Planetary Biosecurity: Applying Invasion Science to Prevent Biological Contamination from Space Travel

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As plans for space exploration and commercial use expand rapidly, biosecurity measures and risk assessments that inform them must adapt. Sophisticated protocols are required to prevent biological contamination of extraterrestrial environments from Earth and vice versa. Such protocols should be informed by research on biological invasions—human-assisted spread of organisms into novel environments—which has revealed, inter alia, that (1) invasion risk is driven by the timing and frequency of introduction events, whose control requires addressing the least secure human activities associated with organismal transport; (2) invasions and their impacts are difficult to predict, because these phenomena are governed by context dependencies involving traits of the organism and the receiving environment; and (3) early detection and rapid response are crucial for prevention but undermined by taxonomic methods that fail to recognize what is "alien" versus what is native. Collaboration among astrobiologists, invasion biologists, and policymakers could greatly enhance planetary biosecurity protocols.

Keywords: astrobiology, biological contamination, biosecurity, disaster preparedness, invasion biology, invasive species, planetary protection, risk management

Biological invasions—the human-assisted spread of organisms into novel environments, in which such species are most often termed "alien"—are a threat to ecosystem sustainability and human well-being (Pyšek et al. 2020). Owing to human activities, the rate of spread of alien microbes, invertebrates, vertebrates, and plants across the planet is unprecedentedly high with no sign of saturation (Seebens et al. 2017, 2021). Even the remotest alpine, polar, and deep ocean regions of the Earth have been invaded (e.g., Voight et al. 2012, Lamsal et al. 2018, McCarthy et al. 2019, Chan et al. 2019); for example, recent evidence indicated that humans have inadvertently introduced drug-resistant enteric bacteria into the Antarctic ecosystem, infecting seabirds and seals (Cerda-Cuellar et al. 2019). Whereas many invasions appear to have had minor impacts, others have contributed to substantial biodiversity loss and detrimental effects on human health and livelihoods (Bellard et al. 2016, Russell et al. 2017, Pyšek et al. 2020). Some have also caused pervasive and profound changes to ecosystem functioning (e.g., nutrient cycling, productivity, carbon sequestration) and evolutionary trajectories (Ricciardi et al. 2013, Underwood et al. 2019, Stigall 2019, Thakur et al. 2019, Xing et al. 2020).

Owing to their massive costs to resource sectors (e.g., agriculture, forestry, aquaculture, apiculture; Diagne et al. 2021) and human health, biological invasions are a global biosecurity issue requiring rigorous transboundary solutions (Hulme 2020, Ricciardi et al. 2021). For example, the International Maritime Organization has implemented regulations and standards to control the discharge of ballast water from transoceanic shipping (Campara et al. 2019), which is the source of many ecologically and socioeconomically damaging marine invasions. To address the science and management of invasive alien species, a highly productive interdisciplinary field has emerged over the past few decades: invasion science (also known as *invasion biology* but embracing nonbiological disciplines)—the study of the causes and consequences of the introduction of organisms beyond their natural evolutionary ranges, with emphasis on the role of humans in these introductions. Research in invasion science has produced novel insights for epidemiology, rapid evolution, the relationship between biodiversity and community stability, and the dynamics of predator–prey and parasite–host interactions, among many other concepts (Hui and Richardson 2017). These insights are being increasingly

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integrated into biosecurity frameworks (e.g., Hulme 2020, Hulme et al. 2020).

Biosecurity at the planetary scale

Space is emerging as a new frontier of biosecurity risk. Space exploration and use is currently undergoing a revolution, led by NewSpace—a global industry of private and public companies seeking to profit from products or services (Profitiliotis and Loizidou 2019). In addition to government-led space missions, the arrival of private companies (e.g., SpaceX) and new international players has caused space to be accessible by a broader range of actors than was ever possible previously (Weinzierl 2018). Therefore, the risk profile associated with space activities has dramatically changed in ways that regulatory and policy frameworks have not anticipated.

Space biosecurity concerns the delivery of living organisms from Earth to extraterrestrial bodies (forward contamination) and, conversely, Earth biota coming into contact with organisms from extraterrestrial bodies in the context of a mission returning to Earth (back contamination; Rummel and Pugel 2019). At present, these are considered to be highly improbable events. For a mission to Mars, for example, the risk of survival and proliferation of introduced terrestrial organisms is thought to be low (e.g., Pavlov et al. 2012, Khodadad et al. 2017), and an even more negligible risk might be ascribed to the event in which a living organism from Mars is transported to Earth, subsequently released, and colonizes its new environment. However, we suggest that these biological invasion scenarios are analogous to extreme natural or technological disasters (e.g., major earthquakes, nuclear meltdowns) that, although typically rare, have potential consequences that are unacceptable and therefore merit unique safeguards (Ricciardi et al. 2011). The risk of accidental forward contamination of extraterrestrial environments was demonstrated recently when an Israeli lunar lander (named Beresheet), belonging to a private organization (SpaceIL), crashed on the moon (Shahar and Greenbaum 2020). It was carrying thousands of dormant tardigrades—invertebrate animals notorious for their ability to withstand harsh conditions, such as extreme desiccation, freezing temperatures, and high doses of ionizing radiation (Møbjerg et al. 2011). Of course, missions to Mars would be subject to more stringent sterilization measures, but the lunar crash illustrated the nonnegligible risk of technological failure and therefore the need for disaster preparedness against biological spills.

