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The Role of Synthetic Biology for *In Situ* Resource Utilization (ISRU)

Michael Montague,[1] George H. McArthur IV,[2] Charles S. Cockell,[3] Jason Held,[4] William Marshall,[5]
Louis A. Sherman,[6] Norman Wang,[7] Wayne L. Nicholson,[8] Daniel R. Tarjan,[9] and John Cumbers[10]

Abstract

A persistent presence in space can either be supported from Earth or generate the required resources for human survival from material already present in space, so called "*in situ* material." Likely, many of these resources such as water or oxygen can best be liberated from *in situ* material by conventional physical and chemical processes. However, there is one critical resource required for human life that can only be produced in quantity by biological processes: high-protein food. Here, recent data concerning the materials available on the Moon and common asteroid types is reviewed with regard to the necessary materials to support the production of food from material *in situ* to those environments. These materials and their suitability as feedstock for the biological production of food are reviewed in a broad and general way such that terminology that is often a barrier to understanding such material by interdisciplinary readers is avoided. The waste products available as *in situ* materials for feasibility studies on the International Space Station are also briefly discussed. The conclusion is that food production in space environments from *in situ* material proven to exist there is quite feasible. Key Words: Astrobiology—Asteroid—Biomineralogy—Cyanobacteria. Astrobiology 12, 1135–1142.

A SINGLE FACT has constrained space exploration since the launch of Sputnik:

$$\begin{array}{c} \text{1 kg of equipment} \\ \text{or consumables to} \\ \text{be used in space} \end{array} = \begin{array}{l} \text{1kg of equipment or consumables} \\ \text{that must be launched from the} \\ \text{Earth into space} \end{array}$$

There are three ways to address this fact. One can partially address it from the left side of the equation by miniaturizing components and using paper-thin, ultralight materials. One can partially address it from the right side of the equation by decreasing the cost of launching material into space. There are organizations, both public and private, that are pursuing both of these approaches. However, the only way to completely address this fact is to change the $=$ sign in the center of the equation to a $\neq$ sign. This can only be done if some or all of the equipment or consumables we use in space come from material that originates in space rather than on Earth. This approach is known as *In Situ* Resource Utilization (ISRU).

On October 31, 2010, NASA Ames hosted a workshop on synthetic biology. One working group in that workshop was tasked with identifying opportunities for using the tools of biology for the use of *in situ* resources in space. This report is the result of the discussions, research, and analysis of that working group.

Strategies for ISRU depend heavily upon the location in which they are to be implemented. Thus, three locations, based upon potential near-term targets of exploration, were considered: the Moon, near-Earth objects (NEOs, which are mostly asteroids), and the International Space Station (ISS).

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[1] Department of Synthetic Biology, The J. Craig Venter Institute, Rockville, Maryland, USA.
[2] Department of Chemical and Life Science Engineering, Virginia Commonwealth University, Richmond, Virginia, USA.
[3] Geomicrobiology Research Group, PSSRI, Open University, Milton Keynes, UK.
[4] Saber Astronautics, Manly, New South Wales, Australia.
[5] NASA Ames Research Center, Moffett Field, California, USA.
[6] Department of Biological Sciences, Purdue University, West Lafayette, Indiana, USA.
[7] Department of Molecular Biosciences & Bioengineering, University of Hawaii-Manoa, Honolulu, Hawaii, USA.
[8] Department of Microbiology & Cell Science, University of Florida, Space Life Sciences Lab, Kennedy Space Center, Florida, USA.
[9] E.O. Lawrence Berkeley National Laboratory, Berkeley, California, USA.
[10] Department of Molecular Biology, Cell Biology and Biochemistry, Brown University, Providence, Rhode Island, USA, and University of California Santa Cruz, University Affiliated Research Center, Synthetic Biology Program, NASA Ames Research Center, Moffett Field, California, USA.

Mars is not addressed in this report due to the fact that any mission to Mars that might meaningfully use martian resources is many decades in the future and that the parameters of martian ISRU are significantly different from the above three cases. Lastly, Mars presents unique issues of planetary protection (concern of contaminating the martian environment with exogenous organisms) that are largely not present in the other locations considered. For these reasons, we feel that the cases for and against martian ISRU are deserving of a completely separate analysis.

