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# PROCEEDINGS OF A WORKSHOP

Joseph R. Schmitt, Rapporteur

Space Studies Board

Division on Engineering and Physical Sciences

The National Academies of SCIENCES • ENGINEERING • MEDICINE

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# Acknowledgment of Reviewers

This Proceedings of a Workshop was reviewed in draft form by individuals chosen for their diverse perspectives and technical expertise. The purpose of this independent review is to provide candid and critical comments that will assist the National Academies of Sciences, Engineering, and Medicine in making each published proceedings as sound as possible and to ensure that it meets the institutional standards for quality, objectivity, evidence, and responsiveness to the charge. The review comments and draft manuscript remain confidential to protect the integrity of the process.

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Courtney Dressing, California Institute of Technology, Marilyn Fogel, University of California, Riverside, Sarah Hörst, Johns Hopkins University, David Paige, University of California, Los Angeles, and David Spergel, NAS,<sup>1</sup> Princeton University.

Although the reviewers listed above provided many constructive comments and suggestions, they were not asked to endorse the content of the proceedings nor did they see the final draft before its release. The review of this proceedings was overseen by Charles F. Kennel, NAS, University of California, San Diego. He was responsible for making certain that an independent examination of this proceedings was carried out in accordance with standards of the National Academies and that all review comments were carefully considered. Responsibility for the final content rests entirely with the rapporteur(s) and the National Academies.

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Searching for Life Across Space and Time: Proceedings of a Workshop

# Contents

| 1 | SETTING THE STAGE<br>Introduction to the Workshop, 1<br>How Likely Is It that Life Exists Beyond Earth?, 3<br>The Limits of Life and Its Interaction with the Environment, 6<br>Is Life a Cosmic Imperative: How Would Thermodynamics Force Life into Existence?, 10  | 1  |
|---|---|----|
| 2 | HABITABLE ENVIRONMENTS IN THE SOLAR SYSTEM<br>Habitable Environments of Ancient Mars: Deciphering the Rock Record, 15<br>On the Habitability of Ocean Worlds, 20<br>Planning for the Exploration of Mars and Ocean Worlds, 25   | 15 |
| 3 | <ul> <li>EXOPLANETS</li> <li>Extrasolar Biosignatures: Developing a Comprehensive Framework for Biosignature Recognition, 27</li> <li>Extrasolar Biosignatures: Thinking Outside the Box, 33</li> <li>Technology Needs to Discover Earth 2.0, 36</li> <li>Prospects for Ground-Based Characterization of Proxima Centauri B, 40</li> <li>General Discussion: Practical Biosignatures that Can Be Exploited to Search for Life In Situ in the Solar System and from Afar on Extrasolar Worlds, 43</li> </ul> | 27 |
| 4 | LIFE DETECTION TECHNIQUES<br>Life Detection: 40 Years After Viking, 49<br>Looking for Life as We Know It on Other Planets, 53<br>Signatures of Life as We Don't Know It, 58   | 49 |
| 5 | INSTRUMENTATION<br>Plume Fly-Through Missions: Detecting Life In Situ at Several Kilometers per Second, 63<br>Life Detection Capabilities of LUVOIR and HabEx, 66<br>In Situ Detection of Organics on Mars, 69  | 63 |

ix

| x   |  | CONTENTS |
|-----|--|----------|
| 6   | FUTURE DIRECTIONS: REPORT OF BREAKOUT GROUPS<br>In Situ Detection of Life as We Know It, 73<br>In Situ Detection of Life as We Don't Know It, 74<br>Remote Detection of Life as We Know It, 75<br>Remote Detection of Life as We Don't Know It, 77<br>General Discussion, 78                       | 73       |
| 7   | WRAP-UP<br>Summary of Chapter 1: Setting the Stage, 81<br>Summary of Chapter 2: Habitable Environments in the Solar System, 82<br>Summary of Chapter 3: Exoplanets, 83<br>Summary of Chapter 4: Life Detection Techniques, 84<br>Summary of Chapter 5: Instrumentation, 84<br>Parting Thoughts, 85 | 81       |
| API | PENDIXES   |          |

| А | Statement of Task                | 89  |
|---|----------------------------------|-----|
| В | Workshop Agenda                  | 90  |
| С | Workshop Participants            | 94  |
| D | Poster Abstracts                 | 99  |
| Е | Biographies of Committee Members | 119 |

# 1

# Setting the Stage

Michael Moloney, the Director of Space and Aeronautics at the National Academies of Sciences, Engineering, and Medicine, opened the December 5-6, 2016, workshop<sup>1</sup> entitled "Searching for Life across Space and Time" by welcoming all those attending the workshop (see Appendix B),<sup>2</sup> both in person at the Arnold and Mabel Beckman Center (Irvine, California) and online watching the webcast.<sup>3,4</sup> The Space Studies Board carries out a major workshop every 2 years for NASA's Science Mission Directorate, who sponsors the workshop. The National Academies' workshops are designed to promote dialogue, not to form a consensus opinion or to make official recommendations. Opinions and recommendations contained within this workshop proceedings are those of the speakers themselves and are not intended to represent the opinions or recommendations of the workshop or the National Academies as a whole. In addition to fostering discussion, Moloney and the Space Studies Board also see this workshop as an early step in preparation for two upcoming decadal surveys—one in astronomy and astrophysics starting in 2018 and one in planetary science starting in 2020.

#### INTRODUCTION TO THE WORKSHOP

The search for life is one of the most active fields in space science and involves a wide variety of scientific disciplines, including planetary science, astronomy and astrophysics, chemistry, biology, chemistry, geoscience, and so forth. These workshop proceedings cover the very stimulating discussions that were held by experts in the various fields about the possibility of habitable environments in the solar system and in exoplanets (i.e., those outside the solar system) and techniques for detecting life and the instrumentation used. The cross-disciplinary discussions were designed to be a highlight of the workshop format.

James Kasting, Pennsylvania State University, and chair of the workshop's organizing committee (see Appendix E), began the workshop by describing the approach the committee took to organizing the workshop, breaking it down into four regions of parameter space (also see Table 1.1):

<sup>&</sup>lt;sup>1</sup> The workshop agenda is included as Appendix A.

<sup>&</sup>lt;sup>2</sup> There were 120 in-person and 92 webcast attendees on day one, and 80 in-person and 72 webcast attendees on day two.

<sup>&</sup>lt;sup>3</sup> The statement of task is included as Appendix C.

<sup>&</sup>lt;sup>4</sup> A summary of the material presented during poster sessions is included as Appendix D.

2

SEARCHING FOR LIFE ACROSS SPACE AND TIME

|                             | In situ detection (solar system bodies)                         | Remote detection (exoplanets)                      |  |  |
|-----------------------------|---|--|--|--|
|                             | Mars  | Artist's concept depicting Kepler 22b              |  |  |
| Life as We<br>Know It       | SOURCE: NASA/JPL/USGS.  | SOURCE: NASA/JPL and NASA/Ames Research<br>Center. |  |  |
|                             | Titan   | S Exoplanet, UCF-1.01                              |  |  |
| Life as We<br>Don't Know It | SOURCE: NASA/JPL/University of Arizona/<br>University of Idaho. | SOURCE: NASA/JPL.                                  |  |  |

#### TABLE 1.1 Four Regions of Parameter Space with an Example of the Type of World in which We Might Find It

- In situ detection in the solar system of life as we know it (e.g., Mars),
- In situ detection in the solar system of life as we don't know it (e.g., Titan),
- Remote detection on exoplanets of life as we know it (e.g., "exo-Earths"), and
- Remote detection on exoplanets of life as we don't know it (target unknown).

These four categories can be used to help guide the approach to detecting life beyond the Earth. Each region has its own characteristic set of biosignatures and will require a different set of technologies, instruments, knowledge, and expertise to determine whether life can or does exist in each environment. This workshop is intended to foster dialogue on the best way to accomplish the goal of detecting life beyond Earth. SETTING THE STAGE

#### HOW LIKELY IS IT THAT LIFE EXISTS BEYOND EARTH?

John Baross of the University of Washington presented the first talk of the workshop on the likelihood that life exists beyond Earth. He started his talk by stating that he would focus on "life as we know it" and leave the "weird life" discussions for later talks. The most important aspect of this workshop, in Baross's opinion, is how life is initially acquired. Panspermia, which includes the notion that life can be transported to Earth by meteorites, comets, or even spacecraft, is one possibility, but the limits of such an occurrence in terms of distance from the source, time of origin, survival, characteristics, and so on are unknown. The biggest challenge in Baross's mind, however, is a de novo origin of life (or abiogenesis). Open questions on abiogenesis include what, if any, are the essential planetary conditions for life and how they might be detected or inferred on other planetary bodies in the solar system and beyond.

#### Life on Earth

Baross then listed the basic requirements for life as we know it. Life uses either light or chemical energy. For example, H<sub>2</sub> was probably the first energy source of microbial systems. Life also requires oxidants. The most abundant elements in the universe (C, H, N, O, P, S, and Fe) are required for life, but also many trace elements like boron, vanadium, tungsten, and nickel. In total, more than 30 elements are required. Baross mentioned that, according to new research, the two earliest catalytic systems on Earth likely required tungsten and molybdenum. *Pyrococcus furiosus*, a high-temperature microorganism from hydrothermal vents, has been found with never-before-seen metalloproteins, including proteins containing lead and uranium. Life also builds catalytic and energy-transfer organic macromolecules around metals and metal-sulfur clusters. Baross said that it is now thought by many that mineral catalysis preceded protein catalysis, providing the backbone of reaction networks that led to metabolism.

Life on Earth has a common ancestor, Baross said. This idea is based on what he called the "unity of biochemistry," which is the fact that all life has the same biochemical and molecular characteristics: the same nucleotide bases, the same 20 amino acids (along with selenocysteine), the same genetic code, lipids with straight, methylbranched chains, and metabolic energetics that use phosphate anhydrides and thioesters. This unity of biochemistry is reflected in the global phylogenetic tree. Baross then asked whether life on other planetary bodies would exhibit a similar sort of unity of biochemistry and if Darwinian selection would allow the most fit genes to survive there too.

Switching topics, Baross said that Earth's geophysical and geochemical characteristics are important because they are the sources of the essential elements and minerals used. He then said he believed that plate tectonics and hydrothermal vent systems are two such essential processes; life as we know it cannot form without them. Plate tectonics can be dated back as far as the end of the Hadean era (~4 billion years [Gyr] ago), according to Baross, although there is wider agreement on a date of 3 Gyr ago. Combining geology with biology is a field Baross calls paleogenetics, which is one of the four ways to approach the issue of abiogenesis. The other three are astrobiology (specifically exobiology and finding alien life), studying prebiotic chemistry to map out the simplest form of life, or creating new life with synthetic biology (see Figure 1.1).

Life is at least 3.7 Gyr old, according to all of the research that has come out, according to Baross.<sup>5</sup> One such paper indicated the existence of stromatolites 3.7 Gyr ago from the Isua sub-crustal belt in SW Greenland, which likely came from a shallow, seawater environment. Three months after the workshop, there was a report of potential 3.77 Gyr old microfossils from a super-crustal belt in Quebec, Canada. The putative fossils were found in iron-containing rocks indicating a seafloor hydrothermal vent setting. If this report is substantiated, Baross said, it would indicate that hydrothermal systems were the earliest habitats for life and perhaps were also instrumental in the origin of life.

Baross said that all evidence points to hydrogen as being the earliest source of chemical energy for both photosynthetic and non-photosynthetic organisms. Methanogens (methane-producing organisms) appear to be ancient and possibly even the root of the Archaea tree. Presumably, hydrothermal vents would have provided

<sup>&</sup>lt;sup>5</sup> There are competing viewpoints. See, for example, B. Grierson, "The Big Debate Over the Oldest Life on Earth," *Discover Magazine*, December 2011.



FIGURE 1.1 The four approaches to studying the origin of life: paleogenetics, astrobiology, prebiotic chemistry, and synthetic biology. SOURCE: John Baross, University of Washington, "How Likely Is It that Life Exists Off the Earth?," presentation to the Workshop on Searching for Life across Space and Time, December 5, 2016.

the ingredients (hydrogen, sulfur, nitrogen, and other compounds) and the needed physical, chemical, and spatial gradients necessary for life.

New discoveries, Baross continued, may lead to changes in the way we think about life on Earth. One such major change is that, according to Baross, the evidence is starting to become overwhelming that the domain of Eukarya might instead reside within the lineage of the Archaea domain (see Figure 4.2), the root of which was likely methanogens. Some of this evidence comes from the Loki hydrothermal vent site in the Arctic Mid-Ocean Ridge, which contained Archaean organisms called Lokiarchaea. Lokiarchaea possess many characteristics originally thought only to be present in eukaryotes, such as a cytoskeleton, membrane remodeling, ubiquitin modification of proteins, and endosymbiosis and/or phagocytosis. The divergence of Lokiarchaeota and Eukaryota may have coincided with a merger with a bacterial endosymbiont (i.e., mitochondria). This paleogenetic approach points to hydrothermal systems as the providers of carbon, energy sources (mainly H<sub>2</sub>), and other essential elements for life, and also possibly as the location of the ancestor of modern-day eukaryotes.

#### Life Beyond Earth

Baross would envisage a search for geophysical processes and water-rock reactions on exoplanets and solar system planets and moons. Enceladus, a moon of Saturn, is an icy body that may have (or have had) active water-rock interactions like the serpentinization systems on Earth that could support life. Serpentinization occurs when olivine  $[(Mg,Fe^{2+})_2SiO_4]$  becomes hydrated and, in the process, produces  $H_2$ ,  $CH_4$ , and other hydrocarbons out to at least  $C_5$ , formate, acetate, and pyruvate. This process results in two things: (1) it raises the temperature to as high as 268°C, and (2) it expands the rock by a factor of about one-third. Serpentinization allows up to 300 kg of water to be taken up by one cubic meter of rock. An oceanic plate undergoing subduction releases this water deep in the lithosphere, which helps give rise to volcanoes and hydrothermal vent systems.

#### SETTING THE STAGE

Enceladus has many of the same properties as the so-called Lost City system of hydrothermal vents in the mid-Atlantic, such as detections of  $CO_2$ ,  $H_2$ ,  $CH_4$ , higher-order hydrocarbons, and a high pH value. However, there are many unknowns in the Enceladus system, like the abundances of important metals, which makes it difficult to say whether there exist the conditions for life. In the Lost City, there is a single species of archaea in the highest-temperature regions involved in methane cycling that can both consume (oxidize) methane and produce it anaerobically. It demonstrates what Baross thinks is a living vestige of mineral catalysis (as opposed to protein catalysis), which he said could be the most ancient catalytic pathway on Earth. Several metals are required though, such as iron, tungsten, selenium, cobalt, zinc, and nickel. Returning to the subject of Enceladus, Baross wondered if tidal heating could mimic subduction in hydrating and dehydrating rocks.

Baross finished by saying that we do not know how life on Earth came to be—whether through abiogenesis or panspermia. Other planets or the moons in the solar system may be habitable if they could acquire the right kind of life. Abiogenesis, he said, would have required a tectonically active, rocky planet with plentiful resources. Finally, he concluded by saying that paleogenetics has inferred that the earliest group of microbes, potentially including the ancestors of modern eukaryotes, were associated with hydrothermal vents.

#### **Audience Participation**

A member of the audience said that, for models of abiogenesis at hydrothermal vents, the  $H_2$  coming from serpentinization reduced CO<sub>2</sub> dissolved in the ocean because there was a large, thick, CO<sub>2</sub> atmosphere at the time. He then asked what the CO<sub>2</sub> source would be on Enceladus and whether this could be the limiting ingredient for life there. Baross said that CO<sub>2</sub> is present there, but was not sure of the concentration. On a similar line of questioning, another audience member then said that the relative abundance of CO and N<sub>2</sub> on Enceladus, both probably primordial, is being debated, considering they both have the same number of atomic mass units. Some CO would convert to CO<sub>2</sub> through OH from water. He then asked what happens when N<sub>2</sub> is put into the system and what kind of biosignature there might be. Baross brought up a proposal from a group who wanted to capture particles in the plume of Enceladus to analyze any organic polymers associated with them. He went on to say that many groups in the international community think that serpentinizing environments were the source of the origin of life. If that were the case, he said, Enceladus would be a good test to see if the correct organic compounds and polymers were there.

A workshop participant then asked whether the trace elements were really necessary for life or if just sulfur and iron were needed. Baross said that there are two approaches to creating life. The "metabolism first" approach is the one most interested in serpentinization because of the gradients present there (like pH). The organisms associated with serpentinization, Baross said, are thought by many to be the most ancient  $CO_2$ -fixing pathway in life. The other approach is making RNA first, but Baross has no idea how to make RNA in a serpentinizing environment. Baross then said that he thinks a global Earth is necessary for the origin of life, not just individual serpentinizing systems that could be habitable for the first living organism.

Moving to the importance of geology for life, another participant at the workshop asked whether having a tectonically active planet was necessary for the origin of life or whether it was only necessary for sustained habitability. Baross answered that, in his opinion, while planets and moons may be able to sustain life, a planet having had a de novo origin of life must have had plate tectonics or other, similar geophysical processes.

A member of the audience then focused on false positives of biosignatures. He said that organics could be produced in serpentinizing systems, but life in these systems could also produce organics. He then asked how one would go about teasing apart these signatures. Baross emphasized that serpentinization is just one of several systems that can produce organics. Greigite  $(Fe_3S_4)$ , for example, can also produce acetate and pyruvate. Even cosmic dust shows abiotic synthesis of organic compounds, such as ether-linked lipids. Baross wished more money would go into understanding the origin of life, such as mineral catalysis and how the synthesis of organics depends on temperature, pH, and other conditions, to better understand how abiotic processes could produce organic materials. The audience member then commented that the very systems that are needed for abiogenesis could also mimic biosignatures. Baross then wondered what the most compelling biosignatures would be to avoid that scenario, thinking maybe that organic biochemistry might have to be combined with other processes, like

6

isotopic analysis or disequilibrium. Another audience member agreed, saying multiple detection techniques are needed to be convincing. She then said that the detection of certain molecules may not be a potent biosignature, but that their relative ratios to other molecules might make them a biosignature. One example she gave was the lipid fatty acid pattern with either a  $C_2$  or  $C_5$  addition, which could suggest life, since abiotic synthesis can only add one carbon at a time.

An example to support Baross's earlier claim that a global Earth is necessary for the origins of life rather than just localized serpentinization systems was then provided by one audience member. He said that trace metals are important for abiogenesis, but at high pH, most of them are very insoluble. Abiogenesis then requires a way to decrease the pH or to produce redox gradients. Atmospheric escape would work for both, and it operates at a global scale. The audience member then said that sugars in prebiotic chemistry are hard to figure out because they're so unstable. He said that reduced carbon, which could have come out of the atmospheric envelopes of red giants, could get irradiated, which would add OH groups to molecules. This cosmic organic matter could provide the unstable carbohydrate-like compounds that would be hard to produce in a hydrothermal system. Baross again said that he supported looking at a global Earth for abiogenesis. He then said that, if the proper minerals are found under certain conditions, sugars will be found. For example, ribose has been found to be catalyzed by borate minerals. He also said that a four-carbon organoboron compound in a ring structure, believed to be very ancient, is used as a signaling compound in bacteria and looks just like ribose. He then wondered whether there could be a compound that is non-ribose that could have formed and served as the early backbone in RNA.

Going back to geology, another workshop participant said that Mars does not have tectonics, but it does contain almost all of the trace elements required for life. He then asked if there was any reason to exclude places like Mars from having abiogenesis just because they don't have plate tectonics, even if they have atmospheric photochemistry and ways to aqueously alter minerals to enhance trace metals. Baross answered by saying that Norm Sleep of Stanford University thinks that they could have been "mushy plate tectonics" in the first tens or even maybe hundreds of millions of years on Mars that could have been a source of subterranean metals. Baross then admitted that any place like Mars that has the key metals and other elements necessary for life could support life, but he still did not think that it could be the origin of life without plate tectonics. He then again emphasized the need to keep these two ideas apart: abiogenesis versus just being able to sustain Earth-like life if it were moved there.

The last person to comment on the talk then posed what he called the "paradox of a biosignature," which he said is the fact that life needs organic compounds that only life can make. Darwinism, however, is the solution to the paradox. For example, he said that homochirality is essential for Darwinism to act on proteins. Isotope effects do not do a good job of this because they only indicate how the molecules, particularly carbon, were fixed. An unusual fixing process could produce a large isotopic effect abiotically. Finishing, he then addressed a previous audience member and said that carbohydrates are indeed unstable in alkaline conditions of pH 12. However, this brings up the question of whether the planet is always flooded. If not, then carbohydrates could be quite stable in areas where there are lots of evaporites, such as boron. However, on Europa, there may never have been dry land to concentrate these elements and molecules. Additionally, these substances would suffer from dilution in the oceans as well.

## THE LIMITS OF LIFE AND ITS INTERACTION WITH THE ENVIRONMENT

Tori Hoehler of the NASA Ames Research Center began his presentation by recalling the previous report by the National Research Council called *The Limits of Organic Life in Planetary Systems*<sup>6</sup> (2007), which has become known as the "Weird Life Report." Although that report explored the limits of all potential life, Hoehler planned to focus his presentation on the limits of life as we know it and trying to create a link between them.

#### **Requirements for Life**

The so-called "Weird Life Report" identified four fundamental requirements for life (in order of decreasing certainty): thermodynamic disequilibrium (Gibbs free energy), an environment capable of maintaining covalent

<sup>&</sup>lt;sup>6</sup> National Research Council, The Limits of Organic Life in Planetary Systems, The National Academies Press, Washington, D.C., 2007.

#### SETTING THE STAGE

bonds (especially between C, H, and other atoms), a liquid environment, and a molecular system that can support Darwinian evolution. The report went on to say that thermodynamic disequilibrium "is not disputable as a requirement for life. Other criteria are not absolute."<sup>7</sup>

Earth life, Hoehler said, only uses a small subset of available energy forms, light and chemical energy, and even then only a small subset of those two forms. For example, life is only known to use light in visible to near-infrared wavelengths and is only known to capture the energy released by oxidation-reduction reactions. Both of these processes create electron flow, which appears to be fundamental to the processes by which Earth life captures and stores energy. It is not clear if this constraint holds for all life. Hoehler then compared life to a laptop computer, saying that both process information at the expense of energy, and both have two types of energy requirements. The laptop requires a minimum voltage (energy per quantity of electrons) and a minimum power (energy per unit time). Life as we know it has analogous requirements: Gibbs energy change (energy per quantity of substrate consumed) and power. Both must be met at minimum levels for life to function properly. There is also a corresponding maximum amount of voltage and power life can handle.

Hoehler then addressed the other three requirements for life from the Weird Life Report. Noting that Darwinism is rarely considered when talking about habitability or biosignatures, Hoehler said that it is nevertheless a fundamental aspect of life as we understand it. The requirement for a molecular system capable of such evolution significantly constrains life's requirements. For example, if Darwinian evolution is fundamental to biology, then information processing is a core attribute of life, and this would require molecular recognition with a very high level of fidelity—a requirement that may limit the range of chemistries, solvents, and environments that are suitable for life. Hoehler described molecular recognition as one example of reversible (i.e., non-covalent) molecular interactions which, although fundamental to the way our life works, are not always considered when defining life's requirements. Hoehler used the example of a ribosome to distinguish covalent and non-covalent interactions. A ribosome is composed of covalent bonds (electrons shared between atoms). Its job, protein synthesis, is ultimately to create covalent bonds. It does this job only by virtue of non-covalent interactions. For example, the folded 3D shape that gives the ribosome catalytic function and supports the recognition of specific amino acids is made possible by a myriad of interactions caused by non-covalent forces within the molecule. The strength and nature of those forces depend as much on the solvent (water) as on the molecule itself. Other liquids that are considered as potential solvents for life must therefore be evaluated on their potential not only to support the synthesis of covalent bonds, but also to support the wide range of non-covalent interactions that confer "life-like" function on a system.

Hoehler then went on to discuss the basic elements that life on Earth needs, the chemical symbols of which spell "CHNOPS" (carbon, hydrogen, nitrogen, oxygen, phosphorus, and sulfur) or "SPONCH" (sulfur, phosphorus, oxygen, nitrogen, carbon, and hydrogen). Carbon is used as the scaffolding element that allows for a large diversity of molecular structures. That diversity is greatest when carbon is in the intermediate oxidation state (between  $CO_2$  and  $CH_4$ ).<sup>8</sup> Heteroatoms (nitrogen, oxygen, phosphorus, and sulfur) support a diverse range of covalent chemistry and also have polar bonds that allow for a variety of non-covalent interactions. Hydrogen provides hydrogen bonding (obviously), which is part of what allows for high-fidelity molecular recognition in our biochemistry. Watson-Crick base-pairing in DNA, for example, is based on hydrogen bonding between complementary nucleobases. Potential alternatives to these elements, Hoehler said, would have to fill these same roles and do so as part of molecules that are stable over meaningful time scales.

Hoehler then described the known environmental limits of life on Earth. Life, he said, is found between -25°C and 122°C, at pH between 0 and 13, at pressures up to at least 200 MPa, and at water activity as low as 0.6. (Water activity is defined as the vapor pressure of water over the solution in question divided by the vapor pressure of pure water at the same temperature; it is a measure of how chemically "available" water is in a given solution and decreases as the concentration of solutes, like salts, increases.) These limits represent the known record holders and are often established in laboratory settings where factors other than the extreme being considered are optimized. Hoehler said that life in natural environments, where other factors are not always optimal, may not be able to reach

<sup>&</sup>lt;sup>7</sup> Ibid, p. 8.

<sup>&</sup>lt;sup>8</sup> Life wants carbon in an intermediate oxidation state, and on Earth, the carbon available is fully oxidized in  $CO_2$ . To perform this chemical reduction, life needs a source of electrons, which it finds abundantly in water (in the bonds between O and H).

these same extremes. It also may not be possible for life to originate at these extremes. Whereas existing life has had the benefit of extensive evolution by which to develop tolerance to extremes, the conditions that foster prebiotic chemistry may be narrower. Life at the limits may sacrifice diversity, abundance, and productivity just to survive. As with solvent and elemental requirements, Hoehler said, the environmental conditions suitable for life must support both covalent bonding and non-covalent interactions. The latter may define a much narrower range of possible environments than the former.

#### How Life Can Alter Its Environment

Hoehler then moved onto the subject of what life can do to alter its environment and the planet as a whole. The potential to do so depends on how abundantly life's requirements are met. For example, biomass densities on land range from practically nothing in the most arid regions of the planet to hundreds of kilograms per square meter in the rain forests, a variation that reflects the availability of water. The oceans have 3 to 4 orders of magnitude variation in the density of photosynthetic biomass, and this variation reflects the availability of nutrients and key elements. Such differences could influence our ability to detect life on another world. Moreover, Hoehler noted, we must be mindful that our intuition about what an inhabited world looks like is based on a world, Earth, in which life's requirements are abundantly met. The same may not be true of some of the worlds currently considered as potential abodes of life. Hoehler pointed to the availability of light energy to Earth's biosphere, but not to worlds like Europa or Enceladus, to quantify this point. The amount of energy available (such as from light) can limit the ability of life to create recognizable biosignatures. On Earth, photosynthetic organisms capture about 1 percent of the Sun's 173,000 TW of power incident on the top-of-atmosphere and create chemical energy in biomass at the rate of 63 to 105 TW. Comparatively, non-photosynthetic chemical energy fluxes on Earth, such as the flux of hydrothermal vent fluids into the oxidizing ocean, amount to only about 0.006 TW in forms that can be utilized by life. The way in which we search for life must take these differences and their likely impact on the abundance and quality of biosignatures into account.

Energy flux, Hoehler continued, places upper limits on several aspects of life. Energy constrains the abundance of biomass sustained. Energy flux also constrains metabolic and biosynthetic rates and thus the rate at which biosignature molecules can accumulate in the environment. This rate is important because low accumulation rates can make it difficult to maintain a pool of biosignatures against physical, chemical, or biological attrition. An example is the racemization of amino acids, which can exist in either of two non-superimposable (i.e., chiral) mirror images called "enantiomers." Earth life typically produces/utilizes one enantiomer exclusively, while abiotic mechanisms generally produce a mixture of both enantiomers. Finding a large excess of one enantiomer over the other would thus serve as good evidence for life. However, chemical processes spontaneously interconvert (racemize) the two enantiomer forms, thereby continuously erasing the signature of life. When energy fluxes are very low, the rate at which the biological signal is replenished by biosynthesis may be overwhelmed by racemization.

When living organisms do have access to abundant energy, Hoehler explained, a commonly mentioned biosignature is the presence of a disequilibrium. He said that, for such cases, the signature of life lies not just in the presence of a disequilibrium, but in its type and magnitude, considered within its environmental context. Oxygen-producing photosynthesis, for example, is not an inevitable outcome of a photosynthetic environment. Its occurrence on Earth is the result of a specific biochemical need expressed in a specific environmental context. Extracting electrons from water yields  $O_2$  as a by-product. Life that either has access to carbon in an intermediate oxidation state, uses a different biochemistry, or has a different solvent might not yield this same by-product of photosynthesis. Moreover, it is important to consider that thermodynamic disequilibrium can result not just from biological processes, but also abiotic ones. For example,  $O_2$  can form abiotically when sufficiently energetic photons fragment oxygen-containing molecules like  $CO_2$  or  $H_2O$ . In an Earth-like setting, abiotic processes can produce about  $10^{11} O_2$  molecules/cm<sup>2</sup>/s. Life on Earth, on the other hand, can produce  $10^{19} O_2$  molecules/cm<sup>2</sup>/s. Thus, the fact that Earth displays evidence of its biosphere in the form of an oxygen-rich atmosphere derives not from the uniqueness of  $O_2$  as a biological product, but from the much greater efficiency (8 orders of magnitude) with which life uses sunlight to create  $O_2$ . More generally, this illustrates that for "disequilibrium biosignatures," it is the irpresence, that constitutes the sign of life.

#### SETTING THE STAGE

Hoehler concluded that, when searching for evidence of life as it appears on Earth, one must consider how and why life does what it does physically and chemically—for example, thinking of the potential for alternative solvents or biochemistries to support non-covalent interactions—which significantly constrains what life can be and do and what evidence it may leave of itself. Lastly, he emphasized the importance of changing our concept of habitability from a binary construct (1 or 0, life either being present or not) to one in which the abundance and productivity of life are seen as a continuum of possibility that depends on a range of environmental factors. Thinking in this way would allow us to distinguish among environments that may have greater or lesser capacity to express evidence of a biosphere.

#### **Audience Participation**

One audience member challenged Hoehler's claim that the limits of life have been established primarily in laboratory settings. He said that there are many organisms in nature that are unknown, saying, "The truth is out there." Hoehler clarified that the currently known limits of life generally correspond to observations made in a laboratory setting, rather than to organisms or biological activity in natural settings. He also said that natural environments provide the feedstock to explore the biochemical breaking points, but that the most extreme limits are often expressed in the laboratory under optimal conditions. The audience member thought that this was still a naïve point of view. He said that he does not think that the conditions these organisms require can be produced in laboratories effectively. For example, microbes are often smothered by what we give them. Hoehler agreed with this point. He said that it is uncertain whether the ultimate limits on life are going to be found in nature or in a laboratory.

A workshop participant then referred to Hoehler's discussion of the upper limits imposed on biosignature formation by energy flux and asked why he placed metabolic rates and biosynthetic rates together on the same line. Part of the reason, Hoehler said, was to save time and space. However, both have also been considered as potential biosignatures. Biosynthesis creates clearer biosignatures, because it manufactures molecules that are abiotically improbable to produce. However, metabolic intermediates and end products can also serve to indicate biological activity if they have not achieved equilibrium with the environment or bear the hallmarks of biological catalysis, such as isotopic discrimination. More properly though, Hoehler agreed that they should be considered separately, as they are not directly coupled.

Another audience member then said that Hoehler had convinced her that using disequilibrium as a biosignature would be impossible because of the difficulty with disentangling the abiotic sources from the biotic sources. Hoehler agreed that it would be hard to say definitively that disequilibrium is a biosignature without better understanding the environmental context, which would be hard to see on a planet that is simply a point of light. However, he thinks that examples like creating  $O_2$ , at which life is 100 million times more efficient than abiotic sources, could be indisputable if placed in the proper context. The audience member countered, saying that if we know the environmental context, we would also already know whether or not the planet had life. The challenge, she said, is for less visible signatures. Hoehler said that that could be true. He gave photosynthesis as an example that has yielded two biosignatures visible from space:  $O_2$  and the "red edge" (the fact that photosynthetic plants are highly reflective in the near-infrared).

A question from an evolutionary biologist asked whether Darwinian evolution is a fundamental feature of life and whether a diversity of life would be possible without it, considering that the process is often included in the definition of life. Hoehler answered that he thinks that it is an essential part of life. He noted Steven Benner's description of evolution as the answer to the "paradox of a biosignature"—the observation that life depends on complex, improbable molecules that only life can create in the first place. Hoehler noted that evolution, despite its central place in biology, is not usually considered when discussing the inherently biological concept of habitability because it is not, in general, directly observable. Lastly, he said that evolution is especially interesting in places where the energy flux is very low, because that would affect the rate at which evolution can explore the possible parameter space of genetics. A simple linear extrapolation, he said, might be a naïve idea. He concluded by speculating whether there may be a critical level of energy flux below which evolution is not possible.

10

#### IS LIFE A COSMIC IMPERATIVE: HOW WOULD THERMODYNAMICS FORCE LIFE INTO EXISTENCE?

Eric Smith of the Santa Fe Institute discussed life as a cosmic imperative and whether the existence of a biosphere might be viewed as the ineluctable result of thermodynamics. He said that there is an idea that the emergence of life is a necessary stage in planetary evolution. Evidence of this may be hidden in biochemistry and higher-level architecture of cellular organization. However, work needs to be done to change this from just hand-waving storytelling into a solid theory. Lastly, he wanted to address how to extend this idea and subsequent analysis to exoplanets.

#### The Ancient Geochemical Landscape for Life

Smith noted the importance of the concept of system-level order to the question of whether life could be a necessary step in a planet's evolution. Addressing this question, he argued, begins with a familiar mathematical concept: the phenomena of breakdown processes, which can be recognized as robust states of dynamical order on short time scales. Familiar physical examples include plasma channels (e.g., lightning), convective storms, and fracture propagation through materials. (In contrast, while diffuse energy stresses of the familiar sort such as heat do not typically create order, breakdown phenomena differ in that they generally result from feedback processes that can focus this energy to create order.) Of the many disequilibria found on Earth, the one that might best explain the emergence of a biochemistry is redox disequilibrium, which is deeply connected to life. On the early Earth, this disequilibrium was driven primarily by hydrogen escape. Photolysis of water vapor can result in the Jeans escape of hydrogen leading to an excess of oxygen in the upper atmosphere. This process has likely occurred throughout Earth's history and maintains a state of persistent redox disequilibrium between the atmosphere/oceans and the bulk Earth. (Such a disequilibrium would not persist in a system dominated by diffusion.)

Smith continued, explaining the consequences of this disequilibrium and how it behaves at large scales. Tracing the emergence of this disequilibrium, he noted the bulk Earth's oxidation state is between Fe and Fe<sup>+2</sup>, which forms a redox gradient between the atmosphere, which shows relatively higher oxidation states, usually between Fe<sup>+2</sup> and Fe<sup>+3</sup>. This is seen in the Earth's two major reservoirs of water: seawater and sub-surface water are each brought to their respective equilibriums, which as noted are at two very different oxidation states. Mixing zones between the two can produce redox potentials of several tenths of an electron volt at distances of just a few atomic diameters. Today's living systems use these potentials to produce chemical order. The question this raises, of relevance to the origin of life, is whether we can infer a link from the earlier abiotic geochemical processes to today's redox potentials of the sort observed in these mixing zones.

Smith then returned to his initial question of whether life is a necessary step in a planet's evolution. If we are correct that the gradient in redox potential is the relevant geochemical disequilibrium that allows for life, where, Smith asked, might we find evidence of this in the biochemistry? He said that one thing to look for is continuity of ancient, universal biochemical signatures with selective organic geochemistry observed today. The ancient signatures include those that either became "locked in" and are unchanging, or are paths of least resistance, or small-molecule chemistry unchanged by modern enzymes. A second place to look for verification of the role of redox potentials is in what Smith calls "upward causation," or metabolic patterns imprinted on higher levels of cellular architecture where you would otherwise not expect such imprints to be, since information usually devolves, in the opposite direction, from higher to lower levels. (This is further discussed below in the subsection, "Upward Causation".)

#### The Core of Metabolic Biochemistry

Smith then went on to discuss metabolism and the degree to which it has or has not experienced innovation during the 4 billion years of life on earth. He showed a slide illustrating a "universal core" of metabolic biochemistry that is visible always at the ecosystem level, sometimes within the level of particular organisms, and that is organized around the citric acid cycle (see Figure 1.2). (The original version of this cycle is now known to be its reductive version, not the oxidative direction of cycling that was first discovered by Krebs.) Of the six autotrophic



includes all six known carbon fixation pathways, the network of sugar-phosphate chemistry, and (schematically) the two major pathways of lipid synthesis (fatty acids and isoprenoid alcohols). The citric acid cycle is highlighted in yellow. Right: the sub-graph containing the four self-amplifying loop-autocatalytic fixation pathways Biology 10(1):011001, 2013 https://doi.org/10.1088/1478-3975/10/1/011001, copyright IOP Publishing, reproduced with permission, all rights reserved; presented in FIGURE 1.2 The citric acid cycle occupies the center of the biosynthetic core of life. Left: A graph covering the universal reactions of CHO-biosynthesis, which that remain close to the citric acid cycle. Each pathway is drawn in a different color. Their overlap shows the segment-wise nature of pathway innovation and the way Eric Smith, Santa Fe Institute, "Is Life a Cosmic Imperative: How Would Thermodynamics Force Life into Existence?," presentation to the Workshop on Searching all pass through the pinch-points of acetate and succinate. SOURCE: R. Braakman and E. Smith, The compositional and evolutionary logic of metabolism, *Physical* for Life across Space and Time, December 5, 2016. carbon fixation pathways that are currently known, five are self-amplifying loops, and four of these remain close to the template of the citric acid cycle. (The only loop that is not closed is the Calvin-Benson cycle.) The evolutionary innovation that appears to have given rise to these other loops usually takes place at the pathway segment level, and the cycles as a whole stay very close to the citric acid cycle intermediates. If these intermediates are not all in a particular cycle that fixes carbon, they are added by auxiliary pathways known as anaplerotic reactions. Two pinch points in the map of cycle diversification are acetic acid in the form of acetyl-CoA and succinic acid, which may or may not be succinyl-CoA.

Smith then opined that there has been almost no innovation in carbon fixation over the last four billion years. This set of autotrophic reactions essentially consists of easy pair transfer chemistry in a water environment (twoelectron reductions) or hydrations and dehydrations. The distinguishing points of departure for different pathways tend to be their opening steps, which are typically a carbon attachment followed immediately by a reduction. Those reactions are also usually associated with strictly conserved proteins with metal centers at the active site. Many researchers have noted the similarity between the metal center proteins and mineral metal centers. Certain transition metals allow for coordination geometries that can be adjusted by evolution. Smith then agreed with Baross's talk that said that mineral chemistry appears to have been placed into a control system.

#### **Upward Causation**

Smith then moved on to upward causation—that is, the imprint of patterns that are native in metabolism being found at higher levels of cellular architecture. The critical question is the compatibility of this evidence of upward hierarchical movement of information with the dogma of the genetic code, which argues for information moving in the opposite direction. In particular, the process of translation from RNA to peptides should acts as a firewall that insulates the fundamental structural non-symmetries of biosynthetic chemistry from the protein sequencespace in order to allow Darwinian selection to pick the best sequence without having to overcome arbitrary biases. The expectation that translation should be a firewall also suggests that the codon assignments of amino acids to nucleobase triplets should be arbitrary, in the sense that they could have been different than they actually are.<sup>9</sup> The actual genetic code, however, does not really have arbitrary assignments; in information-theoretic terms, the genetic code is enormously *compressible* and could, for example, be represented as a decision tree for the selection of amino acids.<sup>10</sup> Thus, it turns out that the first base in the genetic code tells you which citric acid cycle precursor the amino acid that the codon specifies is built from. Thus, for example, if the first base is C, this precursor is glutamate; for A it is aspartate, and for U it is pyruvate. And what the second base is telling you is how these precursors have been modified; for example, if U is the second base, a second copy of pyruvate is added to make hydrophobic amino acids. The evolutionary steps that have produced this type of regularity are a mystery that needs to be explained.

#### Thermodynamics, Kinetics, and Trajectories

Smith's next point was that the maintenance and error correction of systems in disequilibrium are revealed not only by making lists of the structures they form, but more so by considering kinetics and trajectories (i.e., time evolution of a system in phase space). Smith said that thermodynamics is not fundamentally about energy or equilibrium, but rather is about the emergence of a stable macro-world, and that this idea needs to be applied to disequilibrium environments (i.e., those in which the system has a tendency to evolve to a new macrostate) to explain the robustness of the error-correcting processes that operate in the biosphere. A mathematical system to do this, he said, already exists.<sup>11</sup> He went on to explain what is meant when saying the fundamental concept behind thermodynamics is the emergence of macro-phases. The general idea is that, under aggregation of their

<sup>&</sup>lt;sup>9</sup> One can back off slightly from the assumption of arbitrariness by recognizing that coding happens with errors, and error buffering can be provided by grouping similar amino acids at related codons.

<sup>&</sup>lt;sup>10</sup> S.D. Copley, E. Smith, and H.J. Morowitz, 2005, A mechanism for the association of amino acids with their codons and the origin of the genetic code, *Proceedings of the National Academy of Sciences U.S.A.* 102(12): 4442-4447.

<sup>&</sup>lt;sup>11</sup> See, for example: Touchette, H. 2009. The large deviation approach to statistical mechanics. *Physics Reports* 478: 1-69.

#### SETTING THE STAGE

constituent particles and components, systems and the probability distributions that describe their fluctuations can converge on exponential families.<sup>12</sup> This convergence is why systems of large and definite size are possible. The scale of the system separates from the structures the system is capable of taking on—the macrostructures. This is true for equilibrium systems, but, dynamically, the same convergence to exponential families is possible. In the mathematical system noted above, the role played by equilibrium free energy is subsumed by an effective action associated with the trajectory, and dynamical phase transitions are the shifts of the central tendencies of macrostates. Making an analogy with modern electronics and its incorporation of error correction, Smith offered that thermodynamics is in effect an error correction process. The mathematics of fluctuations leads us to expect that error correction in evolution will tend to have a three-way tradeoff between robustness (the probability of error), the complexity of the number of states capable of being maintained, and any associated costs (such as the time or system size needed to take a system back to its central tendency).

In a view of life based on dynamical phase transitions, Smith explained, the abiotic Earth becomes a dynamically metastable condition. The most probable path (also known as "the maximum path entropy condition" or "path of least resistance") will be the path for which there are the most ways to scatter into and the fewest ways to scatter out of. This explains part of the importance of "easy" chemistry, such as water-base pair and group transfer. Catalysis of these reactions is extensively duplicated; enzyme families are either divergent or recurrently evolved. "Hard" chemistry is the electron transfer chemistry, which isn't possible due to radical production in water being disfavored. The need for single-electron transfer processes in biochemistry suggests a mineral or metal-ligand complex origin for these processes. The whole network needs a positive, self-amplifying feedback to concentrate matter and energy flows. Short feedback loops and feedbacks with few alternatives are best suited for this because they have less of a problem with diffusion while also allowing for the evolution of control mechanisms.

#### **Problems and Future Work**

Smith then went through some outstanding problems with this explanation. A major problem is that the pathways that seem specific and necessary biologically do not seem inevitable or special geochemically. Another problem is that reactivity is self-defeating. For example, a one-carbon reduction sequence from CO<sub>2</sub> to CH<sub>4</sub> goes through CH<sub>2</sub>O (formaldehyde), which is extremely reactive. For the origin of life, the process needs a kinetic way to focus reactivity into a thermodynamically disfavored domain because that is the only place where reactivity is available. Recognizing this allows us to see that the formose reaction (i.e., the formation of sugars from formaldehyde) is in a different disequilibrium class than processes of reductive carboxylation, such as Fischer-Tropsch reactions. In the formose reaction, electrons cannot enter or leave the system, so they are trapped on a surface of redox constraint, ensuring that reactivity is preserved. In reductive carboxylations, the flow of electrons into or through the system is the driving force behind creating organic complexity, but there is no natural constraint to preserve reactivity. The contrast between reductive carboxylations and constrained systems such as the formose reaction underscores the problem of understanding what the right concept of disequilibrium should be. Smith gave another example of this problem: the dominant motifs in biochemistry are cascades of group transfers, which can be understood partly in equilibrium-thermodynamic terms because they take place in a context that Yoshitsugu Oono has termed "compartmented quasi-equilibrium." In contrast, most origin-of-life research emphasizes cycling of physical conditions such as temperature or water activity so that the system "chases equilibrium," which differs from the group transfer just mentioned.

Referring to the understanding of thermodynamics showing that little extra structure is needed to make exergonic reactions feasible, Smith said that a similar knowledge is needed for kinetics. He gave a list of things that need to be better known: mineral-metal center catalytic selectivity and activity, including edge, vertex, and face effects; and roles of impurities and mineral-mineral interfaces. He said that we need to combine our knowledge of ligand field theory in a mineral context with the same for soluble metal-ligand complexes. Smith then said that a connection needs to be made between the Hadean atmospheric and oceanic conditions, such as the redox state of the atmosphere (CO vs.  $CO_2$ ).

<sup>&</sup>lt;sup>12</sup> Koopman, B.O. 1936. "On distributions admitting a sufficient statistic". Trans Amer. Math Soc. 39(3):399-409.

In his closing thoughts, Smith said that we need to stop just trying to get to an organic material and instead think about how the organics were created out of equilibrium. He then emphasized that big and random molecules are not necessarily complex. Complexity implies selectivity. Finally, he said that disequilibria (e.g., redox, radio-activity, thermal activation, and dehydration) are not all the same in the context of the origin of life.

