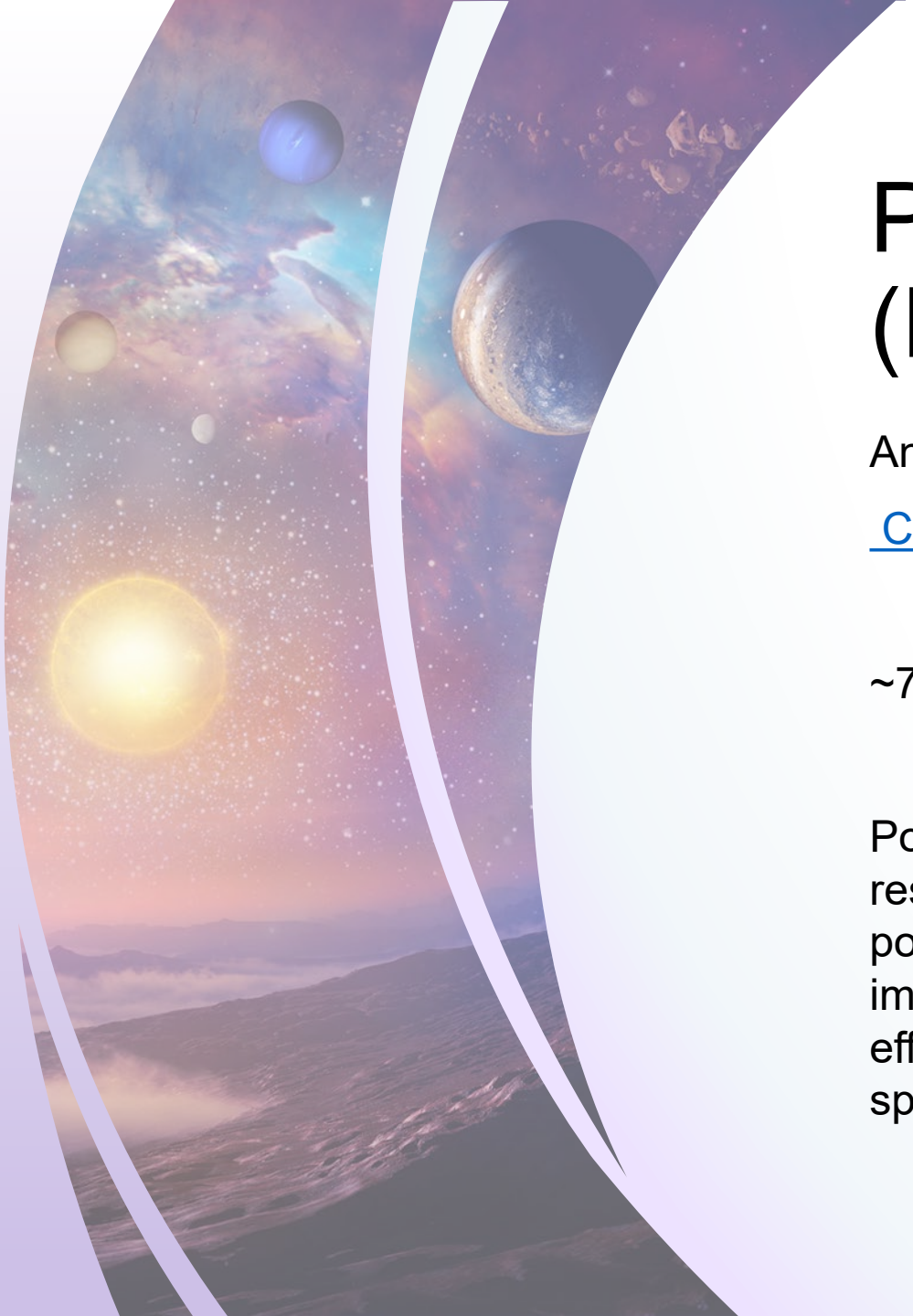
David J. Smith
Program Scientist, Planetary Protection Research

CoPP Fall Meeting Update on Planetary Protection Research Portfolio

October 20, 2023

Program Overview



Planetary Protection Research (PPR)

