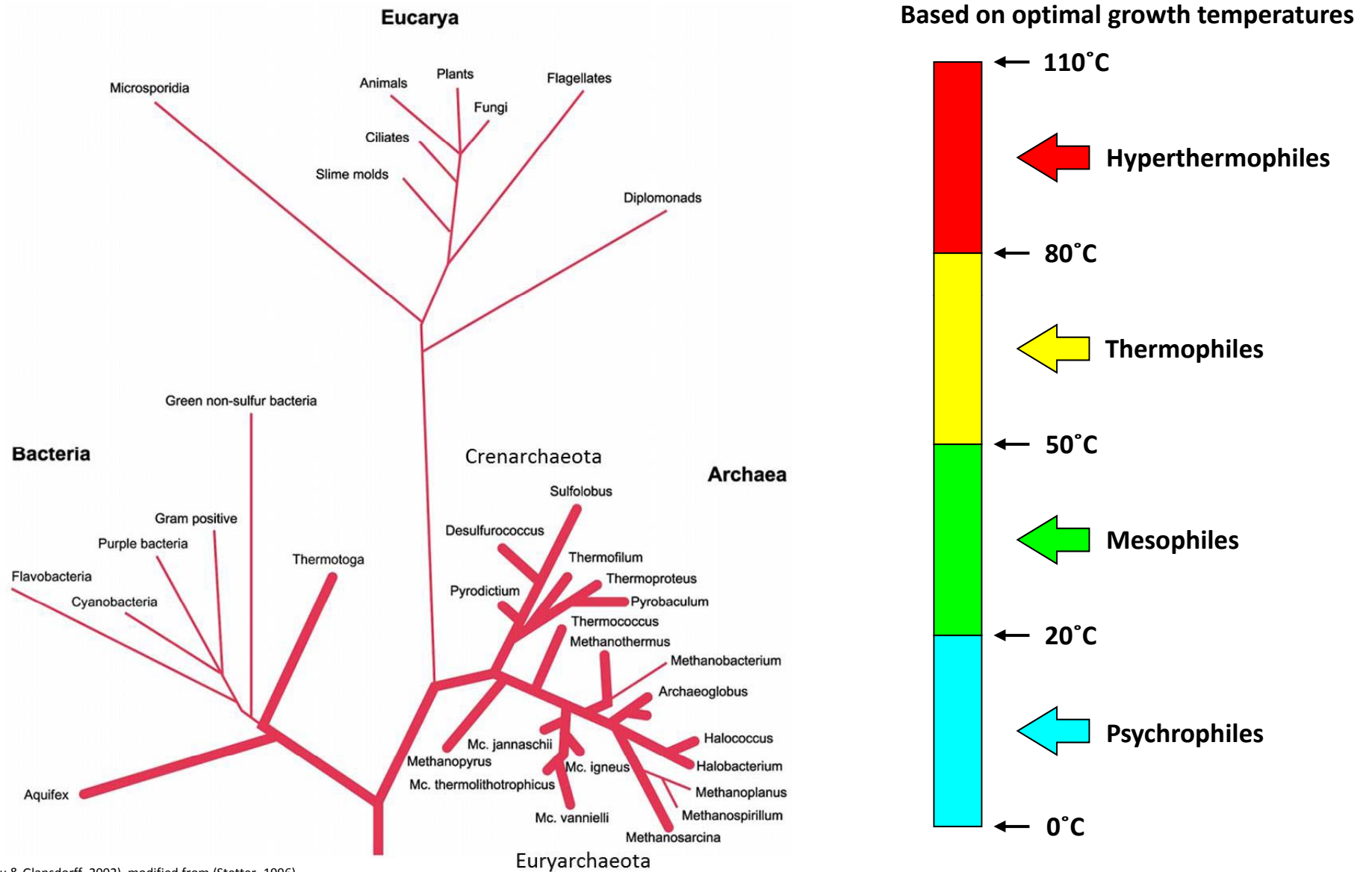
From Whitman et al. (1998), Parkes et al. (1994), Lipp et al. (2008), Heberling et al. (2010), Kallmeyer et al. (2012)

Life in the ocean crust



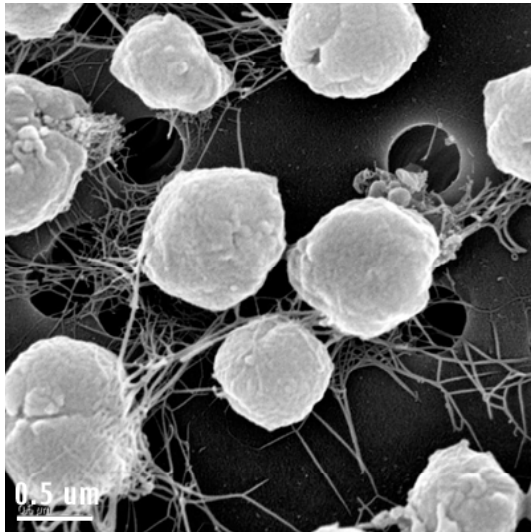


Hyperthermophile background



(Xu & Glansdorff, 2002), modified from (Stetter, 1996)

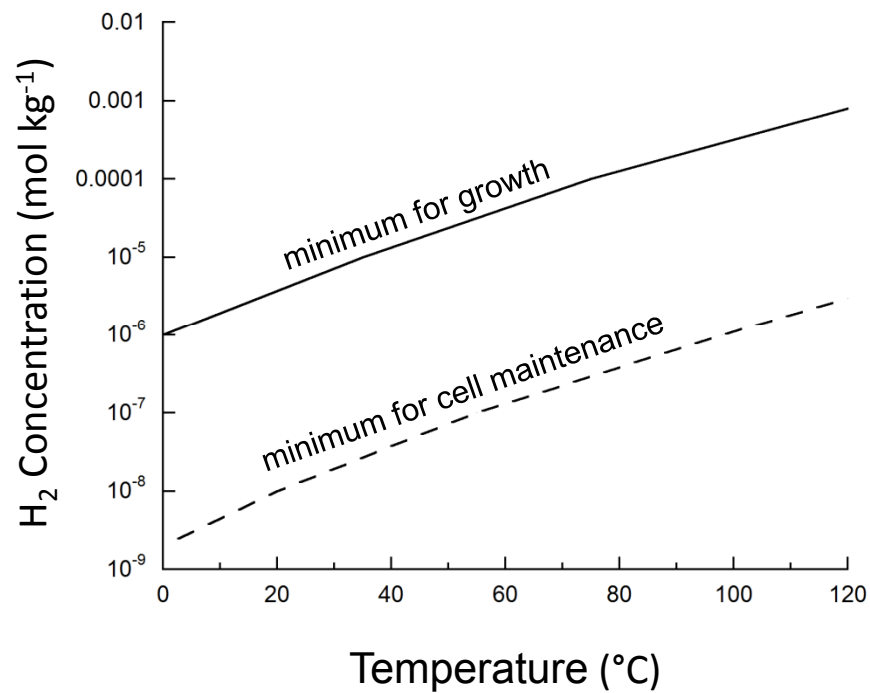
Hydrogenotrophic methanogens



- $4 \text{ H}_2 + \text{CO}_2 \rightarrow \text{CH}_4 + 2 \text{ H}_2\text{O}$
- Found in all thermal classes of organisms
- They are common in deep-sea hydrothermal vents
- They are not found in all vents
 - present in high H_2 vent systems (e.g., serpentinization, recent eruptions)
 - generally absent in low H_2 vent systems

“Energy balance concept” for modeling methanogenesis

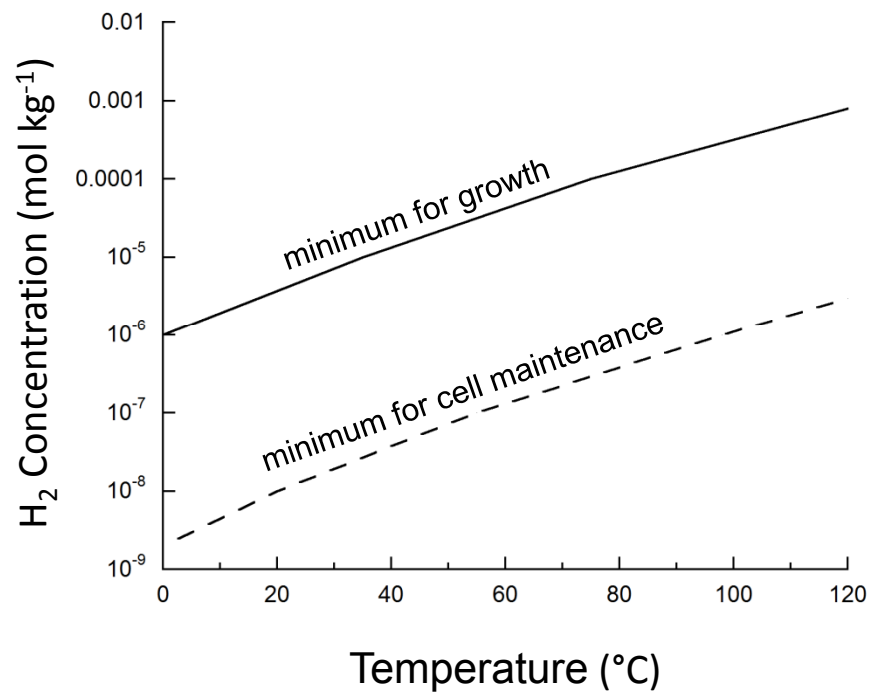
$$\{H_2\}_{\text{BEQ}} = \left(\frac{\{CH_4\}}{\{CO_2\}} \cdot \exp\left(\frac{\Delta G^\circ_{(T,P)} - \Delta G_{\text{BEQ}}}{RT}\right) \right)^{1/4}$$