#### **Audience Participation**

A member of the audience asked Smith whether there is enough of a free energy gradient on Enceladus and Europa for abiogenesis, which is what is needed in a metabolism-first method. Smith said that the Archean era on Earth had a longstanding disequilibrium due to having a reduced interior with a steady process of Jeans escape of hydrogen from the upper atmosphere. He further responded that the important question to answer is where the terminal electron acceptors are produced on Enceladus. (Smith later learned in off-line conversation that the production of peroxides on surface ice through particle bombardment is believed to be the major process generating such acceptors.) There needs to be a flow of electrons. He asked whether there is a way to get a disequilibrium of multiple tenths of an electron volt at distances of just a few atomic diameters, as the key consideration.

A potential problem in reconstructing history using modern day biochemistry, another workshop participant said, could have been flexibility in the earliest stages of Earth and life. Gibbs free energy has been suggested as a way to do this historical reconstruction. She then asked if there was any alternative way to try to reconstruct the past. Smith said that biology is overwhelmingly preoccupied by the role of innovation. However, the discovery of chemoautotrophy in the 1970s led to the realization that there is a chemoautotrophic core in everything with oxygenic shells wrapped around it. There is also the question of the extent to which evolution just wrapped controlling systems around pre-existing processes versus how much it actually built new, innovative pathways. The facts that the deep core of metabolism is so conservative and that the innovations built with it are pretty serial suggest to Smith that error correction is a difficult thing to invent. Therefore, the reconstruction of deep history may not be as difficult as it is usually expected to be by biologists because biology has been dominated by an emphasis on historical contingency. The examples of the architecture of biochemistry were adduced to suggest that in the very deep past, the role of direct historical reconstruction may give way to a science of prediction based on first principles, which are inferred from the structural regularities in this architecture. In agreement with the questioner, Smith actually finds the possibility of flexibility in early stages of Earth and life an important and interesting question.

Moving the discussion away from Earth, an audience member said that, on ocean worlds, icy and solid materials formed together under certain conditions, but once together again at lower temperatures, they were in disequilibrium. He then asked Smith why this long time period where the icy and solid materials are trying to approach equilibrium is not the same as the reductive Earth interior with Jeans escape of hydrogen. Smith said that he is open to the idea that it need not be fundamentally different.

A workshop participant agreed with Smith's talk that there appears to be a serial set of innovations to get to the necessary biomolecules for life. She then said that there is an idea to focus on intermediate molecules as a biosignature that would not necessarily be favored in an abiotic system and asked Smith to comment on that. Smith broadly agreed with the position presented by the questioner. Smith said that there are two views of life, one that is all about innovation and one that is all about conservation with a bit of innovation. One position is that Darwinism will be the smoking gun that there is life. However, Smith thinks that this is too strong of a position and that the statistical mechanics of the biochemical systems can also be informative.

The last comment was about cofactors, which are very geochemically reactive and not well preserved. Smith responded that, in his mind, this is one of the most important questions in the origin of life. He said that a key distinction involves whether cofactors were a stepping stone towards a more structured polymer world or if they are an artifact of a polymer world that was already in place.

2

# Habitable Environments in the Solar System

The second session of the workshop "Searching for Life across Space and Time" was moderated by Bethany Ehlmann of the California Institute of Technology (Caltech) and Britney Schmidt of the Georgia Institute of Technology. Several solar system bodies may have had or may still have habitable environments on the surface, in liquids (both on the surface and underground), or in the atmosphere. The talks in this session focused mainly on Mars and the ocean moons, predominantly Enceladus of Saturn and Europa of Jupiter.

### HABITABLE ENVIRONMENTS OF ANCIENT MARS: DECIPHERING THE ROCK RECORD

John Grotzinger of Caltech began his talk by thinking about Mars as a global system. To illustrate this, he showed a figure made by the session moderator, Ehlmann, showing a timeline of water-related environments on Mars (see Figure 2.1).

#### **Martian Geological Record**

Starting in the Noachian era (~4.1 billion years [Gyr] ago to ~3.7 Gyr ago), there could have been a mixture of aqueous surface environments including hot springs, lakes, and rivers. However, due to planetary processes, like the loss of a geodynamo, which allowed the solar wind to erode away the atmosphere, the surface water environment eventually disappeared. However, it might have come back periodically in pulses (Figure 2.1) that produced a range of elements, minerals, and salts that provide a geological history of Mars. The details of this story, such as the temporal boundaries and the abundance of surface water, are disputed. Grotzinger's main point was that the Curiosity rover mission provides ample evidence that surface water existed into much younger periods of time than previously thought and that the early to middle Hesperian environment is more favorable for habitability than previously regarded. (The Hesperian era began ~3.7 Gyr ago.)

Grotzinger then suggested that there are other aspects of our understanding of ancient Mars that could use some rethinking as well, and these will in turn clarify the question of habitability. One line of thought is that Mars is a volcanic planet. While true, layered rocks are also suggestive of sedimentary geologies, and sedimentary basins are chemical reactors converting heat and fluid flows into aqueous minerals, as shown by Curiosity, the Mars Science Laboratory mission rover. Earth's sedimentary rock, Grotzinger said, is an archive of Earth's earliest



FIGURE 2.1 Water-related environments and alteration minerals in martian history. SOURCE: Adapted by permission from Macmillan Publishers Ltd.: B.L. Ehlmann, J.F. Mustard, S.L. Murchie, J.-P. Bibring, A. Meunier, A.A. Fraeman, and Y. Langevin, 2011, Subsurface water and clay mineral formation during the early history of Mars, *Nature* 479(7371):53-60, copyright 2011; presented in John Grotzinger, California Institute of Technology, "Habitable Environments of Ancient Mars; Deciphering the Rock Record," presentation to the Workshop on Searching for Life across Space and Time, December 5, 2016.

biosphere. The primary feature ensuring that the record of that earlier biosphere has been preserved is silica-rich sedimentary rocks. Another area needing rethinking is seeing Mars as a glacial planet. Opposed to this is the fact that no glacial landforms have been encountered by rovers on the martian surface, and sedimentary deposits lack glacial features. Mars, he said, was apparently warm and wet enough for liquid water to be stable on Mars for  $10^4$  to  $10^6$  years. Another line of thought that needs to be revisited, according to Grotzinger, is that the transition from the Noachian era to the Hesperian era (boundary at ~3.7 Gyr ago) was a global acidification event. Supporting this is the finding that Meridiani Planum (visited by the second Mars Exploration rover, Opportunity) was generally acidic. Challenging this is the fact that Gale Crater (visited by Curiosity) was, in general, pH neutral. Furthermore, the largest river systems on Mars, which spanned the Noachian and Hesperian eras, produced only clays. Additionally, Grotzinger said, when considering biomarker preservation, texture and petrogenesis are just as important as the mineralogy. He said that we need more small and cheap rovers to visit all these various places.

#### **Gale Crater**

The Curiosity rover landed in Gale Crater. In the center of Gale Crater is Mt. Sharp. Grotzinger then showed a geological cross-section of the crater from the central peak to the northern rim. He wanted the audience to appreciate the topography, which shows erosion of the sedimentary deposits that once filled it. Curiosity drove across several geological boundaries in its journey. He said that looking at this mountain is like looking at the layered history of the Grand Canyon. He then showed a view of the crater with plotted results from orbital spectroscopy. Curiosity first traveled across rocks that were mostly covered with dust and that had revealed no minerals from orbit. However, the first hole drilled showed >20 percent clay, which means that much more of Mars may be composed of hydrated

16

minerals than is observed by spectroscopy from orbit. Curiosity then explored the stratigraphically younger and topographically higher Murray formation. This unit showed patches of different materials like silica, hematite, clay, and sulfate from orbit, but when drilled, it again showed a rich bounty of hydrated clays and other hydrated minerals. These minerals exist at abundances much higher than what was predicted from orbit (see Figure 2.2). Curiosity measured a stratigraphic column—layered rocks deposited as a function of time.

Curiosity rover data show that it landed on ancient conglomerates, riverbeds, channels, and rivers with gravelly sandstones. The rover also discovered features interpreted to represent ancient deltas. On Mars, you see river deposits passing into deltas and then on into lake deposits that are strikingly similar to what you see on Earth. This lead Grotzinger to believe that there were long-lived lakes on Mars. He said, however, that the persistence of lakes does not matter critically to habitability. Even when the lake's surface dries out, there is still a habitable, aqueous environment below the surface. They imagine that the basin filled up with alternating lake deposits and maybe some dry deposits. Wind then eroded some of it away and left behind a mountain in the center.

As Curiosity moved up the mountain, it drilled different deposits in progressively younger stratigraphic positions (see Figure 2.2). The Chemical and Mineralogy X-ray Diffraction (CheMin) instrument used X-ray diffraction to examine the lake deposit sample and compared it to Gale soil samples representing primary igneous compositions. At Yellowknife Bay, compared to the soils, the drilled lake deposits showed that igneous minerals have been altered into other minerals, mostly an iron-magnesium clay mineral, but also some magnetite. This looks similar to the results of serpentinization, demonstrating that sedimentary basins are important chemical reactors, favorable



FIGURE 2.2 Composition of martian samples in Gale Crater along Curiosity's path. SOURCE: E.B. Rampe, D.W. Ming, J.P. Grotzinger, R.V. Morris, D.F. Blake, D.T. Vaniman, T.F. Bristow, et al., 2017, "Mineral Trends in Early Hesperian Lacustrine Mudstone at Gale Crater, Mars," LPI Contribution No. 1964, id.2821, presented at Lunar and Planetary Science XLVIII; presented in John Grotzinger, California Institute of Technology, "Habitable Environments of Ancient Mars; Deciphering the Rock Record," presentation to the Workshop on Searching for Life across Space and Time, December 5, 2016.

for microbial habitability. Samples drilled at Yellowknife Bay also revealed a statistically significant quantity of the reduced organic compound chlorobenzene, indicating that this geological environment was favorable for the preservation of organic compounds.

The Curiosity rover started in Yellowknife Bay in the crater basin and drove partway up Mt. Sharp. Along the way, it drilled many holes and analyzed their composition (see Figure 2.2). At the bottom of the stratigraphic sequence, there were chlorobenzene molecules, clay, and magnetite. A younger lake deposit had less magnetite, but Curiosity started to pick up some hematite and a little bit of jarosite that hinted at minor acidity. Moving farther up through the stratigraphy, the lake deposit changed composition again, losing all evidence of acidity and instead increasing magnetite along with a striking amount of both crystalline and amorphous silica. Another rock layer higher up had even more crystalline silica, along with magnetite and minor residual igneous minerals. More recent samples higher up the mountain show a lot of clay and some hematite, but no magnetite. Grotzinger said that this shows that the sedimentary basin acts as a chemical reactor. Primary igneous minerals are being converted into different minerals under different chemical circumstances, which he thinks is pretty exciting for habitability. These compositions might have been able to support several different metabolic pathways.

Grotzinger then showed an image of a striking rock, a very fine-grained chert composed of  $\sim$ 73 percent SiO<sub>2</sub> with a millimeter thick lamination. The rock has a small amount of igneous minerals left (mostly plagioclase), some magnetite, opal CT, and a lot of amorphous materials, probably opal A. All of the mafic materials are gone. Instead, there is a lot of crystalline silica, including an exotic polymorph called tridymite. This, Grotzinger thinks, was likely transported from a felsic volcanic rock. This rock is very important because it is compositionally very similar to early rocks on Earth, which can contain microfossils. Silica is a great material that can survive through a number of geological processes, including, in some cases, thermal metamorphism. There is very strong evidence that this silica was created by primary enrichment, increasing its relevance to preservation of potential biological materials.

Grotzinger then discussed work by Joel Hurowitz on evidence for an ancient redox-stratified lake in Gale Crater. Certain areas of the lake have an abundance of oxidants, which they interpret as ultraviolet (UV) photolysis of water that created oxygen. Meanwhile, reduced iron percolated through the martian crust (i.e., groundwater seeped into the lake). When the level of oxidants exceeded the demand from reduced iron, the oxidants in the water then reacted with reduced iron, which caused the precipitation of hematite. With a little evaporation, some sulfate salts could have been produced as well. The silica-rich rock has a very different story though. In areas of the lake where the oxidant concentration did not exceed the reduced iron, magnetite was created instead. This means that there were multiple oxidation states in the ancient lake; even the lake itself was chemically stratified. This is very important for microbial habitability, which depends on redox gradients.

Grotzinger then said that new research has shown the possibility that the origin of life on Mars could have occurred on the surface. UV radiation could drive some of the chemistry. All that is needed is hydrogen cyanide and hydrogen sulfide, both of which are present on Mars. Gale Crater, he said, gives an opportunity to look at both environments: a long-lived environment possibly thermally warm enough for olivine to dissolve into and maybe even to allow a pathway towards hydrogen production or, alternatively, surface waters that could proceed with a different molecular chemistry.

Grotzinger finished by talking about groundwater. As Curiosity works its way up Mt. Sharp, it is finding fractures that cut across the sedimentary rock that are full of sulfate minerals. The Chemistry and Camera (Chem-Cam) instrument, however, is showing that the fractures are becoming increasingly enriched with boron, meaning that boron may be present as a trace component of other minerals, or perhaps present as amorphous compounds.

#### **Audience Participation**

A member of the audience asked if there was any evidence of carbonate minerals being formed and where the magnesium that was leached from these salts was going. Another audience member said that the question of where the carbonates and magnesium are going may be two separate questions. She said that the magnesium is going into carbon in two places on Mars: the Nili Fossae northeast surface region and the Comanche outcrop. Both have magnesium from olivine going into carbonate, but the Nili Fossae region also has some magnesium going into clay minerals. In the Gale Crater, it seems like the magnesium is primarily going into phyllosilicate minerals, but also maybe some sulfates.

Because no rover has encountered glacial features on Mars, one audience member asked if this was just a site selection issue, considering that some geomorphology implies glacial features. Grotzinger admitted that that could be the case, but he also said that the sedimentary record doesn't provide any evidence for glacial deposits. He went on to say that he's sure glacial features are there, but glaciers probably aren't the dominant paradigm on Mars.

According to a workshop participant, recent origin-of-life work published in *Nature* showed that every detail Grotzinger talked about, even olivines remaining in the residual sedimentary rocks eroding from an igneous facies, is exactly what is needed to go from formaldehyde, generated by the photochemical decomposition of carbon dioxide in the atmosphere, all the way to RNA. The audience member said that he just published a paper showing that opal CT, which was in one of the martian facies, absorbs oligomeric RNA and all of its intermediate steps. He then asked why Grotzinger didn't mention two species, phosphate minerals and borate minerals. Grotzinger said that they do not detect any borate minerals. However, they do see phosphate, with fluorapatite being the dominant phase. They think it's an igneous mineral, but there is phosphorus there. The silica enrichment they see is associated with the retention of phosphorus, which supports a pH-neutral body of water. This is because, if all the igneous minerals were being dissolved at low acidity, it should have been one of the first minerals to dissolve, but they are still seeing it anyway.

A workshop participant then briefly explained that, if intermittent, wet-dry cycles were good for biochemistry, and that the lack of a significant martian moon is an advantage. Earth has glaciation with just a 2° wobble in obliquity, while Mars can move from 10° to 50° and back over just tens of thousands of years.

Underneath the red, highly oxidized martian surface, one audience member said, there is a gray, likely reducing, material underneath. Hearkening back to the earlier talks about how life likes to use redox disequilibria, he asked how this boundary near the martian surface could contribute to habitability. Grotzinger said that the highest altitude drill samples on Mt. Sharp no longer show a gray subsurface. They are red throughout, which means that Gale preserves multiple oxidation states.

An audience member then went back to a previous point on phosphate. He said that there isn't good information on soluble phosphate using X-ray diffraction from CheMin. Elemental analyses, however, do show that phosphorus enrichments are usually accompanied by calcium, and they likely are soluble. Changing topics to salts, he said that as long as there are lakes, the magnesium sulfate salts are not too concentrated for potential life. Only when the salt becomes an evaporite does the water activity become inconsistent with life. Additionally, he has done work showing that many organisms can tolerate high magnesium sulfate levels.

To one workshop participant, martian meteorites are interesting because you can look at the mass-independent fractions of the isotopes of sulfur and of oxygen (i.e., the fractions are not in proportion to the mass of the respective isotopes) in the sulfates, carbonates, and water in the host rock. It therefore looks like a lot of the sulfates are photochemically processed—a known pathway that circumvents mass-dependence. The water might be photochemically processed too because the oxygen in the water is also found to be mass-independent, but it isn't in equilibrium with the sulfate and carbonate that it is found with. Grotzinger replied that a complex mass spectrometer could make that measurement. He then said that it would be a good idea to land in a place with lots of sulfate and to perform that measurement.

A member of the audience then asked Grotzinger which locations on Mars he would most like to land the aforementioned small "boutique" rover on. He said that he would first like to go to a carbonate site. Another place he thought would be a good idea is somewhere in Valles Marineris. Building off this question, another audience member then asked which new instruments Grotzinger would want on it. Grotzinger said that he would really like to see a rover with an imaging spectrometer land on a place with extreme mineral diversity. He also said that a laser Raman spectrometer would be great to have.

The last question asked of Grotzinger was when we were going to drill deep on Mars. Another audience member said that ExoMars, launching in 2020, would drill down 2 m into the martian surface.

20

#### ON THE HABITABILITY OF OCEAN WORLDS

Kevin Hand of the NASA Jet Propulsion Laboratory (JPL) began his talk showing a graphic of all robotic missions, successful and failed, to all bodies of the solar system. One of the most remarkable discoveries from these missions, Hand said, is that at least six worlds beyond Earth likely harbor subsurface, liquid water oceans: Europa, Ganymede, and Callisto of Jupiter; Enceladus and Titan of Saturn; and possibly Triton of Neptune. Additionally, Titan has an atmosphere and surface lakes of hydrocarbon liquids. Triton's ocean may have some ammonia mixed in as well. Hand said that Alan Stern of the Southwest Research Institute, and the principal investigator of the New Horizons mission to Pluto, could make a very strong case for adding Pluto to the list of ocean worlds. Hand said that he would probably agree.

Hand then showed these moons on a grid with potential geophysical properties and processes to illustrate which moons are the most interesting in the context of searching for life beyond Earth (see Table 2.1). In particular, he emphasized the column showing which moons have oceans in contact with rock; a condition that, to the best of our knowledge, only exists on Europa and Enceladus. Europa's ocean has likely been persistently habitable for most of the history of the solar system. Enceladus might also have survived with an ocean for the lifetime of the solar system, although a recent paper has argued that Enceladus is only 100 Myr old and was formed by a Kuiper belt object colliding with a body in the Saturnian system.

What really motivates Hand is the prospect that one of these bodies independently gave rise to life that is still extant. As a point of comparison, Hand said that the search for life on Mars is of great importance, but that the current strategy of searching for ancient life in the rock record of Mars would, if successful, leave many key questions unanswered. What is the fundamental biochemistry? How did that life originate, and was it from an independent, second origin? Was it seeded from life on Earth, or did Mars seed Earth? Answering these kinds of questions requires samples of life that go well beyond what is preserved in rocks billions of years old. The discovery of extant life in an ocean world would allow for a detailed study of life and its biochemistry that would not be possible from martian microfossils. Potential martian life would also be more likely to have been delivered from Earth (i.e., panspermia) than for the outer solar system's icy bodies. For example, out of 600 million rocks produced by an asteroid collision with Earth, only ~30 to 100 rocks would land on Europa, and only ~3 to 20 on Titan, both with impact velocities >10 km/s, which would likely destroy any life transported within the rocks.<sup>1</sup> If DNA were found on these bodies, it would strongly suggest that there is an evolutionary chemical convergence towards using DNA as the information storage molecules for life. This would also help us understand how life originated on Earth. Since the icy ocean worlds do not have continents or tide pools, a discovery of life there would argue against a primordial soup origin on Earth and in favor of a hydrothermal or even an icy origin. Conversely, were life not to be found within ocean worlds, one might be inclined to favor models for the origin of life that include continents and tidal pools. Either way, much about life beyond Earth and how life on Earth may have originated could be learned.

#### **Liquid Water**

Liquid water is the most important aspect of these bodies in terms of habitability. Hand said that the combined water volume in the icy moons, based on conservative estimates, is about 30 times higher than that of Earth. In the Jovian system, the liquid water is maintained through tidal heating and some radiogenic decay. Tidal energy dissipation would usually fade away as the orbits circularize, but the Laplace resonance of the interior three Jovian moons (Io, Europa, and Ganymede) forces an eccentricity that can maintain long-term tidal heating. This is a powerful heating source. While Earth's average surface flux (for only the seafloor) is 60 to 80 mW/m<sup>2</sup>, Europa's possible range is 10 to 800 mW/m<sup>2</sup>, and Io is at 2500 mW/m<sup>2</sup>. Earth's Moon, meanwhile, is at a paltry 9 to 13 mW/m<sup>2</sup> from radiogenic decay.

Hand then explained how plumes can allow for easy confirmation of a subsurface, liquid water ocean. A recent Hubble image revealed a possible liquid water plume around Europa. Enceladus, on the other hand, has an

<sup>&</sup>lt;sup>1</sup> B. Gladman, L. Dones, H.F. Levison, and J.A. Burns, 2005, Impact seeding and reseeding in the inner solar system, Astrobiology 5:483.

|                          | 1 2                  |   |  |                               |                      |                                   |
|--------------------------|----------------------|---|--|-------------------------------|----------------------|-----------------------------------|
| Moon                     | Name,<br>Planet      | Geophysically<br>and<br>Geochemically<br>Plausible? | Significant<br>Tidal Energy<br>to Help<br>Maintain<br>Ocean? | Induced<br>Magnetic<br>Field? | Activity<br>Observed | Ocean in<br>Contact with<br>Rock? |
|                          | Europa,<br>Jupiter   | Yes   | Yes  | Yes                           | No(?)                | Yes                               |
|                          | Ganymede,<br>Jupiter | Yes   | ~Yes   | Yes                           | No                   | No                                |
|                          | Callisto,<br>Jupiter | Yes   | No   | Yes                           | No                   | No                                |
|                          | Enceladus,<br>Saturn | ???   | ???  | ???                           | Yes!                 | Yes                               |
|                          | Titan, Saturn        | Yes   | No   | ???                           | ???                  | No                                |
| SOURCE: Europa (NASA/IPI | Triton,<br>Neptune   | Yes?  | No   | ???                           | Yes                  | No                                |

| TABLE 2.1 Potential | Geophysical | Properties and Processes Releva | int to Origin of Life on Various Moons |
|---------------------|-------------|---------------------------------|--|
|                     |             |                                 |  |

SOURCE: Europa (NASA/JPL/DLR), Ganymede (NASA/JPL), Callisto (NASA/JPL,DLR), Enceladus (NASA/JPL-Caltech/Space Science Institute), Titan (NASA/JPL-Caltech/Space Science Institute), and Triton (NASA/JPL/USGS).

ocean that has been confirmed by the Cassini spacecraft's discovery and fly-throughs of its plume. Enceladus's tidal energy is likely due to the 2:1 tidal resonance it has with the more distant Saturnian moon Dione, although as mentioned before, the long-term nature of this tidal heating is debated.

#### The Availability of Elements

Another keystone for life, Hand suggested, is the availability of elements necessary for building life (CHNOPS [carbon, hydrogen, nitrogen, oxygen, phosphorus, and sulfur] and some metals). These ocean worlds formed in the outer solar systems where the condensing volatiles contained a large quantity of the CHNOPS elements. The interiors and surfaces of the icy worlds therefore retained a lot of these key molecules and elements. Hand noted, ironically, that it is difficult to explain exactly how Earth got so much water and carbon.

In terms of heavier elements, a simple check of bulk density can be informative (see Figure 2.3). Io and Europa (and the Moon) are predominantly rocky and likely silica-rich, with densities  $\geq$ 3,000 kg/m<sup>3</sup>, while all other icy ocean worlds have densities in the approximate range of 1,000 to 2,000 kg/m<sup>3</sup>. The larger bodies (Ganymede, Titan, Callisto, and maybe Triton and Pluto) are massive enough that there is a phase transition to a denser ice (ice III, ice IV, and/or ice VI) that sinks and lines the ocean floor, hindering or altogether preventing water-rock interactions on their seafloors.

Hand said that both Europa and Enceladus have been shown to contain heavier elements. He highlighted the detection of silica in the ice grains of Saturn's E-ring (created by Enceladus) by Cassini's Cosmic Dust Analyzer (CDA). This implies a low-temperature, alkaline, water-rock interaction that provides ~200 ppm of silica to the ocean. On Europa, the discoloration on its surface is believed by many to be salt from within its ocean. Hand, however, had not been fully convinced by the limited spectra supporting the salt hypothesis, instead having largely preferred the sulfuric acid hypothesis, which said that the discoloration is primarily sulfuric acid on the surface derived from sulfur originating from Io's volcanism. At JPL, Hand has a laboratory "Europa-in-a-can" to test for the source of the discoloration. They introduced salt to the sample of artificial Europan material as an evaporite and irradiated it with an electron gun at 10 keV. This turned the white salt into a yellowish brown, which is evidence



FIGURE 2.3 Density of solar system bodies compared to radius. SOURCE: Reprinted from H. Hussmann, F. Sohl, and T. Spohn, 2006, Subsurface oceans and deep interiors of medium-sized outer planet satellites and large trans-neptunian objects, *Icarus* 185(1):258-273, copyright 2006, with permission from Elsevier; presented in Kevin Hand, Jet Propulsion Laboratory, "On the Habitability of Ocean Worlds," presentation to the Workshop on Searching for Life across Space and Time, December 5, 2016.

22

of the so-called F and M color centers that arise when defects are formed in crystals due to the presence of trapped electrons in negative (i.e., anion) vacancies. Hand now predicts that in certain regions on Europa, the discoloration

electrons in negative (i.e., anion) vacancies. Hand now predicts that, in certain regions on Europa, the discoloration is from salts coming up from the ocean and being irradiated and discolored. Keck telescope observations show a spectrally unique region in the Powys Regio. Hubble will soon be looking at this region to check for irradiated salts.

#### **Energetics Needed for Life**

Hand then moved on to the energy needed for life to grow, reproduce, and metabolize. An active seafloor, he said, is not sufficient for chemosynthesis if there is not the right combination of electron donors and acceptors. He argued that the radiation environment of Europa, coupled with geologic overturn of the ice shell, could solve that problem. The radiation has made Europa's surface covered with oxidants. Charged particles split water and create OH radicals, which then combine to make hydrogen peroxide  $(H_2O_2)$ . Some peroxide stays around, some decays (with  $H_2$  escape), and some combines with sulfur. This leads to a surface rich in peroxide, oxygen, sulfate, sulfur dioxide, carbon dioxide, and more. Geological activity could then introduce these molecules into Europa's oceans at a rate that could sustain an active subsurface biosphere within Europa's ocean.

#### **Potential Biosignatures on Europa**

Hand then used a thick ice shell model ( $\sim$ 15 km) for Europa, on top of a thick ocean of water ( $\sim$ 100 km), to examine exchange processes and how potential biosignatures might be preserved on the surface of Europa (see Figure 2.4). The top layer is composed of brittle ice. Underneath this surface is a layer of ductile, convective ice.



FIGURE 2.4 Possible habitable regions, sites of biosignature preservation, and sites of potential biosignature emplacement on Europa. SOURCE: Kevin Hand, Jet Propulsion Laboratory, "On the Habitability of Ocean Worlds," presentation to the Workshop on Searching for Life across Space and Time, December 5, 2016. He said that the seafloor region could potentially be habitable, but that the ice-water interface could also be a chemically rich and potentially habitable interface. Oxidants from the surface could mix with reductants delivered from ocean currents. Fractures and diapirs could provide pathways to deliver material up or down. This could lead to several regions within or at the boundaries of the icy crust that could be habitable.

Hand finished by talking about the possible conditions that lead to the origin of life and how biology might someday be found to have an organizing principle similar to the periodic table of the elements or the Gibbs phase rule. Hand said that revealing these organizing principles, and revolutionizing our understanding of biology, would require finding a second, independent origin of life. The ocean worlds of the outer solar system provide prime locales for testing the biology hypothesis and whether or not biology works beyond Earth.

#### **Audience Participation**

A member of the audience commented that Hand provided an explanation of Europa's surface discoloration that does not require minerals, only salt. Therefore, it does not necessarily have to be from the solid core. He then asked what the latest thinking was on where the energy from tidal forcing is being deposited: Europa's core or its water-ice interface? Hand said that there is still debate about where the energy is deposited but that the key is the partitioning in the mantle or the ice shell. The Europa Clipper (previously known as the Europa Multiple-Flyby Mission) could help reveal this partitioning.

After getting confirmation that the salt irradiation experiment at JPL used only electron radiation, an audience member then asked if the energy flux of the electron radiation was similar to the modern day energy flux on Europa or if it was adjusted to account for historically variable flux levels. Hand said that the total fluence (flux integrated over time) into the salts maps quite well with reality. Furthermore, the reason they focused only on electron radiation is that 80 percent of Europa's radiation comes from energetic electrons in Jupiter's magnetosphere.

The same audience member then asked a different question related to possible hydrothermal activity on Europa. She asked how the chemistry and energy from the hydrothermal vents could be carried through the entire thick ocean to the water-ice interface at the crust. For example, she said, you can't detect the hydrothermal vent systems on Earth at the ocean surface. Hand answered that modeling has looked at what happens to buoyant plumes under different conditions. Results show that plumes could stay contained by traveling vertically through tens of kilometers of ocean—and perhaps all the way to the water-ice interface. He then said that work on Earth is attempting to measure how high Earth's hydrothermal plumes rise in a coherent fashion. The canonical answer, he said, is about 400 to 600 m. They looked for, but did not find, any evidence in the Arctic ice. Another audience member then said that, even if a plume cannot stay self-contained up to the ice-water interface, the by-products could still be delivered through ocean mixing. She then noted that plumes would be the most stable at the poles. Then another audience member went further and showed how hydrothermal systems can influence the whole ocean. He said that a 2015 paper indicated that the overwhelming amount of iron in Earth's ocean comes from hydrothermal systems. Models of trace elements, such as molybdenum, are also starting to show the same thing. He then pointed out that, while there are only about 40 to 50 hydrothermal vent systems known, global budget models show that there could be as many as 900. Extrapolating the ocean abundance models to include unknown hydrothermal systems suggests that 60 to 70 percent of inorganic nutrients could originate from hydrothermal vents. He then said that, even on Earth, there is not a deep understanding of how circulation happens in the deep oceans. How tidal fluxing could affect these circulation patterns on Europa is also unknown. Hand followed this saying that the SiO<sub>2</sub> data from Enceladus, if it is indeed hydrothermal, must have traveled up through at least 50 km of ocean water and 30 km of ice before it could get into space. Even if these bodies do not have life, there is a tremendous amount that could be learned in terms of comparative oceanography.

A workshop participant then pointed out that contamination from Earth life on probes could be problematic. Hand completely agreed and said that planetary protection needs to be a primary concern.

Another participant at the workshop then asked whether a fly-by mission of Enceladus might actually be able to capture cells. She then asked if spores in the plume could travel to other nearby bodies. Hand said that any cells ejected from a plume would likely die after exposure to the space environment. Any cell that gets on the surface would first freeze, which would actually be good in terms of organics and biosignature preservation. However, it

24

would then be irradiated, which could destroy life and alter biosignatures. On the other hand, in the Alum Shale Formation in southern Scandinavia, where there are a lot of uranium-rich materials, strong organic biosignatures are present, despite all of the radiation processing. Changing topics to Europa, Hand finished by saying that, even in its harsh radiation environment, he still thinks that biosignatures in the oceans and the ice could be preserved and detectable.

#### PLANNING FOR THE EXPLORATION OF MARS AND OCEAN WORLDS

Ellen Stofan, the former chief scientist at NASA, began her talk by describing the need for NASA to incorporate research from all of its main scientific fields (astrophysics, heliophysics, planetary science, Earth science, and microgravity) and also from those fields outside of NASA's purview in order to clearly understand what is needed in the search for extraterrestrial life. A recent breakthrough, she said, is the exoplanet revolution due to the Kepler space telescope's discoveries. One of its major discoveries was the huge number of exoplanet candidates in the super Earth to sub-Neptune range. The Transiting Exoplanet Survey Satellite (TESS; expected launch 2018) will build on these discoveries.

The exoplanet revolution has led to a re-examination of what the habitable zone is, both within our solar system and beyond. Stofan emphasized the fact that the habitable zone is not just dependent on spatial location, but also dependent on time. Venus in its early history may have been habitable. Earth's twin almost certainly lost an ocean's worth of water in the past, which may have been stable on its surface before being lost to a runaway greenhouse effect. Two Phase A concepts for going back to Venus were in the works to help answer important questions about Venus, such as the compositions of the atmosphere and the rocky surface, the isotopic composition of the atmosphere, and the minerology of its surface rocks, but neither was ultimately selected. New Frontiers will be the next chance for Venus missions.

For Mars, the history of water on the planet is key, Stofan said. Water was stable on Mars' surface for a long period of time, raising the prospects for past habitability on Mars. She thinks that humans are going to need to land on Mars to fully explore the planet and its potential for past life. This includes drilling deep (below 2 m) and getting samples from multiple locations. Stofan is optimistic about sending humans to Mars. Research on the International Space Station (ISS) has been done to figure out how to keep astronauts healthy for long periods of time spent in space. A plan has been made for using the ISS to investigate how to mitigate bone density loss, muscle wasting, and decreased immune system functioning. Life support systems are also critical. Recycling water, keeping  $CO_2$  levels down, and just keeping the toilets working are all necessary for a successful Mars mission. By the mid-2020s, they plan to put a prototype of the Mars transfer vehicle into orbit around the Moon to test the environmental control and life support systems as a concept demonstration for a crewed Mars mission.

NASA plans to continue sending robotic rover missions to Mars, such as the Mars 2020 rover. In preparation for human missions to Mars, NASA also plans to do robotic landing missions in the late 2020s to test equipment and procedures for future crewed attempts, Stofan said. A crewed martian orbiter mission is planned for 2033. On this mission, they would like to tele-operate a rover on Mars, perhaps going to a region that NASA does not want to send humans. The mission to land the first humans on Mars is slated for the late 2030s. NASA is also partnered with SpaceX for Red Dragon, a planned low-cost, robotic martian lander planning to launch in 2018 or 2020. Planetary protection, however, is a major concern. A decade-old study said it was necessary to do a sample return mission before sending humans, and Stofan thinks that this topic needs to be revisited. Stofan said that a National Academies of Sciences, Engineering, and Medicine study might be useful to determine the best approach.

Stofan then pivoted to the solar system's present (and potential, not yet fully described) Ocean Worlds. The possible plumes on Europa are of particular interest and have sparked greater interest in a Europa lander to be added onto a future orbital mission. A current Jovian mission, the Europa Clipper, is already planned for a launch in the early to mid-2020s. Stofan is excited about the possibility of using the Space Launch System, which cuts travel times to the outer solar system approximately in half, returning data in a much more timely manner (i.e., before the science teams retire).

Stofan mentioned the only currently approved missions related to the exploration of extraterrestrial life in the solar system are the Mars 2020 rover and the Europa Clipper. A Europa lander mission is being extensively
studied at the moment too. On December 9, 2016, NASA announced a call for New Frontiers missions, which now includes a category for oceans worlds (although only for Titan and/or Enceladus at the moment). Stofan finished by saying that she hopes that a robotic demonstration mission that includes sample return in the late 2020s will pave the way for a crewed mission in the 2030s to land scientists on the martian surface to explore the possibility of alien life on Mars.

#### **Audience Participation**

A member of the audience commented that he didn't hear anything about searching for extant life on Mars in Stofan's talk. He said that this might be the first thing we want to do, especially before sending humans, which could possibly contaminate the martian surface. Stofan agreed that it was a good point and again said that a study by the National Academies is needed to determine the necessity of doing a sample return mission before sending humans.

Another member of the audience asked whether the evolution of our understanding of rocky exoplanets changes what we are planning to look for in our own solar system's rocky planets. Stofan said that it changes the questions that are being asked. Looking at other planetary systems, she thinks that we need to better understand our own solar system's habitable (or once habitable) rocky planets: Venus, Earth, and Mars. She said that there are fairly straightforward missions and measurements that can be done, especially for Venus, that have just been ignored.

Several other entities, both state and private, are planning to go to Mars, one audience member said, specifically mentioning India, Europe, Russia, and China. He then asked whether NASA feels that there is a soft space race for going to Mars and whether the United States and its political leaders are aware of that. Stofan mentioned a recent article asking whether NASA was really going to Mars or if it would pivot away. The article also quoted someone saying that there isn't a reason to send people to Mars. Stofan strongly disagreed with that. She also doesn't see the push for Mars as a new, soft space race, but rather, a confluence of interests, including major public interest. She said that sending humans to Mars garners the most public reaction and interest out of all of NASA's projects. She also said that a crewed mission to Mars can be done in collaboration with other countries and with private firms. Mars, she said, will be done and made affordable by cooperation and collaboration rather than competition.

3

# Exoplanets

The afternoon session of the workshop moved beyond the search for life in the solar system and focused instead on the search for life on distant exoplanets. This session was co-chaired by David Des Marais, NASA Ames Research Center, and Dimitar Sasselov, Harvard-Smithsonian Center for Astrophysics.

# EXTRASOLAR BIOSIGNATURES: DEVELOPING A COMPREHENSIVE FRAMEWORK FOR BIOSIGNATURE RECOGNITION

Victoria Meadows of the University of Washington prefaced her talk on extrasolar biosignatures by explaining that she would essentially summarize the 2016 workshop hosted by the Nexus for Exoplanet System Science (NExSS) and the NASA Astrobiology Institute, called the "Exoplanet Biosignatures Workshop Without Walls." This workshop combined the expertise of NExSS, the NASA Astrobiology Institute, and the science and technology definition teams for exoplanet observation mission concepts to focus on the following three main questions:

• What are the known remotely observable biosignatures, the processes that produce them, and their known non-biological sources?

• How can we develop a more comprehensive framework for identifying additional biosignatures and their possible abiotic mimics?

• What standards can we agree to use for assessing biosignature observations, both known biosignatures and those we have yet to identify?

That workshop produced five coordinated papers on topics covered by the workshop: a biosignatures review, work on using  $O_2$  as a biosignature, developing a more general framework for observing and interpreting biosignatures, novel types of biosignatures, and a synthesis paper to guide future research on topics such as modeling and mission development.

# **Exoplanet Biosignature Review**

The major question in exoplanet biosignatures, said Meadows, is how to detect life at great distances. In this case, life must have a global impact to be observable. Identifying biosignatures requires three things: *reliability* 

that the signature is indeed biological, *survivability* of the potential biosignature, and the *detectability* of the possible signature. Meadows said that an alternative way to search for life would be to look for a disequilibrium or some sort of unexpected planetary process that cannot be explained by abiotic processes.

Meadows stated that typical biosignatures are atmospheric gases, such as oxygen in the presence of methane.<sup>1,2</sup> However, she wanted to push the boundaries of what we know by exploring other types of gaseous biosignatures in different contexts and environments. There are also surface biosignatures, such as the "red edge," which is due to the phenomenon that Earth's plants are highly reflective in the near-infrared (see Figure 3.1).<sup>3</sup> Other types of "edges" may be possible with different pigments, which may or may not be related to photosynthesis (e.g., UV protection).<sup>4,5,6</sup> Temporal biosignatures are also possible, such as daily or seasonal changes.<sup>7</sup> An example is the seasonal change in abundance of CO<sub>2</sub> in Earth's atmosphere.<sup>8</sup> A large disequilibrium could also indicate signs of life. The classic example is Earth's high abundance of both O<sub>2</sub> and CH<sub>4</sub>. Since methane's lifetime in the atmosphere is just 10 years,<sup>9</sup> methane's high abundance in the presence of O<sub>2</sub> indicates an active source of the gas, and in the case of Earth, that is due to life (see Table 3.1). Meadows then cited some recent work that showed that the largest Gibbs energy disequilibrium on Earth is the fact that Earth has both N<sub>2</sub> and O<sub>2</sub> with an ocean. Without life, this would end up as nitrate dissolved in the ocean.<sup>10</sup>

She then defined three terms useful in thinking about biosignatures. First, an "antibiosignature" is an aspect of the planetary environment that suggests that life is not present, such as abundant CO on Mars, which would be an attractive energy source for life if it were there.<sup>11</sup> A "false positive" is an abiotic source for a potential biosignature, such as  $O_2$  being produced by photolysis of  $H_2O$  or  $CO_2$ .<sup>12-17</sup> A "false negative" is when processes on the planet work to reduce the detectability of a biosignature, such as oxidation on a planet's surface.<sup>18,19</sup>

<sup>10</sup> J. Krissansen-Totton, D.S. Bergsman, and D.C. Catling, 2015, On detecting biospheres from chemical thermodynamic disequilibrium in planetary atmospheres, *Astrobiology* 16:39.

<sup>11</sup> K. Zahnle, R.S. Freedman, and D.C Catling, 2011, Is there methane on Mars?, *Icarus* 212:493.

<sup>14</sup> R. Wordsworth and R. Pierrehumbert, 2014, Abiotic oxygen-dominated atmospheres on terrestrial habitable zone planets, *The Astro-physical Journal Letters* 785:20.

<sup>15</sup> P. Gao, R. Hu, T.D. Robinson, C. Li, and Y.L. Yung, 2015, Stabilization of CO<sub>2</sub> atmospheres on desiccated M dwarf exoplanets, *The Astrophysical Journal* 806:249.

<sup>&</sup>lt;sup>1</sup> D.R. Hitchcock and J.E. Lovelock, 1967, Life detection by atmospheric analysis, *Icarus* 7:149.

<sup>&</sup>lt;sup>2</sup> V. Meadows, 2017, Reflections on O<sub>2</sub> as a biosignature in exoplanetary atmospheres, *Astrobiology*, accepted.

<sup>&</sup>lt;sup>3</sup> D.M. Gates, H.J. Keegan, J.C. Schleter, and V.R. Weidner, 1965, Spectral properties of plants, Applied Optics 4:11.

<sup>&</sup>lt;sup>4</sup> E.W. Schwieterman, C.S. Cockell, and V.S. Meadows, 2015, Nonphotosynthetic pigments as potential biosignatures, *Astrobiology* 15:341.

<sup>&</sup>lt;sup>5</sup> S. Hegde, I.G. Paulino-Lima, R. Kent, L. Kaltenegger, and L. Rothschild, 2015, Surface biosignatures of exo-Earths: Remote detection of extraterrestrial life, *Proceedings of the National Academy of Sciences of the U.S.A.* 112:3886.

<sup>&</sup>lt;sup>6</sup> N.Y. Kiang, A. Segura, G. Tinetti, Govindjee, R.E. Blankenship, M. Cohen, J. Siefert, D. Crisp, and V.S. Meadows, 2007, Spectral signatures of photosynthesis. II. Coevolution with other stars and the atmosphere on extrasolar worlds, *Astrobiology* 7:252.

<sup>&</sup>lt;sup>7</sup> V.S Meadows, 2005, Modelling the diversity of extrasolar terrestrial planets, *Proceedings of the International Astronomical Union* 1:25.

<sup>&</sup>lt;sup>8</sup> C.D. Keeling, R.B. Bacastow, A.E. Bain-Bridge, C.A. Ekdahl, Jr., P.R. Guenther, LS. Waterman, and J.F.S. Chin, 1976, Atmospheric carbon dioxide variations at Mauna Loa Observatory, Hawaii, *Tellus* 28:538.

<sup>&</sup>lt;sup>9</sup> J.T. Houghton, L.G. Meira Filho, J. Bruce, H. Lee, B.A. Callander, E. Haites, N. Harris, and K. Maskell, eds., 1994, *Climate Change* 1994: *Radiative Forcing of Climate Change and An Evaluation of the IPCC IS92 Emission Scenarios*, Cambridge University Press, Cambridge, U.K.

 $<sup>^{12}</sup>$  R. Luger and R. Barnes, 2015, Extreme water loss and abiotic O<sub>2</sub> buildup on planets throughout the habitable zones of M dwarfs, *Astrobiology* 15:119.

<sup>&</sup>lt;sup>13</sup> F. Tian, 2015, History of water loss and atmospheric  $O_2$  buildup on rocky exoplanets near M dwarfs, *Earth and Planetary Science Letters* 432:126.

<sup>&</sup>lt;sup>16</sup> F. Tian, K. France, J.L. Linsky, P.J.D. Mauas, and M.C. Vieytes, 2014, High stellar FUV/NUV ratio and oxygen contents in the atmospheres of potentially habitable planets, *Earth and Planetary Science Letters* 385:22.

<sup>&</sup>lt;sup>17</sup> C.E. Harman, E.W. Schwieterman, J.C. Schottelkotte, and J.F. Kasting, 2015, Abiotic O<sub>2</sub> levels on planets around F, G, K, and M stars: Possible false positives for life?, *The Astrophysical Journal* 812:137.

<sup>&</sup>lt;sup>18</sup> A.D. Anbar, Y. Duan, T.W. Lyons, G.L. Arnold, B. Kendall, R.A. Creaser, A.J. Kaufman, G.W. Gordon, C. Scott, J. Garvin, and R. Buick, 2007, A whiff of oxygen before the great oxidation event?, *Science* 317:1903.

<sup>&</sup>lt;sup>19</sup> C.T. Reinhard, S.L. Olson, E.W. Schwieterman, and T.W. Lyons, 2017, False negatives for remote life detection on ocean-bearing planets: Lessons from the early Earth, arXiv:1702.01137.



The Red Edge. Synthetic spectrum of a line of sight through the Earth's atmosphere over a conifer forest, with chlorophyll absorption and the rededge reflectivity marked. Chlorophyll, a potentially important biosignature, has strong absorption in the UV and blue (<0.5  $\mu$ m) and in the red (0.6-0.7  $\mu$ m marked in green), and slightly less absorption in the green (0.55  $\mu$ m). Due to changes in the refractive index between air and the internal leaf structure, plants are also highly reflective just beyond the visible range (>0.7  $\mu$ m), resulting in a prominent discontinuity (marked in red) known as "the red edge."

