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END-TO-END MISSION DESIGN FOR MICROBIAL ISRU ACTIVITIES AS PREPARATION FOR A MOON VILLAGE

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Abstract

In situ resource utilization (ISRU) increasingly features as an element of human long-term exploration and settlement missions to the lunar surface. In this study, all requirements to test a novel, biological approach for ISRU are validated, and an end-to-end mission architecture is proposed. The general mission consists of a lander with a fully autonomous bioreactor able to process lunar regolith and extract elemental iron. The elemental iron could either be stored or directly utilized to generate iron wires or construction material. To maximize the success rate of this mission, potential landing sites for future missions are studied, and technical details (thermal radiation, shielding, power-supply) are analyzed. The final section will assess the potential mission architecture (orbit, rocket, lander, timeframe). This design might not only be one step further towards an international "Moon Village", but may also enable similar missions to ultimately colonize Mars and further explore our solar system.

Keywords: microbiology, ISRU, mission architecture, bioreactor

Acronyms/Abbreviations

Earth-Moon Lagrangian Point 2 (EM-L2)
Global Exploration Roadmap (GER)
Isopropyl β-D-1-thiogalactopyranoside (IPTG)
In situ resource utilization (ISRU)
International space exploration coordination group (ISECG)
International Space Station (ISS)
Low Earth Orbit (LEO)
Luria-Bertani (LB)
Mars Oxygen ISRU Experiment (MOXIE)
National Aeronautics and Space Administration (NASA)
Optical density (OD)
Phosphate buffer saline (PBS)
Radioisotope thermoelectric generator (RTG)
Tryptic soy broth (TSB)

1. Introduction

Multiple efforts are now under way to enable sustainable exploration beyond the ISS and LEO. Space agencies are embracing exploration with commercial and international partners in order to return to the Lunar surface, and in doing so, bring back new knowledge and potentially resources, and open up new opportunities for innovation. In this context, new technological concept demonstrations are increasingly sought for surface missions and payloads. In particular, activities that can

potentially enable sustainable exploration for missions on the surface for lengths up to and surpassing 40 days, are being considered. In this work, we present a novel candidate mission concept which leverages modern advances and understanding in synthetic biology to realize an ISRU focused concept that could become an exploration enabling architecture, as well as enabling valuable scientific investigations pertaining to the lunar environment.

1.1 Mission objectives & benefits

Developing the capability to use local planetary resources is critical for future sustainable long-duration missions on the Lunar and Martian surface. The mission here described proposes a novel ISRU biology-based method, which would permit the extraction of metal and gases from lunar regolith, helping to reduce the cost of human missions[1]. While a primary role of ISRU for a Lunar Outpost would be the production of oxygen[2], accessing lunar metals is needed for the development of a lunar infrastructure, and supply from Earth would be mass and thus cost prohibitive[3]. The production of metals and gases from this mission will not enable full independence of Earth supplies, but it would prove the potential of biological ISRU and reduce the resources that are brought from Earth[4].

The objectives of this mission concept are:

- Test a life-support system designed for the bacteria working in the lunar environment.
- Demonstrate the ability to process lunar regolith and extract elemental iron, silicon and more by landing a fully autonomous bioreactor on the lunar surface. The extracted material could be used as raw material, e.g. for manufacturing using 3D printing technology.
- Prove the capability of storing gases (H_2, O_2, CO_2, CH_4) generated by the bacteria during the metabolic process. Over time, this arrangement could store significant amounts of gas-by-products, useful for a permanent human outpost.
- Test the toxicity of lunar dust and the radiation environment on simple organisms.
- Extend the International Space Station (ISS) cooperation model, allowing international partners to develop systems/subsystems of the rover and bioreactor.
- Increasing the number of complex biomolecules on the moon to enable future food supply (The availability for certain biomolecules (carbon, nitrogen-containing) is key to enable sustainable food supply *in situ*) [5].