The risks of back and forward contamination associated with future missions may be higher and more diverse than previously estimated. Indeed, an emerging view is that it is nearly impossible to explore new planets without carrying or delivering microbes (Lopez et al. 2019). Various organisms (some of them extremophilic) are found to exhibit tolerance to conditions necessary for space travel (Tirumalai et al. 2017, 2019, Danko et al. 2021). For example, some bacteria can grow and evolve under microgravity (Tirumalai et al. 2017, 2019), and an extensive list of microbes can survive extreme cold, radiation, and desiccation (e.g., Sánchez et al. 2012, Vaishampayan et al. 2014, Cheptsov et al. 2017, Pacelli et al. 2019, Danko et al. 2021). Cheptsov and colleagues (2017) demonstrated that certain bacteria (e.g., *Arthrobacter* spp.) from ancient Arctic permafrost can survive exposure to gamma radiation (100 kilograys) at low temperature (−50 degrees Celsius), low atmospheric pressure (1 torr), and desiccation—environmental conditions similar to Martian permafrost. Furthermore, recent evidence suggests that certain subsurface regions of Mars could support high microbial cell densities (Tarnas et al. 2021). Such areas of Mars and other extraterrestrial planets and moons deemed potentially habitable would be targeted for missions dedicated to searching for extant life (Carrier et al. 2020), thereby amplifying the risks of forward and back contamination.

Insights from the science of biological invasions: Hard lessons learned

Given the enormous foundation of research in the science and management of invasive species, we contend that greater collaboration between invasion biologists and astrobiologists would enhance existing international protocols for planetary biosecurity—both for Earth and for extraterrestrial bodies that could contain life. Below, we discuss some practical insights from invasion science that could inform policies dedicated to risk reduction in future space missions.

Insular systems are most vulnerable to invasion. A major lesson from invasion science is that ecosystems that have evolved in isolation are exceptionally vulnerable to disruption by introduced alien organisms (Sih et al. 2010, Ricciardi et al. 2013, Carthey and Banks 2014). For insular systems (e.g., islands, lakes, remote habitats), which typically contain endemic species and unique phylogenetic lineages, the consequences of invasion have often been catastrophic, causing cascading ecological effects and extinctions (Ricciardi and MacIsaac 2011, Bellard et al. 2016, Russell et al. 2017). The heightened sensitivity of insular biota is attributable primarily to their lack of adaptations for coexisting with a broad suite of alien life forms (Ricciardi et al. 2013, Saul and Jeschke 2015, Anton et al. 2020). This sensitivity justifies special precautions to protect insular biota against invasion, even when such events are considered to be extremely improbable. In this context, planets and moons that are deemed potentially capable of supporting life should be treated as high-risk insular systems.

Opportunity and weakest-link dynamics drive invasion risk. Theory and empirical evidence correlate the risk of a biological invasion with propagule pressure—the number of release events and the number of organisms released per event (Lockwood et al. 2005, Simberloff 2009, Cassey et al. 2018, Stringham and Lockwood 2021). However, for asexually reproducing taxa with short generation times, such as prokaryotes, the timing of a release event can be more important than the numbers of organisms introduced (Dressler et al. 2019, Peniston 2020). Although such opportunistic events can be modelled (Stringham and Lockwood 2021), they are difficult to manage within complex environmental or technological systems, which typically involve hidden interactions, chain reactions, and tipping points—as are found in natural disasters, technological catastrophes, and invasive species outbreaks (Sornette 2002, Ricciardi et al. 2011, Hui and Richardson 2017). Analogous to disaster management (Ricciardi et al. 2011), the management of biological invasions is challenged by having to identify and strengthen the most vulnerable links in a system in which a single failure of any such link could lead to a species introduction.

When propagule pressure is no longer limiting, areas are less protected by their remoteness and a very small number of opportunities could lead to an invasion. There are a multitude of documented cases of accidental introductions to remote areas, even those under special protection (e.g., Antarctica; Hughes et al. 2010). One noteworthy example is the discovery of a small disjunct population of a marine invertebrate species in a hydrothermal vent field at 2.7 kilometers depth in the northeast Pacific Ocean, which prompted an investigation to determine the population's origin. Using multiple lines of evidence that included genetic sequences and stable isotopic signatures, the species was revealed to have been introduced as a contaminant on a deep-sea exploration vehicle, in spite of attempts to clean the vehicle between multiple dives (Voight et al. 2012).

