

Assessment of Planetary Protection and Contamination Control Technologies for Future Planetary Science Missions

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Jet Propulsion Laboratory California Institute of Technology Pasadena, California

Assessment of Planetary Protection and Contamination Control Technologies for Future Planetary Science Missions

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Andrea Belz, Consultant, Jet Propulsion Laboratory, California Institute of Technology, Lead Author Pat Beauchamp, Jet Propulsion Laboratory, California Institute of Technology, Chair

Advisory Committee

NASA Headquarters

Catharine Conley Perry Stabekis Jack Barengoltz

NASA Ames Research Center Scott Sandford

NASA Goddard Space Flight Center Jason Dworkin Therese Errigo Stephanie Getty Daniel Glavin

NASA Johnson Space Center Michael Zolensky

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John Hopkins University / Applied Physics Laboratory Thomas Magner

Foreword

Planetary protection and organic contamination control, like many technologically rich areas, continually progress. This assessment describes advances in both areas since the first report was generated in 2005, when the primary emphasis was on technologies for *in situ* missions to Mars. As a result of the 2011 Planetary Science Decadal Survey Report, *Vision and Voyages for Planetary Science in the Decade 2013–2022*, the focus is now on a sequence of Mars sample return missions. Thus, in this report, we examine our experiences in returning solar wind and cometary samples, which teach us how to better prepare for returning samples from Mars. It has become clear that linking planetary protection and contamination control requirements and processes together early in the mission development and spacecraft design is key to keeping mission costs in check and returning high-quality samples that are free from biological and organic contaminants. Scientific integrity is a priority.

In addition to Mars, we now have the exciting possibility of a potential mission to the outer planets, most likely Europa. Discussions and debate have occurred in the last few years to firm up the planetary protection requirements for such a mission. At the time of this report's publication, there are three options for the Europa mission ranging from multiple fly-bys, to an orbiter or a lander. The planetary protection and contamination control requirements will, of course, depend heavily on the chosen mission.

This report provides the status of planetary protection and contamination control technologies as they apply to potential missions and provides recommendations to improve our capabilities as we further explore our solar system.

htmin M. Keauchamp

Patricia M. Beauchamp Strategic Missions and Advanced Concepts Office Solar System Exploration Directorate

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Other Reports in This Series

Power Technology

- Advanced Radioisotope Power Systems Report, Report No. JPL D-20757 6/01, March 2001.
- Solar Cell and Array Technology for Future Space Missions, Report No. JPL D–24454, Rev. A, December 2003.
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In Preparation

- Guidance and Control Technology Assessment for Future Space Missions
- Navigation and Mission Design Technology Assessment for Future Space Missions

Table of Contents

Exe	ecutive	Summary	1			
1	Study 1.1 1.2	Overview Introduction Requirements 1.2.1 Planetary Protection and Regulatory Agencies 1.2.2 Requirements for Contamination Control 1.2.3 Exploration Targets	4 5 5 7			
2	Plann 2.1 2.2	ned and Potential Future Mission Concepts Mars Exploration Other Solar System Exploration				
3	Asses 3.1 3.2 3.3	sment of Technology Progress Contaminant Reduction and Assessment. 3.1.1 Microbial Reduction Methodologies. 3.1.2 Bio-burden Detection and Assessment. 3.1.3 Bio-diversity Studies. 3.1.4 Organic Contamination Control and Assessment. 3.1.5 Assured Contamination Control and Assessment. 3.1.4 Organic Contamination Control and Assessment. 3.1.5 Assured Containment Samples Returned from Mars Recontamination Prevention	13 14 18 20 22 23 24 24 24 24 26 28 28 28			
4	Key F 4.1 4.2 4.3	ndings and Recommendations Systems Engineering Technology Development Education and Training	29 30			
Acr	Acronyms					
Ref	ference	5	33			

List of Tables

Table 1-1. COSPAR listing of target body/mission types by planetary protection category.	6
Table 2-1. Planned planetary protection implementation for exploration of Mars	9
Table 2-2. Planned planetary protection implementation for solar system exploration	11

List of Figures

Figure 3-1. Radiation fluxes at Europa. <i>Left:</i> Particle flux for electrons, protons, oxygen, and sulfur ions [49]; <i>right:</i> radiation spectra at geosynchronous (GEO) and Europa orbit [50]. At Europa orbit, the Jupiter radiation environment contains significantly higher fluxes of both high-energy protons and electrons than Earth's orbit	17
Figure 3-2. Summary of the status of microbial reduction technologies, describing sterilization modalities. There are two issues for each modality represented: 1) its progress toward NASA approval as a sterilization technique (fir line) and 2) compilation of a hardware compatibility chart (second line).	st
Figure 3-3. Summary of bio-burden detection and assessment technologies.	20
Figure 3-4. Summary of research in bio-diversity studies	21
Figure 3-5. Summary of technologies for contaminant transport	26
Figure 3-6. Exploded view of the Viking spacecraft illustrating the bio-shield, jettisoned when the descent capsule began entry, descent, and landing. (Courtesy of Flight International)	27
Figure 3-7. Viking engineers preparing the spacecraft for sterilization	27
Figure 3-8. The Phoenix bio-barrier. The robotic arm is shown encased in the bio-barrier, held in place by a series or latches released shortly after landing by a pyro-activated pin puller. Torsion springs at each end then rotated the ribs (right to left in the figure) to open the enclosure [77].	e
Figure 3-9. Summary of isolation technologies	28

Executive Summary

Recent exploration of the solar system has revealed previously unknown extraterrestrial environments on which life could conceivably survive and even thrive. Simultaneously, the understanding of the astonishing diversity of habitable environments on our own planet has increased dramatically; we are beginning to recognize the vast array of living systems able to convert nearly any energetically favorable chemistry locally available into novel forms of metabolism and respiration. Taken together, this breadth of discovery has inspired a new generation of global research designed to seek and understand extraterrestrial habitability.

As exploration begins to hone in on the differences between "prebiotic," "habitable," and "inhabited," strong practices in planetary protection will be critical to guaranteeing the quality of returned science and returning samples safely to Earth. At the same time, the desire to understand the origin and fate of organic molecules in prebiotic systems leads to the need for strong practices in contamination control to protect the integrity of sampling sites. These practices can be daunting in complexity because they dovetail with instrument and spacecraft design, as well as assembly, test, and launch procedures.

The recent era of Mars exploration has already motivated a number of recent advances in planetary protection. Microbiologists have infused modern molecular techniques in planetary protection research. We now have a body of knowledge on the microbial ecology of the spacecraft assembly facility, as well as that of extreme environments previously seen as hostile to life. Similarly, the new generation of *in situ* scientific instruments has led to novel contamination transport models to demonstrate low risk of contamination of scientific experiments.

This document reassesses planetary protection and organic contamination control technologies, which were evaluated in 2005, and provides updates based on new science results, technology development, and programmatic priorities. The study integrates information gathered from interviews of a number of National Aeronautics and Space Administration (NASA) and European Space Agency (ESA) scientists, systems engineers, planetary protection engineers, and consultants, as well as relevant documents, and focuses on the technologies and practices relevant to the current project mission set as presented in the 2011 Planetary Science Decadal Survey.

The Mars Exploration Program's recommendation to build on the successful "Follow the Water" campaign with a new "Seeking Signs of Life" program will likely require meeting increasingly stringent planetary protection requirements. Missions following this new approach include the Trace Gas Orbiter (TGO) and the potential Mars Sample Return (MSR) campaign. While TGO does not require novel planetary protection technologies, the MSR missions, which would cache Mars samples for eventual return to Earth, would require new technology to meet the requirements of sample transportation, biohazard assessment, and time-critical scientific assessment of the samples under both stringent cleanliness conditions and highly reliable bio-containment. Other solar system exploration over the next two decades, including potential missions to Europa and Ganymede, Titan/Enceladus, and comet surfaces, require that planetary protection and contamination control be considered in both the design of the spacecraft and mission.

Although planetary protection and contamination control requirements are derived from different sources, they share the same general approach and many of the same analytical tools,

and benefit from similar education and training programs. One important difference, however, is that planetary protection technologies and procedures have to undergo exacting verification and validation processes in order to comply with international and NASA regulations. The approach to both fields includes contaminant reduction and assessment, recontamination prevention, modeling to support quantitative risk assessments, and development of long-term curation infrastructure. This report highlights the similarities and the need for both communities to work together. Since the last assessment in 2005, many planetary protection technologies have matured, particularly in microbial reduction and validation. There have also been significant improvements in bio-burden detection and assessment, bio-diversity studies, contaminant transport, and isolation techniques. Education and training in this field of research has also increased.

Recommendations stemming from the current assessment include the following improvements over the already impressive progress in planetary protection and contamination control technologies since 2005.

Systems Engineering

<u>Recommendation</u>: The elements of contamination control and planetary protection that are critical to mission planning, science, and hardware design must be a fundamental part of the systems engineering and must be addressed at the earliest stages of the mission to ensure proper flow-down of requirements and cost-effective mission planning. An adequate approved materials/parts list that can accommodate both contamination control and planetary protection considerations should be developed. Integrated modeling tools should be developed to aid systems engineers and designers for future work, particularly in the form of risk assessments for forward- and back-contamination. Also, planetary protection implementers should define and engage systems engineering approaches to determine traceable requirements that can be flowed down within projects early in the process. Cost estimating tools should be developed.

Technology Development

<u>Recommendation A:</u> A streamlined approval process should be developed, as well as instruction on the newly available forward-planetary protection techniques. Plans for MSR technology development related for assured containment must be carefully coordinated with concept studies and formulation efforts.

<u>Recommendation B:</u> The effect of non-uniform molecular contamination on micron and submicron particle contamination levels should be determined.