Near-Term Prospects for Biological* In Situ *Resource Utilization

1. Resources Available on the Moon

1.1. Water and volatiles on the Moon

It has been suspected since Apollo that volatiles of helium and hydrogen are present on, or around, the lunar surface (Hodges, 1973). Evidence was suggested from the Clementine mission (Nozette *et al.,* 1994) and the Moon Mineralogy Mapper (Pieters *et al.,* 2009). The Lunar Crater Observation and Sensing Satellite or LCROSS mission provided direct evidence by crashing an empty rocket stage into one of the permanently shadowed craters on the south pole of the Moon (Schultz *et al.,* 2010). The source of the water on the Moon is thought to have arrived there from impact events from primitive outer solar system bodies (Papike *et al.,* 1982). Parts of the lunar surface act as cold traps with subsurface temperatures estimated at 38 K (Paige *et al.,* 2010). These cold traps at the permanently shadowed craters were targeted by LCROSS, as it was thought they were capable of trapping water ice as well as other volatiles. The spectroscopy of the plume thrown up by the crash indicated not only water but a nitrogen source (ammonia) and carbon dioxide. The light hydrocarbons methane, methanol, and ethylene also were present in small amounts, as were the sulfur-bearing species hydrogen sulfide and sulfur dioxide. The maximum total water vapor and ice in the instrument's field of view was 155 ± 12 kg, estimating the total water content at 5.6 ± 2.9% by mass (Colaprete *et al.,* 2010). Table 1 shows the range of other compounds identified from the spectral analysis relative to the amount of water that was found and by total mass in the regolith that was ejected from the impact.

1.2. Composition of lunar regolith

The lunar regolith consists of a layer of rocks, dust, and particles a number of meters thick, which rest on solid bedrock. During the Moon's lifetime, the repeated processes of comminution (breaking up of rock) and agglutinate formation (aggregation of the particles again) have created the present-day regolith. A number of samples have been collected via Apollo missions and have since been analyzed (Papike *et al.,* 1982). The contents of the particles are dependent on size; the $<10\,\mu$m fraction is enriched in Al_2O_3, CaO, Na_2O, K_2O, light rare earth elements, and thorium but depleted in MgO, FeO, MnO, and scandium. The regolith in the highlands contains higher amounts of aluminum and silica as opposed to the regolith in the maria (the darker flats of the Moon) that are rich in iron and magnesium (Papike *et al.,* 1982). An average lunar regolith composition is shown in Table 2. This is also the composition chosen for the JSC-1 lunar regolith simulant that has been used in a number of ISRU studies (McKay *et al.,* 1993).

Table 1. Compounds Identified by Spectroscopy in the LCROSS Ejecta Plume (Colaprete *et al.,* 2010)

Compound	*Chemical name*	*% Relative to H_2O*	*% by total mass*
H_2O	Water	100	5.60
H_2S	Hydrogen sulfide	16.75	0.94
NH_3	Ammonia	6.03	0.34
SO_2	Sulfur dioxide	3.19	0.18
C_2H_4	Ethylene	3.12	0.17
CO_2	Carbon dioxide	2.17	0.12
CH_3OH	Methanol	1.55	0.09
CH_4	Methane	0.65	0.04
OH	Hydroxide	0.03	0.002

1.3. Useful products that might be manufactured on the Moon

There are a wide variety of products that could be derived from lunar *in situ* resources via biological mechanisms. These products include rocket propellants such as the monopropellants hydrogen peroxide and monomethylhydrazine, or a number of chemicals that can function as rocket fuel when combined with an oxidizer such as hydrogen, ethane, methane, kerosene, or methanol. Other sorts of chemicals that might be useful include binders to stabilize loose regolith, adhesives, or lubricants. However, many of these products could be produced in an easier and less complex manner with the use of chemical and physical techniques. There is, however, one category of material that can only be produced by living systems, in quantity, safely, and economically: food. While these other products might one day also be produced by biological systems, no extended program of human exploration or settlement in space can ever take place if food cannot be produced from locally derived mass.