Although protocols are outlined in existing international policy on planetary protection by the Committee on Space Research (COSPAR Panel on Planetary Protection 2020), humans might have already introduced organisms to Mars during the circa 30 missions that sent spacecraft to the planet (Mason 2021). Within the past decade—an era of relatively heightened planetary protection—bacterial strains exhibiting extreme resistance to ionizing radiation, desiccation, and disinfectants have been isolated in NASA "clean rooms" used for spacecraft assembly (Vaishampayan et al. 2012, Trumalai and Fox 2013). Similarly, a previously undescribed bacterium (*Deinococcus phoenicis*), resistant to extreme doses of ionizing radiation, was discovered in a clean room at the Kennedy Space Center, where the Phoenix spacecraft was assembled (Vaishampayan et al. 2014). In a recent experiment, cells of another extremophile bacterium *Deinococcus radiodurans* survived up to 3 years on the exterior of the International Space Station in orbit (Kawaguchi et al. 2020), suggesting that viable cells could be transported between Earth and Mars. These observations also provide a basis for the hypothesis that stress-induced mortality and selection during space travel (via incomplete sterilization, desiccation, or radiation exposure within or on the spacecraft) could enhance an organism's tolerance and possibly its invasion potential (von Hegner 2020). For these reasons, enhanced protocols need to be developed for the detection of microbes (including bacteria, archaea, protists, fungi, and viruses) during interplanetary missions.

Impacts of invasions are extremely difficult to forecast. Although much progress has been made in understanding invader impacts (environmental changes caused by invasion), a major knowledge gap in the risk assessment of invasive species is the role of species traits, combinations of traits, and trait–environment interactions in determining impact, particularly at the ecosystem level (Ricciardi et al. 2021). This gap is greater for introduced nonpathogenic microbial organisms, for which the links between traits and influence on ecosystem function are not well elucidated (van der Putten et al. 2007, Litchman 2010, Mallon et al. 2015). Nevertheless, evidence shows that introduced microbes can dramatically alter biotic communities (Hewson et al. 2014, Mallon et al. 2018, Scheele et al. 2019, Xing et al. 2020) and produce substantial ecosystem changes over time (Litchman 2010, Mallon et al. 2018).

The evolutionary dynamics of invaders can also confound prediction. Introduced microbial organisms can undergo rapid genetic changes through environmental selection or through hybridization with compatible strains that result in enhanced invasiveness or novel pathogenic behavior (Stuckenbrock et al. 2012, Mallon et al. 2015). More ominously, as was mentioned above, it has been shown that microorganisms could adapt to or be altered by the stressors of a space environment. A study that found that *Escherichia coli* grown over a thousand generations in simulated microgravity conditions in the laboratory underwent mutations, developed adaptive responses, became more competitive than a normal strain of the same species (Tirumalai et al. 2017), and acquired antibiotic resistance under such conditions—even when only trace levels of antibiotics were introduced to the system (Tirumalai et al. 2019). Such cases apparently signify a common phenomenon; for example, adaptations to human-disturbed habitat have been shown to promote the spread of various species into novel environments (Hufbauer et al. 2012). The rapid evolution of invasiveness is an emerging research focus in invasion science (Ricciardi et al. 2017, Sherpa and Després 2021).

Early detection and rapid response are crucial for prevention. Because the costs of impact can far exceed costs of prevention (Leung et al. 2002, Diagne et al. 2021), early detection and rapid response protocols are prioritized in invasive species risk management (Reaser et al. 2020). Alien microbes, in particular, pose management challenges, including detection at very low abundance. At present, biologists have limited ability to detect or identify emerging microbial invasion threats within the time periods necessary to prevent establishment and spread, which has been highlighted as a significant biosecurity gap (Ricciardi et al. 2017). The development of portable real-time DNA sequencing technologies, especially those that can be employed in space (e.g., the Oxford Nanopore MinION sequencer; Castro-Wallace et al. 2017), is crucial for detecting microbial life. A valuable resource to complement these technologies would be a database of all known organismal contaminants found at clean room facilities or likely to be found in facilities built in the future. Although NASA has tested the microbiological profiles of its spacecraft since the early days of the Apollo missions (e.g., Puleo et al. 1970), there is no complete database publicly available yet and we are not aware of any such databases provided by other nations (e.g., Russia, Japan, China).

Other major challenges might be recognizing what is “alien” versus what is native and the possibility of misidentifying pseudoindigenous organisms that were introduced cryptically by humans. Global genomic data provides an essential but incomplete reference (Nayfach et al. 2021), because new microbial life forms continue to be discovered. For example, previously unknown bacterial phylotypes were retrieved from the Antarctic subglacial Lake Vostok in 2012—an endeavor that required strict protocols for controlling contamination, which were implemented through the use of clean-room facilities and a contaminant library. In spite of these protocols, nearly 50 contaminant phylotypes were detected in drill bit and borehole frozen lake water samples (Bulat 2016). New forms of life have also been discovered beyond terrestrial boundaries: A previously undescribed bacterium (*Methylobacterium ajmalii*) found on the International Space Station (Bijlani et al. 2021) is related to a diverse lineage of taxa occurring ubiquitously in air, soil, and fresh water. Several other previously undescribed species have been discovered in NASA clean rooms, including hardy spore-forming taxa that could potentially survive space flight (Danko et al. 2021). Given that rapid adaptive evolution of introduced species is a common phenomenon (Whitney and Gabler 2008, Sherpa and Després 2021), if any of the past missions to Mars have delivered organisms that survived on the planet, it is conceivable that they have since rapidly evolved trait differences with phylogenetically related species on Earth, thereby presenting a new risk of back contamination that would be difficult to recognize and could introduce unknown but potentially disastrous impacts.