Education and Training

<u>Recommendation A</u>: Solicitations for early instrument technology development should include requirements for planetary protection. Education and training should be offered to all interested proposers at a level commensurate with the proposed efforts. In some cases, proposers might be advised to take the excellent planetary protection class offered by the NASA Planetary Protection Office (PPO). In other cases, ensuring the proposer is sufficiently educated with respect to the system implications of planetary protection and contamination control may be adequate. All proposals should be required to delineate their approach to planetary protection and contamination control if applicable.

<u>Recommendation B:</u> NASA should support the creation of a living document detailing experiences with contamination control and curation for previous missions, to help present and

future missions avoid costly mistakes. This document could be constructed around a wiki, permitting information to be collected from the widest possible range of persons. In addition, it could be included in the NASA Lessons Learned program. The timing of this recommendation is critical since the generation of Apollo scientists and technicians is quickly disappearing.

1 Study Overview

The purpose of this document is to describe the technological state of the practice of planetary protection and organic contamination control in order to identify the needs for robotic missions envisioned in the next decade of exploration. The team who authored this document conducted a similar technology and needs assessment in 2005.¹ The current study revisits the original technology assessment with the intention of updating the technology needs in light of new science results, technology development, and programmatic priorities. In addition, the set of missions in the planning stages has been significantly revised since 2005, and thus, this assessment focuses on technologies and practices relevant to the current projected mission set.

Over a six-month period in 2010, the study lead author interviewed a number of scientists, systems engineers, planetary protection engineers, program officers, and consultants from NASA and ESA. The study team collected information describing the state of the art in planetary protection practice and organic analysis. The team then combined this information with the current understanding of missions in the planning stages, and revisited them after the 2011 Planetary Science Decadal Survey was published. This allowed the team to identify the needs with highest priority in meeting the envisioned mission objectives. The Steering Committee jointly created the findings and recommendations.

While human exploration also poses new challenges to planetary protection to Mars² and the Moon,³ this study focuses on the needs for robotic exploration. Although this study does not specifically call out the cross-cutting nature of the technologies described here, some are likely to serve needs in human exploration as well.

This type of study will continue to require revisions and updates as our scientific understanding and technology development both continue to evolve.

1.1 Introduction

In the last twenty years, our exploration of the solar system has revealed previously unknown extraterrestrial environments, both current and ancient, in which life could conceivably survive and even thrive. Simultaneously, we have a deeper understanding of the diversity of habitable environments on our own planet, supporting the opportunistic nature of living systems able to exploit nearly any energetically favorable chemistry.

As a result, international space exploration is now undertaking an extensive experimental program designed to study extraterrestrial chemistries that could shed light on the differences between "prebiotic," "habitable," and "inhabited." This program calls for both increased capabilities in determining habitability prospects *in situ* as well as potentially returning samples from Mars back to Earth for further study. These research objectives bring added design complexity, as we must maintain the scientific integrity of the ever-smaller samples themselves as well as their site of origin. In addition, sample return from potentially habitable planetary environments presents new challenges as we face an increased need for containment systems that protect the native biosphere.

These goals—protecting the scientific integrity of sites on planetary bodies for future research, as well as safely returning extraterrestrial samples to Earth—jointly motivate the field of planetary protection. Planetary protection has policy elements, as it encompasses international agreements governing extraterrestrial research, as well as implementation challenges in the procedures developed to satisfy these top-level requirements. This dual policy-implementation nature of planetary protection makes technology-planning exercises particularly formidable. Certain areas of investigation, such as understanding and characterizing the full scope of

potentially habitable environments, are never "complete" and in fact will only grow in complexity. Technologies to assess and remove contaminants do reach a state of "readiness" but must then be certified by national space agencies wishing to adopt them, causing infusion to lag significantly behind technology maturity.

As the need for planetary protection technologies has grown, NASA has attempted to evaluate the needs through standard scales such as technology readiness levels (TRLs). However, this methodology does not capture the state of the art because of the interplay between technology, policy, and formal NASA approval and certification. Thus, while TRLs were used to assess technology development in the 2005 assessment, this study implemented a simpler scheme to illustrate progress in fundamental research areas and gaps that still remain. In addition, this assessment explicitly highlights the overlap with organic contamination control to emphasize areas where resources can be leveraged to meet multiple needs.

In the spring of 2011, the National Research Council released the Decadal Survey *Visions* and *Voyages for Planetary Science in the Decade 2013–2022*,⁴ identifying the mission concepts of highest priority in the coming years. This document describes a proposed mission set that is flexible within the funding environment. In these missions, planetary protection poses a unique challenge because the work ranges from characterizing the microbial diversity in a specific environment to building sterilizable instrument components and subsystems. Similarly, organic contamination control requires fundamental research in addition to minimization and assessment procedures. As a result, these fields can slip through the cracks because of the natural tension between a programmatic goal of funding long-term cross-cutting capabilities and the challenges of a nuts-and-bolts implementation. Too often these requirements have subsequently been imposed late in the mission life-cycle on flight projects that are poorly informed on the topic, naturally risk-averse, and short of development funds.

Thus, although the requirements flow down from different sources, and the expertise for planetary protection and contamination control reside in separate technical areas within NASA, it is worthwhile to address the needs of both planetary protection and organic contamination control in an integrated manner. This study revisits the 2005 assessment with the intention of updating the technology needs in light of new science results, technology development, and current programmatic priorities. This report emphasizes the technologies and capabilities required by potential future missions.

1.2 Requirements

Planetary protection and organic contamination control share related technologies and educational needs, but stem from requirements imposed by different sources. This section briefly introduces the origin of the requirements.

1.2.1 Planetary Protection and Regulatory Agencies

Several agencies and committees are involved with making and implementing planetary protection policy. These include the Committee on Space Research (COSPAR), NASA, ESA, the Japan Aerospace Exploration Agency (JAXA), the Russian Space Agency, etc., and their advisory boards.

Because Rummel and Meltzer have each written an excellent detailed history of planetary protection,^{5,6} we will simply summarize the key elements of the policies and practice. Signatories to the 1967 "Outer Space Treaty"⁷ must follow guidelines to both preserve the integrity of future exploration studies and protect the terrestrial biosphere.

Category	Architecture	Target
I	Any	Sun; undifferentiated, metamorphosed asteroids; lo; others TBD
II	Any	Venus, Moon (with organic inventory), comets, carbonaceous chondrite asteroids, Jupiter, Saturn, Uranus, Neptune, Ganymede,* Callisto, Titan,* Triton,* Pluto/Charon,* Ceres, Kuiper-Belt Objects > 1/2 the size of Pluto,* Kuiper-Belt Objects < 1/2 the size of Pluto, others TBD
III	Orbiter, flyby	Mars, Europa, Enceladus, others TBD
IV	Lander	Mars, Europa, Enceladus, others TBD
V	Sample return to Earth	"Restricted Earth return": Mars, Europa, others TBD; "Unrestricted Earth return": Venus, Moon, others TBD

 Table 1-1. COSPAR listing of target body/mission types by planetary protection category.

* The mission-specific assignment of these bodies to Category II must be supported by an analysis of the "remote" potential for contamination of the liquid-water environments that may exist beneath their surfaces (a probability of introducing a single viable terrestrial organism of <1 x 10⁻⁴), addressing both the existence of such environments and the prospects of accessing them.

COSPAR

Since 1967, the guidelines have been updated by COSPAR of the International Council of Science Unions (ICSU), which both informs and is informed by advisory groups of the international space exploration community in developing its planetary protection policies. Policies are reviewed every other year to allow the organization to keep pace with scientific discoveries and mission planning. COSPAR's recommendations are then adopted by national space agencies, which, in turn, implement them in their own policies and procedures. COSPAR policy calls for assigning a planetary protection category to each mission, taking into account the mission architecture and target solar system body.

Table 1-1 summarizes the COSPAR categorization for various mission architectures and exploration targets. However, for some missions, such as landed missions on Mars, there may be added nuances due to the selected landing site and spacecraft design (such as the Phoenix mission, in which the sampling arm was subject to more stringent requirements than the rest of the spacecraft because of its contact with the Mars subsurface).

NASA

The NASA Planetary Protection Officer⁸ determines planetary protection categorization for NASA missions, ensures compliance with COSPAR policies, and approves planetary protection plans prior to launch. The Planetary Protection Officer is further advised by the NASA Planetary Protection Subcommittee and by the Space Studies Board of the National Research Council.

NASA requirements for planetary protection are specified in:⁹

- NPD 8020.7, Biological Contamination Control for Outbound and Inbound Planetary Spacecraft
- NPR 8020.12, Planetary Protection Provisions for Robotic Extraterrestrial Missions
- NASA HDBK 6022, NASA Handbook for the Microbial Examination of Space Hardware (formerly issued as NPR 5340.1)

ESA, JAXA, and RosCosmos

ESA has a Planetary Protection Working Group (PPWG), reporting to the Life and Physical Science Advisory Committee (LPSAC),¹⁰ as well as a formally appointed Planetary Protection Officer reporting to TEC-Q. ESA guidelines¹¹ differ slightly from NASA's, but are still consistent with COSPAR regulations. Currently, ESA and NASA are formally coordinating the

requirements documents, with the intent that the wording in the future should be identical. Note that all spacefaring nations are signatories to the Outer Space treaty and are members of COSPAR, which is the governing body for planetary protection; missions such as Nozomi (JAXA) as well as Mars 96 and Phobos-Grunt (RosCosmos) implemented planetary protection strategies for their spacecraft hardware

1.2.2 Requirements for Contamination Control

Unlike planetary protection, organic contamination control requirements flow down principally from science requirements, and thus are implemented in instrument and spacecraft design during the lifetime of the project. These requirements are typically more stringent than those that would normally be imposed on spacecraft design and assembly (for instance, to preserve the performance of optical components or maintain the chemical integrity of a sample to be analyzed *in situ*).

Principal investigators for instruments usually determine the impact of contamination on the integrity of their experiments; if potential sources of contamination from other elements of the spacecraft may be transported to a sample chamber, it may be necessary to engineer a solution in concert with spacecraft engineers. For this reason, systems engineers that understand, monitor, and mitigate the interactions of the instruments with the rest of the spacecraft are a critical element to successful contamination control.

