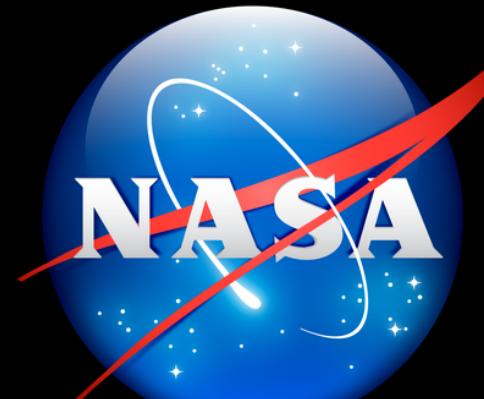


# Simulating Martian Conditions: Methanogen Survivability



## During Freeze/Thaw Cycles

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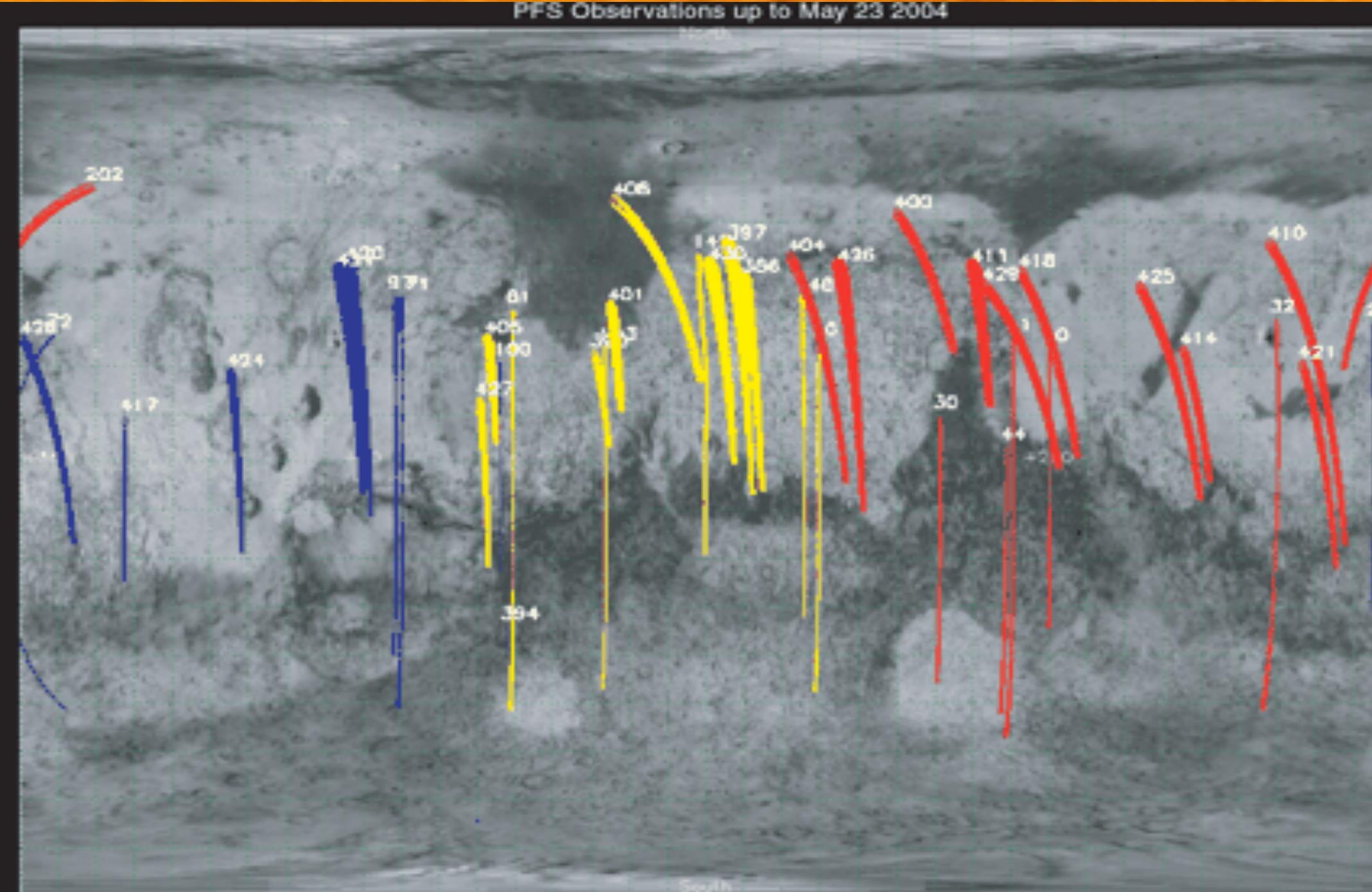
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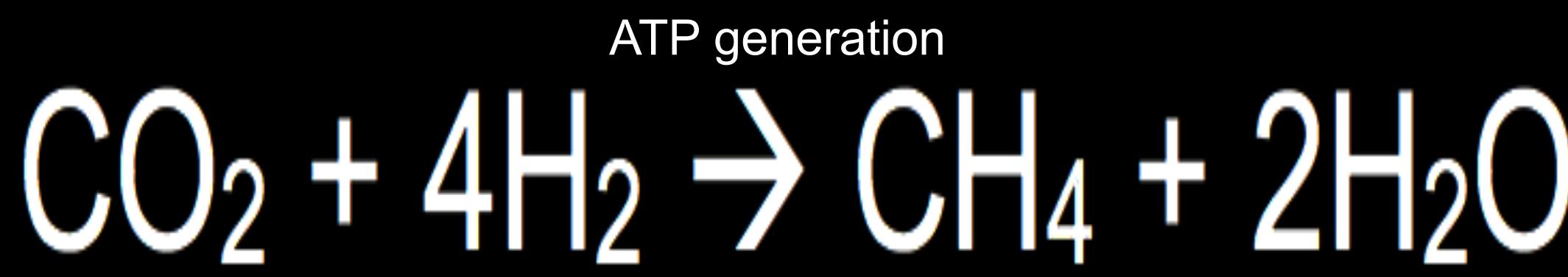
### Background