Planetary protection must keep up with scientific advances

The 1967 Treaty on Principles Governing the Activities of States in the Exploration and Use of Outer Space, including the Moon and Other Celestial Bodies, underpins international space law. There are provisions for biological risks in the treaty, notably article IX: “In the exploration and use of outer space, including the Moon and other celestial bodies, States Parties to the Treaty shall be guided by the principle of cooperation and mutual assistance and… conduct exploration of them so as to avoid their harmful contamination and also adverse changes in the environment of the Earth resulting from the introduction of extraterrestrial matter and, where necessary, shall adopt appropriate measures for this purpose.” (United Nations 1967). Clearly, as we described above, there has been significant advancement of our knowledge of contamination risk since the 1960s when the treaty was first developed. But in some cases there exists a gap between legislated biosecurity and standards necessary to reduce risk. *Hayabusa2* is an asteroid sampling mission that returned to Earth and landed in Australia in December 2020 (figure 1). For the returning jurisdiction, the key regulatory biosecurity requirement is that “the minister is satisfied that the probability of the return or returns causing substantial harm to public health or public safety or causing substantial damage to property is as low as is reasonably practicable” (Australian government 2018). We doubt that this is adequate. In contrast, material from the Mars sample return missions will be subjected to strict containment and biohazard protocols (Viso 2019), although samples returned from Mars’ moons are currently designated as “unrestricted Earth return” (*sensu* COSPAR Panel on Planetary Protection 2020) and can therefore be managed in a standard laboratory without quarantine (National Academies of Sciences, Engineering, and Medicine 2019).

We highlight four essential components of biosecurity and disaster preparedness that should be applied to biological invasions, from regional to planetary scales: vulnerability reduction (recognizing risks and weak links in a sequence of events and having mechanisms in place for preventing biological contamination), rapid response and assessment (ability to detect an alien organism, assessing its risk of colonization, and having a response plan to eradicate or contain the organism), access to reliable information (e.g., through global databases), and coordination among managing authorities (private and public jurisdictions, agencies, and nations; Ricciardi et al. 2011, García-Díaz et al. 2021). Finally, we propose that the COSPAR policy on planetary protection would benefit from interdisciplinary input that enables the identification of critical knowledge gaps. For example, a Donald Rumsfeld approach to risk assessment acknowledges that there are “known knowns,” there are “known unknowns,” and there are “unknown unknowns” (Logan 2009). Most biosecurity principles are focused on known threats, but to achieve more robust protection it is the unknown unknowns that need to be minimized—for example, through scenario planning (Peterson et al. 2003) and horizon scanning (Ricciardi et al. 2017).

Conclusions

International space activities are burgeoning. China has recently landed a rover on Mars. The European Space Agency will launch their ExoMars rover in 2022 (ESA 2021). NASA’s Mars Sample Return mission is planning to bring back the first Martian samples to Earth by the early 2030s (NASA 2020). Japan’s Martian Moons eXploration project will send probes to both of Mars’ moons with the goal of bringing back samples by 2029 (Hyodo and Usui 2021). Other planned missions will target the moons of Jupiter (e.g., Europa) and Saturn (e.g., Enceladus), whose internal liquid oceans might harbor life (Clery 2021). Further in the future, a manned mission to Mars seems inevitable, and this goal will likely drive efforts to bioengineer life (e.g., novel organisms that can generate food, process waste, produce

