FINAL REPORT

3rd COSPAR Meeting on: Refining Planetary Protection Requirements for Human Missions

Addressing Planetary Protection Knowledge Gaps in Microbial and Human Health Monitoring

May 14-16, 2019

Lunar and Planetary Institute, Houston, Texas

Prepared for the COSPAR Panel on Planetary Protection (PPP)

Editors:
Margaret S. Race, J. Andy Spry,
Bette Siegel, Lisa M. Pratt and Gerhard Kminek

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Held under the Auspices of the

Committee on Space Research (COSPAR) of the
International Science Council (ISC)
at the Lunar and Planetary Institute (LPI)
Houston, Texas

Prepared for the COSPAR Panel on Planetary Protection (PPP)

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Editors

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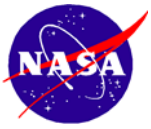

Lisa Pratt, NASA Planetary Protection Officer

Gerhard Kminek, ESA Planetary Protection Officer

With assistance from SETI Institute and LPI

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3rd COSPAR Meeting on

Refining Planetary Protection Requirements for Human Missions 2019

Executive Summary

COSPAR (the Committee on Space Research) and various space agencies are supporting a multi-year stepwise process to identify, prioritize and plan the research and technology development needed to address planetary protection (PP) requirements for human missions beyond Earth orbit. The objective is to incrementally move from the current qualitative COSPAR PP "Principles and Guidelines" towards the development of quantitative PP requirements for future human missions to locations like Mars. The workshops and meetings in this series have involved participants from COSPAR, the US National Aeronautics and Space Administration (NASA), European Space Agency (ESA), Japan Aerospace Exploration Agency (JAXA) and other international space agencies, as well as the scientific and technical community, and commercial/private stakeholders.

This report provides detailed findings of the 3rd COSPAR meeting on Refining PP Requirements for Human Missions, which focused on addressing the specific knowledge gaps (KGs) associated with Microbial and Human Health Monitoring (MHHM). In the planetary protection context, microbial monitoring of the crewed environment is needed to ensure that the systems remain within acceptable limits for:

- Mitigating a contamination threat to Mars (forward planetary protection);
- Being a healthy and functional environment for the crew to live in;
- Mitigating a contamination threat to Earth (backward planetary protection).

Similarly, microbial monitoring for human health is needed to ensure that it is possible to tell if a sick crew member is just sick, or if they are potentially infected by putative Martian biology, prior to their returning to Earth. The three day meeting was held May 14-16, 2019 at Lunar and Planetary Institute (LPI) in Houston, TX, and included a combination of plenary presentations and small group sessions that built upon the findings of earlier workshops in this series (Johnson et al., 2015, Kminek et al., 2016 and Race et al., 2018).

This meeting found that issues of microorganisms and human health will continue to be applicable to both forward contamination (in combination with contamination transport models) and backward contamination for human mission to Mars. There are significant synergisms between Earth safety concerns (planetary protection) and issues relevant to assessing crew health status on long duration missions.

The **findings** are as follows:

1. ISS is the only existing, useful test-bed to obtain long-term baseline data and trends useful for preparing for human missions beyond Earth orbit.
2. Current routine microbial monitoring on ISS is limited in scope (number of crew and locations on ISS), depth (details of microbial populations) and frequency, of data collection.

Based on these findings, the following **recommendations** were formulated:

1. Systematic microbial monitoring of ISS crews and associated ISS environments should be done more frequently in order to obtain statistically relevant data over long periods of time and multiple crew complements.
2. Existing instruments and technologies can be used (ex. MinION, Oxford Nanopore with flight heritage on ISS) to monitor microbial levels for these purposes.
3. It is possible to build upon what is already in use on ISS—including associated processes, consumables and crew time-needs, which are already well understood.

The **way-forward** for addressing MHHM KGs in a timely manner includes a step-wise approach built on:

1. Data-mining activities of existing ISS and other terrestrial databases to establish starting points for ISS sampling (frequency, number of samples) and to write revised sampling and analysis procedures using the MinION equipment (or other flight heritage systems).
2. Integration of the data-mining information and MinION procedure outputs (above) to create an updated microbial monitoring plan for the ISS and crew that would address the MHHM KG.
3. Discussion of flight opportunities with ISS partners.

After gathering data on the ISS and prior to sending humans to Mars, initiate similar microbial monitoring beyond Earth's orbit to study the effects of the radiation environment (e.g. at Gateway) and conditions on a lifeless surface (e.g. the Moon), which the groups considered to be "must-have" additional information for interplanetary missions.

The product of these activities would provide necessary inputs to develop quantitative planetary protection requirements for human missions to Mars. At the same time, the results of these activities would inform the path to engineering of crewed systems and writing operational procedures that would mitigate contamination in the context of forward planetary protection.

1. Background and Context in Human Mission Planetary Protection Requirements

Since the end of the Apollo Program, discussions about planetary protection (PP) policies have focused mainly on avoiding forward contamination of the Moon and other celestial bodies during robotic exploration missions beyond Earth orbit. In the intervening decades, missions with human crews have remained in Low Earth Orbit (e.g., MIR, Space Shuttle, ISS), where COSPAR PP polices and requirements about forward or backward contamination do not apply. When discussions arose in the early 2000's about sending humans beyond Earth orbit and on to Mars, it was apparent that PP policies for human missions would need updating to ensure that that effective controls and safeguards would be integrated in all phases of mission planning and implementation.

In addition to considering the latest astrobiological findings about different planetary locations, there is a need to integrate the evolved understanding about human and environmental microbiomes, in determining how to break the chain of contact (for crew and returned materials) and protect Earth from a potential hazard posed by extraterrestrial matter carried by a spacecraft returning from an interplanetary mission.

1.1 Planetary Protection and Renewed Human Mission Planning

The initial steps toward re-considering PP policies for human missions took place nearly two decades ago, when the international space community initiated a series of studies and workshops to examine issues associated with possible human missions to Mars (e.g., Criswell et al., 2005; Hogan et al., 2006; NRC/SSB 2002; Kminek et al., 2007). These deliberations eventually led to COSPAR's development of a set of qualitative Principles and Implementation Guidelines for Human Extraterrestrial Missions (see Appendix A: COSPAR 2008), which even today remains part of the COSPAR official PP policy (Kminek et al., 2017).

These same principles and guidelines have informed the National Aeronautics and Space Administration's (NASA) Policy for Planetary Protection and Human Extraterrestrial Missions (NPI 8020.7 and NPD 8020.7G). Publication NPI 8020.7 recognizes the need to generate detailed scientific and technological knowledge in order to establish requirements and specifications that would enable NASA to incorporate planetary protection into the development of crewed spacecraft and missions. To move toward this goal, NASA worked with the broader community to develop an incremental path forward. The overall objectives aimed at identifying and addressing key knowledge gaps (KGs) and determining what research and technology development are critical for developing PP requirements for future human spaceflight missions.

A tentative plan based on a series of workshops was outlined as a way to focus on the needs of future human rated flight systems. Three specific areas of importance were highlighted for further examination:

1) Microbial and Human Health Monitoring (MHHM) needs;

2) Technology and Operations needed to implement planetary protection within missions; and

3) Understanding the Natural Transport of Contamination on Mars.

As a first step in the road mapping process, NASA conducted a systematic literature search which identified ~ 100 publications with technical analyses and information related to PP and human missions (e.g., Johnson et al., 2016; Spry et al. 2014).

Subsequently, a series of collaborative meetings have been held, first under NASA auspices, and subsequently through COSPAR and including the broad international community. These have sought to identify and address planetary protection KGs in a stepwise fashion. Figure 1 provides an overview of the evolved approach to address the critical KGs necessary for development of quantitative PP requirements for human extraterrestrial missions:

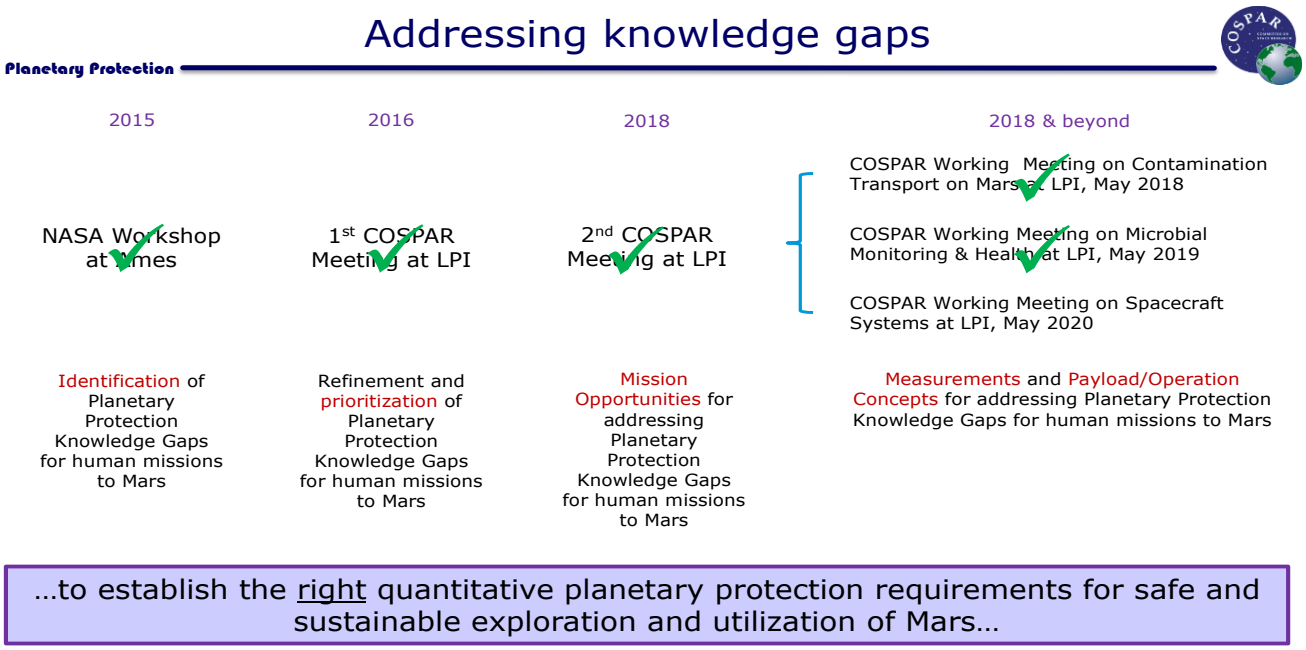

Figure 1:.Evolved NASA-COSPAR Process for Development of Quantitative Planetary Protection Requirements for Human Missions.

1.2 Overview of Earlier Workshops and Findings to Date

Prior to the 2019 working meeting on MHHM, four other PP workshops and meetings were held. The earlier workshops (particularly the one in 2018) provided comprehensive background information and the context for the MHHM workshop reported here. The previous reports and findings can be accessed at https://sma.nasa.gov/sma-disciplines/planetary-protection under the "Conference Reports" tab. The 2018 report consolidated the KGs that would be addressed in this 2019 working meeting.

2. Meeting Overview

The COSPAR Working Meeting (2019) on MHHM was held at Lunar and Planetary Institute in Houston Texas, May 14-16, 2019 with the intent of focusing on KGs identified in earlier workshops:

A. Microbial Monitoring of the Environment
B. Microbial Monitoring of Humans
C. Mitigation of microbial growth in spacecraft systems, and
D. Operational guidelines for PP and crew health.

The overall aim was to produce a meeting report detailing the specific measurements and instrumentation needed for microbial monitoring of crews and crewed vehicles and to consider approaches useful for mitigating microbial growth in spacecraft systems in advance of the first human missions to Mars. This report summarizes the results of the three-day meeting, which was divided into two parts.

The first day of the meeting involved a series of plenary presentations providing the meeting context and objectives, including a review of information on COSPAR and its Planetary Protection Panel (PPP), and explanation of the stepwise workshop process for developing future quantitative PP requirements for human missions. Attendees heard briefing presentations by agency and industry leaders on workshop findings to date, the current state-of-the-art in PP methods, upcoming mission opportunities and proposed timelines for filling identified KGs, and an update on recent changes within NASA relevant to PP implementation. Summary information was also presented on two recent PP reports, one by a National Academies review panel (2018) and another by an Independent Review Board (Stern et al., 2019).

On the morning of the second day, participants toured NASA's Johnson Spaceflight Center facilities to provide the group with context on spacecraft hardware. The tour was followed by a briefing with instructions, templates and context for breakout group discussions ahead.

Breakout Group deliberations began in the afternoon of the second day of the meeting. Building on the findings from the 2016 and 2018 COSPAR workshops and the first COSPAR Work Meeting (Race et al., 2018). Participants at this 2019 meeting were instructed to consider the specific KGs related to MHHM in the context of mission opportunities and timelines identified earlier.

Prior to the start of the meeting, it was determined that, while KG 1D remains a concern for the MHHM group, it should be set aside for later deliberations because operational guidelines will naturally follow from closing the 1A, 1B and 1C KGs. Another driver for setting 1D aside was that additional data from the other two discipline areas (Technology & Operations, and Natural Transport of Contamination) was needed prior to developing operational guidelines for addressing planetary protection and crew health. Thus, the examination of only KGs 1A, 1B and 1C were included in the breakout group deliberations at this meeting.

On the final day of the meeting, NASA astronaut, Kate Rubins provided a virtual presentation where she provided a detailed, first-hand perspective of the challenges of living, and working onboard the International Space Station (ISS). The presentation provided the breakout groups with a fresh perspective and the opportunity to re-evaluate their developing findings in light

of this new knowledge. Following the virtual meeting and a brief plenary progress review from each breakout group, the participants resumed their discussions in breakout sessions. Over the two days of deliberative work, each breakout group had approximately seven hours of focused discussion to consider ways to address KG's 1B, 1A and 1C (see Appendix C-Agenda).

2.1 Meeting Context and Considerations

The first day of plenary presentations set the context for discussions. Before addressing how to fill the KGs for MHHM, attendees were reminded of the multi-phase context and complexity associated with the development of future quantitative requirements for human missions. For example, current robotic missions to Mars incorporate an assortment of PP measures that impact different mission phases and systems—from bioburden reduction of flight hardware using solvent cleaning, dry heat, ionizing radiation and gasses; to re-contamination prevention using flight and non-flight filters and barrier systems; and the required use of bioburden controls and verification assays throughout assembly, test and launch operations (ATLO). In addition, PP planning and implementation also requires information and analysis on the control of spacecraft from launch to orbital and/or landing phases, details on equipment reliability, as well as the potential outcomes for off-nominal scenarios (accidental impacts, navigation errors, etc.).