1.2 Assumptions

Several major assumptions were made in the conceptual design of the mission architecture and are listed below:

1) The mission is assumed to advance the objectives of, and take place within, the overall framework of the Global Exploration Roadmap [6]. Taken as part of a broader effort, the mission would have a higher chance of success, would build key resource utilization knowledge prior to human exploration, and leverage the presence of other capabilities being developed as part of the GER
2) The mission will be a collaboration between several ISECG members with different contributions.
3) The mission will be a technology development and demonstration mission to advance capabilities required for further incremental, sustainable robotic and human-robotic exploration.
4) The bioreactor system will have the capacity of being fully autonomous.
5) Public or private launch vehicles capable of carrying the proposed payload space agencies or private companies will be available by the timeframe of the mission.

1.3 Similar Mission Architectures

NASA, ESA, and other space agencies have for long exposed the importance of ISRU to reduce the cost of the missions. Upcoming missions include robotic capabilities to acquire and process local resources:

- ESA drills and science instruments on Roscosmos's Luna 27 (PROSPECT) will demonstrate the thermochemical extraction of water from lunar regolith[6].
- Selected to fly on NASA's Mars 2020 mission, MOXIE (Mars Oxygen ISRU Experiment) is a payload that will produce oxygen from the Martian atmosphere using solid oxide electrolysis (SOXE)[7].
- Instruments aboard the lander and rover from the ISRO's Chandrayaan-2 mission will collect data on the moon's thin envelope of plasma[8].
- As part of the recently canceled Prospector Mission, a rover would have excavated volatiles such as hydrogen, oxygen, and water from the moon[9].
- Other upcoming missions include biological experiments to perform on the lunar surface: the Chang'e 4 Chinese mission planned for 2018 will deliver a lander to the far side of the Moon carrying, among other instruments and experiments, a container with potatoes, *Arabidopsis thaliana* seeds and silkworm eggs.

Together, the plants and silkworms are expected to create a simple ecosystem[10, 11].

Also, other methods have been studied to evaluate their potential to produce oxygen and metal from lunar resources to support human exploration of space:

- The electrolysis of molten lunar regolith, also termed Magma process, requires an electrolytic cell where the regolith is molten, and in which a potential is applied such that oxygen evolves at the anode and metal deposits at the cathode[12].
- The electrolysis of solid lunar regolith approach, which derived from the FFC-Cambridge process for the electro-deoxidation of metals and metal oxides[13].
- Additionally, there are numerous mission architectures proposed in scientific literature approaching the structure of telerobotic operations[14, 15]

intended to fit within the sustainability principles of the GER including affordability and partnerships, exploration benefit, and capability evolution[6]. A partnership between different nations is proposed to increase affordability and provide opportunities for different partners to contribute in their areas of interest and expertise. ISRU technologies will be demonstrated that will be of use in later space exploration missions, and advancements in the technology would be available for terrestrial uses such as on-demand pharmaceuticals in the longer term.

The mission consists of two main systems to be landed on the lunar surface together; a landing vehicle is containing a bioreactor and all necessary support subsystems, and a robotic rover capable of autonomous and teleoperations. The rover will be capable of collecting and depositing regolith into the bioreactor to perform the main mission objectives and will perform operations throughout the lunar day. During the lunar night, the rover will be housed inside the landing vehicle for protection.

2. The general mission architecture

2.1 Overview

The mission architecture, laid out in this section, is intended to fit within the sustainability principles of the GER including affordability and partnerships, exploration benefit, and capability evolution[6]. A partnership between different nations is proposed to increase affordability and provide opportunities for different partners to contribute in their areas of interest and expertise. ISRU technologies will be demonstrated that will be of use in later space exploration missions, and advancements in the technology would be available for terrestrial uses such as on-demand pharmaceuticals in the longer term.

The major stages of the mission are as follows:

1) Landing of the reactor and gatherer-rover, initial system commissioning. Site selection is covered in