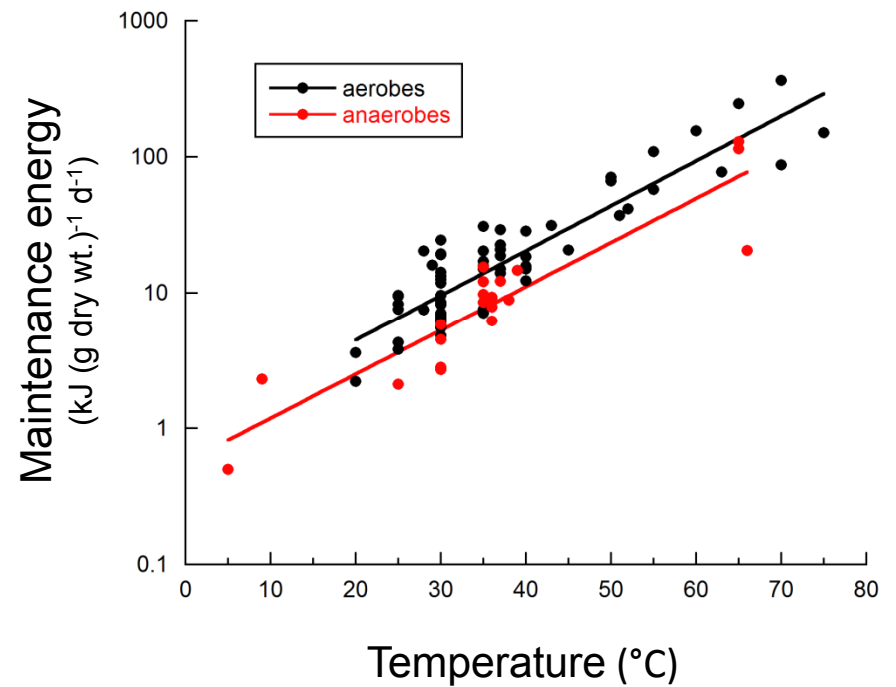
Hoehler (2004)

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Hoehler (2004)



Tijhuis et al. (1993)

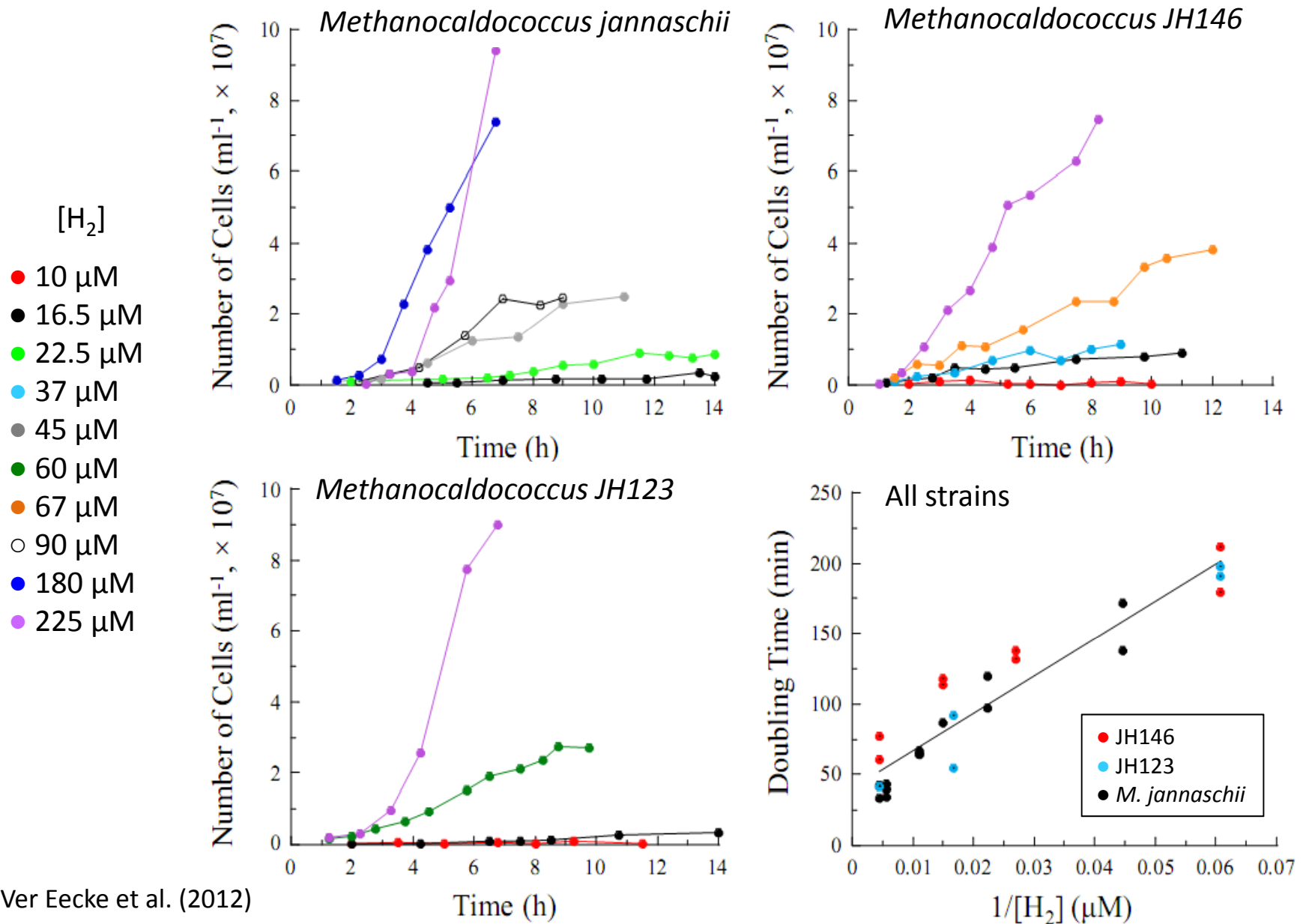
Research questions

- 1a. What is the minimum H₂ concentration for the growth of hyperthermophilic methanogens (70-82°C)? Is it uniform across species?
- 1b. Does field data support this estimate *in situ*?

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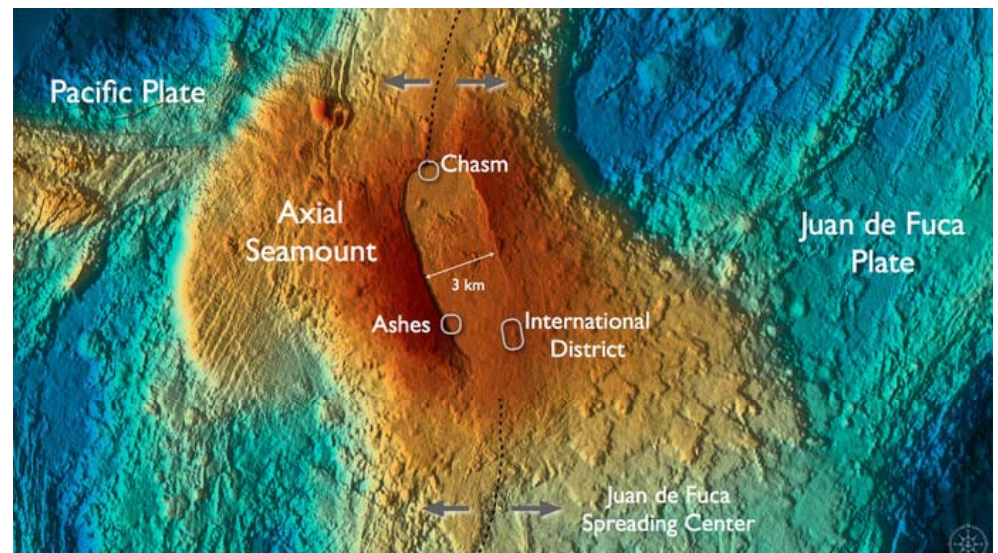
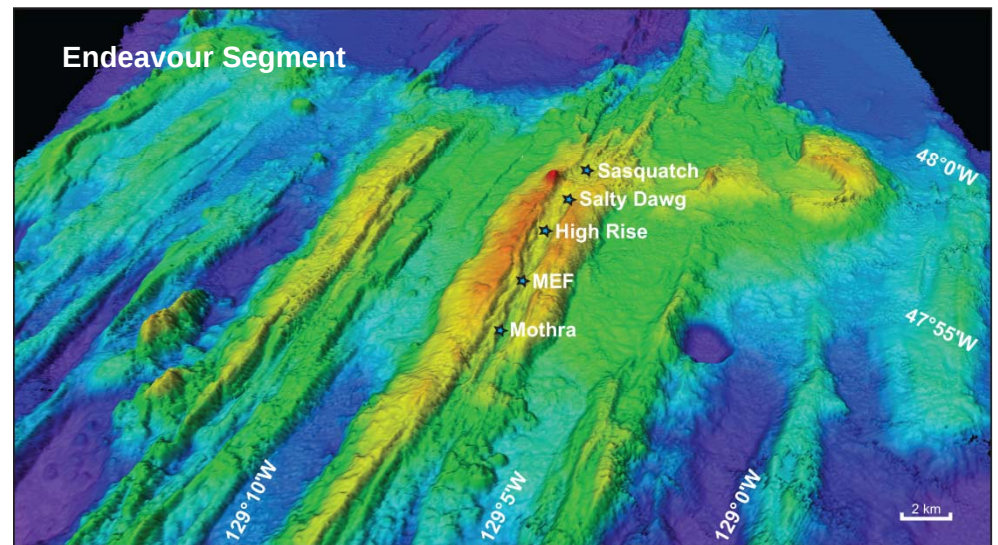
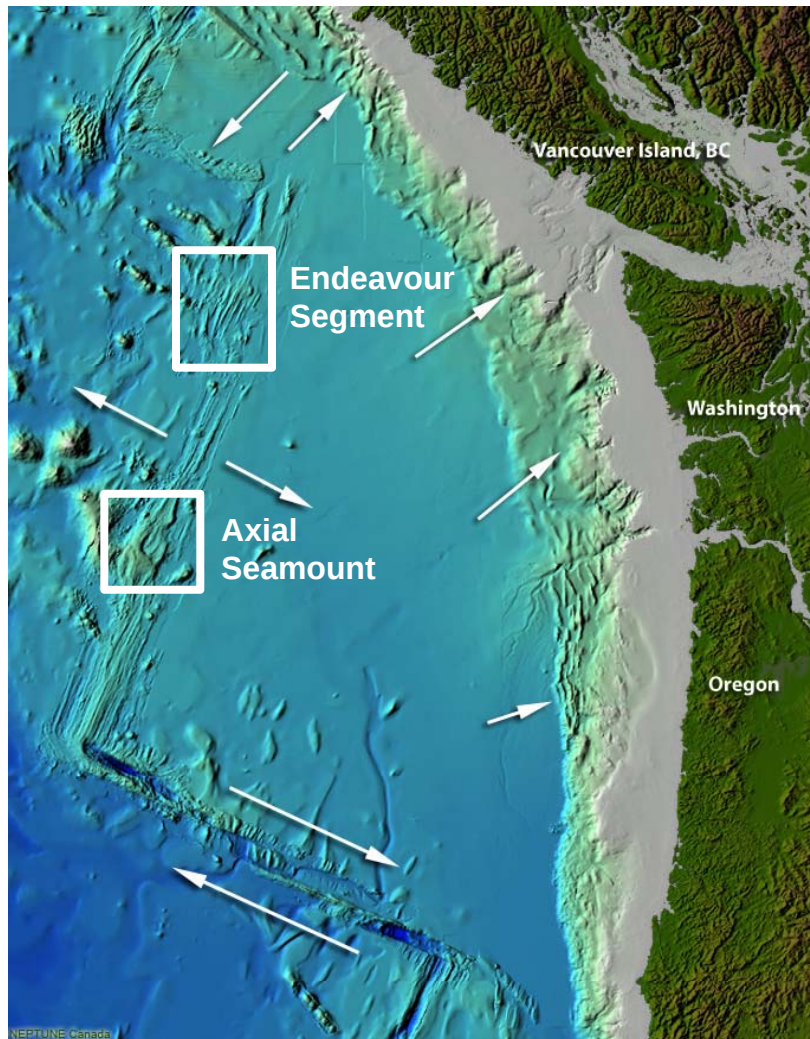
- 3a. Are usable minerals available in vents?
- 3b. Can hyperthermophiles use minerals as a growth substrate?
- 3c. Do these organisms leave behind a mineral biosignature?