1.2.3 Exploration Targets

Several exploration missions in the planning phases pose challenging science requirements that call for extensive requirements for planetary protection and organic contamination control. This subsection summarizes these issues.

Targets of Planned and Potential Missions within the Decade 2013–2023

Mars

Mars continues to be a major exploration target¹² because it is the planet most like Earth in the solar system, and thus motivates a search for extant or extinct life; its surface and subsurface retains a chemical, physical, and geological record of a planet whose environment has changed dramatically over time and thus may help inform about a planet's transition from "habitable" to "inhabited." To date, no organic molecules have been found on Mars, but robotic missions, specifically the current Mars Science Laboratory (MSL), continue to follow signs of water, investigate habitability, and search for life or indications of biological activity on Mars.

In anticipation of the direction of future Mars exploration, the Committee on Preventing the Forward Contamination of Mars identified four key planetary protection objectives in its 2006 report:¹³

- Assessment of spacecraft contaminants
- Definition and development of revised requirements for reduction of bio-burden
- Improvement of bio-burden reduction techniques
- Validation of and transition to new standards and techniques

In 2007, the Committee on the Astrobiology Strategy for the Exploration of Mars issued a report with a number of key recommendations,¹⁴ including suggestions that NASA should target its astrobiology science in locations where liquid water could exist or have existed, and that samples should emphasize those with the best chance of retaining bio-signatures. This

philosophy is echoed by the Mars Exploration Program, which has recommended building on the remarkably successful "Follow the Water" campaign of the last ten years with a "Seeking Signs of Life" program.¹⁵ By its nature, the approach to astrobiology and to Mars exploration will likely require meeting stringent planetary protection requirements.

Europa and Ganymede

Even though the surface of the Jovian satellite Europa is bombarded by Jovian radiation and thus presumed inhospitable to life, a liquid ocean below the icy crust¹⁶ could make a habitable environment for terrestrial organisms, and possibly for indigenous life as well. The liquid ocean, likely warmed by tidal heating of the ice shell and possibly hydrothermal activity on the ocean floor, makes an attractive target for astrobiology studies. Planetary protection analysis for Europa has focused on the need for effective microbial reduction techniques to prevent contamination of the ocean, which may not be fully isolated from the surface,¹⁷ as suggested by recent studies of the Thera Macula region that may harbor a near-surface lake; these requirements are not limited to landed missions, as all current concepts for orbital missions call for disposal onto the surface of Europa.

In contrast to Europa, the oldest terrains of the Jovian moon Ganymede appears to be much older, with ages of various regions estimated to be 400 million years to 4 billion years old.¹⁸ It is thought that Ganymede probably has a thick, cold near-surface icy layer that prevents communication with the interior. Models of their interior based on the complex nature of the water phase diagram suggest that it may possess deep liquid oceans (more than 150 km below the surface), "perched" or "sandwiched" between a thick crust of low-density Ice I and an icy mantle of high-density Ice III, with completely or partially differentiated silicate or silicate plus ice below. Thus, planetary protection requirements will not be as stringent as those required for Europa.

Targets of Potential Future Missions

Titan and Enceladus

Saturn's moon Titan poses challenges because the rich organic environment that may provide clues to prebiotic organic chemistry,¹⁹ particularly because the low ambient temperature of 94 K, acts to retard chemical reactions characteristic of biological systems on Earth. The sheer abundance of methane and its organic products exceeds that of Earth's ocean, biosphere, and fossil fuel reservoirs by more than an order of magnitude.²⁰ As a result, some organic contaminants may interfere with *in situ* analysis, presenting challenges to data interpretation. This mission therefore will likely call for strong organic contamination control. However, as far as planetary protection is concerned, there is a great deal of uncertainty whether there is contact between liquid oceans found deep in the interior of Titan and the surface. The recent discovery of a suspected cryo-volcanic structure, known as Sotra Facula, points to the interaction of the surface with the sub-surface, but it is not clear from what depth this upwelling arises. Further, it is not known whether there is contact between the liquid and the silicate constituents in recent geological times. Consequently, Titan has received a Category II designation for planetary protection.

The tiny moon Enceladus poses different challenges because of the presence of water vapor, ice, sodium, and organics in geysers emanating from its south polar region.²¹ Simple organics including benzene have been observed in the plume, and detected salts imply expulsion of liquid

water that has been in contact with rock. Depending on the exact trajectory and science scenarios, a mission to this body may require more exacting planetary protection requirements.

Comets or Asteroids

The search for habitability calls for developing a better understanding of the inventory and evolution of organic compounds and water throughout the solar system. Both comets and primitive asteroids shed light on these questions because they represent a clean record of the conditions in the early solar system and may have played a role in delivering water and organics to the early Earth.²² Therefore, it would be highly informative to study a pristine sample delivered from the surface of a comet. Like research at Titan, this mission would not fall under strong planetary protection guidelines but would call for conforming to strict organic contamination control requirements.

2 Planned and Potential Future Mission Concepts

This section briefly summarizes the missions facing strong requirements from either planetary protection or organic contamination control. Using the anticipated launch dates currently in NASA plans, this section also presents an integrated project timeline.

2.1 Mars Exploration

The last fifteen years of Mars exploration have revolutionized our understanding of the planet's history. Recent missions implemented the "Follow the Water" theme of NASA's Mars Exploration Program to reveal a complex geological history in which liquid water once flowed on the surface and now may remain in the subsurface. Because it is so similar to Earth and so accessible, Mars provides a compelling platform to test our general models of life-detection experiments. Furthermore, it may serve as an outpost for human exploration of the solar system.

The success of the "Follow the Water" theme has evolved into the new "Seeking Signs of Life" theme proposed recently by the Mars Exploration Program Advisory Group (MEPAG). NASA and ESA are jointly embarking on an exciting program to better elaborate on the differences between "habitable" and "inhabited" worlds. Table 2-1 summarizes the planned missions following this new theme and details the planned planetary protection implementation for each.

Immediately prior to these missions, MSL (launched in 2011) and the Mars Atmosphere and Volatile Evolution Mission (MAVEN) (scheduled for launch in 2013), will further enhance our knowledge of the habitability and history of Mars. Planetary protection implementation technologies for these spacecraft are largely a refinement of previous methodologies. MSL has adopted many heritage technologies from the Mars Exploration Rover (MER) mission, with the additional rigor attendant with having a bigger spacecraft but the same bio-burden cap. MAVEN is planning to meet its planetary protection requirements with a bio-burden reduction approach, similar to that used for the Mars Reconnaissance Orbiter (MRO) mission.

Mission	Study Phase	Category	Planned Implementation
TGO	Phase A	=	Implementation unlikely to require new technologies
Mars 2018 Rover	Pre-Phase A	IVb / V	Forward-protection implementation unlikely to require new
MSR Lander	Pre-Phase A	IVb / V	technologies; significant challenges in sample handling
MSR Orbiter	Pre-Phase A		and back-protection
MSR SRF	N/A	V	Associated with returned sample from Mars; may integrate triage functions

Table 2-1. Planned planetary protection implementation for exploration of Mars.

Trace Gas Orbiter

The Trace Gas Orbiter (TGO) mission consists of an orbiter provided by ESA and a payload consisting, in part, of two NASA instruments. TGO's science objectives focus on a search for evidence of methane and other trace gases in the Martian atmosphere. It also includes the Entry, Descent, and Landing Demonstrator Module to demonstrate new technologies for future Mars missions.

As currently envisioned, TGO does not require novel planetary protection technologies. However, during assembly, test, and launch operations (ATLO), it could provide an opportunity for field testing and validation of new planetary protection implementation technology concepts.

Mars Sample Return Campaign

The Mars Sample Return (MSR) campaign consists of three potential missions—the Mars 2018 Rover mission, the MSR Lander mission, and the MSR Orbiter mission—and a fourth potential component, the MSR sample receiving facility (SRF).

Mars 2018 Rover Mission

(Formerly Mars Astrobiology Explorer-Cacher)

Mars 2018 would be a rover capable of caching a sample for return to Earth, serving as the first flight element of the possible MSR campaign.^{23,24,25} The overall objective of the proposed MSR campaign would be to collect Mars samples and prepare them for return to Earth for in-depth analysis in terrestrial laboratories.

The rover was originally conceived for launch with the ESA ExoMars rover carrying the Pasteur payload for astrobiology and geochemistry research. However, in response to budgetary constraints and other NASA and ESA considerations, in May 2011, the planning assumptions for the 2018 rover mission changed. The current mission concept is for a single joint rover to be delivered to the Mars surface by the MSL sky-crane system, supporting both returned sample science (originally to be performed by the NASA-led caching rover) and *in situ* science (derived from previously defined ExoMars priorities), and carrying both the ExoMars Pasteur payload plus additional payload to support the sample selection and caching objectives of the mission.

MSR Lander

The MSR Lander would be a single-purpose mission with a rover dedicated to retrieving the cache of rock cores collected by the Mars 2018 rover, collecting samples from the local regolith and atmosphere, and launching the sample package into Mars orbit.

MSR Orbiter

The MSR Orbiter mission would have as its primary role to capture the orbiting sample and deliver it to Earth. If this orbiter were launched in the opportunity before the MSR Lander, which is one of the MSR campaign scenarios, it would also be in orbit at Mars to observe and support activities of the MSR Lander.

MSR Sample Receiving Facility and Curation

Planetary protection for a possible sample return from Mars includes requirements for highly reliable sample containment throughout all mission phases, including Earth entry and landing, transport of the returned hardware and samples to an SRF, and operations carried out in the SRF. The SRF would have to provide adequate containment for the Mars-exposed flight hardware and samples returned from Mars until they could be tested for possible biological hazards. These

requirements would pose significant engineering challenges, many requiring new technology, to meet the requirements of sample transportation, all aspects of the biohazard assessment, and some time-critical scientific assessment of the samples under both stringent cleanliness conditions and highly reliable bio-containment.