Addressing MHHM knowledge gaps for human missions will similarly require a multi-phase approach to PP implementation; on the outbound flight, during Mars surface operations, on Earth-return flights, and during Earth based analyses and characterization of returned materials. There is a clear need to monitor initial bioburden loads, gather systematic data on the status and evolution of microbial communities on both the human crew and the human flight systems, and to understand levels of biological and organic contamination release from human support systems. This data is not only critical for establishing quantitative PP standards for human missions to Mars, but will also be needed for the design of hardware and operations in compliance with evolving PP requirements.

For decades, PP requirements have been defined on the basis of spore-based bioburden levels and category-specific requirements for different mission targets and activities. While PP requirements for robotic Mars missions, up to and including planning for the first Mars Sample Return (MSR) mission, are still based on spore counts and culture-based assays, this will undoubtedly be revised well before human missions to Mars. For example, advances in genomic characterization of terrestrial organisms and the human microbiome have spurred development of new chemical and optical tools for rapid detection of bioburden in and on spacecraft. Such methods and instruments are currently being used in parallel with the existing spore based assays currently used on pre-launched hardware and materials and in time, will likely replace them. Planned MSR missions in the early 2030s are likely to integrate new genomics based tools and updated sterilization modalities in different phases. Beyond that, the updated technologies and methods might even become the standard for containment and test facilities performing scientific analyses of returned sample materials following a robotic MSR campaign. New modalities for sterilization and inactivation of biological agents and organisms are also being developed. We can already anticipate how these advances may be used as part of PP implementation for future human missions.

Because of the unique opportunity presented by ISS and its operating life cycle, ISS was identified, during the 2018 workshop, as a key test-bed for systematic long-duration microbial monitoring relevant to both crews and crewed environments on future Mars missions. This may be accomplished using existing systems with known flight heritage (e.g. MinION) to systematically collect statistically relevant microbial data in different locations over long time-periods, and across multiple changes of crew and incoming re-supply shipments. Use of existing ISS hardware and procedures– in addition to relevant Earth analogue facilities—has been acknowledged as a way to address the time-critical "Highest Priority KGs" identified during deliberations after the 2018 COSPAR meeting. This data-gathering could be initiated and incorporated in the established timeline over the next several years.

While there is an archive of microbial and other data from ISS and its associated infrastructure, it is clear this is not sufficient for determining PP requirements for future long duration human missions (see Figure 2). In fact, routine microbial monitoring on ISS is only done quarterly and is limited both in scope (i.e. number of crew and locations sampled) and depth (i.e. limited investigations on different microbial populations). While individual research activities carried out by NASA, the Japan Aerospace Exploration Agency (JAXA) and the European Space Agency (ESA) have been detailed and comprehensive, they are irregular and infrequent, and not always comparable to each other (e.g. investigators using different analytical methods). In order to generate long-term, statistically relevant baseline data and trends for astronauts and crewed flight systems, a more strategic approach is needed.

One desirable outcome is the establishment of a systematic effort to obtain an understanding of the baseline human microbiome and associated spacecraft environmental microbiome and how it changes during spaceflight. Closing these significant knowledge gaps expeditiously, and in a way that aids decision-making concerning human mission PP implementation is key for long duration missions beyond Earth orbit. These types of considerations helped set the context and framework for the deliberative part of the meeting.

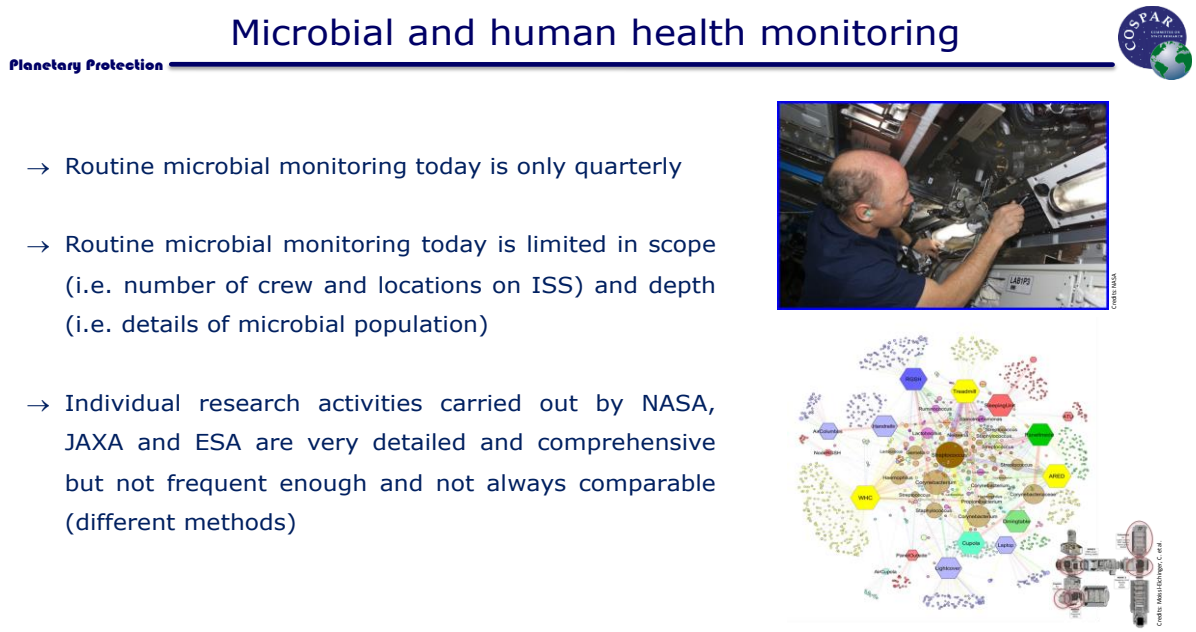

Figure 2: Microbial and human health monitoring

2.2 Breakout Group Tasks and Assignments

For the deliberative portion of the meeting, participants were assigned to one of three breakout groups to examine the MHHM KGs in detail (attendees and breakout group assignments are included in Appendix E). In addition to setting aside KG 1D for a later time, the groups were instructed to examine the KGs in an altered order, focusing first on crew microbial monitoring (KG 1B), then on microbial monitoring of spacecraft environments and crewed vehicles (KG 1A) and lastly, on possible mitigation measures (KG 1C).

KG 1B: Microbial Monitoring of Humans
KG 1A: Microbial Monitoring of the Crewed Environment
KG 1C: Mitigation of microbial growth in spacecraft systems

The groups were asked to use the timeframe of 2019 until ~2024 and reference information from the 2018 Workshop to identify and record the level of operational monitoring of microbes, and associated mitigation strategies needed to address MHHM KGs. In addition to identifying measurements, instruments, and equipment that can be used to fill KGs, the groups were also reminded to give special consideration to opportunities on ISS, Orion, Gateway and subsequent vehicles, as well as relevant ground analogues.

The suggested approach for breakout discussions were as follows:

2.2. A. KG 1B. How do we systematically conduct microbial monitoring of humans?

Goals

Identify systems and consumables needed for a sufficient approach to provide data at the needed frequency and resolution.

Issues

- How much (monitoring) is enough?
- What measurements are needed (e.g. is photon counting preferred to colony counting)?
- Nominal and non-nominal (clinical manifestation) cases? c.f. ordinary health monitoring.
- Suggest methods for: sampling, processing, measurement, analysis, and storage for both data and materials.
- Consider Pre-, During- and Post-exposure to the Martian environment – as well as how to establish baseline, and then monitor normal vs abnormal.
- What are the outcomes (actions?) of abnormal data?

Discussion topics

Break down discussions by categories using the template?

2.2. B. KG 1A. How do we systematically provide for microbial monitoring of the environment?

Goals

Identify systems (instruments?) and consumables needed for a sufficient approach to provide data at the needed frequency and resolution.

Issues

- How much information is enough?
- Different venues for data collection? – Interplanetary transit, surface habitat + leak paths, Vehicular, extra-vehicular activity (EVA) + leak paths, Suit EVA + leak paths etc.
- What measurements (e.g. is photon counting preferred to colony counting)?
- What Methods for: sampling, processing, measurement, analysis, and data (and materials) storage?

Discussion topics

Break down discussions using template.

2.2. C. KG 1C. How do we design spaceflight systems to mitigate microbial growth?

Goal

To ensure flight system compatibility with overall microbial management strategy.

Issues

- How to address/mitigate growth: (review, audit, training; mitigation strategies and approaches)?
- Is this a topic for microbiologists to make recommendations for engineers, but not solve at the workshop?

Discussion topics

- Pre-launch design vs post launch mitigation.
- Mitigation option selection(s).
- Timing of interventions vs graceful decay (of environmental quality).

2.2. D. KG 1D. What operational guidelines are needed to understand planetary protection concerns & crew health?

- Set aside—this will be addressed later when data from all three study areas can be combined.

Templates were provided to guide deliberations related to monitoring of crew (KG 1B) and crewed environments (KG 1A) (see Appendix D). The template categories for consideration, included:

- **Information** type that should be collected (types of organisms and from where)
- **Equipment** needed on board (including considerations about storage), and/or on the ground
- **Consumables**
- **Frequency of sampling**
- **Locations of sampling**
- **Sample Processing**
- **Data Analysis**
- **Other considerations**:
  - What information is needed?
  - Is the measurement sufficient?

- Will backup verification be needed (multiple tools/methods e.g., Metabolism vs. DNA, vs. culture methods)?
- What are the minimum acceptable limits for data collection (time, distance, other)?
- What will it take to close the gap for PP purposes (not for other motivations for monitoring such as science interest)?

Finally, a set of discussions questions were posed surrounding the central question '*when do we know enough?'* —particularly in relation to KG 1A and KG 1B:

- Is data mining 'nice to have' or mandatory to close the KGs?
- Is data gathering post-ISS (e.g., Orion, Gateway, crew on the Moon) 'nice to have' or mandatory to close the KGs?
- What is the natural stopping point for data gathering (when will we know enough about the KGs), or can this only be decided after having started the initial data analysis?
- Are there any short-term ground-based activities necessary to get a system operational (upgrade, delta-qualification, etc.)?

3. Breakout Group Deliberations

Each breakout group undertook discussions in separate rooms over two days, guided by the templates and suggested questions. Brief summaries of group findings were presented in a plenary session at the end of the meeting, followed by submittal of post-workshop reports by the chairs and scribes of each subgroup. The detailed findings of the three breakout groups are included in Appendices E, F and G.

After the meeting, the reports from the three groups were compiled in tables for comparison of the findings for all three KGs. While each group took a slightly different approach, there was consistency across the suggested actions on how to gather data to address KGs on microbial monitoring of human crew, crewed environments, and mitigation concerns. The groups agreed with the initial assertion that the current frequency of routine microbial monitoring on ISS is insufficient and an increased frequency is needed. It was also determined that crew sampling needs to be broadened to capture the diversity of microbial populations, and sampled at multiple anatomical locations and situations, both routine and event driven or off-nominal. The selection and use of standardized methods and technologies will be essential for comparing data information collected at different times and by different international partners. Data mining of archived information from past ISS sampling by national space agencies should also be included as a separate task to determine if obvious trends and findings can be identified.

Overall, the combined findings and comments of all the groups indicated that:

- Closing the PP KGs can be done using ISS as a testbed to obtain systematic microbial monitoring data on crews and crewed space flight systems.
- There is a need to collect and analyze ISS and other analogue data on a more frequent basis to obtain long-term, statistically relevant baseline data and trends for astronauts and crewed flight systems. Decisions about the frequency and number of samples can be determined from data mining and analyses of information from ISS and Earth based isolation test campaigns.
- In addition to beginning a data mining effort, there is a need to initiate a shift to standardized genomics based methods and instrumentation (e.g. the Oxford Nanopore "MinION" system), while continuing the use of culture/spore based methods on the ISS as complementary and comparative data in the near term.

Once there is a broader understanding of the microbial levels and patterns of crew and crewed environment from analysis of ISS data and research analogues, plans for mitigation measures and strategies can be addressed in more detail. Tables 3.1 and 3.2 below provide a cross comparison of the group findings for KG 1B (crew) and KG 1A (crewed environments). A summary of findings on measurements and instrumentation for mitigation (KG 1C) are described in the next section.

3.1. Findings on KG 1B: Routine Microbial Monitoring for Human Health

During discussions about routine monitoring of human crew, the groups agreed that quantification of microbial burden was needed across a broader diversity of microbial taxa, and at greater frequencies than recorded in previous missions, and across multiple locations on the human body during nominal and off-nominal situations. While molecular methods and instrumentation should be used, it is prudent to maintain the option for use of culture-based methods as comparative, supplemental or backup data. In addition to determining baseline data for individual crew members, it is also advisable to consider how crew changes and arrival of cargo, etc. (e.g., plants and accompanying microorganisms, etc.) could alter the human microbiome or be reflected in crew physiological data, immunological response etc. In addition to focusing on molecular and genomic methods for recording microbial levels and types, it is also advisable to develop associated methods and instruments to optimize particle counting, bioburden identification, and appropriate automation, and possibly adapt methods used in other closed systems (e.g. Earth analogue isolation and biomedical, labs, submarines, etc.). Sample processing should consider both on board and on-ground options, with bio-archiving and data mining kept in mind for determining baseline, event driven and alert-levels over the long term. Relevant meta-data should also be recorded along with microbial levels (temperature, humidity, radiation levels, humidity percent, cleanings, etc.).

3.2 Findings on KG 1A: Routine Microbial Monitoring of Crewed Environments (vehicles, spacecraft)

During discussions about the routine monitoring of crewed environments, the groups focused mainly on interior locations, with some comparisons between interior and exterior levels of microbes (alive or dead). Regarding microbial monitoring of crewed environments, all groups indicated that quantification of microbial burden is needed across a broader diversity of microbial taxa, and at greater frequencies than routinely sampled on ISS. In addition to determining more detailed baseline data for ISS, it is also advisable to consider how crew changes and arrival of cargo, etc. (e.g., plants and accompanying microorganisms, etc.) correlate with fluctuations of microbial levels under different micro-environment conditions and on various types of surfaces inside modules. Special attention should be given to strategies for sampling locations inside and between different modules, whether event driven, regularly sampled, or out of the way places. As an additional source of data, discarded items (towels, trash, clothes etc.) should be sampled to gain additional insight into microbial types, fluctuations and baseline vs. perturbation levels in different locations.

Two of the breakout groups also briefly discussed microbial levels and suggested monitoring exterior locations. While such considerations are relevant to leakage and dispersal of microbes during long duration space flight, they are also a particular consideration for future surface mission activities, during EVAs, roving, cache pickups, sample containment, testing and possible mutations may be involved.