FIGURE 3.1 The "red edge" caused by the reflectance of plants at wavelengths longer than detectable by human vision could be considered a potential biosignature. It is seen as a 2 percent effect on Earth between ocean-dominated and forested hemispheres. SOURCE: N. Haghighipour, 2008, "Planetary Environmental Signatures for Habitability and Life," Chapter 10 in *Exoplanets: Detection, Formation, Properties, Habitability* (J.W. Mason, ed.), Chichester, U.K.: Praxis Publishing with permission from Springer; presented in Victoria Meadows, University of Washington/NASA Astrobiology Institute, "Extrasolar Biosignatures: Developing a Comprehensive Framework for Biosignature Recognition: Overview of the NExSS/NAI Biosignatures Workshop 2016," presentation to the Workshop on Searching for Life across Space and Time, December 5, 2016.

| Molecule         | Standard Abundance<br>(Ground-Truth Earth) | Galileo<br>Value <sup>a</sup> | Thermodynamic<br>Equilibrum Value<br>Estimate 1 <sup>b</sup> | Thermodynamic<br>Equilibrum Value<br>Estimate 2 <sup>c</sup> |
|------------------|--|-------------------------------|--|--|
| N <sub>2</sub>   | 0.78                                       |                               | 0.78   |  |
| 0 <sub>2</sub>   | 0.21                                       | 0.19±0.05                     | $0.21^{d}$   |  |
| H <sub>2</sub> O | 0.001 to 0.03                              | 0.001 to 0.01                 | 0.001 to 0.03  |  |
| Ār               | $9 \times 10^{-3}$                         |                               | $9 \times 10^{-3}$   |  |
| CO,              | $3.5 \times 10^{-4}$                       | 5±2.5×10-4                    | $3.5 \times 10^{-4}$   |  |
| CH <sub>4</sub>  | $1.6 \times 10^{-6}$                       | 3±1.5×10-6                    | <10 <sup>-35</sup>   | 10-145   |
| N <sub>2</sub> O | $3 \times 10^{-7}$                         | ~10-6                         | $2 \times 10^{-20}$  | $2 \times 10^{-19}$  |
| $O_3^2$          | $10^{-8}$ to $10^{-7}$                     | >10-8                         | $6 \times 10^{-32}$  | $3 \times 10^{-30}$  |

TABLE 3.1 Constituents of the Earth's Atmosphere (Volume Mixing Ratios)

<sup>a</sup> Galileo values for O<sub>2</sub>, CH<sub>4</sub>, and N<sub>2</sub>O from Near-Infrared Mapping Spectrometer (NIMS) data; O<sub>3</sub> estimate from Ultraviolet Spectrometer (UVS) data.

<sup>b</sup> At P = 1 bar, T = 280 K (see E.R. Lippincot, R.V. Eck, M.O. Dayhoff, and C. Sagan, 1967, Thermodynamic equilibria in planetary atmospheres, *The Astrophysical Journal* 147:753).

<sup>*c*</sup> At P = 1 bar, T = 290 K (see W.L. Chameides and D.D. Davis, 1992, Chemistry in the troposphere, *Chemical and Engineering News* 60:38). <sup>*d*</sup> The observed value; it is in thermodynamic equilibrium only if the under-oxidized state of the Earth's crust is neglected.

SOURCE: Reprinted by permission from Macmillan Publishers Ltd.: C. Sagan, W.R. Thompson, R. Carlson, D. Gurnett, and C. Hord, 1993, A search for life on Earth from the Galileo spacecraft, *Nature* 365:715-721, copyright 1993.

30

Meadows then put the idea of a biosignature in some historical context. Previously,  $O_2$  alone was considered a robust biosignature, as there was no known abiotic source that would produce it in high abundance on Earth. However, she said that it now isn't as simple as that. She said that oxygen is still considered an excellent biosignature because it is produced via photosynthesis; its substrates (water and carbon dioxide) are likely abundant on habitable exoplanets; and  $O_2$  is potentially very detectable because it is evenly mixed throughout the atmosphere and is spectrally active at visible and near-infrared wavelengths. However, recent work has found several abiotic methods that can create a large  $O_2$  atmosphere, often involving photolysis of  $H_2O$  or  $CO_2$ .<sup>20, 21,22</sup> Several of these methods occur on M-dwarf planets, which some consider to be particularly attractive to searches for life.

However, she said, false positives have signatures themselves (see Figure 3.2).<sup>23</sup> For a planet that obtained an atmosphere rich in oxygen by boiling off its oceans while orbiting a young M-dwarf star,<sup>24</sup> the oxygen atmosphere would become so pressurized and dense that a detectable amount of  $O_4$  would form in significant quantities.<sup>25</sup> In a separate example, Meadows said that, if photolysis of CO<sub>2</sub> were the source of  $O_2$  in the atmosphere, then large amounts of CO would also be apparent in the planet's spectrum.<sup>26</sup> In fact, these false positive indicators are often more observable than the biosignature itself, such as the  $O_4$  in the former scenario.<sup>27</sup>

#### Framework for Biosignature Assessment

Using the lessons learned from oxygen, Meadows said that the workshop aimed to develop a general framework for assessing biosignatures. The first step in this process is to characterize the important parameters in the planet's host star and its entire planetary system. Afterwards, the planet's properties must be characterized, and a search for biosignatures can be conducted. If any are found, potential false-positive scenarios must be further scrutinized.

In finding good biosignatures to choose from, Meadows listed three potential starting points. The easiest method would be to simply identify Earth's current biosignatures.<sup>28</sup> A disadvantage of this is that this limits you to the biosignatures of just one planet. Another method would be to explore Earth's past.<sup>29</sup> This expands the types of biosignatures one can search for, but knowledge of Earth's past environments and biosignatures is not fully developed. The most general method would be to explore a large number of potential volatiles that may be biosignatures.<sup>30</sup> However, without an example of a planet with life to analyze in context, this makes the risk of finding a false positive higher.

Giving a preview of the material in the workshop report, she showed a figure demonstrating that having liquid water on the surface is a function of the star, the properties of the planetary system, and the properties of the planet itself. She then listed four processes that could mimic false positives: geological/chemical (e.g., volcanism and serpentinization), mineralogical (e.g., surface reflectivity), photochemical (e.g., photolytic  $O_2$  and seasonal changes in gases), and atmospheric evolution ( $O_2$  produced from water loss). Ruling out these false positives could require additional observations beyond just the detection of a biosignature.

<sup>24</sup> Luger and Barnes, 2015.

<sup>&</sup>lt;sup>20</sup> Luger and Barnes, 2015.

<sup>&</sup>lt;sup>21</sup> Gao et al., 2015.

<sup>&</sup>lt;sup>22</sup> Harman et al., 2015.

<sup>&</sup>lt;sup>23</sup> V. Meadows, 2017, Reflections on O<sub>2</sub> as a biosignature in exoplanetary atmospheres, Astrobiology, accepted.

 $<sup>^{25}</sup>$  E.W. Schwieterman, V.S. Meadows, S.D. Domagal-Goldman, D. Deming, G.N. Arney, R. Luger, C.E. Harman, A. Misra, and R. Barnes, 2016, Identifying planetary biosignature impostors: Spectral features of CO and O<sub>4</sub> resulting from abiotic O<sub>2</sub>/O<sub>3</sub> production, *The Astrophysical Journal Letters* 819:13.

<sup>&</sup>lt;sup>26</sup> Gao et al., 2015.

<sup>&</sup>lt;sup>27</sup> Schwieterman et al., 2016.

<sup>&</sup>lt;sup>28</sup> J.E. Lovelock, 1975, Thermodynamics and the recognition of alien biospheres, *Proceedings of the Royal Society of London. Series B*, *Biological Sciences* 189:167.

<sup>&</sup>lt;sup>29</sup> G. Arney, S.D. Domagal-Goldman, V.S. Meadows, E.T. Wolf, E. Schwieterman, B. Charnay, M. Claire, E. Hébrard, and M.G. Trainer, 2016, The pale orange dot: The spectrum and habitability of hazy Archean Earth, *Astrobiology* 16:873.

<sup>&</sup>lt;sup>30</sup> S. Seager and W. Bains, 2015, The search for signs of life on exoplanets at the interface of chemistry and planetary science, *Science Advances* 1:e1500047.



FIGURE 3.2 Discriminating false positives for different environments. For a photosynthetic biosphere (left), the simultaneous presence of  $O_2, O_3, CH_4$ , and the absence of a large abundance of CO points to a photosynthetic origin for the Earth's  $O_2$ . The remaining panes show the predominant molecules for various false positive scenarios, the molecular discriminants whose presence indicates a false positive mechanism (circled) and the missing molecules (in bottom region) whose absence indicates a likely false positive mechanism. The nominal wavelength ranges needed to observe these discriminants or check for their absence are given at the bottom of each panel. From left to right (after Earth): abiotic O<sub>2</sub> from water photolysis due to a low non-condensable gas inventory (Wordsworth and Pierrehumbert, 2014); a massive O<sub>2</sub> atmosphere from water loss in which H<sub>2</sub>O may or may not still be present (Luger and Barnes, 2015); a habitable world in which a combination of an M-dwarf stellar spectrum and reduced atmospheric and surface sinks for O<sub>2</sub> has resulted in the buildup of abiotic O<sub>2</sub> from CO<sub>2</sub> photolysis or the generation of abiotic  $O_3$  (Harman et al., 2015); and a desiccated, hydrogen-poor environment in which  $O_2$  is a stable photolytic byproduct of CO<sub>2</sub> (Gao et al., 2015; Meadows, 2017). NOTE: See P. Gao, R. Hu, T.D. Robinson, C. Li, and Y.L. Yung, 2015, Stabilization of CO<sub>2</sub> atmospheres on desiccated M-dwarf exoplanets, The Astrophysical Journal 806:249; C.E. Harman, E.W. Schwieterman, J.C. Schottelkotte, and J.F. Kasting, 2015, Abiotic O2 levels on planets around F, G, K, and M stars: Possible false positives for life?, The Astrophysical Journal 812:137; R. Luger and R. Barnes, 2015, Extreme water loss and abiotic O<sub>2</sub> buildup on planets throughout the habitable zones of M dwarfs, Astrobiology 15:119; R. Wordsworth and R. Pierrehumbert, 2014, Abiotic oxygen-dominated atmospheres on terrestrial habitable zone planets, The Astrophysical Journal Letters 785:20. SOURCE: V. Meadows, 2017, Reflections on O<sub>2</sub> as a biosignature in exoplanetary atmospheres, Astrobiology, accepted, https:// creativecommons.org/licenses/by-nc/4.0; courtesy of R. Hasler, V. Meadows, and S. Domagal-Goldman, presented in Victoria Meadows, University of Washington/NASA Astrobiology Institute, "Extrasolar Biosignatures: Developing a Comprehensive Framework for Biosignature Recognition: Overview of the NExSS/NAI Biosignatures Workshop 2016," presentation to the Workshop on Searching for Life across Space and Time, December 5, 2016.

Meadows then briefly mentioned the difficulty of determining the confidence of any detected, potential biosignature and the possibility of novel biosignatures. She then finished with the following list of questions to guide the field forward:

- How do we discover biosignatures with high detection significance?
- How do we know that we're properly interpreting these as biosignatures in the right environmental context?

• Do we have the instrumental capability today or in planned missions to detect and identify biosignatures and their environments in order to put the results in context?

# **Audience Participation**

An audience member asked Meadows whether Earth's biosphere would be detectable before the rise of oxygen. Meadows said that any biosignatures present there would be harder to observe and to properly interpret. However, there could have been a disequilibrium present in the Archean Earth that might have been readily detectable. Pigments might also have existed that did not require photosynthesis to occur, such as for UV protection. However, she then emphasized again that it would be harder to detect life on an Archean Earth.

Responding to that, another participant then cautioned against the idea of just looking for disequilibrium gases. He gave an example of a gas like CO that could build up abiotically in some exoplanet atmospheres. He then stated that, on an Archean Earth, you might have methanogens that would actually drive the system towards equilibrium by metabolizing carbon dioxide and hydrogen into methane. Meadows agreed, but stressed the importance of the abundances of such potential substances. She said that serpentinization would produce maybe 5 ppm of methane naturally,<sup>31</sup> while biotic sources of life could produce enough methane to make up multiple percent of the atmosphere's composition.

Another audience member then said that Earth's history could have had multiple enrichment periods as new types of metabolism evolved into being, particularly in the first couple billions of years before animal predation. For example, methanogenesis could have led to hydrogen-based photosynthesis, then H<sub>2</sub>S-based photosynthesis, and then iron-based photosynthesis, each of which could have greatly enriched the atmosphere with certain biosignatures. Meadows agreed that this was an interesting point and emphasized again the abundance argument, the idea that there would be so much of a substance that it would immediately trigger our interest.

The idea of long-term trajectories was then raised by one member of the audience. For example, planets that develop plate tectonics might all evolve in similar ways. Deviations from this evolution could then be a potential biosignature. Meadows answered that Norm Sleep of Stanford University has looked at the way that life can affect the geology of a planet.<sup>32</sup> Noting the possibility that she might not be remembering it correctly, she said it was about how life could affect mineral assemblages and the behavior of rock in certain situations. However, she thought that geological evolution might be so variable that general statements about a planet's geological state may not be possible. Touching on a couple of asides, she then brought up the great difficulty of dating most stars, which often have uncertainty ranges on the order of billions of years. This then makes it difficult to date the age of the planets in the system. Then she said that habitability is not just a function of location, but of time. A planet that is habitable now may not have been in the past and/or may not be habitable in the future and vice versa.

Another workshop participant then added that Dr. Sleep also talked about a biological carbon pump that could send reductants to the sea floor and therefore affect the redox state of mineral assemblages. He then continued along the path of long-term, geological trajectories, saying that, to him, the habitable zone is the region where feedbacks push the planet toward having liquid water as opposed to away from having liquid water. However, he said, once life begins, it then becomes part of the feedback loops. Additionally, he stated that we need to build up a complete model of a lifeless planet and all its processes and understand how it works in order to obtain a null model. Figuring out how to do this using Earth is difficult, as life is nearly everywhere. Meadows agreed with that, stating

<sup>&</sup>lt;sup>31</sup> A. Guzmán-Marmolejo, A Segura, and E. Escobar-Briones, 2013, Abiotic production of methane in terrestrial planets, *Astrobiology* 13:550.

<sup>&</sup>lt;sup>32</sup> N.H. Sleep, D.K. Bird, and E. Pope, 2012, Paleontology of Earth's mantle, Annual Review of Earth and Planetary Sciences 40:277.

that "life is a planetary process." If life exists, you need to take it into account to get an explanation of the global system. She then stressed that looking at biosignatures must include understanding its environment and context.

An audience member then cautioned against the idea of an antibiosignature as it pertains to carbon monoxide. Abiotic sources, in principle, could create more CO than life could use, and therefore, a detection of CO would not necessarily mean that there was no life. He then cautioned against thinking we know what an abiotic planet might look like, since we cannot explain the amount of water on Earth or know how much water a typical habitable zone rocky planet might contain.

The final question directed toward Dr. Meadows regarded the technological capabilities of future missions being able to resolve the surface of an exoplanet and measure the composition of its optically thin atmosphere and whether it would ever be possible in the near or more distant future. Meadows answered that the Habitable Exoplanet Imaging Mission (HabEx) or the Large UV/Optical/Infrared Surveyor (LUVOIR) should be able to do it, presumably in about 20 years. She said that the Wide Field Infrared Survey Telescope (WFIRST) (expected launch date in the mid-2020s) might be able to do it too, if it is incredibly lucky. Transmission spectroscopy, where the atmosphere is viewed as the planet transits its star, would not be able to see a planetary surface, but direct imaging could see down to the surface, even if the atmosphere is partially cloudy. In fact, she said, if one could take images of a planet as it rotates, one could create a longitudinal map of the surface and potentially measure surface composition. However, disentangling the atmospheric signature from the surface signature would still be necessary.

#### EXTRASOLAR BIOSIGNATURES: THINKING OUTSIDE THE BOX

William Bains of the Massachusetts Institute of Technology continued on the subject of searching for biosignatures in exoplanets, but thought about it in a broader context of what life could produce. In "thinking outside the box," he said that what one really needs to do is to think in a much larger box. He chose this box to be "chemistry." Biosignatures visible over large distances would likely be volatiles in the atmosphere or colors on the surface,<sup>33</sup> so there needs to be an understanding on why life could make volatiles or colors using only the laws of chemistry as starting assumptions. He then gave the following list of three types of biosignatures gases:<sup>34</sup>

- Type I: A byproduct of energy capture (e.g.,  $H_2S + \frac{3}{2}O_2 \rightarrow SO_2 + H_2O)$ )
- Type II: A byproduct of biomass capture (e.g.,  $CO_2 + H_2O + hv \rightarrow [CH_2O] + O_2$ )
- Type III: No chemical "reason" at all (e.g.,  $C_6H_{14}N_4O_2 + 2O_2 + 3[H] \rightarrow NO + C_6H_{13}N_3O_3 + 2H_2O)$

# Type I

Bains said that there is a common expectation that life on other planets would use redox disequilibria to capture energy, the waste product(s) of which is (are) termed Type I biosignatures. How this plays out depends on the atmospheric composition of the planet. One can make predictions on what life might do in an atmosphere dissimilar to Earth's atmosphere of N<sub>2</sub> and O<sub>2</sub>, such as an atmosphere dominated by  $CO_2$ , H<sub>2</sub>, or N<sub>2</sub>. On such a planet, life would presumably react crustal rocks with the atmosphere, reducing them in the case of an H<sub>2</sub>-rich atmosphere. Some of these chemical reactions could produce energy that life could extract. An observable biosignature needs to be one of these energy-extracting chemical reactions that produces a volatile.

In an H<sub>2</sub> atmosphere, the only such volatiles are likely to be CH<sub>4</sub>, H<sub>2</sub>S, H<sub>2</sub>O, and NH<sub>3</sub>.<sup>35</sup> The first three would be expected in an H<sub>2</sub> atmosphere anyway, but Bains said that ammonia could be a good biosignature in an H<sub>2</sub> atmosphere. Ammonia would need to be produced at a rate that, at the very minimum, would maintain a detectable amount of it in the atmosphere despite ammonia removal through atmospheric photochemistry. This requires a certain level of biomass. A calculation of the minimum biomass (under the most favorable conditions) needed to

<sup>&</sup>lt;sup>33</sup> S. Seager and W. Bains, 2015, The search for signs of life on exoplanets at the interface of chemistry and planetary science, *Science Advances* 1:e1500047.

<sup>&</sup>lt;sup>34</sup> S. Seager, M. Schrenk, and W. Bains, 2012, An astrophysical view of Earth-based metabolic biosignature gases, Astrobiology 12:61.

 $<sup>^{35}</sup>$  S. Seager, W. Bains, and R. Hu, 2013, Biosignature gases in H<sub>2</sub>-dominated atmospheres on rocky exoplanets, *The Astrophysical Journal* 777:95.

|                                   | Biosignature<br>Gas Type | Thermal Emission (gm/cm <sup>2</sup> ) |                      |                      | Transmission (gm/cm <sup>2</sup> ) |                      |
|-----------------------------------|--------------------------|--|----------------------|----------------------|------------------------------------|----------------------|
| Compound                          |                          | Sun-like                               | Active M Dwarf       | Quiet M Dwarf        | Active M Dwarf                     | Quiet M Dwarf        |
| NH <sub>3</sub>                   | Туре І                   | $4.0 \times 10^{-4}$                   | $8.0 \times 10^{-6}$ | $9.5 \times 10^{-6}$ | 1.1                                | $1.8 \times 10^{-9}$ |
| CH <sub>3</sub> Cl                | Type III                 | 2,800                                  | 77                   | 0.013                | 860                                | 0.014                |
| (CH <sub>3</sub> ) <sub>2</sub> S | Type III                 | 190                                    | 82                   | 0.0001               | 260                                | 0.00036              |
| CS <sub>2</sub>                   | Type III                 | $5.5 \times 10^{7}$                    | $2.3 \times 10^{7}$  | 37                   | $1.5 \times 10^{7}$                | 24                   |
| ocs                               | Type III                 | $1.3 \times 10^{5}$                    | 5,500                | 0.67                 | $9.9 \times 10^{4}$                | 12                   |

TABLE 3.2 Biomass Needed to Maintain Detectable Levels of Biosignature Gases in an Atmosphere of a Habitable Zone Planet with an H<sub>2</sub>-Dominated Atmosphere at P = 1 bar

maintain a detectable level of ammonia shows that is possible (see Table 3.2). Bains noted that the apparently barren Sechura Desert in Peru has 10 times the biomass necessary for thermal detection on a planet with an  $H_2$  atmosphere orbiting a Sun-like star and orders of magnitude more for an M-dwarf star, if that life used ammonia production as a primary source of energy. However, careful consideration of false positives and false negatives is necessary.

# Type II

Type II gases are produced by biomass capture. On Earth, life needs to grab carbon from  $CO_2$  and throw away the oxygen. On a planet with an H<sub>2</sub>-dominated atmosphere, it would need to take carbon from  $CH_4$ , the most likely dominant carbonaceous gas, and throw out the hydrogen.<sup>36</sup> Bains ran through the likely Type II chemical reactions and came to the conclusion that the most plausible path to get a biosignature is a planet using methane, water, and energy to produce biomass and H<sub>2</sub> gas, a result he called "incredibly disappointing" since the planet already has a hydrogen-dominated atmosphere from abiotic sources. This reaction can use near-infrared photons to power it, meaning that there likely would be not a red edge on this planet either. On the other hand, Bains said, false positives are less of a problem because they are thermodynamically implausible. A false negative could occur if life had not yet evolved to that stage though.

Going more in-depth on the issue of color, Bains noted Earth's red edge, the fact that Earth's plants are very reflective in the near-infrared. However, this is not inevitable. He pointed out that begonia leaves (not petals) are blue. He then showed a figure of a spectral analysis of many different types of things, in which the rocks and living materials were distinct enough that it could be used to classify a spectrum as coming from something living or non-living. A problem with this is that it does not take into account that, in real environments, rocks would be mixed in with plants, and deconvoluting them could be difficult.

Bains then asked what color alien life would be. Photosynthesis merely requires plants to absorb somewhere in the star's spectral range, but that is a weak constraint. Beyond that, he thought that we don't really have an idea what color life would be. He then went through a few examples. UV protection might be generally favorable, but on Earth, melanin is black and looks like rock. Pigments to capture photons evolved at least four independent times (chlorophyll, bacteriorhodopsins, aphid carotenes, and melanized fungi), each pigment having its own absorption characteristics. He then noted that, out of these four cyclic catalytic pathways to capture  $CO_2$ , only one has been exhibited in biomass, and Bains didn't think that we knew why.

# **Type III**

Moving on to Type III, Bains performed the same type of biomass calculation as before (see Table 3.2).<sup>37</sup> Many of them would require huge amounts of biomass to maintain an atmosphere with a detectable level of a Type III biosignature (especially for Sun-like stars), up to an equivalent of "a column of cabbages hundreds of meters high"

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<sup>&</sup>lt;sup>36</sup> W. Bains, S. Seager, and A. Zsom, 2014, Photosynthesis in hydrogen-dominated atmospheres, *Life* 4:716.

<sup>&</sup>lt;sup>37</sup> Seager et al., 2013.

for thermally detected  $CS_2$  around a Sun-like star. All of this was based on gas production rates found in Earth life, however, where there is little understanding of why life chose particular secondary metabolite products. For example, there are 34 halomethanes possible, but life is only known to produce 22 of them, and nobody knows why. Another example is that life does not use fluorine well. He said that, while it is very electronegative, so is oxygen, which life uses aplenty.

In order to approach all of this systematically, Bains noted that Sara Seager suggested building up a catalog of all small molecules possible and then working backwards to filter for stability, volatility, and so on.<sup>38,39</sup> This even includes things not produced by life directly, but molecules produced by industry too. Two of the top-level filters they are applying to these molecules are whether they are detectable and whether they can be produced geochemically. Entropy of formation is also important to look at because bigger molecules are less likely to form spontaneously. This catalog is intended to build up a "geological plausibility index" to determine how likely it is that a molecule might be produced by geology and, inversely, how likely it might be produced by life.

# **Audience Participation**

An audience member asked about whether kinetic arguments, rather than purely thermodynamic arguments, can guide interpretation. He cited  $N_2$  as an example. Treating an  $N_2$  atmosphere only as an energetic sink means that life would be unlikely to exist on such a planet. However, since  $N_2$  is a kinetic sink, this suggests that a detection of  $N_2$  does not preclude life. Referring back to the previous talk by Meadows, Bains said that nitrogen is very robust. While nitrogen and oxygen will eventually get turned into nitrate in the ocean, he said that it will then be broken back down into nitrogen in hydrothermal systems. Therefore, he said, finding a thermodynamic disequilibrium is not really useful without a better understanding of the system's kinetics.

Agreeing with Bains, another member of the audience disputed the idea that, if there is a disequilibrium, it means that there is no life because otherwise life would have used it to make more life. He said that this is a visible fact just by looking at  $O_2$  and  $N_2$  in the atmosphere or seeing a forest outside in an  $O_2$  atmosphere. This is not a failure of life, he said, but a failure of Darwinism. However, after life becomes intelligent, it tries to remove Darwinism in favor of Lamarckism. He then asked what the consequences would be of an intelligent species exploiting Lamarckism. Bains agreed to the first part, saying that a thermodynamic disequilibrium is a red herring. Bains commented that evolution is inefficient at finding the optimal solution, and as the audience member's comment said, it may leave thermodynamic disequilibria unexploited. He also commented that evolution was a poor biomarker, one reason being that you cannot observe evolution of life on other planets.

A workshop participant then brought up the topic of geochemical false positives, saying that one must take into account time. He gave hydrogen as an example. An average-sized planet, he said, would not be able to produce  $H_2$  effectively early on, but after about a billion years, it could then do so. He said that one must look at the planet's age and environment. Bains agreed.

Going back to Bains' point about ammonia being a good biosignature in an  $H_2$ -dominated atmosphere, an audience member suggested that ammonia in the atmosphere could be produced by a comet impact before the observation and asked how to get around these kinds of special events. Bains answered that it requires knowing about the temporal context: how old the world is and how it's changing. This could show that an observation is not a one-off event.

On a similar note, another audience member said that an old paper discovered the reduction of nitrogen to ammonia on desert sands using titanium dioxide as a catalyst, which showed that there is an abiotic way to create ammonia.<sup>40</sup> Bains noted that the audience member had brought this point to Bains' attention before. He then stressed the abundance point from the previous talk, noting that a whole lot of titanium dioxide would be necessary for that scenario.

<sup>&</sup>lt;sup>38</sup> Seager and Bains, 2015.

<sup>&</sup>lt;sup>39</sup> S. Seager, W. Bains, and J.J. Petkowski, 2016, Toward a list of molecules as potential biosignature gases for the search for life on exoplanets and applications to terrestrial biochemistry, *Astrobiology* 16:465.

<sup>&</sup>lt;sup>40</sup> G.N. Schrauzer and T.D. Guth, 1977, Photolysis of water and photoreduction of nitrogen on titanium dioxide. *Journal of the American Chemical Society* 99:7189.

A workshop participant then asked Bains what his practical suggestion was for moving forward. Bains answered that they are trying to categorize all the small molecules to try to rule out things that are likely to have a geological origin. He went on to say that there are major gaps in our understanding in reaction chemistry, such as how stable molecules are in water. The same goes for atmospheric photochemistry. Bains wants a huge database of how different molecules react in different environments under different conditions.

Then an audience member asked for Bains's opinion on the limits of life using different energy sources. He said that, for example, no life extracts mechanical energy. The audience member then said that life is lazy and doesn't want to do anything if there is an available gradient to use instead. Bains replied that there are some energy sources that are just too diffuse to be usable, such as Earth's magnetic field. Bains then agreed that life is lazy (and intelligent life lazier). Difficult steps could take a long time to accomplish. For example, making oxygen from water is chemically difficult to do, and it seems like it took a long time for life to be able to do it. A planet that could support methanogenesis or oxygenesis might have life that has not yet evolved to do it.

A question was then posed to Bains about whether there could be biosignatures in the UV region that could complement the more commonly suggested biosignatures. Bains said that there are biological molecules that absorb in the UV, but he was unsure of geological molecules. Another audience member then answered that ozone photolytically produced by  $O_2$  is the best example. In the Proterozoic era when there was less  $O_2$ , however, the ozone signal may nonetheless be visible in the UV. Methane, on the other hand, absorbs at UV wavelengths that telescopes are unlikely to be able to observe. Other molecules could work too, but they all have better lines in the visible and the infrared. Another audience member then chimed in to say that pigments could absorb in the UV, but that these are not apparent on Earth because little UV radiation makes it to the surface. On other planets, however, these pigments could potentially create a strong surface signature.

#### **TECHNOLOGY NEEDS TO DISCOVER EARTH 2.0**

Nick Siegler of the NASA Jet Propulsion Laboratory (JPL) began his talk by stating that the main goal of the Exoplanet Exploration Program technology effort is to enable future space missions to observe a planetary spectrum of a rocky planet in the habitable zone of its star and understand it in the context of potential life. He went on to say that the main exoplanet discovery tools—the radial velocity and transit techniques, which have discovered more than 95 percent of the more than 3,400 known exoplanets—will not be the techniques to directly image exoplanets, which is needed to get a reflected light spectrum. Spectroscopy will be hard because there simply aren't many photons available to use, but it will not be the biggest problem. The biggest problem will be suppressing the light from the stars, which can be 10 billion times brighter than a rocky planet in the habitable zone of a Sun-like star. Starlight suppression could be done in one of the following three ways: internal occulters (i.e., starshades), and nulling interferometers. The latter option is the least technologically mature of the options and one that NASA is not currently pursuing.

#### Coronagraphs

While the Hubble Space Telescope (HST) and the James Webb Space Telescope (JWST) both have coronagraphs, Siegler explained, WFIRST will be the first space telescope with a coronagraph (or possibly a starshade) specifically designed for directly imaging exoplanets. WFIRST's Wide-Field Instrument (WFI) will arguably help answer questions in three of the biggest astrophysical areas—dark matter, dark energy, and exoplanets (via microlensing and coronagraphy). The telescope's coronagraph instrument (CGI) will be used for the direct imaging and spectroscopy of exoplanets. WFIRST is in its formulation phase (Phase A) at this time. The project, telescope, and WFI are managed by NASA Goddard Space Flight Center, while the CGI is managed by JPL. The project has now also been directed to study the compatibility of a starshade with WFIRST. The current state of the art for coronagraphs is the Gemini Planet Imager (GPI) and the Very Large Telescope Spectro-Polarimetric High-contrast Exoplanet Research instrument (VLT SPHERE). WFIRST would improve upon their contrast ratio capability by 2 to 3 orders of magnitude and also improve upon the ability to probe smaller planet-star separations (see Figure

3.3). Further technological advancement would be required to observe rocky planets in the habitable zone of stars at a distance of 10 parsec (pc) and further.

Siegler then showed a video from JPL about how a classical coronagraph works.<sup>41</sup> As a star's light, depicted in the form of a wavefront, passes through the telescope, it becomes distorted by the slight imperfections inherent in any telescope's optics. Diffraction adds concentric rings to the images. To see the planets, a mask is inserted to block most of the star's light and redirect the rest of the light to the outer edge. A washer-shaped object then blocks most of the redirected light. Because the planet's light comes in at an angle, it misses the first mask and goes through the center hole of the washer-shaped object. At this point, the planet's light is still obscured by the residual starlight leaking through. To reduce the amount of leaking starlight, a deformable mirror is used to correct the distortions in the incoming light beam. This can then reveal the existence of a planet in the image up to a billion times fainter than the star. The video finished by saying that the planet's light can then be directed into a spectrograph for spectral analysis.

Siegler then continued, elaborating a list of what a future telescope with a coronagraph would need in order to study Earth-like planets in Earth-like orbits around Sun-like stars. It would need to improve its contrast ratio sensitivity relative to that of WFIRST's coronagraph by about two orders of magnitude. Deformable mirrors and image post-processing are fairly well advanced but need to go farther. Integration times would be days to weeks typically, so the system needs to be extremely stable. Otherwise, telescope vibrations and thermal distortions can cause blurriness. Siegler said that wavefront sensors would need to be able to measure wavefront distortions up to 10 picometers (pm), a couple of orders of magnitude better than HST (the current best), and correct for them. The technological capability to build large, segmented mirrors in a way that the optics are phase coherent—which may be required to build telescopes with primary mirrors exceeding 4 m—to within at least nanometers is not yet developed. Because of the long integration times, photon rates will be measured in photons per minute, so detectors with ultralow read noise are necessary, especially in the infrared. The size of the telescope is another question, especially with regard to a large, monolithic mirror versus a segmented mirror. Siegler then showed an image of potential telescope architectures for 12-m segmented mirrors of various segment sizes and shapes (hexagonal to a more radial, pie-like structure). The main problem with segmented mirrors is that all the small gaps add additional layers of diffraction, and the primary purpose of a coronagraph is to remove diffraction.

#### Starshades

Siegler then showed an animation of a telescope with a starshade.<sup>42</sup> They were two separate spacecraft with separate propulsion systems. When aligned, the starshade blocked the star's light, revealing the reflected light of the planets. The starshade possesses a petal-like shape which serves to reorient the diffraction, creating a dark shadow for the telescope. He claimed that, in many ways, a starshade is a simpler method than the coronagraph because the starshade is doing all the work. It drastically reduces wavefront-control requirements on sensitivity, segment phasing, and other corrections. It has a higher tolerance for error as long as the starshade performs as designed. The starshade would be tens of meters across and tens of thousands of kilometers away. The starshade needs to be able to deploy and position its petals and maintain its physical stability, suppress the starlight, and fly in formation with a telescope separated from it by tens of thousands of kilometers and maintain the telescope's lateral offset within acceptable limits. He then showed a starshade optical demonstration performed by Northrop Grumman in the Nevada desert, which was able to detect a simulated planet 100 million times fainter. Another experiment used a baseline of 2.4 km with a solar telescope to block out Arcturus and observe background stars. Another test, currently ongoing at Princeton University, has exceeded a contrast ratio of 10<sup>-8</sup> at a single wavelength of 632 nanometers.

The starshade will be challenging to manufacture. The petals, he said, will need to be about 6 to 8 meters in length and fabricated to a tolerance of about 100 microns. The petals will need to be deployed to millimeter-level

<sup>&</sup>lt;sup>41</sup> NASA, "The Search for Alien Earths—How Coronagraphs Find Hidden Planets," video, https://exoplanets.nasa.gov/exep/coronagraph-video/, accessed December 5, 2016.

<sup>&</sup>lt;sup>42</sup> NASA Jet Propulsion Laboratory, Flower Power Starshade Unfurls in Space," March 20, 2014, http://www.jpl.nasa.gov/video/details. php?id=1284.



# Exoplanet Direct Imaging in the Optical and Near-infrared

Separation (arcsec)

FIGURE 3.3 Direct imaging current and future planet-star contrast ratios (ratio of planet brightness to host star brightness) versus the apparent planet-star angular separation. The filled orange circles indicate the direct imaging of young, self-illuminous planets imaged in the near-infrared by ground-based telescopes (all are gas giants). Contrast ratios for the planets of the solar system are for analogous planets placed 10 parsecs away and observed at visible wavelengths. The solid black circles are contrast ratio estimates of measured radial velocity (RV) planets, including Proxima Centauri b. The orange curves show measured instrument performance at near-infrared wavelengths on ground-based coronagraphs. The Gemini Planet Imager (GPI) curve shows typical instrument performance while the SPHERE instrument curve-the extreme adaptive optics system and coronagraphic facility at the Very Large Telescope array (VLT)-shows the best achieved instrument performance to-date on the star Sirius. Achieved performance with the Hubble Space Telescope's (HST's) Advanced Camera for Surveys (ACS) coronagraph masks and the predicted instrument performance of the James Webb Space Telescope (JWST) Near-Infrared Camera (JWST NIRCam) masks are also shown. For consistency, the imaged planets discovered in the near-infrared are shown with vertical arrows pointing to the predicted contrast ratios at visible wavelengths; the Wide Field Infrared Survey Telescope (WFIRST) coronagraph is expected to conduct science between 442 and 980 nm (see Wide-Field Infrared Survey Telescope, WFIRST Simulations, "Spacecraft and Instrument Parameters," https://wfirst.ipac.caltech.edu/sims/Param\_db.html#coronagraph\_ imaging, accessed December 6, 2016). The current threshold requirement at 565 nm for the WFIRST coronagraph instrument (CGI) is shown as the black, solid curve; the black, dashed line is the projected enhanced performance after further technology improvements before launch. SOURCE: NASA Exoplanet Exploration Program, https://exoplanets.nasa.gov/exep/technology/ gap-lists/, accessed December 5, 2016, presented in Nick Siegler, Jet Propulsion Laboratory, "Technology Needs to Discover Earth 2.0," presentation to the Workshop on Searching for Life across Space and Time, December 5, 2016.

precision. JPL tested a deployment method for the petals, showing proof of concept. Another challenge is how to store an opaque starshade for launch and then deploy it without snagging or damaging it, since the starshade relies on its ability to remain opaque. A small, origami-like folding technique worked, so Siegler said that "JPL held back no expense" and performed a larger version (about half the size WFIRST would need) using corrugated cardboard and three interns. A recent prototype demonstrated a smaller starshade, but with more flight-like materials such as Mylar and high-density polyurethane. He then added that they think they have figured out formation flying to meter-level precision using current equipment on WFIRST.

Siegler said that everything in his talk could be found in the Exoplanet Exploration Program Technology Plan Appendix from 2016 (the 2017 update is now released and can be found at their website).<sup>43</sup> He then brought up a slide showing past and future NASA and European Space Agency exoplanet missions, such as CHEOPS and PLATO. He requested that future planning think favorably of exoplanets, since we won't be able to analyze biosignatures and false positives or negatives unless we can directly image these exoplanets. Siegler finished by mentioning two NASA-chartered mission concept studies that will be considered for possible future missions that could dramatically advance the field of exoplanets: the Habitable Exoplanet Imaging Mission (HabEx), a 4-m monolithic mirror or 6.5-m segmented mirror, or the Large UltraViolet/Optical/Infrared Surveyor (LUVOIR), a 9- to 16-m segmented mirror.

#### **Audience Participation**

An audience member asked what the expected lifetime of a starshade would be and how many targets it could reach before running out of fuel. Siegler pointed out that this is a valid question due to the fact that micrometeoroids in space would likely pierce the starshade, limiting its lifetime. He said that, with multiple plies in the starshade, a micrometeoroid is unlikely to pierce perfectly orthogonal to the starshade where leaked light could do the most damage. He estimated a lifetime on the order of years. Pressed on the topic of fuel, Siegler explained a scenario that uses chemical propulsion to keep the starshade aligned with micro-thrusts and uses solar-electric propulsion for slewing to different targets. Another option is having two starshades so that one could be in operation while the other one was slewing.

Staying on the topic of the starshade, another participant pointed out that the tips of the petals have to be precise and sharp and then asked how they would clean dust off of them. Siegler said that they don't know yet, but agreed that the tips need to be razor thin, about one micron thick. Dust, typically on the order of a wavelength, could be a problem.

A biologist then asked why astronomers were so focused on Earth-like planets and so pessimistic about hot Jupiters. He thought that only about 30 percent of the NASA exoplanet program portfolio should be about Earth-like planets, not 100 percent. Siegler said that he embraced that view but explained that by focusing the technology development on detecting Earth-like planets, you get the other planets for free. WFIRST, for example, would be able to detect hundreds of cold Jupiters, Saturns, and Neptunes too. Another audience member then commented that WFIRST will get about 10 times more total planets than rocky planets in the habitable zone by doing an observational sweep in direct imaging.

That same commenter then raised a new question about whether the trick allowing a potential starshade to work with WFIRST would also allow one to work with JWST. Siegler answered that NASA did study whether JWST could be designed to be compatible with a starshade, but they decided against it for technical and programmatic reasons. He then moved back to WFIRST and the collaboration between the JPL starshade and coronagraph teams and the Goddard spacecraft team. He said that the teams had found a relatively simple approach that addressed telescope-starshade alignment requiring minimal modifications to existing instrumentation. The WFIRST project has been asked to continue carrying starshade compatibility in their designs subject to review. A final decision would likely be made by NASA no later than fiscal year 2018.

An audience member then asked about the precision of the stability between the starshade and the telescope. Siegler noted that this is the formation flying issue. He said that the dark shadow of the starshade is about 2 m in

<sup>&</sup>lt;sup>43</sup> NASA, "Technology Needs and Gap Lists," https://exoplanets.nasa.gov/exep/technology/gap-lists/, accessed December 5, 2016.

40

diameter and is cylindrically shaped. The lateral precision needs to be within 1 m, but the on-axis precision can have tolerances of hundreds of kilometers. The 1-m control precision has been done before on other spacecraft, including those docking with the space shuttle, but the angular alignment required with WFIRST is on order of milli-arcseconds, which is in a whole new regime. The audience member then asked him how problematic he felt this was. Siegler answered that recent testbed demonstrations were relieving him of his concern. He no longer thinks that the two spacecraft sensing their relative positions is a problem and said that the necessary control has never been a problem.

The final questioner then asked what WFIRST could do for exomoons. Prompted by the audience, Siegler responded by saying that WFIRST's microlensing capabilities could potentially detect an exomoon, which would have a very unique lensing signature. However, spectral characterization would be impossible.

#### PROSPECTS FOR GROUND-BASED CHARACTERIZATION OF PROXIMA CENTAURI B

Matteo Brogi of the University of Colorado, Boulder, began his talk by saying that he has been using groundbased telescopes to look at hot Jupiters with high-resolution spectroscopy. He said that this could be possible for smaller, fainter planets in the future, such as Proxima Centauri b, which orbits the nearest star to the Sun (1.3 pc away).<sup>44</sup> Proxima Centauri b has a minimum mass (msini) of 1.27 M $\oplus$ , a semi-major axis of 0.05 AU, a period of 11 days, a radial velocity semi-amplitude of 1.4 m/s, and does not appear to transit.

# **M-Dwarf Habitable-Zone Planets**

Brogi then stressed that, not only is there a planet around the closest star, but that the size of the planet represents the most common type of planet around FGK dwarfs<sup>45</sup> and especially M dwarfs,<sup>46</sup> according to Kepler. Having a small planet orbiting a small star gives a higher planet-to-star contrast ratio than if the same planet orbited a larger star. This is true for measurements of the planet's transit depth, reflected light, and thermal emission. The transit depth depends on the relative size of the planet and the star. The thermal emission also depends on the square of the relative planet-to-star radius, and additionally, it has a strong dependence on the temperature of the two bodies. Smaller stars are also cooler, hence they appear fainter when compared to the planet. Finally, habitable-zone planets orbiting M dwarfs need to be very close to the star. With the quadratic dependence on semi-major axis, the reflected light contrast ratio will drastically improve. Brogi then pointed out that M dwarfs are the most common type of stars nearby (about 70 to 80 percent are M dwarfs). Putting all the information together, Dressing and Charbonneau<sup>47</sup> in 2015 predicted that the nearest transiting and non-transiting exoplanets in the habitable zone is about 10.6 pc and 2.6 pc away, respectively. Proxima Centauri b is even closer than that.

From basic energy arguments, Brogi said that the habitable zone will be much closer to the star than for a Sun-like star. Proxima Centauri b is in the middle of the classical habitable zone for an M-dwarf star (like Proxima Centauri).<sup>48</sup> He stressed again that this means that the reflected light signal will be enhanced. The transit probability (radius of the star divided by the semi-major axis, assuming a circular orbit) of an M-dwarf, habitable-zone planet will also be enhanced, and because of the shorter period, transits are more frequent and can be stacked to increase the signal-to-noise ratio.

<sup>&</sup>lt;sup>44</sup> G. Anglada-Escudé, P.J. Amado, J. Barnes, Z.M. Berdiñas, R.P. Butler, G.A.L. Coleman, I. de la Cueva, et al., 2016, A terrestrial planet candidate in a temperate orbit around Proxima Centauri, *Nature* 536:437.

<sup>&</sup>lt;sup>45</sup> F. Fressin, G. Torres, D. Charbonneau, S.T. Bryson, J. Christiansen, C.D. Dressing, J.M. Jenkins, L.M. Walkowicz, and N. Batalha, 2013, The false positive rate of Kepler and the occurrence of planets, *The Astrophysical Journal* 766:81.

<sup>&</sup>lt;sup>46</sup> C.D. Dressing and D. Charbonneau, 2015, The occurrence of potentially habitable planets orbiting M dwarfs estimated from the full Kepler dataset and an empirical measurement of the detection sensitivity, *The Astrophysical Journal* 807:45.

<sup>47</sup> Ibid.

<sup>&</sup>lt;sup>48</sup> R.K. Kopparapu, R. Ramirez, J.F. Kasting, V. Eymet, T.D. Robinson, S. Mahadevan, R.C. Terrien, S. Domagal-Goldman, V. Meadows, and R. Deshpande, 2013, Habitable zones around main-sequence stars: New estimates, *The Astrophysical Journal* 765:131.

# **Characterizing Exoplanet Atmospheres**

Up until a few years ago, transiting planets offered the only opportunity to characterize atmospheres, Brogi said. While one cannot separate a planet's light from the starlight solely based on the system's geometry and differential measurements in time, it is possible to discriminate between the two. When the planet is in front of the star, the star's flux appears to drop, and some of the starlight gets filtered through the planet's atmosphere. The atmosphere's spectral fingerprint is then imprinted onto the observed spectrum, which is called a transmission spectrum. When the planet moves behind the star, the flux from the planet (thermal and reflected) disappears, so one can compare the spectrum before and after to characterize the planet's atmosphere. Stable, sensitive instrumentation can also measure the total light of system as a function of the planet's orbital phase, which is called a phase curve. All of these can be used to help determine the atmosphere's composition, thermal structure, and energy balance.