- Extensive evidence of methane in the martian atmosphere (Fig. 1; avg. 10 ppb) [1-6]
- Methanogens are obligate anaerobes that tolerate a wide range of conditions
- It is proposed that methanogens are able to metabolize and survive in these martian conditions (Fig. 2)
- Temperature and pressure will be analyzed to determine growth and survivability (Fig. 3)



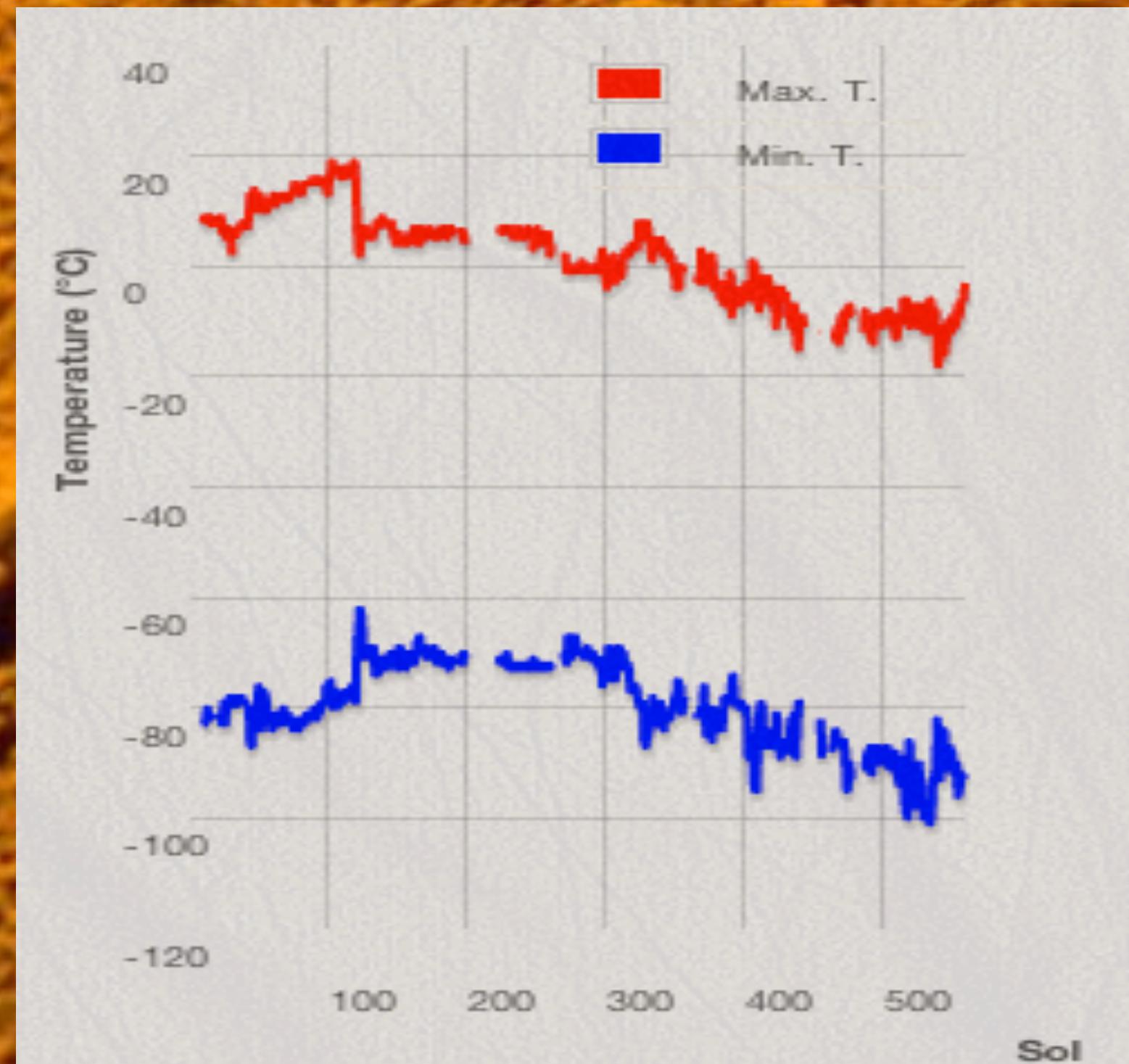
**Figure 1**

Methane distribution on Mars from data obtained from the Mars Express orbiter [1].



**Figure 2**

Methanogens generates methane by using CO<sub>2</sub> as a carbon source and H<sub>2</sub> as an energy source.

