# References (some quotations included to assist verification)

[Uses Harvard reference style, but in this draft, instead of a, b, c etc., I use unique ids like [(NASA, 2020tesgs](#kix.76qmy7dxcqdq)[)](#kix.6aisxl7zz0qc) - the idea is to search / replace these ids with a, b, c etc once the list is complete, after peer review]

## A

Abdo, J.M., Sopko, N.A. and Milner, S.M., 2020. [The applied anatomy of human skin: a model for regeneration](https://www.sciencedirect.com/science/article/pii/S2213909520300033#bib0020). Wound Medicine, 28, p.100179.

*Approximately every 28 days, fully differentiated cuboidal basal keratinocytes with large nuclei, abundant organelles, and a phospholipid membrane migrate apically from the basal layer through the spinous and granular layers [4]. During this turnover process, an accumulation of keratin and lipids ensues which then undergoes terminal differentiation to form the stratum corneum  
…  
Skin is an active immunological organ, and dysfunctional innate defenses have serious clinical implications. Products of the stratum corneum, including free fatty acids, polar lipids, and glycosphingolipids accumulate in the intercellular spaces and horny layer, exhibiting antimicrobial properties, and functioning as a first line of defense. Antimicrobial peptides (AMPs) exhibit potent and targeted resistance against a wide spectrum of common pathogens. When this barrier is breached, second lines of protection are provided by inflammatory cascades in the subepithelial tissue. Approximately sixteen AMPs have been shown to be expressed in the skin (Table 1)*

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New York: Garland Science; .[Cell Biology of Infection](https://www.ncbi.nlm.nih.gov/books/NBK26833/)

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Almeida, M.P., Parteli, E.J., Andrade, J.S. and Herrmann, H.J., 2008. [Giant saltation on Mars](https://www.pnas.org/content/pnas/105/17/6222.full.pdf). *Proceedings of the National Academy of Sciences*, *105*(17), pp.6222-6226.

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American Chemical Society, 2015, [“Cyborg bacteria outperform plants when turning sunlight into useful compounds”](https://phys.org/news/2017-08-cyborg-bacteria-outperform-sunlight-compounds.html), Phys.org

*"A future direction, if this phenomenon exists in nature, would be to bioprospect for these organisms and put them to use,"*

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Andrew, R.G., 2019, [NASA’s Curiosity Rover Finds Unexplained Oxygen on Mars](https://www.scientificamerican.com/article/nasas-curiosity-rover-finds-unexplained-oxygen-on-mars/), Scientific American

*On Earth, photosynthesis and respiration by living things cause tiny fluctuations in our planet’s otherwise steady oxygen concentration. We shouldn’t expect this on Mars, though. “That’s far out,” Telling says: Mars appears too inhospitable for a critical mass of life capable of sustaining either process. “It’s almost certainly going to be a nonbiological chemical reaction.”*

*Trainer herself does not rule out a biological explanation, but nevertheless underscores its unlikeliness. “People in the community like to say that it will be the explanation of last resort, because that would be so monumental,” she says. There are abiotic mechanisms aplenty, both known and unknown, to rule out first before leaping to any more sensational claims.*

Andrews, R.G., 2020, Rocks, Rockets and Robots: [The Plan to Bring Mars Down to Earth](https://www.scientificamerican.com/article/rocks-rockets-and-robots-the-plan-to-bring-mars-down-to-earth1/), Scientific American

*A single U.S. facility ticking all of these boxes could cost around $500 million, Dreier says. And it is not yet clear if others will be built in Europe*

*...*

*MSR’s masters are foregoing parachutes because the devices cannot be guaranteed to work, Vijendran says—something immortalized in 2004 by the solar-wind-particle-gathering Genesis mission, whose sample capsule broke open after an unintentional hard landing. In this case, it is simpler to build a rigid capsule that can withstand such a landing. “It just comes in, and, wham, it hits the ground,” Vago says. “That’s going to be an interesting one.”*

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## B

Bada, J.L., Aubrey, A.D., Grunthaner, F.J., Hecht, M., Quinn, R., Mathies, R., Zent, A. and Chalmers, J.H., 2009. [Seeking signs of life on Mars: In situ investigations as prerequisites to a sample return mission](http://mepag.jpl.nasa.gov/reports/decadal/JeffreyLBada_URS211530.pdf). Planetary science decadal survey White Paper, Scripps Institution of Oceanography, USA.

*"Two strategies have been suggested for seeking signs of life on Mars: The aggressive robotic pursuit of biosignatures with increasingly sophisticated instrumentation vs. the return of samples to Earth (MSR). While the former strategy, typified by the Mars Science Laboratory (MSL), has proven to be painfully expensive, the latter is likely to cripple all other activities within the Mars program, adversely impact the entire Planetary Science program, and discourage young researchers from entering the field."*

*"In this White Paper we argue that it is not yet time to start down the MSR path. We have by no means exhausted our quiver of tools, and we do not yet know enough to intelligently select samples for possible return. In the best possible scenario, advanced instrumentation would identify biomarkers and define for us the nature of potential sample to be returned. In the worst scenario, we would mortgage the exploration program to return an arbitrary sample that proves to be as ambiguous with respect to the search for life as ALH84001."*

Bada, J.L., Ehrenfreund, P., Grunthaner, F., Blaney, D., Coleman, M., Farrington, A., Yen, A., Mathies, R., Amudson, R., Quinn, R. and Zent, A., 2008. [Urey: Mars organic and oxidant detector](http://astrobiology.berkeley.edu/PDFs_articles/08UreySpaceSciRev.pdf). *Strategies of Life Detection*, pp.269-279.

Bahl, J., Lau, M.C., Smith, G.J., Vijaykrishna, D., Cary, S.C., Lacap, D.C., Lee, C.K., Papke, R.T., Warren-Rhodes, K.A., Wong, F.K. and McKay, C.P., 2011. [Ancient origins determine global biogeography of hot and cold desert cyanobacteria](https://www.nature.com/articles/ncomms1167). Nature communications, 2(1), pp.1-6.

Bak, E.N., Larsen, M.G., Jensen, S.K., Nørnberg, P., Moeller, R. and Finster, K., 2019. [Wind-driven saltation: an overlooked challenge for life on Mars.](https://www.researchgate.net/profile/Kai-Finster/publication/328837688_Wind-Driven_Saltation_An_Overlooked_Challenge_for_Life_on_Mars/links/5be5916fa6fdcc3a8dc8fc19/Wind-Driven-Saltation-An-Overlooked-Challenge-for-Life-on-Mars.pdf) Astrobiology, 19(4), pp.497-505.

*Spores in cavities will only be subjected to abrasion when the cavities crack open and the spores can get hit upon by a mineral particle. This process may be slow and explain the long tail of the number of surviving spores.The grain size of the regolith will likely affect the above-mentioned mechanisms and thus would have influence on the survival time of present microorganisms. We will address the effect of grain size in more detail in coming experiments.*

Bains, W. and Schulze-Makuch, D., 2016. [The cosmic zoo: the (near) inevitability of the evolution of complex, macroscopic life](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5041001/). Life, 6(3), p.25.

*Photosynthesis is primarily useful for providing energy for the reduction of environmental carbon ...*

*There are six known pathways for fixing carbon dioxide, of which the Calvin Cycle used in oxygenic phototrophs is the least efficient in terms of the energy and the reducing equivalents (electrons) required per mole of fixed CO₂ ...*

*However, the great advantage provided by oxygenesis was its capacity to liberate life from the need to find rare electron donors such as sulphide, hydrogen or Fe(II) to support the reduction of carbon dioxide, giving oxygenic photosynthesisers an advantage over all other forms of life ...*

*There are six known pathways for fixing atmospheric carbon, of which the Calvin Cycle used in oxygenic phototrophs is the least efficient in terms of the energy and the reducing equivalents (electrons)required per mole of fixed CO₂. Rubisco has a very low turnover for fixing carbon, and its carboxylase activity is compromised by opposing oxygenase activity that uses molecular oxygen to break down Ribulose-1,5-bisphosphate rather than fix CO₂ into it. Despite this, the first inventor of water-splitting was successful, and filled the niche ...*

*Oxygenesis evolved only once. There are two possible explanations for this. One is that it is a Random Walk process, requiring a sequence of unlikely evolutionary steps, which would not have evolved elsewhere. The hypotheses on the origins of oxygenesis above hint this may not be the case, but do not prove it. The other explanation is that the evolution of oxygenesis is a Many Paths process, one which has a high probability of occurring, but is also a Pulling Up the Ladder event, such that once oxygenesis evolved once that evolution removed the preconditions for its evolution again, in this case filling the niche of a photosynthesiser freed from limitation of an electron donor supply. The biochemistry of oxygenic photosynthesis points toward this second explanation.*

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Baqué, M., Napoli, A., Fagliarone, C., Moeller, R., de Vera, J.P. and Billi, D., 2020. [Carotenoid Raman Signatures Are Better Preserved in Dried Cells of the Desert Cyanobacterium Chroococcidiopsis than in Hydrated Counterparts after High-Dose Gamma Irradiation](https://www.mdpi.com/2075-1729/10/6/83/htm). *Life*, *10*(6), p.83.