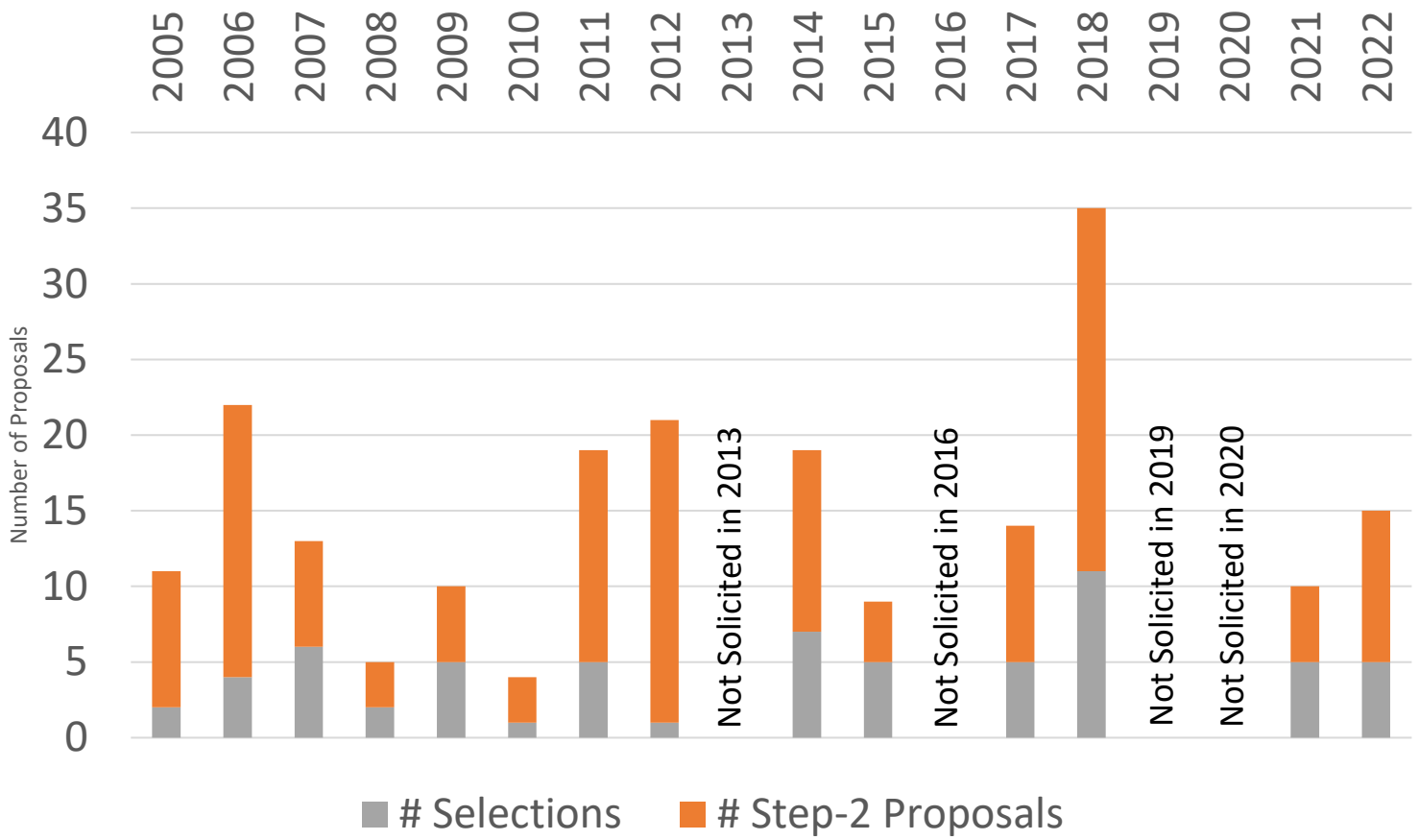
Annual call located in the ROSES solicitation:

C.15 PLANETARY PROTECTION RESEARCH

~7 new awards per year at ~$1M (total)

Portfolio supports mission-enabling and capability-driven research required to improve NASA's understanding of the potential for both forward and backward contamination; and improve methods and technologies for accurate, efficient, and effective minimization of biological contamination for outbound spacecraft and return samples.

Planetary Protection Research – Proposal Submission and Selection Rates



- The previous decade (2012-2021) PPR solicitation cadence was irregular. Variability created challenges for researchers.
- Since ROSES 2021, PPR has been solicited annually. This stability should assist with growing community & pace of impactful research results.
- Selection rates for the past two cycles have been 50% and 33%, respectively.
- Proposal submissions down for ROSES 2023 (*data not shown*)
  - PPR using a dual-anonymous peer review (DAPR) process
  - Open Science Data Management Plan (OSDMP) required

Summary of PPR Solicitation (C.15)

PPR Programmatic Priorities (from C.15)

It should be noted that the evolving planetary protection requirements of NASA's programs may affect the priorities for funding among these areas.