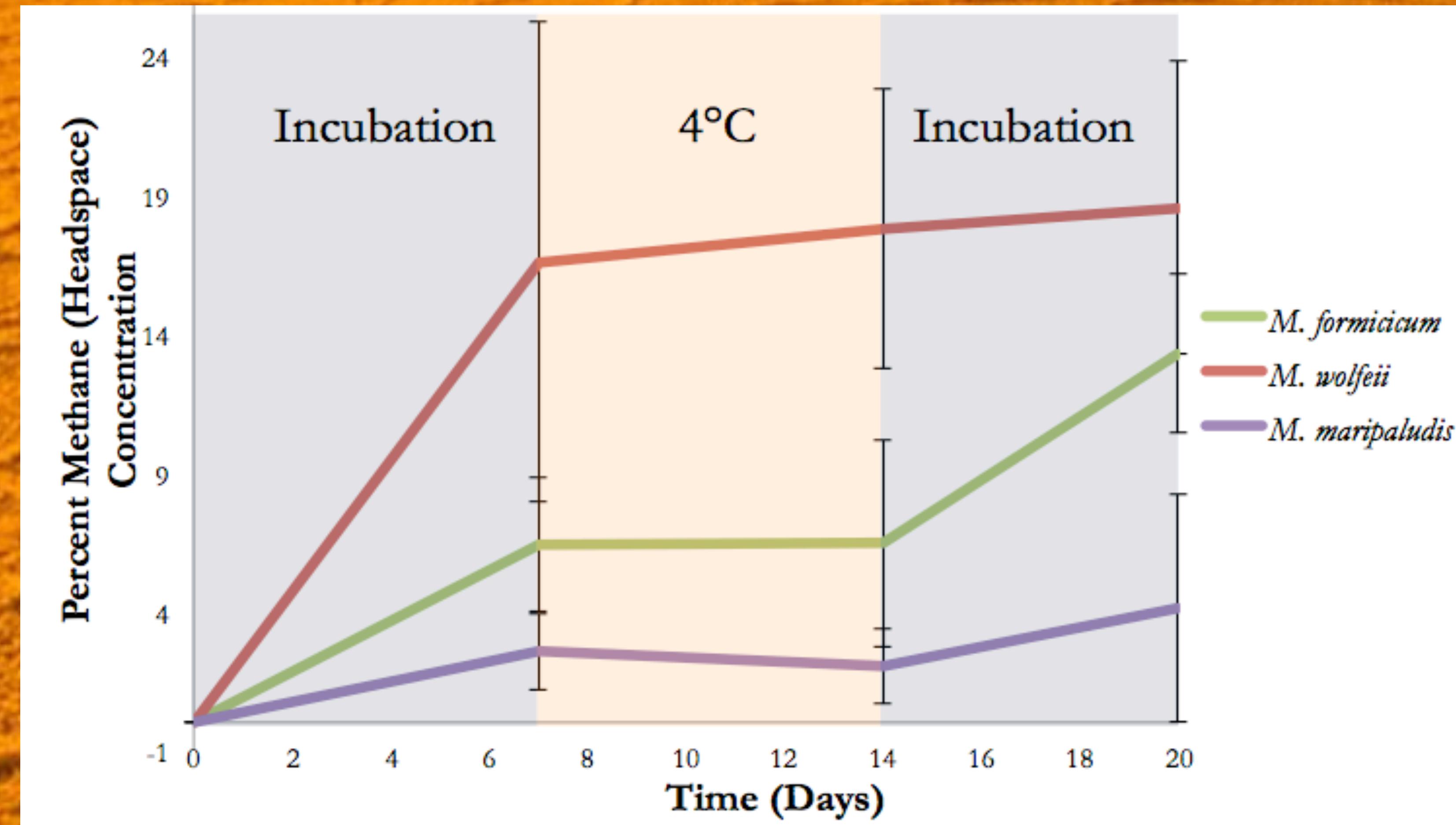


**Figure 3**

Ground temperature data from the Rover Environmental Monitoring System from early-August 2012 to the present time at Mars' Gale Crater [7].