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One such cycle, which is predicted to be two to three times faster than the Calvin–Benson cycle, employs the most effective carboxylating enzyme, phosphoenolpyruvate carboxylase, using the core of the naturally evolved C4 cycle. Although implementing such alternative cycles presents daunting challenges related to expression levels, activity, stability, localization, and regulation, we believe our findings suggest exciting avenues of exploration in the grand challenge of enhancing food and renewable fuel production via metabolic engineering and synthetic biology.

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Baylor University College of Medicine (BUCM), 1967, [Comprehensive Biological Protocol for the Lunar Sample Receiving Laboratory Manned Spacecraft Center](https://ntrs.nasa.gov/archive/nasa/casi.ntrs.nasa.gov/19680021536.pdf)

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Beaty, D.W., Grady, M.M., McSween, H.Y., Sefton‐Nash, E., Carrier, B.L., Altieri, F., Amelin, Y., Ammannito, E., Anand, M., Benning, L.G. and Bishop, J.L., 2019. [The potential science and engineering value of samples delivered to Earth by Mars sample return](https://onlinelibrary.wiley.com/doi/full/10.1111/maps.13232): International MSR Objectives and Samples Team (iMOST). *Meteoritics & Planetary Science*, *54*, pp.S3-S152.

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Benzerara, K., Skouri-Panet, F., Li, J., Férard, C., Gugger, M., Laurent, T., Couradeau, E., Ragon, M., Cosmidis, J., Menguy, N. and Margaret-Oliver, I., 2014. [Intracellular Ca-carbonate biomineralization is widespread in cyanobacteria](https://www.pnas.org/content/111/30/10933). *Proceedings of the National Academy of Sciences*, *111*(30), pp.10933-10938.

*Cyanobacteria are known to promote the precipitation of Ca-carbonate minerals by the photosynthetic uptake of inorganic carbon. This process has resulted in the formation of carbonate deposits and a fossil record of importance for deciphering the evolution of cyanobacteria and their impact on the global carbon cycle. Though the mechanisms of cyanobacterial calcification remain poorly understood, this process is invariably thought of as extracellular and the indirect by-product of metabolic activity. Here, we show that contrary to common belief, several cyanobacterial species perform Ca-carbonate biomineralization intracellularly.*

Berger, E.L., Zega, T.J., Keller, L.P. and Lauretta, D.S., 2011. [Evidence for aqueous activity on comet 81P/Wild 2 from sulfide mineral assemblages in Stardust samples and CI chondrites](https://meteoritegallery.com/wp-content/uploads/2014/04/Evidence-for-aqueous-activity-on-comet-81PWild-2-from-sul%EF%AC%81de-mineral-assemblages-in-Stardust-samples-and-CI-chondrites.pdf). Geochimica et Cosmochimica Acta, 75(12), pp.3501-3513. Press release from the University of Arizona: [Frozen Comet Had a Watery Past, UA Scientists Find](https://news.arizona.edu/story/frozen-comet-had-a-watery-past-ua-scientists-find)

Best, A. and Kwaik, Y.A., 2018. [Evolution of the arsenal of Legionella pneumophila effectors to modulate protist host](https://mbio.asm.org/content/9/5/e01313-18)s. *MBio*, *9*(5).

Bianciardi, G., Miller, J.D., Straat, P.A. and Levin, G.V., 2012. [Complexity analysis of the Viking labelled release experiments](http://central.oak.go.kr/repository/journal/11315/HGJHC0_2012_v13n1_14.pdf). International Journal of Aeronautical and Space Sciences, 13(1), pp.14-26.

Bilen, M., Dufour, J.C., Lagier, J.C., Cadoret, F., Daoud, Z., Dubourg, G. and Raoult, D., 2018. [The contribution of culturomics to the repertoire of isolated human bacterial and archaeal species](https://microbiomejournal.biomedcentral.com/articles/10.1186/s40168-018-0485-5#MOESM1). *Microbiome*, *6*(1), pp.1-11.

Biller, S.J., McDaniel, L.D., Breitbart, M., Rogers, E., Paul, J.H. and Chisholm, S.W., 2017. [Membrane vesicles in sea water: heterogeneous DNA content and implications for viral abundance estimates](https://www.nature.com/articles/ismej2016134). The ISME journal, 11(2), pp.394-404.

*These small, spherical, lipid membrane-bound structures typically range in size from ~20 to 200 nm diameter and provide a means for cells to interact with their environment over both spatial and temporal scales*

*Perhaps one of the most striking features of extracellular vesicles is that they can contain nucleic acids (Dorward et al., 1989; Valadi et al., 2007; Rumbo et al., 2011; Biller et al., 2014). DNA fragments of diverse sizes, ranging from hundreds of bp to >20 kb have been reported in vesicles from Gram-negative bacteria, Gram-positive bacteria, archaea and eukaryotes, and include genomic, plasmid and viral DNA (Dorward and Garon, 1990; Klieve et al., 2005; Soler et al., 2008; Biller et al., 2014; Gaudin et al., 2014; Jiang et al., 2014; Grande et al., 2015; Yáñez-Mó et al., 2015). As such, vesicles can function as vehicles of horizontal gene exchange (Yaron et al., 2000; Renelli et al., 2004; Klieve et al., 2005). Shotgun sequencing of vesicle-associated DNA from ocean samples has revealed sequences from diverse bacteria, archaea and eukaryotes (Biller et al., 2014), suggesting that vesicles could be an important mechanism mediating gene transfer among marine microbes.*

Billi, D., Staibano, C., Verseux, C., Fagliarone, C., Mosca, C., Baqué, M., Rabbow, E. and Rettberg, P., 2019a. [Dried biofilms of desert strains of Chroococcidiopsis survived prolonged exposure to space and Mars-like conditions in low Earth orbit](https://www.researchgate.net/profile/Cyprien-Verseux/publication/331027480_Dried_Biofilms_of_Desert_Strains_of_Chroococcidiopsis_Survived_Prolonged_Exposure_to_Space_and_Mars-like_Conditions_in_Low_Earth_Orbit/links/5ee9e56d299bf1faac5c948f/Dried-Biofilms-of-Desert-Strains-of-Chroococcidiopsis-Survived-Prolonged-Exposure-to-Space-and-Mars-like-Conditions-in-Low-Earth-Orbit.pdf). Astrobiology, 19(8), pp.1008-1017.

*Our results suggest that bacteria might indeed survive on Mars if shielded from UV, for instance by martian dust, since it is known that a few millimeters of soil is enough for UV protection (Mancinelli and Klovstad, 2000; Cockell and Raven, 2004). In view of the resistance of desert strain of Chroococcidiopsis to ionizing radiation (Billi et al., 2000; Verseux et al., 2017), the exposure in LEO to a total dose of 0.5 Gy of ionizing radiation did not affect biofilm survival. Hence, based on the dose of 76 mGy/year measured by the Curiosity rover at Gale Crater’s surface (Hassler et al., 2013), dried biofilms would survive on Mars more than half a decade. In addition, since the UV dose received in LEO corresponds to approximately 8 h under a Mars UV flux at the equator (Cockell et al., 2000), the speculated biofilm survival supports the possible dissemination of viable organisms. If carried, for instance, by winds at 5 m/sec (Gomez-Elvira et al., 2014) with the average flux mentioned above, they could travel more than 100km without dying. However, other factors found on Mars need to be taken into account so as to reduce the planetary protection risk, such as the presence of perchlorates that have been shown to be highly damaging to life (Wadsworth and Cockell, 2017)*

Billi, D., Verseux, C., Fagliarone, C., Napoli, A., Baqué, M. and de Vera, J.P., 2019b. [A desert cyanobacterium under simulated Mars-like conditions in low Earth orbit: implications for the habitability of Mars](https://www.researchgate.net/profile/Daniela-Billi/publication/331027480_Dried_Biofilms_of_Desert_Strains_of_Chroococcidiopsis_Survived_Prolonged_Exposure_to_Space_and_Mars-like_Conditions_in_Low_Earth_Orbit/links/5ca21364a6fdcc1ab5ba0613/Dried-Biofilms-of-Desert-Strains-of-Chroococcidiopsis-Survived-Prolonged-Exposure-to-Space-and-Mars-like-Conditions-in-Low-Earth-Orbit.pdf). *Astrobiology*, *19*(2), pp.158-169.