1. *Model or experimentally measure planetary environmental conditions and transport processes that could permit mobilization of spacecraft-associated contaminants to locations in which Earth organisms might thrive.*
2. *Develop or adapt modern molecular analytical methods to rapidly detect, classify, and/or enumerate Earth microbes carried by spacecraft (on surfaces and/or in bulk materials, especially at low densities) before, during, and after assembly and launch processing.*
3. *Model to understand and predict biological and organic contamination sourcing, transport, survival, and burden level of spacecraft, for both forward and backward contamination.*
4. *Model or experimentally measure space environmental conditions and spacecraft designs that could permit a decrease in biological contamination of spacecraft during the journey (e.g. bioburden credits) to the target destination with emphasis on reduction of organisms currently surviving under cleanroom conditions.*
5. *Identify and provide proof-of-concept on new or improved methods, designs, technologies, techniques, and procedures to support planetary protection requirements for outbound and return sample missions.*
6. *Experimentally measure reduction in viability of hardy terrestrial organisms, including viruses, exposed to high temperatures (e.g. 200 to 500 degrees centigrade) for short periods of time (e.g. seconds to minutes).*
7. *Characterize the limits of life in laboratory simulations of relevant planetary environments or in appropriate Earth analogs.*

PPR Portfolio Snapshot

Recent Planetary Protection Research Awards

ROSES 2018

1. An “Optical Swab” for Direct, Non-Contact Microbial Analysis
2. Reducing bioburden of Europa Lander solid rocket motor (SRM) insulation and assessing bioburden of other SRM nonmetallic materials of concern for planetary protection
3. Determining the Dynamic Limits of Life
4. Metabolic Profiling of Cleanroom-Associated Microorganisms
5. Inactivation of Stable Proteinaceous Particles for Outbound and Return Sample Missions
6. Interactive Effects of Vacuum, Ionizing Radiation, Temperature, and UV Irradiation on Bacterial Survival during Simulated Europan Orbiter and Lander Missions
7. Plasma-Activated Water Sterilization (PAWS): Sub-Micrometer Charged Water Droplet Generation in Non-Equilibrium Plasma for Decontamination of Life Detection Mission Spacecraft
8. Survival of Fungal Conidia in Simulated Mars Conditions and Their Molecular Characterization Using Omics Approach
9. Identifying the extent of spacecraft and sample return bioload reduction from optimized radioisotope power system and galactic cosmic ray radiation.
10. EVA Swab: tools and techniques for collecting aseptic samples from crewed space missions
11. Developing a Fundamental Understanding of Workplace Backgrounds: Organic and Organismal Basics for Life Evaluation and Contamination Knowledge (OOBLECK)

ROSES 2021

1. Investigating Far-UVC Radiation to Reduce Microbial Burden During Spacecraft Assembly
2. Effects of Frost-Cycling on Microbial Survival and Growth under Simulated Martian Conditions
3. Rapid Sterilization of Spacecraft Hardware Using High Photon Fluxes from Femtosecond Pulsed Lasers
4. Survivability of spacecraft-associated microbes under simulated radiation environments of Europa and other icy moons
5. Development of In-Flight and In-Situ Microbial Sterilization System using UV LEDs and Heaters to Prevent Re-contamination and Cross-contamination

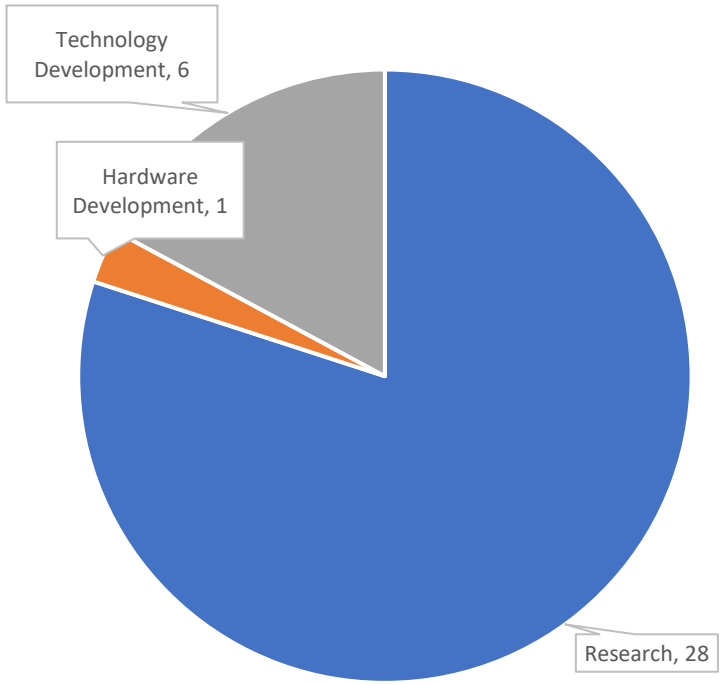
ROSES 2022

1. Metagenomic Methods for Meeting NASA's Planetary Protection Policy Requirements on Future Missions
2. Modeling the Transport of Microbe-Laden Particles in Plume Flows on Mars and Icy Moons
3. A Pilot Study on Forward Contamination in Planetary Analog Environments
4. Windblown Mineral Grains as a Source of Natural Transport of Biological Contamination
5. Responses of Microbial Isolates from Spacecraft Assembly Facilities to the Chemical and Physical Conditions of Mars and the Ocean Worlds

