

Initiating the Biotransformation of Martian Regolith by Dissimilatory Perchlorate-Reducing Microorganisms

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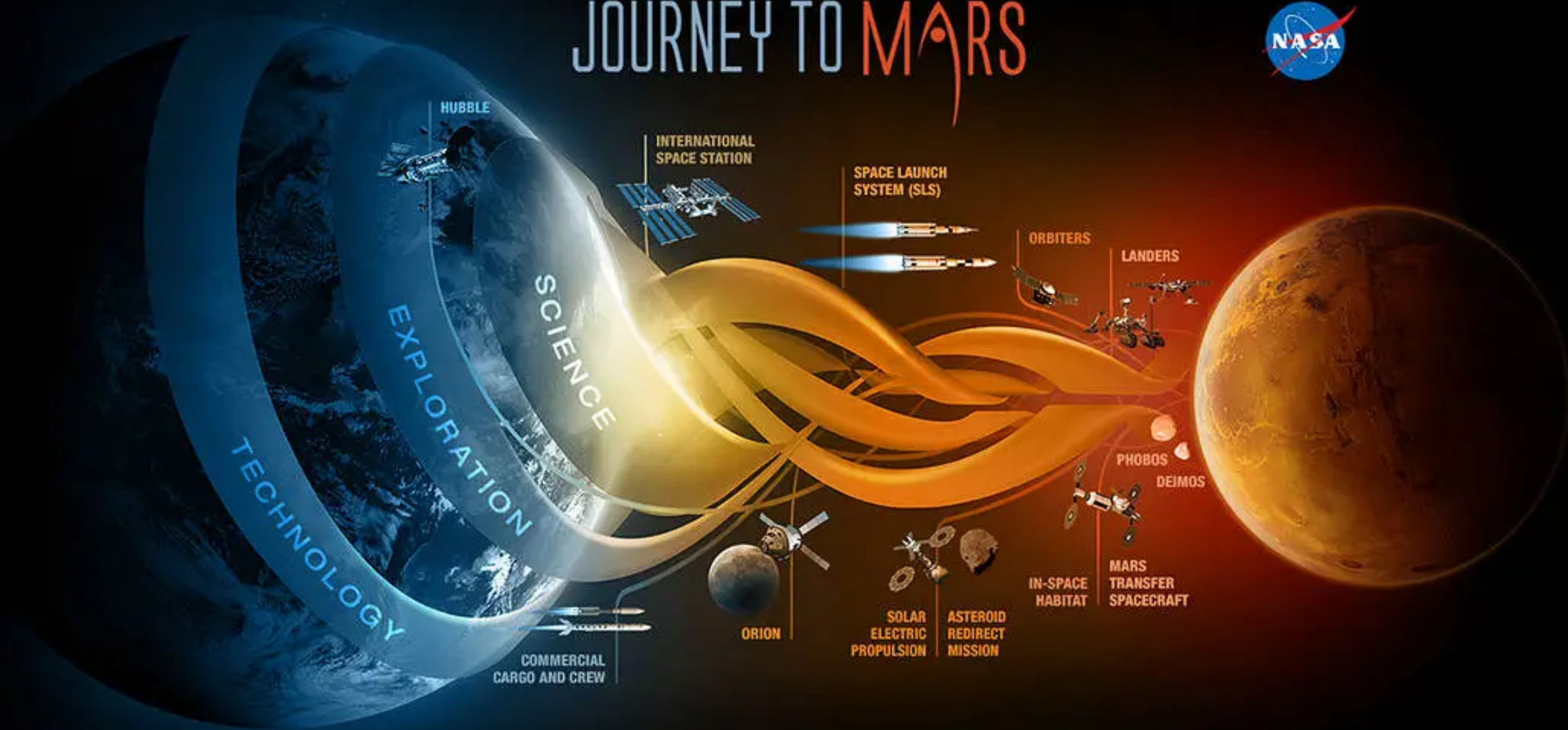
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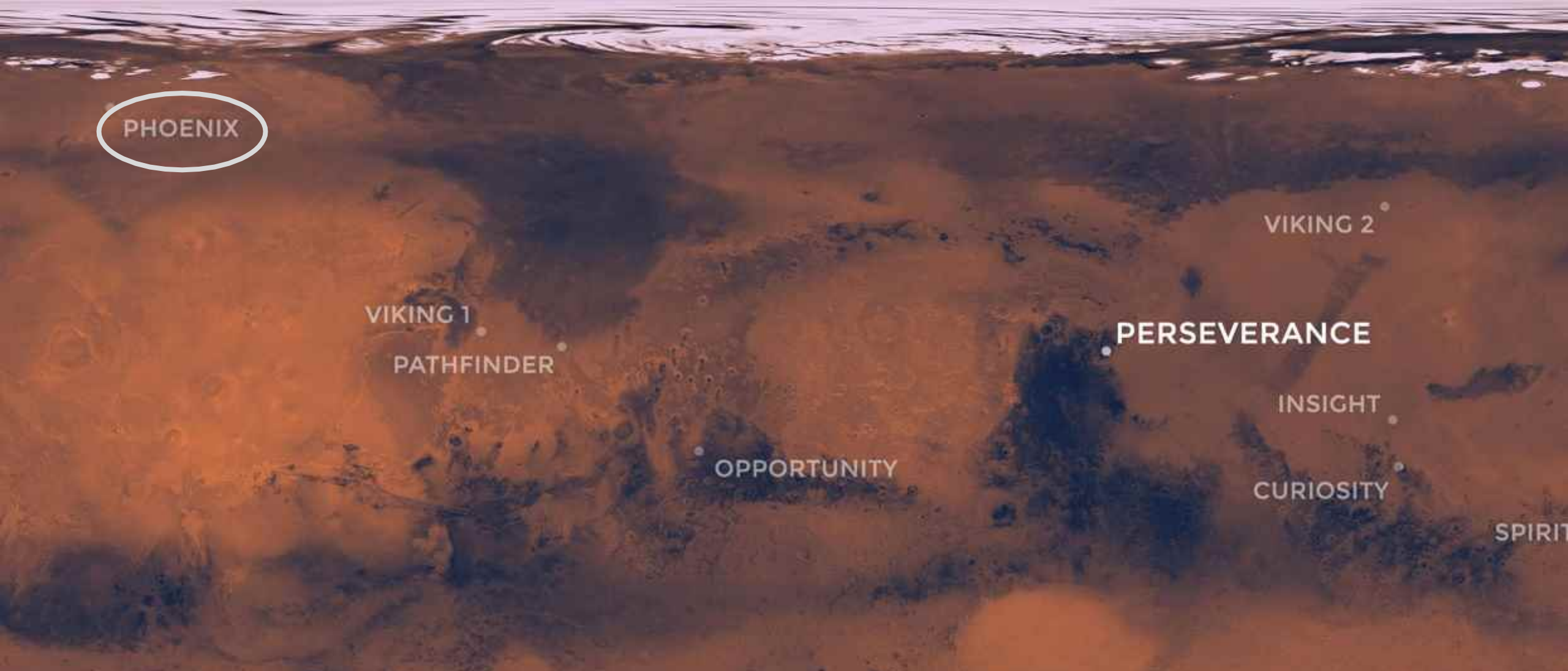
Arizona State University