However, Brogi reminded us that Proxima Centauri b does not transit. For nontransiting planets, he proposes to use high-resolution Doppler spectroscopy to separate the planet's light from the starlight in the spectral domain in addition to the temporal domain. An advantage of high-resolution spectroscopy, he said, is that each molecular species is resolved into the individual lines, resulting in unique and very specific fingerprints. Matching techniques, such as cross-correlation, can be used to detect these species unambiguously. Planets orbiting close to their parent stars also acquire a very distinct Doppler signature due to their orbital motion. Brogi showed a figure demonstrating, with a toy model, the visibility of the planet signature with respect to the telluric lines from Earth's atmosphere (see Figure 3.4). While the planet is moving along the orbit, its radial velocity changes by tens of kilometers per second. In contrast, Earth's atmospheric absorption (telluric absorption) lines remain stationary in velocity (i.e., in wavelength). This duality allows us to effectively disentangle the contaminating telluric signal from the exoplanet signal and to remove the former very effectively without altering the latter. The residual data is then cross-correlated with model spectra for exoplanet atmospheres to combine the signal of all molecular lines. In this way, detections of molecular species also deliver the planet's radial velocity. When compared to the previously known stellar radial velocity, the planet and the star are treated as a spectroscopic binary. This technique allows for a measurement of the planet's mass and inclination without needing the planet to ever transit. A caveat to this, Brogi said, is that the result is not a real planet spectrum, but a likelihood function that is subject to uncertainties in the theoretical models. Brogi and collaborators have used this method successfully, mostly on the Very Large Telescope's Cryogenic High-Resolution InfraRed Echelle Spectrograph (VLT CRIRES) around 2.3 and 3.2 microns. They have been able to measure the mass and orbital inclinations of three hot Jupiters ( $\tau$ Boo b,<sup>49</sup> HD 179949 b,<sup>50</sup> and 51 Peg b<sup>51</sup>). Both CO and H<sub>2</sub>O have been confidently measured in the atmospheres of transiting and nontransiting planets.<sup>52,53,54</sup> (They did not, however, find  $CH_4$ , which is not surprising for these high-temperature planets.) While no thermal inversions have been detected, exoplanet rotation and winds have been measured based on the broadening of the cross-correlation function.55,56

 $<sup>^{49}</sup>$  M. Brogi, I.A.G. Snellen, R.J. de Kok, S. Albrecht, J. Birkby, and E.J.W. de Mooji, 2012, The signature of orbital motion from the dayside of the planet  $\tau$  Boötis b, *Nature* 486:502.

<sup>&</sup>lt;sup>50</sup> M. Brogi, I.A.G. Snellen, R.J. de Kok, S. Albrecht, J.L. Birkby, and E.J.W. de Mooji, 2013, Detection of molecular absorption in the dayside of exoplanet 51 Pegasi b?, *The Astrophysical Journal* 767:27.

<sup>&</sup>lt;sup>51</sup> M. Brogi, R.J. de Kok, J.L. Birkby, H. Schwarz, and I.A.G. Snellen, 2014, Carbon monoxide and water vapor in the atmosphere of the non-transiting exoplanet HD 179949 b, *Astronomy and Astrophysics* 565:124.

<sup>&</sup>lt;sup>52</sup> I.A.G. Snellen, R.J. de Kok, E.J.W. de Mooij, and S. Albrecht, 2010, The orbital motion, absolute mass and high-altitude winds of exoplanet HD209458b, *Nature* 465:1049.

<sup>&</sup>lt;sup>53</sup> J.L. Birkby, R.J. de Kok, M. Brogi, E.J.W. de Mooij, H. Schwarz, S. Albrecht, and I.A.G. Snellen, 2013, Detection of water absorption in the day side atmosphere of HD 189733 b using ground-based high-resolution spectroscopy at 3.2 μm, *Monthly Notices of the Royal Astronomical Society* 436:35.

<sup>&</sup>lt;sup>54</sup> R.J. de Kok, M. Brogi, I.A.G. Snellen, J. Birkby, S. Albrecht, and E.J.W. de Mooij, 2013, Detection of carbon monoxide in the high-resolution day-side spectrum of the exoplanet HD 189733b, *Astronomy and Astrophysics* 554:82.

<sup>&</sup>lt;sup>55</sup> I.A.G. Snellen, B.R. Brandl, R.J. de Kok, M. Brogi, J. Birkby, and H. Schwarz, 2014, Fast spin of the young extrasolar planet β Pictoris b, *Nature* 509:63.

<sup>&</sup>lt;sup>56</sup> M. Brogi, R.J. de Kok, S. Albrecht, I.A.G. Snellen, J.L. Birkby, and H. Schwarz, 2016, Rotation and winds of exoplanet HD 189733 b measured with high-dispersion transmission spectroscopy, *The Astrophysical Journal* 817:106.



FIGURE 3.4 Doppler signature from a toy model of a giant exoplanet orbiting close to its parent star (i.e., a hot Jupiter). The simulated planet spectrum and the telluric absorption lines are shown in white and black, respectively, at a spectral resolution of 100,000. Molecular absorption from planetary CO is resolved into the individual lines, and its Doppler shift due to the planet's orbital motion is detected. This allows (1) an effective disentanglement of the planet and telluric signals, (2) a reliable identification of molecular species via line-matching (i.e., by cross-correlating with models), and (3) a measurement of the planet's radial velocity. For a transiting planet, the maximum signal happens during transit and just before or after secondary eclipse. However, since the method directly targets the planet's thermal emission, nontransiting planets can be studied as well by observing around orbital phase 0.5, and in this way, their masses and orbital inclinations can be determined. SOURCE: Matteo Brogi, University of Colorado, Boulder, "Prospects for groundbased characterization of Proxima Centauri b," presentation to the Workshop on Searching for Life across Space and Time, December 5, 2016.

In order to use this same method on rocky, habitable-zone planets, Brogi illustrated how to combine it with high-spatial resolution via future integral-field, high-resolution spectrographs (IFSs).<sup>57,58</sup> This allows for the same analysis as on the hot Jupiters mentioned previously, but for each individual pixel of the IFS. He said that spreading out the photons spectrally does not cause a loss of signal, because spectra are recombined at a later stage during cross-correlation. Brogi's group tested the method already with VLT CRIRES on known directly imaged planets.<sup>59,60</sup> He said that it worked because of two factors that help increase the signal-to-noise ratio. First, the signal-to-noise ratio increases with the square root of the number of lines analyzed. Second, the contaminating light from the star is suppressed. They ran a simulation of this method using the parameters of the future European Extremely Large Telescope (E-ELT, 39 m diameter) with the Mid-Infrared E-ELT Imager and Spectrograph (METIS), which is a high-resolution, near-infrared spectrograph with an integral field unit. With just classical direct

<sup>&</sup>lt;sup>57</sup> I. Snellen, R. de Kok, J. L. Birkby, B. Brandl, M. Brogi, C. Keller, M. Kenworthy, H. Schwarz, and R. Stuik, 2015, Combining highdispersion spectroscopy with high contrast imaging: Probing rocky planets around our nearest neighbors, *Astronomy and Astrophysics* 576:59.
<sup>58</sup> C. Lovis, I. Snellen, D. Mouillet, F. Pepe, F. Wildi, N. Astudillo-Defru, J.-L. Beuzit, et al. 2017, Atmospheric characterization of Proxima

b by coupling the SPHERE high-contrast imager to the ESPRESSO spectrograph, Astronomy and Astrophysics 599:16.

 $<sup>^{59}</sup>$  I.A.G. Snellen, B.R. Brandl, R.J. de Kok, M. Brogi, J. Birkby, and H. Schwarz, 2014, Fast spin of the young extrasolar planet  $\beta$  Pictoris b, *Nature* 509:63.

<sup>&</sup>lt;sup>60</sup> H. Schwarz, C. Ginski, R.J. de Kok, I.A.G. Snellen, M. Brogi, and J.L. Birkby, 2016, The slow spin of the young substellar companion GQ Lupi b and its orbital configuration, *Astronomy and Astrophysics* 593:74.

imaging, a planet slightly larger and warmer than Earth orbiting Alpha Centauri B would barely be detectable. However, when using cross-correlation filtering, a 5 $\sigma$  detection of the planet would be made after 10 hours (8 $\sigma$  after 30 hours). An exact Earth copy orbiting Alpha Centauri B would be just barely detectable, but a confident detection of Proxima Centauri b would be possible. For optical reflected light, a detection would be more challenging. However, one advantage is that, with an M dwarf's many spectral lines, cross-correlation can result in a gain of 65 to 80 in signal-to-noise ratio. Even so, Proxima Centauri b's reflected light would only be detectable with starlight suppression via extreme adaptive optics. In that case, a 10-hour observation would suffice. Brogi then gave a caveat that this is based on a scaled version of Earth and does not take into account trying to retrieve the planet's properties. However, he has a plan to make this feasible for the next generation of extremely large telescopes.

Brogi then emphasized that Proxima Centauri b is just a prototype. While the habitability of planets orbiting M dwarfs is in question, he said that Proxima Centauri b is the best near-term chance to allow us to test our observational skills and characterize a potentially habitable world. He then concluded by saying that this groundbased, high-resolution technique is capable of getting the masses, inclinations, rotations, and wind speeds for nontransiting hot Jupiters already, but if combined with high spatial resolution, it could allow the same thing for potentially habitable planets in the future.

#### **Audience Participation**

An audience member asked how many M dwarfs the next generation of extremely large telescopes will be able to survey. Brogi answered at least 10. The Giant Magellan Telescope—the smallest of the three planned, extremely large, ground-based telescopes at 25 m—is his favorite because it will have high-resolution spectroscopic capabilities from the start. One issue, however, is how much telescope time will be available for these observations. If the noise is not Gaussian, then it becomes more difficult. He said that 50 to 100 hours of observational time would be a reasonable time investment.

Another audience member then asked about the difficulty of using ground-based telescopes to look for biosignatures due to contamination by Earth's atmosphere. Brogi replied that the Doppler shift of the planet will help distinguish it from Earth's atmosphere, both in terms of where the lines appear and how the lines change due to the planet's orbit. He then said that, at a resolution of 100,000, Earth's atmosphere will not prevent you from detecting these features.

# GENERAL DISCUSSION: PRACTICAL BIOSIGNATURES THAT CAN BE EXPLOITED TO SEARCH FOR LIFE IN SITU IN THE SOLAR SYSTEM AND FROM AFAR ON EXTRASOLAR WORLDS

The moderator for the general discussion on practical biosignatures both in the solar system and for exoplanets was Gary Blackwood from JPL and manager of NASA's Exoplanet Exploration Program. The second question in the workshop's statement of task (see Appendix C) was the focus of this discussion: Are we today positioned to design, build and conduct experiments or observations capable of life detection remotely or in situ in our own solar system and from afar on extrasolar planets? Blackwood then informally polled the audience on their opinion on the answer to that question. There was a mix of yes, no, and unsure. Blackwood then listed five topics to guide the discussion.<sup>61</sup>

# What's Changed Since 2000?

In 2000, the National Research Council released a workshop report called *Signs of Life: A Report Based on the April 2000 Workshop on Life Detection Techniques*.<sup>62</sup> Blackwood asked what is new since then and what has changed in technology, scientific discoveries, and understanding. He then opened the floor for discussion.

<sup>&</sup>lt;sup>61</sup> The text in this section is not necessarily in chronological order. Comments have been moved out of chronological order to improve flow and preserve continuity of thought.

<sup>&</sup>lt;sup>62</sup> National Research Council, Signs of Life: A Report Based on the April 2000 Workshop on Life Detection Techniques, The National Academies Press, Washington, D.C., 2002.

Starting with Earth biology, an audience member said that Robert Hazen has articulately advocated that our understanding of mineral complexity on Earth is largely attributable primarily to life (but also to liquid water). She asked how we should use this to interpret results from other bodies in our solar system and even exoplanets. She brought up the idea of geobiology and emphasized that life is a planetary process.

Another workshop participant then said that more than one example of oxygenic photosynthesis is now known. She said that at longer wavelengths, life co-evolved with its environment. Another person then said that new developments have been made in studying the terrestrial biosphere, such as the extent of the deep biosphere and the ability to detect microbes there and differentiate between active and dormant biomass.

A new discovery since 2000, an audience member said, is the Lost City, a field of hydrothermal vents in the middle of the Atlantic Ocean. This has led to the development of a robust model for life emerging at one of these alkaline, hydrothermal systems.

A member of the audience then said that we now understand what chemical features a universal genetic molecule would have: a one-dimensional biopolymer with a backbone of repeating charges. He then said that work done in the laboratory has shown that RNA was likely the first molecule on Earth to gain access to Darwinism. Changing topics to Mars, he said that we now know that the surface of Mars is not self-sterilizing. Earlier notions to the contrary were attributable to misinterpretations of the 1976 Viking result.

Sticking with the subject of Mars, an audience member said that the Mars rover missions, particularly the Curiosity rover in Gale Crater, have discovered long-lived, aqueous environments. He said that this demonstrated not a biosignature, but the ability of sedimentary rock to preserve organic matter over a long period of time. Another participant followed up saying that there is now lots of evidence for liquid water processes on Mars that were unknown in 2000, such as recurring slope lineae from the Phoenix mission and the High Resolution Imaging Science Experiment (HiRISE) on the Mars Reconnaissance Orbiter (see Figure 3.5). She said that she thinks that the case for modern life on Mars is growing but has been ignored.

A new technological improvement, another participant said, was the ability to perform high-precision measurements of isotopes in molecules on Mars using both Curiosity and the Mars Atmosphere and Volatile EvolutioN (MAVEN) orbiter. This has given us a window into Mars' past in terms of its atmospheric pressure and how it evolved over time. Another innovation, he said, has been the tunable laser spectrometer. The ExoMars Trace Gas Orbiter should be able to do even better for trace gases. Another member of the audience agreed. She said that two classes of measurements have been miniaturized for Mars and can now be used for other bodies in the solar system: the ability to precisely measure stable isotopes and the ability to do precision chemical and mineralogical analysis at the micro scale. She noted that the rovers and orbiters have discovered an amazing diversity on the surface of Mars.

A member of the audience then gave a plug for sample return since 2000. He said that we as a community have returned material from comets and are on our way to an asteroid. There are plans to do the same with Mars. The possibility also exists for a sample return from a plume of Enceladus.

Changing the topic to Venus, another participant said that life could exist in the clouds of Venus. He lamented the focus on the habitable zone as referring to only the surface temperature and not considering habitable temperatures elsewhere in the atmosphere. Since 2000, there has been discussion of exploring Venus's clouds using an unmanned aerial vehicle, which might be possible in the next decade. He said that, although there haven't been any new scientific discoveries, life in the Venusian clouds is reasonable considering that the properties of bacteria on Earth (chemical composition, spectral properties, and size distribution) are similar to the cloud particles on Venus. He suggested we might actually be observing bacteria. He then brought up the UV absorber in Venus's atmosphere, whose origin remains unknown after 50 years. Bacteria, he thought, could be its origin too, and he asked the community to consider this idea.

Moving to the outer solar system, another workshop participant brought up Cassini's discoveries on Enceladus. We now know that it has a liquid water ocean, and there is compelling evidence for hydrothermal vents as well. She then brought up the discoveries on Titan and the possibility of weird life. More generally, she said that community interest in ocean worlds as interesting targets has grown. Another audience member then mentioned the possibility of plate tectonics on the Europan ice shell and progress on researching different types of ice phases.



FIGURE 3.5 Recurring slope lineae (dark streaks in the lower third of the image) from running liquid water on the surface. This is a reprojected image with original data taken by the High Resolution Imaging Science Experiment (HiRISE) camera on NASA's Mars Reconnaissance Orbiter. SOURCE: NASA/JPL/University of Arizona; see HiRISE Operations Center, 2011, "Slope Features on Wall in Newton Crater," https://www.uahirise.org// ESP\_022689\_1380.

Going now beyond the solar system, a member of the audience talked about the discoveries from the Kepler space telescope. She said that it shows that terrestrial exoplanets are probably common, mentioning Proxima Centauri b as an example. Another major thing, she said, was the evolution of our understanding of false positives when it comes to biosignatures. This has removed the idea of  $O_2$  being an easy, straightforward biosignature. Therefore, we now know that we must have an understanding of the entire environmental context and of life as a planetary process. Continuing on this topic, another audience member referred to a debate on martian magnetofossils, which helped us become more skeptical and focus more on potential false positives.

In the final comment about this topic, an audience member talked about what has not yet happened. There is still no magical, Star Trek tricorder. All of our progress has been based on a priori knowledge of our biochemistry that isn't going to be known for life beyond Earth.

#### In Situ and Remote Sensing

Blackwood then moved to the topic of doing in situ and remote sensing of life. He asked what we should search for and why, what processes we should use, and how we could improve the robustness of detection and subsequent interpretation.

An audience member began the discussion by talking about how multiple methods should be used. For example, studies in Antarctica, the Atacama Desert, and the Mojave Desert have revealed hidden microbes, sometimes under just a millimeter of rock. She thought that we should try to identify spectral signatures from not just orbiting satellites, but maybe aircraft and rovers as well.

A member of the audience then mentioned the topic of clumped isotopes, thinking that it would be interesting to do an analysis to learn the sources of the hydrogen and methane on Mars. Going back to the previous discussion, he said that a big discovery since 2000 is life in deep sediments that could have generation times of millions of years. He then asked how we would account for and measure it if it were on Mars. Another person agreed, saying that our frame of reference is one of high energy, but our targets could have very low energy. This should cause us to shift the way we think about biosignatures.

Another participant in the workshop said that, for Mars, the mantra of "follow the water" has been very successful. Expanding this idea to exoplanets, he said that a planet the size of Earth in the habitable zone of a G-dwarf star might not be adequate because it might not be able to get enough water (or, at least, enough water to be detectable). Coming back to Mars, he said that we still do not have evidence of an aquifer on Mars. He thought that we should extend our search for water on Mars deeper below the surface.

A conference participant then said that Mars isn't just telling us to "follow the water," but to "follow the *salty* water." He thinks that the same is probably true for Europa. Therefore, we should focus on biosignatures that can be preserved in salty places. As a brief aside, he also said that a search for extraterrestrial intelligence would be a great place to start doing remote sensing.

Following up on the "follow the water" mantra, another audience member said to "follow the carbon." He said we have been doing this, but not enough has been found to get excited about it. Therefore, we should target the sediments mostly likely to reveal organics with Curiosity's Sample Analysis at Mars (SAM) instrument. In 2020, he said, we will have better remote detection capabilities of organics. He said that if we keep discovering water over and over again, people will eventually catch on.

Again emphasizing the need to look below the surface of Mars, another person commented that we should develop better subsurface exploration techniques using either direct or indirect observations. He said that this was also applicable to Europa. He then moved to exoplanets. He said that, with the Transiting Exoplanet Survey Satellite (TESS), we could have 10 to 50 exoplanets that we could follow up on, but only the time for maybe one or two of them. Because choosing which one(s) could be tricky, he said, we should develop models to predict a planet's environment from only the planet's mass, radius, and host star.

Then a member of the audience said that, while he was excited about directly imaging exoplanets, we should start thinking about instruments and observational techniques to complement direct spectroscopy. Both the planets and the stars, he said, will need to be better characterized in terms of mass and diameter. He gave Kepler as an example, the proposal for which included follow-up observations of ground-based radiovelocity detection for any discoveries made by Kepler itself.

Another member of the audience emphasized the idea of understanding the context of any discoveries. Multiple lines of evidence are important. Another workshop participant then said that the same thing applies to exoplanets, which means having a well-characterized host star.

Referencing Europa, Enceladus, and other outer bodies, another participant then said that we might not have the luxury of multiple mission campaigns to search for life. For these objects, the audience member said that we need to speed up the mission cadence and make careful predictions, so that follow-up missions in the works can handle whatever new discoveries were made in the meantime.

A member of the audience then said that we need more instruments that can do liquid-based analysis, especially if we are going to places looking for water-soluble organic molecules.

# Solar System and Extrasolar Worlds

Blackwood then moved on to the third topic and asked, What is common to both scenarios in regards to the remote or in situ detection of biosignatures?

A workshop participant then said that the questions of what is practical for remote detection and in situ detection are very different. He was confident that any form of life could be detected in situ, even if it did not use DNA as its genetic material.

A WebEx viewer then posed a question about whether Juno could fly through Europa's plumes to try to detect signs of life. An audience member then confirmed that Juno would not be capable of that. It does have a UV spectrometer, but it cannot do the observations in the way that would be needed.

Sticking to the topic of gas giants, a member of the audience then said that he was excited about the elemental abundances of giant planets, and not just hot Jupiters, because we are getting to the point where we can find colder gas giants. Comparison between the elemental abundances between Jupiter and Saturn versus extrasolar giant planets, effectively comparative planetology, could be interesting.

Another member of the audience was struck about the idea of fingerprinting a world. Looking at Venus, Earth, and Mars, she wasn't sure what the lessons were. She wondered what properties of planets really control the composition of the atmosphere and enable a stable redox state through time. The level of coupling between the planet's interior evolution and the nature of degassing, volcanism, and magnetic fields are not understood, she thought.

Continuing on that subject, a workshop participant said that Venus, Earth, and Mars all had liquid water in the past. Venus could have had liquid water for 2 billion years. He said that Venus could still have life in the clouds. Many exoplanets could also be Venus-like. For an in situ detection, he said that an airplane filled with hydrogen or helium could survive up to a year in Venus's clouds. For remote detection, practical techniques could include doing isotopic measurements and looking for disequilibrium using Raman LIDAR. Since no planetary protection precautions for Venus are required per the Committee on Space Research (COSPAR), he said that it would be easy.

Referring back to earlier discussions, an audience member said that "follow the water" is not enough on Mars anymore. He said that we now need to determine which of the sites with water are the best. Similarly, he asked which exoplanets make the best targets. He then wondered what properties of a planetary system could give a sense of the composition of the planets.

A member of the audience wanted to tie "follow the water" together with Venus, Earth, and Mars using observations to see what they all have in common. One such thing is patterns in circulation, and we have general (or global) circulation models (GCMs). 3D GCMs can explore the habitability of a wide range of planets.

Another participant at the workshop emphasized the importance of the planetary interior. He said that processes like plate tectonics and volcanism are not linear or predictable but can be addressed in a probabilistic way depending on things like the planet's mass, composition, evolution, volcanism, and magnetic fields. In this way, we could potentially see what sort of features make certain processes more likely, such as hydrogen outgassing, oxidizing outgassing, dynamo generation, or plate tectonics. In this framework, Venus would be an important data point. A mission to Venus to learn its interior properties, like its rheology, water distribution, seismology, or dynamo generation, would be useful.

#### Are We Ready?

Blackwood then asked if we are ready today to engineer and observe life detection remotely or in situ for either the solar system or exoplanets.

Starting with in situ detection, a member of the audience said that we were technologically ready. However, we were not ready to deal with the environmental context of the detection. Life might have a low signal-to-noise ratio, and the environment could produce a lot of measurement noise.

Another member of the audience then took exception to the phrasing of the question. He said that we are capable of designing a mission to go to an aqueous environment to detect life. However, in situ detection of extinct life is more difficult. It is uncertain whether or not that is possible today. Continuing on, he said that designing a mission to do remote detection of life using just spectroscopy is impossible right now. Another audience member also criticized the phrasing of the question. She said that many instruments could detect something that we interpret as life. However, if we go to a place beyond Earth, which may or may not have Earth-like life, we might not know whether or not we have found life. Many assumptions go into any interpretation.

After Blackwood asked for someone on the side of "no," a workshop participant came out firmly on the negative side. She touched on previous themes and said that detecting life, especially very-low-biomass life, is non-trivial to do in real environments (whereas in a laboratory, it could be easy). Another aspect is the time dimension. Organisms in an environment that don't do much could have a chemical signal. However, we might not be sure if it indicates life, since our assumptions are based on fast microbes. She said that the planet, the crust, and, to some extent, even the oceans and the ocean's sediments are dominated by slow microbes.

Another conference participant said that many people believe that the best places to find life beyond Earth are in completely inaccessible areas, such as deep under the martian surface or below the crust of an icy world. Even sampling a plume is nontrivial. She then said that people never really agree on an unambiguous biosignature. The interpretation will be very difficult.

A participant at the workshop then said that interpretation might not be a black and white issue. That being the case, an instrument relevant to detecting a biosignature should at least give a better characterization of the environment. This would then lead to approaching the issue better for the next mission.

Moving to the exoplanet side, a member of the audience said that we were definitely not ready for in situ detection of life on exoplanets. For remote detection, obtaining a secondary biosignature to constrain the fluxes of the primary biosignature is very difficult, such as methane for oxygen. However, he said, it is completely inadequate compared to Mars, which has an unexplained, variable amount of methane in its atmosphere. In response to this, he stressed the idea of using population statistics in terms of atmospheric detections in order to rule out certain scenarios. This could lead to results showing either life on several worlds or highlighting a profound ignorance of geology and geochemistry.

#### What Can We Learn from One Another?

Blackwood then moved on to the fifth and final topic of the discussion. He asked what we could learn about how to perform in situ and remote sensing of life or potential biosignatures both in the solar system and beyond. He also wanted to know what we could learn from people in other disciplines, such as planetary science, astronomy, biology, geology, oceanography, geochemistry, and others.

One member of the audience said that we have no choice but to learn from people in other disciplines. Otherwise, nobody in the scientific community will believe any claimed detection. Another workshop participant agreed, saying that, no matter how hard one tries, it is very difficult to be an expert in more than one field. Therefore, she thought that we needed to help each other out, particularly in avoiding false positives and false negatives.

Then a workshop participant said that one discipline that is really needed is statistics. The geological context, he said, is all statistical priors, not estimates on the probability that we have detected life. These priors must be understood. He also said that there is an entire industry devoted to designing experiments to optimize the outcome when your knowledge and your priors are uncertain: the clinical trials industry. These people might be useful when designing missions.

Two more disciplines were then added to the list by another audience member. The first was glaciology, since these are clearly visible from space. Glaciers could help concentrate organics or create drainage patterns. The other discipline she suggested would be useful is climatology. Climate change is one example of how life can alter the climate. For example, in addition to just global warming, oceans are rising and becoming warmer and more acidic.

A member of the audience then brought up work he had done that concluded that we should search for evidence of evolution. One thing he said we could learn from one another is how an evolving community of organisms changes the environment. Earth has probably had many previous versions of itself, but we do not understand them. He would like people to understand life as a planetary process and how life has been involved in planetary evolution.

A team member for both the Habitable Exoplanet Imaging Mission (HabEx) and the Large UV/Optical/ Infrared Surveyor (LUVOIR) then extended an invitation for people to communicate with them on what kind of observations they would like to make.

Another audience member brought up the idea of comparative planetology again. There is Venus, Earth, and Mars, but also Mercury and the Moon. He thought we needed to understand how their evolution was different and why they are so different today. Without knowing at least how Venus, Earth, and Mars are different, understanding exoplanets will not be possible.

To finish out the discussion, a member of the audience then addressed how in situ and remote detection could play off one another. For example, the detection of methane is a potential biosignature. In situ detection by the *Curiosity* rover, remote detection by Mars orbiter missions, and remote detection from Earth could answer the question of whether it truly is a biosignature. Something she thought was critical was using a combination of both modeling and laboratory work. The biggest thing that irked her was that we don't know the effect that radiation has on the organic molecules on the martian surface, which she thought needs to be simulated.

# 4

# Life Detection Techniques

John Baross of the University of Washington and Gary Ruvkun of Harvard Medical School were the moderators for the session on life detection techniques: Where we are today and what to look for, in terms of life as we know it and life as we don't know it?

# LIFE DETECTION: 40 YEARS AFTER VIKING

Ben Clark of the Space Science Institute started his talk with an image of a Viking lander with Carl Sagan beside it. The Viking landers were the first and last spacecraft to be subjected to dry heat sterilization. Both were heated to a temperature higher than boiling water. Heat sterilization can now be done cheaper today, which might be necessary for future astrobiological missions. A third Viking lander had been completely built with flight hardware, but eventually, the proposed follow-on mission, which included tracks in place of footpads to transform it into a rover, was dropped because of the high costs of developing the Space Shuttle Program.

# Viking Results

The Viking missions had a gas chromatograph-mass spectrometer (GCMS) to look for organics using hydrogen carrier gas. Initially having four planned biological experiments on Viking, it was reduced to three after serious budget overruns on the overall instrument. The final three selected were very general and simple experiments: pyrolytic release, labeled release, and gas exchange. Despite the tests being simple to perform in the laboratory, they were difficult to implement: all three were packed into a space of about 1 cubic foot. Due to the engineering challenges, Clark said that it was the most expensive instrument that had ever been developed for spaceflight.

The pyrolytic release exposed the soil to a solar simulator light source and radiocarbon monoxide and dioxide and looked for incorporation into complex organics, which would indicate the presence of life. The labeled release provided some common organic substrates used by biology and looked for the conversion of these to any kind of gas, such as  $CO_2$ . The gas exchange used what was called "chicken soup," a mix of everything that they thought life might need or be able to utilize. It then looked for changes in gas concentration. Each experiment had different modes to test in, which together spanned dry, moist, and wet conditions.

The pyrolytic release experiment, Clark said, had an overall negative result for life, although there was one data point that was anomalously positive. The labeled release experiment, on the other hand, did get indications of a

positive result. The result for the gas exchange experiment was negative, but a release of oxygen after humidifying the soil was observed, which indicated the presence of oxidants—a big surprise to the team. It also raised major doubts as to whether the labeled release results were just oxidants undergoing inorganic chemical reactions or if the result was truly of biological origin. An abiological way to simulate those results using oxidants was quickly discovered.<sup>1</sup> The GCMS gas-exchange experiment found no organic molecules with an upper limit of <1 parts per billion (ppb).

Clark said that there was and still is some uncertainty about what was actually found on Mars with the Viking landers. There is the one anomalous data point in the pyrolytic release experiment. In the labeled release experiment, the injection of nutrients in an aqueous solution onto the soil began a rapid evolution of the radioactive gas, which then plateaued as if the nutrient were being used up.<sup>2</sup> This was compared against a control sample of soil that was baked at 160°C for 3 hours, which yielded a null result. When they injected more aqueous nutrients into the soil about 8 days after the initial injection, they would have expected to see life metabolize that injection as well. However, the signal counterintuitively dropped (see Figure 4.1). This divided the community into two camps: those who thought the experiment showed biological activity and those who thought it was all abiotic. The abiotic camp thought that the first injection may have just been oxidants in the soil that were oxidizing the nutrients, specifically formate, while the biological camp said it was life metabolizing it. In the second injection, the data were attributed to uptake by minerals in the wet soil due to temperature variations by the abiotic side, while the biological side suggested it could be a circadian rhythm. The control sample, which was heated to 160°C for 3 hours, was used as proof by the biological camp, but the abiotic side said that the oxidants could be heat labile, that



FIGURE 4.1 Viking Lander 2's labeled release experiment exhibited radioactive uptake after an injection of nutrients, indicating life. A second nutrient release caused a drop, however, rendering the experimental results highly ambiguous. SOURCE: From G. Levin and P.A. Straat, 1976, Viking labeled release biology experiment: Interim results, *Science* 194:1322-1328, reprinted with permission from AAAS; presented in Ben Clark, Space Science Institute, "Life Detection: 40 Years After Viking," presentation to the Workshop on Searching for Life across Space and Time, December 6, 2016.

<sup>&</sup>lt;sup>1</sup> C. Ponnamperuma, A. Shimoyama, M. Yamada, T. Hobo, and R. Pal, 1977, Possible surface reactions on Mars: Implications for Viking biology results, *Science* 197:455.

<sup>&</sup>lt;sup>2</sup> G. Levin and P.A. Straat, 1976, Viking labeled release biology experiment: Interim results, *Science* 194:1322-1328.

#### LIFE DETECTION TECHNIQUES

there could be multiple oxidants, or that the water released by heating the control sample could have destroyed the oxidants. The Curiosity rover, Clark noted, has also seen significant water release from samples heated to 160°C.

Clark said that two of Viking's major accomplishments were that it was the first in situ, robotic mission looking for biomarkers on an extraterrestrial body and that it was also the first search for metabolic activity. No subsequent search for metabolic activity has yet been attempted. Because there are oxidants in the soil, any organics could be reacting with them during the pyrolytic step used to volatilize organics. Curiosity's Sample Analysis at Mars (SAM) instrument is making corrections for this. The best idea to avoid this problem, according to Clark, is to use laser desorption mass spectroscopy, which will fly on the ExoMars rover mission (launch date 2020), led by the European Space Agency (ESA) and the Russian space agency, Roscosmos.

On the labeled release results, Clark said that several people tried simulating the results with different oxidants in the soils and achieved varying degrees of reproducibility. The most successful result so far, according to Clark, has been Quinn et al. (2013).<sup>3</sup> Their experiment used perchlorates, which could not be measured by Viking but were discovered by the Phoenix mission. When irradiated by simulated cosmic rays, perchlorates transform into hypochlorite and trapped oxygen, which can then mimic the Viking results.

There were several things the Viking missions did not explore either. They did not provide all the possible metabolites in their biological experiments, such as  $H_2$ ,  $H_2S$ , NO, or NO<sub>2</sub>. They analyzed only soil, and they did not test inside rocks or salts. The GCMS could not detect a biomass density of <1,000 microbes/cm<sup>3</sup>. Clark thinks that only (cold) sample return will let us determine whether or not martian material contains life or the signs of extinct life. The third, unused Viking lander, he said, was also considered for use in a sample return mission. Sample return is more feasible nowadays. However, as Clark said, the saying in their community is that a "Mars sample return is always 10 years away," consistent with the 2026 launch date for the sample return envisioned by Dr. Stofan in a previous talk (see Chapter 2).

Clark then noted that many comments have been made lamenting that the Viking missions were not able to benefit from today's knowledge of Mars during its mission design phase. However, Clark said that there actually was much known about Mars at the time and that some of the experiments would still be valid today. He then used the National Research Council report *Biology and the Exploration of Mars*<sup>4</sup> (1966) to show what was already known about Mars, such as its incident ultraviolet (UV) flux, low temperature, and dry air, as well as the existence of extremophile organisms on Earth. Clark then cited a statement in the report that a negative result for organic molecules would preclude the existence of biology, which presumably biased the entire endeavor because no organics were measured by the GCMS.

Clark then said that Viking taught us new things about the martian soil. For one, they found high levels of iron in the soil. That in itself was not surprising, since Mars is red. However, they also found that levels of sulfur (in sulfates) and chlorine (in chlorides) were approximately 100 times higher than would normally be expected for soil on Earth, Mars, or the Moon. The Phoenix lander also found the oxidants perchlorate and chlorate in addition to the chlorides. It was also discovered that the soil at the two Viking sites, on opposite sides of the planets, were virtually identical. He then said that there is so much sulfur and chlorine salt in the soils that if you take a regolith, fill the porous space with ice (which happens on Mars), and then melt it, the result would be salt concentrations at Dead Sea levels. The magnesium sulfate salt is different from Earth's NaCl salt, but it still has a high ionic strength. This would require organisms to have a high salt tolerance. Clark is collaborating with biologists who have taken salt-tolerant organisms and exposed them to perchlorates. They have found that alkali perchlorates are tolerated better than alkaline earth perchlorates. However, Mars seems to have alkaline earth magnesium and calcium perchlorates. This might not be relevant though, considering terrestrial microbes have no evolutionary reason to tolerate perchlorates.

# **Possible Life on Mars Today**

Clark then switched to whether or not they now think that Mars has extant life. He said that, with the information they now have, they think it's even more likely than they originally thought back during the Viking mis-

<sup>&</sup>lt;sup>3</sup> R.C. Quinn, H.F.H. Martucci, S.R. Miller, C.E. Bryson, F.J. Grunthaner, and P.J. Grunthaner, 2013, Perchlorate radiolysis on Mars and the origin of martian soil reactivity, *Astrobiology* 13:515.

<sup>&</sup>lt;sup>4</sup> National Research Council, Biology and the Exploration of Mars, National Academy Press, Washington, D.C., 1966.

sions. One bit of new information is that near-surface ice has been discovered at the Phoenix landing site. It was also discovered that Mars has a wide range of obliquity (tilt of the spin axis with respect to orbital plane), which allows for cyclic climate change. It is now in a low obliquity era, meaning that it is in a cold spell. The team actually saw frost in the wintertime at the Viking 2 landing site, which was a surprise. The Shallow Subsurface Radar (SHARAD) instrument on NASA's Mars Reconnaissance Orbiter (MRO) recently discovered a huge quantity of shallow permafrost ice in the Utopia Planitia region where Viking 2 landed.

Conditions on Mars during the Noachian era (~3.7 to 4.1 Gyr ago) were favorable for the origin of life, according to Clark. It had liquid water and a dense atmosphere. Mars also apparently had a significant amount of  $H_2$ , and possibly CO and  $CH_4$ , in the atmosphere to provide energy for life. He then referred to a claim that 99 percent of organisms can metabolically use  $H_2$ . Sulfur, which is found everywhere on Mars in the form of sulfate, could also have been a source of energy for chemoautotrophic life by combining with  $H_2$ . It is even possible to have photosynthesis using sulfur-bearing instead of oxygen-bearing molecules. Using  $H_2S$  instead of  $H_2O$  was actually the first form of photosynthesis on Earth, he said. Mars may also have had organic molecules. Additionally, the planet received enough sunlight (43 percent that of Earth's) to easily allow for photosynthesis. Both iron and manganese can be used as electron acceptors, and both are found on Mars. More broadly, Mars has all the CHNOPS (carbon, hydrogen, nitrogen, oxygen, phosphorus, and sulfur) elements plus other key elements (Fe, S, Ni, Zn, Cu, Mn, and P) necessary for life at concentration levels, which suggest prior interactions with liquid water. Hydrothermal activity on Mars was also present, Clark said.

He then reminded the audience that terrestrial life often goes dormant for long periods of time in "survival mode." Clark suggested that life on Mars could be doing the same thing between obliquity cycles. He also said that abiotic photochemistry on Mars could provide reactants to drive chemoautotrophy without photosynthesis. To search for this life, Clark said that you could go vertically down to perhaps subsurface permafrost ice or even a deep liquid groundwater layer. He also said that you could search horizontally, such as looking in salts for salt-tolerant organisms. There are potentially even caves or lava tubes that could support life.

Clark then finished with lessons learned from the Viking missions. Experiments that are simple in the laboratory can be difficult to implement and expensive to fly, and they might have perplexing results. The environmental context is also important. He then said that understanding Mars requires a sample return mission. Lastly, he said that the Viking mantra of "if life is anywhere, life is everywhere" on Mars may be wrong. It is possible that life only exists in certain regions or environments on Mars.

#### **Audience Participation**

A member of the audience said that it isn't fair to second guess the Viking experiments, saying that it was one of the great accomplishments of civilization. He did, however, say that a big problem with Viking is that it never detected any of the organics that, at the time, might have been thought to come from meteorites. In a paper he published 20 years ago, the audience member showed that with a rebuilt instrument, if they had been sitting on top of partially oxidized meteoritic organics, they would not have seen them either. He then said that the two camps, the biological and the abiological, are equal, and it really depends on which result they thought was the extraordinary one that needed an extraordinary explanation. He then asked why there hasn't been a more sophisticated metabolic experiment. Clark agreed with the need for new metabolic experiments on Mars. One hindrance has been planetary protection, but he said that heat sterilization would now only be needed for the sample acquisition hardware in some cases, not the entire spacecraft. He also repeated that it can be done cheaper today.

An audience member then rebutted the claim that Viking did not detect organics, clarifying that it found chloromethanes. Clark said that it was just the cleaning solvent. The audience member responded by saying that if you heat soil with organics in the presence of perchlorates, chloromethanes are seen. They never found them in the blank samples either. Clark said that he was open to that interpretation, but was presenting the Viking results as they were known at the time. The conclusion by the Viking team was that cleaning solvent was used, although its use was never officially documented or confirmed.

Commenting on the frost found by Viking, a participant asked about the role of deliquescence (the process by which a substance absorbs moisture from the atmosphere until it dissolves in it) in terms of potential habitability

#### LIFE DETECTION TECHNIQUES

on Mars. Clark said that deliquescence was actually predicted. He then said that a deliquescent salt effectively competes against an organism for water. Deliquescence often results in a saturated solution with a water activity below 0.6, which is the most extreme limit known for life. On the other hand, Clark said, maybe deliquescence can attract water even after it becomes saturated, which keeps water nearby that could be used by living organisms. Maybe, he said, life could even outcompete deliquescent salts or they could live in lower water activities than terrestrial life. The same audience member then said that, according to her understanding, the water activity for perchlorate would be too low at <0.6, but for sodium chloride, it would be okay. She noted that this is the habitat found in the Atacama Desert. Clark answered that perchlorates depress the freezing point to about  $-50^{\circ}$ C, whereas sodium chloride only depresses the freezing point to  $-23^{\circ}$ C. At that temperature on Mars, he said, the humidity is still much less than 100 percent. However, a change in obliquity toward warmer conditions would increase the water vapor pressure on Mars, which could make all of these processes easier.

Another audience member mentioned again the likelihood of organic matter from meteoritic impacts, or maybe an igneous, hydrothermal type of abiotic organic matter, which should be all over Mars. Biological organics might also contribute to the organic content on Mars. She said that the Viking experiment would not have broken down all organic macromolecules, because the samples were only heated to 500°C. Therefore, organic macromolecules would not have been detected by Viking. She then said that the release of oxygen complicates the analysis. However, it might be helpful because that means it could combust with the organics allowing for its detection. Clark said that the martian surface should contain organic materials approaching the 1,000 parts per million (ppm) level just from meteorites, judging by both the nickel content of the soil and the impact rates. Therefore, much of it seems to have been oxidized, converted, or degraded to result in the low levels seen today.

A workshop participant asked what the salts (iron sulfates, magnesium sulfates, chlorides, and bromides) found in the Gusev Crater imply about Mars. She said that they have sometimes been interpreted as showing modern mobility of fluids in the top meter of soil. Clark said that the Spirit rover found material that is a mixture of ferric sulfate, magnesium sulfate, and a silica-independent phase at multiple elevations. He has no clue how water could have distributed them this way. Clark also doesn't know why they haven't found more concentrated occurrences of chlorine, especially since perchlorates significantly depress the freezing point of water, whereas sulfates do not. This means that the perchlorates and chlorides should be mobile and become highly concentrated, while sulfates should remain more diffuse. However, the opposite is seen. Bromine, he said, is extremely erratic and could be mobilized just by frost, since bromine is also a powerful freezing point depressor.

The last question to Clark was whether he thought that Wolf Vishniac's "Wolf Trap" should have gone to Mars. The Wolf Trap was designed to place martian dust into a tube containing nutrients in liquid form. Properties such as the pH and turbidity would then be monitored for signs indicating life. However, we now know that martian dust in the atmosphere is in the 3 to 4 micron class, much finer than Earth's dust. Therefore, suspended dust particles would have confounded the experiment. The question was subsequently addressed when the pH of soil was later measured directly by Phoenix. Vishniac did, however, make an enormous contribution to the thought processes behind the search for life on Mars.

# LOOKING FOR LIFE AS WE KNOW IT ON OTHER PLANETS

Gary Ruvkun of Massachusetts General Hospital began his talk by showing the tree of life rooted with the universal common ancestor (see Figure 4.2). How this tree was initially constructed, he said, started with trying to isolate common molecules from cells, which began in the pre-DNA era. The ribosome turned out to be one of the most abundant organelles in a cell. Ribosomes are made out of many different proteins and a few different RNAs. The RNAs are either approximately 1,500 or approximately 2,900 nucleotides long. The ribosome is where a piece of RNA made from the genome is decoded three nucleotides at a time to assemble proteins three nucleotides at a time. Ruvkun called the ribosome a living fossil of the RNA world. These molecules were easy to pull out and perform RNA sequencing on, which could be used to infer relationships between different organisms. He likened the process to the way linguists phylogenetically classify languages and the relationships between them to infer how languages evolved from earlier languages.

SEARCHING FOR LIFE ACROSS SPACE AND TIME



FIGURE 4.2 The "tree of life," rooted at the last universal common ancestor for all life, with Eukarya contained with the domain of Archaea. SOURCE: Courtesy of NASA Astrobiology Institute.

# The History and Current State of Genomic Sequencing

Ruvkun then discussed the modern day genomic landscape. He showed a figure of human ribosomal RNA compared to worm RNA (see Figure 4.3). It showed many regions in the RNA that were identical, justifying the structure of the tree of life (e.g., the animal kingdom on the tree of life is just one small twig). Doing the same analysis between human and archaeal ribosomal RNA, the similarities aren't as strong, but there are still significant stretches where nothing has changed over the last 3 billion years (Gyr) or so. Ruvkun said that evolution had already perfected this part of the RNA. He then said that ribosomal RNA is the most conserved genetic material in life.

Ruvkun then briefly went through the history of genomics. Ribosomal RNA sequencing began in the early 1970s. The discovery of the Archaean branch of the tree of life occurred then. In 1973, recombinant DNA allowed genes to be created one at a time. Gene sequencing was invented in 1976, allowing for a much faster discovery rate (e.g., approximately 3,000 base pairs of DNA in a typical paper). In the 1990s, this process sped up dramatically with the help of machines, during which time the first organism's genome, a bacterial genome, was sequenced. In 1997, the first full animal genome was sequenced, a nematode with  $10^8$  base pairs. In 2001, the human genome was sequences (~ $10^{11}$  base pairs) per year at about \$100 per genome, compared to about \$100 million per genome in 2001 (see Figure 4.4). He said that, although only a small portion of biology has been sequenced, it is a highly diverse portion that allows for the network of relations to be analyzed.

The main machine used for DNA sequencing nowadays is the Ilumina, of which there are about 7,500 in the world. A new genome-sequencing machine by Oxford Nanopore was introduced in 2015. Rather than the 500-pound, power-demanding Illumina machines, the Oxford Nanopore machine has a mass of just 87 grams and can be run on a smartphone. Ruvkun said that there are concerns about its accuracy, but its small size and low power requirement can make it a major asset in genome sequencing. There are already about 1,000 users of it.

There are now about 3,500 eukaryotes with their genomes sequenced, each one having  $5 \times 10^6$  to  $10^{10}$  base pairs. Typical animals have  $10^8$  to  $3 \times 10^9$  base pairs. Larger animals' genomes are usually packed with what is sometimes called "junk DNA," such as the carcasses of viruses. Out of ~25,000 genes in animals, ~10,000 of them are shared between all the different animal species. There are also more than 12,000 bacterial genomes sequenced

#### LIFE DETECTION TECHNIQUES

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FIGURE 4.3 Genetic similarity between worm and human rRNA. SOURCE: Gary Ruvkun, "Looking for Life as We Know It on Other Planets," presentation to the Workshop on Searching for Life across Space and Time, December 6, 2016.

( $\sim 4 \times 10^6$  base pairs each) and  $\sim 700$  archaeal genomes sequenced. A typical bacterium has  $\sim 4,000$  genes. A few hundred genes are universal in all extant life. As an example, he showed a 400 amino acid protein from an animal that encodes a proteasome subunit, a part of an organelle that degrades proteins, and compared it to its counterpart in an archaea. The two were separated for 3 billion years, yet there are still strong similarities. Ruvkun said that it is one of the most conserved proteins in evolution. He then said that there are about 400 genes that are similarly conserved which form the core of biology and were likely present in the last common ancestor of all life on Earth.