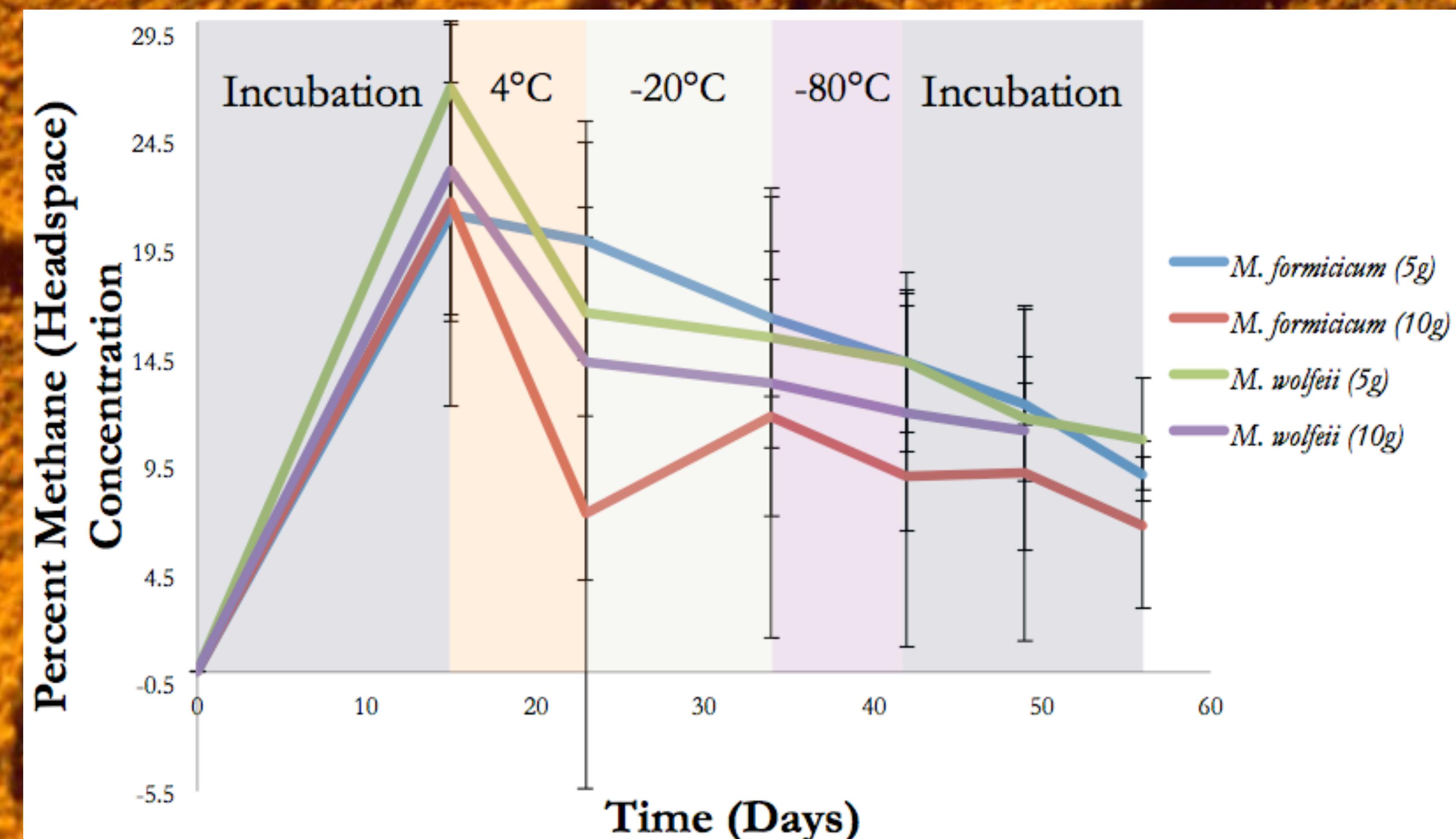
### Methods

- Media prepped according to Kendrick and Kral [8] (Fig. 6)
- Media inoculated with 0.5 mL of respective methanogen (*M. barkeri*, *M. formicicum*, *M. maripaludis*, *M. wolfeii*); grown at ideal growth temperature (*M. barkeri/M. formicicum* at 37°C, *M. maripaludis* at room temperature (22°C), and *M. wolfeii* at 55°C)
- Solutions transferred to various temperatures (Figs. 4, 5) and growth monitored by methane production using gas chromatography
- Solutions were grown in 5 or 10 grams of sand (Fig. 5)



**Figure 4**

Percent methane (headspace) concentrations for each of three methanogen strains (*M. wolfeii*, *M. maripaludis*, *M. formicicum*) following a seven day incubation period, a 4°C freeze-thaw cycle, and subsequent incubation period. Error bars represent one standard deviation.



**Figure 5**

Percent methane concentrations in media inoculations containing either 5 or 10 grams of sand following 7 day incubation period and various freeze/thaw cycles (\**M. barkeri/M. maripaludis* not viable). Error bars represent one standard deviation.



**Figure 6**

Coy Anaerobic Environmental Chamber used for media prep (90% CO<sub>2</sub>/10% H<sub>2</sub>).

### Results and Conclusions

- Methane concentration greater than 5% was interpreted as growth/survivability
- Incubation at 4 degrees slowed or inhibited growth at that temperature, but was not lethal since the organisms continued growing at their ideal growth temperatures afterward
- *M. barkeri/M. maripaludis* displayed little (<5% CH<sub>4</sub>) to no growth during freeze/thaw cycles and their data is not shown here
- Survival indicates that future experiments should be conducted

### Further Studies

- Find temperature/pressure constraints for each species in order to understand implications for life in Martian conditions.
- Longer freeze/thaw studies conducted (Mickol RL. et. al. this conference, Abstract #1603)

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