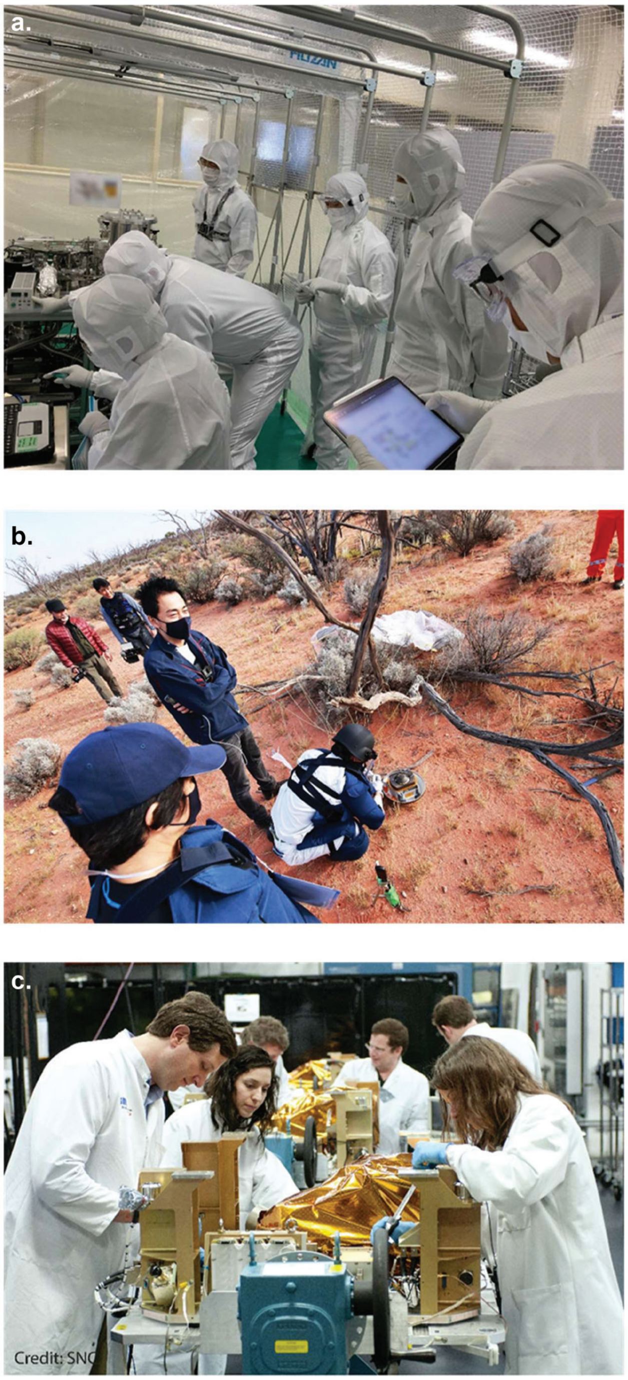

Figure 1. The risk profile and biosecurity preparedness vary considerably across the space exploration sector. (a) After the completion of the setup of the clean booth, a test run is underway for* Hayabusa2 *prior to launch. Despite precautions to mitigate against biological contamination, microorganisms can survive these clean environments (Moissl-Eichinger 2011). Photograph: JAXA. (b) Recovery of the* Hayabusa2 *reentry capsules in Australia 6 December 2020. The focus is on the integrity of the samples. Photograph: JAXA. (c) Sierra Nevada Corporation engineers inspect their OG2 spacecraft. The focus is on the production of technology. Photograph: Sierra Nevada Corporation.

biofuel, or serve as biocomposite building material) to aid in long-term space travel and potential human settlement (Rothschild 2018, Janjic 2019). During this time, expansion of private missions in space will continue. Advances in planetary biosecurity must keep up with the pace of these potential risks, and this goal could be facilitated by collaborations between researchers in invasion science and astrobiology. To our knowledge, invasion biologists have not been involved in development of the COSPAR policy on planetary protection, in spite of obvious conceptual parallels and decades of empirically derived insights that could be applied. Protocols for early detection, hazard assessment, rapid response, and containment procedures currently employed for invasive species on Earth could be adapted for dealing with potential extraterrestrial contaminants on spacecraft or on biological material intentionally transported to Earth for analysis.

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References cited

Anton A, Geraldi NR, Ricciardi A, Dick JTA. 2020. Global determinants of prey naiveté to exotic predators. Proceedings of the Royal Society B 287: 20192978.

Australian government. 2018. Space (Launches and Returns) Act 2018. Australian Federal Register of Legislation. www.legislation.gov.au/Details/C2019C00246.

Bellard C, Cassey P, Blackburn TM. 2016. Alien species as a driver of recent extinctions. Biology Letters 12: 20150623.

Bijlani S, Singh NK, Eedara VVR, Podile AR, Mason CE, Wang CCC, Venkateswaran K. 2021. *Methylobacterium ajmalii* sp. nov., isolated from the International Space Station. Frontiers in Microbiology 12: 639396.

Bulat SA. 2016. Microbiology of the subglacial Lake Vostok: First results of borehole-frozen lake water analysis and prospects for searching for lake inhabitants. Philosophical Transactions of the Royal Society A 374: 20140292.

Campara L, Francic V, Maglic L, Hasanspahic N. 2019. Overview and comparison of the IMO and the US Maritime Administration Ballast Water Management Regulations. Journal of Marine Science and Engineering 7: art.283.

Carrier BL, Beaty DW, Meyer MA, et al. 2020. Mars extant life: What's next? Astrobiology 20: 785–814.

Carthey AJR, Banks PB. 2014. Naïveté in novel ecological interactions: Lessons from theory and experimental evidence. Biological Reviews 89: 932–949.

Cassey P, Delean S, Lockwood JL, Sadowski JS, Blackburn TM. 2018. Dissecting the null model for biological invasions: A meta-analysis of the propagule pressure effect. PLOS Biology 16: e2005987.

Castro-Wallace SL, et al. 2017. Nanopore DNA sequencing and genome assembly on the International Space Station. Scientific Reports 7: 18022.

Cerda-Cuellar M, More E, Ayats T, Aguilera M, Munoz-Gonzalez S, Antilles N, Ryan PG, Gonzalez-Solis, J. 2019. Do humans spread zoonotic enteric bacteria in Antarctic? Science of the Total Environment 654: 190–196.

Chan FT, Stanislawczyk K, Sneekes AC, Dvoretsky A, Gollasch S, Minchin D, David M, Jelmert A, Albretsen J, Bailey SA. 2019. Climate change opens new frontiers for marine species in the Arctic: Current trends and future invasion risks. Global Change Biology 25: 25–38.