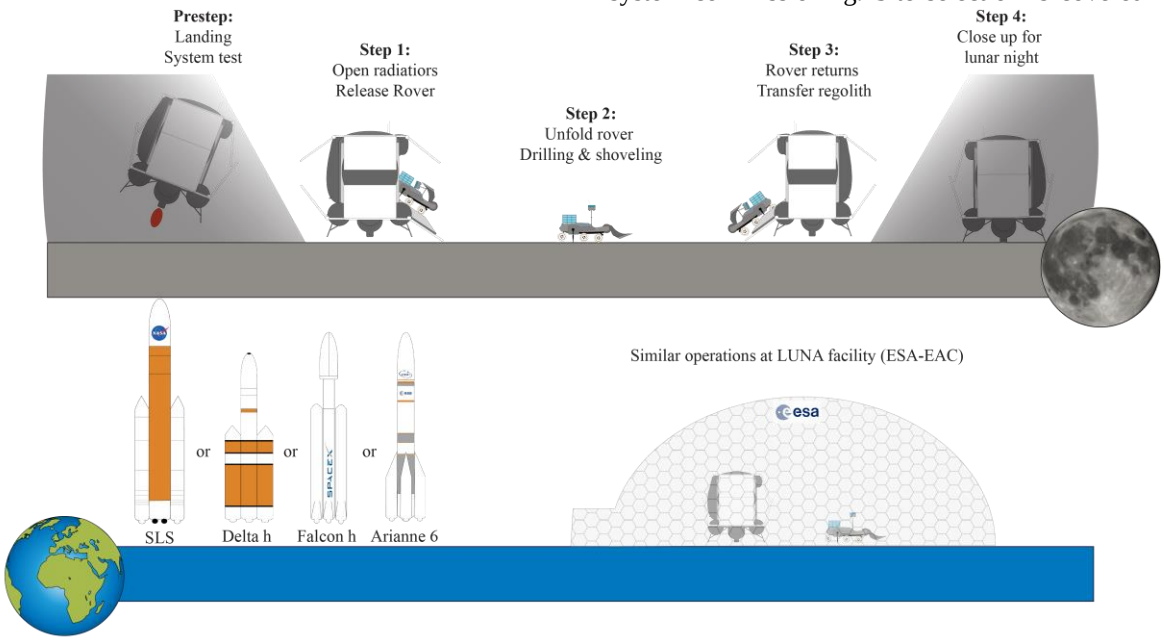


Fig.1: Overview of the mission architecture. A heavy launcher (SLS, Arianne 6, Delta IV heavy or Falcon heavy) will be used to transport the lander to the lunar surface. After the landing and an initial system test during dawn its rover will be released, unfolded and start to collect lunar regolith. The rover returns the samples to the lander/bioreactor and several biological tests can be performed. Throughout lunar night the rover will stay in the lander and start after the night again with step 1. A similar experiment is performed at the LUNA facility at the European Astronaut Centre. This enables direct comparison of the cellular growth.

the following section.

2) Regolith collection operations begin – the rover will gather lunar regolith with a surface drill and with a shovel (collection of regolith with smaller particle sizes is targeted as it is more easily processed by the bioreactor bacteria).

3) Biological experiments are performed in the bioreactor (e.g. material extraction)

4) Repetition of mission stages 2 and 3.

Throughout the mission, the repetition of regolith collection and material extraction will allow different cell systems to be tested with a variety of microorganisms as well as cell-cultures, to maximize science output and test different approaches. Regolith from different areas of interest could also be collected for analysis. Direct comparisons will also be made with terrestrial analog experimentation, for example, experiments performed at the LUNA facility of the European Space Agency[16]. Design of the lander and rover is detailed in sections 3 and 4.

2.2 Landing Site

In order to find a suitable site for a settlement, many parameters have to be taken into account. These parameters can generally be separated into three categories: (1) Operational conditions; (2) Availability of natural resources; (3) Features of scientific interest.

Historically, the operational conditions were always prioritized as the selection of the site was primarily constrained by technical factors. In 1959, the Project Horizon study[17] determined that, given the constraints related to the environment, the communication systems and the energy requirements for the rockets, the site should not be located further than 20° from the optical center of the moon. During the Apollo program, lunar topography was also taken into account for the selection of a landing site[18] as a smooth terrain is critical for landing operations.

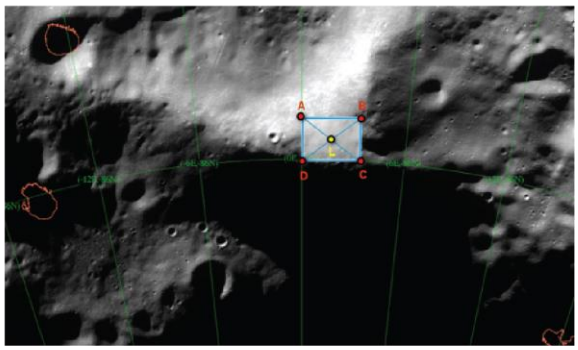
In the near future, more powerful launchers such as the SLS and the Falcon Heavy as well as relay satellites[19] will make it possible for a Lunar base to be located virtually anywhere on the moon's surface. The latest technologies in the domain of autonomous landing and hazard detection[20] will also make accessible sites with rougher terrain.