#### **Ancient Life and Panspermia**

A major problem in the study of evolution, especially early evolution, is determining the time that different branches split off from one another. In the last 500 million years, fossils provide a method to date some branches, but mostly just in the animal kingdom. Fossils from other branches in the phylogenic tree, especially in other domains, are difficult or even impossible to find, particularly as one goes further back in time. However, stromatolites, the fossilized remains of macroscopic mats of bacteria, are known to have existed 3.5 Gyr ago, which is relatively soon after the Late Heavy Bombardment 3.9 Gyr ago, especially for fully developed, rather perfected DNA life-forms. In fact, isotopic evidence for life suggests that life might have existed even earlier (3.9 Gyr ago). By then, life already must have gone through the RNA world, which started out with prebiotic synthesis of proteins and RNA. Life would have already advanced to having cells with RNA as both the coding and catalytic molecule by then. Proteins then took over the catalytic function, and then finally, DNA took over the coding functions. Life, Ruvkun said, already perfected the core biology by about 3.5 Gyr ago. Either it needed to evolve very fast, or it needed to arrive on Earth fully formed already.

A bold belief by Ruvkun is that the tree of life didn't start ~4 Gyr ago here on Earth, but rather ~10 Gyr ago somewhere else and then later brought to Earth. A weaker statement of panspermia, he said, would be to say that life on Earth may have spread to Mars. Maybe, Ruvkun said, the best way to look for life there is the best way to look for life here. The best way here, he said, is with DNA-based surveys. This is a convenient method because of



FIGURE 4.4 Cost per genome over time compared to Moore's Law. The approximate cost by December 2016 is about \$100 per genome. SOURCE: National Institutes of Health, National Human Genome Research Institute, "The Cost of Sequencing a Human Genome," updated July 6, 2016, https://www.genome.gov/sequencingcosts/; presented in Gary Ruvkun, "Looking for Life as We Know It on Other Planets," presentation to the Workshop on Searching for Life across Space and Time, December 6, 2016.

the trillion-dollar investment already made in genomics. With the Oxford Nanopore system, Ruvkun said it would be foolish not to use that method first when exploring Mars. To get life from Earth to Mars, meteorite impacts are necessary to eject material from Earth into space. Within about 30,000 years, 0.001 percent of that ejecta can land on Mars.<sup>5</sup> Interestingly, that same study showed that 1 percent of the ejecta lands back on Earth, which could potentially repopulate an Earth sterilized by the effects of the meteorite impact.

Ruvkun proposed to take martian soil and extract DNA from it. Similar things are already done routinely on Earth, using soil samples from many different types of environments. One can break open any cells present in a sample of martian soil with an electrical disruptor to release the cells' DNA. Next, any DNA that is present in this soil is purified on a solid matrix that specifically binds to DNA. There are then standard ways to randomly fragment DNA and put known DNA sequences (linkers) onto the ends of the unknown sequences of the DNA from the soil. One can then transport these DNA molecules, one at a time, through a nanopore—a commercial technology in which the pore's conductivity is changed as the DNA goes through the pore. By measuring the change in conductivity, each DNA molecule's nucleotide sequence is obtained. This sequence data is then transmitted to Earth as a megabase file.

Contamination is a major issue, however. He recounted how the Neanderthal genome was sequenced.<sup>6</sup> After drilling into the bone and making DNA out of it, 99 percent of the DNA was bacterial. They were then able to

<sup>&</sup>lt;sup>5</sup> B. Gladman, L. Dones, H.F. Levison, and J.A. Burns, 2005, Impact seeding and reseeding in the inner solar system, Astrobiology 5:483.

<sup>&</sup>lt;sup>6</sup> M. Krings, A. Stone, R.W. Schmitz, H. Krainitzki, M. Stoneking, and S. Pääbo, 1997, Neanderthal DNA sequences and the origin of modern humans, *Cell* 90:19.

pull out the 1 percent that was Neanderthal. Ruvkun said that this example shows that, even if there would be contamination on Mars from Earth life, you would still be able to see a real signal of extraterrestrial genomes. He finished by saying that the Oxford Nanopore technique could even read nonstandard nucleic acids, although its best use would be to search for life as we know it.

# **Audience Participation**

A member of the audience commented that he was doing some sequencing for his Ph.D. a couple of years after Viking landed. Working from Ruvkun's numbers, the Oxford Nanopore system could now do his Ph.D. [research] in 0.086 picoseconds. He then took strong exception to Ruvkun's claim that the conservation of key genes for billions of years means that it is perfect. The audience member said that it isn't perfect, but rather, it's just good enough. An example is the vertebrate retina, which is a bad design (it's upside down), but it won't change because it's good enough. Ruvkun said that he strongly disagreed with that. The audience member then noted that the first genome was sequenced in 1977, the phi X 174 (or  $\Phi$ X174), a bacteriophage virus.<sup>7</sup> Two years later, a paper titled "Is bacteriophage phi X174 DNA a message from an extraterrestrial intelligence" was published, which shows that people have been looking at DNA in terms of astrobiology for a long time.<sup>8</sup>

Another conference participant reported that, MinION, the Oxford Nanopore's portable DNA sequencer, has an error rate of 17 percent and asked how it was being improved. Ruvkun said that the 17 percent error rate is per nucleotide. However, there is redundancy built in because you're running the test many times. The accuracy is only a significant problem if you're looking for a one base difference, such as a human cancer gene. A high error rate isn't important if you're just testing to see whether there is a ribosomal gene on Mars or not. Ruvkun tested it on *Bacillus subtilis* and was happy with the results. The 17 percent error rate just wasn't a problem because they got ~100 independent runs of each genomic region.

According to one workshop participant, a ribosomal ancestry reconstruction is characteristically different from protein reconstruction because you never need to put losses into a ribosomal tree. You can, however, always do that with gains, which is almost never the case with a protein reconstruction. This means that there are some things that we don't know about RNA evolution, both after and certainly before translation. The selectionist interpretation can therefore not always be used for genes, because some of them are locked in at the network level. Ruvkun responded that when he envisions sequencing on Mars, he hopes to find that Mars is stuck in the RNA-world stage. He then repeated that the ribosome is a living fossil of the RNA world; it looks like an RNA replicase would before it could do translation. The transfer RNA adapter molecules were probably replicator molecules that were taking RNA segments. The ribosome, he said, is probably a re-engineered replicase. He said that the RNA world hypothesis is pretty well supported by the discovery of catalytic RNAs. He thinks that the RNA world wasn't here on Earth like most scientists believe, but rather, it was somewhere else. Ruvkun just thinks that 100 million years is far too short of a time to go from the RNA world to full-on bacteria.

Another member of the audience asked how to use the Oxford Nanopore system in situ on another body's surface. Ruvkun said it was simple. You just add in a sodium dodecyl sulfate (SDS) solution, a little soap, a hydrophobic disruptor, and then sonicate it. After that, you adhere the DNA and run it into the Oxford Nanopore system. (There is a backup plan if it is RNA.) It's a robust system, but of course, it would need to be prepared in such a way as to survive months or years in the harsh environment of space.

The Neanderthal DNA (1 percent) from all of the bacterial DNA (99 percent) had implications for planetary protection, one workshop participant said. Ruvkun replied that there is still the issue of bringing organisms to Mars. Ideally, he would like to bring DNA with them as a positive control, potentially synthetic DNA. If the goal is really human exploration, Ruvkun said that they should suspend all planetary protection protocols.

A workshop participant then said that radiation damage on the surface of Mars looks bad for finding DNA there. He then asked how old DNA could be on Mars and still be detectable with the Oxford Nanopore technique,

<sup>&</sup>lt;sup>7</sup> F. Sanger, G.M. Air, B.G. Barrell, N.L. Brown, A.R. Coulson, J.C. Fiddes, C.A. Hutchison, P.M. Slocombie, and M. Smith, 1977, Nucleotide sequence of bacteriophage φX174 DNA, *Nature* 265:687.

<sup>&</sup>lt;sup>8</sup> H. Yokoo and T. Oshima, 1979, Is bacteriophage phi X174 DNA a message from an extraterrestrial intelligence, *Icarus* 38:148.

58

considering that organisms might be more common in Mars' past than in Mars' present. Ruvkun said an upper limit on their ability would be finding DNA from about 1 million years ago. However, he wouldn't bet on the extinction of microbes. He thinks microbes are extremely adaptable and is a proponent of the saying "if life is anywhere, it's everywhere." The biggest issue, he said, is whether you're sensitive to life. With DNA, amplification is easy, but there is still the problem of interpreting it in the presence of a background. They're not aiming for a fossil though. On the same subject, another audience member said that concentrating a sample to find a cell might be difficult. Ruvkun finished by saying that DNA is the best at concentrating a sample because it can be amplified, although the audience member still questioned the ability to process a large amount of material.

# SIGNATURES OF LIFE AS WE DON'T KNOW IT

Steven Benner from the Foundation for Applied Molecular Evolution began his talk with the "paradox of molecular signatures" (earlier framed in terms of the "paradox of life" or the "paradox of a biosignature"), which says that a reliable biosignature is a molecular system that cannot arise without life. The paradox is then that this life could never arise. This means that no biosignature can exist to detect life soon after it has arisen—that is, soon after a molecular system has gained access to Darwinian evolution. Instead, we must rely on the subsequent ability of Darwinian evolution to create molecules having structures and complexes that could not possibly have arisen via abiological processes. Darwinian evolution, he said, is a necessary universal feature of life.

# The Universal Genetic Biopolymer Structure

Benner then asked what properties a genetic molecule would need for Darwinism to operate on it. He offered one constraint: it needs to be a one-dimensional biopolymer (attempts to assemble a two-dimensional version have so far failed). To support Darwinism, that biopolymer must be able to change its structure to change its information. However, these changes in its structure cannot substantially change its physical and chemical behaviors (e.g., its solubility, molecular recognition rules, and reactivity). For example, in the one genetic biopolymer that we know of, DNA, replacing a guanine with an adenine does not substantially change its physical and chemical properties. Such systems, he said, are rare. Proteins, polysaccharides, and most other classes of polymers exhibit dramatic physical and chemical changes with just small changes in structure. As an example, changing one amino acid in hemoglobin out of 576 causes sickle-cell anemia. In contrast, a biopolymer able to support Darwinism, the biopolymer has the ability to be imperfectly replicated, where those imperfections are themselves replicable. This is exemplified for both DNA and RNA, whose properties do not greatly change upon changing nucleotide sequences. Almost all DNA and RNA sequences are soluble in water, bind their complements, precipitate in ethanol, and template polymerases.

The property that makes DNA and RNA special, according to Benner, is that they have a repeating backbone of monopoles. For DNA and RNA, this backbone is the negatively charged phosphate unit. Proteins, on the other hand, have a repeating backbone dipole, which is why they fail as a genetic biopolymer. Evidence for this comes from synthetic biology (also called constructive biology). DNA analogs have been made that do not have a repeating backbone charge. These molecules precipitate and act just like proteins in the sense that their physical behavior dramatically changes if even a single base is altered. A repeating dipole backbone can easily fold, while a repeating monopole backbone prevents folding (see Figure 4.5). A repeating monopole backbone also allows for templating, keeps the DNA soluble, and forces strand-strand interactions to occur at the edges of the nucleobases. DNA's backbone keeps its bulk molecular properties the same because the monopole backbone is its dominant property. This polyelectrolyte theory of the gene, Benner said, will be true for all life in water throughout the universe. This, Benner said, is not true for biopolymers that lack a repeating backbone charge. Indeed, synthetic biologists have been able to create alternative forms of DNA with entirely different nucleobases, as long as they have retained the repeating backbone charge. In contrast, they have not been able to remove the backbone charges and obtain an evolvable biopolymer.

Conveniently, Benner said, finding polyelectrolytic genetic biopolymers in a sample of water obtained from an alien locale would be trivially easy and, in fact, is the easiest type of potential genetic molecule to find. The

#### LIFE DETECTION TECHNIQUES



(above) A backbone with a repeating dipole easily folds (below) A backbone with a repeating charge extends to template



FIGURE 4.5 A genetic biopolymer (for water-based life) needs to have a backbone with a repeating charge to prevent folding. SOURCE: Steven Benner, "Paradoxes of Molecular Biosignatures," presentation to the Workshop on Searching for Life across Space and Time, December 6, 2016.

polyelectrolyte will easily bind to a polycharged detector, more easily than other kinds of molecules that carry only a single charge or a small number of charges. As a signaling mechanism, a longer polyelectrolyte will displace shorter polyelectrolytes that are tagged with fluorescence or radio labels. Long polyelectrolytes can be detected in this way even if they are present only at extremely low concentrations in water. After they are detected, they should be examined to determine whether they are random biopolymers or Darwinian biopolymers. Key features to indicate Darwinian biopolymers are homochirality and being built from a "controlled vocabulary" (a small set of building blocks). Thus, this model for the universal life detection system assumes only the universality of Darwinism and the polyelectrolyte theory of the gene based on one-dimensional biopolymers.

#### **Prebiotic Chemistry**

The rest of the genetic biopolymer, Benner said, could be anything. However, ribose is clearly one of the best backbone molecules that have been examined, particularly in terms of molecular recognition, although a handful of other possible structures have been found to work analogously (threose and some bicyclic structures are especially worthy of note). For this reason, most of the prebiotic chemistry community focuses on ribose RNA. While there is not a clear path to get RNA as the first Darwinian biopolymer, Benner said that a path is likely to be found soon, since work over the past decade has offered solutions to many of the problems that were considered insurmountable in the RNA-first model for the origin of Darwinism. One problem arises from the general observation that applying energy to organic matter of the sort that cannot undergo Darwinian evolution just produces tar (i.e., heating up sucrose makes caramel). Additionally, it is hard to obtain an available form of phosphate, especially in the presence of calcium. Further, RNA is unstable in water. Water is essential for life, but it also destroys the RNA biopolymers.

He then said that the C=O group in an organic molecule is a source of horrible reaction complexity that leads to such tars. Ribose has a C=O group, enolizes, reacts with itself, and then forms tar under alkaline conditions. Experiments show that ribose forms tar at pH 7 at 50°C after just 7 years, which was said to preclude the use of ribose and other sugars from being components of the first genetic material.<sup>9</sup> However, mineralogy can mitigate this problem. Borate, Benner said, is a poor mineral-forming element, is concentrated in residual melts and igneous rocks, and is easily leached from these rocks by erosion into aquifers. Sugars have adjacent hydroxyl groups, which borate binds to in an extremely stable way. Borate thus binds to ribose, removing the C=O group and preventing ribose from becoming tar, thus allowing it to accumulate. Further, borate also guides the chemical reactions of smaller carbohydrates that deliver 5-carbon species, including ribose.

Benner then described recent literature that supports a discontinuous model for RNA synthesis. The model starts with gases (CO<sub>2</sub>, H<sub>2</sub>O, N<sub>2</sub>, and CH<sub>4</sub>) that can be converted by UV light and electric discharge to give hydrogen

<sup>&</sup>lt;sup>9</sup> R. Larralde, M.P. Robertson, and S.L. Miller, 1995, Rates of decomposition of ribose and other sugars: Implications for chemical evolution, *Proceedings of the National Academy of Sciences of the U.S.A.* 92:8158.

cyanide (HCN), cynamide (HNCNH), and formaldehyde (CH<sub>2</sub>O), all generally agreed to have been available on the early Earth. Borate moderates the early chemical process that converts formaldehyde to ribose borate. Cyanide and cynamide are hydrolyzed to formamide and urea.

However, all this requires dry land. The entire process is defeated by dilution in a global oceanic system. That means that a submarine origin of life, as in hydrothermal vents, must solve the problem about dilution and the instability in water of many of the bonds in RNA. Benner said that a desert with occasional intermittent water might be ideal. He then referred back to a previous talk about Mars' changing obliquity, which would produce exactly the type of intermittency they need.

The two conditions Benner requires for life, borate and intermittently wet deserts, may not have been available on early Earth, according to some geologists. Further, several geologists have argued that, absent a history of plate tectonics, boron could not have been concentrated in the lithosphere sufficient to attain productive concentrations anywhere on early Earth. Efforts to find residual soluble boron minerals in ancient rocks are not likely to be directly successful, because kernite, ulexite, colemanite, and other boron minerals are not expected to survive for a long time. These and other borate minerals are unstable to metamorphosis, yielding monazite, apatite, and tourmaline. However, we can look for these derived borate and phosphate minerals. For example, a 3.8 Gyr old rock was just recently found to contain monazite, apatite, and tourmaline—suggesting the existence of borate prior to their metamorphic alterations.<sup>10,11</sup>

Benner then addressed the possibility that the inventory of water on early Earth required an ocean world with no dry land. Without dry land, there could be no desert, no borate evaporites, no ribose, and no RNA. If this was so on Earth, the borate-involving prebiotic chemistry should nevertheless have been possible on Mars, which almost certainly had less water. Benner said that there could even be borate-ribose on Mars today. John Grotzinger's earlier talk (see Chapter 2) showed that everything Benner thinks needs to be there for life to arise was actually on Mars. This includes opal CT; Elisa Biondi in Benner's group recently found evidence that RNA adsorbs onto opal in stable form.

Moving to the problem of phosphate concentration, Benner referenced records showing contemporary precipitation of gypsum (CaSO<sub>4</sub>•2H<sub>2</sub>O) and lüneburgite (Mg<sub>3</sub>B<sub>2</sub>(PO<sub>4</sub>)<sub>2</sub>(OH)<sub>6</sub>•8H<sub>2</sub>O) on Earth. This observation is significant with respect to the availability of phosphate and complex geological environments. In simpler environments, when Ca<sup>+2</sup>, Mg<sup>+2</sup>, PO<sub>4</sub>-<sup>3</sup>, and SO<sub>4</sub>-<sup>2</sup> are interacting together in the absence of borate (BO<sub>3</sub>-<sup>3</sup>), they form apatite (composed of Ca<sup>+2</sup> and PO<sub>4</sub>-<sup>3</sup>) and epsomite (composed of Mg<sup>+2</sup> and SO<sub>4</sub>-<sup>2</sup>). Apatite sequesters phosphate, but largely unproductively. In the presence of borate, however, they form gypsum and lüneburgite. This keeps phosphate from being locked away via calcium capture. The phosphate instead joins with the borate into borophosphate. When ribose encounters lüneburgite, it extracts the boron, which disrupts the mineral and releases phosphate. The phosphate is then available to phosphorylate the nucleoside. With the borate coordinated to a specific pair of OH groups, the only products are the five prime phosphorylate ribonucleosides. Without the borate present, many ribonucleoside phosphorylation products are seen in laboratory settings. That all this corresponds with martian geochemistry makes Benner think that panspermia from Mars to Earth is at least plausible, especially if the absence of desert land on early Earth requires this prebiotic chemistry be on Mars. It should be noted, however, that recent work of Stephen Mojzsis supported by the Foundation for Applied Molecular Evolution-Templeton program found that the amount of dry land on early Earth may be sufficient so as to not enforce this requirement of panspermia.

Moving to a different type of chemistry, Benner said that the main problem with life forming in non-aqueous environments, like Titan's hydrocarbon lakes, is solubility. A biopolymer with a backbone of repeated charges will not dissolve in hydrophobic solvents. A potential solution is a repeating dipole that presents only the same end of the dipole all along the biopolymer (e.g., polyethylene glycol). A molecule like this might be able to prevent aggregation and folding while maintaining a genetic function. However, cryosolvents are bad solvents because

<sup>&</sup>lt;sup>10</sup> E.S. Grew, R.F. Dymek, J.C.M. De Hoog, S.L. Harley, J. Boak, R.M. Hazen, and M.G. Yates, 2015, Boron isotopes in tournaline from the ca. 3.7-3.8 Ga Isua supracrustal belt, Greenland: Sources for boron in Eoarchean continental crust and seawater, *Geochimica et Cosmochimica Acta* 163:156.

<sup>&</sup>lt;sup>11</sup> S. Mishima, Y. Ohtomo, and T. Kakegawa, 2016, Occurrence of tourmaline in metasedimentary rocks of the Isua Supracrustal Belt, Greenland: Implications for ribose stabilization in Hadean marine sediments, *Origins of Life and Evolution of Biospheres* 46:247.

they're cold and therefore relatively insoluble. They are unlikely to work on Titan, but they might work on a warm Titan with oceans of hexane or octane.

#### Disequilibrium and the Limitations of Darwinism

Benner's last point related to a claim often made in astrobiology that disequilibrium can be used as a biosignature. For example, a forest in an atmosphere of oxygen is a disequilibrium that can be interpreted as a biosignature. However, Benner said, the disequilibrium exists because Darwinism has been ineffective at creating enzymes that catalyze the destruction of cellulose. Indeed, this ineffectiveness is illustrated by the abundance of uneaten beds of coal. More than a dozen different cellulase families appear in a range of organisms. However, they all appear to have evolved from previously created enzymes that initially had other roles. Darwinism has not created a macroscopic life-form able to exploit the energy in the forest effectively, which illustrates a possible limitation of Darwinism.

One solution to this problem, Benner said, is Lamarckism. For example, humans learned how to use cellulose about 2 million years ago by learning how to build fires and burn wood, and then transmitting this skill to their children not by DNA, but rather by teaching. Benner said that any alien life smart enough to talk to us, or especially to travel to Earth, would also have discovered Lamarckism. Indeed, it would have gained Lamarckian control over its molecular biology, eliminating Darwinism as a mechanism for preserving its core genetic capabilities or creating new ones. Clustered regularly interspaced short palindromic repeats (CRISPR) and germline gene therapy puts humans on the verge of this evolutionary change. Therefore, Lamarckism will have its own biosignatures. For example, carbon-fluorine bonds cannot be generated either biologically or abiologically, but can be manufactured by an intelligent life form. In other words, intelligent life will eschew Darwinism and instead use Lamarckism. This means that the search for extraterrestrial weird life does not necessarily require understanding its unknown molecular biology.

Benner then presented his conclusions. On a planet with water, any life will have a genetic biopolymer with a backbone of repeating charges. This type of molecule can be easily concentrated from alien aqueous environments, such as from the plumes of Enceladus, in order to be detected with today's technology. Further, once concentrated, it can be easily detected in situ. Finally, downstream analysis of its structure will allow us to directly determine whether it is the product of Darwinism or a random, accidental polymer.

There is not yet a reason to believe that RNA is the universal structure, but on a planet with similar geology to the Earth or Mars, it might be likely. However, Benner thinks that, with a desert and an appropriate mineralogy, the abiotic synthesis of RNA will not be an unsolved problem for much longer. He also said that a good non-aqueous environment for life has not yet been discovered and that cryosolvents are particularly bad. Benner said that disequilibrium implicates the impotence, or even the absence, of Darwinism. It is not, if examined out of context, a biosignature. Lastly, Benner said that intelligent life that can move beyond Darwinism to Lamarckism would have its own set of biosignatures, regardless of the underlying molecular biology.

# **Audience Participation**

A member of the audience said that she liked Benner's comparison between the calcium phosphate and the magnesium borate. The geochemical principle behind this, she said, is called the "geochemical divide." She then said that another anion at the time would have been carbonate. She would be interested in an experiment that added various quantities of carbonate to the system to see how that would affect the types and ratios of different minerals precipitating from it. Benner started by saying that we know very little about the early Earth's geology. However, he thinks that it was the borate that was scarce in the early Earth. He then said that people have studied systems with multiple ionic inputs and that he was open to collaboration on future studies.

Emphasizing a caveat to Benner's favorable view of martian geology in terms of the origin of life, another audience member said that the data is not necessarily representative of the entire planet. Gale Crater, for example, had multiple episodes with water and minerals precipitating through it. There are therefore questions about how this stuff formed and whether it was in association with other materials there. She thinks that maybe new types
of work on Mars might be necessary to see whether all these necessary minerals were present and available at the same time sometime in Mars' past. Benner said that he is aware of the environmental heterogeneity on Mars. However, he's ready now to simulate Mars with various minerals to see how hydrogen cyanide and formaldehyde reacts in it. Benner said that doing these experiments on manufactured minerals is required because even samples from Death Valley would contain enough life to eat all the organics they gave it. He said that he's tested their ability to synthesize minerals using 50 rocks from the "Benner Collection of Fine Rock Specimens" in order to show that his synthesized rocks are comparable to natural rocks.

A workshop participant then said that the Mars teams have found mobile phosphate (in the sense that its concentration changes over short distances) in multiple cases. The Curiosity team has also discovered boron in higher than expected concentrations. He then finished saying that all the components to develop life are there.

## Instrumentation

The moderators for the instrumentation session were Phil Neches of Teradata Corporation and Nilton Renno of the University of Michigan. Neches began the session by declaring himself more of a technologist than a scientist. He said that instrumentation is where science, technology, and commerce come together. Neches then introduced the first presenter.

## PLUME FLY-THROUGH MISSIONS: DETECTING LIFE IN SITU AT SEVERAL KILOMETERS PER SECOND

Morgan Cable of the Jet Propulsion Laboratory began her talk by describing three types of plumes in our solar system. Volcanic plumes, such as Io's Loki, are usually rich in sulfur but deficient in water. Cometary plumes are rich in  $CO_2$  and  $H_2O$  and are induced by sunlight. The most interesting type of plume when it comes to life, however, is the type emitted by ocean worlds. Enceladus is one such confirmed example, and Europa also has a potential plume detection.

Enceladus has roughly 100 distinct jets emanating from the tiger stripes on its southern, polar region (see Figure 5.1). These form into a single plume high above the surface. The plume is modulated by diurnal tidal flexing, but appears to be steady at least since the time of Voyager and probably earlier, since it feeds Saturn's E-Ring. This plume contains both gas and solid particles. The Cassini Ion and Neutral Mass Spectrometer (INMS) has determined the plume to be rich in  $H_2O$ ,  $CO_2$ ,  $CH_4$ ,  $NH_3$ , and heavier hydrocarbons all the way up to its mass limit of 100 atomic mass units. Particles observed by the Cassini Cosmic Dust Analyzer (CDA) include water ice, salts, silica, and organics. The size and oxidation state of the silica nanograins suggests hydrothermal activity on Enceladus.

Regarding plumes in general, she said that if one wants to look for biosignatures in the plume, one must measure not only its composition, but also the relative abundances. The source of the plume must also be determined, whether it be from the subsurface ocean or from the surface itself. The plume grains are presumably where biomarkers would be concentrated (potentially even cells). Their size distribution and formation mechanism (e.g., spray aerosols) are also important. The plume's overall structure and dynamics must also be known. Cable said that all of this must be placed in its environmental context.



FIGURE 5.1 Plumes from the southern polar region on Enceladus. SOURCE: NASA/ JPL-Caltech/SSI, presented in Morgan Cable, "Plume Fly-Through Missions: Detecting Life In Situ at Several Kilometers per Second," presentation to the Workshop on Searching for Life across Space and Time, December 6, 2016.

#### **Instrumentation Classes from the Past**

Cable then began to go through several instrument classes that have been used to study plumes or could be used more in the future. She started off with the mass spectrometer, such as the Cassini INMS or the Europa Mass Spectrometer for Planetary Exploration/Europa (MASPEX), which are able to target gas and, occasionally, ice grains. Mass spectrometers have an extensive flight heritage. Recent advancements have been made in mass resolution and sensitivity. The Cassini INMS could not distinguish between CO and  $N_2$  (both 28 atomic mass units), but MASPEX can, since it allows for resolving ambiguities with respect to the atomic mass units of a molecule and also enables isotopic investigations of other molecules, some of which are related to life. Deconvolving complex mixtures of materials can be difficult though. This can be mitigated by trapping material and then slowly releasing them by increasing the temperature. However, an Earth-based laboratory would be best.

Cable then explained that dust detectors, such as those on the Cassini, Stardust, and Europa Clipper missions, are designed for grains containing salts, ions, and organics. The reflectron design has an extensive flight heritage and is made specifically for plumes (i.e., low-density particles sampled at high velocity). They can make rapid measurements, allowing for the measurement of plume structure during a fly-through. A disadvantage to this is that the ionization of incoming grains is dependent on both the mass of the particles and the velocity of the collision. Again though, bringing material back to a laboratory on Earth would be best. However, this is not often possible. Currently, no technology exists to trap and preserve plume particles for long durations other than aerogel, which nonetheless has a lot of organic contamination and is not conducive to life investigations.

Near-infrared spectroscopy, like on the Cassini and Europa Clipper missions, can also target both gaseous and solid particles in plumes, although sensitivity is limited for trace species. Imaging spectroscopy can also provide information on the plume's structure and grain size. However, spectroscopy can only identify certain functional groups, like an N-H or C-O stretch, but cannot unambiguously identify them as a constituent of glycine, for example. Near-infrared spectroscopy can also look for particles being deposited on the surface by looking at the body's surface albedo.

Ultraviolet (UV) spectroscopy also has extensive flight heritage; it has been used on both Cassini and Juno and is planned for the Europa Clipper mission. It can identify plumes at a distance and look for hydrogen and oxygen auroras and simple organics. Similar to near-infrared spectroscopy, the UV technique can also look at the body's surface albedo to look for particles being deposited. Again though, sensitivity for trace species is limited.

Microwave (submillimeter) radiometers, such as the Microwave Instrument for the Rosetta Orbiter (MIRO), also have a long history. They can observe three-dimensional (3D) plume structure and dynamics, but can only

#### INSTRUMENTATION

observe gas-phase polar molecules. A microwave radiometer is able to measure the temperature of water, but like the spectrographs, it has a limited sensitivity to trace species.

#### **Instrument Classes of the Future**

Going back to the mass spectrometer, Cable said that adding a gas or liquid chromatograph could be a very powerful tool. It could allow for the detection of amino acids (including their chirality), proteins, lipids, and other biosignatures. However, these instruments are tailored for certain species, and one has to be aware of potential confounding species as well. The need for greater sensitivity and collection of a sufficient sample could also prove problematic. Another challenge is the complexity of the instrumentation needed to capture and examine particles.

Cable mentioned laser desorption ionization mass spectrometry as another technique—one that she believes will be used on Mars. It uses a soft ionization method that is great for large molecules like DNA, RNA, and proteins. Again though, a very complex instrument is needed to capture and concentrate these plume particles when the instrument is flying at several kilometers per second.

Raman spectrometry would target functional groups, such as amines, carboxylic acids, and ketones. Coupled with microscopic imaging, this could be used to confirm whether a particle is indeed a cell. It is not capable, however, of unambiguously identifying complex organic molecules and again requires complex instrumentation.

Immunoassay-based microfluidic chips, like the LifeMarker Chip, can also identify complex biomolecules like DNA, RNA, and proteins. Disadvantages of the LifeMarker are that it can only target Earth-like molecules and again requires complex instrumentation.

A microchip capillary electrophoresis with laser-induced fluorescence can detect a wide range of organic molecules: chiral amino acids, lipids, amines, thiols, fatty acids, DNA, RNA, and proteins. As with the previous methods discussed above it requires a complex instrument to capture and concentrate these particles.

The last instrument class Cable described was microscopic imaging. This technique would be able to see individual cells and their movements, which would be a "smoking gun" for life. Microscopic imaging, again, requires complex instrumentation to capture and concentrate particles. Another problem may be that whole cells might not be common in plume grains.

#### **Planning Future Missions**

Cable then went through a list of things to consider when designing a plume fly-through mission. The altitude of a fly-through is important, as this determines the plume density and particle size distribution the spacecraft will encounter. For example, grain sizes of 10 microns in the plume of Enceladus can be reached safely at a typical flyby altitude, whereas a grazing swing past Europa necessary to see 10-micron-sized grains could be dangerous, especially considering planetary protection issues. The issue of flythrough speeds has two opposing arguments. High velocities are needed to ionize the species when hitting the collection device, but higher velocities also increase fragmentation. One solution is choosing an intermediate speed, while another option is doing both slow and fast flybys. The capture medium is another issue. Metal plates are simple and can help ionize the particles, but they can cause fragmentation. Aerogel preserves the particles, but it is difficult to then extract the particles from the aerogel afterwards. Another consideration is the number of fly-throughs that are needed. Repeated fly-throughs can either repeat measurements to see variations over time or can be used to build up larger concentrations for later analysis. The choice of which species to target, in terms of molecules or whole cells, gaseous or grains, and whether they are susceptible to fragmentation, is another important consideration. Finally, Cable finished by saying that everything needs to be understood in context and that any in situ life detection would require a sample return mission to confirm it.

## **Audience Participation**

The first question from the audience challenged Cable's concluding thought and asked why a sample return mission would be needed rather than a lander. Cable accepted that a lander could absolutely be included. The

audience member followed up by asking whether a lander is needed before sample return or whether it could be skipped in favor of doing sample return sooner. Cable admitted that that was the process for Mars, but made the distinction that getting to Mars is much quicker than getting to the outer solar system. She then questioned how long we would be willing to wait to follow the progression from fly-through to lander to sample return. The participant said that she is suspicious of the length of time sample return missions require. The martian sample return mission does not plan to do any sample curation prior to return. A further problem with the outer solar system, she said, is that the icy and volatile materials would need to be stabilized. Therefore, she preferred a lander. Another participant then said that Cassini has already justified a sample return and suggested that an in situ and a sample return mission should be planned together. Cable agreed. Referring to the capture and cryogenic stabilization of a sample return, another audience member said that even a non-pristine sample would be interesting. He said that most things don't decompose at 25°C over 10 years anyway.

Some of the instruments Cable mentioned were going to be on the new Europa mission, one workshop participant said. The audience member then asked what type of mission (and timeframe) she is looking for to address these questions for Enceladus. Cable said "5 years ago" because Cassini will be lost soon, which means that no new in situ information about the Saturnian system will be available. While admitting that everybody has their favorite planet or moon, she said that Enceladus is just spewing free samples from its south pole. She said that Cassini has done wonderful things, but it was never designed to be a seafaring or life-detection mission. Its mass spectrometer cut-off at 100 atomic mass units is too low to include more than a couple of tiny amino acids. She wants to extend the mass range and get new instruments with their advancements in sensitivity and maybe even a sample return mission.

Another member of the audience then asked whether the onboard computation capacities and downlink rates could support these new mission instruments. Cable said that the mass spectrometry technique creates a lot of data. However, it also has the processing capacity to quickly look at the data and select the most informative mass spectra to transmit back to Earth.

A member of the audience then requested that Cable elaborate on the importance of measuring not only the abundances of molecules, but also their ratios. He said that the biosignatures would presumably be coming from the bottom of a deep ocean and asked if there were cause for concern that the chemistry in the oceans or plume could alter the signals. Cable answered by saying that these measurements need to be understood in context. Issues like ions dissolved in the ocean, the transport time for molecules to get to the surface, and how they're turning into aerosols and particles in the air must be understood. She said that, barring some tentacle waving hello, there will not be a simple yes or no at first.

Lastly, a participant at the workshop then commented that, with respect to the detection of organic molecules, instrumentation may have different levels of sensitivity to different types of molecules, such as heavy versus light. She then said that instruments are getting down into the parts per trillion range. However, organic contamination must be kept equally as low, which is incredibly challenging to do. This is why, Cable concluded, she wanted to ultimately have a sample return mission where these sensitivity questions could be thoroughly addressed.

## LIFE DETECTION CAPABILITIES OF LUVOIR AND HABEX

Shawn Domagal-Goldman of NASA Goddard Space Flight Center began his talk by emphasizing the need for collaboration and complementarity in the search for biosignatures. He said that the properties of the exoplanet population discovered to date have caused us to reconsider how planetary systems form and evolve.

The next major step, he said, is to characterize their chemical compositions. It has already been done a bit with the Hubble and Spitzer space telescopes, but it will ramp up with the launch of the James Webb Space Telescope (JWST) and then later with the Wide Field Infrared Survey Telescope (WFIRST). The most successful planet discovery techniques to date, the radial velocity and transit techniques, are biased towards planets close to their host stars. Conversely, WFIRST's microlensing mission will be biased towards the outer planets, completing the census of exoplanets. Its coronagraph could also characterize gas giant exoplanets, and a starshade—an option that has been studied but not approved—could enable the search for potential biosignatures. Extremely large ground-based telescopes are now being developed that should be able to do not only transit spectroscopy (which would be biased

#### INSTRUMENTATION

towards the stratosphere), but also direct imaging of planets—perhaps even rocky ones—in the habitable zones of M dwarfs. However, Domagal-Goldman is concerned about the habitability of planets orbiting M dwarfs due to the possibility that they would have lost their atmospheres due to high-energy radiation from their host stars.

## **Potential and Desired Telescope Specifications**

Therefore, Domagal-Goldman said, we should think about how to complement JWST and the extremely large, ground-based telescopes and look beyond M dwarfs. One question is what the wavelength range should be. Ideally, a telescope will need to look at wavelengths shorter than both those JWST is able to observe and those for which ground-based adaptive optics are optimized—that is, the UV and visible. The near-infrared is nonetheless also an interesting region. The wavelength range can help mitigate false positives and increase user knowledge of the environmental context (see Figure 5.2). For exoplanets, this means identifying as many gases and their abundances as possible, which means a wide wavelength range. Domagal-Goldman likes the idea of using the flux or kinetics of biosignatures as a discriminant, a conclusion independently reached in the earlier presentation by Tori Hoehler. Abiotic production of many molecular species proceeds at a rate that is orders of magnitude lower than would be expected through production by life. This contrasts with the concentrations of gases, for which abiotic processes can lead to higher values of oxygen and ozone than biotic ones.

The challenge to identifying or constraining fluxes is that it requires a great deal of environmental context. The teams for LUVOIR and HabEx, he said, want the lower wavelength cutoff to be set at about 100 nm to characterize the far UV starlight that produces a lot of photochemistry as an important constraint on the abiotic sources of oxygen and ozone. However, the wavelength cutoff for directly imaging planets would be 300 to 400 nanometers. Although WFIRST is only planning a maximum wavelength of 1 micron, LUVOIR and HabEx are considering going out to 2 to 3 microns. This would yield the detection of  $O_2$ ,  $O_3$ ,  $H_2O$ ,  $CH_4$ , and high levels of CO and  $CO_2$ . This set of gases would allow for identification of high flux rates of  $O_2$  and  $CH_4$  to modern Earth's atmosphere, and discrimination of Earth's  $O_2$  as biogenic in origin. Obtaining this wide wavelength range is difficult. Maintaining UV capability requires clean mirrors, but when cooling the telescope below 260 K in order to observe in



FIGURE 5.2 Spectrum of Earth as seen from Saturn. SOURCE: NASA Space Telescope Science Institute (STSciI) presented in Shawn Domagal-Goldman, "Life-Detection Capabilities of LUVOIR and HabEX . . . and WFIRST," presentation to the Workshop on Searching for Life across Space and Time, December 6, 2016.

the infrared, precipitates will appear on the mirror's surface. Keeping the telescope above 260 K, however, will degrade observations beyond about 1.8 microns. On the other hand, the extension out to at least 1.8 microns will maintain the ability to detect the suite of gases required to constrain oxygen fluxes.

Another technical challenge, according to Domagal-Goldman, is the starlight suppression. Using a starshade flying in formation significantly lessens the burdens placed on the telescope itself. Another advantage of a starshade over a coronagraph is that there is no outer working angle, meaning that outer planets potentially as far as Kuiper belt distances will remain visible. However, a major disadvantage is that, as the observations move to longer wavelengths or are made with larger telescopes, the starshades become quite large. To observe at 2 microns on a mission like HabEx or to observe at any wavelengths with a large telescope like LUVOIR, a starshade with a diameter of about 100 m is required. Packing a starshade of that size would be challenging. The biggest challenge, he said, may be the edge tolerance of the starshade petals. A coronagraph, on the other hand, suppresses the starlight within the telescope. However, waveform distortions from the optics must be corrected. In a segmented mirror, this can be done by making very stiff segments and ensuring they do not move with respect to each other with active control systems. The mechanisms for doing this have all flown before, and the control systems required are already in operation on ground-based systems such as Keck. Thus, in theory, all the components are in place for this to work. In practice, however, this has not yet been demonstrated at the systems level at the precision required to suppress starlight sufficiently to detect and characterize potential Earth-like worlds.

Both LUVOIR and HabEx are proposed to observe potentially habitable planets and search for potential biosignatures. However, HabEx will be optimized for planets while enabling a broader range of general astrophysical observations. LUVOIR, on the other hand, will be a general observatory for a variety of astrophysical goals, including exoplanets. The two missions also have different levels of ambition. HabEx aims to search for planets around enough stars to have a very good chance at characterizing at least one rocky planet in the habitable zone of another star. LUVOIR, on the other hand, will attempt to characterize dozens of such worlds. LUVOIR will also be able to constrain the abundance of any property on those worlds, including a biosignature or combination of biosignatures, to a level of ~10 percent. Due to the uncertainty of future budgets and scientific and technological discoveries, Domagal-Goldman wants several options to be prepared for different future realities.

Each mission has two different potential architectures depending primarily on the telescope's aperture size. For reference, the Hubble Space Telescope and WFIRST each have a diameter of 2.4 m. The HabEx team is considering using either one 4-m, monolithic mirror or a 6.5-m segmented mirror (either hexagonal or pie-shaped), the same size as JWST. The LUVOIR design team is deciding between a 9-m and a 16-m architecture, both segmented. This is the largest telescope a launch vehicle could reasonably fit. He then simulated an observation of Europa with a ~10-m LUVOIR, which would be able to clearly see the structure of the claimed plume. Domagal-Goldman then simulated the number of potentially Earth-like planets observable as a function of aperture size. For a telescope with a 4-m, 8-m, or 16-m aperture, approximately 6, 25, or 100 potentially Earth-like planets would be observable, respectively. The more candidates observable, the more precise the constraints are on the fraction of rocky planets in the habitable zone. However, these telescopes won't just find potential Earths. They will find everything more detectable than Earth too. Even for the 4-m mission, dozens of other (likely) uninhabitable worlds will also be discovered, such as a warm Titan. A 12-m mission would likely find Jupiter analogs and warm Jupiters. Larger apertures allow for a higher cadence of observations, so more of the temporal domain is observable. This opens up techniques such as longitudinal mapping of planetary surfaces and maybe even latitudinal mapping using seasonal or orbital variations.

## **Notional Instruments**

Instruments are being considered for both telescopes. Domagal-Goldman said that both are likely to have a starlight suppression technique, probably a coronagraph. LUVOIR is planning an instrument called the Optical-IR Band Spectroscopy Coronagraph for Understanding Rocky Atmospheres (OBSCURA). The goal for OBSCURA is to get a contrast ratio of  $<10^{10}$  with low resolution spectroscopy (R > 150) from 0.2 to 0.4 or up to 1.8 to 2.4 µm if the stretch goal is met.

Another LUVOIR instrument is the UV Multi-Object Spectrograph (LUMOS). This would extend from the far- to the near-UV and have a high resolution of about  $R \approx 100,000$ . When used in multi-object mode, its resolu-

#### INSTRUMENTATION

tion would be "medium resolution." It would also have near-UV imaging capabilities. It will be a major upgrade of Hubble's Space Telescope Imaging Spectrograph (STIS). LUMOS would provide contextual information on the host stars of potentially habitable worlds. The High Definition Imager (HDI) would be similar to the Hubble Wide Field Camera 3 (WFC3) and would observe in the optical to near-infrared with a field-of-view of 4 to 6 arcminutes. It could possibly allow for high-precision astrometry to measure planet masses. Domagal-Goldman explained that HabEx is considering a "workhorse" UVOIR camera and a UV spectrograph. The UVOIR camera would deliver similar kinds of science to LUVOIR's HDI instrument, and the UV spectrograph would deliver similar science to LUVOIR's not the HabEx team will select one of these two instruments and leave an "open bay" for a second astrophysics instrument, which could be used for another instrument. This could be the other instrument that was considered, a foreign contribution, or something else.

The fourth and final proposed LUVOIR instrument is a high-resolution spectrograph (up to  $R \approx 100,000$ ) with high photometric precision for transits. Potentially, it could also precisely measure radial velocities in order to obtain planet masses. It can also be combined with the coronagraph via a fiber feed to the spectrograph to deliver ultra-high resolution spectra of exoplanets, which can help identify the presence or absence of specific molecules by the pattern of their individual absorption lines. Domagal-Goldman suggested this could be a powerful way to identify individual molecules even at low abundances. This would help improve the context for any potential biosignatures, thereby improving the confidence that they were sourced from biology.

Each telescope has technological challenges commensurate with their levels of ambition. The biggest challenge, according to Domagal-Goldman, is the starlight suppression. If the telescope uses a coronagraph, it must be highly stable and compatible with the entire telescope, including segmented mirrors if they are used. If it uses a starshade, the problems of deploying the starshade, flying in formation, and manufacturing the petals' edges need to be fixed. Other challenges include needing a heavy-lift launch vehicle and ensuring the compatibility of UV observations with a coronagraph.

## **Audience Participation**

A member of the audience said that his exoplanet friends told him that clouds are a big problem in measuring spectra of hot Jupiters. He then asked about how clouds could confound the detection of biosignatures on terrestrial planets. Domagal-Goldman told the audience member that his friends (with all due respect) were wrong to view clouds only as a problem because the formation of the clouds is an important planetary process in itself. He admitted that they do block and refract photons from lower in the atmosphere, but that it is a bigger problem for transit spectroscopy than it is for direct imaging. He said that simulations show that some photons from the planet's surface do get through the clouds though. Domagal-Goldman finished by wishing that people would think of clouds as conveyors of information.

## IN SITU DETECTION OF ORGANICS ON MARS

Jennifer Eigenbrode of NASA Goddard Space Flight Center started by thanking the members of the Mars Science Laboratory (Curiosity) team and the Sample Analysis at Mars (SAM) team for all their work. She then recounted a story about when Curiosity rover landed. During the landing event, she informally polled the team and found out that the majority of people in the room doubted that they would find organics on Mars, especially not in the top 5 cm of the martian surface. Eigenbrode said that she is now convinced that organics are widely distributed over the martian surface and throughout the rock record.

## **SAM Analysis Techniques**

Eigenbrode described the two in situ analysis techniques on SAM. First is the detection of bulk gas composition via evolved gas analysis (EGA), which can reveal the presence of refractory organic matter. Second is the detection of molecules with gas chromatography–mass spectrometry (GCMS). The SAM measurements of organic volatiles begin by heating up a crushed sample of rocks or soil to about 860°C. As gas comes off, a small portion of it gets

"sniffed" into the EGA. This gives an indication of the evolution of the bulk gas all mixed together. The rest of the gas is trapped, and certain analytes are released into the GCMS. The GCMS can identify specific molecules.

