Mars-2020 Landing Site Workshop

Summary of Finding Signs of Past Rock-Hosted Life

on Behalf of the Rock-Hosted Life Working Group

Bethany Ehlmann (Caltech) and TC Onstott (Princeton) February 8, 2017

NOTE ADDED BY JPL WEBMASTER: This content has not been approved or adopted by NASA, JPL, or the California Institute of Technology. This document is being made available for information purposes only, and any views and opinions expressed herein do not necessarily state or reflect those of NASA, JPL, or the California Institute of Technology.
1. Background and Objectives
2. How knowledge of terrestrial life leads to an exploration strategy
3. Examples of biosignatures
4. Summary of biosignatures and exploration strategy
5. Conclusions & Future Work
<table>
<thead>
<tr>
<th>Name</th>
<th>Affiliation</th>
<th>Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abigail Allwood</td>
<td>JPL</td>
<td>NASA Johnson</td>
</tr>
<tr>
<td>Jan Amend</td>
<td>University of Southern California</td>
<td>Princeton University</td>
</tr>
<tr>
<td>Luther Beegle</td>
<td>JPL</td>
<td>Northwestern University</td>
</tr>
<tr>
<td>Roh Bhartia</td>
<td>NASA Ames</td>
<td>NASA Johnson</td>
</tr>
<tr>
<td>Penny Boston</td>
<td>University of Edinburgh</td>
<td>Caltech</td>
</tr>
<tr>
<td>Charles Cockell</td>
<td>JPL</td>
<td>Caltech, JPL, USC</td>
</tr>
<tr>
<td>Max Coleman (Org.)</td>
<td>Caltech, JPL</td>
<td>University of Toronto</td>
</tr>
<tr>
<td>Bethany Ehlmann (Org.)</td>
<td>NASA Goddard</td>
<td>McMaster University</td>
</tr>
<tr>
<td>Jen Eigenbrode</td>
<td>NASA Goddard</td>
<td>University of Colorado</td>
</tr>
<tr>
<td>Danny Glavin</td>
<td>JPL</td>
<td>JPL</td>
</tr>
<tr>
<td>Lindsay Hays</td>
<td>CNRS-Orleans</td>
<td>CNRS-Orleans</td>
</tr>
<tr>
<td>Keyron Hickman-Lewis (student)</td>
<td>Stony Brook University</td>
<td>University of California Los Angeles</td>
</tr>
<tr>
<td>Kai-Uwe Hinrichs</td>
<td>Univ. of Bremen</td>
<td>University of Colorado</td>
</tr>
<tr>
<td>Joel Hurowitz (Tues. only)</td>
<td>Swedish Museum of Natural History</td>
<td>JPL</td>
</tr>
<tr>
<td>Magnus Ivarsson</td>
<td>Georgetown University</td>
<td>Caltech</td>
</tr>
<tr>
<td>Sarah Stewart Johnson</td>
<td>University of California Los Angeles</td>
<td>University of California Los Angeles</td>
</tr>
<tr>
<td>Issaku Kohl</td>
<td>Cal State University Fullerton</td>
<td>JPL</td>
</tr>
<tr>
<td>Sean Loyd</td>
<td>Harvard University</td>
<td>JPL</td>
</tr>
<tr>
<td>Jeff Marlow (Org.)</td>
<td>Institut de Physique du Globe de Paris</td>
<td>University of Hong Kong</td>
</tr>
<tr>
<td>Benedicte Menez</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Joe Michalski</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anna Neubeck</td>
<td>Stockholm University</td>
<td></td>
</tr>
<tr>
<td>Paul Niles (Org.)</td>
<td></td>
<td>Caltech</td>
</tr>
<tr>
<td>Tullis Onstott (Org.)</td>
<td>Maggie Osburn</td>
<td>Caltech</td>
</tr>
<tr>
<td></td>
<td>Aaron Regberg</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Cecilia Sanders (student)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Haley Sapers (Org.)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Barbara Sherwood-Lollar</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Greg Slater</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Nathan Stein (student)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Alexis Templeton</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Greg Wanger</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Frances Westall</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Reto Wiesendanger</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Ken Williford</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Boswell Wing</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Ed Young</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Jon Zaloumis (student)</td>
<td></td>
</tr>
</tbody>
</table>
Several candidate Mars2020 sites have accessible “rock-hosted” habitats for life, which, if on Earth today, would be inhabited (e.g., deep aquifers in volcanic rock, deep aquifers in sedimentary rock)

The 2nd Mars-2020 Landing Site Workshop (August 2015) had many questions about rock-hosted life, especially past rock-hosted life, e.g.,

➢ “What is the astrobiological potential of the subsurface?”
➢ “How much biomass?”
➢ “What are the biosignatures of rock life?”