Methanocaldococcus growth under varying [H₂]

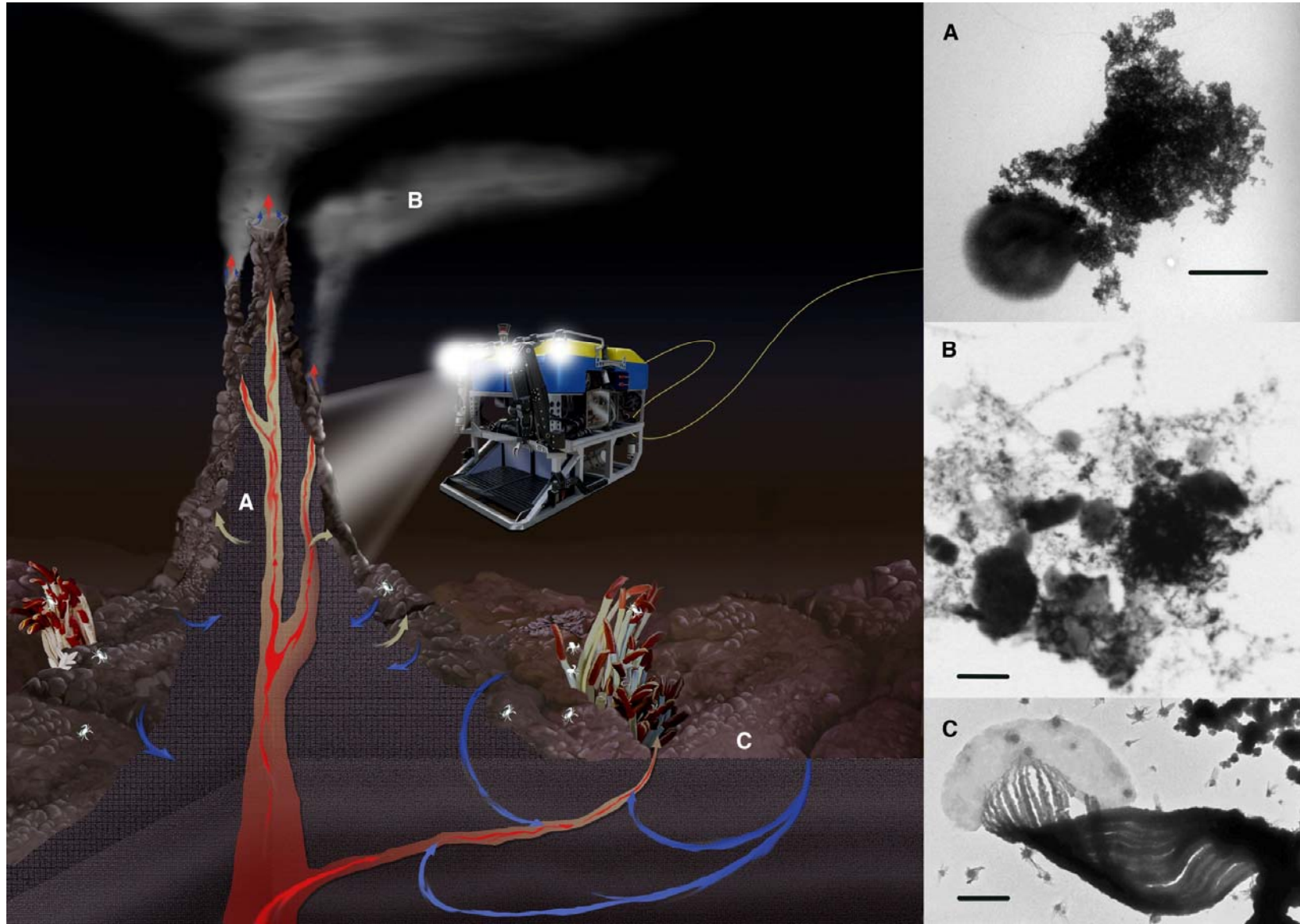


Ver Eecke et al. (2012)

Hydrothermal vents on the Juan de Fuca Ridge

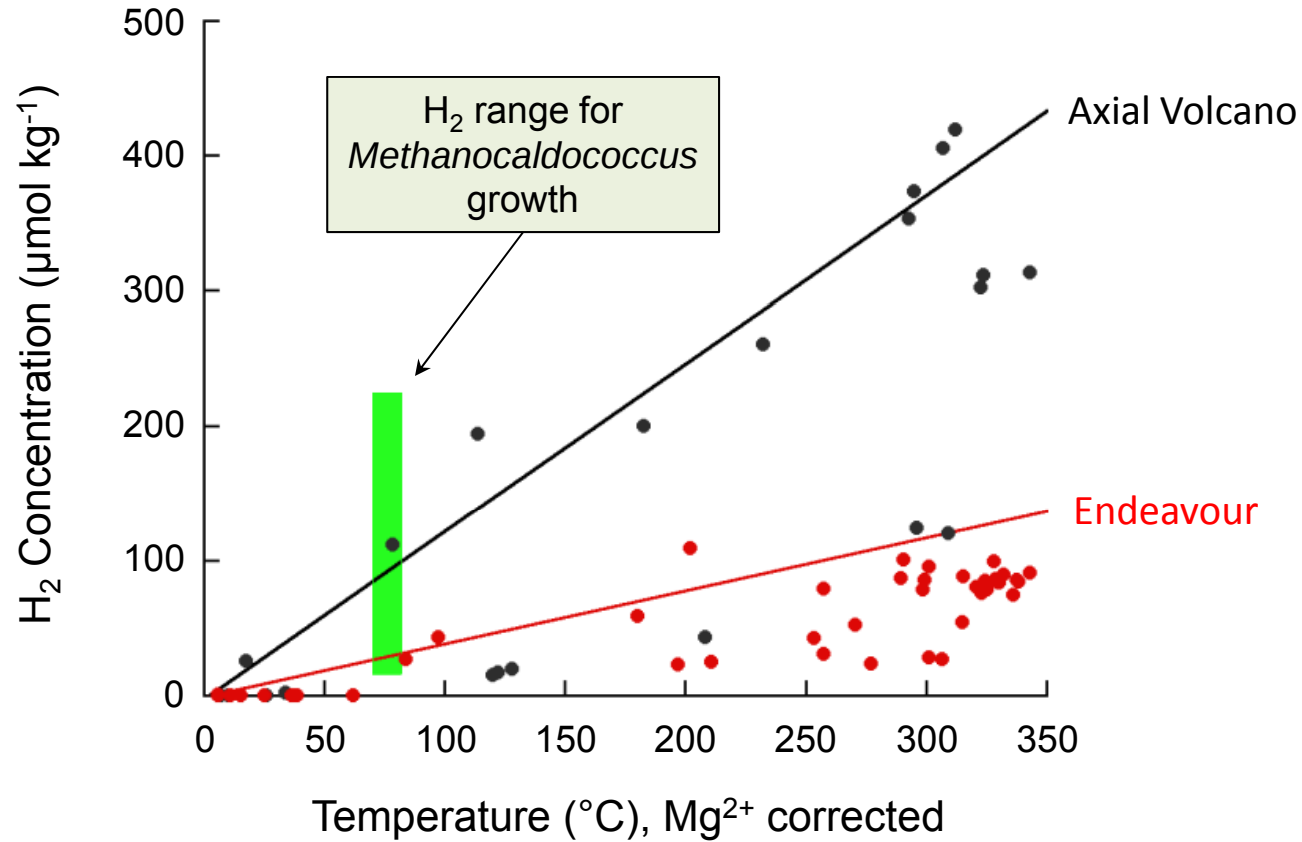


Sampling at deep-sea hydrothermal vents

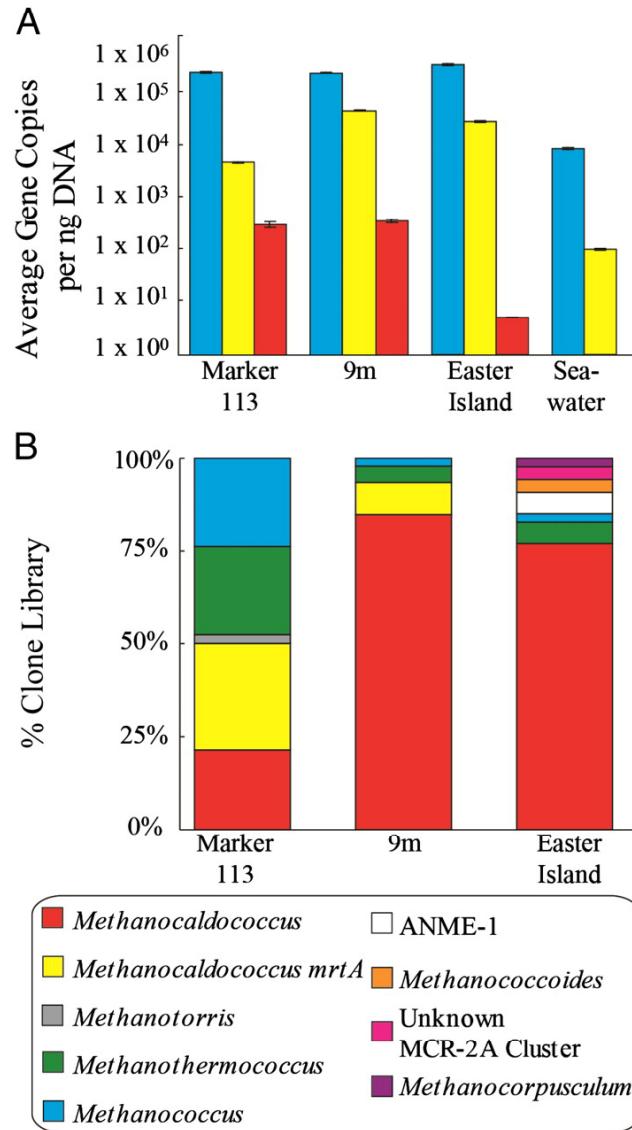


Holden et al. (2012)