Two processes occur, Eigenbrode explained, when heating a sample of pure organic material using helium: thermal desorption and pyrolysis. Thermal desorption is the process where smaller, non-bonded molecules are volatized. When the heated material is purely organic, this process can occur up to about 400°C. Pyrolysis, which proceeds mostly at higher temperatures, is the actual breaking of bonds. This produces two peaks, one for thermal desorption at lower temperatures and one for pyrolysis at higher temperatures (see Figure 5.3). On Mars, the thermal desorption peak is split into two (again, see Figure 5.3). A high abundance of  $O_2$  is evolved at approximately 200°C to 300°C, with thermal desorption being dominant at lower and higher temperatures. A typical source of the  $O_2$  is the breakdown of oxychlorine phases, such as perchlorate. Pyrolysis remains the strongest process at the highest temperatures. It can break macromolecules apart. In natural material that we know of, organic matter is 75 to 90 percent macromolecular. Macromolecules with more functional groups are more easily broken. In the SAM data, there is a well-known, well-characterized background signal that mostly appears during the thermal desorption phase.

## **Rocknest and Mojave 2 Sample Results**

The first site that Curiosity visited was Rocknest, Eigenbrode recalled, an Eolian Drift on the Gale Crater floor often called "martian soil." Using evolved gas analysis (EGA) shows a background signal clearly visible, but there is also a bump at 825°C from the release of refractory organic material. This bump shows  $C_1$ ,  $C_2$ ,  $C_3$ , and possibly  $C_4$  signals. Eigenbrode said that this is probably a reduced carbon source bound up in a mineral, which had to break down in order to release the carbon. If it was just a refractory organic material in a macromolecule, the bump would have been more smeared out. This sample's bulk chemistry as measured by the Alpha Particle X-ray Spectrometer (APXS) is similar to that of Meridiani and Gusev. These locations are mostly basaltic and are thought to be a global signature, implying that the reduced carbon phase might also be global.

Eigenbrode then moved on to stratigraphy, focusing on two samples of mudstones. Mudstones are fine-grained, which is difficult for water to get through. This implies a better chance that organic materials may be preserved inside them. One sample is from Yellowknife Bay, Cumberland, and the other one is at Pahrump Hill/Marias Pass called Mojave2, but both are considered lake lacustrine deposits. However, they were deposited at different times and have different compositions. Thermal desorption of the Cumberland sample showed chlorinated  $C_1$  to  $C_4$  hydrocarbons and benzene.<sup>1</sup> EGA found a set of high temperature (>500°C), correlated peaks for single-ring,



FIGURE 5.3 Response when heating a sample of pure organic material under helium. *Left:* Typical result on Earth. *Right:* Result on Mars. SOURCE: Jennifer Eigenbrode, "In Situ Detection of Organics on Mars," presentation to the Workshop on Searching for Life across Space and Time, December 6, 2016.

<sup>&</sup>lt;sup>1</sup> C. Freissinet, D.P. Glavin, P.R. Mahaffy, K.E. Miller, J.L. Eigenbrode, R.E. Summons, A.E. Brunner, et al., 2015, Organic molecules in the Sheepbed Mudstone, Gale Crater, Mars, *Journal of Geophysical Research: Planets* 120:495.

#### INSTRUMENTATION

aromatic hydrocarbons with masses of up to about 100 atomic mass units, which she said may be macromolecular material. A similar thing is seen for  $C_1$  to  $C_4$  alkyl hydrocarbons, but they are relatively weak signals. Eigenbrode said that this suggests that a macromolecular species (or something else very refractory) is undergoing pyrolysis. If you add in ionizing radiation and metal catalysts, which are known to be present because the material is basaltic, a Fenton-like reaction is possible. When macromolecular material is broken down, smaller, oxygenated molecules are made. These can then be chlorinated to produce  $C_1$  to  $C_4$  chlorinated molecules and chlorobenzene, which are equivalent to the types of molecules detected during thermal desorption.

The Mojave2 sample is at the bottom of the Lower Mound outcrop at Gale Crater. EGA shows large peaks from  $C_1$  to  $C_4$  and potentially even  $C_5$  alkyl hydrocarbons at high temperature. The possibility of nitrogen and oxygen atoms, however, makes this difficult to interpret. The single-ring, aromatic hydrocarbons, such as chlorobenzene and toluene, also display peaks. Organic sulfur volatiles are also detected, such as thiophene, methanthiol, and dimethylsulfide, which are confirmed by GCMS. These are not seen in the Cumberland blank sample. Eigenbrode performed this same analysis on the Tissint martian meteorite, which fell in Morocco in 2011, and found consistent results.

Eigenbrode's conclusion is that refractory organic matter is present on Mars. The source of this material, she said, is still unknown. It could have an abiotic igneous or hydrothermal origin, which has been suggested for the Tissint meteorite. Another potential source of organic matter on Mars is meteorites. After erosion, the organic material could then be concentrated in lake beds. A biological origin is also possible if the material was heavily processed, since heavier molecules were not seen. Meteorite impacts or irradiation might have broken down this material into smaller molecules. The implication of her conclusion is that organic matter might be widespread on the martian surface and in the rock record. The fact that this was discovered in a lake bed supports the hypothesis that the lake in Gale Crater was potentially habitable ~3.6 Gyr ago. Looking forward, Eigenbrode said that these organics could help support habitability on Mars now and in the future.

## **Audience Participation**

An audience member asked about the presence of nitrogen compounds on Mars, or lack thereof, and what this might imply for life. Eigenbrode said that there are nitrates on Mars.<sup>2</sup> However, they were not able to distinguish whether or not the organic material contains nitrogen, especially for the expected  $C_1$  to  $C_5$  hydrocarbons.

According to Dr. Ben Clark's earlier talk, the Viking pyrolysis stopped at 500°C and missed these organic signals. A conference participant then asked why the SAM limit seemed to be 825°C, implying that more interesting data could be revealed at higher temperatures. Eigenbrode answered that they can go to higher temperatures; they hit 860°C regularly. However, it takes a lot of power to go higher. With limited power resources and insufficient initial results to demonstrate the need to go hotter, they usually choose not to. The audience member then asked what the results might be if they went to consistently higher temperatures. Eigenbrode said the amount of organics might be increased if they were trapped in minerals that only break down at hotter temperatures. She then said that this might be an important process on Mars.

In the energy crisis of the 1970s, one audience member noted, the United States put a lot of money into coal structures, which would contain thiophene. He then asked how much coal could diagenize before its biological origin would no longer be apparent. Thiophene, he also noted, is found in some meteorites. The audience member finds thiophene interesting, since it is a diagenesis product of sulfur-containing biology. Eigenbrode responded that thiophene is usually formed by a  $C_4$  structure. In the presence of sulfur materials and at different pH or temperatures, you can sulfurize diene into thiophene. Thiophene is found in abiotic material as well as biological materials. She therefore thinks thiophene is not a good biosignature. However, Eigenbrode said, they also found methylthiophene, a  $C_5$  molecule, and they know that they didn't bring any  $C_5$  molecules with them on SAM.

Another member of the audience then asked what her message is to Gil Levin, the principal investigator of the Viking labeled-release experiment, who in 1997 claimed (and continues to claim) that the 1976 Viking Lander

<sup>&</sup>lt;sup>2</sup> J.C. Stern, B. Sutter, C. Freissinet, R. Navarro-González, C.P. McKay, P.D. Archer, Jr., A. Buch, et al. 2015, Evidence for indigenous nitrogen in sedimentary and aeolian deposits from the Curiosity rover investigations at Gale Crater, Mars, *Proceedings of the National Academy* of Sciences of the U.S.A. 112:4245.

positively detected microbial life on Mars. Eigenbrode answered by saying that GCMS was the right approach for Viking and that the labeled release experiment was a genius idea. An audience member then asked how she would interpret the labeled-release experiment. Eigenbrode said that she doesn't think that her results have any impact on it. However, degraded products, such as smaller carbon compounds like the chlorohydrocarbons in the Cumberland sample, could have easily oxidized under different types of conditions, such as the labeled-release experiment.

A workshop participant then asked what the concentration of organic material is in concretion-rich martian soil and in the mudstones. Eigenbrode said that the answer is still a work in progress, but that it is in the parts per million range.

One participant at the workshop then asked what kind of instrument Eigenbrode would like to send to Mars next in order to study the organic material. She replied that she wants to learn how the organic molecules are preserved, what mineral associations they are contained in, and how they got in them. One way to answer this question, she said, is to take the refractory organic material, break it up, and get more mineralogical information out of it. Another way is to look at the material with spectroscopy. It could even require sample return. She is concerned, however, that the organic material might be heterogeneously distributed, so that follow-up missions might not see organics depending on the technique and the sample location. Eigenbrode wants more details on the structure and the context of the organics.

A member of the audience then asked how they know that the peak at 825°C is not contamination from Earth and asked if that experiment has been done on soil or minerals on Earth to see if similar peaks occur. Eigenbrode said that the Rocknest sample was analyzed four times, two of which showed the peak. Prior to those tests, they did a blank that used just the instruments. The peak also was not seen in other samples. This shows heterogeneity on the martian surface. They tried looking at this material with a GCMS, but because the temperature is so hot, the particles were hard to trap, especially with a sample containing a lot of chlorine and sulfate, which alter the trapping conditions. Therefore, the level of trapped material may not have been sufficient for detection. They did see some chloromethane, but are not sure what the origin was or whether it could be from the instrument. As far as Earth-based tests, she has tried, but has been unable to reproduce the signal. They have maybe found CO,  $CO_2$ , and  $CH_4$ , but nothing with more carbon atoms. Every other sample they have tried has exhibited traditional behavior with only two peaks: thermal desorption and pyrolysis.

Lastly, a participant at the workshop then asked how to get around the problem of perchlorate chemistry when heating the sample. Eigenbrode said that, in order to avoid the oxidation of organics by the oxygen produced by perchlorate when heating, you have to stick the oxygen somewhere, such as providing other materials for the oxygen to latch onto, like strongly alkaline-like materials or reductants to buffer the oxidation. She thinks that the tetramethylammonium hydroxide (TMAH) experiment on SAM could diminish perchlorate's effects, but they haven't yet run those experiments with martian samples. Lastly, Eigenbrode offered another strategy, which would be to do a two-step process: (1) heat up the sample to lower temperatures to drive off the perchlorate oxygen, and (2) continue to higher temperatures to examine the pyrolysis aspect of the experiment.

## Future Directions: Report of Breakout Groups

John Grunsfeld, a former astronaut who until April 2016 served as associate administrator of NASA's Science Mission Directorate (SMD), moderated the session on future directions for the search for alien life and the report-outs from the breakout groups that met for 2 hours earlier in the day. Grunsfeld started by explaining why he had asked the National Academies of Sciences, Engineering, and Medicine to organize this workshop. Grunsfeld wanted to know about our origins here on Earth and our place in the universe. He finds the question of whether we are alone, or if there is life beyond Earth, to be truly compelling—and a question which can be answered by scientific inquiry. In order to delve deeper into this question, he wanted to bring together scientists from many different disciplines and communities. Grunsfeld said that one takeaway message from this workshop was that the search for life in the universe would "make NASA great again."

Grunsfeld then introduced the four breakout groups, each of which, while not offering consensus summaries, reported back their key points to the wider audience. These four breakout groups were as follows:

- In situ detection of life as we know it,
- In situ detection of life as we don't know it,
- · Remote detection of life as we know it, and
- Remote detection of life as we don't know it.

The question each group had been given in order to guide them in their discussion was the following: How could targeted research over the next 5 to 10 years help advance the state of the art for life detection, including instrumentation and precursor research?

## IN SITU DETECTION OF LIFE AS WE KNOW IT

Tanja Bosak of the Massachusetts Institute of Technology (MIT) and Nita Sahai of the University of Akron led the breakout group focusing on the in situ detection of life as we know it. Bosak began by saying that significant developments have been made in the field of DNA sequencing and analog environments. Current instrumentation, she said, could be used in upcoming missions on the in situ detection of life.

She then described some plans on the horizon: the Mars sample return mission, its associated analytical tools, and a holographic microscope. Planetary targets for the next generation of missions are also clear: Mars, Venus, and the icy ocean moons.

Most of the time in this breakout group was spent figuring out what they need in order to perform an in situ detection of life. A microscope was at the top of the list. There was also a strong emphasis on equipment to extract and handle samples. They would also like deep drills in order to test the material below what has currently been examined. Tools to assist in the capture and analysis of sprays (i.e., plumes) were also desired. Instruments that can perform clumped isotope analysis were also mentioned, although Bosak said that some already exist that could analyze the returned martian samples. She then gave a list of molecular types that an instrument should be able to detect: native fluorescents, adenosine tri-phosphate (ATP), dipicolinic acid (which is in spores), lipids, and more. These instruments should be cable of capillary microchip electrofluorosis and fluorescent immunoassay experiments. Any instrument sent into space must also be radiation hardened to prevent degradation, especially on longer missions. Other types of instruments that some members of the breakout group desired were an aerial sampling mission in Venus's atmosphere and subsurface water detection on other bodies.

The breakout group also emphasized the necessity for precursor research to develop in situ detection of life. Of critical importance for life detection are positive and negative controls, a better understanding of false positives and false negatives, and how to detect different types of species and molecules related to life (and what they would even be).

Bosak then discussed the possible approaches to detecting life. The detection of life requires a spatially resolved analysis that combines multiple techniques. In a sample return mission, particularly from the outer solar system, the issue of how to preserve ices needs to be solved. Any sample return mission needs to have multiple laboratories independently analyze the samples as a hedge against false positives or negatives. Any mission focusing on the detection of life must also have collaboration between the instruments being flown. The discussion within the breakout group identified the need for funding to allow for such collaborations; currently, there is too much emphasis on competition among instrument teams. She also conveyed a desire that funding should be available for instrument development to increase the technology readiness level (TRL) and explore NASA Innovative Advanced Concepts (NIAC). Missions should also have greater mobility on the surfaces of extraterrestrial bodies. Conversely, they also want the ability to sample the same place at multiple times to test for temporal variations.

Sahai then summarized the wish list. Effectively, the breakout group wants to be able to quickly analyze a lot of samples and then triage them to pick out the specific samples that require more detailed analysis. A suite of biosignature analysis instruments would then extensively analyze the selected samples.

## IN SITU DETECTION OF LIFE AS WE DON'T KNOW IT

Steven Benner of the Foundation for Applied Molecular Evolution and Mark Thiemens of the University of California, San Diego, led the breakout group on in situ detection of life as we don't know it. Benner started out by saying that their breakout group first tried to figure how and where to look for life. Two camps arose, the people who want to "follow the water" and the people who want to "follow the carbon."

The breakout group then tried to come up with a set of attributes they value in calling something "life." One example is compartmentalization (or isolation) as a universal feature of life. For life as we know it, compartmentalization means a structured membrane. However, they decided that this was too constraining, so they chose to generalize the definition of compartmentalization to isolation such that Darwinism could still occur. Isolation is required both for controlling the flow of energy and for Darwinism. It allows for replication while preventing both parasitism and losing important biochemical molecules. Another feature of life is that it contains information. Other attributes life has are motion, the ability to use energy, and being in a community of life.

Benner said that there were three steps in detecting life: finding it, seeing it, and determining its composition. However, technical problems are associated with finding an isolated system, since we do not really know what could be allowed. As already mentioned, they decided to focus on looking for carbon and water as (likely) universal requirements for life. After finding life, they would want to see it do something. This can also be problematic. For example, Brownian motion of a particle could be easily deceiving. Benner said that maybe we would have to see

#### FUTURE DIRECTIONS: REPORT OF BREAKOUT GROUPS

it do something more interesting, like dividing and replicating. He also worried that we might accidentally kill any life we found due to our ignorance of how to keep it alive, although that could also be used to check whether it was alive in the first place. Benner said that a good idea coming from the discussion would be to just take a sample of it and determine its entire molecular inventory.

Testing for life also depends on what kind of life it is—micro versus macro and extant versus extinct. Macro life would probably be pretty easy to find no matter what it was made of. Finding extant microbes might be possible. The biggest difficulty, of course, would be detecting extinct microbes.

The last topic Benner touched on was the possibility of searching for weird life on Earth. A potential type of compartmentalization, Benner said, was a mineral hole in a rock. There is no known life that goes into one of these holes and then gives up its membrane layers and chooses to instead use the hole itself as its compartment. We could search for that kind of life here on Earth, so maybe it should be done, especially considering it's easier than doing the same experiment on Mars or another body.

They also discussed the genetic information molecule and the energy source. He said that everybody agreed that a thermal equilibrium is necessary. The energy used by known life is chemical or visible light. They therefore discussed abnormal sources of energy for life, such as ionizing gamma radiation. This type of life might also be extant on Earth, so he said that it would be best to search for it here first. Benner said that they agreed that a von Neumann automaton (capable of self-replication) should be classified as living or, at the very least, a biosignature.

Benner then finished by saying that any planet with life would likely have a biosphere filled with different species. Patterns and textures created by this biosphere could itself be a biosignature.

## **Audience Participation**

A member of the breakout group then said that the list of general attributes required for life was formulated in the context of ancient life on a rocky planet. They then tried to expand the list to be inclusive of extant life, extinct life, and life on icy worlds too. She then went through the list. First they rejected the idea of a cellular morphology in favor of the compartmentalization or isolation morphology. They also wanted to eschew the idea of just organic matter being a biosignature, instead wanting to look for organic molecular compositions that indicate selectivity, patterns, and complexity. The mineralogy of the body is also important. The traditional mineralogy required for life, she said, needs to be broadened. For example, she said that iron metal or sulfate ions on Europa would be a potential biosignature. She called this "contextual chemistry and structures." Biofabrics are another attribute of life, but on an icy world, one might expect cellular aggregates instead. One item on the list, isotopic knowledge, was controversial as to whether it was important in terms of determining biological processing. The breakout group also listed activity (in terms of metabolism, motility, and reproduction) and information carrying molecules. She said that this list is designed so that future missions could select for instruments than can address each of these points. Preferably, each point could be addressed by multiple instruments independently to ensure the validity of the results. Another audience member then agreed that missions should focus on these, even those missions searching for life as we know it.

## **REMOTE DETECTION OF LIFE AS WE KNOW IT**

Vikki Meadows of the University of Washington and Sushil Atreya of the University of Michigan were the leaders of this breakout group. Edward Schwieterman of the University of California, Riverside, summarized the breakout session. He started with what we already know. He quoted Grunsfeld, "Atmospheric spectroscopy would show external observers that Earth is inhabited." At a minimum, therefore, the goal should be having the ability to detect and recognize Earth life. Schwieterman said that the discussion fit into two categories: precursor science/ interpretation and technology/engineering.

There was consensus within the breakout group that more work needs to be done to determine the ratios of trace gases that can be biosignatures, but which can also be produced abiotically (e.g.,  $CH_4$ ,  $O_2$ ,  $O_3$ , and  $N_2O$ ). This requires knowledge of the environmental context to avoid false positives that could be caused by geochemical

or photochemical processes.<sup>1</sup> A broader environmental context requires knowledge of the planetary architecture and correlations between different parameters, which could inform the interpretation of biosignatures. In atmospheres with a potential biosignature gas (e.g.,  $O_2$ ), certain other gases could instead indicate an abiotic origin. Schwieterman also said that the feasibility of using isotopic measurements needs to be explored (e.g.,  $^{13}C/^{12}C$  and D/H ratios), including how to interpret the results. However, this would require very-high-resolution spectroscopy (R ~ 100,000). Biosignature gases might also have seasonal changes modulated by life, which could potentially be measured. Clouds and aerosols might obscure biosignatures, but they also provide information about the planet's geophysical and atmospheric processes and the planet's potential habitability.<sup>2</sup> Another factor in a planet's habitability is tectonic and volcanic activity. Sulfur gases, he said, could be used to infer these properties about the planet.<sup>3</sup> Polarimetry could also be informative in terms of both biological chirality and scattering processes in the atmosphere.<sup>4,5</sup>

Schwieterman then listed a number of technological advancements that are required to enable or enhance the scientific return of future missions. The wavelength range is one of the most important properties of any instrument. It determines which molecules you can detect. Because many molecules have overlapping spectral lines and bands, multi-band measurements should be pursued, particularly if done alongside low-resolution spectroscopy. Detector technologies and telescope size limit the wavelength range accessible for each target (which is also a function of the angular separation between planet and star), so new technologies or larger telescopes would expand the number of observable targets.<sup>6</sup> The noise sources, such as exozodiacal light, also vary as a function of wavelength. Improvements to cooling technology would keep thermal noise down in the near- and mid-infrared, enhancing the science return at these wavelengths.

Another technological advancement that could increase scientific return is high-resolution spectroscopy ( $R \sim 10,000-100,000$ ), which is necessary for uniquely fingerprinting molecules (and especially isotopes) and their mixing ratios. Pushing high-resolution spectroscopy into space requires miniaturization of existing and developing technology. This will be difficult and expensive. A potential partial solution is to use high-resolution facilities on the ground and complement them with lower-resolution instruments in space.<sup>7</sup>

The group also suggested that more advanced technologies should also be pursued. A photon detector that can resolve energies in the ultraviolet, visible, and infrared regions of the spectrum could vastly reduce the noise and technical hurdles of data reduction. Coronagraph technology, currently under development, has a throughput problem. Only 1 to 3 percent of the total light gets through. This also needs to be improved. Lastly, the breakout group discussed the technical aspects of polarimetry.

## **Audience Participation**

A member of the audience then asked whether they thought that Venus-like exoplanets could be observed in the search for life with near-future technologies. Schwieterman said in response that he hopes that the James Webb Space Telescope (JWST) will get some transit spectra of Venus-like worlds, considering that there is a (geometric

<sup>&</sup>lt;sup>1</sup> S.D. Domagal-Goldman, A. Segura, M.W. Claire, T.D. Robinson, and V.S. Meadows, 2014, Abiotic ozone and oxygen in atmospheres similar to prebiotic Earth, *The Astrophysical Journal* 792:43.

<sup>&</sup>lt;sup>2</sup> G. Arney, S.D. Domagal-Goldman, V.S. Meadows, E.T. Wolf, E. Schwieterman, B. Charnay, M. Clare, E. Hébrard, and M.G. Trainer, 2016, The pale orange dot: The spectrum and habitability of hazy Archean Earth, *Astrobiology* 16:873.

<sup>&</sup>lt;sup>3</sup> L. Kaltenegger and D. Sasselov, 2010, Detecting planetary geochemical cycles on exoplanets: Atmospheric signatures and the case of SO<sub>2</sub>, *The Astrophysical Journal* 708:1162.

<sup>&</sup>lt;sup>4</sup> W.B. Sparks, J.H. Hough, L. Kolokolova, T.A. Germer, F. Chen, S. DasSarma, P. DasSarma, F.T. Robb, N. Manset, I.N. Reid, F.D.

Macchetto, and W. Martin, 2009, Circular polarization in scattered light as a possible biomarker, *Journal of Quantitative Spectroscopy and Radiative Transfer* 110:1771.

<sup>&</sup>lt;sup>5</sup> J. Takahashi, Y. Itoh, H. Akitaya, A. Okazaki, K. Kawabata, Y. Oasa, and M. Isogai, 2013, Phase variation of Earthshine polarization spectra, *Publications of the Astronomical Society of Japan* 65:38.

<sup>&</sup>lt;sup>6</sup> C.C. Stark, A. Roberge, A. Mandell, M. Clampin, S.D. Domagal-Goldman, M.W. McElwain, and K.R. Stapelfeldt, 2015, Lower limits on aperture size for an exoearth detecting coronagraphic mission, *The Astrophysical Journal* 808:149.

<sup>&</sup>lt;sup>7</sup> M. Brogi, M. Line, J. Bean, J.-M. Désert, and H. Schwarz, 2016, A framework to combine low- and high-resolution spectroscopy for the atmospheres of transiting exoplanets, submitted to *The Astrophysical Journal Letters*, arXiv preprint, arXiv: 1612.07008.

transit probability) bias towards finding them compared to Earth-like worlds. A direct image of a Venus-like world, however, would be more difficult due to the small angular separation and inner working angle constraints. Meadows then also said that a potential super-Venus (Venus-like world, but with a larger radius), GJ 1132 b, will be one of the first targets of JWST. Schwieterman said that one challenge with the spectroscopy of Venus-like worlds will be the small scale height ( $H = (kT)/(\mu g)$ ) due to the high mean molecular weight ( $\mu$ ) of the atmosphere. The scale height determines the magnitude of the transit features, which would be about 20 times smaller for a CO<sub>2</sub>-dominated atmosphere ( $\mu = 44$  g/mol) than an H<sub>2</sub>-dominated atmosphere ( $\mu = 2$  g/mol). The most promising molecular bands for characterizing Venus-like worlds are the 4.3 and 15 micron CO<sub>2</sub> bands, which are in the range of the JWST Near-Infrared Spectrograph and Mid-Infrared Instrument, respectively.

## **REMOTE DETECTION OF LIFE AS WE DON'T KNOW IT**

William Bains of MIT and John Baross of the University of Washington led the breakout group to discuss the remote detection of life as we don't know it, otherwise known as "weird life." Bains said that the group started out discussing just how weird to get, deciding against the extremely weird life, such as life made out of neutronium or interstellar clouds. They instead used the National Research Council (NRC) report *The Limits of Organic Life in Planetary Systems*,<sup>8</sup> which has become known as the "Weird Life Report," as their framework. They thus focused on carbon- and water-based life. Two new ideas since that report, Bains said, were a discussion of energy sources as a precursor to life and a growing realization of the importance of the statistical background information on whether a signature is biological.

Instead of looking at the mechanism of how biosignatures are created, the group decided to look for inputs and outputs. In that sense, there was a question of whether life's possible weirdness was even relevant. A lot of what the group would want to look for to discover weird life would be equally valid as a sign of normal life as well, with a few exceptions.

However, weird life allows for an expansion of the definition of "habitable." The habitable zone could be much wider and weirder. Examples include a planet outside the conventional habitable zone with a large greenhouse effect from  $H_2$ , an ultra-cold ocean world (e.g., an ocean composed of water plus ammonia and salt), and a very hot world with a few habitable locations (e.g., the clouds of Venus). This wider range of planets allows for a wider range of the planetary system's possible architecture and evolution. However, Bains said that the search for life doesn't have to happen only on other bodies. Earth itself could harbor weird life.

Bains then outlined some research goals suggested by the group. One broad category was to move away from looking for just an Earth-like world in an Earth-like orbit around a Sun-like star, but to look for other combinations of planets and environments that could support life. It is not enough to just identify geochemistry of these alternative types of planets. Inputs, outputs, and rates of production also need to be modeled to check for detectability.

Another research goal is to further explore energy capture, specifically the relationship between photon flux, energy per photon, plausible photon capture mechanisms and efficiencies, and oxidants and reductants available in the environment. This relates to looking for a "blip" in the data at certain wavelengths. The terrestrial "red edge" is just one example of such a blip. Any dips, edges, peaks, or other blips need to be examined in the search for life. A biological origin of a weird blip might be ruled out in this way. Revisiting early Earth could be a useful exercise to explore whether different photosynthetic or energy capture processes were used. Bains then again emphasized that there could be a type of life here on Earth using a radically different source of energy, such as thermal, magnetic, or mechanical energy (although previous sessions had been skeptical about these).

An obvious sign of life would be any sort of technosignature, a biosignature that requires technology, such as gases that are very unlikely to be formed naturally. Other indications of life would be large-scale differences from what is expected. Bains gave an example of a Mars-sized planet in a Mars-like orbit, but with the climate of Los Angeles. Even more bizarre examples of technosignatures include rearranging planetary systems, Dyson spheres, Alderson disks, von Neumann probes, and machine civilizations.

<sup>&</sup>lt;sup>8</sup> National Research Council, 2007, The Limits of Organic Life in Planetary Systems, Washington, D.C.: The National Academies Press.

## **Audience Participation**

A member of the audience then said that hot Jupiters are weird and wondered if that could be evidence of an advanced civilization. Bains said that the idea was interesting, but that the planetary migration people would consider natural (i.e., non-technological) explanations for planetary migration to be more plausible. Another audience member then emphasized that they do not have a "life between the gaps" approach to weird life. He compared their approach to how climatologists proved not just that climate change is happening, but that it's anthropogenic. Basically, they proved that it was anthropogenic by showing that robust, trustworthy models of the global climate could only represent reality when anthropogenic-induced warming was included. He said that the astronomical and astrobiological communities need a similar set of robust, trustworthy models that only invoke biological processes when all other explanations are insufficient to explain the data.

Another commenter then brought up Venus again. He said that we still cannot explain how Venus became the way it is today, suggesting that maybe life caused the changes.

## GENERAL DISCUSSION

Grunsfeld continued as the moderator for the general discussion portion. (The text in this section is not necessarily in chronological order. Comments have been moved out of chronological order to improve flow and preserve continuity of thought.)

## Microscopy and Cellular Morphology

Grunsfeld started off the general discussion by bringing up microscopy, specifically on Mars. The Curiosity rover has a microscopic camera called the Mars Hand Lens Imager (MAHLI).

Two discussion items the first audience member brought up came from the NRC report Signs of Life<sup>9</sup> (2002), which identified microscopy as a technology that needed further development. Microscopy at the <1 micron level still needs to be developed, she said. The other discussion item was to move beyond single-purpose instruments and instead move toward instruments that combine multiple experiments for biosignatures or that allow for the chemical analysis of specific samples identified through microscopy. Another commenter quipped that that's why we just need to send astronauts there to do analysis in situ or at least in a nearby analytical laboratory.

Another member of the audience talked about imaging new places in greater detail. Every time we have done so, we have discovered amazing new and unexpected things. Only once we see these new things can we start theorizing about them and developing new experiments. He also said that imaging is important for public support. He thought that the Apollo image of Earth taken from the Moon was especially powerful. Furthermore, he said, continuing to image new things, big or small, is important for maintaining public support. Grunsfeld specifically noted the Hubble Space Telescope's role in this. Then another audience member brought up microscopy in the same sense, noting that high-resolution spectroscopy has allowed for the imaging of single molecules, and saying that efforts should be focused on making more powerful microscopes too.

A workshop participant said that, following the claim being made that the martian meteorite Allan Hills 84001 contained signs of martian life, there has been a fear of using, talking about, or searching for morphology as a biosignature on Mars. She thought that this is why microscopy has not been used enough and worries that people will continue to avoid using morphology as a sign of life. Another audience member agreed and was puzzled why microscopes haven't been used. Not only can they increase scientific information, but he said that the public would love to see beautiful, microscopic images. Then another conference participant agreed with the need for microscopes, but said it needed to be combined with chemical analyses of the same samples.

At the Biosignatures of Extant Life on Ocean Worlds Workshop earlier in the year, one participant from that workshop said that everybody just wanted to see a cell. However, morphology (i.e., looking like a cell) was not enough. They also needed to know its chemical and/or molecular composition, its structure, and what it does.

<sup>&</sup>lt;sup>9</sup> National Research Council, Signs of Life: A Report Based on the April 2000 Workshop on Life Detection Techniques, The National Academies Press, Washington, D.C., 2002.

#### FUTURE DIRECTIONS: REPORT OF BREAKOUT GROUPS

A member of the audience then continued, saying that there is much more you can do with microscopy than just taking a monochrome image of a cell. Simple additions can detect things like proteins, lipids, saccharides, auto-fluorescence, chemotaxis, and index of refraction. Then he said that the microscope should input samples into a chemical analysis instrument.

Agreeing with the previous commenter, another workshop participant said that it was a good first step. However, all microbial morphology is dominated by the physical forces at the micro scale. This means that cells are almost always spheres or tubes. Additional information beyond just morphology must be extracted to conclude whether it is life.

In more exotic environments, such as below ground, another audience member said that cells can have a variety of very complex shapes (e.g., chrysanthemum or polygonal). Surface microbes, she said, might have environmental selection effects pushing them to be spheres or tubes, but more protected microbes could have a more diverse set of shapes. She then agreed with earlier comments for the need of an instrument capable of doing a variety of analyses on the same sample either simultaneously or one immediately after the other. Another workshop participant agreed. He said that we need to consider that cells could have irregular shapes and could be very sparse. What is really needed, he said, was an algorithmic search that could scan the entire field and look for repeated shapes that it could then make available to the instruments.

A microbiologist in the audience who has worked on microscopy in extremely dilute environments (and selfproclaimed "Debbie Downer") said that a cell being 1 micron large was optimistic. She said that they're usually about 0.1 to 0.2 microns or could even be smaller in oligotrophic environments. She also said that the pretty visualizations of cells on Earth have already been filtered and stained. Cells aren't visible with a regular microscope. She also said that morphology is difficult to determine when it is just a dot of light. If the cell is in water, there is a chance that the cells could be filtered out. She studied life in the Vostok ice core and had to filter at least 100 ml—and often more, up to a full liter—of water to be able to count them with a microscope and extract their DNA. Even looking for things like auto-fluorescence is not a strong biosignature as minerals can exhibit the same property. In sediments on Earth, they have abandoned using microscopes in favor of using rigorous extraction techniques instead. She did agree that you would need to do a chemical analysis experiment on the same sample of material that was put under a microscope. However, she said that this would be very difficult to do, which is why she thinks we need astronauts on Mars to do it.

## Weird Life

Shifting to the topic of weird life, another participant in the workshop wondered about the possibility of life on Titan. He said that there has been discussion about life using acetylene and hydrogen as nutrients and existing in hydrocarbon fluids instead of water. Another audience member said that it would necessarily be very cold life. Therefore, getting complex molecules dissolved into a solution would be almost impossible. The diversity of molecules would be limited only to small molecules. The solubility just isn't there. Another member of the audience continued this discussion, saying that there was an entire group dedicated to potential life on Titan. For example, the solubility of argon in methane at 95 K is extremely low. He said that in the report Signs of Life<sup>10</sup> a liquid solvent was deemed to be required for life as we know it. Life in the gas phase has issues of gravitational collapse versus dispersion, while life in the solid phase would be very slow because of its metabolism. He said that there could be life in the Oort Cloud living on the occasional photon, but it would be slow. Going back to Titan, he said that practically nothing dissolves in its surface hydrocarbon lakes, but he did say that its subsurface water-ammonia ocean would be more favorable for solubility and therefore life. In general, however, cryosolvents are bad for life because they're cold. The audience member posing the initial question about life on Titan then asked whether a water ocean with 5 to 10 percent ammonia was problematic. The other audience member then responded that it would not be a problem. A strong acid or base deprotonates thymine and guanine so that they can no longer bind to adenine or cytosine, respectively. Ammonia, however, is not that basic, having a pH of only about 10 to 12.

A member of the audience then brought up a 2016 study by Kan et al. in which proteins from an Icelandic bacterium were used to coax microbes into producing a carbon-silicon bond in a process called directed evolu-

<sup>&</sup>lt;sup>10</sup> NRC, Signs of Life, 2002.

tion.<sup>11</sup> He then asked whether it was feasible that there could be silicon-based life. If the answer is yes, he asked if there is a way to model or predict biosignatures of silicon-based life. A member of the audience then responded by saying that you first need to decide whether we are carbon-based life. Elaborating, he said that a peptide's backbone is C-C-N-C-C-N-C-C-N and that DNA has chains of O-C-C-C-O-P. More elements than just carbon are needed. He then said that the Kan et al. study set up a high-energy process that allowed for the C-Si bond to form. The fact that the result was a C-Si bond makes it difficult to say whether it is a carbon- or silicon-based life. He said that the study was still a spectacular result though. If, however, you wanted to make silicon-based life with a backbone of repeating silicon atoms, these molecules are already known. However, because the d orbitals of silicon conduct electricity, these molecules are quite unusual. This kind of silicon-based life is unconvincing. However, he thinks that a Si-C-C-O kind of life would be productive. The Kan et al. (2016) paper is a very reasonable way to get this C-Si bond. The problem with silicon-based life is how much silicon is present. The more silicon in life, the more the life deviates from the natural chemical reactivity we're familiar with. He then said that Earth life isn't really carbon-based; the interesting things life does are actually done by the nitrogen and oxygen associated with the carbon.

The key point of Kan et al. (2016), another audience member said, was that it used Darwinian evolution. The selection criteria were set artificially, but the rest of it was Darwinian mutation and selection. It shows that evolution can produce a C-Si bond given the right environment. Another conference participant replied saying that the experiment provided an evolutionary pressure on an enzyme that already could create C-Si bonds (but extremely inefficiently) and evolved it to make it much more efficient (although still pretty inefficient). A key point, he said, was that the starting materials were artificially selected. They fed the bacteria silanes instead of silicates. It is a reaction that happens on its own, and they took an enzyme that already catalyzed it and made it more efficient. Enzymes exist that can handle more than C-Si bonds. Other enzymes can handle carbon-fluorine bonds, carboniodine bonds, and even one that makes cyanide-bromine bonds. However, it needs an evolutionary pressure to create these bonds. The carbon-fluorine bond, for example, creates toxic molecules in plants that kill animals that eat them. If there were an environment where a C-Si bond was advantageous and no other sort of bond was, he absolutely thinks that there would be life making C-Si bonds there.

## The Term "Biosignature"

A participant in the workshop then said that it is imperative that the community continues and increases communication with Congress, the public, and the world about astrobiology. As such, she feels that the word "biosignature" has been used in too many different and ambiguous ways. It's usually referred to as a *possible* signature of life by scientists, not a definite signature of life. However, the public and policymakers interpret "biosignature" as being a definite sign of life. In this light, she thinks that the word "biosignature" should be abandoned and replaced by something that is clearer.

This same issue was covered about 10 years ago, according to one workshop participant. There, they decided on two terms: "biosignature" and "potential signature." A biosignature is a definite sign of life, while a potential biosignature is a feature of interest that demands further investigation. He said that he thinks it would be a mistake to completely abandon the term "biosignature."

The "Exoplanet Biosignatures Workshop Without Walls" hosted by the Nexus for Exoplanet System Science (NExSS) and the NASA Astrobiology Institute coined the term "biohint," according to one audience member who had gone to both workshops. He feels that these terms address this issue appropriately. The general sense of the assembled audience was that more thinking is needed to come up with a term that conveys the uncertainty of "potential biosignature" for the interested public.

An audience member then finished the general discussion with a thought that it would be a great idea to design an app like the Star Trek tricorder to educate people on what biosignatures/biohints were. The general public could use it to learn about potential habitable environments or look at trace gases and minerals in the context of potential biosignatures.

<sup>&</sup>lt;sup>11</sup> S.B.J. Kan, R.D. Lewsis, K. Chen, and F.H. Arnold, 2016, Directed evolution of cytochrome c for carbon-silicon bond formation: Bringing silicon to life, *Science* 354:1048.

## Wrap-Up

James Kasting, from Pennsylvania State University and chair of the workshop's organizing committee, was charged with summarizing up the workshop. Kasting began by explaining, from his perspective, what the National Academies of Sciences, Engineering, and Medicine wanted from the workshop. Desired outputs from this workshop were answers to the following two major questions:

• What are the key scientific and technological challenges in astrobiology as they relate to the search for life in the solar system and exoplanetary systems in the next decades?

• To what extent will current and planned NASA missions, such as the Transiting Exoplanet Survey Satellite (TESS), the James Webb Space Telescope (JWST), the Wide Field Infrared Survey Telescope (WFIRST), Mars 2020, the Europa Clipper (and a possible Europa lander), international missions, ground-based telescopes, and other facilities, play in addressing the key questions relating to the search for life in the solar system and exoplanetary systems?

## SUMMARY OF CHAPTER 1: SETTING THE STAGE

Kasting then summarized the sessions one by one and asked audience members to chime in with any additional remarks. In the first session, it was said that the search for biosignatures must also consider both the origin of life and the maintenance of life. John Baross said that hydrothermal vents may have played a role in the origin, or at least the early evolution, of life. Baross also said that serpentinization was a key process for providing the hydrogen, trace metals, and surface area needed for life.

Kasting then named free energy as an important consideration in the context of searching for life. Tori Hoehler said that this is the most fundamental requirement for life. Hoehler also said that life as we know it only uses redox chemistry (the transfer of electrons), as opposed to other sources of free energy. A member of the audience then clarified that light energy in photosynthesis is used, but the light energy just gets electrons moving. Life relies on transfer of electrons; light provides additional energy for these electrons in some forms of biosynthesis. Kasting then said that Eric Smith's presentation stated that free energy gradients are also an essential factor in the metabolism-first origin of life theory.

Another key point was raised by Baross. He had stated that our understanding of the evolutionary relationships between extant organisms is still changing. Baross showed that the ribosomal RNA (rRNA) tree may contain only

two domains instead of three, with eukaryotes included in the Archaeal domain. This has implications as to the nature of the last common ancestor, which Baross said might have been a methanogen.

## **Audience Participation**

A member of the audience said that he would like to see a greater emphasis placed on understanding mineral catalysis, particularly for minerals composed of cations that are associated with some of the most ancient enzyme proteins (e.g., molybdenum and tungsten). A greater understanding of boron is also necessary, considering that some oceanic serpentinizing environments had a high concentration of boron. Related to that, the audience member thinks that the origin of life is the most fundamental question remaining in science. He wants to see scientists from every discipline working together to find the solution. He then made a prediction that an organism using a third energy source (not chemical or visible light) would be discovered on Earth, even if it was slow life, dividing once every tens of thousands of years. This prediction is the result of recent discoveries of Archaea growing on some of the lowest levels of energy sources ever imagined.

One important thing to worry about, an audience member said, is that complex order can depend on the details of the boundary conditions and on the substrate. Life, he said, fools us into thinking it can exist anywhere, but this is only because cells are protected by membranes, which have been tuned by a long period of Darwinian evolution. Because of membranes, a homogeneous biochemistry can exist in heterogeneous environments. If the details of the boundary conditions are important, understanding the organic geochemistry in the Hadean era and the earliest part of the Archean era should be a high priority.

## SUMMARY OF CHAPTER 2: HABITABLE ENVIRONMENTS IN THE SOLAR SYSTEM

Kasting said that one theme from this session was that Mars remains a possible abode for either extant or extinct life. John Grotzinger said that the Curiosity rover has provided new evidence for the repeated formation of long-lived lakes. There is also direct evidence from clays and magnetite that suggests that serpentinization-like reactions took place on Mars. Grotzinger thought that more small rovers were needed to explore a more diverse set of environments. Kasting then said that there is no consensus on how to explain the prolonged periods of warmth on early Mars.

Moving on to ocean worlds, Kasting repeated Kevin Hand in saying that the detection of life on an ocean world would almost certainly indicate an independent origin of life. Therefore, finding DNA there would imply a convergence towards a universal type of biology. Europa and Enceladus are particularly intriguing moons because of the presence of subsurface oceans. Many of the requirements for life are already present on ocean worlds: water, biologically important elements, water-rock interactions, and maybe even hydrothermal vents. Free energy is still an important consideration. Reduced materials could be provided by the mantle, while oxidants could be provided by crustal overturn. Kasting then wondered whether the free energy available was sufficient to create or sustain life on these icy ocean worlds.

Referring to Ellen Stofan's presentation, Kasting said that NASA has multiple different pathways that it might pursue in order to detect biosignatures. Stofan thought that humans can play a key role in looking for life on Mars, such as enabling deep drilling. Others, however, have argued that human exploration should be delayed due to its cost or to avoid biological contamination until after it has been studied more thoroughly.

## **Audience Participation**

A workshop participant clarified that serpentinization itself has not been found on Mars, but rather, a sort of iron redox chemistry where you take reduced iron in an olivine and transfer it to magnetite. This releases hydrogen gas. The latter process is similar to serpentinization, but is not the same thing. She then emphasized and built upon two of Kasting's other remarks on Mars. Not only has the Curiosity rover discovered long-lived lakes, it has also found evidence for even longer-lived groundwater. She then said that, with Curiosity's discovery of boron, all of the necessary trace elements for life have now been found on the martian surface.

WRAP-UP

Bringing up Venus again, a workshop participant opined that an important question that needs to be answered is why Venus never had life (assuming it didn't) despite having water. If life did exist, he wondered if it could have moved to Venus's potentially habitable atmosphere. Kasting replied that Venus could have life in the clouds, but he thinks it unlikely. Kasting instead thinks that the best theory is that Venus went into a runaway greenhouse effect during the accretion phase and never came out of it.<sup>1</sup> Kasting likes this because it gets rid of all the oxygen. However, other recent calculations have conversely claimed that there might have been water on the surface of Venus.

A member of the audience then wondered what could be done in the laboratory in terms of synthesizing biological molecules from prebiotic chemicals. For example, one type of RNA synthesis requires 1-molar dissolved phosphate. Another requires 25 milli-Molar (mM) dissolved borate. She does not know of any geological environments that are able to concentrate that much phosphate or borate. She then told the synthesizers to give her a scenario of a mineral exposed to an atmosphere with certain physical properties (e.g., temperature) and specified partial pressures of different molecules. She can plug these into well-established water-rock interaction-reaction programs to simulate weathering of the primary rock. These programs can give a rough idea of the ranges of solution chemistry as well as of any secondary minerals produced by the weathering process. Then the primary and secondary minerals are known, along with quantitative estimates of the ions. All the thermodynamic equilibria are then calculated for hundreds of minerals. Then, using this plausible range of ion concentrations, one could attempt a plausible synthesis experiment. Elements like boron will be found anywhere with a good detector, but one needs to also know its other properties, like its abundance and oxidation state.

## **SUMMARY OF CHAPTER 3: EXOPLANETS**

Kasting then moved on to exoplanets. He said that the remote detection of biosignature gases has been studied much more intensively in recent years. In Vikki Meadows' talk, she said that the combination of  $O_2$  and  $CH_4$ (or N<sub>2</sub>O) was still the best available remote biosignature.  $O_2$  by itself is ambiguous. However,  $O_2$  false positives could hopefully be identified from the planetary and environmental context of it. Thermodynamic disequilibrium by itself is not necessarily a biosignature because chemoautotrophic life (e.g., methanogens) would tend to drive an atmosphere towards equilibrium.

Describing William Bains's talk, Kasting said that we should be aware that life on exoplanets might be "weird." On a hydrogen-rich super-Earth, ammonia might be a possible biosignature, according to Bains. Humans cannot be too Earth-centric and focus only on life as we know it.