We set out to answer these and other questions, with funding from NASA HQ (M. Meyer, M. Voytek) and logistical support from the NASA Astrobiology Institute and the JPL Mars Program Office - thank you!

➢ 4 Community Webinars, recorded
➢ In-person meeting of invited experts at Caltech, February 6-7, 2017
➢ Dissemination: This Presentation and A Publication(s)

For more detailed information, go to  http://web.gps.caltech.edu/~rocklife2017/
Successful, Well-Attended Community Webinar Series

Advertised on LPI, PEN, NAI, and C-DEBI email newsletter lists; 30-60 independent logins per telecon

Telecon 1: Martian Environments, Facies, and Ages: Evidence for Rock-Hosted Waters
December 19, 8:30AM PST // facilitated by Bethany Ehlmann, Paul Niles
What is the evidence for ancient Mars environmental conditions? What is the likelihood of habitats for rock-hosted life?
   ppt here | recording here | cited refs list

Telecon 2: Metabolisms and Niches for Terrestrial Rock-Hosted Life
December 20, 8:30AM PST // facilitated by Tullis Onstott, Jeff Marlow
Where rock-hosted life found on earth today? What are its metabolisms and products?
   ppt here | recording here | cited refs list

Telecon 3: Paleo-Rock-Hosted Life Biosignature Detection and Characterization
January 13, 8:30AM PST // facilitated by Barbara Sherwood-Lollar, Haley Sapers
How do we detect signs of paleo (non-extant) rock-hosted life on Earth?
   ppt here | recording here | cited refs list

Telecon 4: Advanced Instrumentation Techniques for Finding Biosignatures
January 23, 9:30AM PST // facilitated by Max Coleman, Paul Niles
What are the latest techniques in biosignature detection, including new capabilities expected in the next decades? (e.g. in mass spectrometry, synchrotron-based analyses, nano-SIMS)
   ppt here | recording here | cited refs list

For recordings, presentations, reading lists, go to  http://web.gps.caltech.edu/~rocklife2017/
Why Focus on An Exploration Strategy for Martian Rock-Hosted Life?

One hypothesis is that the record of ancient Martian life might look much like some aspects of the presently-known early terrestrial record (~3.0-3.7 Ga), i.e., mineralized, (+/-oxygenic) photosynthetic mats, forming laminated structures in near-shore, marine facies on a mostly ocean world.

By 3.5 Ga, Mars’ surface environment had evolved to conditions different and more challenging to life (vs. Earth)

- Earth had had an ocean in continuous existence for 1 Ga. Mars did not.
  - Instead, 8 southern highlands landing sites under consideration had subsurface aquifers and/or systems of episodic lakes/rivers fed by runoff from precipitation or ice melt.
- Mars lost much of its radiation protection early (3.9-4.1 Ga). Loss of magnetic field; thin atmosphere (~1 bar or less)

Martian surface habitats at all 8 landing sites are both more episodic and more extreme than age-equivalent surface habitats on the Earth. Early Martian organisms at the surface faced
  - Cold (at least seasonally sub-freezing temperatures)
  - Surface aridity
  - Surface radiation doses many times higher than that present on the early Earth
  - Low pN$_2$ limiting nitrogen uptake

There is thus a “risk” photosynthetic life would have been rare to absent

On the other hand, subsurface environments were comparatively stable. Data from orbital and landed missions suggest widespread subsurface waters. Consequently, rock-hosted habitats showing evidence of persistent water warrant attention in the search for Martian life.
Specific Objectives and Methods

Our objectives are to develop an end-to-end (living organism to biosignature) understanding of potential traces of past rock-hosted life and then

1. articulate the suite of biosignatures produced by paleo rock-hosted life
2. establish which facies types may preserve them
3. describe measurements can Mars-2020 can make \textit{in situ} to identify potential biosignatures and collect samples with a high probability for hosting biosignatures, identifiable in terrestrial laboratories
4. disseminate findings via presentation at the 3rd Mars Landing Site workshop, a peer-reviewed publication

Key Challenges for Earth Rock-Hosted Life Analogs

- High temperature alteration of the older rocks by metamorphism
- Modern rock-hosted life is common and modern terrestrial organisms eat their older ancestors in the rock for key nutrients. Consequently, most research so far has focus on the relatively near-term past
- Mars may be better for preservation of ancient rock-hosted life!
How Knowledge of Terrestrial Life Leads to an Exploration Strategy
What do we know about terrestrial rock-hosted life?