Portfolio currently includes 28 awards that received funding in FY 2023

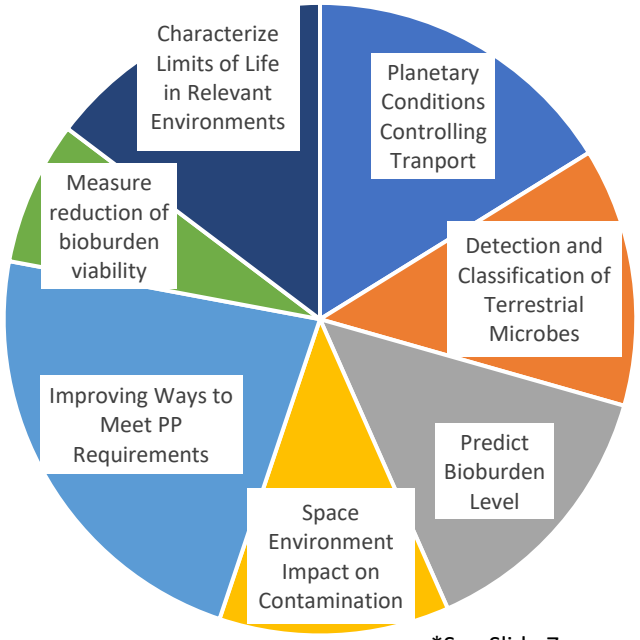
PPR Portfolio Balance

PPR supports both research and tech development



# of Current Awards Binned by Type of Work

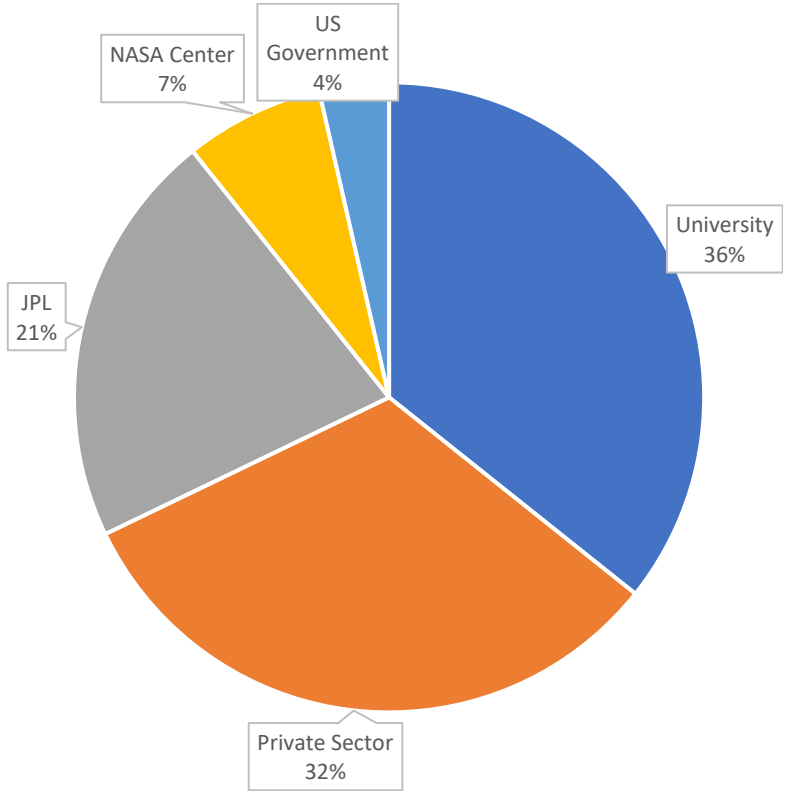
PPR awards distributed across research target areas