*In this experiment, survival of the Chroococcidiopsis strain occurred only with those cells that were mixed with martian regolith simulant and plated as thin layers (about 15–30 μm, corresponding to 4–5 cell layers).*

*… Our finding suggests that a putative microbial life-form at least as resistant to desiccation and radiation as the investigated desert cyanobacterium could withstand some exposure to UV on the martian surface.*

*… Our findings support the hypothesis that opportunistic colonization of protected niches on Mars, such as in fissures, cracks, and microcaves in rocks or soil, could have enabled life to remain viable while being transported to a new habitat*

Billings, L., 2015, Making Space for Everyone: [A Q&A with BoldlyGo's Jon Morse](https://www.scientificamerican.com/article/making-space-for-everyone-a-q-a-with-boldlygo-s-jon-morse), Scientific American

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*"Despite suggestions to the contrary, it is simply not possible, on the basis of current knowledge, to determine whether viable Martian life forms have already been delivered to Earth. Certainly in the modern era, there is no evidence for large-scale or other negative effects that are attributable to the frequent deliveries to Earth of essentially unaltered Martian rocks. However the possibility that such effects occurred in the distant past cannot be discounted.”*

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***“Revised Finding 3-1:*** *Cell division by Earth microbes has not been reported below –18°C (255K). The very low rate of metabolic reactions at low temperature result in doubling times ranging from several months to year(s). Current experiments have not been conducted on sufficiently long timescales to study extremely slow-growing microorganisms.”*

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*From the perspective of planetary protection, Conley is also concerned about terrestrial organisms that can absorb water from the air. She recalls fieldwork she did in the Atacama Desert in Chile, which is one of the driest places on Earth, with less than 0.04 inch of rain a year.*

*Even in this dessicated place, she found life: photosynthetic bacteria that had made a home in tiny chambers within halite salt crystals. There’s a small amount of water retained inside the halite and, at night, it cools down and condenses both on the walls of the chambers and on the surface of the organisms that are sitting there.*

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***Section 3.1 final para:*** *Simply scaling by the time between molecular collisions, corresponding to a 125 m diameter ice patch at φ = 0, we find at the base of the regolith a 160 m patch at φ = 26◦ (Aristarchus Plateau), 580 m at φ = 51◦.6 (Plato), 2.3 km at φ = 65◦ (10% polar cap), and an essentially divergent value, 522 km at φ = 82◦ (1% polar cap). If in fact the regolith layer is much deeper than suspected, the added depth of low diffusivity dust significantly increases the patch area: 170 m at φ = 26◦, 830 m at φ = 51◦.6, and 4 km at φ = 65◦.*

***Section 3.3:*** *In the Moon’s formation temperatures of proto-Earth and progenitor impactor material in simulations grow to thousands of Kelvins, sufficient to drive off the great majority of all volatiles, but these are not necessarily the only masses in the system. Either body might have been orbited by satellites containing appreciable volatiles, which would likely not be heated to a great degree and which would have had a significant probability of being incorporated into the final moon. Furthermore, there is recent discussion of significant water being delivered to Earth/Moon distances from the Sun in the minerals themselves (Lunine et al. 2007, Drake & Stimpfl 2007), and these remaining mineral-bound even at high temperatures up to 1000K (Stimpfl et al. 2007). The volume of surface water on Earth is at least 1.4 × 109 km3, so even if the specific abundance of lunar water is depleted to 10−6 terrestrial, one should still expect over 1010 tonnes endogenous to the Moon, and it is unclear that later differentiation would eliminate this. This residual quantity of water would be more than sufficient to concern us with the regolith seepage processes outlined above.*

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*The U.S. takes a different approach for filter classification of HEPA filters. The mother of all test procedures for these filters in the U.S. is MIL-STD-282, which was introduced in 1956. Other test procedures include e.g. IEST-RP-CC001 and IEST-RP-CC007. Each test procedure specifies certain particle sizes at which efficiency is evaluated. Depending on the filter class evaluated, this is done at 0.3 µm, 0.1 - 0.2 µm or 0.2 - 0.3 µm.*

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*“Only recently, with the reality of a Mars Sample Return project, have we started to revisit and think in depth about implementation of backwards planetary protection,” said Lisa Pratt, NASA’s planetary protection officer. The last time NASA seriously thought about backwards planetary protection, she noted at the MEPAG meeting, was during the Apollo program a half century ago.*

*...*

*A concept review and development milestone known as Key Decision Point (KDP) A are scheduled before the end of the current fiscal year.*

*“We’re not prepared to discuss that at this point in time,” Watzin said when asked at the MEPAG meeting for the cost of the overall program. “As we go forward into KDP-A, we’ll have to start talking about that. Towards the end of this fiscal year is when we’ll be ready to have that conversation.”*

Foust, J., 2021, [The multi-decade challenge of Mars Sample Return](https://spacenews.com/the-multi-decade-challenge-of-mars-sample-return/), Space News

*That schedule was too aggressive for the independent panel. “The schedules required to support launches in 2026 were substantially shorter than the actual experience from recent, somewhat similar programs,” like Mars 2020 and Curiosity, Thompson said.*

*Under a revised schedule recommended by the panel, the lander mission would launch in 2028. The orbiter could launch in either 2027 or 2028, since its use of electric propulsion gives it the flexibility to pursue alternative trajectories. That revised schedule would delay the return of the samples until 2033.*

*At the same time, the study warned about delaying the missions beyond 2028 [because of risk of landing before a global dust storm with solar power issues]*

*...*

*It also recommended looking at adding a radioisotope thermoelectric generator (RTG) to the lander, or at least the lander with the MAV, to ensure sufficient power and to keep the rocket’s propulsion system from getting too cold.*

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*Examples of such “molecular fossils” are 1.64 Ga old carotenoid derivatives (Lee and Brocks, 2011) and degradation products of chlorophylls and hemes (geoporphyrins; Callot and Ocampo, 2000) which have been reported, for example, from ∼500 Ma old oil shales (Serebrennikova and Mozzhelina, 1994). Hence, the extraordinary stability of certain molecular fossils opens the prospect of detecting chemical traces of life on other planets and moons even if it became extinct a long time ago.*

*It is highly unlikely that a natural abiotic process generates long chain molecules that have precisely defined lengths, ordered sequences, and homochiral building blocks. Therefore, proteins and nucleic acids can certainly be regarded as strong chemical biosignatures.*

*Low to moderate enantiomeric excesses, as they occur, for example, in meteoritic α,α-dialkyl amino acids (Pizzarello and Cronin, 2000), are definitely not indicative of a biological origin.*

*On the other hand, a lack of enantiopurity can be a false-negative result because the initial enantiopurity could have been lost by racemization, a process well-known for the proteinogenic L-amino acids (Bada and Schroeder, 1975; Bada, 1985). Furthermore, one should not discard the possibility that an extraterrestrial organism synthesizes both enantiomers. In fact, terrestrial bacteria produce diverse D-amino acids (e.g., D-Ala, D-Glu, D-Leu, D-Met, D-Phe, and D-Tyr) which have effects on the peptidoglycan of the cell wall, both directly by incorporation into the polymer and indirectly by regulating enzymes that synthesize and modify peptidoglycan (Höltje, 1998; Lam et al., 2009). Another intriguing example from terrestrial life is the simultaneous presence of L- and D-isovaline in some fungal peptides (Degenkolb et al., 2007).*

*No natural non-biological processes that generate them have been observed in nature, but there are some indications that, at least in rare instances, natural abiotic compounds might be enantiopure. For example, there is a single case where, under laboratory conditions, a small enantiomeric excess of an amino acid was amplified to near enantiopurity (>99%; Klussmann et al., 2006). This amino acid was serine under solid–liquid equilibrium conditions in water at the eutectic point. However, for all other amino acids tested, enantiopurity was not achieved. Also, this mechanism will not work with chiral compounds that crystallize as conglomerates (i.e., mixtures of pure L and pure D crystals). Because of the special conditions and compounds necessary, it is unclear if this physical process is relevant to the generation of enantiopurity (i.e., enantiomeric excesses near 100%) in extraterrestrial environments.*

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*"The method originates from the USA, and is used to remove paint from aircraft fuselage. A powerful jet of frozen carbon dioxide (CO₂) crystals, about the size of a rice kernel, blasts the paint right off the metal. The researchers made this crude instrument substantially more refined. Instead of CO₂ pellets, they use carbon dioxide snow to work on each individual component – from the highly sophisticated aluminum workbench to the ring washers. Here’s the rub: the beam that the jet emits is additionally accelerated with a blast of CDA (clean dry air) that encases it. This is how it penetrates into every nook and cranny, removing even the minuscule pollutant. As soon as the tiny snowflakes hit the relatively hot surface, they become gaseous, causing their volume to explosively expand 800-fold. The detonation pressure completely sweeps away every single bit of dust, even fingerprints which the cold gas had just turned brittle. “This approach involves a dry process that does not warp surfaces. When cleaning, these can be gently treated with CO₂. That makes it unnecessary to apply heat or chemicals,” Gommel says when explaining the advantages of this method.* "

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*“RNA is the key to the ribosome, which is what makes proteins. There’s almost no question that RNA, which is a molecule involved in catalysis, arose before proteins arose,” Benner explains. The difficulty is that for RNA to assemble into long strands–which is needed for genetics – you can’t have the assembly taking place in water. “Most people think that water is essential for life. Very few people understand how corrosive water is,” Benner says. For RNA, water is extremely corrosive – bonds cannot be made within water, preventing long-strands from forming.*