Long-term curation of Mars samples brought to Earth as part of a future MSR mission would require one or more dedicated laboratories and associated staff. Curation of returned Mars samples would likely call for technology development to meet possible new requirements involving storage conditions and extreme organic and inorganic cleanliness to preserve the scientific value of the samples.

Each element of the proposed MSR campaign—the two landed missions, the orbiter returning to Earth, and the receiving facility—requires special attention to planetary protection and contamination control. However, the three-launch architecture provides a platform to proceed with sample collection while the technologies required to handle the returned samples are still reaching maturity.

2.2 Other Solar System Exploration

Solar system exploration plans over the next two decades include potential missions to Europa, Uranus, Enceladus, and Titan. The planetary protection requirement for the icy bodies is identical—do not contaminate liquid water where terrestrial organisms might be able to survive and thrive. Implementation of the requirements for bodies with very thick shells is likely to be less challenging. The Juno mission to Jupiter, launched in August 2011, will not be discussed because it is unaffected by technologies and plans discussed here. Several mission concepts have been studied over the past few years but two have been studied extensively and provide scenarios whereby planetary protection and contamination control have to be considered as important to design of the spacecraft and mission.

Table 2-2 lists the envisioned set of solar system exploration missions (outside of Mars) and planned implementation. The distinction between V_r , Restricted Earth Return, and V_u , Unrestricted Earth Return, should be noted, due to the significant difference in implementation.

Mission	Study Phase	Category	Planned Implementation
Europa JUICE	Pre-Phase A TBD		 Forward contamination would exploit ambient radiation of Jovian system to provide further bio-burden reduction with assumed system-level sterilization. Europa orbiter or lander planned contact with Europa; must demonstrate sufficiently low probability of contamination of subsurface ocean. JUICE or Europa multiple fly-by planned impact with Ganymede; must also demonstrate sufficiently low probability of impact on Europa and/or contamination of subsurface Europa or Ganymede ocean.
Titan/Enceladus	Study Phase, CML 4	11/111	Organic contamination control critical concern; Enceladus may motivate other requirements.
Comet Sample Return	Study Phase, CML 3 / 4	II/V	Must meet requirements for returned sample.
OSIRIS-REx	Phase B	II/V unrestricted	Organic contamination control is a high priority; Must meet requirements for returned sample.

 Table 2-2. Planned planetary protection implementation for solar system exploration.

Europa and Ganymede Missions²⁶

Both NASA and ESA are contemplating missions to the satellites of Jupiter: NASA to Europa and ESA to Ganymede. These missions would carry instruments to characterize the satellites as well as their subsurface oceans.²⁷ Currently, NASA is considering Europa options that include a Europa orbiter, a multiple fly-by spacecraft that would characterize Europa from Jupiter orbit, and/or a Europa lander. The ESA concept, called the JUpiter ICy moon Explorer (JUICE), would characterize the Jupiter system from Jupiter orbit and then would characterize Ganymede from Ganymede orbit.

Because Europa is considered much more likely to harbor life than Ganymede, the key planetary protection requirements on the ESA JUICE mission would be to demonstrate that the probability of impact on Europa meets COSPAR requirements, and that the chance for contamination of Ganymede's putative ocean are negligible. Missions to the Jupiter system would likely be able to exploit the ambient radiation to further reduce bio-burden after launch.

On the other hand, it may be necessary for a Europa mission to undergo terminal sterilization to protect the sub-surface ocean on Europa. This would require the spacecraft to be assembled and then heated in appropriate facilities. In addition, this process would require a careful inventory of the biological and organic materials present, as well as the careful use of witness plates, material archiving, and other processes used successfully by the Stardust sample return mission.

The planned approach to planetary protection compliance can be summarized as follows:

- · Pre-launch sterilization to control bio-burden for those areas not sterilized in flight
- In-flight sterilization via radiation prior to entering the close vicinity of Europa.

Titan/Enceladus Mission²⁸

The envisioned Titan/Enceladus mission would conduct remote observations of both satellites, as well as *in situ* analysis of the Titan atmosphere and surface. A flight element landing on Titan would have to meet strong contamination control requirements in order to avoid mixing contaminants with the heavy organic load present in acquired samples. The presence of waterice, sodium-organics emanating from Enceladus may motivate additional requirements for planetary protection and contamination control, depending on the mission architecture.

Comet Surface Science Return Mission²⁹

The Comet Surface Science Return (CSSR) mission would return a sample from the surface of a cometary nucleus with techniques preserving complex organic compounds for study of the contribution of comets to the Earth's volatile inventory. The requirement of a cryogenic sample return mission is expected to greatly increase the complexity of contamination control since contaminants may cryopump into the sample.

3 Assessment of Technology Progress

Although requirements for planetary protection and organic contamination control flow from different sources, they share the same general approach and many of the same analytical tools, and can benefit from similar education and training programs. The approach to both fields includes:

• Reducing initial (biological and/or organic) contaminant levels and validating the reduction (including assessment of the remainder)

- Preventing the recontamination of a cleaned surface, including modeling to support quantitative risk assessments (this sometimes requires research to support model development)
- Developing long-term curation infrastructure

As the sensitivity of current instruments improves and the sample size for *in situ* organic analysis is reduced, new challenges are faced because contamination levels previously viewed as acceptable now represent sources of concern for interpreting scientific data. Therefore, the contamination control processes must improve in parallel. For example, a study by the MEPAG Organic Contamination Science Steering Group described *in situ* astrobiology experiments (emphasis on MSL) looking for organic signatures at levels of 1–10 parts-per-billion (ppb), requiring reduced carbon contaminants to fall below 40 ng/g of sample.³⁰

This section summarizes the activities and needs in both planetary protection and organic contamination control. Summary charts are provided for research in planetary protection technologies, ranked on a simple 1–3 scale for convenience. In addition, this section shows the progress in these fields since the 2005 assessment.

3.1 Contaminant Reduction and Assessment

Techniques for reduction and assessment are central to meeting requirements in both planetary protection and organic contamination control. Many planetary protection technologies, particularly in microbial reduction and validation, have matured significantly in the five years since the last assessment, lacking only formal NASA approval before they can be fully utilized by mission design teams. However, a project can request permission to use a "non-approved" process by providing adequate documentation regarding efficacy.

Similarly, effective organic contamination control calls for materials compatibility studies early in the instrument and spacecraft design, so that detrimental outgassing can be minimized and spacecraft can be designed to reduce the possibility of transporting contaminants from one region of the spacecraft to an instrument analysis suite. It should be noted that miniaturization of future mission detectors and sample sizes can increase their sensitivity to contamination. For terrestrial laboratory experiments, this can be effectively addressed through cleaning and minimizing sample and equipment exposure times post-cleaning. However, maintaining ultralow levels of contaminants over the extensive period of time required to build and launch spacecraft can be difficult and expensive. Another important factor to consider in the initial design and planning stages is the achievability of target contamination levels to ensure the level of permitted contaminants are compatible with existing commercial and government analytical facilities and processes. Further, early mission planning must also consider transport mechanisms and sources that are unique to the environment of the foreign body.

Missions with astrobiological instruments requiring high levels of both bio-burden reduction and organic contamination control to meet planetary protection and science requirements must be designed to be mutually compliant; for instance, because dry heat microbial reduction (DHMR) sterilizes but does not remove biological material, it may be inappropriate or insufficient for certain instrument designs. In addition, care must be taken not to recontaminate the instruments on the spacecraft after a contamination-reduction activity.

A problem that continues to plague planners of sample return missions is the reality that all forms of organic contamination cannot be analyzed during contamination control assessments, and so a critical cross-section of families of organic compounds must be selected for analysis.

The obvious problem is that intelligent selection of the organics to be analyzed requires some knowledge of the compounds to be expected on the target planetary body, whereas no such knowledge may exist. When the analysis of organics in returned samples reveals unexpected compounds, there is the very real hazard that this class of organics may not have been assayed on the spacecraft before launch, or guarded against during all phases of the mission. Lessons learned from LDEF, Stardust, Genesis, and Hayabusa have illuminated shortcomings in mission contamination-control process, procedure and project systems engineering that must be addressed early in the mission planning process for future missions involving sample return.³¹ In this regard, it is critical to have a living document that details planetary protection-related lessons learned in previous missions. Such a dedicated living document does not exist. For OSIRIS-Rex, which is in Phase B, the contamination control documents are already in development. It is critical, however, to create a contamination control primer for all missions including sample curation, especially sample return missions. This would aid the Hayabusa II and Marco Polo missions now in development.

3.1.1 Microbial Reduction Methodologies

Microbial reduction can be implemented prior to launch or, under selected conditions, after launch if the ambient environment is detrimental to microbial survival and replication. This assessment describes research and development relevant to both approaches.

Pre-launch Bio-burden Reduction

At the time of this document's publication, the only pre-launch sterilization technique approved by NASA is DHMR; NASA policy specifies parameters for various D-values, defined as the time required at a given temperature to reduce the microbial population by a factor of 10 (i.e., 90% of the population is destroyed), and ranging from approximately 30 minutes exposure for exposed surfaces to 5 hours for encapsulated bio-burden. While the nominal temperature of exposure is 125°C, NASA procedures provide for extensions to temperatures as low as 104°C.

This protocol was defined in concert with Viking, the first mission to face the most stringent planetary protection requirements; its implementation remains the gold standard today. Because the probability of an incomplete sterilization had to be less than 1.4×10^{-5} , the Viking landers were enclosed in a bio-shield and exposed to approximately 112°C for roughly 30 hours.^{32,33} This posed two challenges:³⁴ 1) the radioisotope thermoelectric generators (RTGs) required cooling during the heating cycle; and 2) the interior of the lander, insulated to protect instruments from the extremes of Mars diurnal cycling, heated very slowly. Engineers thus devised a scheme using a formaldehyde-isopropanol mixture as an RTG coolant, distributing it to provide an interior heat source.