*See Slide 7 for further description

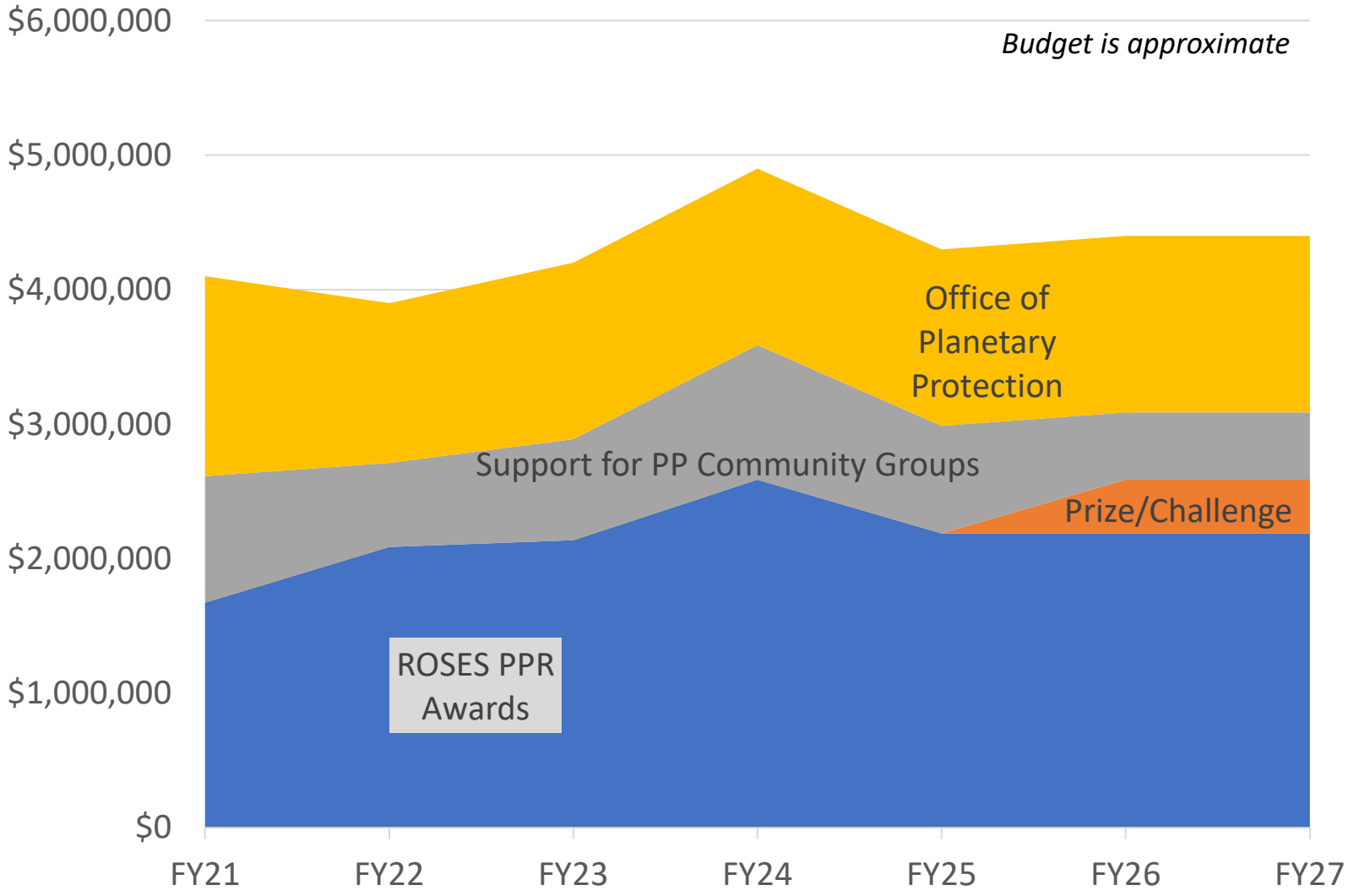
# of Current Awards Binned by Research Target Area