Table 3.1 All Groups: KG 1B: Routine Microbial Monitoring for Crew Health

	GROUP 1	GROUP 2	GROUP 3
Information	Bioburden: what is it? Quantification of constituents; What is the risk potential? Constituents of human microbiome (Bacteria, Fungi, Viruses, archaea, Eukaryotes) Level of detail? Spore formers + radiation resistant (Type C-via Space Studies Board) Also account for microbes associated with plants brought on board Molecular analysis preferred- cultures still needed as backup & on ground. Metadata for sample collection- Include physiological data and medications taken;	Bacteria? Fungi? (impt) Viruses? Archaea? Taxonomic Level- as deep as possible; Constraints from database and technology. Also shotgun info (non-targeted) Culture/- keep option to grow OR Instead: assess viability (KG to improve these assays) Molecular- MinION; need to resolve test sampling methods and data- processing methods (machine learning, autonomy required.) Associated Human physiological data: maybe even more important. Correlate with microbiological info; KG- which human parameters are important? (Medical? Psychological? Need to be requested/included as additional data areas? Include additional medical checks for return flights	Bacteria? Fungi? Viruses? All Plus Archaea Taxonomic Level: Highest Resolution possible- species level required Molecular,- but retain the ability to culture and use these isolates for further analysis Need to determine baselines for individual. crew members, (normal/abnormal, and links to other physiological responses); Frequency should be more often than on ISS Routine Monitoring w/new techniques—On Board Must consider high background radiation. (monitor to detect alien infection?; changes to microbiome; immunological responses; transcriptomics- biomarker monitoring) Study animal colony analogs and ISS Microbiome analysis alone not sufficient—need orthogonal data to understand ongoing changes (ex. nominal/off-nominal; and patterns over time) Identify patterns & variability over time w/ large data sets—for individual humans (crew); also create software package to provide risk assessment.
Equipment	On-board (incl. storage): Need to improve /optimize techniques before analysis by MinION. How to isolate microbiome in collected samples in orbit? on Ground- Microfluidics and automated sample processing	On-board (incl. storage) - very small for Mars (limitations but selection) different requirements for data gathering (than ISS?); same equipment for life detection? On-ground- Need analogue research such as Concordia, submarines etc.)	On-board - Eventually Need Onboard capabilities for microbiome identification needed; multiple methods Big data analytics and AI/machine learning – automated system On-ground- Additional ground-based human and animal colony assessments (animal analogues) to develop baseline. Multiple methods Other Miniaturized hardware – e.g., Particle counter airborne system (NAD/NADP); biowarfare agent sampling systems?
Consumables	Use Non-liquid & Liquid-- prefer non-liquid depending on shelf life	Non-liquid- swabs, tubes etc. Liquid- extraction buffers- lyophilized, water-UV, DNA-free) Consider recycling of consumables.	Non-liquid- lyophilized wherever possible Liquid- kept to a minimum Need to consider e.g., crew of 6; 4 sample sites per crewmember; 1 sample a week for 6 months…

Frequency	Nominal based on Human Microbiome Project data. Event driven during critical events (e.g., rash, fever etc.) Locations, frequencies and revisit rate will depend on specific event(s).	Before, during, post flight Fixed (daily, weekly, monthly,…) and Event driven Test on-board/analogue Necessary to identify frequency, replications (triplicated?); Data limited for confined systems;	Before, during, post flight Fixed (daily, weekly, monthly): At a minimum, sampling every 30 days ; 7 days optimal for routine sampling Event driven- YES. In addition to nominal sampling, do prior to every crew/cargo arrival and departure [event driven plus routine];
Locations	In situ-on board: skin, nose, mouth, hair, urine, blood, fecal; event driven (e.g. tears, ears, throat.)	Surface (body)- forward contamination Liquid (saliva, urine); Solid (stool) TEST ALL- then down -select for long-term monitoring	Surface (body), Liquid (saliva, urine) , solid (stool); Understand leakage of contaminants too
Sample Processing	On-board and On-ground	On-board KG: processing samples & DATA (machine learning required,) On-ground- Bioarchiving of samples and parallel analysis on ground	Ground Based for analog studies On board with new methods – HERA (analogue environment) for short term analysis?
Data Analysis	Current On-board analysis is enough to meet info needs for generic phylogenetic analysis; Shotgun meta-genomic analysis and species level analysis—requires more powerful on-ground capabilities Machine learning is avenue for improving capability	On-board (incl. expected link budget) autonomous as much as possible *Looking ahead:* On-ground—Data Mining is mandatory to close KGs—Also need short term ground-based activities to get a system operational (e.g., focus on upgrades, nanopore, sample processing, data analysis, testing of biocompatible materials and engineering solutions) On Board: short-term data gathering on ISS (and Gateway, Artemis/ Moon etc.) On Board use is mandatory to close KGs – and to develop analogue and test facilities for Mars.	Use Metadata analysis of all previously collected data to: Design and implement ground-based analog study to determine microbiome baseline (w/ fluctuations & anomalies) Immunological monitoring could be key data from analog Need to compare ground based with ISS data. Start immediately on ISS and run concurrently with suggested ground studies

Figure 3: Table 3.1: KG 1B: Routine Microbial Monitoring for Crew Health

Table 3.2a All Groups: KG 1A: Routine Microbial Monitoring of Crewed Environment (vehicles, spacecraft,) INTERIOR

	GROUP 1	GROUP 2	GROUP 3
Information	Identify constituents of crew vehicle-microbiome (bacteria, fungi, archaea, eukaryotes) Spore formers + radiation resistant (Type C-via Space Studies Board)? Distinguish between microbial sources (human, plants, cargo) Quantify constituents of bioburden Relevant metadata (temp., humidity, materials/surface type; atmospheric composition; visible sites), other? Before sample collection-- assess active air, UV assessment of surfaces, filters, biofilms	Bacteria fungi (impt) Viruses? Archaea Cult/ molecular- BOTH methods needed initially Air/(active/.passive); surfaces (dry/wet) filters (if available? dust on filter) Water-liquid systems (how to sample & process—sensitivity is an issue, biomass too low?) Particulates counting in background- amount of microbes, steady-state situation. Alert-level—what action is necessary? Real time monitoring needed. Cultivation still necessary and comparisons to sequencing (e.g. Bacillus e.g. might not be well detected by sequencing) - Determine correlation between both techniques. How to address the unknowns? Many biosignatures unknown…Another KG- data to define the decision criteria?	See below—under frequency See comments in text version under Crew Health screening too, Understand what's going on inside—and compare with outside, Molecular data needed to support
Equipment/ Collection/ Processing	On-board (incl. storage)- Molecular analysis preferred Nonspecific biol. monitoring first to assess number of non-viable samples to be collected (c.f. LALR, biovigilance approaches) Automated DNA extract. w/ multiple samples (Library prep before Min); Thermophoretic Sampler (for airborne samples); and MinION On-ground- cultures maintained as backup	On-board (incl. storage) On-ground? Disturbance events to study (cargo, crew exchange etc.) to test flexibility and effectiveness of system	Include multiple techniques: - MinION (up front sample prep) - PhyloChips (what are the probes?) - Build a MALDI-TOF (multiple databases) - Culture methods (how much?) - Fluorescence data Should cleaning processes of hardware/cargo and vehicles be assessed?
Consumables	-	Non-liquid Liquid	Need to consider-
Frequency	Based on terrestrial sampling from Human Microbiome Project (may not be relevant to space environment?) Likely schedule- weekly?	Fixed + event driven (also confirm after cleaning) -Increase info, as much as possible -Ground Analogues to determine frequency\ -Gateway monitoring (as relevant to Mars trip) -Check with info from Pharmaceutical industry	-Data exists for other outside confined environments—need to look there before determine frequency -Current monitoring is insufficient—need frequency more often than monthly—(weekly?)

	Unplanned events: (growths, leaks, smells, analysis anomalies) EVAs will require their own sampling routines (suits, airlocks, vehicle exteriors); Frequency driven by results of initial sampling	-Need establish *target/baselines (for* monitor & follow up on actions)	-Monitor/sample before/after crew, cargo changes & other event related situations
Locations	Event Driven locations (incoming cargo; Spacecraft exterior (vent sites, high traffic areas) Suits (before/after EVAs) Airlocks ;) Regularly Sampled: filters; bathrooms, automated air samples near high traffic areas; food, water, waste stream, sleeping quarters; near biological experiments (rodents: need baseline and genomes pre-flight) also Plants. 'Out of the way" places (e.g. behind racks; likely need less frequent sampling)	All modules; One module Cycle between modules	All modules – anticipate richest biodiversity near galley and WHC Inlets and outlets of systems; leak points (consider condensation / wet spots on surfaces of ISS)? Maybe also external samples? (compare inside vs. outside), microbiota (airlocks.; venting, areas spacesuit activities;) leak points Use discarded items to gain microbial insight (e.g. clothes, towel, wipes, trash etc.)? Maybe compare with Navy data &/ or ISS Make risk assessments for different areas—what are baselines & perturbations? HEPA filters as early warning systems prior to symptoms? Screen for virulence genes within crew microbiome
Sample Processing	--	On-board? Data analysis & interpretation (how severe? Define what is normal, what not) Machine learning gives info, but doesn't decide. Bioarchiving,- making sure 'waste' is exploited	On-board? On-ground? Archival data. Both- On board sampling, analysis and ID methods required, but maintain option to do additional assessments on ground.
Data Analysis	Archived biological samples from waste and disposables Metadata- particle count	On-board (incl. storage) On-ground Disturbance events to study (cargo, crew exchange etc.) to test flexibility of system	On-board (incl. expected link budget) On-ground Include multiple techniques & methods: on board capability definitely required (May need separate workshop to consider plant microbiota)

Figure 4: KG 1A: Routine Microbial Monitoring of Crewed Environment (vehicles, spacecraft,) INTERIOR

Table 3.2b KG 1A: Routine microbial monitoring of crewed environment (vehicles, spacecraft,) EXTERIOR

	GROUP 1	GROUP 2	GROUP 3
Information	**No info for Exterior**	-Use Witness Plates; Check for level of human contamination outside -Determine microbial leaks (level, type), -Ground simulation might be needed (already on ISS?) to address: -Suit sampling (inside/outside); -exterior ISS sampling) -Suitable Tests on Moon? - New PP requirements needed for Human Missions (Represents Final KG to be filled) Disturbance events to study (e.g., when cargo, crew exchange, etc.)- to test flexibility of system	
Equipment		On-board (incl. storage) On-ground Witness Plates Detection of human contamination outside (ongoing research on Concordia) Rovers sample outside for contamination (what distance?) Analyze outside surface of spacecraft	Multiple techniques Portable sampling devices
Consumables			
Frequency			
Locations			On ISS: Inlets & outlets of systems; leak points? Maybe also external samples? (compare inside vs. outside), microbiota (airlocks.; venting, areas with spacesuit activities) Also: Think beyond ISS to Mars surface—e.g. consider dusts, biofilms, leakage areas from vehicles; external contamination of suits, seals, skin cells released? Viability/persistence outside? Alive/dead? Need technology for external sampling; Contaminant /dispersal? Shadowed areas? Radiation shielded? Short vs. long term? Mutations over time?

			(Hypothetical situations – use modelling) Induced/ habitable environments? Landing areas; leak/mutate/re-infect? Sheltered niches? (consider back contamination too) Microbial degassing?
Sample Processing	=		
Data Analysis			

Figure 5: KG 1A: Routine microbial monitoring of crewed environment (vehicles, spacecraft,) EXTERIOR

3.3 Findings on KG 1C: Measurements and Instruments for Mitigation

The three breakout groups addressed KG 1C (mitigation measures) after completing their deliberations on microbial monitoring for crew and crewed environments. The detailed findings on mitigation are included near the end of their respective breakout reports (Appendices E, F and G). The overall findings on mitigation are summarized below.

In general, the groups approached mitigation deliberations in different ways (i.e. making lists of comments and findings rather than specific recommendations for R&TD). While a consistent set of actionable steps to address mitigation was not generated, all groups emphasized the need to build upon the data and findings related to microbial monitoring of crew and crewed environments. Once progress has been made in understanding the types and levels of microbes, changes associated with crew members and the microbial data related to monitoring of crewed environments, then the combined data can serve as an updated baseline to evaluate what mitigation measures are needed to develop mission operations plans.

The primary focus was on forward contamination, en-route microbial questions (during the long duration in interplanetary space) and Mars surface activities. However, several comments were noted about longer-term mission architecture and operations concerns. These concerns included habitat shutdown/abandonment and/or quiescence and re-use; and crew habitat facilities as an unintended incubation facility (in hard to reach areas, or in the absence of crew). The groups also discussed monitoring efforts that focused on backward contamination of Earth by crew and samples. While these topics and concerns were outside the general topics assigned for this meeting, they will clearly need detailed analysis and consideration to satisfy KG 1D.

Consistent findings for KG 1C: Mitigation across all groups included the following:

- It is important to get data about the Mars environment from Mars 2020 and other upcoming missions, as well as from future meteorological stations on Mars. Such data will help in understanding the levels and nature of terrestrial-sourced microbiota (forward contamination) and the anticipated biocidal effects of the natural Martian environment (what will the Mars environment take care of naturally?).
- Microbiological assessment of Mars landing sites should be done prior to egress and human activity in order to acquire a baseline of the planetary surface conditions and data on the microbiological and organic content of Mars surface – which are relevant to later handling and testing of materials in labs—whether in situ or upon return to Earth.
- No human missions to the Mars surface should be allowed before the comprehensive organic/biological assessment of the surface is done.
- No additional microbial monitoring (beyond a baseline – like on ISS) should be required on the way to Mars. Attention should focus on maintaining crew health and environmental conditions, and would not include additional microbiological requirements imposed on the way to Mars.
- Sampling methods, instruments and modalities are needed to assess cleanliness, disinfection, sterilization methods, and approaches for monitoring microbes and cleanliness during human missions. Decontamination protocols; ingress-egress

interfaces, and other concerns (isolated nooks; unusual conditions for microbial growth, etc.) will need to be addressed to develop an acceptable baseline of bioburden at Mars, and effective methods for maintaining healthy conditions during the mission.

- It was suggested that consideration be given to employing an approach similar to Integrated Pest Management (IPM) which does not aim to get rid of all microbes or symbionts, but rather to minimize problematic or hazardous organisms. There is a need for strategic level vs. operational level thinking in considering control of microbes—an integrated concept with active mitigation—based on understanding human and crewed environments from a microbial perspective.
- There is a need to gather relevant information on various strategies used in terrestrial analogue situations. Some of examples of this are biocontainment/biosafety labs, hospital isolation areas, how to repair and maintain habitats, background monitoring of life support equipment, etc. (e.g., from the disease control agencies, hospitals, submarines, defense departments, etc.).
- Sampling will also be required to assess unknown threats.

4. Summary

To establish quantitative planetary protection requirements for human missions beyond Earth orbit and an informed partitioning of the Martian surface for safe and sustainable exploration and utilization of Mars requires new and more information about the following elements:

- A source term: the microbial community of crew and crewed vehicles and how it develops over time.
- A distribution term: how microbial contamination spreads and the threat posed to Mars.