*However, Benner says that these paradoxes can be resolved with the help of two very important groups of minerals. The first are borate minerals. Borate minerals–which contain the element boron–prevent life’s building blocks from devolving into tar if incorporated into organic compounds. Boron, as an element, is seeking electrons to make itself stable. It finds these in oxygen, and together the oxygen and boron form the mineral borate. But if the oxygen boron finds is already bonded to carbohydrates, the carbohydrates linked with boron form a complex organic molecule dotted with borate that’s less resistant to decomposition.*

*The second group of minerals that come into play involve those that contain molybdate, a compound that consists of molybdenum and oxygen. Molybdenum, more famous for its conspiratorial relation to the Douglas Adams classic A Hitchhiker’s Guide to the Galaxy than for its other properties, is crucial, because it takes the carbohydrates that borate stabilized, bonds to them and catalyzes a reaction which rearranges them into ribose: the R in RNA.*

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(by visual count, I make it 7 not operational yet out of 59 mapped)

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*Fortunately, cells make a variety of antioxidant enzymes to fight the dangerous side-effects of life with oxygen. Two important players are superoxide dismutase, which converts superoxide radicals into hydrogen peroxide, and catalase, which converts hydrogen peroxide into water and oxygen gas. The importance of these enzymes is demonstrated by their prevalence, ranging from about 0.1% of the protein in an Escherichia coli cell to upwards of a quarter of the protein in susceptible cell types. These many catalase molecules patrol the cell, counteracting the steady production of hydrogen peroxide and keeping it at a safe level.*

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*Soils from the hyper-arid core of the Atacama Desert have cell numbers and culturable counts similar to University Valley permafrost (*[*Supplementary Table S3*](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4918446/#sup1)*), but small, viable microbial communities are activated and detected when Atacama soils are wetted (*[*Navarro-González et al., 2003*](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4918446/#bib22)*;* [*Crits-Christoph et al., 2013*](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4918446/#bib6)*). Our results suggest that microorganisms in the University Valley permafrost soils analysed here are not exposed to sufficiently long and frequent clement conditions to allow for metabolism or growth. Instead, our results suggest that a fundamental threshold may be crossed in some University Valley permafrost soils, where the combination of permanently subfreezing temperatures, low water activity, oligotrophy and age are severely constraining the evolution of functional cold-adapted organisms*

*Very low microbial biomass was found by direct microscopic cell counts (1.4−5.7 × 10^3 cells per g soil) in both the dry and ice-cemented permafrost using DTAF stain as described by Steven et al. (2008). Comparatively, 2 orders of magnitude higher cell counts (1.2−4.5 × 10^5 cells per g soil) were detected in the active layer and permafrost soils from the Antarctic Peninsula.*

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...

*The disadvantages of removing a sample from its environment prior to analysis revolve around changes that might occur because the sample is no longer in thermal or redox equilibrium with its surroundings.*

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Administrative discretion is necessary to weight these factors on a case-by-case basis. No magic risk number can substitute for informed and thoughtful consideration by accountable officials who work with the public to make balanced decisions.

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*Improved instrumentation on rovers that might detect and identify a diversity of potential in situ biosignatures, including ancient organic molecular biosignatures, designed with the ability to differentiate biotic and abiotic signals in micro- or macrostructures. Instrumentation could also be better attuned to the unique complications of biosignature preservation on Mars (e.g., deeper drilling to access potentially better preserved organics)*

*The fluorescence spectrometers on SHERLOC can detect condensed carbon and aromatic organics by deep UV-induced fluorescence, and SHERLOC's Raman spectrometer will allow classification of aromatic and aliphatic organics. Raman spectrometry can also be used to detect minerals relevant to aqueous chemistry. While these measurements would allow us to identify reduced carbon compounds, there may not be sufficient structural information to distinguish between a biological signal and extraterrestrial organic input.*

*A major knowledge gap that will directly impact our ability to choose an appropriate landing site is what terrestrial analog environments might look like—what the biosignature signals might be—if photosynthetic microorganisms had not evolved and instead the environments were only inhabited by chemosynthetic microorganisms*

*4.4. Strategies and priorities*

*In many of the environments discussed, there is a dichotomy between habitability and preservation—many of the conditions that make an environment more habitable are destructive to one or more of the biosignatures of interest. For example, fluid flow in the subsurface of hydrothermal environments helps create the redox gradients that support communities that inhabit the outflow channel. Fluids are also essential for lithification and the associated decrease in permeability essential for long-term preservation. Preservation is enhanced by rapid burial and mineral precipitation that encases and lithifies biological materials in less permeable matrices—in these cases, silica from hydrothermal environments, or silica-enriched aqueous environments, is an important material for preservation. However, these same fluids can degrade biosignatures such as mineralogy, chemistry, and micro- and macrostructures. One strategy for astrobiological exploration has to be to seek out a “sweet spot” where these two balance each other so that long-term preservation is possible. This sweet spot may occur as conditions change through time.*

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*As shown in Table 2, when a sample containing 100,000 spores was analyzed, either Bleach Rite® or 10% bleach was able to dramatically reduce (<0.0001% remaining) the number of viable spores at the earliest time point, and no viable spores were detected after 20 minutes of treatment. Complete sterilization was not attained until 20 minutes post-inoculation due to 1 cfu being present at 10 minutes in the 10% bleach-treated groups.*

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With consideration of upcoming Mars-targeted missions, we conclude that gas collected in a newly designed and purpose-built valved sample-tube sized vessel, which could be flown on either SFR or SRL, would be considered of higher priority than either the head space gas or a sealed M2020 sample tube. Conceptually, this vessel would require no more physical space to return than a sealed empty sample tube and alleviate concerns about the manufacturing and history of a non-purpose-built vessel, and the valving would provide a more robust mechanism for sealing the vessel and testing the seal upon return.

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*“What are the mechanisms by which HGT occurs? Currently these include: transduction, a process whereby a viral capsule is used to transfer genetic material from one cell to another; conjugation, a process exhibited by microbes during which a plasmid or a small piece of a plasmid from one donor cell is transferred to another recipient cell (Prof. Matxalen Llosa—see summary report); transformation, which occurs when a competent cell takes up a “naked” strands of nucleic acid from the environment—such strands of nucleic acids may not necessarily have been exuded by living entities (e.g., mitochondrion genes transferred to eukaryote chromosomes), they could also be from recently dead cells, as well as from long extinct organisms; gene transfer agents (GTA), which are bacteriophage-like particles containing random cellular genomic segments intended for transduction to another living recipient cell; and membrane vesicle transfer (MVT), in which small membrane sacs emanating from the surface of a cell contain genetic material for transfer to another living recipient cell.”*

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*Its distinct impacts on the spectral signature of our planet are, most significantly, oxygen in the atmosphere and the surface reflectance spectrum of land plants. The latter is notable not only for a "green bump" but also a "red edge", the steep contrast between absorbance by chlorophyll in the red and high reflectance of plant leaves in the near-infrared (NIR). However, purple bacteria perform photosynthesis with NIR radiation and produce no oxygen, and lichens do not have a strong red edge. Scientists still puzzle over why plants are green, because it seems this wastes the light where our Sun produces the most energy.*

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*The tube voltage is 50 kV and the electron beam current is 200 μA in the calculation.*

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*At the Spirit landing site, half the year is dominated by dust deposition, the other half by dust removal, usually in brief, sharp events. At the Opportunity landing site the Martian year has a semiannual dust cycle with dust removal happening gradually throughout two removal seasons each year.*

*On Spirit there is a yearly pattern with steady dust deposition throughout roughly the colder half year from late southern summer to late southern winter, which encompasses the Martian aphelion, and overall dust removal during the warmer and windier perihelion season from late southern winter to late southern summer.*

*On Opportunity ... the overall variation between highs and lows is smaller, and there are two periods of overall dust deposition and two periods of overall dust removal every year. The deposited dust thickness peaks once in the middle of the northern hemisphere spring. This peak recurs very regularly 6 times. ... There is also a peak roughly in the middle of the southern spring. This peak is clear in the first year, but the pattern becomes more irregular later in the mission and is entirely absent in the last year.*

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**LM:** Pamela and I disagree on this, but there’s a provision in the Outer Space Treaty, Article VI, which says that each country must supervise and authorize the activities of its nongovernmental entities. This is not a self-executing provision, and the U.S. Supreme Court has held that a non-self-executing treaty is not domestically enforceable. ...

**PM:** I disagree with Laura on this. Article VI of the Outer Space Treaty provides that all state parties to the treaty are responsible for their activities in outer space, whether they’re carried out by government agencies or private companies. Countries are required to subject private companies within their jurisdiction that engage in space activities to an authorization requirement and continuing supervision. So, the United States is responsible for compliance with the Outer Space Treaty by our private companies or entities that go into space.