For this mission, the priority is set on operational factors. In this context, one of the biggest constraints is power production. Therefore, we consider sites located at the poles, where particular topographic features combined with orbital mechanics allow good illumination conditions for long periods of time. These areas are also characterized by relatively stable temperature conditions, which may be critical to the mission operations. It is also the region chosen by NASA for its lunar outpost reference design[21].

A particularly interesting possibility is the summit of the Malapert mountain, close to the south pole. Indeed, it has been estimated that this site receives sunlight for 93% of the lunar year[22]. Due to its elevation of about 4700 meters above the lunar reference radius[23], this location also allows for a line of sight communication with the earth, possibly a second base on the rim of the Shackleton crater and many regions of scientific interest around the South Pole. Features of scientific interest are also located in the direct vicinity of Mons Malapert. As an example, a crater located at the south of the peak is considered a possible "cold trap"[24].

A portion of the south pole lunar map was evaluated regarding a suitable area for mission operations and landing. (Fig.2A). An important factor was the average slopes and distances in this area to allow for a smooth movement of the rover (Table1).

A



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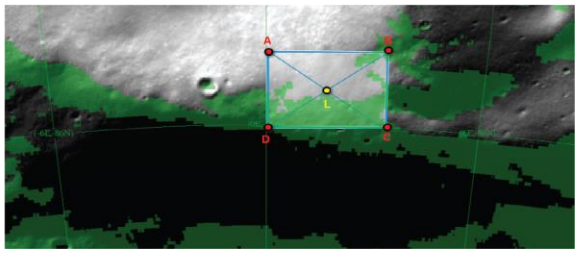

Fig. 2: (A) Position of the landing site (point L), and approximative area of operations (square ABCD)
(B) The marked areas (green) indicate terrain slope values below 10 degrees. [http://bit.ly/2PE69Ru, quickmap.lroc.asu.edu]

Point	Coordinates
A	85.8° S, 0.0°E
B	85.8° S, 3.4° E
C	86.0° S, 3.4° E
D	86.0° S, 0.0° E
L	85.9° S, 1.7° E

Path	Distance	Average Slope
AB	7250 m	5°
BC	5130 m	8°
CD	6970 m	-3°
DA	5130 m	-12°
AC	8570 m	9°
BD	8870 m	4°

Table 1: Coordinates, distances and average slopes in between the field of operation (see Fig.2).

The area chosen for mission operation is characterized by an average slope of about 12° (Table 1 and Fig. 2B). This approximated value is obtained by modeling the region as a geometrical plane passing through points A, B, L (Fig. 2A). The region close to the point C shows a flatter terrain and is more suitable for lander operation (Fig. 2B). Point C is also close to the summit of Malapert Mountain, reaching better illumination condition, and avoiding shadows generated by surrounding reliefs. Nevertheless, the area around point C is characterized by a rough terrain and several impact craters that can be easily explored and avoided by rover operations, but not during landing. As a result, even if point C remains a good candidate as a landing site, point L is preferred in this mission since it supposed that illumination conditions are not very different with respect to point C.

3.Bioreactor/Lander

3.1 Lander design

The approximate lander dimensions are 5 metric tons (+ rocket fuel for landing), 4.5 m wide and 6 m high. Based on these dimensions, a heavy lifter would be needed to transport this reactor to the moon[25-28]. A main part of the reactor is its resupply tanks and RTG power source to ensure continuous operations for years (Fig. 3A). All tanks would be equipped with an external valve allowing them for refilling if needed.