Estimates for the consumption of food by a crew in space have been made based on nutritional and caloric requirements (Allen *et al.,* 2003). The amount of dry food mass required per person per day was initially estimated at 674 g with certain assumptions of food preparation. However, this static daily rate of consumption does not take into account the probability that different crew members will have

Table 2. Average Composition of Lunar Regolith (Papike *et al.,* 1982; McKay *et al.,* 1993)

Chemical symbol	*Chemical name*	*Lunar soil composition wt %*
SiO_2	Silicon dioxide	47.3
Al_2O_3	Aluminum oxide	17.8
CaO	Calcium oxide	11.4
FeO	Iron(II) oxide	10.5
MgO	Magnesium oxide	9.6
TiO_2	Titanium dioxide	1.6
Na_2O	Sodium peroxide	0.7
K_2O	Potassium oxide	0.6
Cr_2O_3	Chromium(III) oxide	0.2
MnO	Manganese(II) oxide	0.1

different dietary needs. For example, food consumption predictions for the ISS crew have been confounded by factors such as variable tastes, moods, and workloads (McAlary, 2005). An additional study was able to estimate the requirement function variation due to the crew's workload and mood (Held *et al.*, 2007), suggesting that additional food mass might mitigate the risk, bringing the total required per day to 682 g.

1.4. Ways in which known lunar resources can be turned into useful products

The dry lunar regolith does not offer biology much in the way of resources. As Table 2 shows, the composition of regolith is mainly oxidized metals, 47.3% of which is silicon dioxide. Previous studies have proposed using biology for ISRU on the Moon (Johansson, 1995; Brown *et al.*, 2008). A recent NASA workshop (Dalton and Roberto, 2008) concluded that ISRU of lunar regolith would not be a feasible approach until enough oxygen and carbon dioxide were available as waste products from a human presence. However, the LCROSS mission has since changed the view of the Moon. Table 1 shows numerous compounds that could be used as feedstocks for biology. All the main elements required for life (C, H, N, O, S, K, Ca), except phosphorus, were identified in substantial quantities in the top 10 compounds by mass from the LCROSS spectra. Phosphorus, however, has already been found in lunar regolith in a fraction referred to as KREEP (Potassium, Rare Earth Elements, and Phosphorus) (Warren and Wasson, 1979). As organisms are primarily made of carbon, the sources of carbon should be noted in particular. The percent total masses of carbon-containing molecules are C_2H_4 (ethylene) 0.17%, CO_2 (carbon dioxide) 0.12%, CH_3OH (methanol) 0.09%, and CH_4 (methane) 0.04%. When summed, these carbon-bearing molecules in the LCROSS ejecta plume represent 0.4% total by mass.

Due to the earlier view that the Moon did not have the carbon or water to sustain life, previous studies have focused on using biology for closed-loop life support (*e.g.*, Gòdia *et al.*, 2002). The following presents a quantified concept mission for utilizing carbon, water, and trace metals to produce food from resources found in the lunar ice.

Spirulina is the common name for human and animal food produced primarily from two species of cyanobacteria, *Arthrospira platensis* and *Arthrospira maxima*. As with all cyanobacteria, Spirulina uses photosynthesis to fix carbon dioxide to make food. As carbon dioxide is 0.012% of total mass of lunar ice regolith, lunar ice carbon dioxide could be used as a feedstock for cyanobacteria grown in an appropriate photobioreactor. On Earth, Spirulina is sold as a dietary supplement; it is a complete protein source and approximately 50% protein by mass (Pandey and Tiwari, 2010).

It is estimated that Spirulina would require 2.67 kg of carbon dioxide to produce 682 g of food, which is the amount required in space per person per day. This is based on the following equation:

$$\begin{aligned}&80\%\ \text{of PAR photons} \times 78\%\ \text{used for carbon fixation}\\&\quad\times 10\%\ \text{Glucose per photon} \times \text{MASS}\ CO_2\\&\quad\times (\text{Glucose mass}/CO_2\ \text{mass}) = \text{max total biomass}\end{aligned}$$

It takes 8 to 12 photons to make a single glucose (Chain and Arnon, 1977; Bolton and Hall, 1991). Skillman (2008) indicated that in macroscopic plants 22% of incoming energy is diverted to other systems since it cannot be accounted for in carbon fixation.