Since Viking, the materials and processes used to fabricate spacecraft have advanced steadily. In addition, there are often reasons to use commercial-off-the-shelf products or hardware designs from prior (but recent) missions. These trends often create technical obstacles for planetary protection when the hardware as designed cannot survive exposure to DHMR temperatures. Plastic packaging, nanometer-scale features only a few atoms thick, and conductive epoxy attachment methods, are a few of the design features of modern spacecraft electronics that, at best, make DHMR a risk to long-term reliability, or at worst, impossible without risking immediate damage. In addition, many instrument sensors cannot be exposed to elevated temperatures without risk of permanent damage. Some instrument and spacecraft structures have critical alignment requirements that limit maximum allowable temperatures as

well. The availability of alternate microbial reduction methods and approaches is necessary to accomplish science goals for certain strategic missions (especially those involving the search for life) and to do so within programmatic schedule and cost constraints.

Today, alternative techniques (namely, specification for surface sterilization using vapor hydrogen peroxide and an expanded DHMR specification) are close to approval by NASA; although none has yet been included in the policy documentation. Precision cleaning is not included because it is a familiar practice, varying from institution to institution. ESA has also adopted many of these techniques in its documented procedures.^{35,36}

Extension of DHMR Parameters

Because many flight hardware manufacturing processes (e.g., curing processes or qualification testing) expose components to elevated temperatures as high as 300°C, it is desirable to be able to take sterilization credit for such processes. This can reduce planetary protection-specific processing to surface sterilization at assembly only, or obviate the need for processing at all. In addition, implementation may be simpler if process humidity is not a parameter that needs to be controlled. These considerations, in addition to others, prompted the Mars Exploration Program to conduct in-depth assessments of the lethality of heat exposures outside the allowed range of parameters in the current DHMR specification. The NASA Planetary Protection Officer, in partnership with her ESA counterpart, then undertook an experimental validation effort, followed by an intensive evaluation of all experimental data, to enable development of new and expanded specifications for use of heat processing as a bio-burden reduction method.

<u>Status</u>: NASA has undertaken a suite of experiments to determine the efficacy of DHMR beyond the currently approved temperature and humidity specifications.^{37,38,39} ESA has worked with NASA to validate the experimental results as an important step toward formal acceptance and approval. The PPO is nearing completion of rigorous statistical analysis of the NASA and ESA data and creation of new formal specifications for the reduction of bio-burden for achievement of planetary protection compliance by flight projects.

Radiation Sterilization

Gamma radiation and electron beams are used extensively in the medical industry for sterilization and thus are logical candidates for planetary protection implementation. Gamma radiation is a low-temperature penetrating technology and was therefore selected for the Beagle 2 implementation.³⁴ Pillai et al.⁴⁰ suggested that the 10 MeV electron beams used in the food industry could conceivably be used in efficient implementation; electrons of these energies would penetrate fairly deeply and low doses could likely be used because of the sparse density (10⁶ cells/m²) relative to those in typical medical sterilization. The authors' preliminary work suggested an effective 6-log reduction (i.e., reduction by 99.9999%) of *Bacillus* spores on aluminum coupons.

Urgiles and collaborators described a related technology⁴¹ using 100 keV electron beams for surface applications. Here, the shorter penetration depth of several microns as the radiation is well matched to bacterial spore size. A second benefit would be the capability of using a portable 100 keV electron source.

<u>Status</u>: This technology has not been brought to maturity. Its compatibility with various types of materials and geometries of hardware needs to be determined.

Vapor Hydrogen Peroxide

While hydrogen peroxide has been recognized as a sterilant for well over a century, the use of vapor-phase hydrogen peroxide dates to approximately twenty years ago.⁴² Although its demonstrated efficacy made it logical to consider this technology for planetary protection compliance, it took many years to validate it for this purpose and determine the effects of ambient temperature, humidity, and material substrate on the process. As a strong oxidizer, high levels of hydrogen peroxide can affect finishes, lubricants, or other materials with chemistries susceptible to free radical attack such as aromatic rings and sulfur bonds; the technique's compatibility with complex systems needs to be demonstrated

<u>Status</u>: This technology has been reported as effective^{43,44} when controlling for conditions such as peroxide concentration and exposure duration. As of early 2011, ESA and NASA are finalizing specifications for use of this technique.

Ethylene Oxide

Ethylene oxide is widely used in the medical industry for sterilization.⁴⁵ It has the advantage that, while it is highly effective against organisms, it has a different spectrum of material/process compatibility compared to hydrogen peroxide. Developments within the medical device industry have addressed many of the limitations (temperature, humidity, vacuum requirements) associated with earlier EtO process approaches.

<u>Status</u>: This technology is under consideration by GSFC for several extraterrestrial missions, including the planned ExoMars mission in collaboration with ESA.

Post-Launch Sterilization through Environmental Effects

Bio-burden reduction can also be achieved through exposure to a sterilizing environment. Missions to both Mars and Europa provide opportunities to use this passive exposure to obtain additional bio-burden reduction. However, passive ultraviolet (UV) sterilization at Mars has already been considered in the development of the "at launch" numerical bio-burden requirements for Mars; thus, it cannot be used by a mission to argue (pre-launch) for a further reduction of bio-burden post-launch.

Passive Sterilization through Ultraviolet Radiation at Mars

In contrast to Earth, the thin atmosphere at Mars allows most solar UV radiation to be transmitted to the surface, making this an attractive sterilization alternative for missions to Mars. Several years ago, Schuerger and collaborators⁴⁶ reported that in Mars ambient conditions, most bacterial populations will be sterilized in a few minutes. More detailed studies, however, have shown more complex results. Newcombe et al. elaborated on sterilizing UV radiation, demonstrating highest effectiveness at 254 nm⁴⁷; however, they also identified a *Bacillus* strain found in spacecraft assembly facilities that is particularly resistant to sterilization by exposure to UV radiation. The effect of solar UV radiation is harmful to microbes on un-shadowed spacecraft surfaces in interplanetary space. On the surface of Mars, this effect is complicated by possible shadowing by dust particles,⁴⁸ the regolith, or spacecraft hardware.

<u>Status</u>: These studies suggest a need for further research into the limitations and constraints of these effects in ambient Mars conditions; this will be discussed further in the section on fundamental microbiology.



Figure 3-1. Radiation fluxes at Europa. *Left:* Particle flux for electrons, protons, oxygen, and sulfur ions [49]; *right:* radiation spectra at geosynchronous (GEO) and Europa orbit [50]. At Europa orbit, the Jupiter radiation environment contains significantly higher fluxes of both high-energy protons and electrons than Earth's orbit.

Passive Sterilization through Particle Radiation at Europa

Use of radiation as a contributing factor to meet the probabilistic requirement for contamination of all bodies other than Mars is accepted practice. However, this will require re-evaluation for future missions, since organisms exist that can resist this radiation dose. Passive environmental sterilization is anticipated to be particularly useful for missions to Europa because of the strong Jovian radiation environment. Figure 3-1 characterizes the environment at Europa in terms of high fluxes of electrons, protons, and sulfur and oxygen ions⁴⁹ generated by volcanic plumes at Io and swept to Europa. The electrons and protons present particular challenges to spacecraft because they are significantly more energetic than those encountered by spacecraft in low Earth orbit,⁵⁰ as shown in Figure 3-1, but the high energy electrons in particular have the potential for bio-burden reduction utilizing this effect.

<u>Status</u>: While current implementation plans for missions to Europa provide credit for hardware exposed to 7 MRad levels,^{51,52} further measurements of the microbial reduction in high-radiation environments may suggest that lower exposure levels are sufficiently lethal, thus affecting planetary protection implementation as well as mission architecture.

Summary of Microbial Reduction Technologies

Since the 2005 assessment, extensions to DHMR parameters have been validated and are used extensively for MSL. In addition, hydrogen peroxide studies have been concluded and the technique is ready for adoption upon approval. Environmental radiation also can be used during a mission and this is being seriously considered for a potential Europa mission using highly conservative assumptions. In fact, a mission to Europa could use all of these techniques.

Hardware compatibility studies are expected to continue for each project, as it is expected that certain natural incompatibilities (such as surface finishes exposed to hydrogen peroxide) will not be overcome. These studies will continue at the project level as parts lists are revised and should be included in allocating both cost and time. In addition, the use of passive environmental radiation, particularly in the Jovian environment, is currently being investigated; although this changes as alternative trajectories are identified.

Figure 3-2 summarizes the status of all the microbial reduction methodologies.



Figure 3-2. Summary of the status of microbial reduction technologies, describing sterilization modalities. There are two topics for each modality represented: 1) its progress toward NASA approval as a sterilization technique (first line) and 2) compilation of a hardware compatibility chart (second line).

3.1.2 Bio-burden Detection and Assessment

Planetary protection includes analytical methods to assess surfaces for bio-burden and the effectiveness of microbial reduction techniques. The NASA standard assay uses classical microbiological methods to detect the presence of aerobic spore-forming organisms. In addition, novel rapid detection techniques have been approved for some bio-burden assessment applications and others are in the process of being approved for use.

Cultivation-Based Methods

Rapid Spore Assay

Commercial rapid assay systems can be modified with different reagents to be compatible with NASA's standard sampling method. This allows for the detection of spores, not just viable organisms, and gives results in just 5 hours (compared to the 72 hours of the NASA standard method).⁵³

<u>Status</u>: This method has been demonstrated to be equivalent to the NASA standard assay, but faster.