Vent field H₂ fluid chemistry



Molecular analyses for methanogens in diffuse fluids



Axial Seamount:

- 6 fluids tested
- 2 showed evidence of methanogens
- one site was Marker 113 vent

Endeavour Segment:

- 11 fluids tested
- 1 showed evidence of methanogens

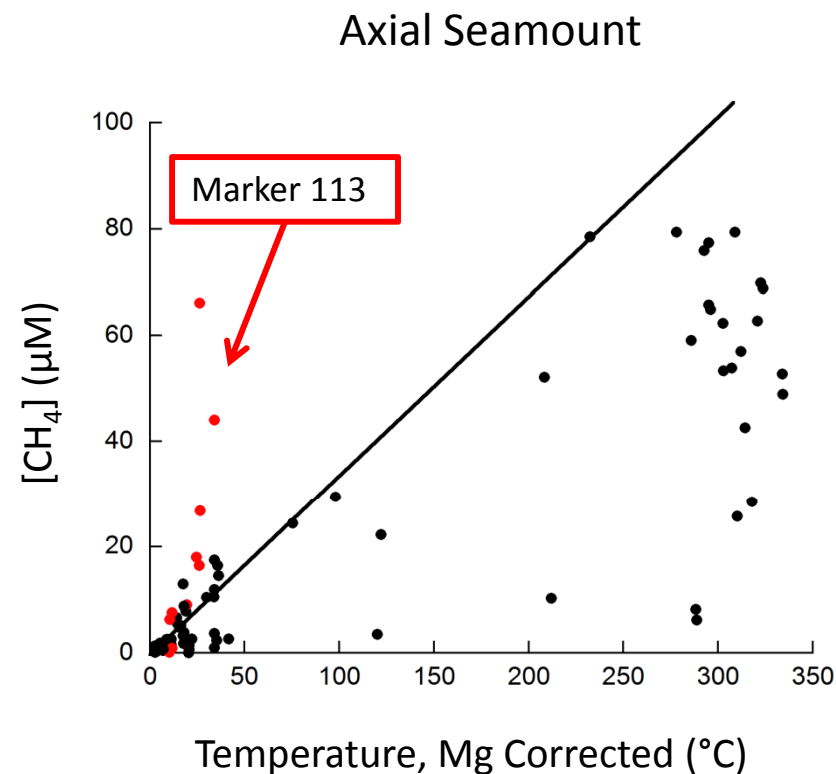
Fluid chemistry and culture-dependent survey for hyperthermophilic methanogens

Axial Seamount MPNs

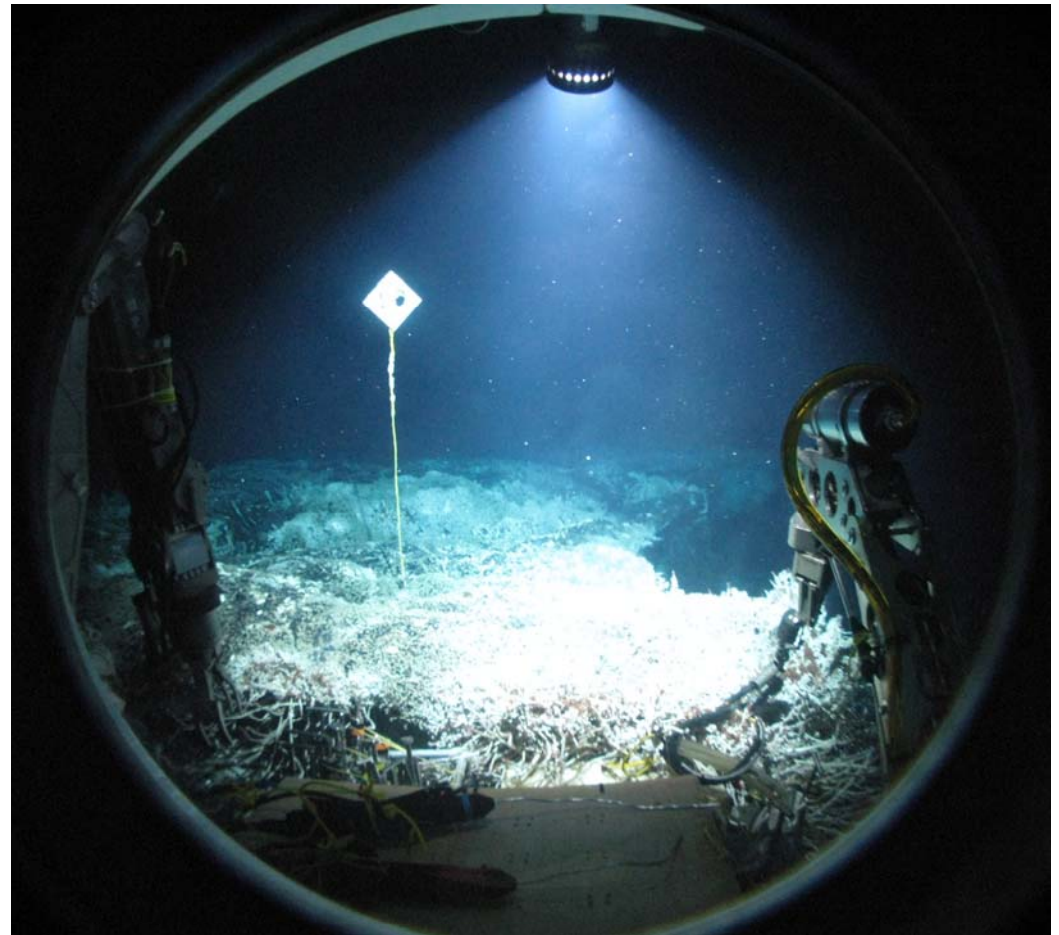
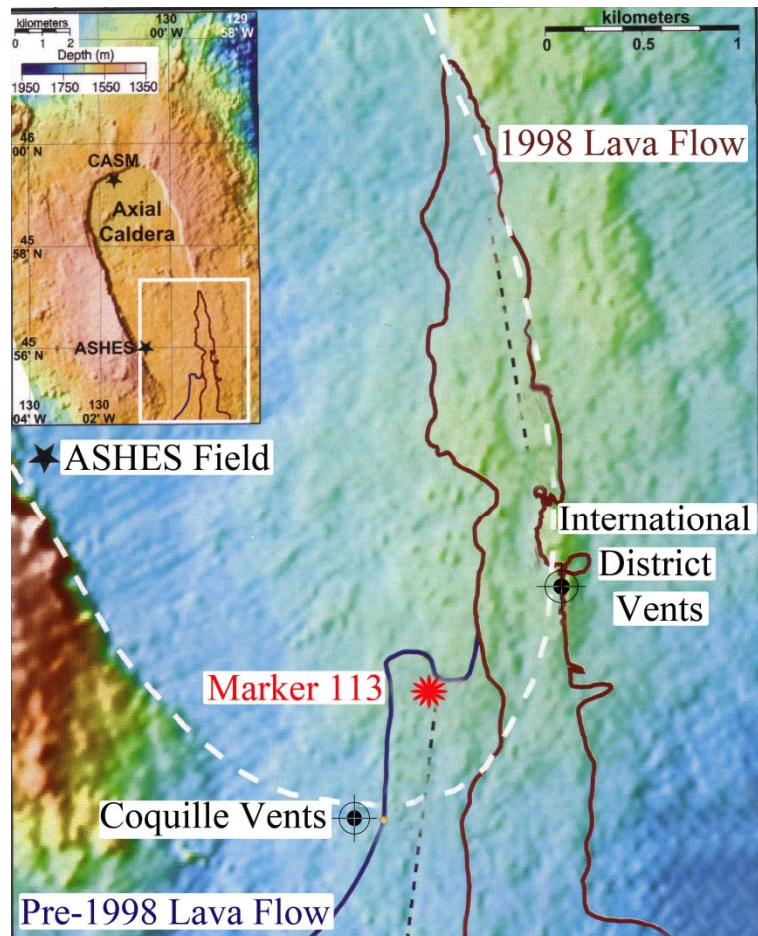
- 2 of 10 fluid samples had methanogens
- both were from Marker 113
- 1 of 2 sulfide chimney samples had methanogens

Endeavour Segment MPNs

- 2 of 18 fluid samples had methanogens
- both came from high H_2 vents
- 3 of 6 sulfide chimney samples had methanogens
- evidence of H_2 syntrophy



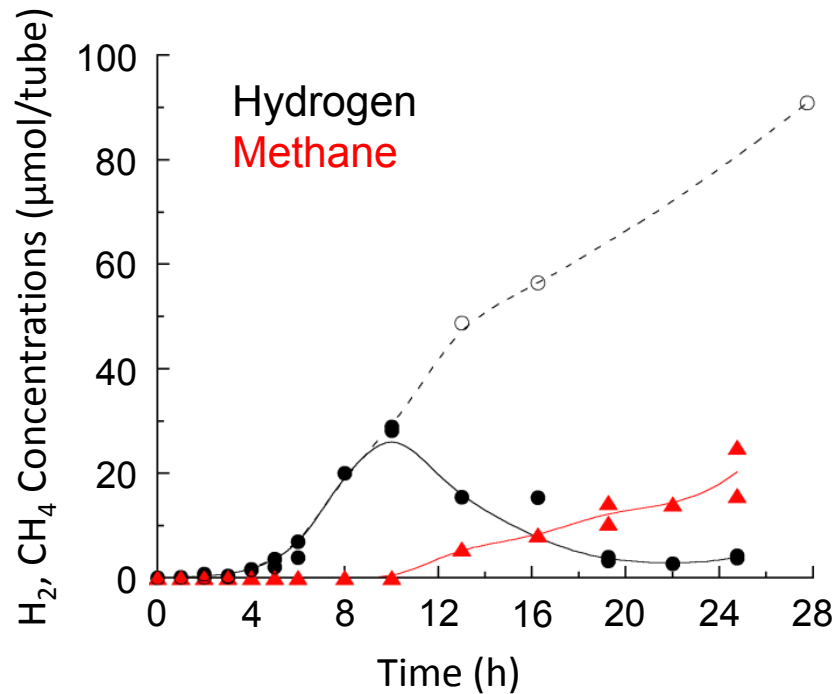
Marker 113 at Axial: Methanogen hot spot