Biomass concentration varies from $<10 \text{ cells/cm}^3$ to $>10^9 \text{ cells/cm}^3$.

- High cell concentrations and microbial activity occur at redox interfaces where nutrient fluxes (both diffusive and advective, energy and essential trace elements) are greatest.

- Deep subsurface biomass abundance is similar for sedimentary, igneous and metamorphic rocks and usually does not correlate with organic carbon content of rock (with exception of seafloor sediments).

Taxonomic biodiversity varies from location to location and environment to environment from simple to extremely complex, but **functional diversity** has common components.

- **Primary Production** - The **primary producers** are chemolithotrophs many of which use $H_2$ that is produced by multiple abiotic processes (e.g. serpentinization, radiolysis, cataclastic reactions). Metal/sulfide oxidizers also leach/oxidize minerals and glass.

- **Syntrophy** - Complexity appears to build upon recycling of metabolic products to reduce thermodynamic limitations and increase activity between organisms at the same trophic level.

- **Mobility**: Subsurface microorganisms are mobile and will migrate to new food sources or comrades.

- **Evolution**: Subsurface microorganisms and communities evolve through selection and gene transfer to gain functional diversity.

_for more information see telecon #2 and its reading list on the website_
What do we know about terrestrial rock-hosted life?

**Key point 1** SHERLOC hot pixel requires 10 cells; equivalent over volume of observation is $10^3$ cells/gm

Brown = sedimentary rocks; Gray = igneous and metamorphic rocks

compiled from the literature by TC Onstott (see webinar #2)
What do we know about terrestrial rock-hosted life?

Biomass concentration varies from <10 cells/cm³ to >10⁹ cells/cm³.

- High cell concentrations and microbial activity occur at redox interfaces where nutrient fluxes (both diffusive and advective, energy and essential trace elements) are greatest.

- Deep subsurface biomass abundance is similar for sedimentary, igneous and metamorphic rocks and usually does not correlate with organic carbon content of rock (with exception of seafloor sediments).

Taxonomic biodiversity varies from location to location and environment to environment from simple to extremely complex, but functional diversity has common components.

- Primary Production - The primary producers are chemolithotrophs many of which use H₂ that is produced by multiple abiotic processes (e.g. serpentinization, radiolysis, cataclastic reactions). Metal/sulfide oxidizers also leach/oxidize minerals and glass.

- Syntrophy - Complexity appears to build upon recycling of metabolic products to reduce thermodynamic limitations and increase activity between organisms at the same trophic level.

- Mobility: Subsurface microorganisms are mobile and will migrate to new food sources or comrades.

- Evolution: Subsurface microorganisms and communities evolve through selection and gene transfer to gain functional diversity.

*for more information see telecon #2 and its reading list on the website*
Seek – redox interfaces at a range of spatial scales because redox disequilibria drives metabolism
➢ This could start at the orbital scale by identifying lithological boundaries and continue to the rover scale and down even to the PIXL/SHERLOC scale (e.g. sulfate deposits adjacent to serpentinite) or small scale diffusive redox gradients (no fluid flow, just diffusive exchange, alteration haloes).

Seek - lithologic interfaces that indicate high permeability zones for focused fluid flow
➢ Fault zones, dykes swarms, fracture networks, connected vesicles.