PPR work is ongoing at a variety of institutions



# of Current Awards Binned by Institution Type (PI)

Planetary Protection Budget

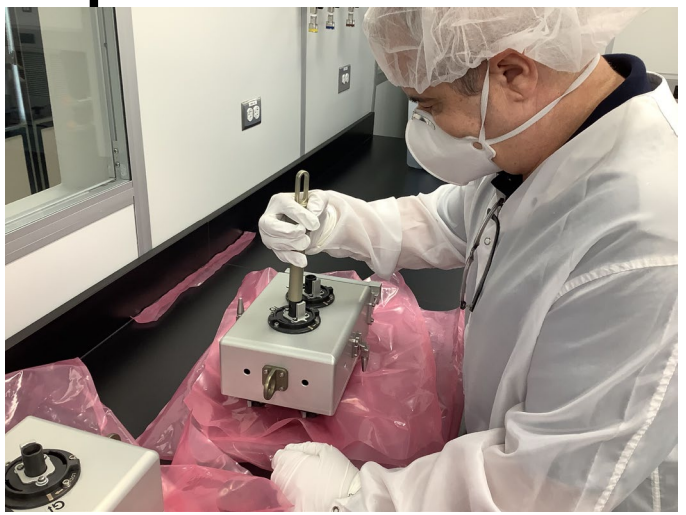


- PSD budget is healthy for support of ROSES PPR awards
- Plans to start supporting Prizes/Challenges in collaboration with STMD
- SMD is continuing robust support for the Office of Planetary Protection
- Ongoing support the community inputs, such as NASEM CoPP, PPIRB
- Examining ways to leverage staff at NASA Centers (i.e., directed work packages relevant to PPR)

PPR Science Highlights

ISS External Microorganisms: A tool to Collect microbes from External Surfaces on the International Space Station (ISS)

PI: Dr. Aaron Regberg, NASA JSC



Some microbes can survive prolonged exposure to space. The ISS releases microbes to space through vents & airlocks. Does the ISS have an external microbiome? What does it look like?

https://www.nasa.gov/mission/station/research-explorer/investigation/?#id=7715

Project Goals:

- Collect 6 samples + 2 controls from ISS external surfaces.
- Return samples to Earth for next generation (amplicon and or metagenomic) DNA sequencing.

Expected Results

- Address planetary protection knowledge gaps and allow NASA to design life support systems that can be used on Mars without introducing unwanted contamination.
- Teach us how microbes evolve in response to exposure to space.
- The tool can be used on future robotic and crewed missions to collect biological, chemical, and geological samples that address Space Biology and Astrobiology questions.

ISS EVA planned for October 2023

PPR Research Area: Develop or adapt modern molecular analytical methods to rapidly detect, classify, and/or enumerate Earth microbes carried by spacecraft (on surfaces and/or in bulk materials, especially at low densities) before, during, and after assembly and launch processing.

Assessing the Potential for the Forward Contamination of a Robotic or Human Exploration Zone by Aeolian Processes on Mars

Dr. Lori Fenton, Principal Investigator, SETI Inst.

Despite rigorous cleaning of spacecraft prior to launch, it is likely that a few stowaway microorganisms manage to survive the rigors of launch, space travel, and landing on the surface of Mars. The ever-blowing wind on Mars routinely blasts sand, builds ripples and dunes, creates whirlwind dust devils, and can form into dust storms that range in size from a few kilometers to planet-encircling. Landed spacecraft and rovers often accumulate sand and dust.

What is the likelihood that windblown sand could knock off any stowaway microorganisms? We are testing what kinetic energy and momentum of sand is needed to blast away killed spores of the bacterium Bacillus subtilis HA101. Figure 1a shows a "sand gun" blowing sand onto an aluminum target coated with B. subtilis spores. The bombarded targets are then inspected with a scanning electron microscope (SEM) to determine how many spores are removed or crushed by the sand. We find that, for similar areal coverage of grain impacts, faster sand (12.4±1.3 m/s) removes or crushes all spores (Fig. 1c), whereas slower sand (2.4±0.2 m/s) only removes or crushes some of the spores (Fig. 1d). We are establishing a relationship between spore removal and sand grain impact speed. Because most spacecraft surfaces are not vertical, we will next investigate the effect of impact angle on spore removal.