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*The resistance of D. radiodurans is not exclusive to radiation and desiccation but extends also to many toxic chemicals and conditions. Therefore, Dra is called a polyextremophile, a robust “generalist,” to be distinguished from specialized extremophiles with an evolutionary redesign of their proteome (e.g., proteins purified from thermophiles are thermostable in vitro). Unlike specialized extremophiles, Dra does not thrive on extreme conditions—indeed, it does not grow while desiccated or when heavily* irradiated—but it can reproduce under standard growth conditions after recovering from damage inflicted by chronic moderate, or acute intense, exposures to cytotoxic conditions.

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been a substantial connection between its import and the occasion of  
my visit to you November 6, 1957….*

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*Hydrated sulfates may form through notable acid weathering of dust and sand inside the ice deposit when volcanic aerosols dissolve in the thin films of water to create acidic solutions (*[*36*](https://www.science.org/doi/10.1126/sciadv.abn8555#core-R36)*); however, this process has difficulty explaining the duricrust features. Therefore, one scenario that we prefer is that the predepositional regolith underwent cementation and lithification during the rising or infiltration of briny groundwater to form the observed platy rocks (*[*Fig. 5*](https://www.science.org/doi/10.1126/sciadv.abn8555#F5)*). The salt cements (e.g., sulfates or opaline silica) precipitate from the groundwater in the capillary fringe zone, where active evaporation and accumulation can occur (*[*37*](https://www.science.org/doi/10.1126/sciadv.abn8555#core-R37)*). Episodic fluctuation of the groundwater table may further thicken the indurated section and result in a fine-layered structure. After evaporation, the regolith overlying the duricrust is subject to deflation and erosion, while the duricrusts are resistant to aeolian erosion (*[*38*](https://www.science.org/doi/10.1126/sciadv.abn8555#core-R38)*). In this scenario, kilometer-scale briny groundwater may have been episodically active and interacting with the colluvium at the landing site. Alternatively, aqueous minerals such as hydrated silica have been observed to be associated with flow features and pitted cones elsewhere in the northern plains (*[*12*](https://www.science.org/doi/10.1126/sciadv.abn8555#core-R12)*), and the observed mineralogy and duricrust in this work may have some generic link with the pitted cones in the vicinity of the rover (*[*Fig. 1*](https://www.science.org/doi/10.1126/sciadv.abn8555#F1)*), which requires further investigation by the Tianwen-1 orbiter and Zhurong rover*

*The hydrated minerals and widespread salt cementations imply the presence of briny liquid water in the subsurface, which may have been generated by melting the ground ice during temporary climate perturbations (e.g., volcanism and impacts).*

*Specifically, possible dike swarms responsible for landform formation or recent volcanism from the Elysium region could have been a heat source for maintaining the groundwater system or melting the ice. Alternatively, local transient liquid water under current climate condition may be responsible for local melting of subsurface ground ice, forming indurated duricrust, in which case the water-rock interaction and the spatial extent would be limited.*

*Determining the mineralogy and spatial extent of the platy rocks in future traverse would provide clues to distinguish different climate conditions for these water activities. Regardless of the potential heat source, the in situ observations manifest recent aqueous activities on Mars, suggesting that the cold and dry late Amazonian epoch may have been episodically punctuated by short-duration climatic warming events that result in melting of ground ice at latitude less than 30°N. The in situ identification of such environments points to a more active Amazonian surface hydrosphere for Mars than previously considered.*

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*“We have discovered the substance calcium perchlorate in the soil and, under the right conditions, it absorbs water vapour from the atmosphere. Our measurements from the Curiosity rover’s weather monitoring station show that these conditions exist at night and just after sunrise in the winter. Based on measurements of humidity and the temperature at a height of 1.6 meters and at the surface of the planet, we can estimate the amount of water that is absorbed. When night falls, some of the water vapour in the atmosphere condenses on the planet surface as frost, but calcium perchlorate is very absorbent and it forms a brine with the water, so the freezing point is lowered and the frost can turn into a liquid. The soil is porous, so what we are seeing is that the water seeps down through the soil. Over time, other salts may also dissolve in the soil and now that they are liquid, they can move and precipitate elsewhere under the surface,” explains Morten Bo Madsen, associate professor and head of the Mars Group at the Niels Bohr Institute at the University of Copenhagen.*

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*page 419: Category 1: Small and medium-sized meteoroids (with radii from 2 to 80 cm and masses from 0.1 kg to~6:5 tons (if 𝜌≈3g/cm3). These meteoroids provide no shielding against the galactic cosmic rays, on the contrary they increase the dose rates caused by unshielded GCR by the creation of more particles in-side the meteoroids. However, they could still serve as vehicles for viable transfers from Mars to Earth lasting 1 million years* for D. radiodurans R1 and 0.3 million years for B. subtilis (wild type) if DNA decay is not limiting.

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*A temperature-regulated change in CO2 solubility could at least partially account for the amplitude of the LR oscillation. However, the HT oscillation phase leads the LR oscillation by as much as two hours, an unusual circumstance if this were simply a chemical oscillation driven by thermal fluctuation.*

*(Admittedly there is uncertainty concerning the delay between change in temperature at the head end assembly, perhaps one inch over the 0.5 cc soil sample, and soil sample temperature per se. However, a two-hour lag seems quite long for what is presumably a convective and radiative process. Similarly, thermal-induced movement of gas between the soil sample and the beta detector requires only about 20 minutes.)*

*Furthermore, the LR oscillation does not slavishly follow the thermal variation; rather, it seems that the LR rhythm is extracted from the HT oscillation, while high frequency noise is not. This is very common in terrestrial organisms in which a low frequency periodic stimulus (i.e., a zeitgeber) such as a 12:12 light/dark cycle can entrain a circadian rhythm, while high frequency transients in the same stimulus are ignored (e.g., turning on the light in the bathroom at night for a minute or two does not alter normal entrainment to the light/dark cycle).*

*Furthermore, there is abundant evidence that as little as a 2º C temperature cycle can entrain circadian rhythms in terrestrial organisms such as lizards, fruit flies, and bread molds and entrainment can be preferential to the diminution phase of the temperature cycle, in analogy to the temperature fall that occurs at sunset on Mars).*

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*After 7 years of air-drying, Chroococcidiopsis not only avoided genome degradation but preserved at least a sub-set of mRNAs and 16S ribosomal RNA.*

*... In the present work, the occurrence of survivors in dried biofilms and dried-UV-irradiated biofilms was proved by growth after transfer into liquid BG-11 medium (not shown) and by INT reduction after 72 h of rewetting.*

*Reshaping the boundaries of Chroococcidiopsis desiccation and UV tolerance has implications in the search for extra-terrestrial life since it contributes to defining the habitability of Mars and planets orbiting other stars. In fact, the UV dose used here corresponds to that of a few hours at Mars’s equator (Cockell et al., 2000). Hence, considering that survivors occurred in the bottom layers of the biofilms (Baqué et al., 2013), it might be hypothesized that if a biofilm life form ever appeared during Mars’s climatic history, it might have been transported in a dried state under UV radiation, from niches that had become unfavorable to niches that were inhabitable (Westall et al., 2013). The reported survival also suggests that intense UV radiation fluxes would not prevent the presence of phototrophic biofilms or their colonizing of the landmass of other planets.*

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*D-1: NASA and ESA should replan the baseline MSR program for SRL and ERO launches in 2028, with the potential of a 2027 ERO launch continuing to be studied for feasibility and potential benefits.*

*NASA Response: NASA partially concurs with this recommendation. The MSR team will continue to examine the 2026, 2027 and 2028 launch opportunities during Phase A, while working to maintain current schedules to mature the design and retire risk as quickly as possible during Phase A, while also working to minimize program impacts due to COVID.*

*C-3:This study should be augmented to include a strong focus on potential Radioisotope Thermoelectric Generator(RTG)incorporation on either a single-lander or two-lander approach, to achieve the following benefits:Type1 launch option in 2028 Possible longer surface timeline RTG-sourced heating of the MAV   
NASA Response: NASA concurs with this recommendation*

NASA, 2020tesgs, [The Extraordinary Sample-Gathering System of NASA's Perseverance Mars Rover](https://mars.nasa.gov/news/8682/the-extraordinary-sample-gathering-system-of-nasas-perseverance-mars-rover/).

NASA, 2021nmttm, [NASA Moves to the Next Phase in a Campaign to Return Mars Samples to Earth](https://scitechdaily.com/nasa-moves-to-the-next-phase-in-a-campaign-to-return-mars-samples-to-earth/), SciTechDaily

NASA, 2021mpb, Marscopter press briefings.

Marscopter altitude: **Bob Balaram** says looking at 10 meters, limited by the range of the laser altimeter. MiMi Aung says 600 to 700 meters.