Most subsurface cell concentrations, if like Earth and clustered, would be detectable (1 SHERLOC hot pixel requires 10 cells; over volume of observation is $10^3$ cells/gm)

Products of life are more volumetrically significant than life itself (detectable by PIXL and SHERLOC)
➢ Sulfide, carbonate, oxides and other mineral by products
➢ Gas trapped in fluid inclusions
➢ Organics

Model scales spatially from landscape-scale, to hand-scale, to microscopic
Scaling the Exploration Strategy

Seeking boundaries and interfaces at all spatial scales

- from orbit
- Landscape-scale
- Hand sample
- Microscopy
- Thin section
Examples of Biosignatures and the Exploration Strategy from Terrestrial Data
Zones to target for Potential Biosignatures: Example, sedimentary aquifer Fe-redox interfaces

Proterozoic vanadium-enriched reduction spot from sandstone aquifer

PIXL map of 12x12mm area shows concentration of biologically significant elements

Data courtesy of the PIXL team

Sample courtesy Spinks et al. 2010, J. Astrobio.
How biosignatures are preserved for rock-hosted life: Example, Clay/Fe-ox. Mineralization

In the Holocene Hellisheidi cores through Icelandic basalt, microbial cells are associated with clay minerals and Fe oxides in vesicles.

Here, microbial activity facilitates the creation of permeability by dissolution of primary materials (contrast with the “self-sealing” idea of mineralization in hydrothermal systems).

Feed-zones (made of fracture and rubbles) provided flow pathway for CO$_2$ charged ground waters.
Dissolving the rock and feeding microbes (including iron-oxidizers) with aromatic compounds and metals

Trias et al, Nat Com under rev.; Moore, Menez, Gérard, in prep.
Preserved Biosignatures of Rock-Hosted Life: Example, Ancient Colonized Basalt

Fossilized prokaryotes and heterotrophic fungal colonies in basaltic subsurface basalt (8-43 Myr old) Bengtson et al., Geobiology, 2014; Ivarsson et al., PLoS One, 2015
Preserved Biosignatures of Rock-Hosted Life: Example, Ancient Colonized Basalt

Fossilized prokaryotes and heterotrophic fungal colonies in basaltic subsurface basalt (8-43 Myr old) Bengtson et al., Geobiology, 2014; Ivarsson et al., PLoS One, 2015
Preserved Biosignatures of Rock-Hosted Life: Example, Organics from Trace Fossils in Impact Glasses

Ries impact structure, ~15Ma

Quinoic

C=O

Pervasive microtubules in zones of hydrothermal alteration

Organics co-located with morphology

Redox patterns consistent with metabolism

Sapers et al., 2015, Geology; Sapers et al., 2015, EPSL

C K-edge NEXAFS

~Energy (eV) 283.5 285 286.5 288.5 290.3

Fe L-edge NEXAFS

Fe(II) Fe(I) Fe(III)
Preserved Biosignatures of Rock-Hosted Life: Example, Fe-sulfide mineralization

Pyrites (incl. framboidal) are a possible indicator of an ‘active’ sulfur cycle in the presence of organics (as indicated by DUV fluorescence). Sulfides indicate need for further examination for organics and collection.

Framboidal pyrite

Abundant, active endolithic communities in these rocks. Marlow et al., Nature Comm., 2014

Data courtesy of G. Wanger/SHERLOC team
Preserved Biosignatures of Rock-Hosted Life: Example from Deep Carbonate-Serpentine Interface

Klein et al. 2015, PNAS
Fossil Lost City Hydrothermal System, deep rocks
Lw. Cretaceous (Aptian; 125 - 113 Ma)

Lipid analysis standard samples
C32:0 DEG Standard 1pg
C30:0 DEG
C32:0 DEG
C34:0 DEG
standard
standard Archaeol
GDGT-0

Klein et al. 2015, PNAS
Fossil Lost City Hydrothermal System, deep rocks
Lw. Cretaceous (Aptian; 125 - 113 Ma)
Summary of Biosignatures and Exploration Strategy
### Summary: How the Exploration Strategy Leads to Biosignatures in the Examples

<table>
<thead>
<tr>
<th>Initial Observables</th>
<th>Biosignatures</th>
</tr>
</thead>
<tbody>
<tr>
<td>Redox interface, local concentration of trace metals</td>
<td>$\delta^{34}S$ evidence in framboïds (potential)</td>
</tr>
<tr>
<td>Fracture Interface, Clay/Fe oxides, Abiotic Organics</td>
<td>DNA discovery</td>
</tr>
<tr>
<td>Mineralized vesicles, Complex spongiform textures Fe/Mn oxides, microstromatolite</td>
<td>cell-like morphologies, organic matter</td>
</tr>
<tr>
<td>Interface between altered and unaltered amorphous material</td>
<td>microtubules w/ biogenic characteristics, redox gradients/organics co-located w/ tubules, spectral signatures of redox-active cofactors (quinones)</td>
</tr>
<tr>
<td>Redox interface with carbonate mineralization at methane seep, pyrite, organics</td>
<td>aromatic and aliphatic amino acids, DNA</td>
</tr>
<tr>
<td>Mineral interface of serpentine and carbonate, organics</td>
<td>lipids, $\delta^{13}C$ evidence</td>
</tr>
</tbody>
</table>
Characteristics to Look for From Orbit and Rover