Cheptsov VS, Vorobyova EA, Manucharova NA, Gorlenkon MV, Anatoli K, Pavlov AK, Vdovina MA, Lomasov VN, Bulat SA. 2017. 100 kGy gamma-affected microbial communities within the ancient Arctic permafrost under simulated Martian conditions. Extremophiles 21: 1057–1067.

COSPAR Panel on Planetary Protection. 2020. COSPAR policy on planetary protection. Space Research Today 208: 10–22.

Clery D. 2021. Decades ahead, Europe picks goals for big space missions. Science 372: 1252–1253.

Danko DC, Sierra MA, Benardini JN, Guan L, Wood JM, Singh N, et al. 2021. A comprehensive metagenomics framework to characterize organisms relevant for planetary protection. Microbiome 9: 82.

Diagne C, Leroy B, Vaissière A-C, Gozlan RE, Roiz D, Jarić I, et al. 2021. High and rising economic costs of biological invasions worldwide. Nature 592: 571–576.

Dressler MD, Conde J, Eldakar OT, Smith RP. 2019. Timing between successive introduction events determines establishment success in bacteria with an Allee effect. Proceedings of the Royal Society B 286: 20190598.

[ESA] European Space Agency. 2021. ExoMars orbiter continues hunt for key signs of life on Mars. Phys.org (20 July 2021). https://phys.org/news/2021-07-exomars-orbiter-key-life-mars.html.

García-Díaz P, et al. 2021. Management policies for invasive alien species: Addressing the impacts rather than the species. BioScience 71: 174–185.

Hewson I, et al. 2014. Densovirus associated with sea-star wasting disease and mass mortality. Proceedings of the National Academy of Science 111: 17278–17283.

Hufbauer RA, Facon B, Ravigné V, Turgeon J, Foucaud J, Lee CE, Rey O, Estoup A. 2012. Anthropogenically induced adaptation to invade (AIAI): Contemporary adaptation to human-altered habitats within the native range can promote invasions. Evolutionary Applications 5: 89–101.

Hughes KA, Conway P, Masel NR, Smith RIL. 2010. Accidental transfer of non-native soil organisms into Antarctica on construction vehicles. Biological Invasions 12: 875–891.

Hui C, Richardson DM. 2017. Invasion Dynamics. Oxford University Press.

Hulme PE. 2020. One Biosecurity: A unified concept to integrate human, animal, plant, and environmental health. Emerging topics in Life Sciences 4: 539–549.

Hulme PE, Baker R, Freckleton RP, Hails RS, Hartley M, Harwood J, Marion G, Smith GC, Williamson M. 2020. The Epidemiological Framework for Biological Invasions (EFBI): An interdisciplinary foundation for the assessment of biosecurity threats. NeoBiota 62: 161–192.

Hyodo R, Usui T. 2021. Searching for life on Mars and its moons. Science 373: 742.

Janjic A. 2019. Assisted evolution in astrobiology: Convergence of ecology and evolutionary biology within the context of planetary colonization. Astrobiology 19: 1410–1417.

Kawaguchi Y, et al. 2020. DNA damage and survival time course of Deinococcal cell pellets during 3 years of exposure to outer space. Frontiers in Microbiology 11: 2050.

Khodadad CL, Wong GM, James LM, Thakrar PJ, Lane MA, Catechis JA, Smith DJ. 2017. Stratosphere conditions inactivate bacterial endospores from a Mars spacecraft assembly facility. Astrobiology 17: 337–350.

Lamsal P, Kumar L, Aryal A, Astreya K. 2018. Invasive alien plant species dynamics in the Himalayan region under climate change. Ambio 47: 697–710.

Leung B, Lodge DM, Finnoff D, Shogren JF, Lewis MA, Lamberti G. 2002. An ounce of prevention or a pound of cure: Bioeconomic risk analysis of invasive species. Proceedings of the Royal Society B 269: 2407–2413.

Litchman E. 2010. Invisible invaders: Non-pathogenic invasive microbes in aquatic and terrestrial ecosystems. Ecology Letters 13: 1560–1572.

Lockwood JL, Cassey P, Blackburn T. 2005. The role of propagule pressure in explaining species invasions. Trends in Ecology and Evolution 20: 223–228.

Logan DC. 2009. Known knowns, known unknowns, unknown unknowns and the propagation of scientific enquiry. Journal of Experimental Botany 60: 712–714.

Lopez JV, Peixoto RS, Rosado AS. 2019. Inevitable future: Space colonization beyond Earth with microbes first. FEMS Microbiology Ecology 95: fiz127.

Mallon CA, Le Roux X, van Doorn GS, Dini-Andreote F, Poly F, Salles JF. 2018. The impact of failure: Unsuccessful bacterial invasions steer the soil microbial community away from the invader's niche. ISME Journal 12: 728–741.

Mallon CA, van Elsas JD, Falcão Salles J. 2015. Microbial invasions: The process, patterns, and mechanisms. Trends in Microbiology 23: 719–729.