Addressing both these terms through the identified knowledge gaps will allow a proper risk assessment that at the end will drive the engineering and operational mitigation measures that need to be implemented.

This meeting specifically addressed the source term. A previous meeting (Race et al., 2019) addressed specifically the distribution term and an upcoming meeting will address issues associated with engineering and operational mitigation measures.

In terms of results of this meeting, we have identified the following findings, recommendations and way-forward:

The **findings** are as follows:

1. ISS is the only existing, useful test-bed to obtain long-term baseline data and trends useful for preparing for human missions beyond Earth orbit.
2. Current routine microbial monitoring on ISS is limited in scope (number of crew and locations on ISS), depth (details of microbial populations) and frequency, of data collection.

Based on these findings, the following **recommendations** were formulated:

1. Systematic microbial monitoring of ISS crews and associated ISS environments should be done more frequently in order to obtain statistically relevant data over long periods of time and multiple crew complements.
2. Existing instruments and technologies can be used (ex. MinION, Oxford Nanopore with flight heritage on ISS) to monitor microbial levels for these purposes.
3. It is possible to build upon what is already in use on ISS—including associated processes, consumables and crew time-needs, which are already well understood.

The **way-forward** for addressing MHHM KGs in a timely manner includes a step-wise approach built on:

1. Data-mining activities of existing ISS and other terrestrial databases to establish starting points for ISS sampling (frequency, number of samples) and to write revised sampling and analysis procedures using the MinION equipment (or other flight heritage systems).
2. Integration of the data-mining information and MinION procedure outputs (above) to create an updated microbial monitoring plan for the ISS and crew that would address the MHHM KG.
3. Discussion of flight opportunities with ISS partners.

After gathering data on the ISS and prior to sending humans to Mars, initiate similar microbial monitoring beyond Earth's orbit to study the effects of the radiation environment (e.g. at Gateway) and conditions on a lifeless surface (e.g. the Moon), which the groups considered to be "must-have" additional information for interplanetary missions.

The product of these activities would provide necessary inputs to develop quantitative planetary protection requirements for human missions to Mars. At the same time, the results of these activities would inform the path to engineering of crewed systems and writing operational procedures that would mitigate contamination in the context of forward planetary protection.

References

Criswell, M.E. et al., (2005). *Planetary Protection Issues in the Human Exploration of Mars, Pingree Park Final Workshop Report.* NASA Ames Research Center. doi:NASA/CP-2005-213461

Hogan, J. et al., (2006). *Life Support and Habitation and Planetary Protection Workshop, Final Report.* NASA Ames Research Center. doi:NASA/TM-2006-213485

Johnson, J., Spry, J., Race, M., Conley, C., Siegel, B. (2016). *NASA's path to planetary protection requirements for human exploration missions: Update on recent progress.* Conference Report, 2016 IEEE Aerospace Conference.

Johnson, J., Spry, J., Siegel, B., Conley, C., Race, M.S. (2015). *Workshop Report: Planetary Protection KGs for Human Extraterrestrial Missions.* NASA. doi:DAA_TN36403

Kminek, G., Conley, C., Hipkin, V., Yano, H. (2017). COSPAR's Planetary Protection Policy. *Space Research Today* (200), 12.

Kminek, G., Rummel J., Race M.,. (2007). *Planetary Protection and Human System Research and Technology, Joint ESA-NASA Workshop Report.* Nordwijk: ESTEC. doi:WPP-276

Kminek, G. et al., (2016). *Report of the 1st COSPAR Workshop on Refining Planetary Protection Requiremmnts for Human Missions.* Retrieved from: https://sma.nasa.gov/docs/default-source/sma-disciplines-and-programs/planetary-protection/report-cospar-workshop-refining-planetary-protection-requirements-for-human-missions-final20180424.pdf?sfvrsn=a3fcfff8_2

NASA (1999). NPD 8020.7G: Biological Contamination Control for Outbound and Inbound Planetary Spacecraft: https://nodis3.gsfc.nasa.gov/displayDir.cfm?Internal_ID=N_PD_8020_007G_&page_name=main&search_term=8020.7

NASA (2014). NPI 8020.7 On Planetary Protection Requirements for Human Extraterrestrial Missions. Retrieved from https://nodis3.gsfc.nasa.gov/OPD_docs/NPI_8020_7.pdf

National Academies of Sciences, Engineering, and Medicine. (2018). Review and Assessment of Planetary Protection Policy Development Processes. Washington, DC: The National Academies Press. https://doi.org/10.17226/25172

National Research Council. (2002). Safe on Mars: Precursor Measurements Necessary to Support Human Operations on the Martian Surface. Washington, DC: The National Academies Press. https://doi.org/10.17226/10360

Race et al., (2019). *Report of the 2nd COSPAR Workshop on Refining Planetary Protection Requirements for Human Missions, and COSPAR Work Meeting on Developing Payload Requirements for Addressing Planetary Protection Gaps on Natural Transport of Contaminants on Mars*. Retrieved from: https://sma.nasa.gov/docs/default-source/sma-disciplines-and-programs/planetary-protection/cospar-2019-2nd-workshop-on-refining-planetary-protection-requirements-for-human-missions-and-work-meeting-on-developing-payload-requirements-for-addressing-planetary-protection-gaps-on-nat.pdf?sfvrsn=507ff8f8_8

Spry, J., Conley, C., Race, M. (2014). Advanced Planetary Protection Technologies for the Proposed Future Mission Set. *44th ICES*. Tuscon. doi:ICES-2014-254

Stern, A. et al., (2019) NASA Planetary Protection Independent Review Board (PPIRB) Report to NASA/SMD https://www.nasa.gov/sites/default/files/atoms/files/planetary_protection_board_report_20191018.pdf

U.S. Department of State. (1967). *Outer Space Treaty*. Retrieved from Treaty on Principles Governing the Activities of States in the Exploration and Use of Outer Space, Including the Moon and Other Celestial Bodies: https://2009-2017.state.gov/t/isn/5181.htm

Appendices

Appendix A: COSPAR Principles and Guidelines for Human Mission to Mars (2008)

BOX 1: COSPAR Planetary Protection Principles and Implementation Guidelines for Human Missions to Mars
http://cosparhq.cnes.fr/Scistr/PPPolicy(20-July-08).pdf

The intent of this planetary protection policy is the same whether a mission to Mars is conducted robotically or with human explorers. Accordingly, planetary protection goals should not be relaxed to accommodate a human mission to Mars. Rather, they become even more directly relevant to such missions—even if specific implementation requirements must differ. **General principles include**:

- Safeguarding the Earth from potential back contamination is the highest planetary protection priority in Mars exploration.
- The greater capability of human explorers can contribute to the astrobiological exploration of Mars only if human-associated contamination is controlled and understood.
- For a landed mission conducting surface operations, it will not be possible for all human associated processes and mission operations to be conducted within entirely closed systems.
- Crewmembers exploring Mars, or their support systems, will inevitably be exposed to Martian materials.

In accordance with these principles**, specific implementation guidelines** for human missions to Mars include:

- Human missions will carry microbial populations that will vary in both kind and quantity, and it will not be practicable to specify all aspects of an allowable microbial population or potential contaminants at launch. Once any baseline conditions for launch are established and met, continued monitoring and evaluation of microbes carried by human missions will be required to address both forward and backward contamination concerns.
- A quarantine capability for both the entire crew and for individual crewmembers shall be provided during and after the mission, in case potential contact with a Martian life-form occurs.
- A comprehensive planetary protection protocol for human missions should be developed that encompasses both forward and backward contamination concerns, and addresses the combined human and robotic aspects of the mission, including subsurface exploration, sample handling, and the return of the samples and crew to Earth.
- Neither robotic systems nor human activities should contaminate "Special Regions" on Mars, as defined by this COSPAR policy.
- Any uncharacterized Martian site should be evaluated by robotic precursors prior to crew access. Information may be obtained by either precursor robotic missions or a robotic component on a human mission.
- Any pristine samples or sampling components from any uncharacterized sites or Special Regions on Mars should be treated according to current planetary protection category V, restricted Earth return, with the proper handling and testing protocols.
- An onboard crewmember should be given primary responsibility for the implementation of planetary protection provisions affecting the crew during the mission.
- Planetary protection requirements for initial human missions should be based on a conservative approach consistent with a lack of knowledge of Martian environments and possible life, as well as the performance of human support systems in those environments. Planetary protection requirements for later missions should not be relaxed without scientific review, justification, and consensus.

Figure 6: COSPAR Planetary Protection Principles and Implementation Guidelines

Appendix B: Information from prior workshops and meetings:

1st COSPAR Workshop (2016)

Each Group
Analyzed & Ranked
Knowledge Gaps (KGs)
by
Time Priority
& Mission Criticality

TABLE X: All Splinter Group Findings	Priority/Criticality	
GROUP 1: Microbial & Human Health Monitoring	**TIME**	**MISSION**
1A. Microbial Monitoring of Environment	H	
1B. Microbial Monitoring of Humans	H	
1C. Mitigation of Microbial Growth in Spacecraft Systems	H	
1D. Operational Guidelines for PP and Crew Health	L	
GROUP 2: Technology & Operations for Contamination Control		
2A. Bioburden/Transport /Ops during Short v. Long Stays	M	M
2B. Microbial/Organic Releases from humans and support systems	H	H
2C. Protocols (Decontam/Verific/Monitor) to Remediate Releases	M	H
2D. Design of Quaratine Facilities/Methods -for different phases	L	L
2E. How do MarsEnv Conditions vary over time wrt growth of Earth mcirobes	L	H
2F. Res. needed to make ISRU & PP goals compatible	M	M
2G. "acceptable contam' of wastes left behind? Constraints on vented matls.	L	L
FORMER 2H. DELETED		
2 I. Approach to Achieve 'Break the Chain" Requirements?	L	L
2J. Global Distrib/Depth of subsurf. Ice-- and evidence of Extant life?	H	H
2K. Evolution of PP Reqmets/goals from robotic to Human Missions & zones'	H	M
GROUP 3: Natural Transport of Contamination on Mars	**Time/Mission**	
3A. Measurements/Models for Mars atm. transport of contaminants	H	
3B. Measurements/Models for subsurf. transport of contaminants	M	
3C. Effect of Biocidal Factors on surv./growth/adapt of microbes on Mars	H	
3D. Determine Acceptable Contam. Rates & Thresholds	H	
3E. Protection Mechanisms for organisms on Mars	M	
3F. Degradation of Landed Materials by martian envmt	M	
3G. Induced Env Conditiosn around Struture?	M	
3H. Sensitivity of non-culturable spp to biocidal factors	M	

Figure 7: 1st COSPAR Workshop (2016)

2nd COSPAR Workshop (2018)

Identify *Mission/ Location/ Test Opportunities* now through 2030's

Potential Opportunities Considered

- Ground based research
- ISS
- SLS – EM1 and EM2
- Cubesats
- Gateway
- Lunar landers
- Precursor missions to Mars
- Commercial missions
- International missions

- *Time Periods* Considered
  - Near Term (2018-19)
  - 2020-24
  - 2025-30
  - 2031 Onward

Data to Excel Files:

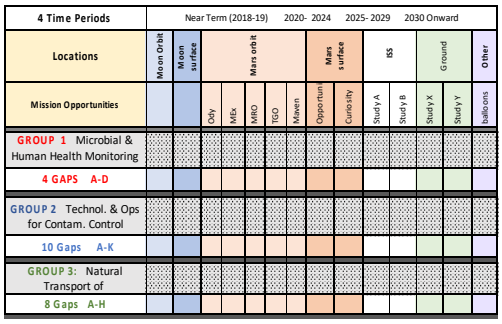

Figure 8: 2nd COSPAR Workshop (2018)