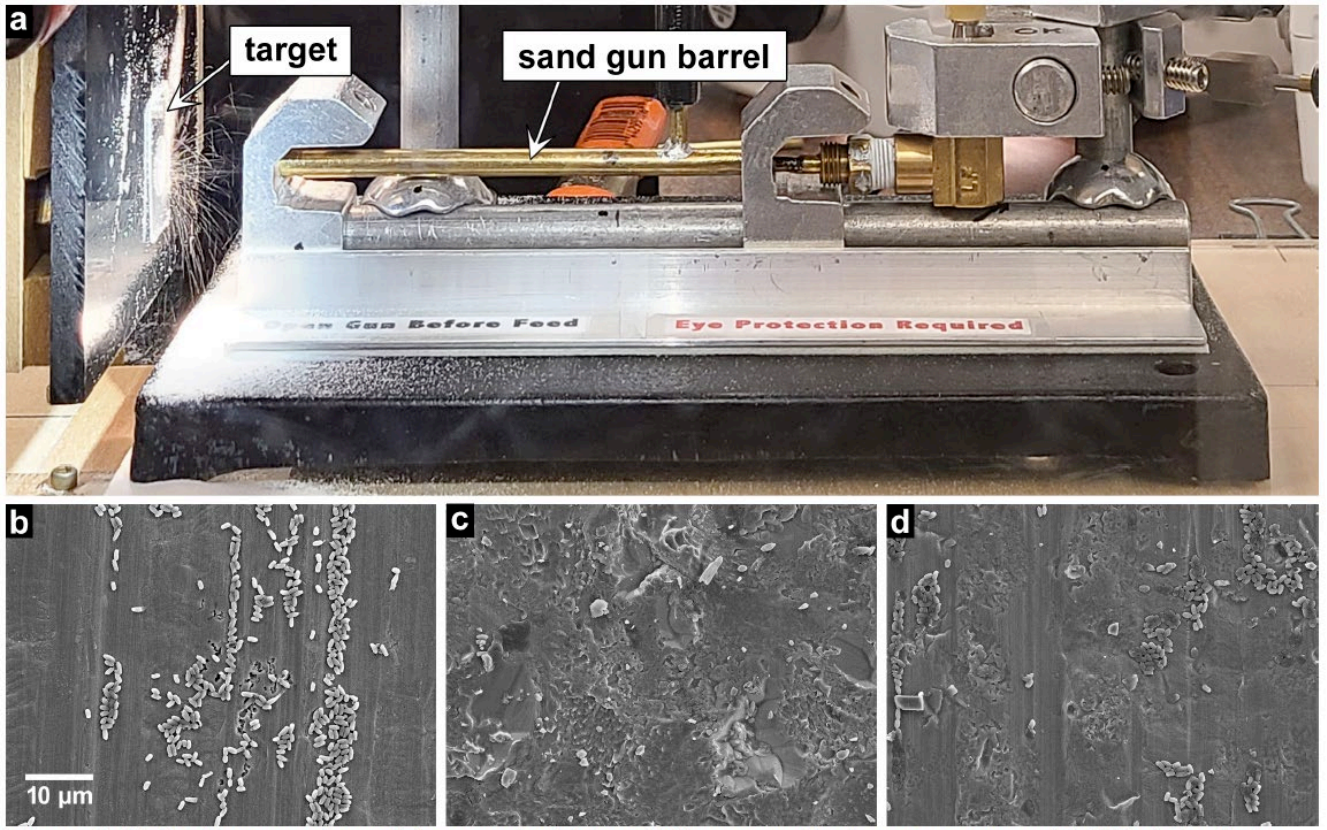


Figure 1. (a) Sand was accelerated down a tube (right to left) onto an aluminum target coated in killed *B. subtilis* HA101 spores. Beneath are portions of SEM images from three coupons: **(b)** Spores on an Al-6061 coupon that was not bombarded by sand (control case). **(c)** Near 100% spore removal by ~99% coverage by sand impacts at ~12.4±1.3 m/s. **(d)** Approximately 30% spore removal by ~99% coverage by sand impacts at ~2.4±0.2 m/s.

PPR Research Area: Model to understand and predict biological and organic contamination sourcing, transport, survival, and burden level of spacecraft, for both forward and backward contamination.

Heat inactivation of stable proteinaceous particles for future sample return mission architecture

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Sec. Extreme Microbiology
Volume 13 - 2022 | https://doi.org/10.3389/fmicb.2022.911091

"Our data show that Sup35NM aggregates are reproducibly inactivated by the exposures to dry heat at 250°C or 350°C for 11.5 min."