Closed Spirulina bioreactors on Earth can produce around 1 g/L per day, dry weight biomass (Vonshak, 1997; Cornet and Dussap, 2009). If run on a 4-day batch cycle in space, a 682 L bioreactor could provide enough food per day for a single astronaut. To produce 682 L of water, 12,179 kg of lunar regolith would need to be processed every 4 days (as H_2O is 5.6% by mass in lunar regolith ice). Processing of the lunar regolith ice for this much water would also provide 14.61 kg of CO_2 (as CO_2 is 0.0012% by mass in lunar regolith ice) and other essential elements to supply the growing cyanobacteria. Because the Spirulina only needs approximately 10.68 kg of CO_2 to produce 4 days' worth of food, there would be an excess of CO_2, which could be stored for future use. The water estimations given in this example are conservative, as they do not account for any water recycling. The carbon requirement is also conservative, as it only takes into account CO_2 and does not utilize the other carbon sources available in the regolith ice.

Many of the other elements that would be required are found in Table 2, which shows that there is plenty of nitrogen, potassium, calcium, iron, and manganese in the regolith. Zinc was also found from rocks brought back from Apollo 16 (Cadogan, 1981).

It is likely that other trace elements required for Spirulina growth such as cobalt, iodide, molybdate, vanadium, selenium, and copper will also be found on the Moon in the future because there is no reason to think that elements found on Earth would not also be found on the Moon in some quantity. But if not, then these elements are only required in very small amounts and could be launched from Earth. Likewise, for the micronutrients required in cyanobacterial media such as cyanocobalamin, biotin, and thiamin, if these cannot be produced *in situ* then they could also be launched from Earth.

The Spirulina could be shielded from the lunar radiation and temperature fluxes by physical containment inside the bioreactor. Previous bioreactor designs have been proposed that transfer solar light through optical fibers to growing cultures inside the bioreactor (Mori, 1985). A detailed energy budget for this system is beyond the scope of this study, which focuses on the resources available, but the energy requirements for the bioreactor could either come from solar or nuclear.

The example given here involves the excavation and processing of large amounts of lunar regolith, which may at first seem unreasonable. However, the ability to excavate these quantities is well within the range of current lunar excavation technology. NASA's recent Lunar Regolith Challenge competition was won by a robot weighing 80 kg that has the capacity to excavate 1000 kg of lunar regolith in a 30 min period (Dunbar, 2009). Although there is no current technology designed for capturing volatiles from lunar evaporated ice, it seems unlikely that the excavation or processing of the lunar regolith ice will be a concern for biological ISRU. Considerable research is still needed, however, on the robotics required to run this as an automated process that could operate and store products from the bioreactor without human presence.

NASA has some experience already in sending containers suitable for use as bioreactors to the Moon. The Centaur upper rocket stage that was crashed into the pole as part of the LCROSS mission was a cylindrical fuel tank 9.14 m tall of diameter 3.04 m, a total volume of 66,342 L. The Centaur fuel tank, however, is very thin and would need to be maintained at high pressure to maintain its rigidity. This does, however, demonstrate the capability of transporting such hardware to the lunar surface.

1.5. Using synthetic biology to enhance Spirulina

The example given so far, of Spirulina, is a naturally occurring cyanobacterium that can fix CO_2. However, it cannot use most of the carbon sources found in the LCROSS data. Synthetic biology might be used to enhance Spirulina so that it could utilize carbon fixation pathways for C_2H_4, CH_3OH, and CH_4. This is not as outlandish an engineering target as one might expect. There are numerous organisms that do use these chemicals as carbon sources, and they can and have been isolated by selection from the environment and even used in industrial settings (Schink, 1985; Thalasso *et al.*, 1997; Tsukamoto and Miller, 1999). From the genomes of such organisms, pathways that provide the capacity to metabolize such carbon sources can then be searched for with bioinformatic techniques as well as library transformation experiments. Because the ability to use a particular carbon source is a selectable trait, large libraries could be screened in such efforts. Alternatively, these other forms that carbon takes on the Moon (C_2H_4, CH_3OH, and CH_4) can all be easily rectified into CO_2 by the simple expedient of combusting them. The oxygen to do so could in turn be liberated from water.

In its current, un-engineered form, Spirulina is a complete nutritional source except for vitamin C and essential oils. Way and coworkers, who have previously engineered cyanobacteria for the synthesis and export of hydrophilic products (Niederholtmeyer, 2010), have also suggested Spirulina as a suitable nutrient source for humans in space (Way *et al.*, 2011). They made note of the potential of cyanobacteria to be grown in relatively low volumes and yet support relatively high biomass yields with fast growth cycles while relying only upon solar energy. This is a capacity unequaled by other biological or nonbiological systems. They propose that cyanobacteria could be engineered with pathways to produce hydrophilic products such as missing nutrients; increase carbohydrate content; and produce particular flavors, colors, and textures to increase palatability.