Dipicolinic Acid (DPA) Detection

One novel technique previously under study for planetary protection applications is a microscopy-based assay that uses terbium-dipicolinate triggered by UV excitation as DPA molecules are released during germination.⁵⁴

<u>Status</u>: Development was partially funded by the NASA PPO. The technique was demonstrated to be effective, but did not significantly enhance the state of the art compared to the NASA standard assay and the rapid spore assay, and was not adopted for planetary protection implementation. Documentation for this technique is convincing; therefore, there is no reason a mission could not request approval to use the DPA assay; however, a rigorous comparison with the standard assay would be needed.

Molecular Methods

Two molecular methods have been approved for assessment of microbial contamination on spacecraft surfaces.⁵⁵

Adenosine Triphosphate Detection

Adenosine triphosphate (ATP) is a key molecule in cellular metabolism and can be measured through spectrophotometric detection of its bioluminescence. It is somewhat superior to older colony-counting protocols because of its ability to report the presence of non-cultivable organisms,⁵⁶ but can be confounded by the presence of ATP from biological material irrelevant to planetary protection compliance.

<u>Status</u>: This method has been validated and can be used for general indication of spacecraft hardware biocontaminant cleanliness levels and real-time process control.

Limulus Amoebocyte Lysate Assays

Lipopolysaccharides (LPS), found in the cell walls of Gram-negative bacteria, can be measured with a limulus amoebocyte lysate (LAL) assay. LPS is present only in Gram-negative bacteria, making it difficult to relate to the standard assay that analyzes spores from microbes that are primarily Gram-positive.

<u>Status</u>: This method has been validated and can be used for general indication of spacecraft bio-contaminant hardware cleanliness levels and real-time process control.

Quantitative Polymerase Chain Reaction

This method does not measure products of microbial metabolism, but rather quantifies the genetic material present. Nellen⁵⁷ discusses the useful role of the quantitative polymerase chain reaction (Q-PCR) in characterizing environmental systems with populations of uncultivable organisms. However, Rawsthorne et al. report that it is difficult to use this method to quantify bio-burden because of the challenge of releasing nucleic acid from spores; furthermore, the presence of DNA does not necessarily imply the presence of viable spores.⁵⁸ Nevertheless, Q-PCR has also aided bio-diversity research, illuminating the microbial ecology of the spacecraft assembly facility by identifying uncultivable organisms.

<u>Status</u>: This method is not used for planetary protection compliance (i.e., to quantify bioburden), but is used in research in microbial diversity. Current research at the Jet Propulsion Laboratory (JPL) includes the development of a PCR-based approach with a special dye to differentiate between viable and nonviable spores.⁵⁶

Summary of Bio-burden Detection and Assessment Technologies

Figure 3-3 summarizes the work in bio-burden assessment since 2005.



Figure 3-3. Summary of bio-burden detection and assessment technologies.

3.1.3 Bio-diversity Studies

Bio-diversity encompasses a wide range of research topics, including the microbial ecology of anthropogenic extreme environments (namely, spacecraft assembly facilities), natural extreme environments, and transport mechanisms between them. This line of research is concerned with characterizing the microbial populations in these environments, determining the effects of environmental change on these populations, and evaluating the transport mechanisms of both microorganisms and biological material.

Understanding bio-diversity and the transport of organic material is critical for organic contamination control studies concerned with the fate and presence of organic materials originating from biological sources. Today, bio-diversity studies use the full range of improved bio-burden detection and assessment technologies, ranging from improved cultures to the molecular methods used for organisms that are not easily cultivated.

Bio-diversity in Spacecraft Assembly Facilities

A number of studies have assessed the genomic inventories of spacecraft assembly clean rooms, treating them as extreme environments.^{59,60,61,62,63} Phylogenetic analyses and cultivation experiments have evaluated the populations and diversity in these environments. One analysis by Crawford⁶⁴ called for studies in the genetics and structural biology of organisms in clean rooms because the ones isolated tend to be unusually resistant to treatments such as desiccation, peroxide, UV light, and gamma radiation. Furthermore, because the communities found in spacecraft assembly (and thus most likely to "hitch-hike") typically reflect the environment surrounding the assembly facility, extensive bio-diversity studies near spacecraft facilities are still critical, although these studies have not yet motivated a change in how we approach microbial reduction.

<u>Status</u>: Studies to date have identified both Gram-positive and Gram-negative organisms, including thermophiles, obligate anaerobes, alkaophiles, and non-spore-forming microbes, tolerant to environmental stress. While this field is fairly advanced and good techniques exist to provide further detail, a good path to data interpretation is still needed.

Genetic Inventory (Sampling in a Low Bio-mass Environment)

Bio-burden assessment and bio-diversity studies for planetary protection have typically emphasized live, cultivable microorganisms with an emphasis on spore-forming microbes. In response to a call for NASA to develop a genetic inventory capability, the Mars Program Office initiated a research program to determine the potential "passenger list," developing the capability to sample in a low-biomass environment. Resource limitations required the task to be limited to a qualitative inventory without discrimination between live and dead organisms.

<u>Status</u>: The Mars Program Office sponsored a five-year task in 2007 to develop the capability for collecting DNA samples from low bio-mass environments. It is scheduled for completion during 2012, with possible follow-on work focused on specific planetary protection requirements for future planetary missions.

Microbial Diversity in Extreme Environments

Fundamental research in microbial diversity continues to dramatically expand our understanding of environments that are potentially habitable.^{65,66} Some of this research motivates new solutions to microbial contamination by elucidating resistance mechanisms.⁶⁷ In addition, this field of research adds to the body of work describing potentially "special regions" on Mars and on other bodies.

Status: This field will continue to inform research in extraterrestrial environments.

Summary of Research in Microbial Diversity

Research in these fields is ongoing, as shown in Figure 3-4. The genetic inventory task has achieved significant progress since 2005.



Figure 3-4. Summary of research in bio-diversity studies.

3.1.4 Organic Contamination Control and Assessment

Although organic contamination control is a natural consideration for spacecraft design and assembly,⁶⁸ standards have been concerned with the higher contamination levels affecting factors such as optical performance rather than the levels acceptable for chemical analysis and other instrumentation needed for life detection missions envisioned here. The principal contamination source in typical spacecraft operation is material outgassing, leading to release of volatile organic compounds. All materials and processes must be controlled and monitored to minimize contamination; typically, this is funded by the project and takes place as the project progresses with little technology or research done outside of the project. For sample return missions, however, contamination control will have to be considered early in the process and particular notice must be given to the requirements of archiving, curation, and sample distribution. For IVb life detection missions, planetary protection is also concerned with non-viable organic contamination that has the potential to interfere with life detection experiments. Consequently, it is necessary to anticipate that there would be planetary protection requirements for control of non-living organic contamination and compliance verification by the Planetary Protection Officer.

NASA has developed significant capabilities and procedures to understand and limit organic contamination. The MSL team has undertaken extensive work to understand and limit organic contamination for *in situ* analysis, and the successful Stardust sample return mission published its procedures and recommendations.⁶⁹ The Stardust analysis was particularly challenging because the aerogel matrix could, in principle, have confused the scientific results. However, even though the contamination level was higher than one would wish, careful use of witness plates, excellent curation, and continuous monitoring of contamination throughout the assembly and operation processes contributed to the return of quality science. This was further enabled by the inclusion of curation experts and systems engineers at all phases including planning and implementation. The Sample Analysis at Mars (SAM) instrument, launched on MSL, has similarly included contamination control at all phases of the assembly process. Furthermore, the MSL contamination control and planetary protection teams have attempted to integrate planetary protection compliance and contamination control processes where it was possible to satisfy the respective planetary protection and contamination control requirements simultaneously.

Organic contamination control is typically assessed with instruments used for other standard analyses. The Mars Organic Contamination Science Steering Group report²⁵ summarizes the techniques relevant to analyzing spacecraft contamination during ATLO:

- *Chromatographic mass spectrometry methods (GC-MS):* Separates and determines the mass of volatile organic compounds.
- *Ion chromatography (IC):* Separates by molecular charge; appropriate for compounds such as proteins, carbohydrates and other molecules are charged in solution.
- *Fourier transform infrared spectroscopy (FTIR):* Analyzes functional groups (but has limited sensitivity).
- Time-of-flight secondary ion mass spectrometry (TOF-SIMS): Performs elemental analysis with spatial distributions.
- Getter Technology: Protects flight hardware against recontamination.

Further, the use of quartz crystal monitors and residual gas analyzers are suitable tools for quantifying total outgassing in vacuum and molecular mass of outgassed constituents. Additionally, the chromatographic mass spectrometry methods can be expanded to include liquid chromatography–mass spectrometry (LC-MS) and supercritical fluid chromatography–mass

spectrometry (SFC-MS), which can then separate and determine the mass of most organic compounds. It should be noted that ion chromatography is also appropriate for inorganic ions.

Like planetary protection, organic contamination control calls for extensive material compatibility testing, as well as making sensible implementation decisions such as the use of synthetic swab materials instead of cotton. An additional area of overlap is the need for methods to remove trace amounts of biological and organic materials with methods such as supercritical CO₂ cleaning⁷⁰; in the case of Mars, this provides the interesting opportunity of using the atmosphere for cleaning. Similar methodologies should be actively pursued as they can be leveraged for both contamination concerns. While a particular cleaning method may be optimized for the removal of specific types of contaminants, it is important to be mindful that, cleaning methodologies must be applicable to a wide range of hardware geometries and materials types. Thus, cleaning development and hardware designs (materials of construction and assembly sequence) must be considered simultaneously to yield an implementable outcome.

The following are two critical differences between organic contamination control and planetary protection:

- 1. The source of volatile organic contamination is primarily man-made polymers used in the construction of a spacecraft. Non-volatile organics may be the result of volatile organics that have formed a film on a surface via thermal accommodation or other interaction with the surface or they may have their origin in biological sources (skin cells, bacteria, viruses, etc.). Non-volatile organics that contaminate sample path surfaces or the sample itself can seriously contaminate the sample and can degrade the science or even end an experiment. Material outgassing in low-pressure environments does not violate planetary protection standards, but may compromise returned science.
- 2. Planetary protection methodologies, particularly DHMR, may not remove organic material, and thus the choice of a microbial reduction technique impacts the organic contamination threat. Recent research has demonstrated that biological molecules may persist on spacecraft surfaces^{71,72}; therefore, sterilization techniques may be insufficient to meet contamination control requirements and additional methods may be needed.