Mars Mission Opportunities 2001-2030+

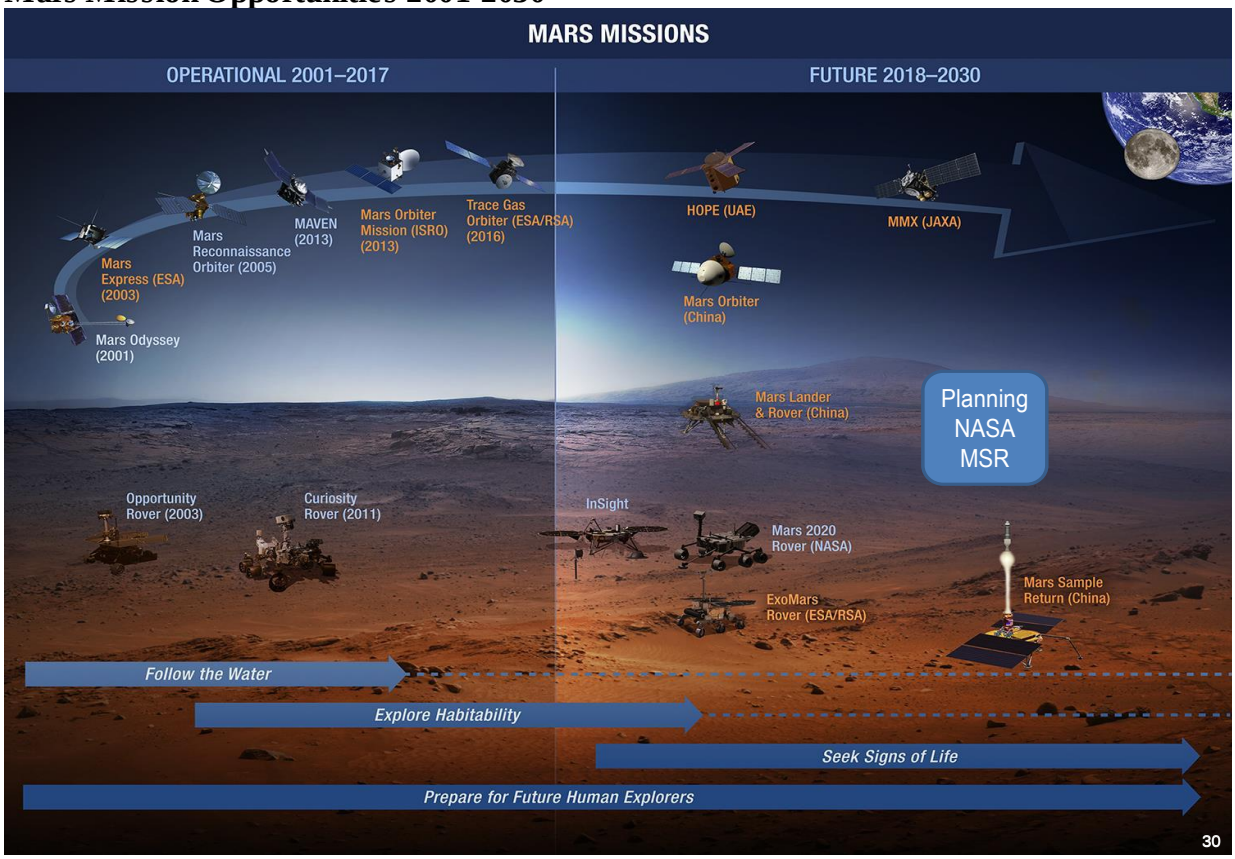

Figure 9: Mars Mission Opportunities 2001-2030+

Lunar Mission Opportunities 2018-2030

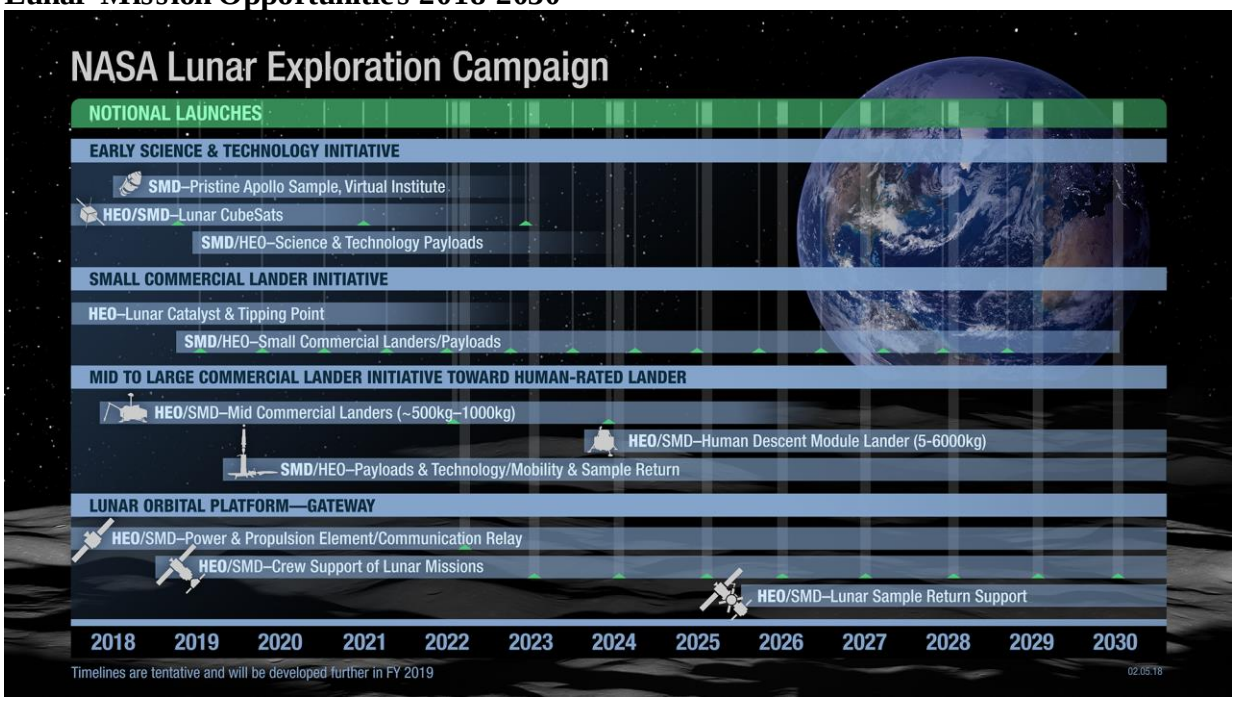

Figure 10: Lunar Mission Opportunities 2018-2030

Appendix C: Meeting Agenda

3rd* COSPAR Meeting to Address Planetary Protection Knowledge Gaps for Human Missions and Working Meeting on *Microbial and Human Health Monitoring
LPI, MAY 14-16, 2019

CONTEXT: This meeting is part of a multi-year series of workshops & associated meetings that collectively aim to refine COSPAR's current *qualitative* Planetary Protection (PP) policies for Human Missions to Mars and incrementally contribute to the development of detailed, *quantitative* PP requirements in the timeframe between now and the first crewed flight to the Martian surface. Already, this step-wise process has identified and prioritized knowledge gaps (KGs) in three study areas and developed a list of potential mission opportunities, locations and ground based research & test concepts that represent feed-forward prospects for gathering needed data.

GOAL: This 2019 COSPAR work meeting will build upon the findings from earlier workshops in the series. The overall agenda for this meeting has two parts - the first part (Day 1), is a reexamination of the state-of-the-art and refinement of the KG timeline. The second part (Days 2-3) will be more focused, with the objective to address KGs related to Microbial and Human Health Monitoring (MHHM).

The overall output of the meeting will be a document describing the necessary level of microbial monitoring of crew and crewed vehicles to close the MHHM KGs, together with a brief meeting report.

Venue/dates Lunar & Planetary Institute, Houston, TX; May 14-16, 2019

Chairs: Gerhard Kminek & Bette Siegel

> For those unable to attend the meeting in person, there will be a virtual viewing accommodations provided for Day 1 deliberations and presentations:
> **Teleconference Number: 888-889-6566 Teleconference Passcode: 8213219**
> **Meeting Number: 902 184 006 Meeting Passcode: MeetingMay14!**
> **Meeting Link:**
> ***https://nasaenterprise.webex.com/nasaenterprise/j.php?MTID=m07e11ba23d92910084b40e4394fa9c49***

Agenda – Day 1, May 14 - Plenary Information

9:00 – 9:30	Coffee and Registration (Foyer)
9:35 – 10:00	Welcome, Meeting Objectives and Introductions (Auditorium) - Bette Siegel (NASA HQ)
10:00 - 10:20	Review of COSPAR Planetary Protection Policy/Planetary Protection Panel Organization – Gerhard Kminek (Co-Chair: COSPAR Planetary Protection Panel/ESA)
10:20 – 10:30	Planetary Protection at JAXA – Kazuhisa Fujita (JAXA)
10:30 - 10:45	Planetary Protection at NASA – Lisa Pratt (NASA HQ)
10:45 - 11:00	Coffee Break (Foyer)
11:00 - 11:20	Why Mars – John Rummel (SETI Institute)
11:20-11:40	The Path to Development of COSPAR Planetary Protection Requirements for Crewed Missions Beyond Earth Orbit – J Andy Spry (SETI Institute)
11:40 - 11:50	Update on Strategic Knowledge Gaps for Human Missions – Bette Siegel (NASA HQ)
11:50 - 1:00	Lunch

1:00 - 2:00 Overview / Update on 2018 Meeting Findings

- Microbial and Human Health Monitoring – (Mark Ott (NASA JSC)/ David Pearce (Univ. Northumbria)) - 15 minutes
- Technology & Operations for Biol. Contam. Control – Mark Lupisella (NASA GSFC) - 20 minutes (include update on architecture changes since last year)
- Natural transport of biological contamination on Mars Manish Patel (Open University)/ - 25 minutes

2:00 - 2:30 Review of 2018 synthesis product – J Andy Spry (SETI Institute)

2:30 - 2:45 Coffee Break (Foyer)

2:45 - 5:25 Invited presentations from Agency and Industry leaders related to the state of the art and future mission opportunities in MHHM KG closure

- 2:45 JAXA ISS MHHM work to date – Kazuhisa Fujita (JAXA)
- 3:05 ESA ISS MHHM work to date – Jason Hatton, Guillaume Weerts (ESA)
- 3:25 NASA ISS MHHM work to date – Sarah Wallace (NASA JSC)
- 3:45 NASA Gateway Opportunity– Paul Niles (NASA JSC)
- 4:05 Mars 500 data – Petra Rettberg (DLR)
- 4:25 ISS sampling – Andrew Schuerger (University of Florida)
- 4:45 ESA ISS Monitoring – Christine Moissl (Med University Graz)
- 5:05 ISS Microbial Observatory – Kasthuri Venkateswaran (JPL)

5:25 – 5:35 Tour Logistics – Larry Toups/Kevin Watts (NASA JSC)

5:35 – 7:00 Reception

Agenda – Day 2-3, May 15-16

8:30 - 11:30 **JSC Tour - Tour "behind the scenes**" at NASA's JSC facilities
This tour is open to all Workshop attendees, however (for non-NASA badge-holders) visitor's names, affiliation, nationality, and (for foreign nationals) a copy of the visitor's passport photo page, must have been provided by the due date.

Agenda – Day 2-3 Working Sessions

COSPAR Working Meeting on Requirements for Addressing Planetary Protection Knowledge Gaps in <u>Microbial and Human Health Monitoring</u>

Scope: The second part of the meeting will focus on Microbial and Human Health Monitoring (MHHM). Using the KG's identified and prioritized during the 1st COSPAR Workshop Report on Refining Planetary Protection Requirements for Human Missions (2016), and the mission opportunities and locations identified in the 2nd COSPAR Workshop (2018). The intent is to describe the necessary level of microbial monitoring of crew and crewed vehicles (and the measurements and instrumentation needed to do that) for closing the MHHM KG's.

11:30 – 11:50 Instructions for MHHM Breakout Discussions; review of last year by Manish Patel (Open University), templates and tools by Margaret Race (SETI Institute), charge to the group by J Andy Spry (SETI Institute)

11:50 - 1:00 Lunch at LPI

1:00 – 4:30 Breakout group discussions
Measurement and instrumentation for operation monitoring of crew, with particular emphasis on gathering baseline data on ISS, Orion, Gateway and subsequent vehicles

3:15 - 3:30 Afternoon refreshments (at option of subgroups)

4:30 – 5:00 Breakout reports

5:00 Adjourn

6:30 Group Dinner (Frenchies)

Agenda – Day 3, May 16

8:10- 8:30 Future microbial monitoring of crew and spacecraft: experience and issues - Kate Rubins (NASA JSC) –Remote from Moscow

8:30 – 12:00 Plenary review then return to breakout groups – J Andy Spry (SETI Institute) and Bette Siegel (NASA HQ)

Measurement and instrumentation for operational monitoring of crewed environments, with particular emphasis on gathering baseline data on ISS, Orion, Gateway and subsequent vehicles
and
Measurement and instrumentation for Mitigation

10:15 – 10:30 Coffee break (at option of subgroups)

12:00 – 1:00 Lunch at LPI

1:00 - 2:30 Plenary consolidation of work products

2:30 Adjourn

APPENDIX D: Templates

Table D.1: Template for Monitoring of Crew

Routine microbial monitoring of crew

Information	Equipment	Consumables	Frequency	Locations	Sample processing	Data analysis
• Bacteria? Fungi? Viruses? • Taxonomic level • Culture/molecular • Associated physiological data	• On-board (incl. storage) • On-ground	• Non-liquid • Liquid	• Before, during, post flight • Fixed (daily, weekly, monthly,…) • Event driven • Fixed + event driven	• Surface (body) • Liquid (saliva, urine) • Solid (stool)	• On-board • On-ground	• On-board (incl. expected link budget) • On-ground

Figure 11: Template for Crew Monitoring

Table D.2 Template for Monitoring of Crewed Environments /Vehicles

Routine microbial monitoring of crewed vehicles

Information	Equipment	Consumables	Frequency	Locations	Sample processing	Data analysis
• Bacteria? Fungi? Viruses? • Taxonomic level • Culture/molecular • Air (active or passive), surfaces (dry or wet), filters	• On-board (incl. storage) • On-ground	• Non-liquid • Liquid	• Fixed (daily, weekly, monthly, …) • Event driven • Fixed + event driven	• All modules • One module • Cycle between modules	• On-board • On-ground	• On-board (incl. expected link budget) • On-ground

Figure 12: Template for Monitoring of Crewed Environments/Vehicles

APPENDIX E: Deliberations - Breakout Group 1

E.1.0 Breakout Group 1:

Chair: CRAIG KUNDROT Rapporteur: BOB COLLUM

Preliminary discussions: Before considering the specific measurements and payload instruments necessary for understanding the microbiome on human missions, Breakout Group 1 discussed a broad set of questions associated with the MHHM knowledge gap, including the following topics and questions:

- What do we need to understand about the human microbiome, and what does a "normal" human microbiome look like?
- What are the contamination risks of different microbes, and how are different microbes distributed across the body?
    - What is the potential for the human microbiome to survive outside of the human?
        - Nominally?
        - In an extreme environment?
    - How many sampling locations are needed to build a complete and accurate picture of the questions above?
- What can we accomplish in different environments—in space or using terrestrial data? (ISS, submarines, Antarctica, laboratory etc.)? Are there other study areas and analogs that could also be useful for research of relevance?
- What are the differences between a habitat environment and a space suit environment?

In addition, they identified a list of what they considered High-Level Assumptions:

- Data mining of existing studies can help answer many of these questions
    - Specifically the Human Microbiome Project
- Other Potential data sources include:
    - Analog environments both in space (currently ISS; and future deep space analogues) as well as on the Earth.
    - General Population (Human Microbiome Project)

After their general discussions above, Breakout Group 1 shifted to deliberation of what specific measurements and instruments are needed for monitoring of the microbes associated with crew health vs. the spacecraft environment. Using the template as a guideline, they considered questions about monitoring needs (information, equipment, consumables, frequency, locations, sample processing, and data analysis) and assessed the types of monitoring and information needed, and how these relate to future equipment and sample collection. Their summary findings are as follows:

E1.1 Breakout Group 1: KG 1B Crew Health

Before addressing the template topics for Crew Health, they compiled the following introductory notes and comments:

Monitoring the human microbiome and identifying the microbes therein makes it easier to

- Develop contamination mitigation techniques
- Avoid false positives in life detection (clearly important)—but will need more information , including

        - Understanding of the potential contaminants, and
        - Knowledge of how biocidal effects may impact the composition of the human microbiome
    - Understand the potential of different microbes for microbial proliferation and organic contamination,
    - Determine how to Identify novel microbes from the Martian environment, and
    - Monitor the effects of a space environment (adaptation, resistances, etc.) on the human the microbiome

They then used the template guidelines for further deliberations on:

Information Needed:

- Identification of the constituents of the human microbiome
    - Need to consider Bacteria, Fungi, Viruses, Archaea, Eukaryotes
        - What is the appropriate level of detail?
            - How to deal with Spore Formers + Radiation resistant (c.f. Type C organisms per the Space Studies Board "Preventing the Forward Contamination of Europa" Report
    - How do we account for microbes associated with plants and materials that the crew bring along?
- Quantification of the constituents of the microbiome to establish
    - What is the bioburden?
- Assessment of the risk potential for the constituent microbiome.
- Molecular analysis is preferred, but
    - Cultures should be maintained as a back-up and performed on the ground
- Metadata for sample collection – to be drawn from the Human Microbiome Project
    - Include data on Medications taken
    - Other considerations/ unusual factors?
- Physiological data including:
    - Continue using data already being taken on astronauts
    - Meta-analysis (of both astronaut data and general population data)
    - Sample collection during off-nominal events (fevers etc.,)
    - Data recording of Astronaut symptoms
    - Also noted that additional desirable data (not currently taken) may be identified later