Session G4: Bioremediation: Advances in Amendment Formulations

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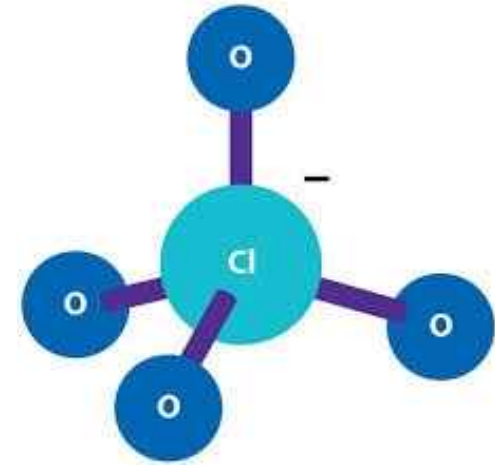
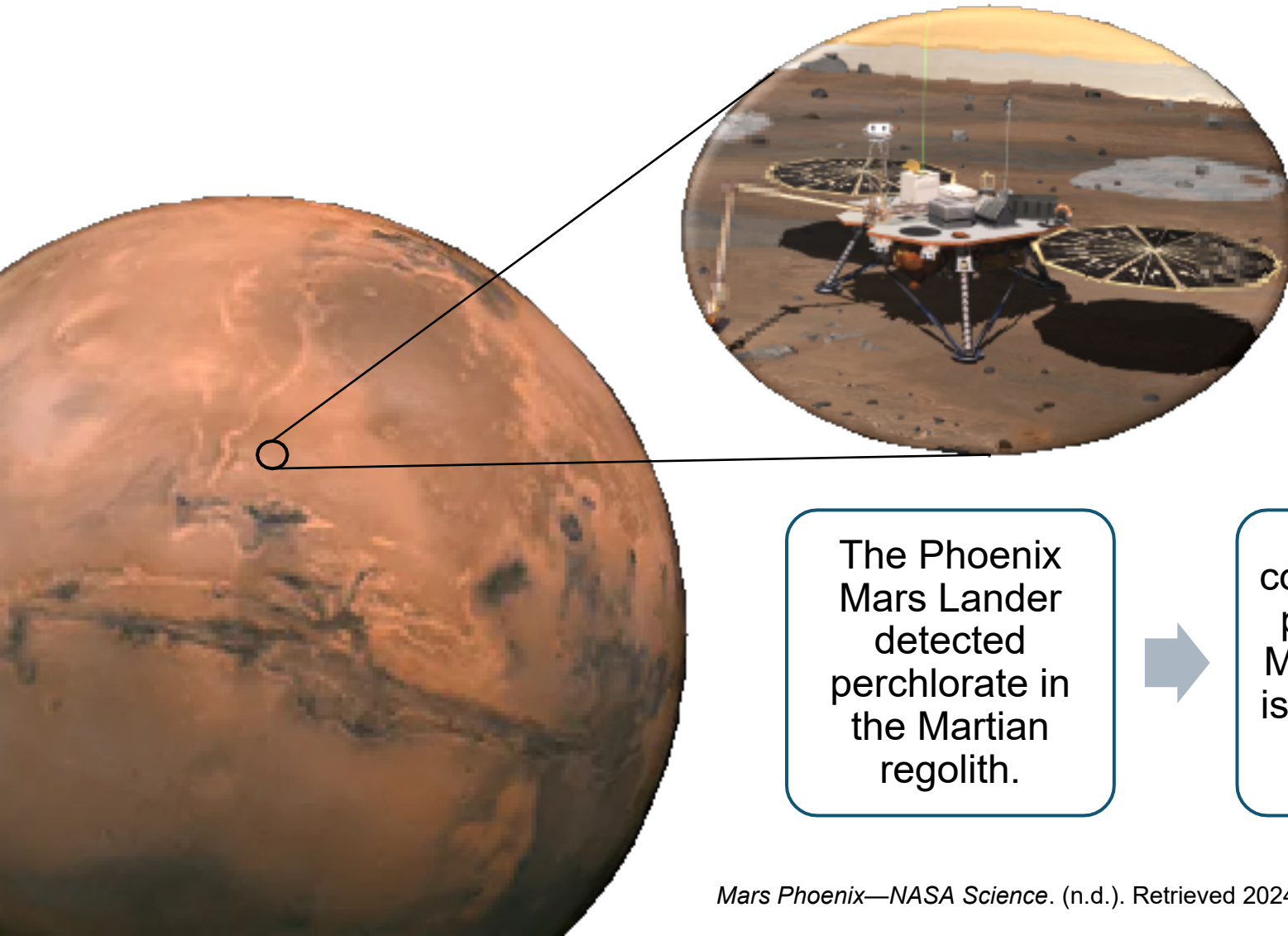






Mars Exploration

Perchlorate on Mars



The Phoenix Mars Lander detected perchlorate in the Martian regolith.



The concentration of perchlorate in Martian regolith is **0.5-2 wt%** (5-20 g/kg regolith).