Key to effective project implementation is recognizing that both planetary protection and contamination control need to be prominently addressed during the pre-project phase, so that instrument and spacecraft providers can be proactive during the design of the mission and the hardware, and maintain the appropriate level of vigilance.

3.1.5 Assured Containment Samples Returned from Mars

MSR in the 2020s would require assured containment of return samples (commonly referred to as backward or back-planetary protection) and would be a significant focus area for Mars technology development. MSR would be classified as Category V, restricted Earth return, and would thus have both stringent forward- and back-planetary protection requirements. Unlike forward-planetary protection, which is intended to protect exploration targets (and sometimes the scientific samples) from Earth-originating contamination, back-planetary protection is intended to ensure a very low probability of inadvertent release of Mars material in order to provide extra protection against the extremely unlikely possibility of biological hazards in the returned sample. Such a mission would require the highest degree of containment of the samples and all returned hardware that had been in direct contact with Mars until completion of a review (consistent with the reports and review requirements described in NASA procedural requirements for planetary

protection [NPR 8020.12D]) to ascertain that all planetary protection requirements, including the execution of prescribed life detection and biohazard protocols have been met.

Containment assurance requires "breaking-the-chain" of contact with Mars: the exterior of the sample container must not be contaminated with Mars material. Also, the sample container and its seals must survive Earth impact, the Earth return vehicle must provide accurate delivery to the Earth entry corridor, and the Earth entry vehicle (EEV) must withstand the thermal and structural rigors of Earth atmosphere entry (all with an unprecedented degree of confidence).

Mission concepts with various break-the-chain methodologies are under development; some concepts break the chain of contact once the samples reach the MSR Orbiter, while others start to break the chain of contact before the cached samples leave the surface. In either case, technologies to verifiably meet requirements for back-planetary protection will be needed. Robust sealing technologies as well as technologies to protect the sealed sample container during Earth return, entry, and landing will also be needed.

<u>Status</u>: A comprehensive program for developing back-planetary protection technology was initiated in fiscal year (FY) 2005 in conjunction with a pre-project activity, only to be discontinued part way through the year when the work on Mars Sample Return was postponed as part of a realignment of the Mars Program. The program was guided by a detailed probabilistic risk assessment (PRA) of threats to sample containment and included integrated container sealing/break-the-chain techniques and corresponding verification techniques, robust EEV concepts, and methods for protecting the EEV heat shield from micrometeoroids (identified as a key risk by the PRA). A portion of the EEV work has recently been re-started under the In-Space Propulsion Program.

3.2 Recontamination Prevention

Once a surface has been cleaned of spores or chemical contaminants, maintaining its state of cleanliness is critical. This section describes technologies and facilities required for the upcoming missions described here.

3.2.1 Aseptic or Ultra-Clean Assembly

In this context, aseptic or ultra-clean assembly refers to a dedicated enclosure free of biological contamination. This definition may be more stringent than the conventional meaning of a sterile unit, but one in which biological material may be present. This assembly may take place in small units, such as glove boxes, or large facilities capable of handling spacecraft; in addition, it may refer to specialized techniques to integrate cleaned subsystems in environments with restrictions on both particulate levels and the presence of biological material. ESA previously implemented aseptic assembly for the Beagle 2 life detection mission.³⁴ Therefore, this is a Mars program-level issue to be resolved prior to project start.

<u>Status</u>: Use of aseptic assembly facilities is determined at the project level, but must be determined far (many years) in advance in order to build and certify the facility.

3.2.2 Modeling Contaminant Transport

Sources of contamination include transport from another part of the spacecraft or other post-assessment deposition.

1. *Materials outgassing transport in planetary environments*. This field is critical for *in situ* studies on the surfaces of Mars, Titan, and the environment on Europa. Recent work has included a contamination transport model for the SAM payload for MSL,⁷³ designed to

conduct organic analysis of Mars soil samples. Because the instruments have sub-ppb detection capability, total organic carbon contamination may not exceed sub ng/g for smaller sample sizes. To estimate and mitigate the effect of *in situ* self-contamination, a geometric model of the spacecraft was developed for use with simulations estimating the likelihood of outgassed materials escaping a vent and entering the sample chamber under Mars ambient wind conditions. The simulation results were then used as a basis for modifications to the spacecraft design, and derivation of allowable outgassing levels for spacecraft components, so that the requirements could be met.

- 2. *Materials outgassing transport in a vacuum environment.* This field is well-developed and the techniques have been used on numerous planetary and non-planetary spacecraft and observatories. It is used to ensure the proper functioning of spacecraft hardware as well as scientific instruments with sensitivities to sub-monolayer depositions. System-level studies to preclude contamination during system-level testing prior to launch and during post-launch and cruise phases for planetary missions have been developed. Recent work has included a contamination transport model for the Juno Flight System to estimate molecular deposition from outgassing materials onto contamination-sensitive surfaces (instrument optics, radiators, detectors, etc.) during post-launch, cruise, and science phases. The modeling results were used as a basis for modifications to the flight system design (i.e., bake-out of solar arrays, vault vent moved, and propulsion bay vent customized) to meet the end-of-life (EOL) contamination requirements. Evaluation of outgassing via *in situ* measurements such as on Rosetta⁷⁴ is crucial in both validating these models and understanding the photochemical breakdown of contaminants as well as their expected effect on the scientific products.
- 3. *Particle redistribution*. This field is critical for system-level studies to preclude particulate contamination of contamination-sensitive surfaces during system-level testing prior to launch (acoustics and vibration), launch (launch vehicle acoustic and vibration environment), and on-orbit, atmospheric entry, or post-landing acoustic and vibration environments (deployments, aero-braking, surface landing, etc.).
- 4. *Transport to subsurface regions.* Contamination transport may, in principle, refer to transport from the orbit to the surface, or from the surface to the subsurface of an extraterrestrial body. In this context, we are concerned principally with modeling transport on the extraterrestrial body itself. On the surface of Mars, transport to subsurface regions is estimated through models of localized ice melting, triggered by a spacecraft heat source. On the other hand, the Juno mission to Jupiter calls for demonstrating that the spacecraft would not accidentally land on the surface of Europa such that contaminants could be transported through the ice to the ocean. Relevant analyses suggest that the probability of contaminating the ocean is estimated at less than 9×10^{-6} , an order of magnitude below the maximum of 1×10^{-4} required by COSPAR.^{75,76} These system-level models demand capabilities in systems engineering, microbiology, and planetary surfaces to create an integrated approach, unlike the conventional analysis of point designs.
- 5. *Spore adhesion*. In order to estimate the risk of spores transported from one part of the spacecraft to another, it is critical to understand the mechanism and magnitude of spore adhesion to spacecraft surfaces under various environmental conditions. This work is in its infancy.



Figure 3-5. Summary of technologies for contaminant transport.

Figure 3-5 summarizes the progress in these areas.

3.2.3 Isolation Technologies

Isolation technologies protect cleaned surfaces from recontamination and are thus systemspecific, and may be implemented at the systems level or for a subsystem. Use of isolation technologies in Mars exploration dates back to Viking; it had a bio-shield architecture protecting the spacecraft after sterilization and until deployment on the surface of Mars, as the bio-shield was jettisoned in flight. Figure 3-6 shows an exploded view of the Viking spacecraft in its bioshield enclosure⁷⁷ and Figure 3-7 shows the preparation for system-level sterilization.

After Viking yielded negative life detection results and showed no organic materials, subsequent missions were subject to different implementation requirements. The Phoenix mission to polar latitudes on Mars did not require system-level sterilization, but it did require a bio-barrier to protect the integrity of analysis of samples extracted by the sampling arm. This bio-barrier was successfully deployed on the Martian surface, protecting the arm after sterilization,⁷⁸ as shown in Figure 3-8.

Future missions will require a combination of approaches to isolation and recontamination prevention. A mission to Europa may require system-level sterilization; like Viking, it could be the first spacecraft to land on a body without the sophisticated understanding of previous missions. In addition, although bio-barrier technology is likely to be necessary for a number of missions to Mars and other targets to prevent recontamination, it does not require significant developments in novel technology, as shown in Figure 3-9.



Figure 3-6. Exploded view of the Viking spacecraft illustrating the bio-shield, jettisoned when the descent capsule began entry, descent, and landing. (Courtesy of Flight International)



Figure 3-7. Viking engineers preparing the spacecraft for sterilization.



Figure 3-8. The Phoenix bio-barrier. The robotic arm is shown encased in the bio-barrier, held in place by a series of latches released shortly after landing by a pyro-activated pin puller. Torsion springs at each end then rotated the ribs (right to left in the figure) to open the enclosure [77].



Figure 3-9. Summary of isolation technologies.

3.3 Organizational Needs

Many of the needs in planetary protection and contamination control revolve around organizational changes or initiatives, rather than technology development alone. This section summarizes several central issues.

3.3.1 Mars Sample Return Facility

A sample returned from Mars would require retrieval, analysis, and curation. Any MSR facility would call for an integration of all the technological and organizational improvements discussed in this document, plus others. Previous studies⁷⁹ suggest that such a facility, with requirements consistent with biosafety level 4 (BSL-4) laboratories, could take as long as fifteen years for candidate site evaluation and the potential selection, design, commissioning, and testing of any approved site.

However, none of these planetary protection approaches mitigates the need for strong contamination control procedures to guarantee the scientific integrity of the returned sample. Planetary protection procedures, particularly sample handling and receiving on Earth, are under discussion now⁸⁰ and would need to reach maturity significantly ahead of a launch date.

3.3.2 Curation

Curation of any returned samples is a central element of contamination assessment and planetary protection, with five major aspects that must be considered.