Equipment/Sample Collection & Processing

- Improve sample collection techniques to better mimic techniques on the ground
    - How do we isolate the microbiome in collected samples on orbit?
- Sample collection and processing techniques for on board samples need to be optimized and improved before they are analyzed in MinION
- Samples should be divided to be processed both on board and on the ground as validation
    - On Board equipment and techniques
        - MinION aboard the ISS – available now
    - On Ground equipment and techniques
        - Microfluidics based technology
        - Automated sample processing

Consumables

- Use both Non-liquid & Liquid, with the notation that
    - Depending on the shelf life, non-liquid is preferred

        - Good because their smaller

Frequency

- Nominal sampling frequencies should be based on Human Microbiome Project
- In addition, include Event Driven Considerations for Sampling
    - Some locations may be sampled at higher frequencies during critical events (e.g. swabbing a rash during the rash)
        - Locations, frequencies, and revisit rate will depend on the specific event(s)

Locations for Crew Microbiome Monitoring

- In Situ/On Board sampling of varied body locations:
    - Skin
    - Nose
    - Mouth
    - Hair
    - Urine
    - Blood
    - Fecal, and
    - Relevant Event Driven Locations (e.g. tears, ears, throat)

Data Analysis

- Current on-board analysis is enough to meet information needs for providing a generic phylogenetic analysis
- Shotgun meta-genomic analysis and Species level analysis will require more powerful capabilities that can only be carried out on the ground
    - Machine Learning is a potential avenue for improving this capability (which they categorized as 'A Nice to Have')

E1.2 Breakout Group 1: KG 1A - Routine Microbial Monitoring of Crewed Environment

The group then began discussions about **microbial monitoring of the spacecraft and vehicles** as follows:

- What is the potential for fungus to produce deleterious effects (corrosion, biofilms, etc.) within the crew vehicle?
    - How does the development of fungal sites influence the proliferation of other taxa?
- How uniform is the microbiome across various spacecraft modules?
    - How does that change over time?
- What is the difference in the distribution of airborne microbes between the ISS, the Earth, and Mars?
- How do we approximate the environment on the surface of Mars?
    - How does it (the Martian Surface) change the distribution of (terrestrial?) microbes
- Are vehicle air filter samples representative of the entire vehicle or a smaller subset?
    - How do we validate them?

They also considered on notion of monitoring **Inside *vs*. Outside of vehicles** to characterize and understand their respective microbiomes, to make it easier to:

- Develop strategies for understanding the biocidal rates in the crew environment
- Understand the long term changes/adaptations of the microbes
- Avoid false positives in life detection
  - What are the potential contaminants?
- Understand bioload likely to develop in pre-emplaced robotic cargo on way to Mars
- Understand biologically caused deterioration (corrosion, biofilms, etc.)
- Understand what is likely to be vented
- Understand the dispersion and transfer mechanisms of microbes
  - **Including How far do they propagate?**

Using the template categories as guidelines for further deliberations, they filled in details as follows about crewed environments**:**

Information Needed:

- Identification of the constituents of the crew vehicle microbiome
  - Bacteria, Fungi, Viruses, Archaea, Eukaryotes
    - What is the appropriate level of detail?
      - Spore Formers + Radiation resistant (Type C – via Space Studies Board)
  - Do we need to distinguish between the sources of the microbes (human, plants, cargo)?
- Quantification of the constituents of the microbiome
  - What is the bioburden?
- Assessment of the risk potential (organic contamination, proliferation) for the constituents
- Relevant Metadata
  - Temperature
  - Humidity
  - Materials/Surface type
  - Atmospheric composition
  - Visible sites
  - Others?
- Molecular analysis is preferred
  - Cultures should be maintained as a back-up and performed on the ground
- Assessments before sample collection occurs
  - Active Air
  - UV assessment of surfaces
  - Filter
  - Biofilms
  -

Equipment/Sample Collection & Processing Needs

- On Board
  - Nonspecific biological monitoring needs to be the first step to limit the number of nonviable samples collected
    - LALR approaches
    - Biovigilance
  - Automated DNA extraction with multiple samples
    - Library prep before MinION
  - Thermophoretic Sampler (Augi's recommendation) – not available now
    - For airborne samples - A
  - MinION

        - Limited by required sample size

Consumables- No notes/details provided

Frequency

- Frequency could be based on terrestrial sampling from the Human Microbiome Project
    - May not be as relevant in a space environment
- The protocol should be on a set schedule that recurs regularly
    - Most likely weekly
- Unplanned events may drive more frequent sampling (e.g. visible growths, leaks, smells, anomalies in analysis)
- EVAs will require their own sampling routines
    - For suits, airlocks, vehicle exteriors
    - Frequency will likely be driven by the results of initial sampling

Locations (for environmental microbe monitoring)

- Event Driven locations
    - Incoming cargo
    - Spacecraft Exterior
        - Vent Sites
        - High traffic areas
    - Suits
        - Before and after EVAs
    - Airlock
- Regularly sampled locations should include:
    - Filters
    - Automated air samples near High traffic areas
    - Bathroom
    - Food
    - Water
    - Waste Stream
    - Sleeping Quarters
    - Areas Near biological experiments, including those with
        - Rodents
            - Helps set a baseline given that we understand the relative cleanliness and genome of the rodents pre-flight
        - Plants
- "Out of the way places"
    - Likely need to be sampled less frequently
    - E.g. Behind racks

Data Analysis

- On the ground
    - Archived biological samples from waste and disposables
- Metadata
    - Particle count

E1.3 Breakout Group 1: KG 1C Measurements & Instrumentation for Mitigation:

Finally, Breakout Group 1 considered questions related to possible Mitigation measures, by focusing on the following concerns:

- What are the *natural Biocidal factors* on the in-flight and on the surface of Mars?
    - Are they dying or just hibernating?
- What is the probability of *inducing a special region* whether via heat or a different mechanism?
    - Is it a problem because the site will be both isolated physically and temporally?
        - If it reactivates something terrestrial?
        - If it reactivates something native?
        - What is the likelihood for adaptation over the limited timescales being discussed?
- What are *temperature changes at the human outpost*?
    - Under the habitat?
    - In a rodwell?
    - During landing or take-off?
- As mitigation, Can we encourage the growth of Type A or B microbes to discourage the growth of Type C microbes in areas where microbial growth is unavoidable (e.g. food areas)?
    - What can we do to the environment?
    - To the design of the habitat?
    - Are there interventions we can carry out to encourage growth of favorable microbes
- What is the appropriate quarantine duration for the crew coming back?
    - Is the time they would spend on the MTV for return sufficient?
- What is the feasibility of detecting changes in the human genome caused by exposure to things in the Martian environment?

Discussions then shifted to considering what **Engineering and Operational Strategies** could address **mitigation** concerns, noting

- Human explorers will continue to contaminate the surface and local area regardless of how well sterilized things are going in
- There is need to have both nominal and off-nominal mitigation strategies
    - Nominal:
        - Venting
        - EVAs
    - Off-nominal:
        - Induced special regions
        - Habitat failure
- Different **tiers of mitigation strategies** were then identified (in order of approximate burden on the mission):
    - Design
        - A suit with fewer folds
        - A suit that can be cleaned
    - Operational
        - Limiting exposure through activity management
    - Low consumable strategies
        - UV lights
    - ISRU consumable strategies
        - Perchlorates

    - High consumable strategies
        - Disinfectants
- Possible use of a microbial "Bear Fence"[1]
    - Potentially a high energy perimeter to keep human microbes within a designated area
- Need to Distinguish the difference between clean and sterile
    - Where and when is one necessary vs the other?
- In addition, there are a number of pre-flight and in-flight mitigation strategies that have been developed for robotic Mars missions. They should be assessed to determine their adaptability for future human missions
- There are also a number of mitigation strategies that can be taken from existing Biosafety Lab (BSL) procedures on Earth— They should be assessed to determine their adaptability for various aspects of future human missions
- Also need to assess utility of different mitigation strategies for human missions:
    - Are the procedures adaptable or too onerous?
        - On astronaut time?
        - On the composition of the materials?
        - On consumables?
        - On the design of the habitat?

E1.4 Additional Recommendations of Breakout Group 1:

In addition to the specific recommendations on microbial monitoring of crew and spacecraft environments—and for mitigation measurements and instruments-- the group added the following concluding recommendations (note: 1. & 2. were reinforcements of the 1st (2016) and 2nd (2018) COSPAR meeting findings)

1. We need next generation meteorological stations on Mars
2. We need studies to assess the biocidal effects of the Martian environment
    a. More specifically:
        i. Oxidation
3. Need an assessment of relevant existing terrestrial strategies (for microbial monitoring) and their applicability to Mars missions. In particular, suggest look at
    a. BSL labs—biosafety; biosecurity
    b. Relevant Military procedures , and
4. Need an assessment of what constitutes a healthy microbiome (in a home, in an office, etc.)

[1] A microbial 'bear fence' was used metaphorically – like some type of barrier to separate or protect humans from a recognized hazard (in this case microbes).

APPENDIX F: Deliberations - Breakout Group 2

F.1.0 Breakout Group 2:

Rapporteur: Ben Clark Scribe/Co-Chair: Christine Moissl-Eichinger

(Participants: see Appendix D)

Breakout Group 2 first generated a list of comments, questions and starting information, before filling out the templates and mitigation information (see Tables 3.1, 3.2, 3.3 and 3.4) check numbers for tables..:

- Considering the differences between Open vs. closed testing systems (completely confined environment), -- they noted that perhaps a new testbed system is needed? Is it possible before we fly?
- What are Mutation rates for microbes and do we need to worry about them?
- How do microbes behave under flight conditions? There is only limited data on phenotypic variations (& adaptations etc.) in flight environments. Looking at single species of microbe does not make sense. How to study microbes in general? They represent a complex system, together with host.
- Is cleaning contradictory? If microbial balance is important, how to maintain it?
- Crew might possibly exchange microbiome during flight? What are the implication
- We need to know the materials etc. that might be eaten by microbes—including info on fungi, slime molds, growth etc.
- Another possible important KG: What is the material and microbial interaction?
- What concerns are associated with coming home again --might it be the major issue?

Recognizing the importance of fungi, they asserted the need to improve fungi monitoring (current focus predominantly on Bacteria)—and compiled the following considerations:

- Why are fungi and their interactions with humans still a question? Fungi are complicated… not easy to assess
- Need to Test engineering components, architecture, physics… and Antifungal coatings?
- Need to improve fungi characterization, archaea, viruses.
- To monitor fungi, could shotgun sequencing target them?
- Note: Food is not sterilized before sent up.
- Do we need to distinguish environmental and human monitoring? (perhaps they are Overlapping?… ex., fungi growing on walls could produce harmful compounds)
- How do planetary protection concerns overlap with human health?

- How do we address KGs in the area of microbial monitoring
    - Monitoring of Bacteria, AND Archaea, and fungi and viruses
    - Monitoring of microbial interaction with materials
    - Monitoring of the human microbiome, in parallel to minim. Medical diagnostics
- What is going to challenge the human immune system, when everything is clean?
- Assessment of Function preferred over focus on microbial composition, --more informative!
- Should we focus on harmful microorganisms to Mars? Do we Characterize contamination of interest… or all contamination?

- They noted that sequencing is not done on a high frequency, but medical screening, event-based sequencing is.

They also included comments about **frequency** of monitoring, and indicated that there is need to establish a test series (e.g., once a day, once a week, once a month) to optimize data acquisition. Specific items noted for consideration:

- Astronauts: Allergies, skin infections, urinary tract infections, respiratory tract infections; 1-2% of all people
- For metadata: do not rely on standard medical checks, but lot of metadata needed
- Astronaut should be his/her own control (the focus is on an *individual*—not populations)
- Crew time?
- KG: Need understand physiology of microorganisms, and determine if able to grow under Martian conditions?

Finally, Breakout Group 2 addressed questions of when we will know enough about Crew Microbiology. They noted that:

- **Data Mining is Mandatory** to close the KG about crew
- **Data Gathering post ISS** (Orion, Gateway, crew on Moon–) **is likewise mandatory**—and should be done along with studies in analogue and test facilities
- The **natural stopping point** for microbial data collection about Crew can only be determined **after having started the data analysis** , which should be begin soon—and continue .
- S**hort term ground based activities can be used to address** the diverse data needs for **system & operational uses**. -–e.g., nanopore, sample processing, data collection (?), testing of biocompatible materials and engineering solutions etc.
- Short term **ground based activities** are needed to get a system operational (upgrade, delta qualification/ etc.) And considerations should include such things as nanopore, sample processing, data, testing of biocompatible materials and engineering solutions etc.