Kasting then said that the detection of life on exoplanets might start to become possible within the next few years. JWST, for example, might be able to do transit spectroscopy of an Earth-like star around an M dwarf. Nick Siegler had said in his talk that WFIRST will not find Earth-like planets, but it will find other non-transiting planets. It will also test either a space-based coronagraph or a starshade. Matteo Brogi in his talk said that, with the next generation of extremely large telescopes, Proxima Centauri b might be characterized from the ground.

### **Audience Participation**

Again opening it up to the audience, a participant in the conference agreed with an earlier point and said that the community had become sloppy with the term "biosignature." He cautioned again to use the word "possible" or "potential" before the term "biosignature."

Another member of the audience then suggested that the community move away from short, snappy descriptions of what a biosignature is. The entire environmental context must always be taken into account in order to avoid false positives. Furthermore, just the detection of a molecule is not enough evidence. The abundances are also important in order to discriminate its origin as being biological or abiological. Thermodynamic signatures, on the other hand, are more of a tool rather than a definitive thing. She said that the detection of thermodynamic disequilibrium only indicates the existence of a process that we still need to interpret and explain.

<sup>&</sup>lt;sup>1</sup> K. Hamano, Y. Abe, and H. Genda, 2013, Emergence of two types of terrestrial planet on solidification of magma ocean, *Nature* 497:607-610.

SEARCHING FOR LIFE ACROSS SPACE AND TIME

## **SUMMARY OF CHAPTER 4: LIFE DETECTION TECHNIQUES**

Kasting stated that NASA has been looking for extraterrestrial life for at least 40 years. According to Ben Clark, the Viking life detection experiments did not give us the knowledge we hoped they would, but they still resulted in a lot of useful information. Clark also said that the Viking results provide a cautionary tale about thinking about false positives carefully before announcing any results.

In his talk, Gary Ruvkun explained that looking for DNA-based life is easy. All we need to do, according to Ruvkun, is to send an Oxford Nanopore machine to Mars. Since we already have a large database of DNA for Earth life, discriminating between terrestrial life and alien life should be easy. However, extracting the DNA and preparing the sample might be difficult. Additionally, it can be difficult to discriminate one microbe from another one. Another issue is that convergent evolution might complicate the interpretation.

Kasting then mentioned Steve Benner's "paradox of life" from his talk, the idea that molecular systems required for life could not have arisen without life. Kasting sees this as a challenge to the metabolism-first hypothesis, which says that some molecular systems arose abiotically. Benner had also said that life in liquid water would be based on biopolymers with a backbone of repeating charges that would be easy to detect even if it was not DNA-based. According to Benner, dry land may have been a requirement to allow for the formation of borate evaporites to build up a high enough concentration of boron needed for life. If that logic is correct, then water worlds might be a bad place to search for (RNA-based) life.

## **Audience Participation**

A workshop participant said that there are many orders of magnitude separating the selectivity of what is possible at the functional group level in synthetic chemistry versus what is actually used in our biosphere. The big question, he said, is figuring out what mechanisms produce these selection effects and whether they all require a Darwinian dynamic and the emergence of individuality.

Benner then made an offer that he said he has made many times before. He thinks that the metabolism-first model lacks sufficient actionable substance, meaning that they cannot actually test it. However, he offered to synthesize and ship (for free) any necessary molecules to anybody with a specific, actionable chemical system in which a metabolism-first model might operate.

Another participant said that the statement that water worlds might not be good places to search for life is too strong. The water worlds, he said, may have all the ingredients we're looking for. Enceladus has both organic material and lots of water. Although Enceladus doesn't have dry land, it does have a rocky core. The boundary layer may provide a template and mineral interactions that could allow for life. He thinks that these are still promising places. Another member of the audience then responded, saying that the water problem applies to RNA and other molecules that have bonds that are thermodynamically unstable in water. Dry land, he said, is the way to make these bonds stable. Therefore, water worlds would not be a good place to search for RNA-based life. If there were a genetic biopolymer that was not very sensitive to water hydrolysis, a water world could be a great place for that kind of life. However, if you are going to have a genetic biopolymer that is sensitive to water hydrolysis, there has to be a compartmentalization mechanism that can dehydrate a small volume. He said that it is a tough problem for which he does not have a solution.

A participant at the workshop then said that, when considering the origins of life on other planets, the environmental context has to include more than just the planet. It has to include the entire planetary system and the star. For example, a water world could host life that was originally created on another world with dry land and then shipped over via impact.

## **SUMMARY OF CHAPTER 5: INSTRUMENTATION**

In the interests of time, Kasting then briefly went through the instrumentation chapter. Morgan Cable described missions and instruments that could allow for the detection of life in plumes. Shawn Domagal-Goldman said that the Habitable Exoplanet Imaging Mission (HabEx) or the Large UV/Optical/Infrared Surveyor (LUVOIR) could

WRAP-UP

potentially detect life on exoplanets. Jen Eigenbrode detailed missions and instruments designed to detect organic materials on Mars.

## **Audience Participation**

A member of the audience then said that the one thing that they cannot constrain about martian organic geochemistry is what the ionizing radiation does to organics at the surface, both at the time when it was first put into the ground and at the time that it became exposed to radiation. However, life could have also contributed to this. She then said that the one thing we know about how radiation affects organics is that the surface material is the worst place to look for life's potential contribution to the supply of organics. To answer this, she thinks we need to excavate or drill deeper than previous missions ever have.

### PARTING THOUGHTS

Kasting then concluded by saying that there are many questions remaining concerning the detection and interpretation of biosignatures. He then hoped that the astrobiological community would organize itself ahead of the two upcoming decadal surveys facilitated by the National Academies (astronomy and astrophysics and planetary science) in order to provide them with a coherent set of principles and suggestions.

Michael Moloney, the director of Space and Aeronautics at the National Academies, then thanked the committee, the workshop participants, and the members of the Space Studies Board staff for a great two days of discussions. Moloney then closed the workshop. Searching for Life Across Space and Time: Proceedings of a Workshop

# Appendixes

Searching for Life Across Space and Time: Proceedings of a Workshop

## A

## Statement of Task

The National Academies of Sciences, Engineering, and Medicine will appoint an ad hoc planning committee to organize a workshop that will focus on facilitating an expert dialogue on the current status of extraterrestrial life detection and related issues. Based on our current understanding of the nature and physical and chemical limits of life on Earth, the characteristics of worlds in our solar system and planets orbiting other stars, and the state of the art of relevant technologies, the workshop will address the following questions:

• What is our current understanding of the limits of life and life's interactions with the environments of planets and moons?

• Are we today positioned to design, build and conduct experiments or observations capable of life detection remotely or in situ in our own solar system and from afar on extrasolar worlds?

• How could targeted research help advance the state of the art for life detection, including instrumentation and precursor research, to successfully address these challenges?

A workshop report will document the workshop, including summaries of individual presentations and ensuing discussions. This report will not present consensus conclusions or recommendations.

## В

# Workshop Agenda

## **DECEMBER 5, 2016**

| 8:30 a.m. | Welcome  |
|-----------|--|
|           | Michael Moloney, Director, Space Studies Board and Aeronautics and Space<br>Engineering Board          |
| 8:35      | Introduction to the Workshop   |
|           | Jim Kasting, Workshop Planning Committee Chair   |
|           | Session 1: Setting the Stage   |
|           | Moderator: Jim Kasting, Penn State University  |
| 8:40      | How Likely Is It that Life Exists off the Earth?   |
|           | John Baross, University of Washington  |
| 9:15      | Current Understanding of the Environmental Limits of Life and Life's Interactions with Its Environment |
|           | Tori Hoehler, NASA Ames  |
| 9:50      | Is Life a Cosmic Imperative: How Did Thermodynamics Force Life into Existence?                         |
|           | Eric Smith, Santa Fe Institute   |
| 10:25     | Break  |

APPENDIX B

|            | Session 2: Habitable Environments in the Solar System  |
|------------|--|
|            | Moderators: Bethany Ehlmann, Caltech and Britney Schmidt, Georgia Tech   |
| 11:00      | Habitable Environments on Present and Ancient Mars   |
|            | John Grotzinger, Caltech   |
| 11:35      | Habitable Environments on Ocean Worlds   |
|            | Kevin Hand, Jet Propulsion Laboratory  |
| 12:10 p.m. | Planning for the Exploration of Mars and Ocean Worlds  |
|            | Ellen Stofan, NASA   |
| 12:30      | Lunch (Poster Viewing Opportunity)   |
|            | Session 3: Exoplanets  |
|            | Moderators: David Des Marais, NASA Ames and<br>Dimitar Sasselov, Harvard-Smithsonian Center for Astrophysics   |
| 2:00       | Current State of Knowledge of Exoplanets and Their Habitability  |
|            | Vikki Meadows, University of Washington  |
| 2:35       | Extrasolar Biosignatures: Thinking Out of the Box  |
|            | William Bains, Massachusetts Institute of Technology   |
| 3:10       | Break (Poster Viewing Opportunity)   |
| 3:30       | Technology Needs to Discover Earth 2.0   |
|            | Nick Siegler, Jet Propulsion Laboratory  |
| 4:05       | Prospects for Ground-Based Characterization of Proxima Centauri b  |
|            | Matteo Brogi, University of Colorado, Boulder  |
| 4:40       | <i>General Discussion:</i> Practical Biosignatures that Can Be Exploited to Search for Life In Situ in the Solar System and from Afar on Extrasolar Worlds |
|            | Moderator: Gary Blackwood, Jet Propulsion Laboratory   |

| 92            | SEARCHING FOR LIFE ACROSS SPACE AND TIME                                      |
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|               | Session 4: Poster Session   |
| 5:30          | Small Bites, and Reception in Atrium  |
| 8:30          | Adjourn for the Day   |
|               | DECEMBER 6, 2016  |
|               | Session 5: Life Detection Techniques  |
| Moderators: ( | Gary Ruvkun, Harvard Medical School and John Baross, University of Washington |
| 8:30 a.m.     | Life Detection: 40 Years after Viking   |
|               | Ben Clark, Space Science Institute  |
| 9:05          | Signatures of Life as We Know It  |
|               | Gary Ruvkun, Massachusetts General Hospital                                   |
| 9:40          | Signatures of Life as We Don't Know It  |
|               | Steve Benner, the Foundation for Applied Molecular Evolution                  |
| 10:15         | Break: Poster Viewing Opportunity   |
|               | Session 6: Instrumentation  |
| Moderators    | s: Phil Neches, Teradata Corporation and Nilton Renno, University of Michigan |
| 10:45         | In Situ Life Detection in the Context of Plume Fly Thru Missions              |
|               | Morgan Cable, Jet Propulsion Laboratory                                       |
| 11:15         | Life Detection Capabilities of LUVOIR and HabEx                               |
|               | Shawn Domagal-Goldman, NASA Goddard Space Flight Center                       |
| 11:45         | In Situ Detection of Organics on Mars   |
|               | Jen Eigenbrode, NASA Goddard Space Flight Center                              |
| 12:15 p.m.    | Instructions for Afternoon Breakout Groups                                    |
|               | Jim Kasting, Workshop Planning Committee Chair                                |
| 12:25         | Lunch (Poster Viewing Opportunity)  |
|               |   |

| APPENDIX B | 93   |
|------------|--|
|            | Session 7: Future Directions   |
| 2:00       | Breakout Groups  |
|            | <i>Focus Question:</i> How Could Targeted Research Over the Next 5-10 Years Help Advance the State of the Art for Life Detection, Including Instrumentation and Precursor Research                                   |
|            | Discussion Topics:<br>1. In Situ Detection of Life as We Know It<br>2. In Situ Detection of Life as We Don't Know It<br>3. Remote Detection of Life as We Know It<br>4. Remote Detection of Life as We Don't Know It |
|            | <ul> <li>Moderators:</li> <li>1. Nita Sahai and Tanja Bosak</li> <li>2. Mark Thiemens and Steve Benner</li> <li>3. Vikki Meadows and Sushil Atreya</li> <li>4. William Bains and John Baross</li> </ul>              |
| 3:15       | Break (Poster Viewing Opportunity)   |
|            | Session Moderators to Collect Thoughts in Preparation for Report Back to the Group   |
| 4:00       | Report Back from Breakout Group  |
|            | General Discussion   |
|            | Moderator: John Grunsfeld, NASA (retired)  |
|            | Session 8: Conclusions   |
| 5:30       | Summary of Key Points Raised During the Workshop and Concluding Remarks  |
|            | Jim Kasting, Workshop Planning Committee Chair   |
| 6:00       | Workshop Adjourned   |
|            |  |

## С

# Workshop Participants

| Abdulaziz    | Albarakah    | University of California, Irvine   |
|--------------|--------------|--|
| Sushil       | Atreya       | University of Michigan   |
| William      | Bains        | Massachusetts Institute of Technology  |
| Stuart       | Bale         | University of California, Berkeley   |
| Saibal       | Barik        | IGIT Sarang  |
| John         | Baross       | University of Washington   |
| David        | Beaty        | Jet Propulsion Laboratory (JPL)/California Institute of Technology (Caltech) |
| Patricia     | Beauchamp    | JPL/Caltech  |
| Steve        | Benner       | Foundation for Applied Molecular Evolution                                   |
| Dana         | Berry        | SkyWorks Digital Inc.  |
| William      | Bertch       | JPL (NASA Facility)  |
| Doris        | Bertch       |  |
| Janice       | Bishop       | SETI Institute   |
| Gary         | Blackwood    | JPL  |
| David        | Blake        | Exobiology Branch /NASA Ames Research Center                                 |
| Rosalba      | Bonaccorsi   | SETI Institute/NASA Ames Research Center                                     |
| Tanja        | Bosak        | Massachusetts Institute of Technology  |
| Penelope     | Boston       | NASA Astrobiology Institute  |
| William      | Brinckerhoff | NASA Goddard Space Flight Center   |
| Matteo       | Brogi        | University of Colorado, Boulder  |
| Eric         | Brown        | TechnoView IP Inc.   |
| Celena Diana | Bumpus       | American Cancer Society  |
| Shirleye     | Bumpus       | American Cancer Society  |

APPENDIX C

| BileenChinaBrianChowUniversity of California, San DiegoShiyunChungVortex PlusBentonClarkSpace Science InstituteCallaCofieldSpace comCatharineConleyNASA HQCatharineCraftJohns Hopkins University Applied Physics LaboratoryJessicaCreamerJPLBrendanCrillJPLKarenCrow-RoarkNASA Ames Research Center/Texas State NASA STEM Educator<br>Professional DevelopmentPunamDalaiUniversity of AkronOmkarDalviSpace Studies BoardLeonardDavidSpace com; Inside Outer SpaceClaudioDe-FrajaAmeriTech AdvisorsMaryDeiomaDesertSensorsBethanyDomagal-GoldmanNASA Goddard Space Flight CenterAndrewDuncanDesertSensorsBethanyEhlmannCaltechJenniferEigenbrodeNASA Goddard Space Flight CenterBradGeinerUniversity of California, RiversideYishalGajjarUniversity of California, RiversideYishalGajaraUniversity of California, RiversideYishalGalateBrazilian Synchrotron Light Laboratory (LNLS/CNPEM)RichGillockNorthrop Grumman (retired)CatherineGiradeyUniversity of Southern California   | Morgan    | Cable           | JPL   |
|---|-----------|-----------------|---|
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| JessicaCreamerJPLBrendanCrillJPLKarenCrow-RoarkNASA Ames Research Center/Texas State NASA STEM Educator<br>Professional DevelopmentPunamDalaiUniversity of AkronOmkarDalviKatieKatieDaudSpace Studies BoardLeonardDavidSpace.com; Inside Outer SpaceClaudioDe-FrajaAmeriTech AdvisorsMaryDeiomaDavidDes MaraisNASA Ames Research CenterShawnDomgal-GoldmanNASA Goddard Space Flight CenterAndrewDuncanDesertSensorsBethanyEhlmannCaltechJenniferEigenbrodeNast Goddard Space Flight CenterBradFeinnerPedroFlechaConsultant/ResearcherMarilynFogelUniversity of California, RiversideVishalGajjarUniversity of Southern Light Laboratory (LNLS/CNPEM)RichGillockNorthrop Grumman (retired)CatherineGirardeyUniversity of Southern CaliforniaDanielGahamNASA Goddard Space Flight Center  | Catharine | Conley          | NASA HQ   |
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## D

## Poster Abstracts

## CURRENT UNDERSTANDING OF THE PHYSICAL AND CHEMICAL LIMITS OF LIFE

## Ion Tolerance and Preferential Selectivity of a Lipid in Mixed Lipid Systems: An Evolutionary Approach to Modern Membranes

Punam Dala, Putu Ustriyana, and Nita Sahai, University of Akron

Protocell membranes may have been composed of single chain amphiphiles (SCAs) due to their prebiotic availability, but SCA membranes are disrupted by divalent cations in aqueous solutions. Mixed SCA vesicles are known to be more resistant to the fatal effects of dissolved Mg<sup>2+</sup> and Ca<sup>2+</sup>. Here we examined the potential role of Mg<sup>2+</sup> as an environmental selection pressure in the transition of fatty acid membranes to mixed SCA-phospholipid membranes and, finally, to phospholipid membranes. The Mg-tolerance of binary mixtures of oleic acid (OA) with palmitoyl-2-oleoylphosphatidylcholine (POPC) was determined. The fatal magnesium concentration, [Mg<sup>2+</sup>]fatal, was defined as the concentration of Mg<sup>2+</sup> required to disrupt ~100% of vesicles (200 nm extruded). Membrane disruption was determined by measuring the decrease in fluorescence intensity of a membrane-soluble dye, naphthopyrene, that had been previously entrapped in the vesicle membranes. The relative distribution of lipid into vesicles and amorphous aggregates was also estimated by dynamic light scattering and optical microscopy. The  $[Mg^{2+}]$  fatal increased drastically with increasing relative POPC content from 5 mM for pure OA, to 40 mM for [OA]/[POPC] = 1:1, and >80 mM for pure POPC (total lipid concentration = 2 mM, pH 8.5). We propose two distinct mechanisms by which magnesium-tolerance of the mixed lipid systems increases. First, as confirmed by zeta potential measurements, POPC (zwitterionic head group) stabilizes the mixed-lipid vesicles by decreasing the relative negative charge density of the vesicles, so more  $Mg^{2+}$  is needed to disrupt the vesicles. Second,  $Mg^{2+}$ was found to preferentially bind to and abstract OA from OA-POPC mixed lipid membranes, resulting in lower [OA]/[POPC] ratio in the vesicles as compared to the initial ratio. Quantitation of OA and POPC concentration in the vesicles was achieved by filtration (220 nm pore) to remove Mg<sup>2+</sup>-lipid aggregates and high-performance liquid chromatography (HPLC) analysis of the filtrate. This is the first time that a cation has been shown to directly change the composition of a lipid membrane. The significantly greater Mg-tolerance of SCA-phospholipid vesicles may hold implications for the evolutionary selection of phospholipid membranes and for accommodating Mg<sup>2+</sup>promoted processes such as RNA polymerization.

## Non-Enzymatic RNA Polymerization at the Mineral-Water Interface: A Search for a Potential Adsorption-Polymerization Relationship

Hussein Kaddour, Selim Gerislioglu, Toshi Miyoshi, Chrys Wedemiotis, and Nita Sahai, University of Akron

The extent of adsorption of nucleotide or amino acid monomers at mineral surfaces and subsequent surface-catalyzed polymerization to RNA or peptides is a widely assumed potential role of minerals in the origins of life. Few studies, however, have critically examined this assumption. Here, we investigated the relationship, if any, between the adsorption of adenosine monophosphate (AMP) nucleotide on a wide range of minerals (oxides, oxyhydroxide, carbonate, sulfide, aluminosilicate) and the potential catalytic activity of these minerals in the nonenzymatic polymerization of AMP, by using ultraviolet-visible (UV/Vis) spectroscopy, HPLC, mass spectrometry (MALDI) and solid-state 31P NMR spectroscopy. Adsorption on AMP, which is negatively charged at pH 8, increased with isoelectric point (positive surface charge) of the mineral reaching a maximum with zincite (ZnO, IEP ~ 8). However, polymerization on montmorillonite, a negatively charged clay mineral, was better than on ZnO, consistent with the work of Jim Ferris, Prakash Joshi, and co-workers. The nuclear magnetic resonance spectroscopy results showed that the AMP monomer was bonded via the phosphate moiety to the ZnO surface, thereby preventing condensation between adjacent AMP monomers. In contrast, the phosphate moiety was relatively unconstrained at the montmorillonite surface, which is interpreted to indicate that the adsorbed conformation allowed interaction between phosphate moieties of adjacent AMP monomers. Thus, the configuration of the adsorbed AMP monomer with respect to the mineral surface and to the neighboring AMP molecule is more important than the total mass of adsorbed monomer for surface-catalyzed polymerization.

## HABITABLE ENVIRONMENTS IN THE SOLAR SYSTEM AND EXTRASOLAR PLANETARY SYSTEMS

## The Icebreaker Life Mission: Why Search for Modern Life on Mars and How to Do It

## Carol Stoker and C.P. McKay, NASA

Ground ice in the northern plains of Mars hosts habitable conditions for life periodically, most recently during high obliquity, 0.5 to 10 Myr ago. Habitable conditions include (1) pressure above the triple point of liquid water; (2) ice near the surface as a source of liquid water; and (3) high summer insolation at orbital tilts >35° (present 25°), equivalent to levels of summer sunlight in Earth's polar regions at the present time. Terrestrial permafrost communities are examples of possible life in the ground ice. Studies in permafrost have shown that microorganisms can function in ice-soil mixtures at temperatures as low as  $-20^{\circ}$ C, living in thin films of interfacial water. In addition, it is well established that ground ice preserves living cells, biological material, and organic compounds for long periods of time, and living microorganisms have been preserved under frozen conditions for thousands and sometimes millions of years. Similar biomolecular evidence of life could have accumulated in the ice-rich regolith on Mars. The Mars Icebreaker Life mission has been proposed to search for life there. Science goals are: (1) search for biomolecular evidence of life; (2) search for organic matter from either exogeneous or endogeneous sources using methods that are not affected by the presence of perchlorate; (3) characterize oxidative species that produced reactivity of soils seen by Viking; and (4) assess the habitability of the ice-bearing soils. The payload includes a 1-m drill that brings cuttings samples to the surface where they are delivered to three instruments: the Signs of Life Detector (SOLID) for biomolecular analysis, Laser Desorption Mass Spectrometer (LDMS) for broad spectrum organic analysis, and Wet Chemistry Laboratory (WCL) for detecting soluble species of nutrients and reactive oxidants. The poster will describe the mission and instruments.

## How Can We Know that We Are Not Sampling Bacteria in the Clouds of Venus If Their Physical Properties Are Similar?

### Sanjay Limaye, University of Wisconsin

There is a great similarity in the physical properties of *Th. ferrooxidans* and similar species and those of the cloud particles on Venus. Further, these types of bacteria also absorb ultraviolet below 400 nm and have variable transmittance at 2 to 3 microns, key characteristics of Venus clouds. Recent research suggests that Venus could have harbored liquid water on its surface for about 2 billion years, so it is conceivable that life migrated to the clouds when surface became warmer. How can we look for the distinctive properties of the clouds to know if the UV absorber is organic or inorganic?

#### The Habitable Zone: A Planetary Scientist's Perspective

## David Paige, UCLA

The habitable zone is generally defined as the region around a star that can support liquid water given sufficient pressure. In our own solar system, this region is not limited to a small range of distances from the Sun, but includes a diverse-range of surface, subsurface, and atmospheric environments that extend from Mercury to the outer solar system. Conversely, not all planetary bodies within the Sun's "Goldilocks Zone" are actually habitable, as evidenced by the Earth's Moon. What we know about the habitability of our own solar system through time can help guide our search for habitable environments around other stars.

#### Plausible Organic Chemistry Might Precede the Early Development of Life?

Sayali Mulay, TYBSc. Biotechnology, Fergusson College, Pune, India

Objective: The Urey-Miller-Miller experiment gave a content explanation of the possibility of formation of organic molecules from the inorganic molecules by depicting the early Earth conditions. By simulating the same experiment with Venusian gases and conditions, potential molecules regarded as the early building blocks of life that might precede the development of life in the Venusian clouds can be stated.

Introduction: The Venusian clouds present at 50 to 60 km from the ground have favourable environment for life to originate and sustain. These clouds are rich in carbon dioxide and nitrogen. Traces of other gases such as water vapour, carbon monoxide, and sulphur dioxide are present. Acidic environment due to presence of hydrochloric acid and sulphuric acid is seen. Atmospheric pressure is 1 atm, and temperature ranges from 76°C to 10°C with the altitude. With these conditions, experiments can be carried out, and the formation of organic compounds can be observed, if any.

Methods: sterile glassware must be used to carry out this experiment. The gases used will be carbon dioxide, nitrogen, water vapour, carbon monoxide, and sulphur dioxide in ratio as per the presence of the same in the Venusian clouds. Fluctuating low electric current can be used as there are few evidences of lightning strikes on Venus and Whistler waves. Otherwise, UV radiation can also be used, although the UV flux penetrating into the clouds at 50 to 60 km from the ground is much less. Water vapour at about 76°C can be used.

Results: Hypothesis is such that, due to presence of carbon and nitrogen with a source of electricity, organic compounds should be obtained. These organic compounds studied show that possible microbial world that can exist in the clouds can be found out using characters of different extremophilic microbes on Earth. This resulting "organic tar" (hypothesised) can also be used as a medium to isolate Venusian microbes in the clouds (hypothesised) as a sample return mission.

Conclusion: Simulation of Urey-Miller experiment in the laboratory with Venusian environmental factors can result into following hypothesis: (1) organic chemistry is flourishing in the Venusian clouds, (2) obtained organic compounds can be responsible factors for the plausible preceding life on Venus, and (3) Venus sample return mis-

sions can be planned with the obtained "organic tar" as the source in the media to isolate already hypothesised microbial life on Venus.

#### Identification of Clays on Mars and Why They Are Important for Astrobiology

## Janice Bishop, SETI Institute

Phyllosilicate deposits on Mars provide an opportunity to evaluate aqueous activity and the possibility that habitable environments may have existed during Mars' early history. Analysis of visible/near-infrared (VNIR) reflectance spectra acquired by the Mars Reconnaissance Orbiter (MRO) Compact Reconnaissance Imaging Spectrometer for Mars (CRISM) instrument has revealed thick, complex profiles of phyllosilicates on the surface of Mars that are consistent with long-term aqueous activity and active chemistry. The ancient phyllosilicates in places such as Mawrth Vallis could have served as reaction centers for organic molecules. Previous experiments even suggest that phyllosilicates could have played a role in the origin of life. Regardless of whether life formed on early Mars or not, evaluating the type and thickness of clay-bearing units on Mars provides insights into plausible aqueous processes and chemical conditions both during the time of formation of the phyllosilicates, but also the subsequent period following their formation. Changes in iron redox state and in phyllosilicate chemistry indicate an active geochemical environment during the time of clay formation. In some environments, clays are associated with carbonates and neutral environments, while in others they are associated with sulfates and acid alteration. Also, recent identification of poorly crystalline aluminosilicates at the top of the clay profile indicates a change in climate from the environment supporting liquid water and formation of clay minerals to an environment where liquid water was no longer abundant on the surface. Thus, characterizing the type of clays and associated minerals on Mars from orbit provides clues to when and where Mars may have been habitable in the past.

## Finding a Planet's Heartbeat: Unexpected Results from Patient Mars

Vlada Stamenkovic, Lewis Ward, and Woody Fischer, Caltech, and Michael Mischna and Michael Russell, JPL

A planet, from deep interior to atmosphere, has the potential to generate essential nutrients and redox gradients critical for the emergence and the evolution of life. Here, I will present preliminary results on two very different large-scale planetary processes that generate nutrients and redox gradients and discuss the implications for Earth and Mars. Using time-dependent geodynamical and atmospheric models, I will show results on how (1) geodynamically produced hydrogen and methane via serpentinization generates a flux of reducing elements within the lithosphere and (2) how oxygen-rich oases can be formed on the surface and in the crust as a result of brine-atmosphere interactions. This allows us to study the ability of planets, like Earth and Mars, to generate nutrient-rich and redox-rich oases as a function of planet mass, composition, age, and tectonic mode. Moreover, our model also opens the doors to make predictions where and when such oases could have existed or could still exist on Mars. The latter does not only help to better understand how Mars evolved but can become a promising tool to help guide future landing site selections and even manned missions that are looking for hydrogen-rich and oxygen-rich reservoirs on the red planet.

### Searching for Life on Mars: Consensus Input from the Mars Exploration Community

Lindsay Hays, JPL/Caltech; Jennifer L. Eigenbrode, NASA Goddard Space Flight Center; Sarah Stewart Johnson, Georgetown University; Tori Hoehler, NASA ARC; David Des Marais, NASA ARC; Lindsay Hays, JPL/Caltech; David Beaty, JPL/Caltech; and Victoria E. Hamilton, Southwest Research Institute

The Mars Exploration Program Analysis Group's (MEPAG's) "Goals Document" is regularly updated to reflect the exploration community's consensus regarding its scientific priorities for robotic investigations at Mars.

Goal I—to determine if Mars ever supported life—is one of four unprioritized goals and has been a key driver of the Mars Exploration Program. Goal I is broken down into two objectives that focus on searching for evidence of past (objective A) and extant life (objective B), which are kept separate because there are significant differences in the strategies, technologies, target environments, and forms of evidence involved in those searches. For both objectives, "A clear scientific strategy (i.e., an investigative plan built on target-specific hypotheses and measurements) can only be formulated once an environmental record or environment is understood in sufficient detail" (MEPAG goals document, 2015). Although the prioritization and definition of the Goal I objectives, sub-objectives, and investigations are subject to change in response to mission discoveries and community discussions, priority is currently placed on objective A (evidence for past life), because previous mission observations have largely provided details on past environmental conditions.

Objectives A and B are further broken down into sub-objectives focused on (1) identification of habitable environments (past or present) and characterization of the conditions and processes that may have influenced the degree or nature of habitability therein, (2) assessing the potential of specific conditions and processes that may influence the expression and/or degradation of signatures of life and habitability, and identify favorable deposits for their detection, and (3) determining if biosignatures of a prior or extant ecosystem are present. Finally, each sub-objective lists investigations that would collectively enable the achievement of the sub-objective while avoiding discussion of implementation.

#### The NASA Exoplanet Exploration Program

#### Douglas Hudgins and John Gagosian, NASA HQ, and Gary Blackwood, JPL

The NASA Exoplanet Exploration Program (ExEP) is chartered to implement the NASA space science goals of detecting and characterizing exoplanets and to search for signs of life. The ExEP manages space missions, future studies, technology investments, and ground-based science that either enables future missions or completes mission science. The exoplanet science community is engaged by the program through science definition teams and through the Exoplanet Program Analysis Group. The ExEP includes the space science missions of Kepler, K2, and the proposed WFIRST-AFTA, which includes dark energy science, a widefield infrared survey, a microlensing survey for outer-exoplanet demographics, and a coronagraph for direct imaging of cool outer gas- and ice-giants around nearby stars. Studies of probe-scale (medium-class) missions for a coronagraph (internal occulter) and starshade (external occulter) explore the trades of cost and science and provide motivation for a technology investment program to enable consideration of missions at the next decadal survey for NASA Astrophysics. Program elements include follow-up observations using the Keck Observatory, which contribute to the science yield of Kepler and K2, and include mid-infrared observations of exo-zodiacal dust by the Large Binocular Telescope Interferometer, which provide parameters critical to the design and predicted science yield of the next generation of direct imaging missions. ExEP includes the NASA Exoplanet Science Institute, which provides archives, tools, and professional education for the exoplanet community. Each of these program elements contribute to the goal of detecting and characterizing Earth-like planets orbiting other stars and seeks to respond to rapid evolution in this discovery-driven field and to ongoing programmatic challenges through engagement of the scientific and technical communities.

#### Subsurface Mars as the Longest Continually Available Habitat: Implications for the Search for Life

## Bethany Ehlmann, California Institute of Technology

The last decade of Mars exploration has revealed a dozen aqueous, potentially habitable environments, ranging from lacustrine to hydrothermal to weathering. These environments varied in space and time and imply a warm and wet subsurface with punctuated periods of more clement conditions that allowed liquid water at a cold surface. Mineralogical evidence for past liquid water is widespread, but lack of evidence for terrestrial-style, open-system chemical weathering in most terrains points to subsurface water-rock interactions, water-limited weathering under

cold conditions, or both (Hurowitz and McLennan, 2007; Ehlmann et al., 2011; McLennan et al., 2014; Arvidson et al., 2014), supported by climate models (Wordsworth et al., 2015).

Searches for martian life must be informed by these conditions, different from those of early Earth. Unlike on Earth, little geologic evidence exists for a martian northern ocean or individual lakes that persisted continuously as stable habitats for billions of years. Instead, Mars may have episodically hosted a northern ocean (Pan et al., 2017), and in the southern highlands, lakes, and rivers fed by runoff from ice melt or precipitation were episodic (10 kyr to 10 Myr; Barnhart et al., 2009; Grotzinger et al., 2015). Second, Mars lost its magnetic field early (3.9-4.1 Ga) and also likely had a thin atmosphere (<1 bar) by the time periods accessible in rock strata (Ehlmann et al., 2016). Consequently, martian organisms dealing with challenges of cold and surface aridity also faced surface radiation doses many times higher than on Earth. Thus, martian surface habitats have always been more episodic and more extreme than age-equivalent surface habitats on Earth. Consequently, rock-hosted habitats, shielded from radiation and showing evidence of persistent water, warrant particular attention in the search for life: groundwater aquifers, hydrothermal systems, and weathering profiles. Here we describe habitats for past and present rock-hosted Mars life and potential biosignatures. Similarly, data for liquid water on modern Mars point to the importance of the subsurface. Salts excavated in soils by the Spirit rover and climate data coupled with perchlorate detections by Curiosity show likely brine creation and brief stability a few to tens of cm beneath the surface (Wang et al., 2006; Arvidson et al., 2008; Martin-Torres et al., 2015). Recurring slope lineae show temperature-correlated activity consistent with a role for liquid water or salt deliquescence (McEwen et al., 2011; Stillman et al., 2014) but a dry surface (Edwards and Piquex, 2016).

## Social and Conceptual Issues in Astrobiology

## Kelly Smith, Clemson University

A very successful off-year workshop of the International Society for the History, Philosophy and Social Studies of Biology (http://kcs098.wixsite.com/socia) was held at Clemson University in September to explore the social and conceptual issues surrounding astrobiology. The workshop mixed younger scholars and graduate students with established scholars from a wide variety of disciplines, including history, philosophy, communications, biology, astronomy, engineering, theology, medicine, chemistry, geology, and education.

Such a diverse group of researchers produced an equally diverse set of presentations, both in terms of focus and approach. But they fell into five broad categories: (1) philosophy of science (e.g., evidentiary considerations and the risks of anthropocentrism), (2) intelligence/consciousness (e.g., the evolution of cognitive capacity and conceptual difficulties for communication), (3) life concepts (e.g., universality in biology versus conceptual pluralism), (4) ethical issues (e.g., planetary protection policies and extraterrestrial wilderness), and (5) social/cultural issues (e.g., the interplay between astrobiology, religion, education, and politics). The quality of presentations was excellent, and many will appear in a forthcoming conference volume.

The organizers plan to leverage the success of this first meeting with a second meeting at the University of Nevada, Reno in the spring of 2018, with the ultimate goal of founding a new society dedicated to scholarship and outreach on these exciting issues.

## IN SITU BIOSIGNATURES

#### Resilience of Molecular Biosignatures under Simulated and Analogue Planetary Environments

Douglas Galante, Brazilian Synchrotron Light Laboratory (LNLS/CNPEM); Fabio Rodrigues, Instituto de Química da Universidade de São Paulo; and Tamires Gallo, Maria Fernanda Cerini, and Nathalie Rivas, LNLS/CNPEM)

This work will present an overview of the resilience of biomolecules that could be used as indicative of the past or present presence of life on exposed planetary surfaces, such as that of Mars or the icy moons of the solar system.

Different classes of molecules have been used, but especially biological pigments—carotenoids, chlorophyll, and porphyrins. These have been tested in laboratory under simulated conditions, especially radiation, pressure and temperature, and the response has been measured using in situ and ex situ spectroscopic methods—UV-Vis, Raman, and Fourier transform infrared spectroscopy. The molecules have also been exposed to the stratospheric environment using balloons, to produce a more complete martian analogue environment.

#### Life Detection in Planetary Analog Materials: Applications to the Search for Life in the Solar System

Rosalba Bonaccorsi, SETI Institute/NASA ARC; Christopher McKay, NASA ARC; Alfonso Davila, SETI Institute/NASA ARC; and David Willson, Keck Institute of Space Studies/NASA ARC

Detection of molecular proxies for life in planetary environments depends on four conditions: (1) their initial presence due to current and past biological production, (2) their concentration in measurable amount through sedimentary processes in geological materials, (3) their long-term preservation within the material, and (4) the analytical ability of payload instruments to detect and identify them. The analytical requirement is a very key one. False negatives (null or incomplete recovery) can result from the analysis of both biologically lean and biologically rich environmental samples. To test effectiveness of life-detection assays, we have analyzed lipopolysaccaride (LPS) Lipid A and Adenosin Triphosphate (ATP) biomarkers in a variety of planetary-like environments (e.g., hypersaline lakes, fine-grained clay-rich sediments, ice-cemented ground, cyanobacteria-colonized soil crust, and hydrothermal sinters). We present here results from the in situ analysis of Lipid A and ATP using lab-on-the chip/ wet chemistry assays. In geological and water samples, LPS- and ATP-based biomass range from 10<sup>2</sup> to 10<sup>9</sup> cells/ gram. Most importantly, LPS and ATP detection can be affected by the mineralogical (i.e., clay minerals, nanophase iron oxyhydroxides) and physico-chemical composition (salts, pH, T, organics) of the geological matrix. Failing to detect life in modern terrestrial environments that we know have abundant life is a chief concern for our ability to detect life on Earth and other planets as well. Learning how to assess and mitigate matrix-related interference is key to the success of future life detection missions to our solar system, including Mars and the ocean world icy moons, Enceladus and Europa.

#### **Biosignatures of a Hyper Saline Environment**

### Heather Smith, Keck Institute of Space Studies Institute of Practical Robotics

We report on changes in the salt crust photosynthetic microbial community measured when exposed to 1 week of simulated martian conditions (UV, pressure, and temperature) in a Mars chamber. Halophile ecosystems are models for life in extreme environments including planetary surfaces. Our research was on the microbial preservation potential of salt subjected to martian pressure, UV, and temperature. Figure 1 is a picture of the research site with the inset showing the microbial stratigraphy within the salt crust. Visual changes within the stratigraphic layering and phospholipid fatty acid (PLFA) analysis were used to determine changes in microbial community.

#### Linking Microbial Communities to Preserved Biosignatures

Scott Perl, NASA/JPL and USC; P. A. Vaishampayan, Caltech/JPL; F. A. Corsetti, USC; O. Piazza, USC; K. W. Williford, Caltech/JPL); M. L. Tuite, Caltech/JPL; B. K. Baxter, Westminster College; J. Butler, Westminster College; W. M. Berelson, USC; and K. H. Nealson, USC

Determination of potential in situ biology in the martian subsurface in the form of biosignatures and/or biomarkers can be extremely difficult to detect due to their likely physical location embedded within mineralogy and sedimentary outcrops. References to terrestrial extreme environments that have similar geologic, geochemical, and aqueous histories are necessary to distinguish between abiotic and biotic samples (not from and from organic life, respectively). The precipitation of evaporate minerals from ancient and receding lakebeds allows for evidence

of in situ biogenic material to be preserved and/or recorded in the structure of minerals that use the evaporating lakebeds as input fluids for evaporate formation. The depth of preserved biotic information from the perspective of a rover's payload can be complemented by independent microbiological analysis. These complementary analyses would behoove future planetary rovers that have mineralogical and organic detection instruments due to comparisons that can be made from the mineralogy at the elemental level and biogenically with DNA extraction and sequencing as well as fatty acid extraction. This would help confine the types of organics that a planetary rover could discover in situ while providing biological evidence that a rover's toolset cannot currently achieve. The purpose of this investigation is to classify, validate, and quantify organics (archaea and bacteria) that have been preserved within mineralogy, formed in situ, from the evaporation of saline lake waters. The methodology focuses on entombed biology within evaporates employing two principal initiatives that support microbiology laboratory experiments and rover instrument quantification.

#### Sulfur Redox States among High Arctic Sites and Carbonaceous Chondrites

Ted K. Raab, Carnegie Institution for Science; Darren Locke, Jacobs/NASA Johnson Space Center; and Trudy Bolin, University of Illinois, Chicago

Biochemical energy can be generated through transformation of redox states. On Earth, such elements include Fe, S, Mn, Cl, and Cu. In the hazardous radiation environment of space, life requires shielding from charged particles and intense UV. In a series of field and laboratory experiments, we explore the "niche space" for sulfur transformations among bacteria and viruses—an element with the largest number of accessible redox states. We also identify pore networks within carbonaceous chondrites meteorites. Both questions rely on X-ray methods that can eventually be developed for unmanned exploration.

## It's Alive! (But Is It Local?) Planetary Protection Considerations in Distinguishing Extraterrestrial Life from Earthly Contamination

### John Rummel, SETI Institute

While much can be made of the search for in situ biosignatures representing either life as we know it, or life as we don't know it, the spectre of detecting life from Earth when looking for life from (name your favorite extraterrestrial habitat) haunts the field. Earth life, in its profusion, has made it difficult to "see" extraterrestrial life unless strict measures are taken to avoid and/or remove the biological and organic contamination affecting spacecraft carrying life-detection instrumentation. Given the challenges associated with reaching potential life-sites on other worlds and the revival of interest in novel techniques to detect life in samples that can be held in a robotic "hand," the planetary protection measures designed to protect against false indications of life on planetary bodies may seem daunting. Nonetheless, searching for life while distributing contamination widely into the surrounding environment is both counterproductive and, for most spacefaring nations, illegal in their adherence to the United Nations Outer Space Treaty, *res ipsa loquitur*. Even more obviously, false-positive results about life in a particular location can have the unfortunate result of drowning out the actual detections of extraterrestrial life that the taxpayers are funding and that we are pursuing with our own lives. This presentation will discuss the current status of planetary protection measures available to prevent Earth contamination of spacecraft searching for extraterrestrial life.

## Ammonium in Clays—A Biosignature

#### Eva Stueeken, University of California, Riverside, and University of Washington, Seattle

Some of the oldest rocks on Earth – 3.8 billion-year-old metasediments from Isua – contain significant amounts of nitrogen with concentrations of several hundred parts per million. Such concentrations are commonly found in

younger mudrocks, where they are usually thought to be derived from the degradation of biomass and incorporation of  $NH_4^+$  into clay minerals. Whether or not an abiotic nitrogen cycle could mimic such high concentrations is so far unknown. This question is addressed with a numerical box model that simulates an abiotic nitrogen cycle with inputs of fixed nitrogen through lightning, impacts, and hydrothermal activity. Abiotic sinks include volatilization of  $NH_3$  back into the atmosphere and adsorption of  $NH_4^+$  on mineral surfaces. The results suggest that abiotic pathways are unlikely to produce nitrogen concentrations greater than a few parts per million under realistic pH conditions and source fluxes. The observed abundances are thus most plausibly interpreted as a relic of an early Archean biosphere. In conclusion, nitrogen concentrations may serve as a useful biosignature on other planets.

#### Signs of Life 2002

#### David Smith, National Academies of Sciences, Engineering, and Medicine

In April 2000, the National Academies' Space Studies Board and Board on Life Sciences jointly organized a workshop to discuss a variety of topics, including the following: the search for extraterrestrial life in situ and in the laboratory; extant life and the signature of extinct life; and determination of the point of origin (terrestrial or not) of detected organisms. The material presented during the workshop and in follow-on study were published in the 2002 report *Signs of Life*.

The report was organized around four general questions. First, how does one determine if living terrestrial organisms are on a spacecraft before launch? Second, how does one determine if there are living organisms in a returned sample? Third, how does one determine if living organisms have been present at some earlier epoch and have left fossil remnants behind in a returned sample? Fourth, how does one determine whether there are living organisms or fossils in samples examined robotically on another solar system body?

Significant progress has been made in the last 16 years in addressing many of the questions above. Indeed, much of the material contained in *Signs of Life* might now be considered dated. However, the report's concluding chapter contained two very useful tables summarizing life-detection techniques that were promising at the time and gave assessments of their likely sensitivities and areas of applicability. These tables are reproduced in the current poster and are available online at the National Academies Space Studies Board website. 2016 workshop participants are encouraged to review specific entries and suggest updates and/or other amendments, as appropriate.

## NASA's Life Detection Ladder 2016

#### David Smith, National Academies of Sciences, Engineering, and Medicine

The direct detection of extant life has not been attempted by NASA since the Viking missions in the late 1970s. NASA's Ladder of Life Detection (http://astrobiology.nasa.gov/research/life-detection) was generated to stimulate and support discussions among scientists and engineers about how one would detect extant life beyond Earth but within the solar system (particularly on Europa and the other "ocean worlds"). In creating the Ladder, we started with the NASA definition of life, "Life is a self-sustaining chemical system capable of Darwinian evolution" and considered the specific features of the one life we know—terran life. Please e-mail any suggestions to arc-nai@ mail.nasa.gov.

## The 2007 Astrobiology Strategy for the Exploration of Mars

David Smith, National Academies of Sciences, Engineering, and Medicine, and Bruce Jakosky, Laboratory for Atmospheric and Space Physics, University of Colorado, Boulder

The last decade of the 20th century and the first decade of the 21st witnessed a rebirth in interest in the exploration of, and search for life on, Mars and a spate of new spacecraft missions. Mars Pathfinder, Mars Odyssey, Mars Express, Mars Reconnaissance Orbiter, and the Mars Exploration rovers Spirit and Opportunity provided a wealth of new information about the planet's environment, including strong evidence of a watery past and the possible discovery of atmospheric methane. In addition, new developments in our understanding of life in extreme conditions on Earth suggest the possibility of microbial viability in the harsh martian environment. Together, these results have greatly increased interest in the search for life on Mars, both within the scientific community and beyond. Given the enhanced scientific and political interest in the search for life on Mars, it is surprising that NASA's then most recent end-to-end strategy for the detection of martian life, contained in the report *An Exobiological Strategy for Mars Exploration*, was published as long ago as 1995.

