One of the central, long-standing goals of NASA that holds promise for both major scientific discoveries and for exciting the general public is to understand life in space. The main strategy towards achieving this goal has been to search for life beyond earth. This is a long-term approach that might yield results through direct and long-term observations and explorations extending to Mars, the moons of Jupiter and Saturn and even beyond our solar system.

To pursue a better understanding of life in space, and link it to future missions we propose an additional strategy aimed at determining the potential for terrestrial microbial life to adapt and evolve in space environments. Although this idea is not new, it has not been pursued systematically. Consequently, relatively little is known currently about the ability of microorganisms to survive and thrive in space, where the conditions are distinctly different from those on earth. An understanding of life processes at the extremes and the accompanying molecular adaptations is a prerequisite for developing predictions and hypotheses about life on other worlds. Defining the limits of terrestrial life in space will also provide a theoretical foundation for developing strategies to search for life beyond earth. If microorganisms showed the ability to adapt to and survive in a wide range of conditions encountered in space it would indicate that terrestrial life might not be a local planetary phenomenon, but instead could expand its
evolutionary trajectory beyond its planet of origin. This would further imply that terrestrial life may not be unique and similar life forms might exist elsewhere in the Universe.

The proposed strategy fills the void in the current planetary science portfolio at NASA by combining rigorous, astrobiology-oriented research that exploits revolutionary progress in modern molecular and cellular biology with space missions involving living organisms. The absence of biologically oriented missions in previous decadal plans has been their weakness that can and should be remedied. The proposed agenda is timely because many relevant biological tools and exploration capabilities have been developed recently or are in development. For these reasons, considerable progress can be achieved in the next decade at only moderate costs.

A logical starting point for this strategy is to focus on key groups of terrestrial microbes that are of great evolutionary significance and have demonstrated tenacity and versatility of life on earth. By these credentials, that cyanobacteria naturally come to mind. Representatives of the phylum cyanobacteria, survive and flourish in an amazing range of hostile environments on earth. Their global distribution is, at least in part, the reflection of their ability, as a group, to cope with wide fluctuations in temperature, nutrient and light levels. The fossil record and other biogeochemical evidence indicate that cyanobacteria have existed in microbial communities for at least three billion years and are the pioneers of oxygenic photosynthesis on earth. During this period they have adapted to survive major changes in climate and atmosphere involving geo-chemical events with the attendant changes in global temperature, irradiance including high UV dosages and desiccation. The evolutionary history of cyanobacteria and their global distribution suggest that these organisms are important for understanding life and its adaptive capabilities at molecular, biochemical and physiological level.
Cyanobacteria are currently the targets of wide-ranging research efforts on earth focused on astrobiology, ecology, physiology and biotechnology. Because they are subjects of conjectures regarding their capabilities in space environments (including panspermia and terraforming), some work has also been done in short-term flight, or ground-based space simulation studies. However, ground-based experiments, no matter how well designed, cannot reproduce all the features of space environments. What is imminently required is a well-designed research and exploration program that best informs us about cyanobacterial survival, potential for adaptation and evolution in space-like environments. What is even more critical is to assess the effects of combinations of stress factors, such as temperature and desiccation, which we know much about from earth studies, in conjunction with “space-unique” stressors, such as microgravity, cosmic radiation and non-terrestrial fluxes in photosynthetically active radiation.

Cyanobacteria are discussed here as an example rather than the only group of microorganisms that are of interest for space studies. Other phyla, such as proteobacteria might also be of considerable value as model microorganisms, and in fact some of them have already been studied in space environments. Most technologies that have been and need to be developed for space based experiments, such as support on small satellite vehicles, devices for measuring gene expression in space, other sensors and a number of ground-based molecular biology and bioinformatics tools are applicable to most microbial models. This means that technological constraints are not a barrier to investigating different microbial taxa in space. Another systems of great interest are consortia, especially that they might be able to survive in space environments better than pure cultures. It should be, however, recognized that analyzing and interpreting results of studies on bacterial communities might be considerably more difficult than for a single organism.

To achieve the highest scientific return, the research agenda for studying terrestrial life in space should be well coordinated with NASA's plans for space
exploration and take advantage of synergy with various missions and payload launch opportunities. An essential component of this agenda are small satellite missions, which provide frequent, low cost access to space that is required for meaningful microbiological studies. In this approach we build on GeneSat and PharmaSat missions, in which microorganisms were successfully flown in space and the upcoming OREOS mission, which is aimed at monitoring the negative effects of space on biology and will be the first demonstration flight of the ASTID small payloads initiative. A number of tools for biological research in space are in development, including a number of sensors and a fully automated, miniaturized, integrated fluidic system for *in situ* measurements of gene expression in bacterial cultures. Once deployed, these tools will yield high value data, orders of magnitude richer than what is currently available.

As the long-term goal we envision establishing a lunar research laboratory that would be compatible with NASA’s strategy for human and/or robotic exploration of the Moon. The lunar platform will provide the necessary components to assess with full scientific rigor potential of microorganisms for NASA missions and astrobiology program. These components are necessarily limited in other space venues. The lunar platform will also provide the volume needed to conduct highly reproducible studies, and the ability to study microbial adaptation to the space environments. Further, it will present the opportunity to conduct studies, including gradual “training”, on adaptation of microorganisms to survive under specific stress conditions, such as radiation regimes, low levels of different nutrients, low pressures and high/low pH values. Such studies would not only yield a systematic evaluation of limits for life in space but could also serve as Mars simulation program until biological experiments on Mars are cleared by planetary protection agreements. Finally, the lunar laboratory would allow for evaluating the potential of using microorganisms for life support and *in situ* resource utilization in future exploration of the Moon.
To accomplish the goals of the proposed initiative, we recommend establishing a vigorous research and exploration program at NASA. The program should contain (but not necessarily be limited to) three main components:

1. **Ground-based research.** This research should be clearly aimed at (a) elucidating molecular, physiological and ecological responses to a range of conditions potentially encountered in space, using a range of available and in-development tools (b) helping identify organisms best suited to survive extreme conditions, (c) re-engineering them for improved resistance to space environments, and (d) evaluating factors related to long-term survivability of bacterial *communities* under conditions in space.

2. **Small satellite missions.** This component should be aimed at (a) the development of miniaturized, autonomous, reusable and cost-effective technologies for measuring survival, growth, metabolic behavior and genetic adaptations of microorganisms to conditions in space and (b) collecting data on the effects of space-induced stress factors on microorganisms in short and medium-length (up to 12 months) flights. The results of small satellite missions will provide essential information for further ground-based research and, conversely, ground-based studies will help to define high value mission experiments.

3. **Lunar laboratory for biological research.** In the next decade, this component will be aimed at developing research plans and biological and space technologies for the laboratory that are aligned with NASA’s strategies for the exploration of the Moon. The objectives of the laboratory will be to (a) determine long term survival and adaptation of microorganisms to conditions in space, (b) develop and apply *in situ* resource utilization and life support technologies involving microorganisms, (c) investigate survival of microbes in simulated Martian environments.
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