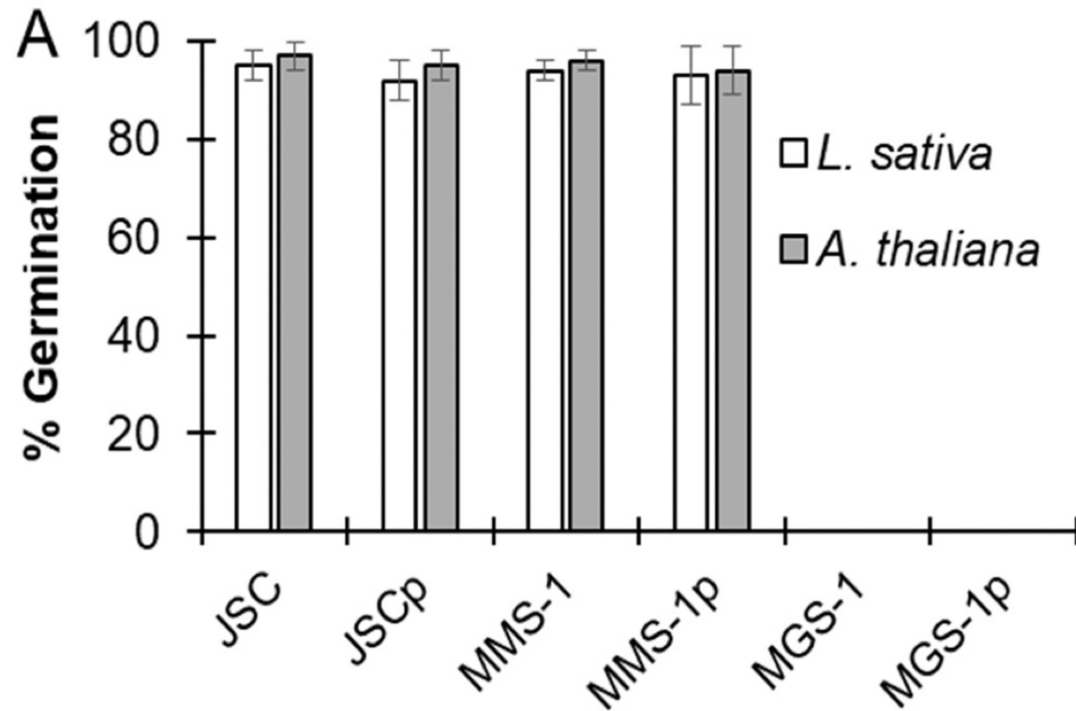


Perchlorate has been detected as sodium, calcium, and magnesium salts.

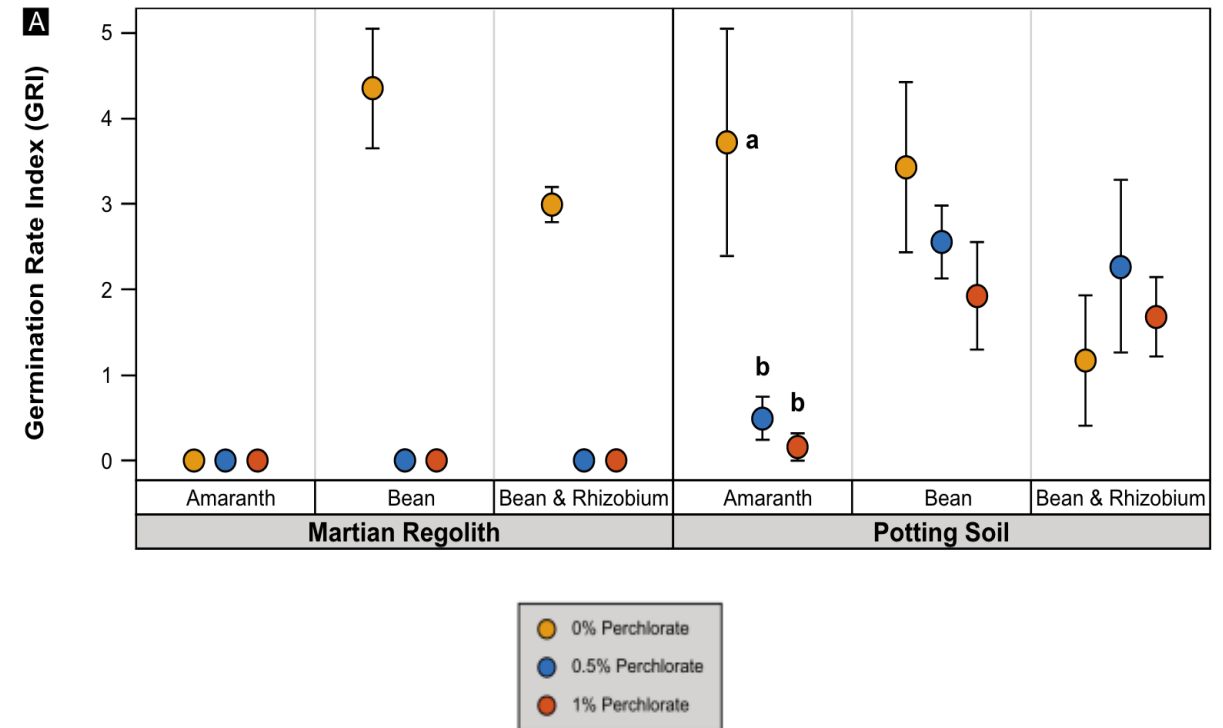
How will a human crew on a mission to Mars
grow plants for crops and bio-regenerative
life support?



Plant Growth in the Presence of Perchlorate



Plants used: romaine lettuce (*Lactuca sativa*), thale cress (*Arabidopsis thaliana*)

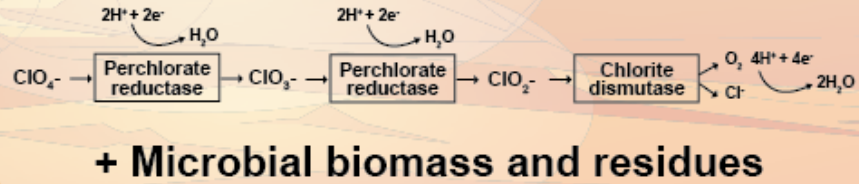


Plants used: Grain amaranth (*Amaranthus cruentus*) and common bean (*Phaseolus vulgaris*)