Heterotrophs a syntrophic source of H₂

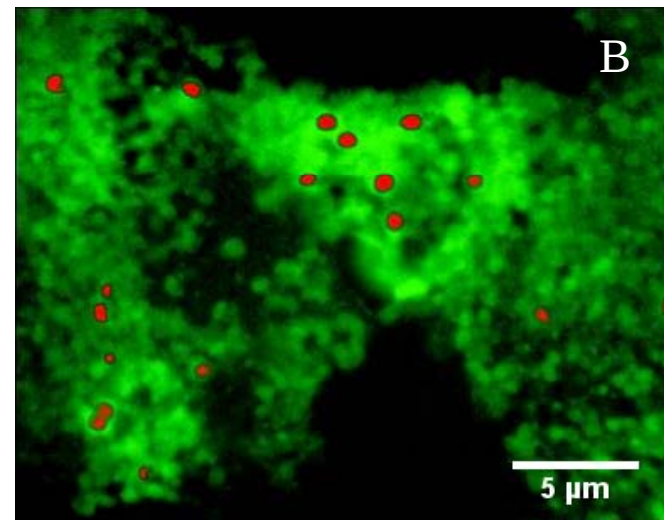
Hyperthermophilic heterotrophs were found in 27 of 28 diffuse vent fluids and in all sulfide chimney samples, and were >100x higher in concentration than methanogens

— — — *Thermococcus* CL1, pure culture
— — — *Thermococcus* CL1, co-culture
with *Methanocaldococcus* JH146



Fluorescent *in situ* hybridization (FISH)

Thermococcus
Methanocaldococcus

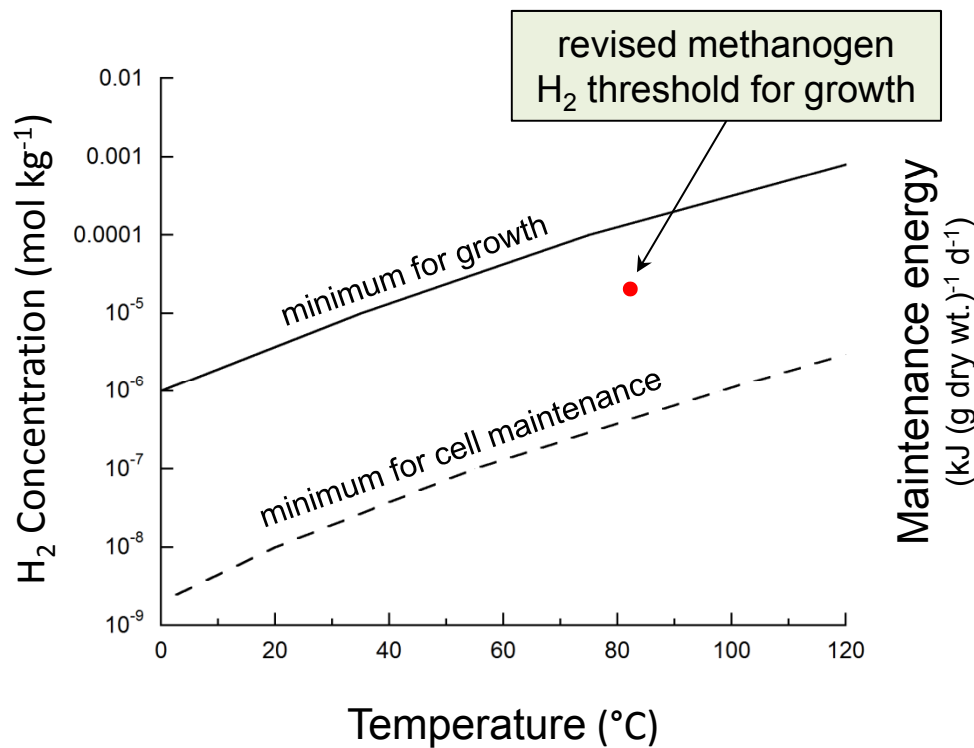


17-23 μM H_2 : Global threshold for methanogens

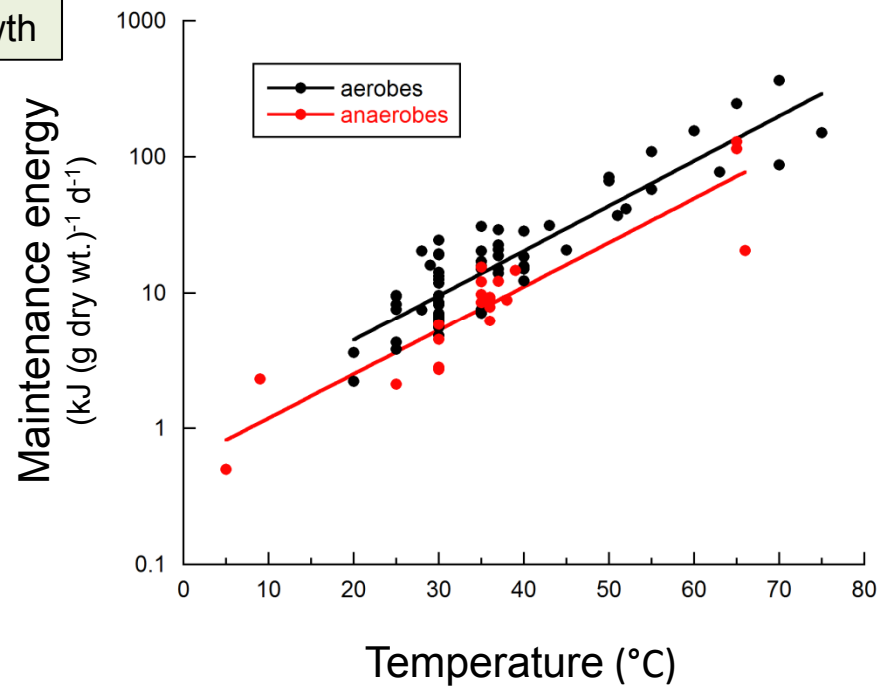
	Axial Volcano	Endeavour Segment	Mariner Field	TOTO Caldera	Brothers Volcano	Iheya North Field	Kairei Field
H_2 concentration, μM (82°C)	17-162	0-31	13	5	5	~50	550
Hyperthermophilic methanogens (max.)	1,290 L^{-1} 4 g^{-1}	690 L^{-1} 34 g^{-1}	not detected	not detected	not detected	650 g^{-1}	2,500 g^{-1}
Hyperthermophilic heterotrophs (max.)	>33,000 L^{-1} >45,200 g^{-1}	>72,000 L^{-1} 7,200 g^{-1}	10^7 g^{-1}	10^7 g^{-1}	10^7 g^{-1}	$3.5 \times 10^7 \text{ g}^{-1}$	$6 \times 10^6 \text{ g}^{-1}$

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$$\{H_2\}_{BEQ} = \left(\frac{\{CH_4\}}{\{CO_2\}} \cdot \exp\left(\frac{\Delta G^\circ_{(T,P)} - \Delta G_{BEQ}}{RT}\right) \right)^{1/4}$$



Hoehler (2004)



Tijhuis et al. (1993)

Research questions

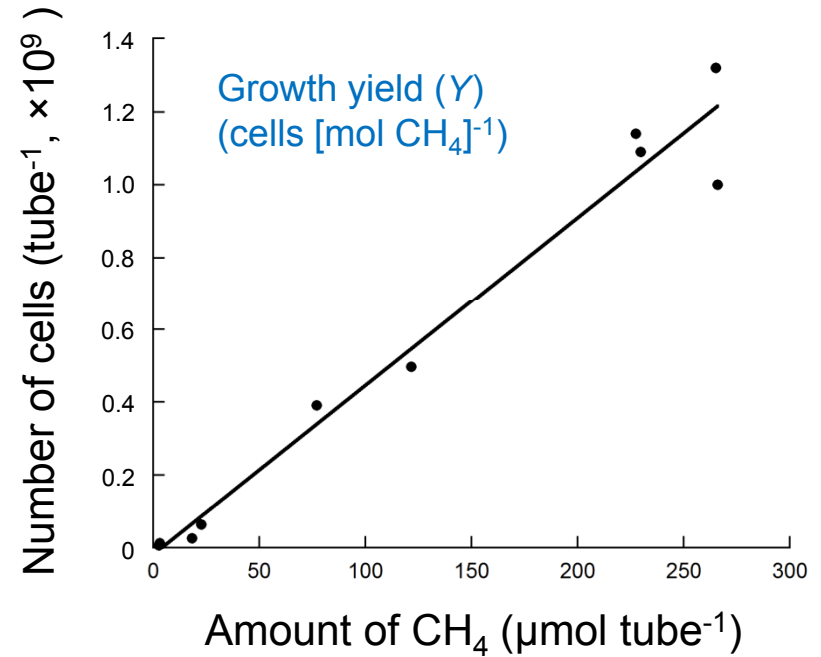
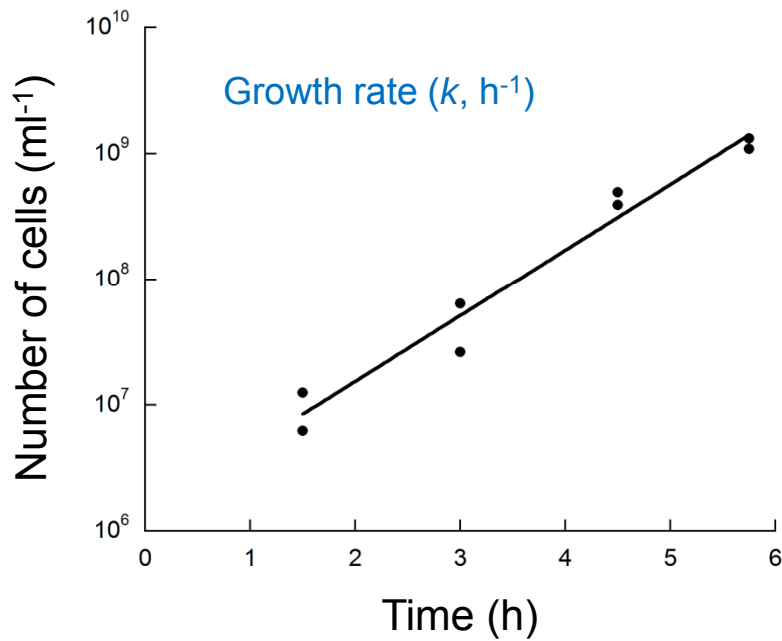
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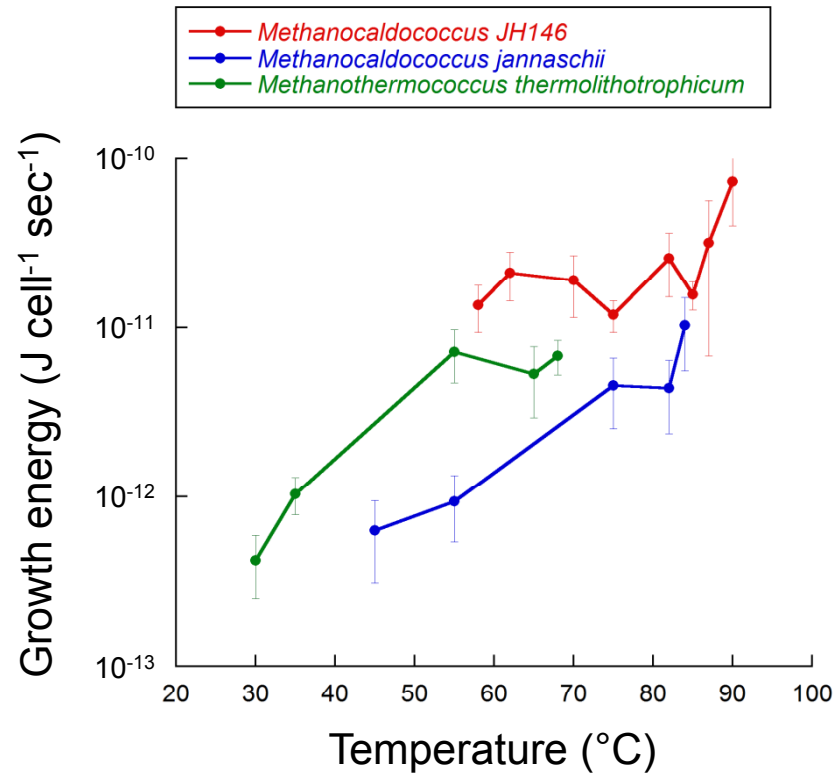
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Growth energy calculations for methanogens