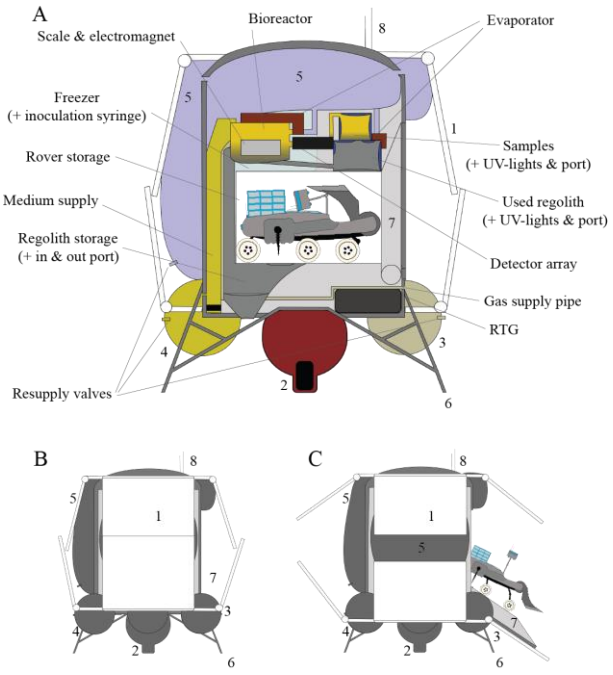

Fig. 3: Overview of the lander with its external structures: heat radiators (1) landing engine (2) gas tank (3) medium tank (4) water tank (5) landing legs (6) rover ramp (7) and communication antenna (8). (A) Central cut of the lander showing the main internal structures to resupply and run the bioreactor as well as store the rover throughout space-travel and lunar nights. (B) External structures of the lander in its closed position (left) with the radiators (1) attached to the body and the ramp (7) still closed as well as in its open position (7) (right) with the radiators (1) spread out and the rover being released.

3.1.1 Tanks

The main tank (top and mid of the reactor) is filled with sterilized water (Fig. 3 (5)) which has several functions. Passively it will be used as a radiation shield against cosmic rays and a heat reservoir throughout the lunar night. For the latter function, heat coils connected to the RTG are used to keep the water above its freezing point and to store heat there before the night starts. Water used in the bioreactor will be transported back into the tank via evaporation after each growth experiment. The main loss of water per iteration will be caused by the liquid used during the microbial growth. An optional design would store the water in separate hydrogen and oxygen tanks and use a fuel cell to produce water and energy on demand (not shown here).

The tanks at the lower part of the reactor are filled with growth medium (Fig. 3 (4)), a gas mixture (Fig. 3 (3)) and rocket fuel (Fig. 3 (2)). The growth medium is in the form of a compressed, sterilized powder and it will

be premixed with water while being pushed into the bioreactor. The gas (mainly O_2, N_2) is used to mix the reactor liquid (bubble reactor) continuously [29].

Assuming a delta-v of 1870 m/s and a mass of 5 tons, approximately 4300 kg of fuel will be needed for the initial landing. Five spherical tanks with a volume of one m^3 each are providing the necessary space for this fuel. The emptied fuel tanks will be reused after the landing to store excess gas produced throughout the experiments.

3.1.2 Bioreactor

The central piece of equipment is the 5 L bioreactor coupled to the scientific instruments. All supply valves and ports are installed in redundancy to ensure proper function of the system. This reactor is filled up in several steps (Fig.4):

1. Lunar regolith (10 - 1000 g) gathered by the rover is transported into the reactor with an Archimedean screw mounted in a 45° angle.
2. Water and medium are premixed and used to fill up the reactor
3. The gas flow is initiated to achieve improved mixing.
4. A syringe is used to inoculate the bioreactor with the organism of interest (stored in a freezer).
5. Start of the actual experiment: Every hour the optical density at 650 nm (OD_{650}) and total cell counts are observed (see chapter 3.3). The tested material is stored in an extra area and regularly sterilized with UV radiation. The experiment runs between 12 and 48 hours at a constant temperature depending on the scientific or operational question (see chapter 3.2). A scale, centrifuge, ChemCam[30] and electromagnet can be used for further analysis or extraction. The gas is reused for the entire experiment duration.
6. UV lamps will be used to sterilize the bioreactor and terminate the processes. Two additional optical density and cell count measurements should verify this termination.
7. The temperature in the reactor is increased, and the gases are evacuated and measured (see chapter 3.2.3).
8. Water will be evaporated and fed back into the water tanks. The solid material (cells, rest of the medium and regolith) will be transported in a storage compartment and further sterilization processes are initiated. The compartment has an airlock to remove material from the storage and transport it outside.
9. The whole system will be sterilized again and is ready for a new experiment. If a different type of regolith is used (other area or different drilling depth), all regolith storage chambers must be emptied.