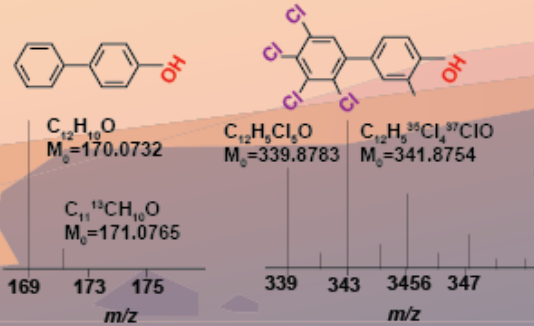
Project approach



Martian regolith
simulant with ClO_4^-



Regolith bioweathering/
SOM stabilization
and diversification



SOM/Bioweathering
product characterization

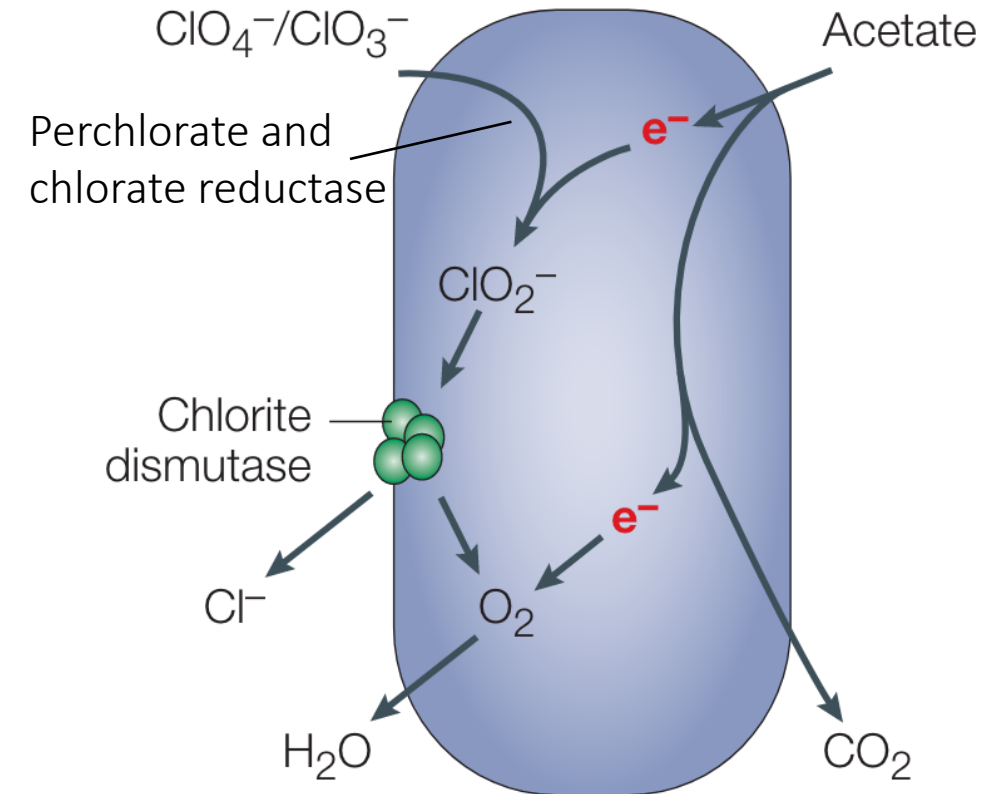
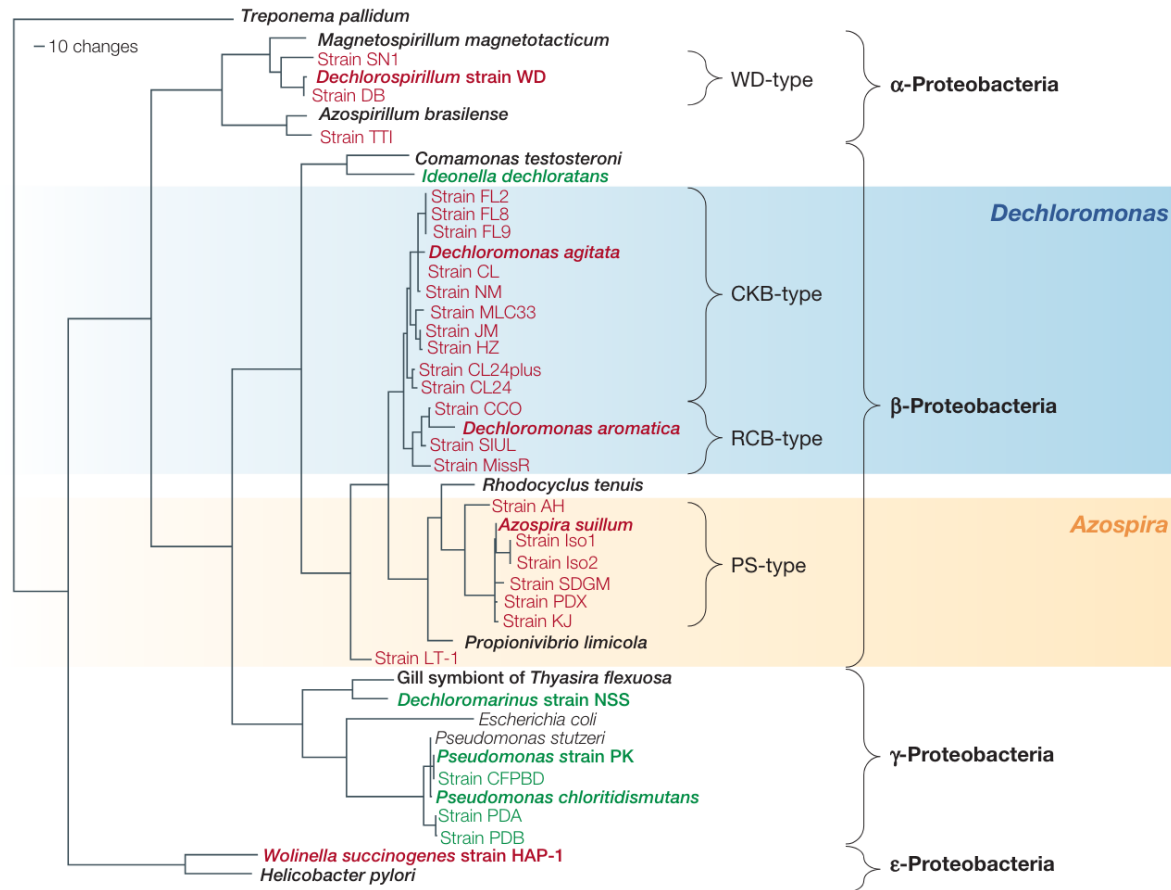
Seed germination
and plant growth