[1:01:32](https://youtu.be/JM_2hmdRnfQ?t=3692) from: [After NASA's Historic First Flight: Ingenuity Mars Helicopter Update Streamed live on 19 Apr 2021](https://youtu.be/JM_2hmdRnfQ)

Maximum separation: **MiMi Aung:** "The vehicles can be apart up to a kilometer or further ... The signal to noise ratio is extremely good. We can go beyond 1 kilometer distance."

[33:41](https://www.youtube.com/watch?v=BAlXe-U0ws4&t=2021) from: [NASA’s Ingenuity Mars Helicopter’s Next Steps (Media Briefing) Streamed live on 30 Apr 2021](https://youtu.be/BAlXe-U0ws4)

Distance for a single flight: **Bob Balaram:** “I think a total of 600 meters is not unreasonable. 2 minutes flight at 5 meters per second is a possibility. That’s probably where we'll see how well it does, and if there is more margin we can use for the flights. That's probably a good place to think of the limit.”

[1:01:20](https://youtu.be/BAlXe-U0ws4?t=3680) from: [NASA’s Ingenuity Mars Helicopter’s Next Steps (Media Briefing) Streamed live on 30 Apr 2021](https://youtu.be/BAlXe-U0ws4)

NASA, 2021prmtl, [Perseverance Rover, Mission Timeline › Landing](https://mars.nasa.gov/mars2020/timeline/landing/)

NASA, 2021wnpr, Watch NASA’s Perseverance Rover Land on Mars, Thomas Zurbukin at [14:45](https://youtu.be/gm0b_ijaYMQ?t=885)

***Macy Ragsdale:*** *Is anything alive on Mars?*

***Thomas Zurbuchen (NASA associate administrator):*** *That's a question i ask myself, is anything alive there, and frankly at the surface where we're going right now with Perseverance we do not believe there's anything alive right there, because of the radiation that's there, it's chilling cold and there's really no water there. But guess what we think that three billion years ago this looked like a stream that you may see on earth and frankly a lot more similar than Earth but water with a magnetic field just like the earth with an atmosphere and the question is at that time three billion years ago were there single cell organisms just off the type that developed on earth so is there life on on Mars overall we don't know but where we're going right now we're really looking for ancient life and that's what we're so excited about.*

NASA, 2022mpfs, [Fact Sheet Proposed Action](https://downloads.regulations.gov/NASA-2022-0002-0002/attachment_5.pdf), [MSR PEIS Fact Sheets](https://www.regulations.gov/document/NASA-2022-0002-0002)

NASA, 2022msr, [public comments](https://www.regulations.gov/document/NASA-2022-0002-0001), MSR, PEIS

NASA, 2022smsr [The Safety of Mars Sample Return](https://downloads.regulations.gov/NASA-2022-0002-0002/attachment_7.pdf), [MSR PEIS Fact Sheets](https://www.regulations.gov/document/NASA-2022-0002-0002)

*Such a Mars sample receiving facility would have design and sample handling requirements equivalent to those of biological safety laboratories used for research studies of infectious diseases. The well-established safety protocols and engineering controls used to isolate hazardous biological materials in such laboratories address issues that are very similar to those involved in Mars sample return. At this time, there are several options under study for implementing a Mars sample receiving facility.*

NASA, 2022nepa, [National Environmental Policy Act; Mars Sample Return Campaign](https://downloads.regulations.gov/NASA-2022-0002-0001/content.pdf) Federal Register / Vol. 87, No. 73 / Friday, April 15, 2022 / Notices

*The general scientific consensus is that the Martian surface is too inhospitable for life to survive there today. It is a freezing landscape with no liquid water that is continually bombarded with harsh radiation.*

*Scientists are interested in returning samples that may reveal what the Martian environment was like billions of years ago, when the planet was wetter and may have supported microbial life.*

*There is no current evidence that the samples collected by the Mars 2020 mission from the first few inches of the Martian surface could contain microorganisms that would be harmful to Earth’s environment.*

*Nevertheless, out of an abundance of caution and in accordance with NASA policy and regulations, NASA would implement measures to ensure that the Mars samples are contained (with redundant layers of containment) so that they could not impact humans or Earth’s environment, and the samples would remain contained until they are examined and confirmed safe for distribution to terrestrial science laboratories. NASA and its partners would use many of the basic principles that biological laboratories use today to contain, handle, and study materials that are known or suspected to be dangerous.*

NASA, 2022nic, [NASA Invites Comment on Initial Plans for Mars Sample Return Program](https://www.nasa.gov/press-release/nasa-invites-comment-on-initial-plans-for-mars-sample-return-program)

*NASA will consider all comments received during the scoping process in the subsequent development of the MSR Draft Environmental Impact Statement, which is currently scheduled to be released for public comment later this year.*

NASA, 2022wip, [*"Where is Perseverance?"*](https://mars.nasa.gov/mars2020/mission/where-is-the-rover/). Mars 2020 Mission Perseverance Rover

NASA, n.d.ame, [Atmosphere](https://marsed.asu.edu/mep/atmosphere), Mars Education

NASA, n.d.cls, [Curiosity's Landing Site: Gale Crater](https://mars.nasa.gov/msl/timeline/prelaunch/gale-crater/)

NASA, n.d.cm, [Chris McKay, at NASA Ames](https://www.nasa.gov/content/chris-mckay/)

NASA, n.d.dan, [Dynamic Albedo of Neutrons (DAN)](https://mars.nasa.gov/msl/spacecraft/instruments/dan/), see also archived page for scientists: [Dynamic Albedo of Neutrons (DAN)](https://web.archive.org/web/20210224030220/https://mars.nasa.gov/msl/spacecraft/instruments/dan/for-scientists/)

NASA, n.d.ecilm, [Eugene Cernan in Lunar Module](https://www.nasa.gov/content/images-of-astronaut-gene-cernan)

NASA, n.d.hsp, [Health Stabilization Program](https://www.nasa.gov/sites/default/files/atoms/files/health_stabilization_program_technical_brief_ochmo_021020.pdf)

NASA, n.d. mbtn, [Mars, by the numbers](https://solarsystem.nasa.gov/planets/mars/by-the-numbers/) surface area 144,371,391km2. This seems to be based on the volumetric mean radius of 3389.5 kilometers as 4\*pi\*3389.5^2. See [NASA n.d. Mars Fact Sheet](https://nssdc.gsfc.nasa.gov/planetary/factsheet/marsfact.html)/ Since a sphere has the minimum surface area to volume ratio of any spheroid then the Martian area is at least this much.

NASA, n.d.mfs, [Mars Fact sheet](https://nssdc.gsfc.nasa.gov/planetary/factsheet/marsfact.html)

NASA, n.d.monm, [Map of NASA's Mars Landing Sites](https://mars.nasa.gov/resources/24729/map-of-nasas-mars-landing-sites/)

NASA, n.d.MOXIE [MOXIE](https://mars.nasa.gov/mars2020/spacecraft/instruments/moxie/), and [MOXIE for scientists](https://mars.nasa.gov/mars2020/spacecraft/instruments/moxie/for-scientists/).

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NASA, n.d. PRLS, [Perseverance Rover's Landing Site: Jezero Crater](https://mars.nasa.gov/mars2020/mission/science/landing-site/), accessed at <https://mars.nasa.gov/mars2020/mission/science/landing-site/>, accessed on 17 July 2020.

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NASA, n.d.,SEH,, [System Engineering Handbook](https://www.nasa.gov/seh/index.html), see particularly

2.5 [Cost Effectiveness considerations](https://www.nasa.gov/seh/2-5-cost-effectiveness-considerations#Fig2-5-1https://www.nasa.gov/seh/2-5-cost-effectiveness-considerations)

3.3 [Project Pre-Phase A: Concept Studies](https://www.nasa.gov/seh/3-3-project-pre-phase-a-concept-studies)

3.4 [Project Phase A: Concept and Technology Development](https://www.nasa.gov/seh/3-4-project-phase-a-concept-and-technology-development)

3.5 [Project Phase B: Preliminary Design and Technology Completion](https://www.nasa.gov/seh/3-5-project-phase-b-preliminary-design-and-technology-completion)

NASA, n.d.WiC, [Curiosity: Mission: Where is the rover?](https://mars.nasa.gov/msl/mission/where-is-the-rover/)

Curiosity landing site: 137.44°E, 4.589°S

NASA, n.d.WiP, [Where is Perseverance?](https://mars.nasa.gov/mars2020/mission/where-is-the-rover/)

Perseverance landing site: 18.45°N 77.45°E,

NASA, n.d. WISO, [What is Surface Operations?](https://mars.nasa.gov/mars2020/timeline/surface-operations/)

*drills core samples from about 30 promising rock and “soil” (regolith) targets and caches them on the Martian surface (Objective C)*

Naseem, M., Osmanoglu, Ö. and Dandekar, T., 2020. [Synthetic Rewiring of Plant CO₂ Sequestration Galvanizes Plant Biomass Production](https://www.sciencedirect.com/science/article/pii/S0167779919303142). *Trends in Biotechnology*, *38*(4), pp.354-359.