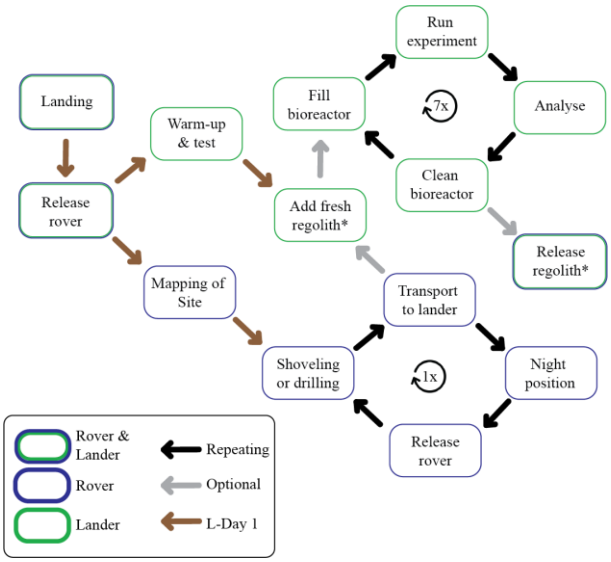

Fig. 4: Overview of the rover and lander operations. Arrows in light brown indicate operations only done on the first lunar day. Grey arrows are optional operations depending on the exact experiment, and black ones show repeating processes. The rover has normally only one iteration per lunar day/night cycle, while the bioreactor can have up to 7 iterations. (*) All regolith stored and used in the bioreactor can be released to allow testing of different locations and drilling depths.

3.1.3 Additional systems

The lander has four landing legs similar to the Apollo missions[31] and eight radiators (two each side) to get rid of excess heat during lunar day and shield it during lunar night. The rover storage area is used for initial transport of the rover, safeguarding the rover throughout lunar night and transfer regolith from and to the rover. Antennas are used to enable communication between the rover and the lander as well as to transfer data to the earth (see chapter 6.)

3.2 Extraction process

The rover supplies the lander with regolith to be mixed with bacteria, water, and growth medium. The modified bacteria extract or alter certain elements from the regolith (e.g., iron, silicon, gases). The extracted materials will be stored in an additional tank and can be accessed via an airlock. The produced gas will be stored in the empty fuel tanks. Growth medium can only be used once, but water can be evaporated and reused for multiple runs. Bacterial kinetics and total number are continuously observed by optical density.

After the extraction process, a small amount of bacteria can be stored for an additional batch, while the rest of the reactor is decontaminated, and the materials (iron, silicon, gases) are extracted.

3.2.1 Silicon extraction

Silicon is an important building block for all types of electronic devices and in particular energy devices (solar cells, fuel cells). Genetic modifications of *E. coli* (Top 10) allowed the bacterium to express the enzyme silicatein-alpha at the surface. This enzyme is used by marine demosponges to build layers of poly-silicate, a complex of SiO_4^{2-} salts bound to a protein[32, 33]. The TU Delft iGEM team 2016 visualized this formation of a poly-silicate using a sodium silicate solution and fluorescence microscopy[34].