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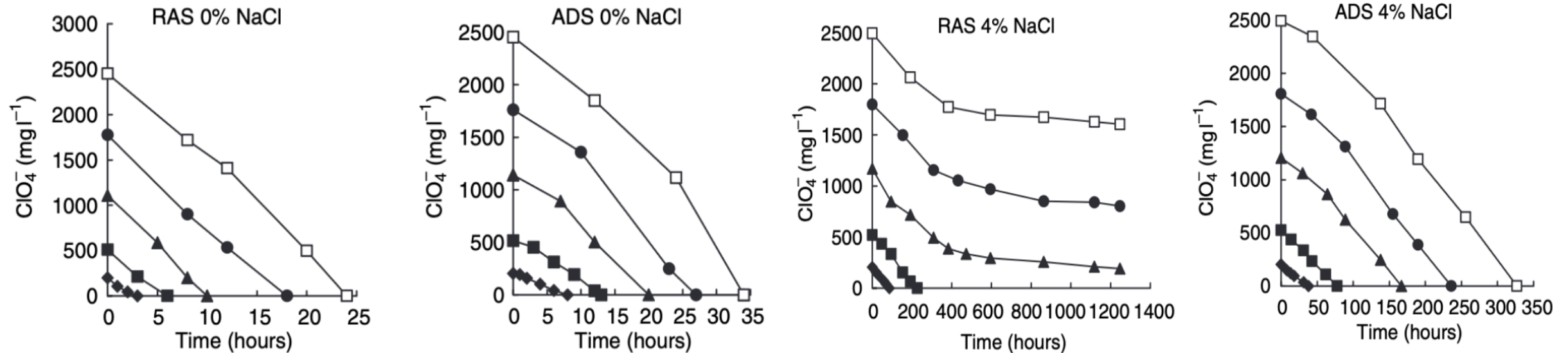
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Microbial Perchlorate Reduction



Perchlorate Reduction at High Concentrations

RAS – Return Activated Sludge, ADS – Anaerobic Digester Sludge



The ADS culture reduced perchlorate at salinities up to 4% NaCl, while the RAS culture exhibited significant inhibition of perchlorate reduction at 4% NaCl.

Given the high concentration of perchlorate in Martian regolith, is perchlorate reduction possible? If yes, to what extent?



Martian Regolith Simulant MGS-1

Characterization



MGS-1 Mars Global Simulant | **Fact Sheet**
003-05-001-1223

Simulant Name: MGS-1 Mars Global Simulant
Simulant Type: General purpose
Reference Material: Rocknest soil
Uncompressed Bulk Density: 1.40 g/cm³
Median Particle Size: 63 µm
Particle Size Range: >0.04 µm – 1000 µm



Parameter

MGS-1

MGS-1 +
1 wt.%
ClO₄⁻

pH (s.u.)

7.64 ± 0.03

7.81 ± 0.03

Conductivity (mS cm⁻¹)

9.52 ± 0.26

14.82 ± 0.04

Total dissolved solids (g/L)

4.32 ± 0.03

6.50 ± 0.06

Salinity (g/L)

3.97 ± 0.06

7.18 ± 0.08

Total organic carbon (%)

0.234 ±
0.001

0.156 ±
0.004

Moisture content as received
(%)

2.65 ± 0.37

Moisture content at 100%
water holding capacity (%)

19.06 ± 12.39

Geotechnical Properties

Avg Angle of Repose: 38.9°
Max Angle of Repose: 43.6°

More coming soon!

Safety

See SDS for details.
Primary hazard is dust
inhalation; wear a
respirator in dusty
conditions.

Mineralogy

As mixed.

Component	Wt.%
Anorthosite	27.1
Glass-rich basalt	22.9
Bronzite	20.3
Olivine	13.7
Mg- sulfate	4.0
Ferrihydrite	3.5
Hydrated silica	3.0
Magnetite	1.9
Anhydrite	1.7
Fe-carbonate	1.4
Hematite	0.5

Bulk Chemistry

¹Relative abundances
Measured by XRF.

Oxide	Wt.%
SiO ₂	43.90
TiO ₂	0.46
Al ₂ O ₃	12.84
FeO	10.60
MnO	0.11
MgO	14.81
CaO	7.91
Na ₂ O	1.49
K ₂ O	0.29
P ₂ O ₅	0.17
LOI	4.90
Total	97.48

Experimental Approach



Martian regolith simulant: 20 – 80 g/bottle of Martian regolith simulant MGS-1 + 1 wt.% perchlorate