Barriers to making genetic modification in Spirulina in the past have included the presence of nucleases upon the surface of the Spirulina that destroy foreign DNA before it can be transformed into the cells, and the fact that Spirulina species are motile, which makes isolation of clones from single colonies problematic. However, nonmotile strains have been isolates, and successful construction of vectors, selectable markers, and stable transformation of these strains have been reported, opening the way for larger-scale engineering of Spirulina species (Wang *et al.*, 2009).

The sorts of genetic modification that could be targeted include improving the productivity of such organisms at different temperature ranges, which would allow for greater stability inside the bioreactor. This might be done by simple strain-improvement techniques (mutagenesis and selection). Spirulina could additionally be designed to be radiation resistant if needed by transforming in genes associated with repair pathways. One challenge to farming food-organisms from lunar materials is that metal toxins in the regolith would have to be removed. This too could be done in a biological step similar to bioremediation efforts on Earth either by a separate organism or by adding heavy metal export and resistance genes to Spirulina. If the trace metals currently not shown to exist on the Moon are really not present at all in the lunar regolith, such as cobalt, iodide, molybdate, vanadium, selenium, and copper, then it might be possible to reengineer the enzymes that require them as cofactors to be able to function without them. By providing the necessary biosynthetic pathways from other organisms, Spirulina could also be engineered to produce its own micronutrients that it currently cannot produce, such as cyanocobalamin, biotin, and thiamin. Even the poor flavor of Spirulina might be altered by random transposon-based knock-out and screening approaches.

Over the last decade, the increased ease of DNA sequencing has provided us with many sequenced genomes in digital form. Although current tools for the manipulation of Spirulina are limited, recent advances such as the booting up of a synthetic genome (Gibson *et al.*, 2010) indicate that genetic manipulation will not be a barrier to the field of synthetic biology. This allows us to imagine much broader applications for synthetic biology in space. The ability to send genetic constructs or even whole genomes digitally to a bioreactor on the Moon or to a remote location in our solar system to have them synthesized *in situ* opens up exciting opportunities and important ethical questions for humanity.

2. Resources Available on Asteroids

2.1. Water and volatiles on asteroids

Asteroids have a variety of compositions. About 80% of asteroids at the outer edge of the asteroid belt are carbon-rich C-type asteroids, which are thought to be composed of similar, or identical, material to the carbonaceous chondrite meteorites that fall on Earth. These asteroids are a complex mix of hydrated minerals such as serpentines [hydrous magnesium iron phyllosilicates $(Mg, Fe)_3Si_2O_5(OH)_4$], sulfides, and carbon compounds, among other constituents. Asteroidal material is known to harbor platinum-group elements, volatiles, and a variety of useful resources for human space settlements and terrestrial industries (Kryzanowski and Mardon, 1990; Sonter, 1997).

One factor that might limit the use of asteroid material via biological mechanisms is the presence or absence of water. However, water has been directly detected upon at least two asteroids, 24 Themis (Campins *et al.*, 2010) and 64 Cybele (Licandro *et al.*, 2011). These studies also detected the presence of organic molecules on both asteroids, thus establishing that the two primary prerequisites for feeding a bioreactor with asteroid-derived raw materials, water and carbon, can be found coexisting in individual asteroids. There is some evidence to suggest that between 8% and 13% of NEOs in asteroid-like orbits had their origins as comets (DeMeo and Binzel, 2008). Lastly, water might be manufactured from hydrated minerals that are present and even common in claylike minerals found in asteroids (Rivkin *et al.*, 2002).