$$\text{Growth energy} = \Delta G_r \times \frac{k}{Y \times 0.693}$$

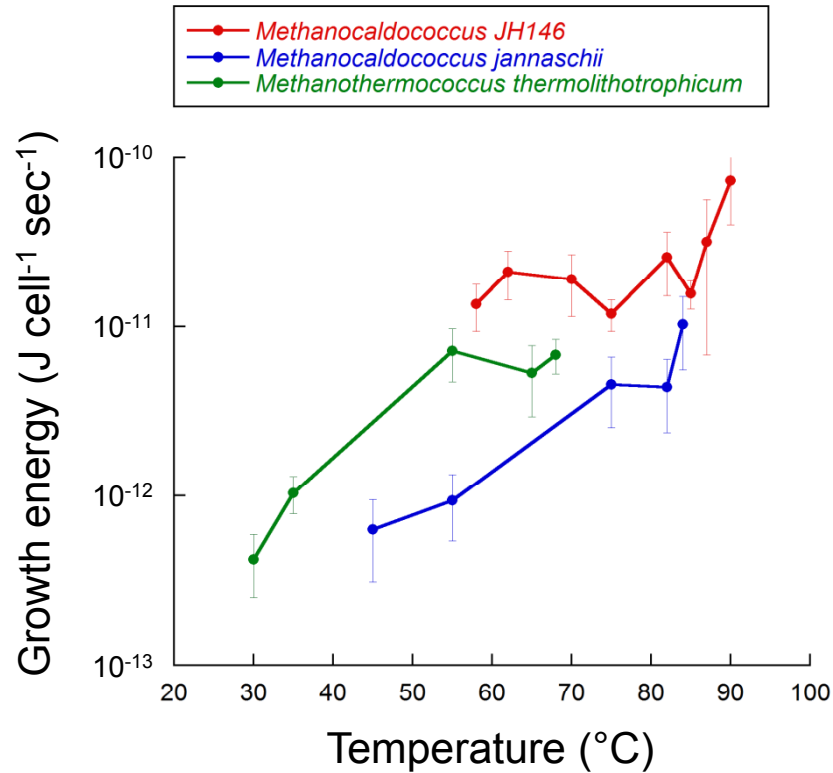


Effect of temperature on growth energy

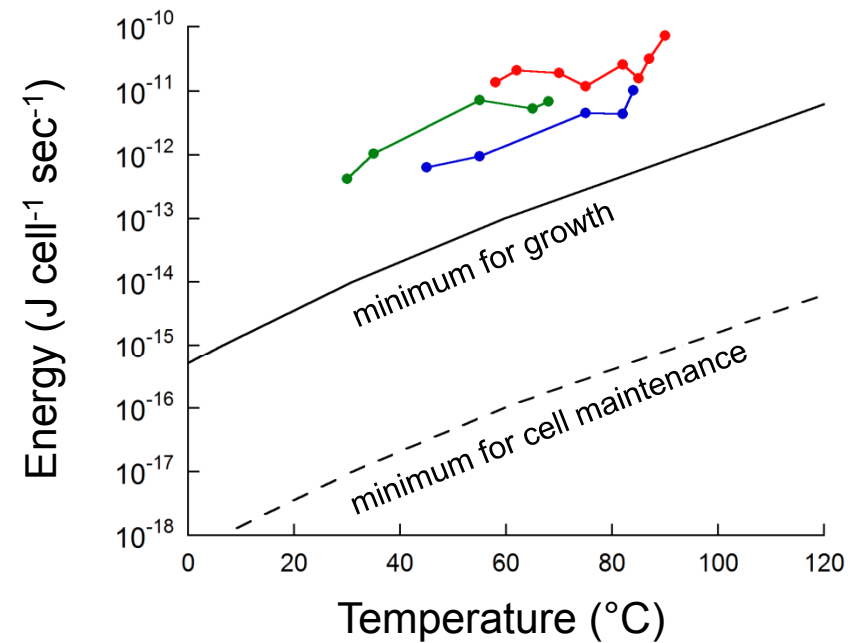


Ver Eecke et al. (in prep.), Stewart & Holden (unpubl.)