Mason C. 2021. Could humans have contaminated Mars with life? BBC Future (10 May 2021). www.bbc.com/future/article/20210510-could-the-perseverance-rover-have-carried-life-to-mars.

McCarthy A, Peck LS, Hughes KA, Aldridge DC. 2019. Antarctica: The final frontier for marine biological invasions. Global Change Biology 25: 2221–2241.

Møbjerg N, Halberg KA, Jørgensen A, Persson D, Bjørn M, Ramløv H, Kristensen RM. 2011. Survival in extreme environments: On the current knowledge of adaptations in tardigrades. Acta Physiologica 202: 409–420.

Moissl-Eichinger C. 2011. Archaea in artificial environments: Their presence in global spacecraft clean rooms and impact on planetary protection. ISME Journal 5: 209–219.

NASA. 2020. NASA Moves Forward with Campaign to Return Mars Samples to Earth. NASA Press Releases (17 December 2020). www.nasa.gov/press-release/nasa-moves-forward-with-campaign-to-return-mars-samples-to-earth.

National Academies of Sciences, Engineering, and Medicine. 2019. Planetary Protection Classification of Sample Return Missions from the Martian Moons. National Academies Press. doi:10.17226/25357.

Nayfach S, Roux S, Seshadri R, Udwary D, Varghese N, Schulz F, et al. 2021. A genomic catalog of Earth's microbiomes. Nature Biotechnology 39: 499–509.

Pacelli C, Selbmann L, Zucconi L, Coleine C, de Vera J-P, Rabbow E, Böttger U, Dadachova E, Onofri S. 2019. Responses of the black fungus *Cryomyces antarcticus* to simulated martian and space conditions on rock analogs. Astrobiology 19: 209–220.

Pavlov AA, Vasilyev G, Ostryakov VM, Pavlov AK, Mahaffy P. 2012. Degradation of the organic molecules in the shallow subsurface of Mars due to irradiation by cosmic rays. Geophysical Research Letters 39: L1320.

Peniston JH. 2020. Digest: Propagule pressure might not matter in the establishment of invasive prokaryotes. Evolution 74: 203–204.

Peterson GD, Cumming GS, Carpenter SR. 2003. Scenario planning: A tool for conservation in an uncertain world. Conservation Biology 17: 358–366.

Profitiliotis G, Loizidou M. 2019. Planetary protection issues of private endeavours in research, exploration, and human access to space: An environmental economics approach to forward contamination. Advances in Space Research 63: 598–605.

Puleo JR, Fields FD, Moore B, Graves RC. 1970. Microbial contamination associated with the Apollo 6 spacecraft during final assembly and testing. Space Life Sciences 2: 48–56.

Pyšek P, et al. 2020. Scientists' warning on invasive alien species. Biological Reviews 95: 1511–1534.

Reaser JK, Burgiel SW, Kirkey J, Brantley KA, Veatch SD, Burgos-Rodriguez J. 2020. The early detection of and rapid response (EDRR) to invasive species: A conceptual framework and federal capacities assessment. Biological Invasions 22: 1–19.

Ricciardi A, et al. 2017. Invasion science: A horizon scan of emerging challenges and opportunities. Trends in Ecology and Evolution 32: 464–474.

Ricciardi A, Hoopes MF, Marchetti MP, Lockwood JL. 2013. Progress toward understanding the ecological impacts of non-native species. Ecological Monographs 83: 263–282.

Ricciardi A, et al. 2021. Four priority areas to advance invasion science in the face of rapid environmental change. Environmental Reviews 29: 119–141.

Ricciardi A, MacIsaac HJ. 2011. Impacts of biological invasions on freshwater ecosystems. Pages 211–224 in Richardson DM, ed. Fifty Years of Invasion Ecology: The Legacy of Charles Elton. Wiley-Blackwell.

Ricciardi A, Palmer ME, Yan ND. 2011. Should biological invasions be managed as natural disasters? BioScience 61: 312–317.

Rothschild L. 2018. Myco-architecture Off Planet: Growing Surface Structures at Destination. NASA. www.nasa.gov/directorates/spacetech/niac/2018_Phase_I_Phase_II/Myco-architecture_off_planet.

Rummel JD, Pugel DE. 2019. Planetary protection technologies for planetary science instruments, spacecraft, and missions: Report of the NASA Planetary Protection Technology Definition Team (PPTDT). Life Sciences in Space Research 23: 60–68.

Russell JC, Meyer J-Y, Holmes ND, Pagad S. 2017. Invasive alien species on islands: Impacts, distribution, interactions and management. Environmental Conservation 44: 359–370.

Sánchez FJ, Mateo-Martíb E, Raggioc J, Meeßend J, Martínez-Fríasb J, Sanchoc LG, Ott S, de la Torrea R. 2012. The resistance of the lichen *Circinaria gyrosa* (nom. provis.) towards simulated Mars conditions: A model test for the survival capacity of an eukaryotic extremophile. Planetary and Space Science 72: 102–110.

Saul W-C, Jeschke JM. 2015. Eco-evolutionary experience in novel species interactions. Ecology Letters 18: 236–245.

Scheele BC, et al. 2019. Amphibian fungal panzootic causes catastrophic and ongoing loss of biodiversity. Science 363: 1459–1463.