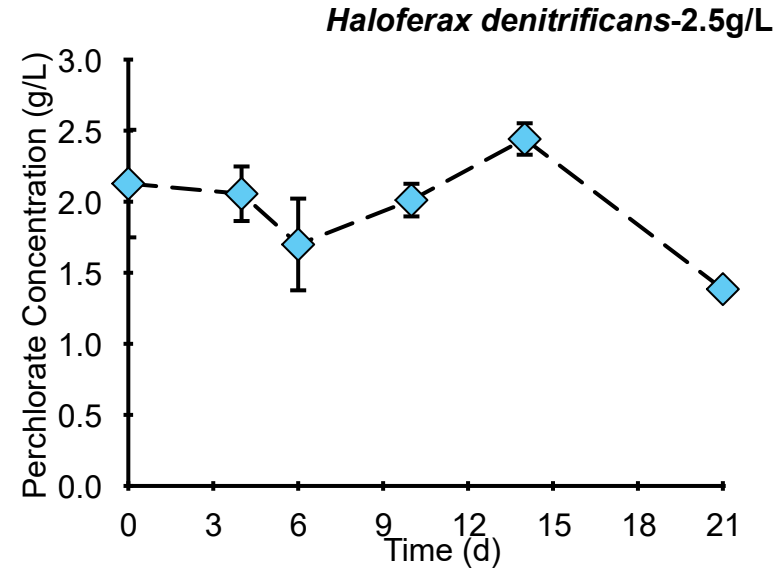
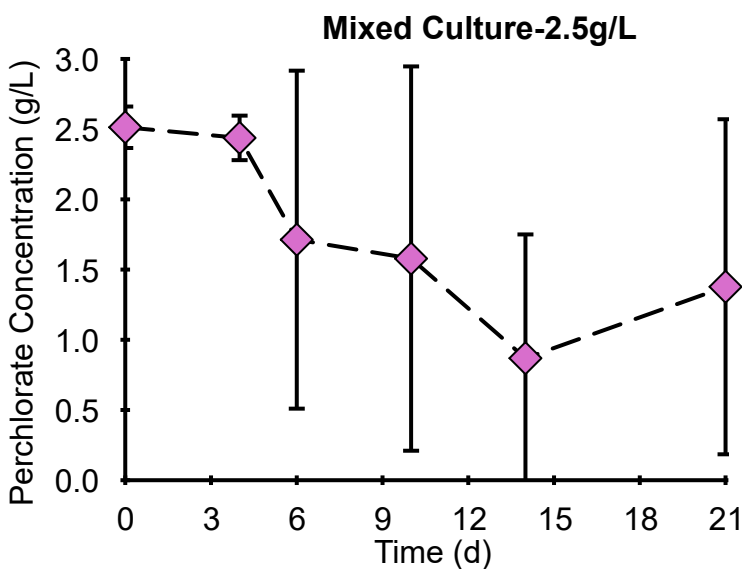
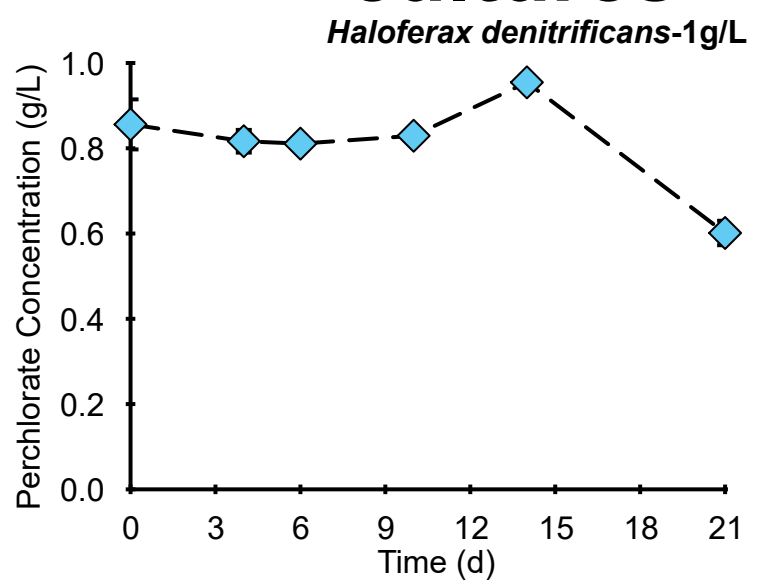
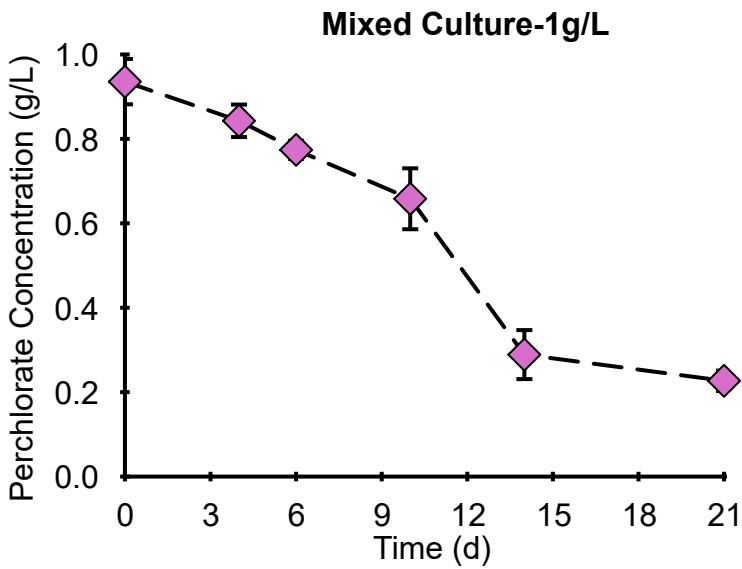
Perchlorate target concentration in the regolith simulant water: 1-8 g/L

Gas composition: 95% CO₂, 3% N₂, 2% Ar (approximating Mars atmosphere)

Acetate as electron donor

Mixed culture enriched from anaerobic digester sludge and pure cultures of *Haloferax denitrificans* and *Dechloromonas denitrificans*