Mineral assemblages that indicate habitable waters. Present at all sites

Where to look for the surface expression of the subsurface?
Answer: Ample at some of the landing sites due to faulting and erosion into deep rock units
e.g., Olivine-carbonate/serpentine contacts and zones of discharging waters
e.g., Fe/Mg clays in mineralized fractures within basalts indicating the roots of springs
e.g., Fe redox reaction zones    |    e.g., Fe sulfides

Given heterogeneity (and sometimes low abundance), how are you sure you’ve sampled the right places?
Answer: Seek the interfaces. Seek specific chemolithologic signatures; they are larger than the biomass itself. Sample prospective areas and also employ payload for organics.

How do you know the millions-of-years-old, already discovered rock-hosted life biosignatures are preserved over billions of years?
Answer: The race is currently on on Earth to find the oldest rock-hosted life. Oldest biosignature 125 Ma [Klein et al. 2015], oldest potential (debated) biosignature 3.5 Ga [Stuadigal et al., 2008]. The preservation mechanism is mineral entombment/formation (e.g., in silica, carbonate, or clay). Organics can be preserved, minerals, e.g. sulfide, record a biogenic metabolism. **Same principles as surface life preservation.** A geologically less active planet makes rock-hosted life preservation easier on Mars than on Earth.
Conclusions & Future Work

➢ Ancient Mars aqueous environments included stable, spatially widespread, long lived habitats within rocks. Mars surface was more harsh than time-equivalent ancient environments on Earth (no magnetic field, atmosphere was thin, obliquity cycled, arid, sometimes freezing)
  ○ Aquifers in crystalline rock, aquifers in sedimentary rock should be explored for life

➢ The Exploration Strategy for Rock-Hosted Life is to seek the interfaces (redox and paleo-permeability), has been demonstrated on Earth, and should be conducted at scales ranging from orbit to microscopic on M2020. Also,
  ○ The metabolic waste products (minerals) of rock-hosted life are more numerous that the life itself and are most likely to be identified by the rover
  ○ The spatial clustering of organisms means they are detectable at $\sim 10^3$ cells/gram
  ○ These are a guidepost for sampling for isotopic biosignatures, further terrestrial work

➢ Future investigations of terrestrial analogs
  ○ Further exploration stepping backward in time to equivalent Archaean habitats both to look for biosignatures and to understand the factors that overprint them on Earth, leading to determination of the sweet spot of preservation.
Scaling the Exploration Strategy

Seeking boundaries and interfaces at all spatial scales: A case study at of the sulfate-serpentinite at NE Syrtis

Marlow et al., 2014, Astrobio.
### Summary: How the Exploration Strategy Leads to Biosignatures in the Examples

<table>
<thead>
<tr>
<th>Initial Observables</th>
<th>Biosignatures</th>
</tr>
</thead>
<tbody>
<tr>
<td>Redox interface, local concentration of trace metals</td>
<td><em>d</em>$_{34}S$ evidence in framboids <em>(potential)</em></td>
</tr>
<tr>
<td>Fracture Interface, Clay/Fe oxides, Abiotic Organics</td>
<td>DNA discovery</td>
</tr>
<tr>
<td>Mineralized vesicles, Complex spongiform textures Fe/Mn oxides, microstromatolite</td>
<td>cell-like morphologies, <em>organic matter</em></td>
</tr>
<tr>
<td>Interface between altered and unaltered amorphous material</td>
<td>microtubules w/ biogenic characteristics, redox gradients/organics co-located w/ tubules, spectral signatures of redox-active cofactors (quinones)</td>
</tr>
<tr>
<td>Redox interface with carbonate mineralization at methane seep, pyrite, organics</td>
<td>aromatic and aliphatic amino acids, DNA</td>
</tr>
<tr>
<td>Mineral interface of serpentine and carbonate, organics</td>
<td>lipids, d$_{13}$C evidence</td>
</tr>
</tbody>
</table>