Seebens H, et al. 2021. Projecting the continental accumulation of alien species through to 2050. Global Change Biol. 27: 970–982.

Seebens H, et al. 2017. No saturation in the accumulation of alien species worldwide. Nature Communications 8: 14435.

Shahar K, Greenbaum D. 2020. Lessons in space regulations from the lunar tardigrades of the Beresheet hard landing. Nature Astronomy 4: 208–209.

Sherpa S, Després L. 2021. The evolutionary dynamics of biological invasions: A multi-approach perspective. Ecological Applications 14: 1463–1484.

Sih A, Bolnick DI, Luttbeg B, Orrock JL, Peacor SD, Pintor LM, Preisser E, Rehage JS, Vonesh JR. 2010. Predator–prey naïveté, antipredator behavior, and the ecology of predator invasions. Oikos 119: 610–621.

Simberloff D. 2009. The role of propagule pressure in biological invasions. Annual Review of Ecology, Evolution, and Systematics 40: 81–102.

Sornette D. 2002. Predictability of catastrophic events: Material rupture, earthquakes, turbulence, financial crashes, and human birth. Proceedings of the National Academy of Sciences 99: 2522–2529.

Stigall AL. 2019. The invasion hierarchy: Ecological and evolutionary consequences of invasions in the fossil record. Annual Review of Ecology, Evolution, and Systematics 50: 355–380.

Stringham OC, Lockwood JL. 2021. Managing propagule pressure to prevent invasive species establishments: Propagule size, number, and risk–release curve. Ecological Applications 31: e02314.

Stukenbrock EH, Christiansen FB, Hansen TT, Dutheil JY, Schierup MH. 2012. Fusion of two divergent fungal individuals led to recent emergence of a unique widespread pathogen species. Proceedings of the National Academy of Sciences 109: 10954–10959.

Thakur MP, van der Putten WH, Cobben MMP, van Kleunen M, Geisen S. 2019. Microbial invasions in terrestrial ecosystems. Nature Reviews Microbiology 17: 621–631.

Tarnas JD, et al. 2021. Earth-like habitable environments in the subsurface of Mars. Astrobiology 21: 741–756.

Tirumalai MR, Fox GE. 2013. An ICEBs1-like element may be associated with the extreme radiation and desiccation resistance of *Bacillus pumilus* SAFR-032 spores. Extremophiles 17: 767–774.

Tirumalai MR, Karouia F, Tran Q, Stepanov VG, Bruce RJ, Ott M, Pierson DL, Fox GE. 2017. The adaptation of Escherichia coli cells grown in simulated microgravity for an extended period is both phenotypic and genomic. npj Microgravity 3: 15.

Tirumalai MR, Karouia F, Tran Q, Stepanov VG, Bruce RJ, Ott CM, Pierson DL, Fox GE. 2019. Evaluation of acquired antibiotic resistance in *Escherichia coli* exposed to long-term low-shear modeled microgravity and background antibiotic exposure. mBio 10: e02637–18.

Underwood EC, Klinger RC, Brooks ML. 2019. Effects of invasive plants on fire regimes and postfire vegetation diversity in an arid ecosystem. Ecology and Evolution 9: 12421–12435.

United Nations. 1967. Treaty on principles governing the activities of states in the exploration and use of outer space, including the moon and other celestial bodies. Article IX, U.N. Doc. A/RES/2222/(XXI).

Vaishampayan PA, Rabbow E, Horneck G, Venkateswaran KJ. 2012. Survival of *Bacillus pumilus* spores for a prolonged period of time in real space conditions. Astrobiology 12: 487–497.

Vaishampayan P, Roberts AH, Augustus A, Pukall R, Schumann P, Schwendner P, Mayilraj S, Salmassi T, Venkateswaran K. 2014. *Deinococcus phoenicis* sp. nov., an extreme ionizing radiation resistant bacterium isolated from the Phoenix Lander assembly facility. International Journal of Systematic and Evolutionary Microbiology 64: 3441–3446.

van der Putten WH, Klironomos JN, Wardle DA. 2007. Microbial ecology of biological invasions. ISME Journal 1: 28–37.

Viso M. 2019. Mars sample receiving facility or facilities? That is the question. Life Sciences in Space Research 23: 69–72.

Voight JR, Lee RW, Reft AJ, Bates AE. 2012. Scientific gear as a vector for non-native species at deep-sea hydrothermal vents. Conservation Biology 26: 938–942.

von Hegner I. 2020. Interplanetary transmissions of life in an evolutionary context. International Journal of Astrobiology 19: 335–348.

Weinzierl M. 2018. Space, the final economic frontier. Journal of Economic Perspectives 32: 173–192.

Whitney KD, Gabler CA. 2008. Rapid evolution in introduced species, 'invasive traits' and recipient communities: Challenges for predicting invasive potential. Diversity and Distributions 14: 569–580.

Xing J, Jia J, Wang H, Ma B, Falcão Salles J, Xu J. 2020. The legacy of bacterial invasions on soil native communities. Environmental Microbiology 23: 669–681.

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