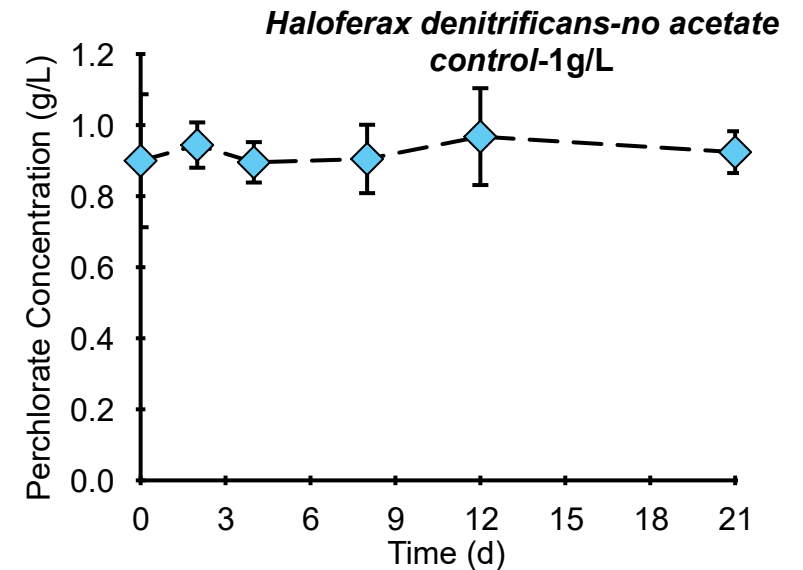
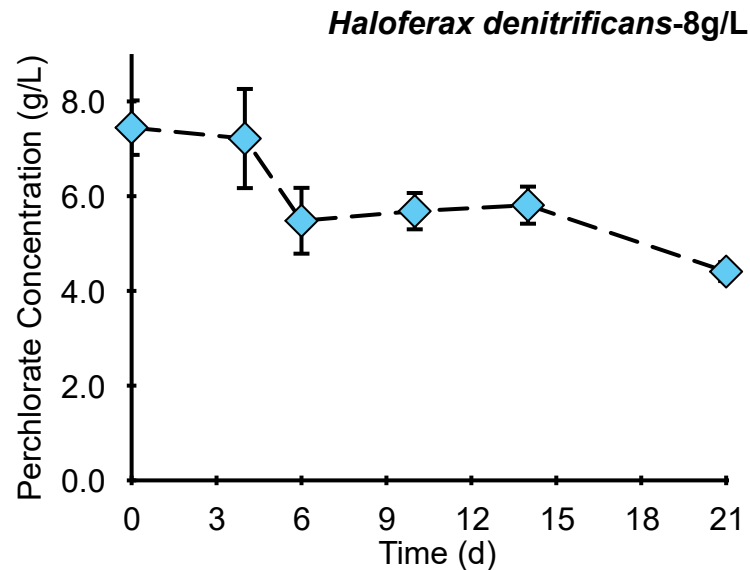
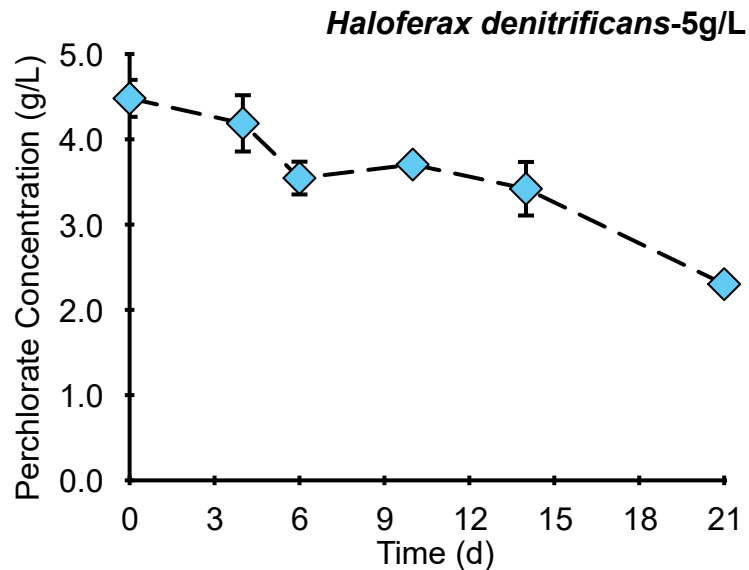
Perchlorate reduction at 1 and 2.5 g/L by mixed and pure cultures



Condition	Mixed Culture		<i>Haloferax denitrificans</i>	
	% reduction	Reduction rate (mg L ⁻¹ d ⁻¹)	% reduction	Reduction rate (mg L ⁻¹ d ⁻¹)
1 g/L perchlorate	75.8%	50.6 ± 2.6	31.4%	13.4 ± 3.6
2.5 g/L perchlorate	45.2%	175.2 ± 148.2	36.0%	35.4 ± 16.9

Between 31 and 75% of reduction was achieved both at 1 and 2.5 g/L perchlorate with the mixed and pure cultures.

Perchlorate reduction at 5 and 8 g/L by the halophilic archaeon, *Haloferax denitrificans*

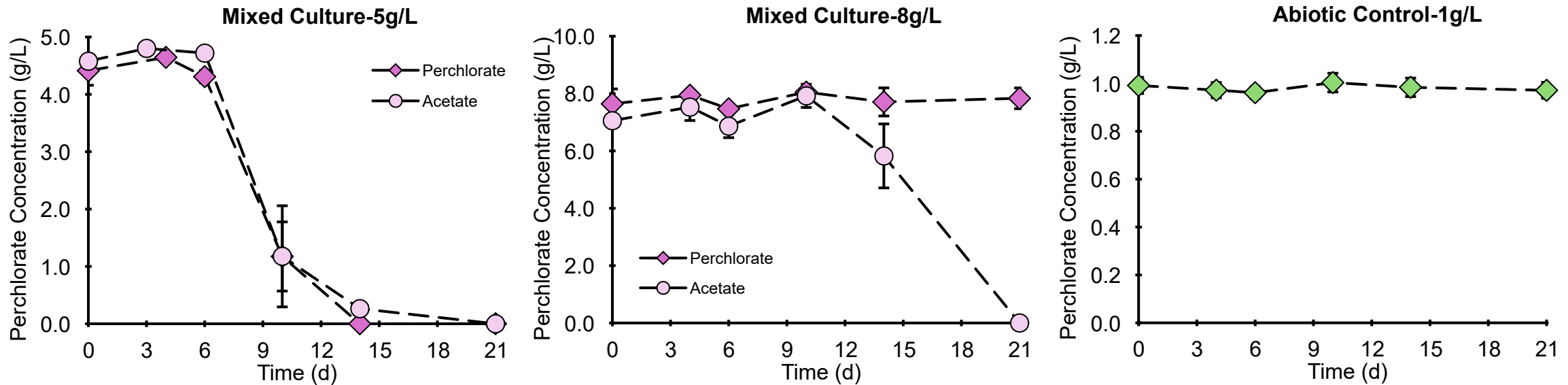


Between 40 and 48% of reduction was achieved with *Haloferax denitrificans* at 5 and 8 g/L.

The reduction rate for the pure culture was the highest at the 8 g/L condition which is the highest ever successfully reduced.

Condition	<i>Haloferax denitrificans</i>	
	% reduction	Reduction rate (mg L ⁻¹ d ⁻¹)
5 g/L perchlorate	48.6%	103.8 ± 9.0
8 g/L perchlorate	40.8%	144.7 ± 29.3