- 1. All spacecraft hardware (i.e., solar panels, spacecraft shielding, and the spacecraft bus materials) that could interact in any significant way with the collected samples, including obvious contaminant sources such as outgassing species, particles, and propellant contaminants. Especially critical are hardware items that have been anodized or allodyned, since these routine hardware preparation techniques introduce a significant range of inorganic contamination.
- 2. All witness surfaces exposed during spacecraft hardware manufacture, integration, testing, and cleaning right up to launch—these surfaces must be archived for analysis during all aspects of the mission (as part of the contamination control verification process) and far into the future for comparison with the returned samples.
- 3. All witness surfaces flown on the mission, to record the contamination suffered by the collected samples from spacecraft flight operations, the space environment, and reentry into Earth's atmosphere.
- 4. Samples of air and earth from the capsule recovery site.
- 5. The returned scientific samples themselves.

Returned sample missions also include the challenges of containers used to transport samples as well as to maintain the integrity of the scientific samples during initial screening, transport, storage, and distribution to the scientific community.

Issues of concern for curation include design of the curation facility and archiving of relevant materials during manufacture and assembly. Ideally, a curation specialist would be part of the spacecraft design team from the start of all sample return missions.

3.3.3 Education and Training and Transfer of Knowledge

Designing spacecraft instruments for cleanliness begins during the early phase of a mission with clear architectural and systems engineering plans. When the 2005 assessment was conducted, most of the planetary protection and contamination control expertise then resided at JPL, with only a limited number of institutions engaged in this field of research. In the last five years, other organizations have been designing instruments destined for targets with stringent planetary protection and/or contamination control requirements, and even more institutions are proposing or implementing instruments for such missions.

These procedures require an investment in education and training of principal investigators (PIs), systems engineers, mission design engineers, and spacecraft assembly engineers. The NASA PPO offers an excellent class in planetary protection but earlier education is also required. Knowledge transfer between institutions is also important to inform pre-project personnel and instrument providers, so as not to duplicate research and development.

PIs conducting organic investigations will likely provide for their own contamination control programs. However, PIs may not have a sophisticated understanding of planetary protection requirements. Therefore, any requests for instrument proposals, such as those found in Research Opportunities for Space and Earth Sciences (ROSES), must call for development of instruments compatible with planetary protection implementation if the instrument or technology is a candidate for one of the missions described here or for future life detection missions. Similarly, these proposers must consider the methods used during contamination control early in their instrument development, so that instrument designs are compatible with maintaining sample integrity.

4 Key Findings and Recommendations

Planetary protection and organic contamination control are related disciplines that ensure the integrity of returned scientific data and the protection of all planetary bodies. The importance of these disciplines will only increase as our missions converge on determining the differences between "prebiotic," "habitable" and "inhabited."

The following recommendations, stemming from the current assessment, include suggested improvements over the already impressive progress in planetary protection and contamination control technologies since 2005.

4.1 Systems Engineering

<u>Finding</u>: Systems engineering education, tools, and capabilities typically do not extend to contamination control and planetary protection. The 2005 report recommended the development of integrated tool sets for planetary protection and contamination control but there has been little progress in the intervening years. Although we have the capability to carry out trade studies for point designs, we cannot yet rapidly estimate the cost, risk, and schedule impact of different architectures. Early integration of system engineering approaches to mitigate planetary

protection and contamination control concerns is vital as we progress to ever more complex missions where these could become costly add-ons if not considered at the inception of a mission project.

<u>Recommendation</u>: The elements of contamination control and planetary protection that are critical to mission planning, science, and hardware design must be a fundamental part of the systems engineering and must be addressed at the earliest stages of the mission to ensure proper flow-down of requirements and cost-effective mission planning. An adequate approved materials/parts list that can accommodate both contamination control and planetary protection considerations should be developed. Integrated modeling tools should be developed to aid systems engineers and designers for future work, particularly in the form of risk assessments for forward- and back-contamination. Also, planetary protection implementers should define and engage systems engineering approaches to determine traceable requirements that can be flowed down within projects early in the process. Cost estimating tools should be developed.

4.2 Technology Development

Finding A: In the last five years, there has been impressive progress in certain areas of forwardplanetary protection technology. There have been advances in sterilization methods most notably in the use of vapor hydrogen peroxide as a surface sterilant. Commercial rapid assay methods now allow for the detection of spores, not just viable organisms, and give results in 5 hours compared to 72 hours of the NASA standard method. NASA and ESA have just completed work on those methods. This time saving not only translates into cost savings for planetary protection but in the overall spacecraft assembly process. Finally, extensive research was recently concluded at both JPL and ESA aiming at the revision and extension of the current specifications for dry heat sterilization and providing alternatives to the requirement for complete sterilization (currently 500°C for 0.5 seconds).

However, the process for validating and approving new technologies for NASA applications is cumbersome, due to the multiple research organizations involved in the development of the new data. As a result, since the last report, no new technologies have been actually approved for use by NASA. The NASA and ESA PPOs expect that this will soon be corrected with all of the above mentioned research leading to specifications in the first half of the coming year. In the meantime, projects have been granted permission to use conservative versions of the anticipated specifications in their planning and implementation.

Advances in assured containment of any returned samples will be critical to the formulation of the MSR campaign, the highest priority for planetary flagship missions in the latest Decadal Survey.

<u>Recommendation A:</u> A streamlined approval process should be developed, as well as instruction on the newly available forward-planetary protection techniques. Plans for MSR technology development for assured containment must be carefully coordinated with concept studies and formulation efforts.

Finding B: Levels of interest for particle redistribution models have been based on optical performance of contamination-sensitive systems. These models generally ignored the redistribution of particles smaller than 50 μ as they were not a large contribution to the loss of performance (mainly caused by particles larger than 100 μ). These smaller particles are difficult to control because they are not visible to the naked eye during assembly build, integration, and test activities. However, they will be the largest contributor of biological contamination.

In addition, the interaction of micron and submicron particles with very small levels of molecular contamination (non-uniform below 100 Angstroms) is not well understood. There are many competing physical phenomena occurring when a particle interacts with a surface. If there is molecular contamination, the particle may be accommodated on the surface by physisorption, chemisorption, electrostatic forces, etc. These phenomena may have a significant effect on meeting the astrobiology experiments requirements for reduced carbon contaminants. Additional factors for consideration: movement of particles in lower or higher than terrestrial atmospheric pressures, lower or higher than terrestrial gravity, interaction with planetary atmospheres (charging, chemical interactions), and planetary dust storms.

<u>Recommendation B:</u> The effect of non-uniform molecular contamination on micron and submicron particle contamination levels should be determined.

4.3 Education and Training

Finding A: Solicitations for low-TRL instrument technology development proposals do not address planetary protection; therefore, technologists looking toward potential MSR or Europa missions may not be aware of the planetary protection requirements or implementation techniques, and thus are not designing their technologies to meet these requirements even though the technology is still at an early stage and consideration of these requirements at an early stage could significantly reduce overall instrument costs.

<u>Recommendation A</u>: Solicitations for early instrument technology development should include requirements for planetary protection. Education and training should be offered to all interested proposers at a level commensurate with the proposed efforts. In some cases, proposers might be advised to take the excellent planetary protection class offered by the NASA PPO. In other cases, ensuring the proposer is sufficiently educated with respect to the system implications of planetary protection and contamination control may be adequate. All proposals should be required to delineate their approach to planetary protection and contamination control if applicable.

Finding B: Contamination control experiences are not being captured in adequate form. This experience base stretches back to the Apollo era, is held by dozens of widely dispersed persons (many now retired), and is not recorded in any convenient place. In fact, most of the information is effectively not recorded at all.

Recommendation B: NASA should support the creation of a living document detailing experiences with contamination control and curation for previous missions, to help present and future missions avoid costly mistakes. This document could be constructed around a wiki, permitting information to be collected from the widest possible range of persons. In addition, it could be included in the NASA Lessons Learned program. The timing of this recommendation is critical since the generation of Apollo scientists and technicians is quickly disappearing.

Acronyms

ATLO	assembly, test, and launch operations
ATP	adenosine triphosphate
BSL	biosafety level
COSPAR	•
	Committee on Space Research
CSSR	Comet Surface Science Return
DHMR	dry heat microbial reduction
DPA	dipicolinic acid
EEV	Earth entry vehicle
EOL	end-of-life
ESA	European Space Agency
FTIR	Fourier transform infrared spectroscopy
FY	fiscal year
GC-MS	gas chromatographic mass spectrometry
GSFC	Goddard Space Flight Center
IC	ion chromatography
ICSU	International Council of Science Unions
JAXA	Japanese Aerospace Exploration Agency
JUICE	Jupiter Icy Moon Explorer
JPL	Jet Propulsion Laboratory
LAL	limulus amoebocyte lysate
LC-MS	liquid chromatography-mass spectrometry
LPS	lipopolysaccharides
LPSAC	Life and Physical Science Advisory Committee
MAVEN	Mars Atmosphere and Volatile Evolution Mission
MEPAG	Mars Exploration Program Advisory Group
MER	Mars Exploration Rover
MRO	Mars Reconnaissance Orbiter
MSL	Mars Science Laboratory
MSR	Mars Sample Return
NASA	National Aeronautics and Space Administration
PCR	polymerase chain reaction
PPO	Planetary Protection Office
PPWG	Planetary Protection Working Group
PRA	probabilistic risk assessment
Q-PCR	quantitative polymerase chain reaction
ROSES	Research Opportunities for Space and Earth Sciences
RTG	radioisotope thermoelectric generator
SAM	Sample Analysis at Mars
SFC-MS	supercritical fluid chromatography-mass spectrometry
SRF	sample receiving facility
TGO	Trace Gas Orbiter
TOF-SIMS	time-of-flight secondary ion mass spectrometry
TRL	technological readiness level
UV	ultraviolet

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