*The CETCH cycle requires less energy to operate than other aerobic CO₂ -fixation pathways. One limitation of CETCH is the production of glyoxylate, a less active metabolic intermediate that requires acetyl-CoA (AcCoA) or propanoyl-CoA [*[*3*](https://www.sciencedirect.com/science/article/pii/S0167779919303142#bb0015)*] for conversion into other metabolites. Also, glyoxylate is not well connected to other metabolic pathways. Despite functional impediments associated with any synthetically designed pathway, CETCH is the most efficient artificial cycle that fixes (in vitro) several-fold more CO₂ than does the natural CBB. The incorporation of CETCH-based enoyl-CoA carboxylase/reductases (ECRs) should be an excellent alternative to the native Calvin cycle. It can sequester approximately 80 CO₂ molecules per second (in vitro) compared with RuBisCO, which fixes two to five CO₂ molecules per second in plants.*

National Research Council. 2009. [Assessment of Planetary Protection Requirements for Mars Sample Return Missions (Report)](http://www.nap.edu/openbook.php?record_id=12576&page=28). p. 59.

"*It has been estimated that the planning, design, site selection, environmental reviews, approvals, construction, commissioning, and pre-testing of a proposed SRF will occur 7 to 10 years before actual operations begin. In addition, 5 to 6 years will likely be required for refinement and maturation of SRF-associated technologies for safely containing and handling samples to avoid contamination and to further develop and refine biohazard-test protocols. Many of the capabilities and technologies will either be entirely new or will be required to meet the unusual challenges of integration into an overall (end-to-end) Mars sample return program.*"

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Nealson, K.H., Inagaki, F. and Takai, K., 2005. [Hydrogen-driven subsurface lithoautotrophic microbial ecosystems (SLiMEs): do they exist and why should we care?](http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.1078.4608&rep=rep1&type=pdf). *Trends in microbiology*, *13*(9), pp.405-410.

Negi, S., Perrine, Z., Friedland, N., Kumar, A., Tokutsu, R., Minagawa, J., Berg, H., Barry, A.N., Govindjee, G. and Sayre, R., 2020. [Light regulation of light‐harvesting antenna size substantially enhances photosynthetic efficiency and biomass yield in green algae](https://lodgbot.com/wp-content/uploads/2020/05/13May20-The-Plant-Journal-Algae-photosynthesis-improvement.pdf). *The Plant Journal*.

*page 15: The NC-77 transgenic line, however, had a three-fold increase in bio-mass yield compared with wild-type. This increased bio-mass production in NC transgenics with adjustable light harvesting antenna sizes, however, raises the question why have algae and plants evolved large, less effi-cient, fixed light-harvesting antenna systems that oversaturate downstream electron transfer processes during most (80%) of the day. In mixed species environments, the abil-ity to shade or reduce the light available to competing spe-cies may offer a selective advantage, because limiting light availability to other species would reduce their growth rates and presumably their fitness (Zhuet al., 2008; Ortet al., 2015). Species competing for light are clearly impacted by shading as plant canopies close or as algal cultures reach high cell densities. Thus, having large light-harvesting antenna systems may reduce light availability for competitors and enhance fitness for plants or algae thatshade competitors as is the case in high-density algal cul-tures. In addition, plants living lower in the canopy or algae growing deeper in the water column often experi-ence very low light conditions.*

*Having a large light-harvesting antenna would allow photosynthesis and growth at light intensities that could not support the growth of algae with smaller antenna sizes optimized for growth at higher light intensities. In fact, algae that grow at extreme depths in the oceans have among the largest light-harvesting antenna sizes known in photosynthetic organisms (Yamazakiet al., 2005).*

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Niles, P.B., Catling, D.C., Berger, G., Chassefière, E., Ehlmann, B.L., Michalski, J.R., Morris, R., Ruff, S.W. and Sutter, B., 2013. [Geochemistry of carbonates on Mars: implications for climate history and nature of aqueous environments](http://faculty.washington.edu/dcatling/Niles2012_CarbonatesOnMarsReview.pdf). Space Science Reviews, 174(1), pp.301-328.

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A critical nutrient to the expansion of both subsurface and surface life on any planet is the availability of nitrogen as an aqueous species. On Earth, microorganisms evolved the ability to fix N2 into ammonia with the development of nitrogenase to overcome this constraint. Nitrogenases, Nif proteins, are complex enzymes, utilizing iron, molybdenum, and/or vanadium, that exist in both bacterial and archaeal domains. Phylogenetic comparison of genes that comprise nitrogenases and a complement of proteins required for their regulation indicate that nitrogenases emerged in anoxic sulfidic environments on Earth within obligate anaerobic thermophilic methanogens and were transferred to obligate anaerobic clostridia (Boyd et al., 2015), both common subsurface microorganisms. As Nif proteins were adopted first by the aerobic diazotrophic lineage Actinobacteria and then by the more recently evolved aerobic Proteobacterial and Cyanobacterial lineages, the Nif protein suite became more complex to protect the core MoFe-bearing proteins from O2 (Boyd et al., 2015). Although it is not clear whether the emergence of the more complex protein occurred prior to or after the Great Oxidation Event, it is certain that the ancestral protein emerged in an anoxic environment when the demands for aqueous nitrogen species exceeded the abiotic supply. The implications for martian ecosystems are that nitrogenase would have also likely emerged within an anaerobic subsurface environment, not in the oxic surface environment.

Experiments on the effects of low pN2 on diazotrophic nitrogen-fixing soil bacteria have shown that they could grow in N2 partial pressures of 5 mbar but not 1 mbar (Klingler et al., 1989). This result suggests that further experiments on wild-type species are required to determine whether the evolution of pN2 in the martian atmosphere was a significant deterrent to the expansion of early life, especially after Mars lost most of its atmosphere. Analyses of the nitrogen budget and of nitrogen cycling from deep subsurface environments in South Africa indicate that the pN2 is higher at depth than on the surface, that most of this N2 originates from the rock formations through nitrogen cycling, and that N2 is being actively fixed in the subsurface by microbial communities (Silver et al., 2012; Lau et al., 2016b). Given the presence of a cryosphere barrier to diffusion on Mars, the nitrogen availability and perhaps even the pN2 of subsurface brines are likely to be higher there than on the martian surface.

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*Meter-sized Fe(II)-rich carbonate/iron oxide concretions (Fig. 4) are found in Jurassic sandstone deposits of southwest Colorado that were formed at hundreds of meters' depth between 2 and 0.5 Ma as the Colorado River Basin was uplifted (McBride et al., 2003; Loope et al., 2010). Similar-sized ferroan calcite and siderite concretions occur in Late Paleocene/Early Eocene Wasatch Group sandstones, and siderite nodule-bearing cores from the formation (Lorenz et al., 1996) yielded thermophilic Fe(III)-reducing bacteria that were capable of producing prodigious quantities of siderite (Roh et al., 2002). In subaqueous systems unconstrained by rock matrix, authigenic carbonate mounds at CH4 and hydrocarbon seeps, formed from carbon mobilized by methane- and alkane-oxidizing microorganisms (Greinert et al., 2001; Formolo et al., 2004; Ussler and Paull, 2008), can be hundreds of meters tall and more than a kilometer wide (Klaucke et al., 2008).*

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* ***A Case for Caution*** by John Rummel, NASA'S planetary protection officer at the time, and previously, NASA senior scientist for Astrobiology
* ***Hazardous Until Proven Otherwise***, by Margaret Race, a biologist working on planetary protection and Mars sample return for the SETI Institute and specialist in environment impact analysis
* ***Practical Safe Science*** by Kenneth Nealson, Director of the Center of Life Detection at NASA's JPL at the time.

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*Pages 94-5: Questions about the adequacy of the SRF to maintain the new life form must also be addressed, including the possible need to add equipment, change operations, review emergency plans, or upgrade the facilities because of what has been found.*

*Concerns about security should also be reconsidered, especially in view of the potential disruptive activities of any terrorists or ‘radical’ groups that may be opposed to sample return. The advisability of allowing distribution of untested sample material outside the SRF2684* may need to be reconsidered, as well.

*Plans should be developed well in advance in order to avoid a frenzied, reactive mode of communications between government officials, the scientific community, the mass media, and the public. Any plan that is developed should avoid a NASA-centric focus by including linkages with other government agencies, international partners, and external organizations, as appropriate. It will also be advisable to anticipate the kinds of questions the public might ask, and to disclose information early and often to address their concerns, whether scientific or non-scientific.  
...*

*Evaluations of the proposal should be conducted both internal and external to NASA and Centre National d’Etudes Spatiale (CNES) and the space research communities in the nations participating in the mission. An ethical review should be conducted at least at the level of the Agencies participating and these reviews made public early in the process (in France, the national bioethics committee, Comité Consultatif National d'Ethique pour les Sciences de la Vie et de la Santé, CCNE, is the appropriate organization). The final protocol should be announced broadly to the scientific community with a request for comments and input from scientific societies and other interested organizations. Broad acceptance at both lay public and scientific levels is essential to the overall success of this research effort.*

In the long term, the discovery of extraterrestrial life, whether extant or extinct, in situ or within returned sample materials, will also have implications beyond science and the SRF per se. Such a discovery would likely trigger a review of sample return missions, and plans for both robotic and human missions. Legal questions could arise about ownership of the data, or of the entity itself, potentially compounded by differences in laws between the United States and the countries of international partners. In any event, ethical, legal and social issues should be considered seriously. Expertise in these areas should be reflected in the membership on appropriate oversight committee(s).