Perchlorate reduction at 5 and 8 g/L by mixed bacterial culture



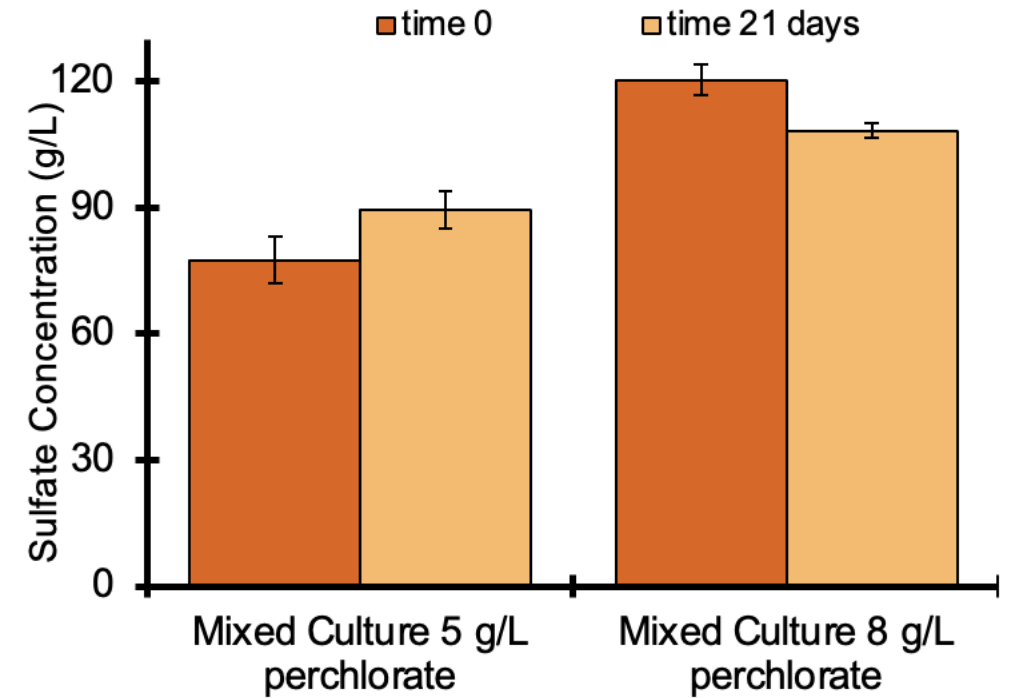
The mixed culture was able to fully reduce 5 g/L but had no reduction at 8g/L.

Acetate was completely consumed in both conditions, indicative of another microbial active process.

Condition	Mixed Culture	
	% reduction	Reduction rate (mg L ⁻¹ day ⁻¹)
5 g/L perchlorate	100%	323.9 ± 34.8
8 g/L perchlorate	0%	N/A

Sulfate reduction was the main process in the mixed culture at

8 g/L perchlorate



Magnesium sulfate is present in Martian regolith simulant at 4 wt.% (40 g/kg regolith)

Key Messages



Microbial perchlorate reduction is **achievable** in simulant at high concentrations relevant to Martian regolith.



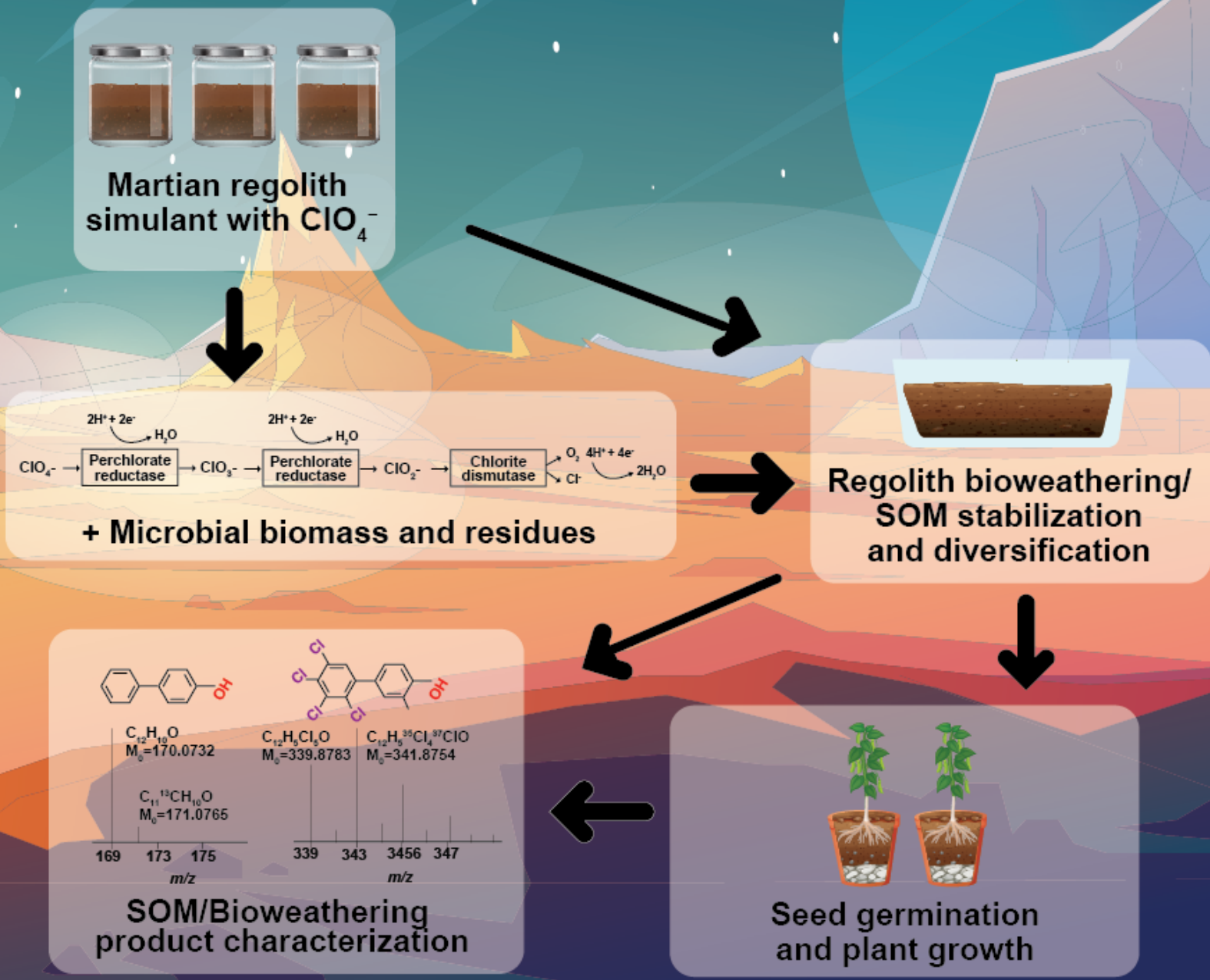
The **choice of culture** will depend on other environmental conditions such as salinity, the presence of sulfate or pH.



Perchlorate in Mars regolith is both a **challenge and a resource**.



Results from this study are also relevant to **bioremediation** efforts at perchlorate contaminated sites.



Delgado Lab



<https://biodesign.asu.edu/environmental-biotechnology/>



NSF EFRI ELiS Award
Number: 2223829

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Thank you!

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