Breakout Group 2 assembled their findings in template tables for Crew (1B), Crewed Environment (1A) (inside and outside), and included text suggestions on Mitigation. See Tables F1.1-F1.4

Table F1.1 Breakout Group 2 - Findings on KG 1B: Monitoring of Crew

Information	Equipment	Consumables	Frequency	Locations	Sample processing	Data analysis
• Bacteria? Fungi (important!)? Viruses? Archaea • Taxonomic level: as deep as possible, constraints from database and technology. Additional: shotgun information (non-targeted) • *Culture (keep the option to grow) or instead: viability test (KG to improve those assays)*/molecular: Minion, solve: processing samples and DATA (machine learning, autonomy, req) • Associated human physiological data maybe even more important! Correlate with microbial information. KG which human parameters (medical, psychological) are important? Need to be requested	• On-board (incl. storage): very small for Mars (limitations, but selection), different requirements for data gathering(ISS); same equipment for life detection? • On-ground (need analogue research, such as Concordia, etc., Submarines!)	• Non-liquid (swab, tubes etc.) • Liquid (extraction buffers - lyophilized, water-UV: DNA-free)	• Before, during, post flight • Fixed (daily, weekly, monthly,…) • Event driven • Fixed + event driven • Test run onboard/analogue is necessary to identify frequency, replications are necessary (triplicates?), data limited for confined systems • Consider recycling of consumables	• Surface (body) (forward contamination) • Liquid (saliva, urine) • Solid (stool) • (test all, down select for long-term monitoring)	• On-board KG: processing samples and DATA (machine learning, req) • On-ground • Bioarchiving of samples and parallel analysis on ground	• On-board (incl. expected link budget) autonomous as much as possible • On-ground

Figure 13: Breakout Group 2 - Findings on KG 1B: MONITORING OF CREW

Table F1.2: Breakout GROUP 2-- Findings on KG 1A Routine Microbial Monitoring of Crewed Vehicles—Interior

Information	Equipment	Consumables	Frequency	Locations	Sample processing	Data analysis
• Bacteria? Fungi (important!)? Viruses? Archaea • Taxonomic level • Culture/molecular (both needed initially) • Air (active or passive), surfaces (dry or wet), filters (available? dust on filter), water/ liquid systems (how to sample and process) (sensitivity (!) is an issue, biomass to low) • Particulates counting in background, amount of microbes, steady-state situation. Alert-level, what action is necessary? Real-time monitoring needed • Cultivation still necessary and comparisons with sequencing (*Bacillus* e.g. might not be well detected by sequencing), correlation of both techniques • Include additional medical checks for return flight • How to address the unknowns? A lot of signatures unknown… KG. Data to define the criteria for decision	• On-board (incl. storage) • On-ground • Disturbance events to study (cargo, crew exchange, etc.) to test flexibility of the system	• Non-liquid • Liquid	• Fixed (daily, weekly, monthly, …) • Event driven • Fixed + event driven (confirmation after cleaning action) • Increase information, as much as possible • Again ground analogs for determ. of frequency • Gateway monitoring, as connected to Mars trip • Check with information from pharmaceutical industry • Target has to be established, to be able to monitor it, and for subsequent action	• All modules • One module • Cycle between modules	• On-board • Data analysis and interpretation (how severe? Define what is normal what not) Machine learning, gives information, but does not decide • Bioarchiving, making sure 'waste' is exploited • On-ground	• On-board (incl. expected link budget) • On-ground

Figure 14: Breakout Group 2—Findings on KG 1A Routine Microbial Monitoring of Crewed Vehicles—Interior

Table F1.3 Breakout Group 2—Findings on KG 1A: Routine Microbial Monitoring of Crewed Vehicles - Exterior

Information	Equipment	Consumables	Frequency	Locations	Sample processing	Data analysis
• Bacteria? Fungi? Viruses? • Taxonomic level • Culture/molecular • Air (active or passive), surfaces (dry or wet), filters; witness plates, check for range of human contamination outside • Determine the (microbial) leaks (level, type), ground simulation might be needed (already on ISS! Suit sampling (inside, outside), exterior ISS sampling…): KG • Tests on moon? • New pp requirements are needed for human missions (final KG to be filled)	• On-board (incl. storage) • On-ground • Witness plates • Detection of human contamination outside (ongoing research at Concordia) • Rovers sample outside for contamination, which distance? • Analyze outer surface of spacecraft	• Non-liquid • Liquid	• Fixed (daily, weekly, monthly, …) • Event driven • Fixed + event driven	• All modules • One module • Cycle between modules	• On-board • On-ground	• On-board (incl. expected link budget) • On-ground

Figure 15: Breakout Group 2—Findings on KG 1A: Routine Microbial Monitoring of Crewed Vehicles - Exterior

Breakout Group 2—KG 1C—Comments on Mitigation

Mitigation

- Sampling necessary to assess cleanliness, modalities stil tbd
- Cleaning/disinfection needed but research needed for process for manned missions
- Sampling required to assess unknown threat
- No additional microbiological requirements on the way to Mars for crew health (on top of ISS requirement if baseline acquired on the ISS) Monitoring required for baseline
- Landing site microbiological assessment prior to egress and human activity on the surface (planetary baseline) / sample return
- Evaluation microbiological /organic content of Mars Sample Return e.g. Mars 2020
- No human mission to Mars surface before this organic/biological assessment is done

Figure 16: Breakout Group 2—KG 1C—Comments on Mitigation

APPENDIX G: Deliberations - Breakout Group 3

Chair/Rapporteur: David Pearce; **Co-Chair/Scribe**: Sarah Wallace-Castro

G1.1 BREAKOUT GROUP 3 KG 1B: Routine Microbial Monitoring of CREW

The group compiled a recording of their collective deliberations on varied topics related to crew microbiology: They began by noting "We don't know what we don't know." There remain many unknowns, some of which we may be unaware of… so it is important to get a comprehensive baseline**.** Moreover, in order to consider long term effects, we need to know what's 'normal' – and consider what is needed to set a baseline:

"Important" topics are highlighted in text boxes

Considerations:

Need to focus on Simple characterization tools. Indicator organisms. Is the population really changing? Ratios. Baseline establishment. Routine monitoring.
Try to consolidate redundant studies and repetitive actions!!!!

Need routine monitoring with new techniques and do it on board – even if there is no sample return. Questions included-- How often to sample crew??? What is Unknown?

NOTE: there was **disagreement** on the use of indicator organisms. Suggested there is no reason to down select—rather focus on data analysis.

Analog environments. More controlled environment. Combination of in situ analysis, but also collect and have samples for offline analysis. To be able to go back to those samples.
WHAT ABOUT WHAT WE HAVE? Need examine Archived samples.

Microbiome changes ALL the time. There is not a baseline in a normal human's life. You can monitor, but patterns are going to be difficult to determine. We can't do that now here on Earth. What are we looking for in all this microbiome data?

Collecting as many resolution points as you can on a regular time frame is a meaningful experiment – there are probably ways of doing it. Need to identify patterns and variability within these large datasets. Create bounds.

Overall: We need to Get to a point where we can find range that is "normal" and "abnormal"—and how they relate to other physiological responses.

Microbiome analysis alone is not sufficient. Need orthogonal data for confirmation. Not worried about populations (humans), rather, we're worried about *a few individuals.*

Need Front-end and back-end study to establish baselines for THOSE specific individuals and their individual microbiomes.

Not feasible to do entirely on ISS. So use a ground analog as well

In some areas, we do know what we don't know!

Much focus is on 'business as usual ops' (nominal)

– But what about off nominal ops? Radiation. All information is based on ISS data, which is a well-protected environment. What about crew in higher radiation? Need more time series data?

What data do we need from EVA? Space / power limited / leaks!

Need more regular collection of data. Currently, a guessing game with limited data points. More swabs from craft and crew.

Data analysis. How do we know when 'a bad thing is starting to happen (to crew environments)?

Need to characterize the microbiome and when is the difference meaningful.

What is going on inside – what is getting outside??

Need a lot more sampling around the ISS outside at points that are representative of airlocks, venting, and spacesuit activities. What is being released on Mars? ISS can help us with what is happening at those leak points. The known leak points and fully characterize there. **Develop the baseline.** Leaking – get a grip on that.

MinION – low power, low mass, very fast, usable in situ. Collecting many data well within reach. Mutations?? Sequencer should have the power to do that.

1. We don't know what the baseline is.
2. Frequency of current monitoring is inadequate.
   a. Once a month, on board analysis
   b. Shorter timeframe with Earth-based system (analog) to get baseline and assess risk
   c. Talk to the Navy (about closed systems and life support)
3. Specific methodologies… TBD

What is common collection of species vs what is changing? Correlate change with risk to astronaut health. What does the data mean (microbiome changes)?

How is risk determined?

Standard microbiome and standard microbiome fluctuations…but also looking for acquiring unknown organism that you could not detect, but detected it through changes in the crew microbiome.

Need Study Normal microbiome and their adaptations from a crew health perspective—also with a high background radiation.

Human Health Monitoring

- Monitor human health over microbiome to determine alien infection.
- Lagging indicator vs a leading indicator (human health)
- Immunological response monitored
- Transcriptomics – biomarker tracking in the crew

Anomalies --on the equipment and surfaces

Scout the system looking for anomalies

Automated systems doing the scouting not crew

Consider Techniques for big data analytics and AI / machine learning Suggest an analog – recommend a microbiome study in an analog

Animal colony analogs?

HERA – short term analysis?

Exposure to extraterrestrial material in Gateway.

Suggestions for getting the baseline…

Use the ISS as long as long as it is there.

Do the analysis on existing data sets – then write the experiment to be done on the ISS.

Propose timeframe…

Start as soon as possible with regular measurements of the crew

Use multiple techniques / methods

Summary Recommendations re CREW Monitoring: KG 1B

- Metadata analysis of all previously collected data. use this analysis to:
- Design and implement a ground-based analog study to determine the microbiome baseline (including fluctuations and anomalies)
- Immunological monitoring could be key data from the analog
- Many feel increased sampling should be started immediately and run concurrently with our suggested ground study

G1.2 Breakout Group 3: KG 1A: Monitoring of crew vehicle/ environment

Group 3 began their deliberations about KG 1A by compiling comments and questions related to crew vehicles and their environments:

Information --Data exists outside of confined environments and we should look there first before determining frequency.

FREQUENCY --Quarterly monitoring is inadequate. Longest once a month and shortest practical time needs to be identified (weekly / biweekly)…

Is one month too long: cry from the group = YES!

Clarify = environment vs crewmember sampling @ once/month?

Crew VS environmental sampling?

Monitoring right before crew / cargo arrival and right after crew / cargo departure

More frequent and event related sampling.

Min 7 day Max 30 window for sampling

Experiment to establish a baseline of how frequently to sample to capture the variability.

LOCATIONS-- Terrestrial metadata analysis to back up our recommendation!
Richest biodiversity is near the galley and/or WHC (opinion from individual)
Human sampling = comprehensive / gut / etc.?
Bring in folks to educate us on where on the human would be in the sample locations!
Inlets and outlets of the system are best (opinion from one individual)

Can Crew clothes be used for sample analysis?

What discarded items could be used to gain this microbial insight?

- Clothes
- Towels
- Wipes
- And so on…

For comparative data, the suggestion was made to analogue data from the Navy! Would ISS be any different? Can we just use that?

Need to **screen for virulence genes** within the crew microbiome (and possibly plants)…

Back to risk assessments = what will it all mean? How do we make risk assessments? Should risk assessment experts and engineers be involved? YES!

WE NEED THE BASELINE and the definition of WHAT IS A PERTURBATION TO THE BASELINE as well as tolerance level to risk and to changes

Regarding Monitoring of Spacecraft Environments:

Increase sampling within **the Standard Measures project.**
Some folks really want external samples for forward contamination and are very passionate about this.

Referring to earlier comments, they noted-- "**What's going on inside – what is getting outside??** A lot more sampling around the ISS outside at points that are representative of airlocks, venting, spacesuit activities. What is being released on Mars? ISS can help us with what is happening at those leak points. Examine the known leak points and fully characterize there."

We are looking at all organisms at the highest taxonomic resolution we can get! We are moving toward molecular data.

NASA's archived isolates should be in a repository for all researchers.

It's the molecular data that is needed to support the KGs.

Cultures are still desired for analysis, so we should keep culturing—and can use cultures to close other KGs, but we don't need them as much as the molecular to close the KGs.

Include multiple techniques:

- MinION (up front sample prep)
- PhyloChips (what are the probes?)
- Build a MALDI-TOF (limited databases)
- Culture (how much?)
- Fluorescence data

We should include all of these methods in the ground-based study(ies)

What is the consumable mass (for MinION)?

- 6 crew
- 4 sample sites/crewmember
- 1 sample a week for 6 months

Considerations for On-board **sampling**-- some analysis on-board, and some sample return.

Metadata analysis should be used to create software package to provide some level of risk assessment based on the data.

Need to consider Plants and non-human microbes as well --should be a separate workshop.

What other miniaturized hardware is out there?

- Particle counter airborne system: NAD/NADP?? They aren't super useful and not as good as they would have thought it would have been.
- Bio-warfare agent sampling systems (maybe compare sampling methods etc. with Department of Defense and agencies doing work with biosafety/biosecurity/ and bioagent sampling)?

In terms of **use of the ISS,** the group compiled a list of **additional questions related to crew vehicles and environments:**

- Ask Kate what we should be sampling
- Again, send us your trash! These items could prove very useful for microbial analysis.
- Condensation on surfaces in the ISS…analyze cold spots, as water = growth.

- Can we use Robotic (autonomous?) sampling (e.g., swabbing) or a Roomba (vacuum cleaner-type approach) to sample larger areas?
- Big question: Is the current sampling scheme sufficient? Need to determine if we are we under-sampling *or* over-sampling?
- Where do wet towels and clothes go to dry?
- Sample sharing could be utilized (the operational, culture-based samples routinely collected).

- What if we changed the cleaning processes of hardware/cargo/vehicles? Should this be assessed? How?
- Public health – looking for early indicators of problems-- HEPA filters could be used as an early warning system prior to symptoms

Moving Beyond ISS (thinking about the Mars Surface and other considerations)

- Dust (external on surfaces)
- Priority on sampling the vehicle outside should be logical points of escapes from microbiota from inside (vents, airlocks, etc.) where leakage is likely to occur.
- Having a portable sampling device that will collect the vent things and sample regularly.
- Understand external contamination on spacesuits (fabrics, seals, etc.) large quantities of human skin cells have the potential to be released
- We need to know what comes out, but ISS might not be the right model, as the environment on the Moon and Mars is very different and the data are not necessarily extractable.
- Viability – Do we care about what's viable outside of the ISS? Do we want to know if the stuff *(vented materials) we* are venting is alive or dead? What Technology to develop in the area of viable vs nonviable with a molecular approach?
- How do samples migrate? Shadowed areas and every place an EVA crew touches should be sampled.
- # of spores per area – not very scientific, do we need to define a better approach?
- Data have to be extrapolated to evaluate a hypothetical/ real scenario with release on surfaces (using ISS or ground-analogs) and modeled for Mars. (consider both inside /outside)
- Understand microbial dispersal.
- Long terms persistence of cells/spores with dispersal that they end up in a niche in which proliferation could occur.
- The systems that we send and land on Mars will induce habitable environments. Landing system should be addressed sooner rather than later.

- **Don't lose sight of back contamination issues**—need to keep population of Earth safe from something on Mars. (Still need to address comprehensively)
- Microbial viability in biofilms and levels of persistence need to be understood.
- Use exposure facilities on ISS to work towards understanding these questions as well as active processes *within* cells.
- Surface of Mars is incredibly harsh, BUT we don't know if there might be habitable niches on Mars—and where?
- 1 – 2 meters depth provides protection and a habitable zone in terms of radiation protection – we know nothing about this depth.
- What if the Earth microbes mutate due to the creation of habitable zones and then infect the crew? What if, indeed.
- Monitor the degassing of microbes into the presumably sterile environment of Mars

G1.3 Breakout Group 3—KG 1C: Mitigation Strategies

Cleaning: How clean is clean enough? All venues for thought. All the interfaces need to be managed.
"Sterile Rover Zone" (is it needed?)

Decontamination protocols? What equipment can go with us? Are there things you can do within an airlock interface to reduce microbial load?

General cleaning vs sterilization processes. 110 C+ for robotic bake out.
Interior of spacecraft – if you over clean are you going to create superbugs? What is the balance? Super sterile is generally not conducive to a healthy environment.

Will the system be engineered to allow a high degree of bioburden reduction?
Use prelaunch quarantine as a baseline for bioburden?

Forward contamination is a philosophical question – so what is acceptable if you can't mitigate the risk to zero (see concept of 'Contaminatometer - Appendix I)?