2.2. Ways in which asteroid-derived resources can be turned into useful products

The sorts of materials that might be useful to manufacture on asteroids are very similar to those that might be manufactured on the Moon. However, the high metal content of some asteroids demands special attention and invites comparison with terrestrial biomining operations. Microorganisms might be used to facilitate access to this asteroid-derived material by enhancing the breakdown of the constituent minerals. In an experiment in which the acidophilic biomining organism was used, *Acidithiobacillus ferrooxidans*, researchers demonstrated that the reduced iron phases within carbonaceous meteorites (the Murchinson and Cold Bokkveld meteorites) could be used as a source of energy by these microbes (Gronstal *et al.,* 2009), and that in the process they degraded the meteoritic material and accelerated its weathering. The existence of sulfides within the meteorites could generate sulfuric acid (in an artificially oxygenated atmosphere), analogous to terrestrial biomining, to assist in rock dissolution and extraction of economically important elements.

The compatibility of microbial growth of biomining organisms with carbonaceous asteroidal material is consistent with experiments conducted by Mautner (Mautner, 1997, 2002; Mautner *et al.,* 1997). In experiments in which extracts of Murchison meteorite were used, soil microorganisms (*Pseudomonas*, *Arthrobacter,* and *Nocardia* spp.) were shown to achieve populations in excess of 10^7 colony-forming units/mL in meteorite extracts. In this case, the heterotrophic organisms use the carbon from the meteoritic extracts. Plants were shown to change elemental composition, but their growth was enhanced with nutrients provided by meteoritic extracts. Later experiments led to a prediction that asteroidal material could sustain 10^{32} microorganisms and 10^{14} humans based on biomass calculations resulting from these experiments (Mautner, 2002).

Toward the inner regions of the asteroid belt, the carbonaceous asteroids are rarer, and most of the material is similar to the stony chondrites, materials comprised of pyroxenes, olivines, and other Fe-, Mg-, and Ca-containing silicate minerals. Pyroxenes are silicon or aluminum oxides complexed with a pair of metal ions, one large and one small. The large ion can be derived from elements such as calcium, sodium, ferrous iron, magnesium, zinc, manganese, and lithium, while the smaller from ion elements such as chromium, aluminum, ferric iron, magnesium, manganese, scandium, titanium, and vanadium. Olivine is $(Mg,Fe)_2SiO_4$. These materials do not contain the organic compounds that would sustain organic-utilizing microorganisms investigated in the studies by Mautner. However, the abundant bioessential cations could provide nutrients for other types of microorganisms and plants. The reduced iron in olivines and pyroxenes could act as an electron donor for iron-oxidizing bacteria, similarly to the experiments conducted by Gronstal and coworkers in biomining operations (Gronstal *et al.,* 2009). These authors also showed the capacity for anaerobic biological iron-oxidation by using carbonaceous chondrites, which might also be used to drive microbial growth and processing of asteroidal non-carbonaceous material.

Two elements that might not require biomining operations are iron and nickel, which are abundant in some asteroids. Indeed, some bodies contain solid iron and nickel metal and correspond to the iron meteorites or the nickel and iron found in stony chondrites collected on Earth. This material, if it can be collected, could be melted to yield pure iron or nickel. Nevertheless, it should be noted that this material has been used as a source of energy for bacteria in laboratory experiments (González-Toril *et al.,* 2005; Gronstal *et al.,* 2009) and could be used as a source of additional iron-derived energy for iron-oxidizing bacteria in asteroidal biomining operations, perhaps supplementing bioreactors used to biomine asteroidal material with low iron concentrations.

3. Resources Available on the International Space Station

The environmental control and life support (ECLS) system on board the ISS offers a unique opportunity to immediately implement and test novel biological ISRU methodologies relevant to life-support systems. Integrating biological components within the ISS ECLS system will not only benefit existing systems such as wastewater recycling and air revitalization but also will provide approaches for new avenues of ISRU. For example, undesirable biofilm formation on the surface of urine-processing equipment may be prevented by intentionally inoculating the system with a benign, biofilm-degrading microbe engineered to assist in treating urine, rather than focusing on developing an anti-biofouling material. Unlike lunar or NEO environments, carbon fixation is not the limiting factor for the development of products such as food. As biological engineers move toward designing synthetic microbial ecosystems on the ISS, solutions to more complex problems such as extracting edible biomass from solid waste and generating breathable oxygen could possibly be developed.

3.1. Biological urea processing

Fresh human urine is composed of 95% water and nitrogen concentration ranging from 7 to 9 g/L (Kirchmann and Pettersson, 1994). Urea, ammonia, and uric acid amount to 90–95% of the total nitrogen in urine. Urea is the main nitrogenous compound, accounting for 85%, and various amino acids contribute between 1% and 5%. During storage, urea decomposes primarily into ammoniacal nitrogen, the ammonium being paired with bicarbonate (Kirchmann and Pettersson, 1994). Therefore, urea presents itself as a logical target for nitrogen reclamation in the ISS.