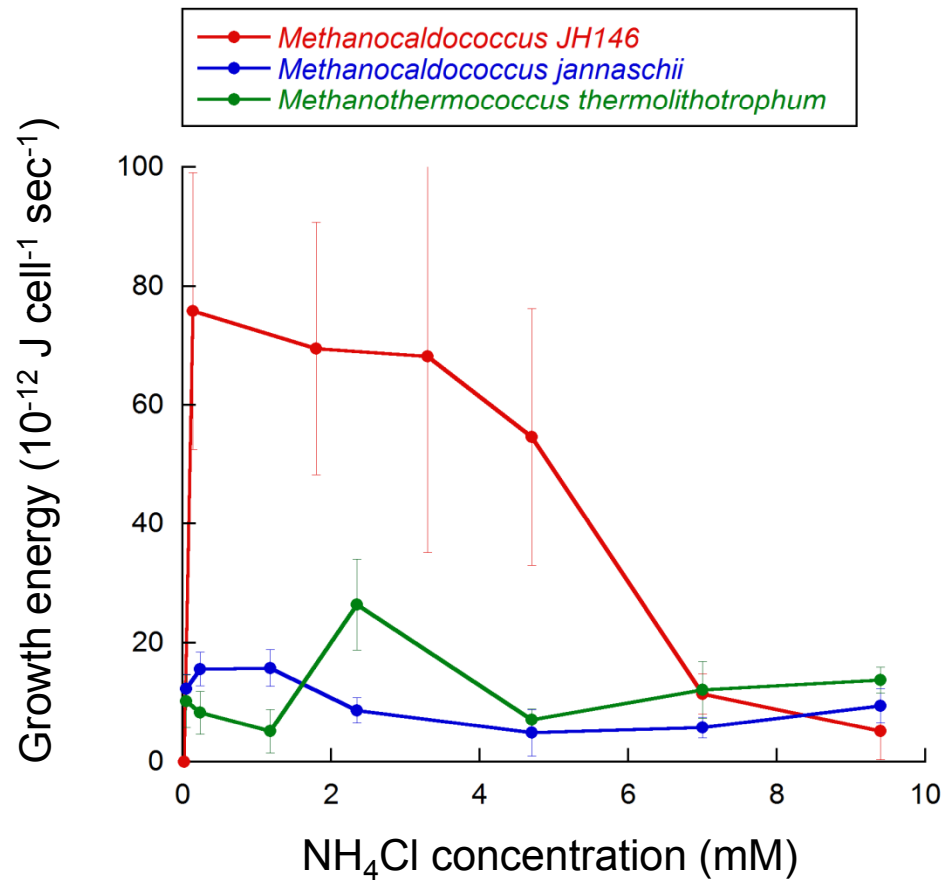
Effect of temperature on growth energy



Future studies: chemostat experiments to refine 'energy balance' model



Effect of N availability on growth energy

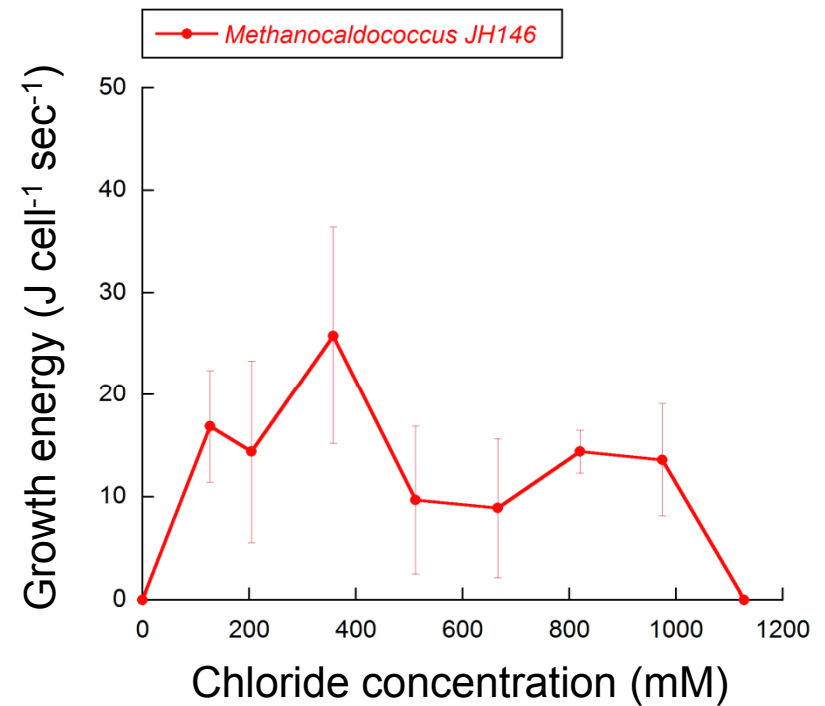
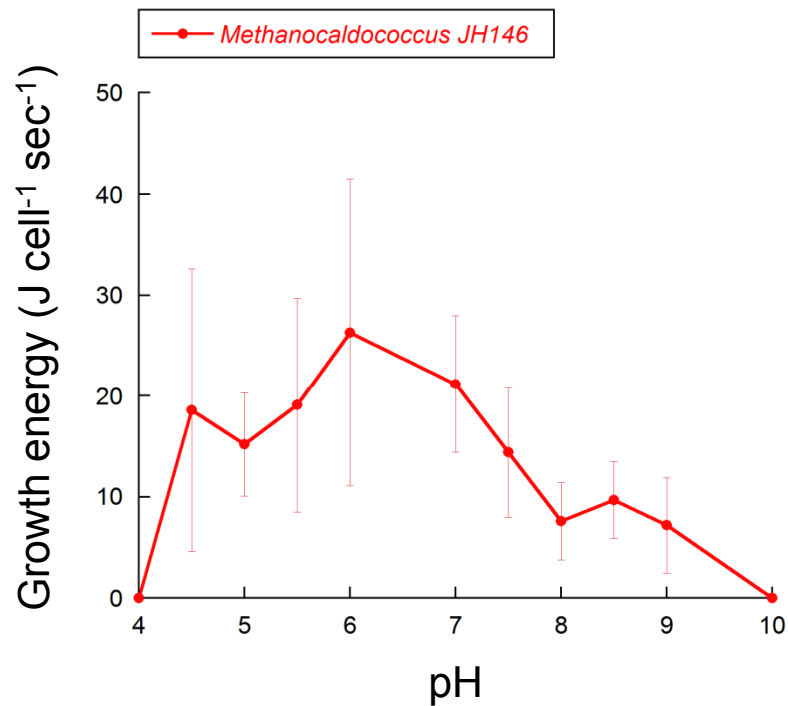


Hydrothermal fluid-seawater mix
 NH_4^+ concentration $\sim 15 \mu\text{M}$

Standard growth medium
 NH_4^+ concentration = 4.7 mM

Effects of pH and chlorinity on growth energy

No effect at environmentally relevant pH and Cl⁻ concentrations



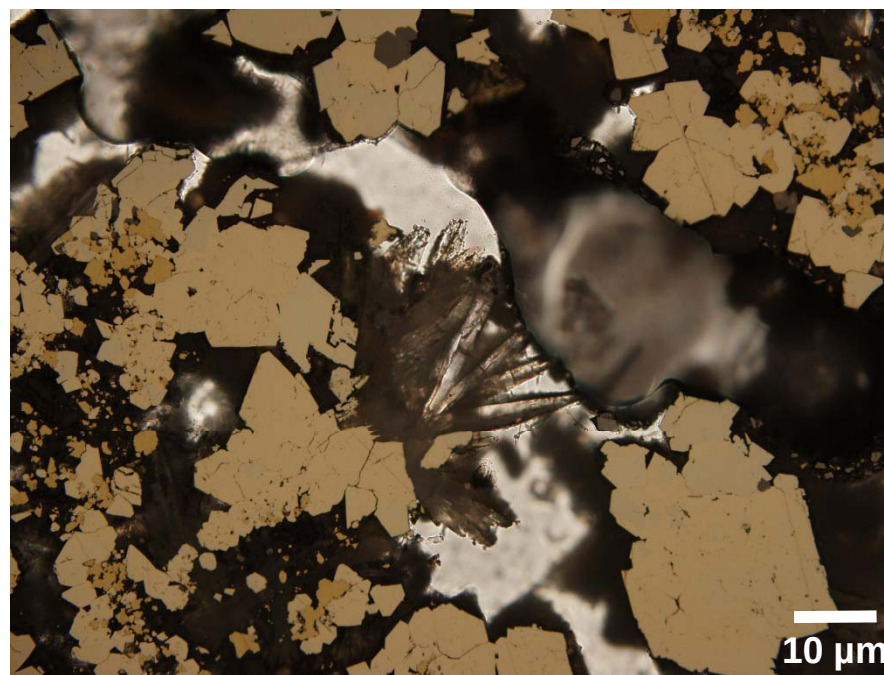
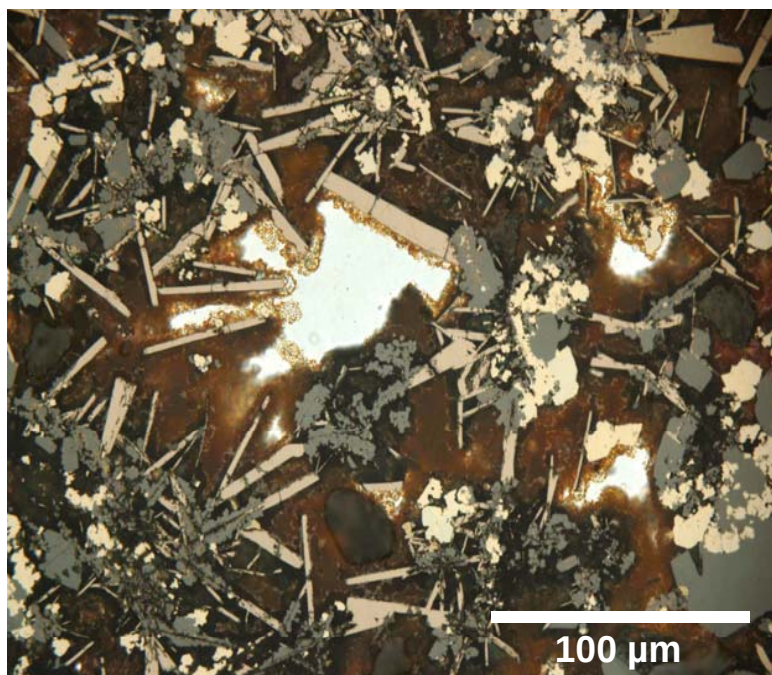
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- 2a. Do growth energy requirements vary with temperature for any one methanogen? **2 yes, 1 no**
- 2b. Do they vary within and between thermal classes of methanogens? **Yes and no**
- 2c. How do other environmental factors (pH, Cl⁻, N availability) impact methanogen growth energy requirements? **N affects 1 species, no effect of pH or Cl⁻ at environmental levels**

- 3a. Are usable minerals available in vents?
- 3b. Can hyperthermophiles use minerals as a growth substrate?
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Petrography and mineralogy of active hydrothermal sulfide deposits



August 2012 MPN results for Axial Seamount diffuse vents

Location (Temp.)	Methanogens (L ⁻¹)		Sulfur reducers (L ⁻¹)		Iron reducers (L ⁻¹)		Heterotrophs (L ⁻¹)		Total cells (L ⁻¹)
	55°C	80°C	55°C	80°C	55°C	80°C	55°C	80°C	
Anemone (33°C)	n.d.	n.d.	n.d.	n.d.	n.d.	90	690	7,200	7.9×10 ⁷
Fuzzy Tubeworm Bush (28°C)	n.d.	n.d.	270	n.d.	90	n.d.	270	2,790	4.7×10 ⁷
Boca (10°C)	120	n.d.	90	n.d.	450	n.d.	270	690	2.3×10 ⁸
Marker 113 (28°C)	n.d.	120	n.d.	n.d.	n.d.	13,800	n.d.	2,790	3.4×10 ⁸
Off-summit, 1,300 m (~2°C)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	2.5×10 ⁷

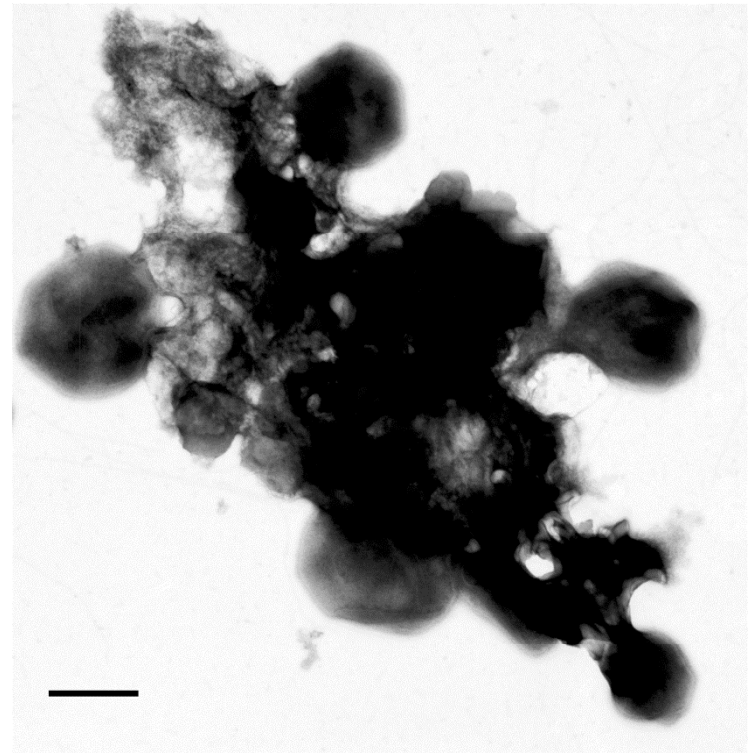
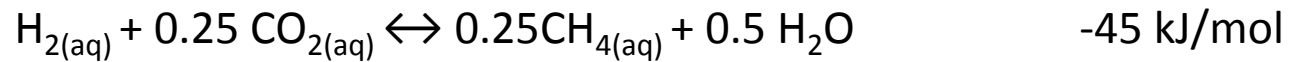
n.d., not detected

J.F. Holden (unpublished data)