Strategic level vs operational level thinking…
Surfaces that inhibit bioburden levels / growth (antimicrobials, functionalized surfaces…)
Use Cold atmospheric plasma?

Combo of engineering and operational levels of control to minimize exposure. This, combined with cleaning and sterilization procedures, might yield the desired outcome

Consider analogue issues in **Integrated Pest Management** (IPM). Use everything at your disposal to combat known problems. Keep known threats at 0 and have the system colonized by what is very difficult to control and sterilize (human microbiome).

See: IPM: https://www.epa.gov/managing-pests-schools/introduction-integrated-pest-management

In practice, IPM uses a multi-tiered approach—identify and monitor; set action thresholds (ex. nuisance, health hazards; environ/economic concern etc.); prevent/remove (treat); and control over time.

Long Duration considerations: The first few years then should be pretty clean, but by the end of the spacecraft lifetime, things increasingly get worse. BUT something will still go wrong…so we **need an active way of sterilizing the nooks**.

- An integrated concept with active mitigation.
- Analog research to understand what the Mars environment will naturally take care of. Is enough being done here, though?
- Work back from robotic standards?
- *Recognize that Habitat will be an incubation facility in our absence.*
- Do we have an obligation to monitor the habitat / what about contamination footprint after we have left it?

- The next return won't be to a pristine location.

Level of Cleaning? Is there a place for shock treatment at key places in the mission timeline?

Cleaning/ Biocontrol Differences and needs based on the hardware being cleaned…

Reduction in bioburden by more than an order of magnitude. Allow stronger biocides on occasion without continual input into the system.

Abandon phase: are there already requirements to prevent this? No, not for landers. After 100 years, microbes may still be viable (e.g. Hughes & Nobbs 2004 based on Antarctic samples)?
Suggest: Do a hard shutdown of the Mars base. Vent everything through a HEPA. Sterility is neither practical nor achievable. Use IPM approach, in general.

Analogue Studies – for mitigation information
Let's go back to the Apollo sites and sample!!!
Gateway is a test case for this and there are experiments that could be left up during Gateway shutdowns.
Using water and electricity to create H2O2 directly. Catalytic systems for destroying H2O2. Airlocks closed – then run H2O2 vapor through needed modules.
'Special regions' on Mars have a fixed limit on the number of Earth microbes-- that is the same for human and robotic missions.

Appendix H: Group Photo, List of Attendees and Breakout Group Assignments

Figure 17: 3rd COSPAR Meeting, Group Photo

Attendees –3rd COSPAR Meeting, May 14-16, 2019
Lunar & Planetary Institute, Houston, TX

	First Name	Last Name	Organization	Email
1	Peter	Doran	LA State University.	pdoran@lsu.edu
2	Esther	Beltran	U of Cen. FL	Esther.Beltran@ucf.edu
3	Natacha	Chough	NASA JSC	natacha.g.chough@nasa.gov
4	Kazu	Fujita	JAXA	Fujita.kazuhisa@jaxa.jp
5	Brian	Glass	Lithic	brian1789@lithic.net
6	Corien	Bakermans	Penn State	cub21@psu.edu
7	Monsi	Roman	NASA MSFC	monsi.roman@nasa.gov
8	Christine	Moissl-Eichinger	University Graz	christine.moissl-eichinger@medunigraz.at
9	Aaron	Regberg	NASA JSC	Aaron.b.regberg@nasa.gov
10	Heather	Smith	NASA ARC	Heather.d.smith@nasa.gov
11	John	Rummel	SETI Institute	jrummel@seti.org
12	John	Canham	NGC	John.canham@ngc.com
13	Christina	Johnson	Univ. Miami	Ckj12@med.miami.edu
14	Petra	Rettberg	DLR	Petra.Rettberg@dlr.de
15	Bette	Siegel	NASA HQ	Bette.siegel@nasa.gov
16	Gerhard	Kminek	ESA	Gerhard.kminek@esa.int
17	Margaret	Race	SETI Institute	mracemom@aol.com
18	Andy	Spry	SETI Institute	aspry@seti.org
19	Mark	Ott	NASA JSC	c.m.ott@nasa.gov
20	Sara	Wallace-Castro	NASA JSC	sarah.wallace@nasa.gov

21	Jericho	Locke	IDA Science &Technology. Policy Institute.	jlocke@ida.org
22	Jeffrey Trauberman or	Cara Cavanaugh	IDA Science & Tech. Policy Inst.	
23	Mark	Lupisella	NASA GSFC	Mark.l.lupisella@nasa.gov
24	Richard	Quinn	NASA ARC	richard.c.quinn@nasa.gov
25	Andrew	Schuerger	Space Life Science Lab	schuerg@ufl.edu
26	Lisa	Pratt	NASA HQ	Lisa.m.pratt@nasa.gov
27	Manish	Patel	Open Univ	Manish.patel@open.ac.uk
28	Bob	Gershman	JPL	Robert.gershman@jpl.nasa.gov
29	Ben	Clark	Space Science Institute.	bclark@SpaceScience.org
30	Vera	Kolb	U.. Wisconsin	kolb@uwp.edu
31	Madhan	Tirumalai	Houston Science Center	madhanrt@gmail.com
32	David	Pearce	U. Northumbria	David.pearce@northumbria.ac.uk
33	Jason	Hatton	ESA	Jason.hatton@esa.int
34	Perry	Stabekis	SETI Institute	pstabekis@gmail.com
35	Stan	Love	NASA JSC	stanley.g.love@nasa.gov
36	Tom	Kieft	New Mexico Tech	Thomas.kieft@nmt.edu
37	Javier	Martin-Torres	Luleå University &Technology, SWE	Javier.martin-torres@ltu,se
38	Cassie	Conley	NASA ARC	cassie.conley@nasa.gov
39	Craig	Kundrot	NASA HQ	Craig.e.kundrot@nasa.gov
40	Tullis	Onstott	Princeton	tullis@princeton.edu
41	Paul	Niles	NASA JSC	Paul.b.niles@nasa.gov
42	Mark	Fonda	NASA ARC	Mark.l.fonda@nasa.gov
43	Juan	Agui	NASA GRC	Juan.h.agui@nasa.gov
44	David	Coil	UC Davis	coil.david@gmail.com
45	Amy	Ross	NASA JSC	Amy.j.ross@nasa.gov
46	George	Fox	U. of Houston	fox@uh.edu
47	Judith	Allton	NASA JSC	Judith.h.Allton@nasa.gov
48	Penelope	Boston	NASA ARC	Penelope.j.boston@nasa.gov
59	Fathi	Karouia	NASA ARC	fathi.karouia@nasa.gov
50	John	Connolly	NASA JSC	John.connolly-1@nasa.gov
51	Kimberly	Jenks	UC Davis	kjjenks@ucdavis.edu
52	Kasthuri	Venkateswaran	JPL	kjvenkat@jpl.nasa.gov
53	Nick	Benardini	JPL	James.n.benardini@jpl.nasa.gov
54	Bob	Lindberg	UVA	Robert.lindberg@cox.net
55	Kevin	Watts	NASA JSC	Kevin.d.watts@nasa.gov
56	Margarita	Marinova	Space X	Margarita.Marinova@spacex.com
57	Valerie	Chabot		
58	Robert	Zimmerman	On his own	<rrzimmer@gmail.com>
59	John	Hallsworth		
60	Guillaume	Weert		<Guillaume.Weerts@esa.int>
61	Sandra	Graham		sgraham@nas.edu

Figure 18: 3rd COSPAR Meeting List of Attendees

Breakout Group Assignments – Days 2-3

R= Rapporteur **S** = Scribe

Breakout Group 1	Breakout Group 2	Breakout Group 3
Agui	Bakermans	Bell
Allton	Boston	Benardini
Beltran	Cavanaugh	Canham
Collom (S)	**Clark (R)**	Chough
Doran	Gershman	Coil
Garcia Robles	Hallsworth	Conley
Glass	Jenks	Fonda
Gunderson	Kminek	Fox
Johnson	Kolb	Fujita
Kundrot (R)	Marinova	Graham
Lindberg	**Moissl-Eichinger (S)**	Hatton
Locke	Niles	Lange
Patel	Ott	Lupisella
Roman	Race	**Pearce (R)**
Rummel	Regberg	Rettberg
Siegel	Smith	Ross
Sturtz	Stabekis	Schuerger
Tirumalai	Weerts	Spry
Venkateswaran	Zaruba	**Wallace-Castro (S)**
		Zimmerman

Figure 19: 3rd COSPAR Meeting Breakout Group Assignments

Appendix I: Contaminate-ometer Conceptual Approach

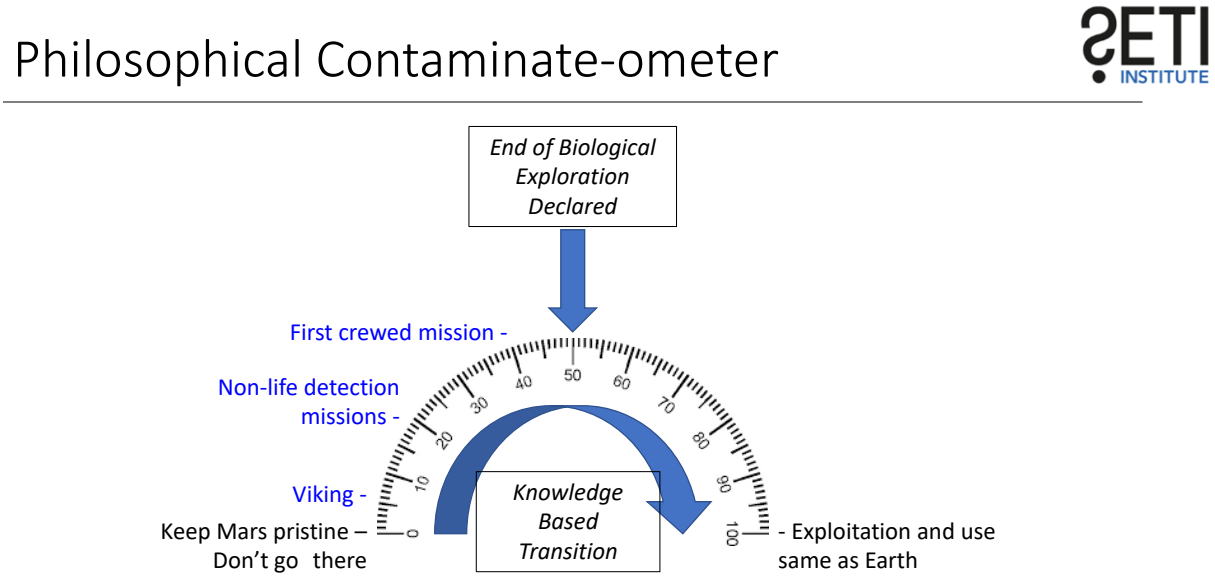

Figure 20: Philosophical Contaminate-ometer

By Andy Spry COSPAR 2019 workshop meeting

The Contaminatometer approach was introduced as a concept to illustrate that COSPAR planetary protection implementation approaches are not fixed in time. In the concept, the scale is the contamination tolerance level for Mars, where zero is where even the risk of contamination is unacceptable (so no missions to Mars are tolerated), and 100 is where Mars is treated as an extension of Earth, and exploration, exploitation and use there is on the same level as for terrestrial environments.

The issue is that the contamination tolerance (ability to cope with the contamination we send there without causing the Harmful Contamination of the kind prohibited in the Outer Space Treaty) of Mars is a knowledge-based assessment: where we have no ground truth of the Mars environment, extreme caution against contamination is warranted, as was executed by the Viking mission. Since the Viking data, we understand that Mars is less contaminatable, so the planetary protection cleanliness requirements for robotic missions have been somewhat relaxed.

Despite this, as expressed in this COSPAR meeting series, we still really have only very limited information on the "contaminatability" of Mars by terrestrial organisms, particularly at the scale of contamination associated with crewed exploration. The desire would be for us to get that information before the irreversible gross contamination that might possibly result from a broad campaign of crewed missions to the Martian surface, protecting the Martian scientific harvest from the blight of terrestrial microbiology. However, at some point in time in a knowledge-based transition, enough will be known about the habitability of Mars; whether it is indeed inhabited, and; who those inhabitants are (if any), that an end to the "period of biological exploration" can be declared. From that point forward, COSPAR planetary protection protocols would be unnecessary for Mars, and sophisticated decisions on permitted contamination levels for Martian environments, perhaps based on a regional, geographic or even hydrological basis can be made by the stakeholders of the day.

Appendix J: List of Acronyms/Glossary

Abbreviations & Acronyms	Term Explanation
ATLO	Assembly, Test and Launch Operations
BSL	Biosafety Level
CDC	Centers for Disease Control
COSPAR	Committee on Space Research
DNA	Deoxyribonucleic Acid
ECLSS	Environmental Control and Life Support System
ESA	European Space Agency
EVA	Extra-vehicular Activity
HEOMD	Human Exploration Mission Directorate
HERA	Human Exploration Research Analog
ICSU	International Council for Science
ID	Identification
IPM	Integrated Pest Management
IR	Infrared radiation
ISRU	In-situ Resource Utilization
ISS	International Space Station
JAXA	Japan Aerospace Exploration Agency
KG	Knowledge Gap
JSC	Johnson Space Center
LALR	Look Ahead Left/Right (algorithm)
LEO	Low-Earth Orbit
LPI	Lunar and Planetary Institute
Meta-analysis	Statistical analysis combining multiple scientific studies to yield weighted average and identify patterns based on multiple contexts.
MHHM	Microbial and Human Health Monitoring
MinION sequencing	Portable, real-time device for DNA and RNA sequencing (instrument by Oxford Nanopore Technologies)
NAD/NADP	Nicotinamide Adenine Dinucleotide (Phosphate)
NASA	National Aeronautics and Space Administration
NASEM	National Academies of Science Engineering and Medicine
NPI	NASA Policy Instruction
NPR	NASA Policy Requirements
Ops.	Operations
Oxford Nanopore	see MinION sequencing
Phylochip	DNA microarray for identifying organisms in complex (community) samples

PP	Planetary Protection
PPIRB	PP Independent Research Board
PPP	Panel on Planetary Protection (COSPAR)
Rodwell	Contraction of Rodriguez well, where snow or ice is melted and stored in place at some depth below the surface of the ice, eliminating the need for fabricated storage tanks
R&TD	Research and Technology Development
SETI	Search for Extraterrestrial Intelligence (Institute)
SMD	Science Mission Directorate
SSB	Space Studies Board (National Academy of Sciences)
TBD	To Be Determined
UV	Ultra Violet radiation/light
WHC	Waste & Hygiene Compartment (on ISS)
wrt	with respect to

Figure 21: List of Acronyms/Glossary