Various soil and gut microbes, including *Bacillus subtilis, Helicobactor pylori,* and *Escherichia coli,* produce ureases that catalyze the hydrolysis of urea into ammonia and carbon dioxide. The ammonia can be further converted into nitrite. These bacteria may prove beneficial as the starting point for developing an optimized strain for ISS waste-stream processing. For example, the urease gene cluster found in *B. subtilis* encodes functional urease apparently without known accessory proteins such as those encoded in the *ureDABCEFG* cluster in *E. coli*, although it is possible that unidentified accessory proteins are required for activation (Kim *et al.,* 2005). This gene cluster presents an excellent foundation for optimizing controlled urea breakdown into usable compounds within an appropriate microbial host.

One can imagine utilization of a microorganism to efficiently convert urea into ammonia and carbon dioxide while existing in the ISS wastewater stream, without any potential harm to the humans on board. The CO_2 and ammonia could potentially serve as carbon and nitrogen sources for Spirulina. Alternatively, released carbon dioxide could possibly react with hydrogen over an appropriate metal catalyst (in what is known as the Sabatier reaction) to produce water and methane.

3.2. Solid waste recovery

The ECLS system aboard the ISS performs regenerative functions for atmosphere revitalization and water recovery by way of physiochemical modules of finite life span that require periodic component exchange (Wieland, 1998). Human metabolic waste is stored and returned to Earth for disposal. A closed-loop, biological regenerative life-support system able to more completely recover resources from the waste stream could alleviate the demand for resupply missions while increasing the station's independence. The European Space Agency's Micro Ecological Life Support System Alternative (MELiSSA) has described and partially realized such a system for future space habitats (Gòdia *et al.*, 2002).

Significant challenges remain before such a system could be considered for implementation on the ISS. Research questions involving the effects of microgravity, radiation, and the robustness to environmental and genetic perturbations of the microbial cultures remain open (Hendrickx *et al.*, 2006). A space station imposes severe volumetric and power limitations that would need to be met as well as operational considerations. The crew needs to be able to fully service the components of the life-support system. The development of MELiSSA for planetary or lunar habitats will bring solutions to these obstacles closer as experience accumulates and eventually allow for closed-loop life support on space stations such as the ISS.

Conclusion

A persistent human presence in space currently exists aboard the ISS. Ongoing Earth-based supply of the ISS is barely feasible and comes at a great cost. However, such a supply chain operating continuously across deep space (such as between Earth and an asteroid) is hard to imagine. Consequently, as the persistent human presence expands further into the solar system, the need to be able to "live off the land" will become increasingly vital. Even before we reach the conceivable end of an ISS-style Earth-based supply chain, economic pressures to reduce the cost of exploration missions will make ISRU unavoidable.

Humans have always depended upon a plethora of associated organisms: gathering nutrients, harnessing energy, and detoxifying waste streams. It should therefore come as no surprise that any attempt at creating systems capable of supporting human habitation beyond low-Earth orbit will require a biological component. No mechanical or biological system can continue to function in a completely closed manner. So energy, matter, or both must be introduced to keep any such system running. The engineering and control of such systems to use *in situ* resources is therefore absolutely essential to any long-term presence of humans in this solar system beyond the immediate vicinity of Earth. This includes the engineering of the biological components of such a system. As outlined in this article, achieving this is neither impossible nor even especially ambitious. Efforts to achieve this goal will likely begin with waste processing on the ISS and eventually extend to the demonstration of food production both from waste streams and raw materials that are abundant on the Moon and asteroids.

Abbreviations

ECLS, environmental control and life support; ISRU, *In Situ* Resource Utilization; ISS, International Space Station; LCROSS, Lunar Crater Observation and Sensing Satellite; MELiSSA, Micro Ecological Life Support System Alternative; NEOs, near-Earth objects.

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Address correspondence to:
Michael Montague
The J. Craig Venter Institute
Department of Synthetic Biology
Bldg1 345E
9704 Medical Center Drive
Rockville, MD 20850
USA

E-mail: mmontagu@jcvi.org

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