Alternative to methanogens in low H₂ hydrothermal systems

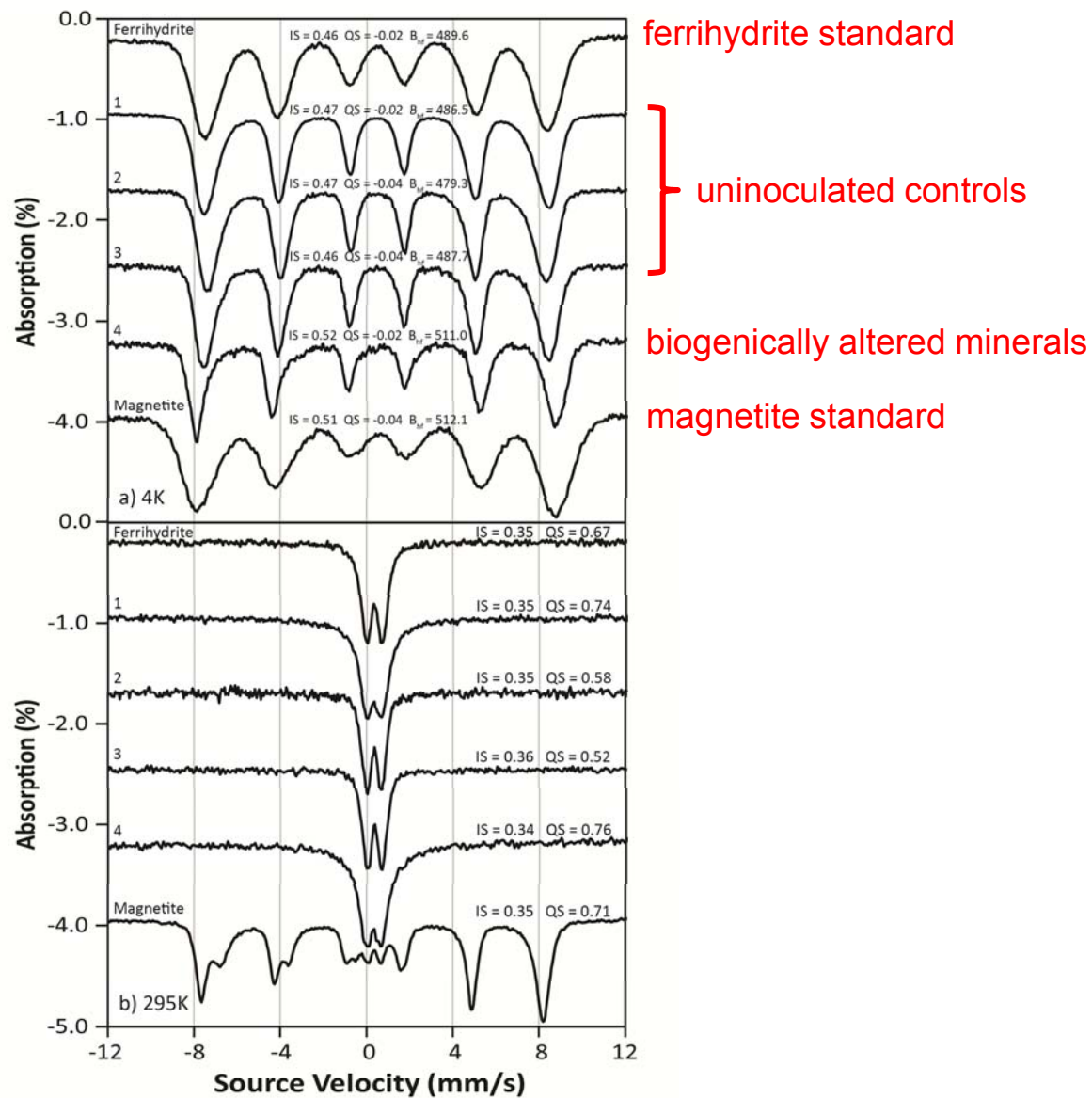
Growth Reaction

ΔG_r° 100°C



Biogenic mineral transformations as determined by Mössbauer spectroscopy

4 K



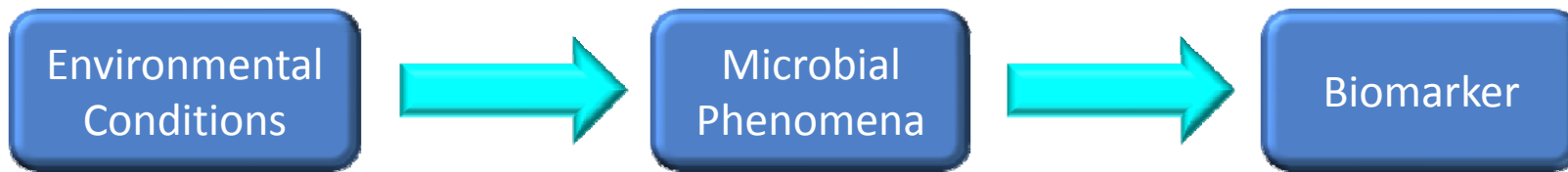
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- 3a. Are usable minerals available in vents? **Yes, in mixing zones**
- 3b. Can hyperthermophiles use minerals as a growth substrate? **Yes**
- 3c. Do these organisms leave behind a mineral biosignature? **Yes**

How is this actually done?

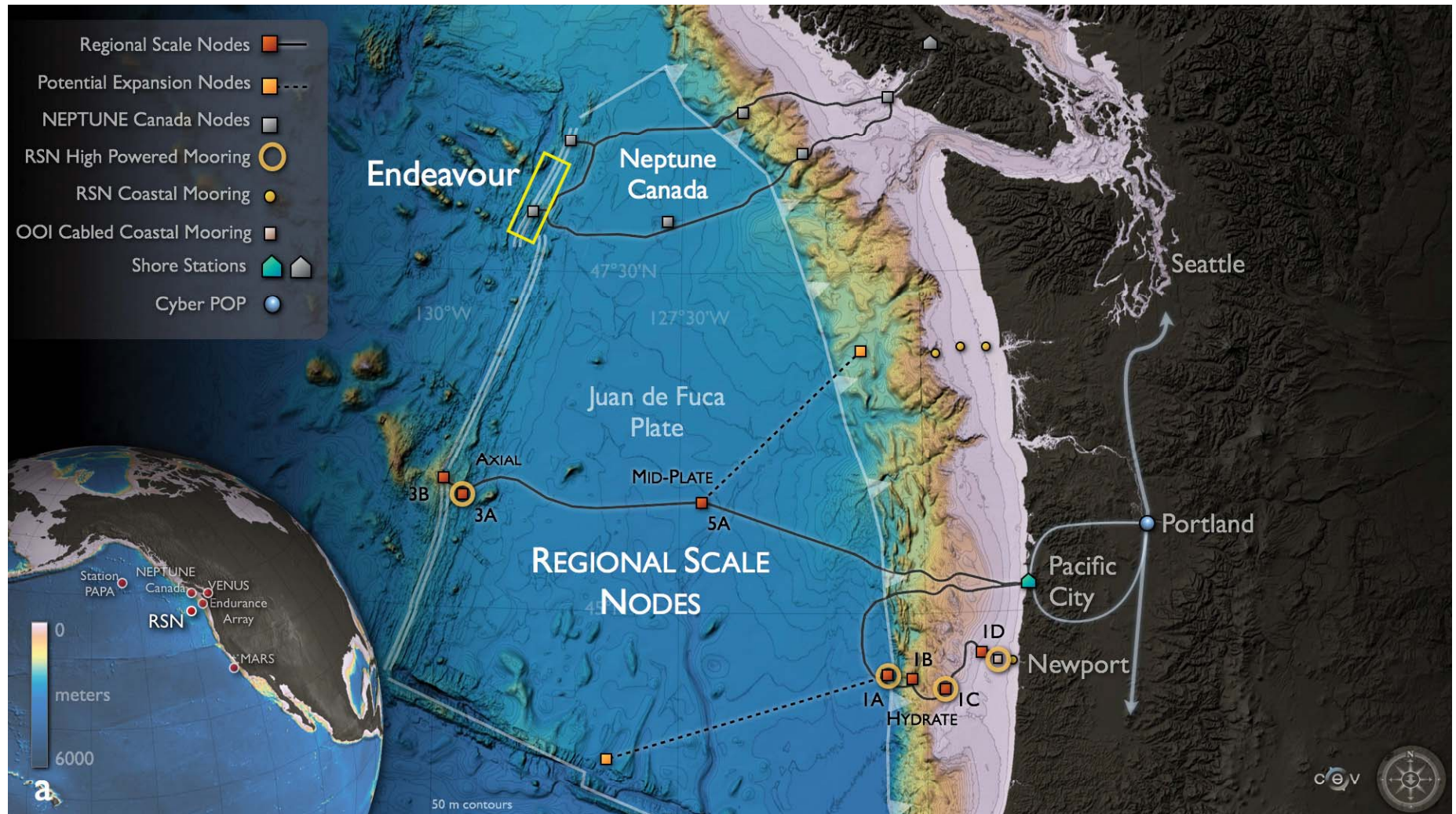


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and ...

Cabled observatories in the northeastern Pacific Ocean



Biologically relevant chemistry of end-member hydrothermal fluids from various sites and host-rock environments

LOCATION	RANGE OF CONCENTRATIONS				
	pH	H ₂ (mM)	CH ₄ (mM)	H ₂ S (mM)	Fe (mM)
MAFIC ROCK					
Main Endeavour Field	4.2-4.5	0.16-1.5	1.8-4.5	2-29	0.53-1.36
ASHES, Axial Volcano	3.5-4.4	0.07-0.27	0.07-0.28	7.1	0.01-1.07
9°50'N East Pacific Rise	ND	0.33-8.91	0.05-0.75	6.2-23.2	0.2-6.64
Kilo Moana, Lau Basin	2.9-4.0	ND	ND	5.4-6.3	2.5-3.8
Lucky Strike, Mid-Atlantic Ridge	3.5-4.9	0.02-0.73	0.30-0.85	1.4-4.6	0.03-0.86
ULTRAMAFIC ROCK					
Rainbow, Mid-Atlantic Ridge	2.8	16	2.5	1.2	24.1
Logatchev, Mid-Atlantic Ridge	3.3-3.9	12-19	2.1-3.5	0.8-2.5	2.4-2.5
Kairei, Central Indian Ridge	3.4-3.5	7.9-8.2	0.08-0.20	3.9-4.1	3.5-6.0
DACITIC/ANDESITIC ROCK					
Mariner Field, Lau Basin	2.4-2.7	0.04-0.10	0.01	6.9-9.0	10.5-13.0
DESMOS Caldera, Manus Basin	2.1-2.7	ND	ND	5.3-9.7	0.01-4.4
Brothers Volcano, Kermadec Arc	2.8-3.0	0.01-0.02	< 0.01	2.3-7.9	4.2-7.3
Seawater	7.8	0.0004	0.00001	0	0.000061

ND = not determined.