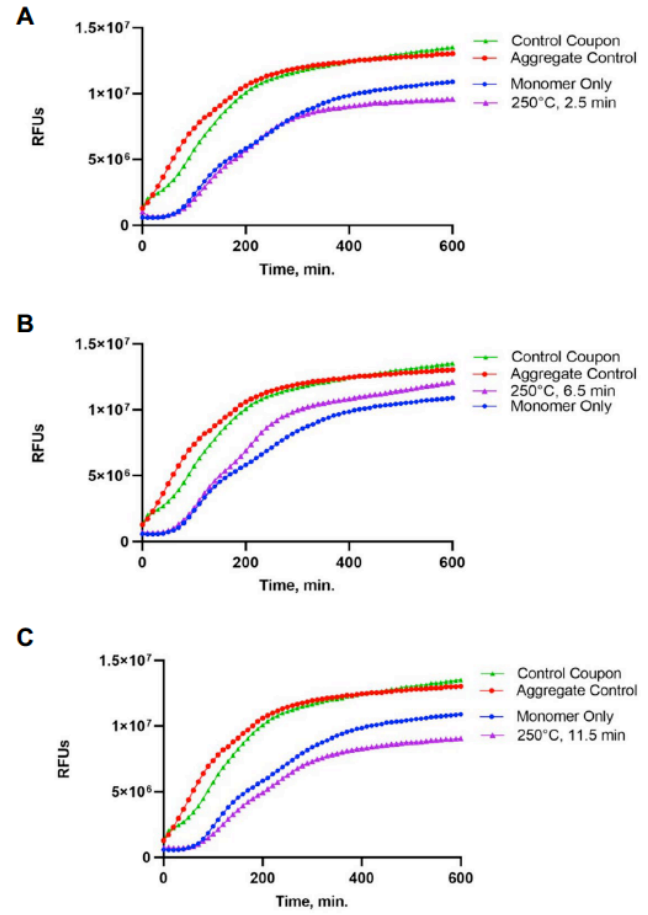

FIGURE 3
Impact of the 250°C treatment on the seeding activity of Sup35NM-His_6 aggregates. Sup35NM-His_6 recovered from coupons treated at 250°C for 2.5min **(A)**, 6.5min **(B)** and 11.5min **(C)** was analyzed. Conditions of the seeding assay and designations are the same as on Figure 2. After 250°C treatment for 2.5min (typical example is shown on **A**) or 11.5min (typical example is shown on **C**), seeding activity was not detected, however it was detected in one of two samples treated for 6.5min (shown on **B**).

PPR Research Area: Experimentally measure reduction in viability of hardy terrestrial organisms, including viruses, exposed to high temperatures (e.g. 200 to 500 degrees centigrade) for short periods of time (e.g. seconds to minutes). Of particular interest are mission enabling time/temperature experiments with greater than 7 decimal reductions in viability.

Delrin Off-Gasses Biocidal VOCs

Problem: Delrin (a polymer) is used in electronic components for planetary spacecraft. Delrin has been noted to produce formaldehyde (a known biocidal VOC). Can Delrin be used to fumigate sealed compartments in spacecraft?

Methods: Spores of three *Bacillus* spp. were exposed to biocidal volatile organic compounds (VOCs) off-gassed by Delrin in sealed chambers to investigate the biocidal nature of the VOCs.

Results: In all assays, 100% of spores from *B. subtilis* 168, *B. pumilus* SAFR-032, and *B. atrophaeus* 9372 were killed within 5 days when the spores were sealed in a small chamber with blocks of Delrin. The figure shows the results for *B. subtilis*.

Delrin may be useable as a 'fumigating' agent for sealed spacecraft subsystems prior to launch.

Schuerger et al., 2022, *IJA*, doi:10.1017/S1473550422000349.

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doi:10.1017/S1473550422000349

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RESEARCH ARTICLE

Microbial protocols for spacecraft: 2. Biocidal effects of Delrin and nylon in sealed compartments may enhance bioburden reductions in planetary spacecraft

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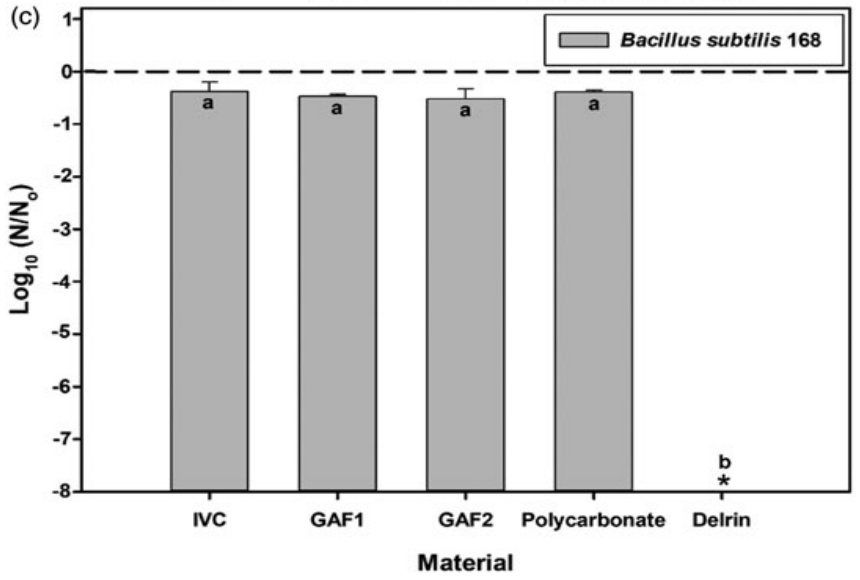

PPR Research Area: Identify and provide proof-of-concept on new or improved methods, designs, technologies, techniques, and procedures to support planetary protection requirements for outbound and return sample missions.

Questions?

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