Page 101: **Communications** Unusual or unprecedented scientific activities are often subject to extreme scrutiny at both the scientific and political levels. Therefore, a communication plan must be developed as early as possible to ensure timely, and accurate dissemination of information to the public about the sample return mission, and to address concerns and perceptions about associated risks. The communication plan should be pro-active and designed in a manner that allows the public and stakeholders to participate in an open, honest dialogue about all phases of the mission with NASA, policy makers, and international partners. Risk management and planetary protection information should be balanced with education/outreach from the scientific perspective about the anticipated benefits and uncertainties associated with Mars exploration and sample return.

The communication plan should also address how the public and scientific community will be informed of results and findings during Life Detection and Biohazard testing, including the potential discovery of extraterrestrial life. Because of the intense interest likely during initial sample receipt, containment, and testing, procedures and criteria should be developed in advance for determining when and how observations or data may be designated as “results suitable for formal announcement.” Details about the release of SRF information, the management of the communication plan, and its relationship to the overall communications effort of the international Mars exploration program should be decided well in advance of the implementation of this protocol.

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Rutkin, A., 2014. [X-ray medicine blasts off to space](https://www.newscientist.com/article/2004086-first-medical-x-ray-scanner-heads-for-space-station/). *New scientist*, (2974), p.14.

## S

Sagan, C, 1961. [Organic matter and the Moon](https://www.nap.edu/download/18476)., National Academy of Sciences.

[*Page 23*](https://www.nap.edu/read/18476/chapter/5#23)*: It is remarkable that the depth at which surviving lunar organic matter is expected to be localized (section II) is just the depth at which temperatures appear to be optimum for familiar organisms (section IV). At such temperatures and depths, some moisture should be expected, arising from meteoritic and organic bound water. Watson, Murray and Brown (1961) have recently pointed out that ice could have been retained on permanently shaded areas of the Moon. These circumstances provide all the survival requirements of many terrestrial organisms - water and their metabolites, appropriate temperature, and negligible radiation. That autochthons evolving with the changing environment could also survive under these conditions is far from inconceivable.*

Sagan, C., Levinthal, E.C. and Lederberg, J., 1968. [Contamination of Mars](https://profiles.nlm.nih.gov/ps/access/BBABJH.ocr). *Science*, *159*(3820), pp.1191-1196.

*"The prominent dust storms and high wind velocities previously referred to imply that aerial transport of contaminants will occur on Mars. While it is probably true that a single unshielded terrestrial microorganism on the Martian surface ... would rapidly be enervated and killed by the ultraviolet flux, ... The Martian surface material certainly contains a substantial fraction of ferric oxides, which are extremely strongly absorbing in the near ultraviolet. ... A terrestrial microorganism imbedded in such a particle can be shielded from ultraviolet light and still be transported about the planet."*

*…*

*"A single terrestrial microorganism reproducing as slowly as once a month on Mars would, in the absence of other ecological limitations, result in less than a decade in a microbial population of the Martian soil comparable to that of the Earth's. This is an example of heuristic interest only, but it does indicate that the errors in problems of planetary contamination may be extremely serious."*

Sagan, C., 1973, [*The Cosmic Connection - an Extraterrestrial Perspective*](https://www.e-reading.life/bookreader.php/148581/Sagan_-_The_Cosmic_Connection___An_Extraterrestrial_Perspective.pdf)

*I reach this conclusion reluctantly. I, myself, would love to be involved in the first manned expedition to Mars. But an exhaustive program of unmanned biological exploration of Mars is necessary first****. The likelihood that such pathogens exist is probably small, but we cannot take even a small risk with a billion lives.*** *Nevertheless, I believe that people will be treading the Martian surface near the beginning of the twenty-first century.*

Sagan, C., 1977. [Reducing greenhouses and the temperature history of Earth and Mars](https://www.nature.com/articles/269224a0). *Nature*, *269*(5625), pp.224-226.

Sagan, C., 1980., *Cosmos: The Story of Cosmic Evolution, Science and Civilisation*

full quote:

*The surface area of Mars is exactly as large as the land area of the Earth. A thorough reconnaissance will clearly occupy us for centuries. But there will be a time when Mars is all explored; a time after robot aircraft have mapped it from aloft, a time after rovers have combed the surface, a time after samples have been returned safely to Earth, a time after human beings have walked the sands of Mars. What then? What shall we do with Mars?*

*There are so many examples of human misuse of the Earth that even phrasing this question chills me. If there is life on Mars, I believe we should do nothing with Mars. Mars then belongs to the Martians, even if the Martians are only microbes. The existence of an independent biology on a nearby planet is a treasure beyond assessing, and the preservation of that life must, I think, supersede any other possible use of Mars.*

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See Table 1, final row, delta v 1230.6 m/s. This patent is based on the rescue mission for the HGS-1 geostationary satellite using a lunar flyby described in [(Ocampo, 2005)](#kix.dhfxkcwvvr1s)

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*While isolating psychrophilic strains would likely provide a better analog for the Martian surface, the generation times are prohibitively slow for research purposes in such exploratory experiments*

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*page 37: Normal wild-type algae have large chlorophyll-bindingLHCII antenna systems and consequently the culture is dark green. Cell lines with small LHCII antenna systems yield cultures which are a much lighter green at the same cell density (Fig.7a). In the wild-type case, algal cells at the illuminated surface of the bioreactor that are exposed to high light levels capture the bulk of the light, but waste upto∼90% of the energy as fluorescence and heat [122,134].*

*As a result the wild-type cells located deeper in the culture are exposed to ever decreasing levels of light the further they are from the illuminated surface (see“Open PondSystems”section). These shaded cells are prevented from capturing enough solar energy to drive photosynthesis efficiently. This in turn drastically reduces the efficiency of the overall culture.In contrast, small antenna cell lines with reduced LHCIIlevels have the advantage that they improve the light penetration into the bioreactor (Fig.7a) and better match itto the energy requirements of each photosynthesizing cell. Thus small antenna cells at the bioreactor surface absorb only the light that they need, largely eliminating fluores-cence of excess energy. This in turn allows more light (i.e.the light wasted in wild-type as fluorescence and heat) to penetrate into the bioreactor so that even cells deeper in the culture have a near optimal exposure to light*

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*HEPA Filtration is the “Best Available Control Technology” at 99.99 percent at 0.3-micron efficiency level and is “Generally Accepted Control Technology” at 99.97 percent at 0.1-micron efficiency level. The added feature of the new 0.1-micron advanced filters is the “gel” seal and micro fiberglass construction that allows combining these filters with UV light disinfection. HEPA filters combined with charcoal and prefilters are the highest approved filters available for NIOSH-certified respirators.*

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*… Legionella pneumophila enters mononuclear phagocytes by depositing complement C3b on its surfaces and using that host protein to serve as a ligand for binding to macrophage cell surfaces. After ingestion, the bacteria remain in vacuoles that do not fuse with lysosomes, apparently due to the influence of soluble substances produced by the bacteria.*

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*Indeed, not even all infectious human pathogens—let alone non-infectious pathogens— on Earth require a multicellular, macroscopic host to evolve harmful capabilities.*

*July, 1976, the month that VL1 [Viking Lander 1] landed on theMartian surface, was also the month of the outbreak of Legionnaires’ disease at the American Legion convention in Philadelphia.*

*The cause, Legionella pneumophila, is a facultative, Gram-negative rod that is one of several human pathogens now known to be carried in the intracellular environments of protozoan hosts. L. pneumophila can also persist, even outside of any host, as part of biofilms.*

*In essence, all that a potentially infectious human pathogen needs to emerge and persist is to grow and live naturally under conditions that are similar to those that it might later encounter in a human host. On Mars, these conditions might be met in a particular niche within the extracellular environment of a biofilm, or within the intracellular environment of another single-celled Martian organism. It is important to note the numerous biofilms observed aboard the Mir space station, which were found on surfaces and within water plumbing. These films were often multi-species and included bacteria, fungi, and protozoa.*

*To be sure, the genetic similarity between humans and protozoa is much greater than could be expected between humans and the Martian host of a Martian microbe.*

*However, the L. pneumophila example does bring into question the rationale of the need for host-pathogen coevolution. Even in the context of a planetary bio-sphere that is limited to single-celled life, and even where there is unlikely to have been a co-evolution between agent and host organism, the possibility of infectious agents, even an invasive type, cannot be ruled out.*

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