Against this backdrop, NASA's Science Mission Directorate requested that the Space Studies Board develop an up-to-date integrated astrobiology strategy for Mars exploration that brings together all the threads of this diverse topic into a single source for science mission planning. The resulting report, *An Astrobiology Strategy for the Exploration of Mars*, published in 2007, addressed the following topics:

• The characteristics of potential targets for Mars exploration particularly suited for elucidating the prebiotic and possibly biotic history of Mars, and methods for identifying these targets;

• A catalog of biosignatures that reflect fundamental and universal characteristics of life (i.e., not limited to an Earth-centric perspective);

• Research activities that would improve exploration methodology and instrumentation capabilities to enhance the chances of astrobiological discovery; and

Approaches to the exploration of Mars that would maximize the astrobiological science return.

## Biosignatures of Extant Life on Ocean Worlds (BELOW) Workshop

Jennifer Eigenbrode, NASA GSFC; Stephanie Getty, NASA GSFC; Tori Hoeler, NASA ARC; John Priscu, Montana State University; Andrew Steele, Carnegie Institution of Science; and the Working Group Chairs of BELOW

The aim of BELOW was to evolve our understanding of the detectability of extant life on ocean worlds, such as Europa and Enceladus. The event brought together astrobiologists, biologists, chemists, geologists, oceanographers, and mission and instrument developers to discuss the informational value of different types of biosignatures, the importance of context and the concept of ecology in the search for extant life, and as well as exploration criteria that would support a productive search for extant biology in future missions. The workshop successfully identified signals deemed important for the search for life on ocean worlds as well as issues of noise and processing concerns. A summary of the results of the workshop will be presented.

#### A Probabilistic Intrinsically Calibrated Framework for Recognizing Complex Molecules as Biosignatures

## Jennifer Eigenbrode, NASA GSFC, and Lee Cronin, University of Glasgow

The ability of living systems to replicate and evolve allows for the generation of complex molecules, such as metabolites and co-factors, which would be highly unlikely to form in any significant quantity in the absence of biology. We have developed an intrinsic general complexity measure that can predict the likelihood of a molecule (and by extension complex polymers) to have formed by a non-biological process, that can be assessed using analytical methods. By comparing the complexity of simple molecules to that of complex ones (found in biology), we aim to establish a threshold beyond which the molecules are increasingly unlikely to form without supporting biological machinery. Then by evaluating the complexity of unknown molecules found in a given environment, we use the threshold to assign the probability that the molecules in question were generated by a living system, either directly as a metabolite, or indirectly by a person or robot. The advantage in searching for complexity, rather than specific chemical features, is that it is completely general and agnostic to the specific chemical or biological details.

108

#### **Detecting Motility and Morphology as Biosignatures**

## Chris Lindensmith, JPL; Jay Nadeau, Caltech; Manuel Bedrossian, Caltech; Marwan Elkholy, McGill University; Jody Deming, University of Washington; and Max Showalter, University of Washington

Meaningful activity (motility, cell division, biofilm formation) is an unambiguous biosignature. Because life in the solar system is most likely to be microbial, the question is whether activity may be detected effectively on the micrometer scale, and whether inactive or dormant organisms may be stimulated to become active. Recent results on microbial motility in oligotrophic Earth environments, such as the open ocean, have provided insight into the physics and biology that determine whether and how microorganisms as small as bacteria and archaea swim, under which conditions, and at which speeds. These discoveries are only starting to be reviewed in an astrobiological context. This poster discusses these findings in the context of Earth analog environments and environments expected to be encountered in the outer solar system, particularly the Jovian and Saturnian moons. We present several imaging technologies, including holographic and Fourier ptychographic microscopy capable of observing activity of sub-micrometer-sized organisms, and discuss how an instrument would interface with several types of sample-collection strategies and with chemical biosignature detection.

## Capillary Electrophoresis Separations of Chiral Amino Acids for Biosignature Detection

Jessica Creamer, Maria F. Mora, and Peter A. Willis, JPL

Amino acids are fundamental building blocks of terrestrial life as well as ubiquitous byproducts of abiotic reactions. In order to distinguish between amino acids formed by abiotic versus biotic processes, it is possible to use chemical distributions to identify patterns unique to life. This article describes two capillary electrophoresis methods capable of resolving 17 amino acids found in high abundance in both biotic and abiotic samples (seven enantiomer pairs D/L-Ala, -Asp, -Glu, -His, -Leu, -Ser, -Val, and the three achiral amino acids Gly,  $\beta$ -Ala, and GABA). To resolve the 13 neutral amino acids, one method utilizes a background electrolyte containing  $\gamma$ -cyclodextrin and sodium taurocholate micelles. The acidic amino acid enantiomers were resolved with  $\gamma$ -cyclodextrin alone. These methods allow detection limits down to 5 nM for the neutral amino acids and 500 nM for acidic amino acids and were validated by analyzing samples collected from Mono Lake with minimal sample preparation.

### **Biosignatures from a Deep Biosphere: Lessons Learning from Earth**

Haley Sapers, Caltech; Jan Amend, USC; David Beaty, JPL; Rohit Bhartia, JPL; Kevin Cannon, Brown University; Charles Cockell, University of Edinburgh; Max Coleman, JPL; Dave Des Marais, NASA ARC; J. Marlow, Harvard; B. Ehlmann, Caltech; Tori Hoehler, NASA ARC; Tom McCollom, University of Colorado; Joe Michalski, Planetary Science Institute; John Mustard, Brown University; Ken Nealson, USC; Paul Niles, NASA Johnson Space Center; G. R. Osinski, Western University; Tullis Onstott, Princeton University; Victoria Orphan, Caltech; Barbara Sherwood-Lollar, University of Toronto; Alexis Templeton, University of Colorado; Greg Wanger, JPL

Introduction: The current surface conditions on Mars are incompatible with life as we know it: the surface atmospheric pressure precludes standing water. Harsh UV and gamma radiation destroy complex organic molecules in the surface and near-surface environment, hindering detection of organic biosignatures. These harsh surface conditions potentially extended to the Noachian/Hesperian boundary, so surface environments, including lakes/ deltas, may not have habitable at the surface. However, subsurface refugia may have extended the window of habitability, and putative subsurface pockets of habitable conditions could potentially still harbor extant life and their biosignatures. Understanding the biological processes in the terrestrial subsurface will yield insight into the identification, detection, and characterization of potential subsurface martian biosignatures.

Subsurface habitats on Earth: The minimum requirements for subsurface life include space, carbon, and energy linked in a substrate allowing for an adequate supply of nutrients and removal of toxic waste products. Subsurface environments may harbor the majority of microbial life on Earth and Archaean biosignatures suggest the existence of a terrestrial biosphere for billions of years. Recent findings from multiple sulfur isotope investigations of fracture waters in 2.7 Ga rock from the Canadian Shield demonstrate the potential for isotopic evidence of subsurface microbial activity to be preserved on long geologic timescales. Subsurface microbial communities are sustained through chemolithoautotrophic metabolic processes adapted to energy limitations limited by the geothermal gradient reaching the upper temperature limits of life. Extant subsurface metabolisms in these terrestrial Mars analogue habitats include coupling oxidation of  $H_2$  generated by serpentinization reactions to reduction of oxidized iron and sulphate minerals. The dynamic biogeochemical reactions defining life processes in the subsurface constantly process abiotic materials resulting in biosignatures with the potential to be preserved in the rock record. Understanding the biologically mediated processes that result in geological biosignatures not only extends our knowledge of the limits of life on Earth, but provides a framework with which to search for life on Mars.

## **INSTRUMENTATION**

## In Situ Resource Utilization for Environmental Protection

#### Yu Qiao and Brian J. Chow, University of California, San Diego

As searching for life on Mars draws increasing attention, environmental protection associated with unmanned and manned space exploration missions must be carefully investigated. One major technical issue is the lack of infrastructural materials to build relatively large-scale insulation layers, protective walls, separation zones, permanent waste-disposal containers, and sealed storage units, among others. In a recent experimental research, we discovered that both primordial and secondary martian soils could be directly compacted into strong and dense "bricks," with appropriate processing conditions. The compaction procedure was simple, fast, and more importantly, energy efficient. It was conducted under ambient condition; no heating/calcination or any additives was involved. This technique may also have important relevance to expansion and maintenance of martian bases/ outposts/habitats, as well as massive and bulky parts in space research facilities and equipment, such as launch/ landing platforms and supports of space telescopes.

## **Ocean Biomolecule Explorer for Astrobiology**

Heather Smith, Keck Institute of Space Studies Institute of Practical Robotics; Andrew Duncan, Desert Sensors; and Chris Lloyd, Retego Labs

The Ocean Biomolecule Experiments for Astrobiology is a life detection instrument suite designed towards an Ocean Worlds surface mission. The instrument suite relies on the modification of commercial off-the-shelf instruments combined with newly developed biochemical analysis methods to paint a picture of the biological realm on Europa's Ocean World. This search for extant life relies on our understanding and assumptions of Europa, Enceladus, and Titan within the context of Earth's biochemistry and known metabolic process. To gain an initial picture of Europa life, if present, the instrument suite is designed to detect a range of targets associated with life on Earth, including basic biomolecules as well as the yield from complex metabolic process. The instrument suite will both detect the presence of extant life and provide insight into evolutionary process on the Ocean World. While the instrument suite in this proposal is designed for a Europa lander, the fundamental method of detection could also be applied to Enceladus and Titan. As such, when relevant, a brief analysis on the modification of the instruments for Enceladus and Titan is also included.

#### **Non-Contact Detection of Biomolecules**

## Andrew Duncan, DesertSensors; Heather Smith, Keck Institute of Space Studies Institute of Practical Robotics; and Chris Lloyd, Retego Labs

For this project, we designed an instrument to detect bacteria via biomolecular (amino acids, metabolites) fluorescence. We proposed a novel technique for searching for direct evidence of life on planetary bodies. Fluorescence laboratory measurements using the portable instrument reveal microbial concentration in desert soil to range from 102 to 107 bacteria per gram of soil equivalent. Biomolecules and polycyclic aromatic hydrocarbons are highly fluorescent at wavelengths in the ultraviolet (266 nm, 355 nm), but not as much in the visible 532 nm range. Preliminary results show minerals discovered, such as perchlorate, fluoresce highest when excited by 355 nm light. Overall, we conclude the fluorescence instrument described is suitable to detect microbes, organics, biomolecules, and some minerals via fluorescence, offering a high scientific return for minimal cost with non-contact applications in extreme environments on Earth and on future planetary missions.

#### Accelerating Our Search for Life Beyond Earth with Privately Funded Robotic Space Missions

## Jon Morse, BoldlyGo Institute

Basic research in the space sciences holds essentially limitless potential for tackling profound questions of our existence and opening the doors of exploration, innovation and future economic opportunity. The search for life beyond Earth is a particularly compelling endeavor that is attracting significant private funding. The BoldlyGo Institute seeks to conduct privately funded, world-class space science missions that would tangibly accelerate our search for extraterrestrial life, feeding forward scientifically and technologically to future missions. BoldlyGo's initial portfolio includes a Mars robotic dust sample return mission and a UV-visible space telescope for the post-Hubble era that could host a coronagraph and be paired with a starshade. We describe the mission plans and the opportunities that such missions could provide in filling funding-driven gaps in the space science portfolio.

## iSEE: In-Situ Spectroscopic Europa Explorer

#### Pablo Sobron, SETI Institute

The in-situ Spectroscopic Europa Explorer (iSEE) is a next-generation ultra-compact Raman Spectrometer with superior performance that meets the top-level scientific requirements of the 2022 Europa lander mission. Our motivation is to build a small, versatile instrument that can address priority science goals in different spacecraft configurations (orbiters, flybys, landers, rovers). iSEE utilizes an innovative combination of light source, adaptive spatial coding optics, and detector. It integrates a high-performance signal processor and data processing algorithms that enable unprecedented measurements: in situ chemical identification and quantitation of complex organic compounds, including pre-biotic compounds (e.g. amino acids), biomolecules (organic biomarkers including proteins, lipids, and nucleic acid polymers), minerals, and volatiles. iSEE also provides sample context, including ice composition, crystallinity, and ice phase distribution. iSEE has potential to become a critical new instrument in NASA's exploration toolbox that can replace already-flown in-situ sensing technologies in future mission opportunities. It will deliver three game-changing advantages: (a) unprecedented Raman analytical capabilities-on-spectrometer quantitative analysis of organic content, minerals, and volatiles at or <1 ppb; (b) minimization of the cost and complexity of the light source system; and (c) possibility for novel mission architectures - organic, mineral, volatile analysis, and sample context, are offered within a single, ultra-compact instrument. The following missions highlighted by the Planetary Science Directorate will specifically benefit from iSEE: (a) landed exploration missions to Venus, the Moon, Mars, Europa, Titan, comets, and asteroids and (b) sample return missions to the Moon, Mars, comets and asteroids. In addition, iSEE may be used to identify and map available planetary in situ resources and to spur the development of autonomous in-situ resource utilization devices for robotic and human missions.

#### MapX: An In Situ, Full-Frame X-ray Spectroscopic Imager for the Biogenic Elements

## Dave Blake, Exobiology Branch/NASA ARC; Philippe Sarrazin, SETI; Kathy Thompson, SETI; and Thomas Bristow, NASA

Microbial life exploits microscale disequilibria at boundaries where valence, chemical potential, pH, Eh, etc. vary on a length scale commensurate with the organisms themselves-tens to hundreds of micrometers. These disequilibria can exist within cracks or veins in rocks and ice, at inter- or intra-crystalline boundaries, at sediment/ water or sediment/atmosphere interfaces, or even within fluid inclusions trapped inside minerals. The detection of accumulations of the biogenic elements C, N, O, P, and S at appropriate concentrations on or in a mineral/ice substrate would constitute permissive evidence of extant life, but context is also required. Does the putative biosignature exist in a habitable environment? Under what conditions of pressure, temperature, and chemical potential was the host mineralogy formed? MapX is an arm-deployed contact instrument that directly images the biogenic elements C, N, O, P, and S, as well as the cations of the rock-forming minerals (Na, Mg, Al, Si, K, Ca, Ti, Cr, Mn, and Fe) and important anions such as Cl and Fl. The instrument provides element images having  $\leq 100 \ \mu m$ lateral spatial resolution over a 2.5 cm by 2.5 cm area, as well as quantitative XRF spectra from ground-selected or instrument-selected Regions of Interest (ROI) on the sample. Quantitative XRF spectra from ROI can be translated into mineralogies using ground- or instrument-based algorithms. Either an X-ray tube source (X-ray fluorescence) or a radioisotope source such as 244-Cm ( $\alpha$ -particle and  $\gamma$ -ray fluorescence) can be used, and characteristic X-rays emitted from the sample are imaged onto an X-ray sensitive charge-coupled device through an X-ray MicroPore Optic. As a fluorescent source, 244-Cm is highly desirable in a MapX instrument intended for life detection since high-energy  $\alpha$ -particles are unrivaled in fluorescence yield for the low-Z elements. The MapX design as well as baseline performance requirements for a MapX instrument intended for life detection/identification of habitable environments will be presented.

#### **Curation of Deep Space Samples in Transit**

## Madhu Thangavelu, University of Southern California

Samples retrieved from deep space (e.g. asteroid or comet) by virtue of trajectories and energies required to bring them back to Earth, can take many months to years to reach Earth. During that period, changes can occur to those samples. Curation procedures are sought that can preserve the sample in as pristine of a condition as possible. Some ideas and recommendations are proposed.

#### Astrobionibbler: In Situ Microfluidic Subcritical Water Extraction

Aaron Noell, JPL; Anita M. Fisher, JPL; Nobuyuki Takano, JPL; Kisa Fors-Francis, Oklahoma State University; Stewart Sherrit, JPL; and Frank Grunthaner, JPL (retired)

Searching for trace levels of organic molecules on Mars or other rocky bodies is a formidable challenge, but impressive capabilities are being developed for reducing instrument size without losing performance for techniques such as gas chromatography with mass spectrometric detection or capillary electrophoresis with laser induced fluorescence detection. However, less work has been done to develop suitable instrumentation for analyte extraction and extract delivery to these analytical instruments. On Mars, the main driver for new extraction techniques is the difficulty that both the Viking Landers and Curiosity Rover have experienced with pyrolysis of samples; where degradation of the indigenous organics has occurred because of the high temperature breakdown and subsequent reactions of perchlorate salts.

The Astrobionibbler instrument (ABN) focuses on this problem, with the primary aim of developing a chip based fluidic device for subcritical water extraction (SCWE) from powder samples. In SCWE a pressurized system allows water to remain liquid at temperatures greater than 100°C (but less than the critical point at 374°C) and

perform accelerated extractions on a variety of samples. The high temperature allows water to behave like other less polar solvents because the dielectric constant of water changes dramatically with temperature. This enables molecular class targeted extractions based on polarity, a useful feature when trying to eliminate unwanted interferences for downstream instruments. The high temperatures reached in SCWE can also be used to hydrolyze biopolymers such as proteins into their constituent amino acids, increasing our ability to separate and conclusively detect potentially very small amounts of material. The work described here will focus on the development of the chip based ABN instrument, and tests performed on amino acid/protein extraction/hydrolysis.

## In-Situ Liquid Extraction and Analysis Platform for Mars and Ocean Worlds

Florian Kehl, D. Wu, M.F. Mora, J.S. Creamer, and P.A. Willis, JPL

Mars, Europa, Enceladus, and Titan are the most auspicious worlds to search for signatures of past or present alien life in our solar system. Here we present a compact, integrated sample extractor and analysis unit that could be used to support robotic missions seeking these chemical signatures of life. This wet chemistry instrument addresses habitability and the potential to preserve biosignatures by characterizing the local geochemical environment. In a first step, inorganic and putative organic compounds are automatically extracted from 1 cm<sup>3</sup> of regolith or ice/soil mixtures by subcritical water extraction at 175°C to 200°C and elevated pressures. Inline, miniaturized electrochemical probes quantify the eluate's pH, redox potential and electrical conductivity to better understand the ice or soil chemistry and mineralogy. Colorimetric measurements by flow injection analysis in a fully integrated mesofluidic manifold furthermore allow additional assessment of the soil's ionic composition. Besides the evaluation of the potential for past or present biology, this system can be employed as a front-end instrument for subsequent, more sophisticated organic analyzers, such as capillary electrophoresis or mass spectrometer units, to put these down-stream measurements in context.

## Capillary Electrophoresis Instrumentation for Determination of Chemical Distributions Indicative of Life on Future Spaceflight Missions

## Maria Mora, JPL/Caltech; F. Kehl, JPL; E. Tavares da Costa, JPL; J. Creamer, JPL; J. Chapman, SCIEX; D. Arnold, SCIEX; T. Horton, SCIEX; M. Darrach, JPL; A. Ricco; and P.A. Willis, JPL

The search for evidence of life beyond Earth is among the highest-level goals in planetary exploration. However, despite multiple orbiter and landed missions to extraterrestrial bodies in the solar system, we still haven't found evidence of life. A powerful approach in the search for life involves seeking biochemical signatures of life at the molecular level, as distributions of organic molecules. The liquid-based separation techniques capillary electrophoresis (CE) and its miniaturized version, microchip electrophoresis (ME) overcome the limitations of gasphase techniques and hold unique promise in the search for signatures of life on other worlds. Although multiple detection methods can be coupled to CE and ME, we focused on the two most powerful organic detection and characterization techniques: mass spectrometry (MS) and laser-induced fluorescence (LIF). LIF offers the highest sensitivity to organics, while MS allows complete identification. These techniques are complementary of each other and would allow full characterization of a sample in situ. Here we describe the status of instrumentation developed at JPL and the steps we are taking to someday enable its implementation on other worlds.

We describe a ME-LIF system we dub "The Chemical Laptop," which would provide the sample-processing capabilities required for in situ analysis with sub parts-per-billion sensitivity in a compact, low-mass, and low-power package. This instrument concept could be adapted to a variety of astrobiologically interesting targets like Europa, Enceladus, or Titan. This instrument is the first battery-powered and truly portable "end-to-end" ME-LIF astrobiology instrument capable of receiving an unlabeled liquid sample and performing all operations required for analysis.

We also present here the Organic Capillary Electrophoresis Analysis System (OCEANS) that couples capillary electrophoresis with electrospray ionization mass spectrometry (CESI-MS), in order to enable the characterization of distributions of organic compounds on future in situ planetary missions to ocean worlds.

#### The Search for Life with a Large Segmented-Aperture Space Telescope

Christopher Stark, Neil T. Zimmerman, Mamadou N'Diaye, Kathryn St. Laurent, Rémi Soummer, Laurent Pueyo, Anand Sivaramakrishnan, and Marshall Perrin, Space Telescope Science Institute (STScI), and Robert Vanderbei, Princeton University

The search for life on extrasolar planets requires the ability to detect and spectrally characterize planets 10 billion times fainter than their host stars. These measurements will likely be performed, at least in part, using a coronagraph to block stellar light. Historically, coronagraphs have been thought to perform at an acceptable level only with geometrically simple monolithic apertures, limiting the telescope diameter to roughly <4 m. Here I present a number of fundamental reasons why the search for life may require larger segmented apertures, ~12 m or greater, and show that new coronagraph designs may enable adequate performance for a large range of segmentation patterns.

## Sample Preparation Enabling Characterization of In Situ Biosignatures

Kathleen Craft, Christopher Bradburne, Matthew Hagedon, Jason Tiffany, and Matthew Grey, Johns Hopkins University Applied Physics Laboratory, and Antonio Ricco, Stanford University/NASA ARC

The detection of life outside Earth would be an incredible discovery, revolutionizing our perception of life and providing insight into how life develops and persists in various environments. The 2011 Planetary Science Decadal Survey puts emphasis on developing capabilities to enable the search for extraterrestrial life, including sampling environments for organisms possibly living now. However, no flight-qualified instrument currently exists that has the capability to definitively test for life in these environments, nor does the astrobiological community agree on one analysis technique/instrument that would determine, without a doubt, that life exists or that there is absence of life in a planetary environment.

The most robust strategy for searching for life in extraterrestrial environments would be to employ several techniques on a mission to corroborate the detections/non-detections. Possible techniques include chirality ratios, electron-transfer/redox gradients/disequilibrium, polymer detections, physical morphology characterizations, and organic detections. Adequate sample preparation for these analyses includes removals of salts and inhibitors. We present here a sample preparation and characterization process called COOL (Characterization of Organic Life), developed for detection of long-chained molecules in planetary in situ samples. COOL has been proven on planetary analog samples [1-3] and will reach flight readiness through evaluation of a low size weight and power sequencer that can detect extracted polymers and maturation of the sample separation and extraction preparation components [4].

Another important application of COOL is investigating how terrestrial organisms taken into space change within those environmental conditions. Increasing our understanding of biological adaptations to micro-g, radiation, various pressures, extreme temperature swings, etc., would provide insight into life that may have evolved on extraterrestrial bodies.

[1] Craft et al. (2014), LPSC 45, #2929; [2] Neish et al. (2012), AbSciCon, Atlanta, GA; [3] Bradburne et al. (2012), LPSC 43, #6043; [4] Craft et al. (2016), LPSC 47, #3035.

#### **REMOTE BIOSIGNATURES**

## **ExoPAG SAG 16 Report on Remote Biosignatures for Exoplanets**

Shawn Domagal-Goldman, NASA GSFC; Nancy Kiang, NASA Goddard Institute for Space Studies; Niki Parenteau, NASA ARC; and SAG 16

Future exoplanet observations will soon focus on the search for life beyond the solar system. Biosignatures to be sought are those with global, potentially detectable, impacts on a planet. Biosignatures occur in an environ-

mental context in which geological, atmospheric, and stellar processes, and interactions may work to enhance, suppress, or mimic these biosignatures. Thus biosignature science is inherently interdisciplinary. Its advance is necessary to inform the design of the next flagship missions that will obtain spectra of habitable extrasolar planets. The Exoplanet Biosignatures Workshop brought together the astrobiology, exoplanet, and mission concept communities to review, discuss, debate, and advance the science of biosignatures. This process engaged a broad range of experts by merging the interdisciplinary reaches of Nexus for Exoplanet System Science (NEXSS), the NASA Astrobiology Institute (NAI), NASA's Exoplanet Exploration Program (ExEP), and international partners, such as the European Astrobiology Network Association (EANA) and Japan's Earth Life Science Institute (ELSI). Between these groups, we had expertise in astronomy, planetary science, Earth sciences, heliophysics, biology, instrument/ mission development, and engineering. The workshop gathered these communities in the pursuit of three goals: (1) State of the Science Review: What are known remotely observable biosignatures. How can we develop a more comprehensive conceptual framework for identifying additional biosignatures and their possible abiotic mimics? (3) Confidence Standards for Biosignature Observation and Interpretation: What paradigm informed by both scientists and technologists could establish confidence standards for biosignature detection?

## Searching for Technosignatures

Jill Tarter, SETI Institute (retired) Co-authors: Martin Rees, Institute of Astronomy, Cambridge University; Michael Garrett, Jodrell Bank Centre for Astrophysics

Modifications to distant planetary environments by intelligent, technological life may be discoverable in ways not routinely investigated by astrobiologists intent on finding biosignatures. The detection of mathematicians in addition to microbes may be feasible with a search for technosignatures. Often such searches utilize the telescopic resources of the astronomers, across the electromagnetic spectrum. However, big data analytics focused on archival data from a wide range of scientific explorations, or the inclusion of detectors sensitive to artifacts among the toolkits deployed for in situ searches for biomarkers may also uncover evidence of technological civilizations. This poster summarizes historical and ongoing searches and forecasts those that may become possible with new facilities, detectors, and/or software.

#### On the Potential Use of Returned Samples from Mars in the Search for Life

David Beaty, JPL/Caltech, Hap McSween, University of Tennessee; Andy Czaja, University of Cincinnati; Yulia Goreva, JPL/Caltech; Libby Hausrath, University of Nevada; Lindsay Hays, JPL/Caltech; Chris Herd, University of Alberta; Munir Humayan, Florida State University; Francis McCubbin, NASA Johnson Space Center; Scott McLenna, SUNY at Stony Brook; Lisa Pratt, Indiana University; Mark Sephton, Imperial College; Andrew Steele, Carnegie; Ben Weiss, MIT; and Michael Meyer, NASA HQ

As recommended by the decadal survey *Visions and Voyages for Planetary Science in the Decade 2013-2022* (2011), a crucial element of our strategy to find evidence of life of Mars is Mars sample return. The scientific planning for this is currently being led by the Returned Sample Science Board within the M-2020 Project. A summary of this planning will be presented.

Key issues/topics/questions include:

• Discovering definitive biosignatures on Mars is judged by most to be not possible with current technology, due to the heavy burden of proof by the scientific community. Therefore, *studies of returned samples* in laboratories on Earth are key to confirming potential biosignatures identified by rovers on Mars, and turning them into definitive biosignatures.

• There is often spatial heterogeneity of biomarker distribution and preservation about any environment, and therefore *a suite* of returned samples from any region of interest is key to increasing the chances of identifying definitive biosignatures.

• Martian life is likely to be microbial. Therefore the biosignatures are likely to be *microscopic*, and require in situ and returned sample science analyses that function on that scale. Fine-scale observations are key.

• Organic molecules are crucially important type of potential biosignature, but since contamination is a given, it is important to have well-developed strategies for *contamination knowledge*. The Returned Sample Science Board has considered several potential options for mitigating this problem and presented them to the Mars 2020 project.

## False Negatives in Remote Life Detection: Lessons from Early Earth

## Stephanie Olson, University of California, Riverside; Christopher T. Reinhard, Georgia Institute of Technology; and Timothy W. Lyons, University of California, Riverside

The Earth's atmosphere has been an unfaithful reflection of its evolving surface chemistry and biology throughout its nearly 4-billion-year history of inhabitation. A particularly striking example from our history is the  $\sim$ 2.5-billion-year discrepancy between the earliest evidence for biological oxygen production and utilization on Earth and the accumulation of sufficient atmospheric oxygen to facilitate the remote recognition of an aerobic biosphere. Although the reasons for the delayed oxygenation of the atmosphere are not well understood, this delay highlights the likelihood of "false negatives" in the remote detection of life on exoplanets based on the identification of atmospheric biosignatures.

We have used an Earth system model to explore the fate of biogenic oxygen and other potential biosignature gases early in our history, and we evaluate the utility of classic biosignatures (e.g., the co-detection of oxygen and methane) for identifying and characterizing Earth's biosphere through time. We find that extended intervals of Earth's history may have appeared sterile based on atmospheric composition—despite major biological innovation within the ocean, including the origin of multicelluarity. At present, no single spectral feature could continuously identify life throughout Earth's history, and no combination of existing biosignature gases could reliably characterize any stage in the evolution of life on Earth. Importantly, Earth's cryptic biosphere arises naturally from the dynamics of ocean-atmosphere interaction in our model, and false negatives would not be unique to the early Earth; instead, false negatives may hinder remote detection of an aquatic biosphere on any Earth-like planet with an ocean at its surface. An implication is that the extrasolar bodies most likely to host life may actually be the worst candidates for remotely detecting and characterizing life.

## Oxygen in Exoplanet Atmospheres: Identifying True and "False Positive" Astronomical Biosignatures

Edward Schwieterman, University of California, Riverside; Victoria Meadows, NASA Astrobiology Institute Virtual Planetary Laboratory (NAI VPL) and University of Washington; Shawn Domagal-Goldman, NAI VPL and NASA GSFC; Timothy Lyons, NAI Alternative Earths and University of California, Riverside; Giada Arney, NAI VPL and NASA GSFC; Rory Barnes, NAI VPL and University of Washington; Chester Harman, NAI VPL and Pennsylvania State University; Rodrigo Luger, NAI VPL and University of Washington; Stephanie Olson, Alternative Earths and University of California, Riverside

The spectral signatures of molecular oxygen  $(O_2)$  and its photochemical byproduct ozone  $(O_3)$  are the most highly referenced and studied potential biosignatures in terrestrial exoplanet atmospheres. In previous years, mechanisms for generating oxygen by abiotic planetary processes, possible "false positives" for life, were believed to be limited to planets outside of the traditional habitable zone and therefore distinguishable by simple observables such as semi-major axis. However, recent modeling work has illuminated several plausible channels for generating detectable abiotic oxygen on planets inside the habitable zone, especially for those with M-dwarf host stars. These abiotic processes would produce potentially observable independent signatures that would fingerprint the abiotic source of  $O_2$ , such as CO from CO<sub>2</sub> photolysis and  $O_4$  from the accumulation of many bars of  $O_2$  from

116

117

massive water loss. Conversely, the detection of reduced gases, such as  $CH_4$ , in conjunction with  $O_2$  or  $O_3$ , would establish the presence of chemical disequilibrium and a more robust signature of life. In either case, we argue that strategies for detecting life on exoplanets must include characterization of a broad enough wavelength range to capture multiple gaseous absorbing species to provide maximal context. This poster details strategies for mitigating against "false positives" by identifying the complementary signatures whose presence or absence would strengthen the case for the photosynthetic (biogenic) origin of oxygen detected in an exoplanet atmosphere. We find that the near-infrared region of the planetary spectrum contains critical contextual information with ambiguity most reduced by extending spectral analysis to 5.0 microns.

## Global Surface Photosynthetic Biosignatures Prior to the Rise of Oxygen

Mary Parenteau, NASA ARC; Nancy Kiang, NASA Goddard Institute for Space Studies; Robert Blankenship, Washington University in St. Louis; Esther Sanromá, Instituto de Astrofísica de Canarias; Enric Pallé, Instituto de Astrofísica de Canarias; Tori Hoehler, NASA ARC; Beverly Pierson, University of Puget Sound; and Victoria Meadows, University of Washington

The study of potential exoplanet biosignatures—the global impact of life on a planetary environment—has been informed primarily by the modern Earth, with little yet explored beyond atmospheric  $O_2$  from oxygenic photosynthesis out of chemical equilibrium, and its accompanying planetary surface reflectance feature, the vegetation "red edge" reflectance. However, these biosignatures have only been present for less than half the Earth's history, and recent geochemical evidence suggests that atmospheric  $O_2$  may have been at very low—likely undetectable levels, until 0.8 Ga (Planavsky et al., 2014, Science 346:635-638). Given that our planet was inhabited for very long periods prior to the rise of oxygen, and that a similar period of anoxygenic life may occur on exoplanets, more studies are needed to characterize remotely detectable biosignatures associated with more evolutionarily ancient anoxygenic phototrophs.

We measured the surface reflectance spectra of pure cultures of anoxygenic phototrophs, and used these spectra to deconvolve complex spectra of environmental microbial mats from a variety of marine and continental environments. Rather than the "red edge," we observed "NIR edge(s)" due to absorption of NIR light by bacteriochlorophyll (Bchl) pigments. We initially expected only to detect the pigments in the surface layer of the mats. Surprisingly, we detected cyanobacterial Chl a in the surface layer, as well as Bchl c and Bchl a in the anoxygenic underlayers. This suggests that it does not matter "who's on top," as we were able to observe pigments through all mat layers due to their different absorption maxima. The presence of multiple "NIR edges" could signify layered phototrophic communities in marine and continental settings, which could possibly strengthen the support for the detection of life on the surface of an exoplanet.

#### The Limits of Organic Life in Planetary Systems

## David Smith, National Academies

The search for life beyond the Earth via in situ or remote-sensing techniques has a highly scientific, popular, programmatic, and political priority, but nothing would be more unfortunate than to expend considerable resources in the search for alien life and then not recognize it if it is encountered! To date, the search for life (e.g., by the Viking spacecraft on Mars in the 1970s) and/or planning for future searches has been governed by a model for life as we know it, so-called terran life. This approach is defensible in the absence of a general understanding of how life might appear if it had an origin independent of life on Earth. Plausible arguments can be made that if life originated independently, even within the solar system, it may not be detectable by missions carrying in situ or remote-sensing instruments designed explicitly to detect terran biosignatures. A committee established by the National Academies' Space Studies Board in 2006 published a report, *The Limits of Organic Life in Planetary Systems*, which attempts to address issues relating to the detection of hypothetical non-terran life. The motivation for the study, details concerning how the committee went about its task, and the report's principal findings and recommendations are discussed.

#### TARGETED PRECURSOR RESEARCH

#### **Strategies for Life Detection in Extraterrestrial Samples**

#### Catharine Conley, NASA HQ; Andrew Steele and Gerhard Kminek

Current international policy on performing biohazard assessments on samples brought from Mars to Earth is framed in the context of a concern for false-positive results. However, as noted during the 2012 Workshop for Life Detection in Samples from Mars (Kminek et al., 2014), a more significant concern for planetary samples brought to Earth is false-negative results because an undetected biohazard could increase risk to the Earth. This is the reason that stringent contamination control must be a high priority for all Category V Restricted Earth Return missions. A useful conceptual framework for addressing these concerns involves two complementary "null" hypotheses: testing both of them together would allow statistical and community confidence to be developed regarding one or the other conclusion. As noted above, false negatives are of primary concern for safety of Earth, so the "Earth safety null hypothesis"—which must be disproved to assure low risk to Earth from samples introduced by Category V Restricted Earth Return missions—is that "there is native life in these samples." False positives are primarily a concern for astrobiology, so the "astrobiology null hypothesis" - which must be disproved in order to demonstrate the existence of extraterrestrial life—is that "there is no life in these samples." The presence of Earth contamination would render both of these hypotheses more difficult to disprove. Both these hypotheses can be tested following a strict science protocol: perform analyses, interpret results, and select subsequent analyses that would increase confidence in the interpretation. The science measurements undertaken are done in an iterative fashion that responds to discoveries made, with both hypotheses testable by interpretation of the scientific data. This is a robust, community involved activity that ensures maximum science return with minimal sample use.

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# **Biographies of Committee Members**

JAMES KASTING, *Chair*, is a Distinguished Professor of Geosciences at Pennsylvania State University. He is well known as a world leader in the field of planetary habitability for his efforts to define the liquid water habitable zone around stars using one-dimensional, globally averaged climate models. His research focuses on atmospheric evolution, planetary atmospheres, and paleoclimates. He has published three books and more than 140 research papers and is a fellow of the American Geophysical Union (AGU), the American Association for the Advancement of Science, the Geochemical Society, the American Academy of Arts and Science, and the International Society for the Study of the Origin of Life (ISSOL). He was awarded the LExEN Award for his work "Collaborative Research: Methanogenesis and the Climate of Early Mars" and in 2008 received the Oparin Medal from ISSOL for "significant career contributions to the origin of life field."

WILLIAM BAINS is a visiting scientist researching astrobiology at the Massachusetts Institute of Technology (MIT), as well as an entrepreneur and teacher in the life sciences. His work touches several fields: regenerative medicine, astrobiology, business, and entrepreneurship. He is currently working with Sara Seager at MIT on what life could look like on planets with hydrogen atmospheres. In 1999 he founded Amedis Pharmaceuticals Ltd. (which was later acquired by Paradigm Therapeutics), and has since founded four other biotech companies and helped create over 10 others, as well as sitting on the advisory boards of the SULIS Fund, Iceni Fund, and Bath Ventures. Dr. Bains also runs Rufus Scientific, helping entrepreneurs, universities, and start-ups identify how to generate value from visionary science and technology.

TANJA BOSAK is an associate professor of geobiology in the Department of Earth, Atmospheric and Planetary Sciences at MIT. Her research is concerned with microbial fossils that reveal the parallel evolution of life and the environment. Her laboratory, which is part of the MIT NASA Astrobiology Team, Foundations of Complex Life, seeks to develop a quantitative understanding of the various morphological and geochemical biosignatures found in sedimentary rocks, in addition to studying the microfossil record associated with certain major climatic and geochemical oscillations in the Neoproterozoic Era. Her Ph.D. research investigated the role of microbial processes in the formation of laminated limestone rocks that were common for the first 80 percent of Earth's history. That work won her the 2007 Subaru Outstanding Woman in Science Award, which is presented to a woman whose Ph.D. research has impacted the field of the geosciences in a major way. Dr. Bosak spent 2 years as a microbial

sciences initiative fellow at Harvard University before she joined the faculty at MIT in 2007. She is an investigator on the Simons Collaboration on the Origins of Life and was awarded the James B. Macelwane Medal by the AGU, of which she is a fellow.

KEVIN P. HAND is the deputy chief scientist of the Solar System Exploration Directorate at the Jet Propulsion Laboratory (JPL) where he helps guide JPL's future for the robotic exploration of the solar system. He is also the founder of Cosmos Education and was its president until 2007. Dr. Hand studied psychology and physics as an undergraduate at Dartmouth College. He then went on to earn a master's degree at Stanford University in mechanical engineering while also working as a public policy research associate at Stanford's Center for International Security and Cooperation. Dr. Hand later completed his Ph.D. in geological and environmental sciences, also at Stanford. While a Ph.D. student, he was chosen by James Cameron to take marine biology samples from hydrothermal vents in subsea expeditions to the mid-Atlantic ridge and East Pacific Rise and was a featured scientist in Cameron's 2005 IMAX documentary "Aliens of the Deep." Dr. Hand is a recipient of the national Geographic Society Emerging Explorer Award and the Lew Allen Award for Excellence.

VICTORIA MEADOWS is a professor with the astronomy department and director of the Astrobiology Program at the University of Washington. She is also the principal investigator for the NASA Astrobiology Institute's (NAI's) Virtual Planetary Laboratory Lead Team. Dr. Meadows' primary research interests are in using modeling and observations to determine how to recognize whether a distant extrasolar planet is able to harbor life. Her NAI Virtual Planetary Laboratory team develops innovative computer models that can be used to understand the terrestrial planet formation process, test planetary dynamical stability and orbital evolution, and simulate the environment and spectra of present day and early Earth, other solar system planets, and plausible extrasolar terrestrial environments. In addition to her astrobiology research, Dr. Meadows remains a planetary astronomer, and her research interests also encompass remote-sensing observations and radiative transfer modeling of the lower atmosphere and clouds of Venus, the variable Earth, spectra of Titan and Neptune's atmospheres, and the impacts of Comet SL-9 with Jupiter.

PHILIP M. NECHES is the founder of Teradata Corporation and is a lead mentor and venture partner at Entrepreneurs Roundtable Accelerator in New York City. He is chairman of Foundation Ventures, LLC, an investment bank serving information technology and life science companies. Previously, Dr. Neches was vice president and chief technology officer of AT&T's Multimedia Products and Services Group and senior vice president and chief scientist at NCR. He is a director of Evolving Systems, Inc. and a trustee of the California Institute of Technology, where he earned his B.S., M.S., and Ph.D. in computer science.

NILTON O. RENNO is a professor in the Department of Climate and Space Sciences and Engineering at the University of Michigan. He is also chair of the department's master's programs and director of the master of engineering program in space engineering. Dr. Renno's research interests include aerosols and climate, astrobiology, instrument development, planetary science, systems engineering, and thermodynamics. He studies the physical processes that control the climate of Earth and other planets, and works on the design and fabrication of instruments for this purpose. Previously, Dr. Renno was a tenured associate professor in the Department of Planetary Sciences at the University of Arizona. He has received the Space Foundation John L. "Jack" Swigert Jr. Award for Space Exploration, the American Institute of Aeronautics and Astronautics Foundation's Award for Excellence, and the National Aeronautic Associations 2012 Robert J. Collier Trophy for his work on NASA's Mars Science Laboratory Team and the Curiosity Rover Mission, as well as several NASA Group Achievement Awards.

GARY RUVKUN is a molecular biologist at Massachusetts General Hospital and professor of genetics at Harvard Medical School. He discovered the mechanism by which lin-4 (the first microRNA) regulates the translation of target messenger RNAs via imperfect base-pairing to those targets, and discovered the second miRNA, let-7, and that it is conserved across animal phylogeny, including in humans. These miRNA discoveries revealed a new world of RNA regulation at an unprecedented small size scale, and the mechanism of that regulation. Dr. Ruvkun also discovered many features of insulin-like signaling in the regulation of aging and metabolism. The Ruvkun

APPENDIX E

laboratory has started work with the Church Laboratory and engineers at MJ Research and the MIT Center for Space Research to develop a miniature thermal cycler and protocols to send to Mars in search of microbial life. Dr. Ruvkun has received numerous awards for his contributions to medical science, particularly his study of microR-NAs. He is a recipient of the Lasker Award for Basic Medical Research, the Gairdner Foundation International Award, and the Benjamin Franklin Medal in Life Science. In 2008, Dr. Ruvkun was elected as a member of the National Academy of Sciences.

NITA SAHAI is an Ohio Research Scholar Professor in the Department of Polymer Science at the University of Akron. Her research interests include biomolecular and cellular interactions with biomaterials and minerals, interfacial chemistry, the origins and early evolution of life, and the relationship between molecular-level, nanoscale, and macroscopic properties. Her research group is working determine the potential role of mineral surfaces in the evolution of cell surfaces. Previously, Dr. Sahai was a professor of geochemistry at the University of Wisconsin, Madison, where she received the Romnes Faculty Fellowship. She has served as an editor on a number of publications, including *Medical Mineralogy and Geochemistry*, *Reviews in Mineralogy and Geochemistry Series*, *American Mineralogist*, and *Geochemical Transactions*. She is an investigator on the Simons Collaboration on the Origins of Life, a recipient of the NSF CAREER award, a fellow of the Mineralogical Society of America, as well as the latter Society's Distinguished Lecturer for 2013-2014.

DIMITAR SASSELOV is a professor of astronomy at Harvard University and the founding director of the Harvard Origins of Life Initiative, an interdisciplinary institute that joins biologists, chemists, and astronomers in searching for the starting points of life on Earth. A co-investigator for Kepler, in 2002, Dr. Sasselov and his team sighted OGLE-TR-56b, a planet in the constellation Sagittarius that was the farthest planet from the Earth discovered until then (1500 pc away). His research interests include both exoplanets and the interaction between radiation and matter. He also studies how planetary conditions may act as the seedbed of life, and how knowing the composition and conditions of a planet could teach us how life might form there. Dr. Sasselov is the author of the book *The Life of Super-Earths: How the Hunt for Alien Worlds and Artificial Cells Will Revolutionize Life on Our Planet*.

MARK H. THIEMENS is the dean of physical sciences, Distinguished Professor of Chemistry, and Chancellor's Associates Chair in the Department of Chemistry and Biochemistry at the University of California, San Diego. His research is centered on use of the mass-independent fractionation process for stable isotopes to study the origin and evolution of the solar system from meteorite analysis, definition of the source and transformation of greenhouse gases in the troposphere, chemistry of the stratosphere and mesosphere, chemistry of the ancient martian atmosphere, and the origin and evolution of oxygen-ozone and life in the Earth's Precambrian. His climate work has included field work at the South Pole, Greenland summit, Mt. Everest, and the rainforests of South America. His work also includes studies of the origin and evolution of life on Earth, especially the oxygen evolution, and includes field sampling in China. Dr. Thiemens is a member of the National Academy of Sciences and American Academy of Arts and Sciences and has been recognized with the E.O. Lawrence Medal from the Department of Energy, the Goldschmidt Medal of the Geochemical Society, and several honorary professorships.

MARGARET TURNBULL is an astrobiologist at the SETI Institute. Her research expertise is in identifying planetary systems that are capable of supporting life as we know it. She is currently principal investigator for the Wide Field Infrared Survey Telescope (WFIRST) exoplanet imaging coronagraph, and for a WFIRST Preparatory Science Team on habitable exoplanet colors and spectral signatures, in addition to chairing the WFIRST Coronagraph Target Selection Working Group. She also serves on the Hab-Ex Science and Technology Definition team to define a flagship scale space telescope plus starshade mission to find habitable worlds amongst the Sun's nearest neighbors. Previously, Dr. Turnbull developed a Catalog of Habitable Stellar Systems with Jill Tarter (called HabCat) for use in the search for extraterrestrial intelligence, and she has studied the spectrum of the Earth to identify telltale signatures of life. She is a member of NASA's Exoplanet Planning and Analysis (ExoPAG) Executive Committee, a co-investigator on the Arizona State University's "Exoplanetary Ecosystems" NExSS team, and a co-author of NASA's probe-scale Exo-S telescope plus starshade concept study. Searching for Life Across Space and Time: Proceedings of a Workshop