We applied the same methodology to test the capability of this genetically modified *E. coli* to extract silicon from the lunar regolith simulant EAC-1. The cells were grown with a lunar simulant concentration of 4 g/L in Luria-Bertani (LB) medium under continuous shaking (250 rpm) and 37 °C. The optical density (OD_{600}) value was observed hourly in a photospectrometer, and 0,1 mM IPTG solution was added after the OD_{600} reached 0.4 (an indication that the bacteria are in exponential growth and are having a maximal metabolic activity). After 4 hours of incubation, the cells were stained with 2 µg/mL rhodamine 123 for 10 minutes and kept in the dark to prevent bleaching. Six washing steps (exchanging the whole volume with phosphate buffer saline (PBS)) were performed and the cells were resuspended in PBS. An inverted fluorescent microscope (Nikon Eclipse Ti inverted microscope with A1R confocal module) was used to visualize the stained and unstained cells (Fig. 5A). Quantification of the cells (n > 19) was automated using a macro implemented in the image analyzing software *Fiji*. The quantification of stained cells is selected based on intensity, size and shape. The findings indicate that the genetically modified bacteria can utilize the alkoxy silicates present in the simulant regolith to encapsulate themselves with a poly-silicate layer (Fig. 5B). In the presence of 4 g/L regolith simulant, 13.4±0.9% of the genetically modified bacteria and 1.3±0.2% of the wild-type bacteria are covered with a poly-silicate layer. Without regolith simulant, 6.7±1.3% of the genetically modified bacteria show silicate expression (errors are standard errors of the mean).

The extraction of silicon-covered bacteria from the remaining regolith is problematic because they share the mechanochemical properties of regolith particles. An agglomeration of the silicon-rich surfaces based on changing the pH could resolve this issue but still requires optimization.

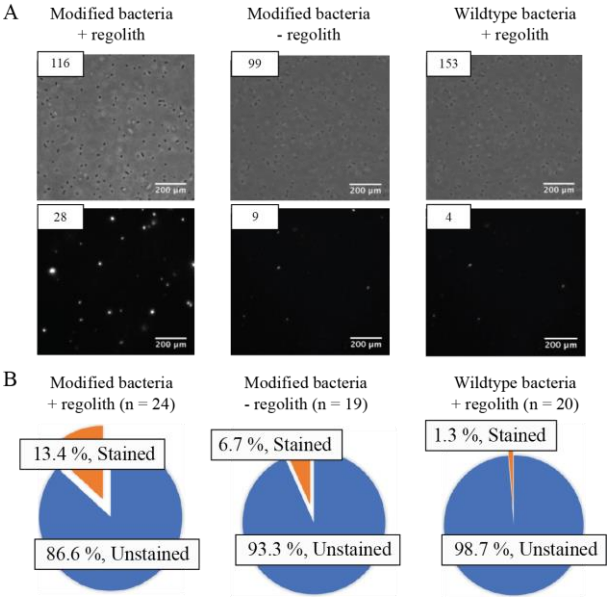

Fig. 5: Fluorescence microscopy showed the modified *E. coli* to be able to form a silicone layer from lunar regolith simulant (EAC-1) around its cell membrane. (A) Exemplary light microscopy (top) and fluorescence pictures (bottom) of rhodamine 123 stained *E. coli* cells. The left two refer to the genetically modified, the right one to the wild-type *E. coli*. The numbers in the left top show the total number of visible cells. (B) Quantitative analysis of the modified and unmodified with rhodamine 123 stained bacteria.

3.2.2 Iron extraction

Stable structures on earth rely strongly on iron and its derivatives. Construction material, as well as replacement parts on another planetary surface, can be built from iron as well. The main disadvantage of iron derivatives, the high mass, plays a minor role in in-space applications on low-gravity planets and moons, where their weight is lower. Iron is also very abundant in Lunar and even more in Martian regolith and a very bioactive molecule.

Our collaborators at Newcastle University engineered an additional *E. coli* strain (DH5α) to incorporate a high amount of iron ions from a liquid solution. Magnetic forces (neodymium magnets) separate the iron stored within the bacteria from the rest of the material (Fig. 6A). The bacterial growth was performed aerobically in a 30 °C incubator under constant shaking with 250 rpm. Colony forming units and OD_{650} measurements were done every 3 hours. For the magnetic extraction, 3 mL were pipetted onto a cover glass slide and washed with 5 mL distilled water. The extracted amount of regolith increased significantly from 3.34±0.58 mg to 7.25±1.83 mg after a 48-hour treatment with bacteria, but showed

no significant difference in the control (3.34±1.15 mg at 0h and 4.57±1.13 mg after 40h) (ANOVA Turkey PostHoc test: $p_{T0\text{-}T48}$: 0.600, $p_{E0\text{-}E48}$: 0.004, $p_{E0\text{-}T0}$: 0.999, $p_{E48\text{-}T48}$: 0.010; errors are standard deviation).

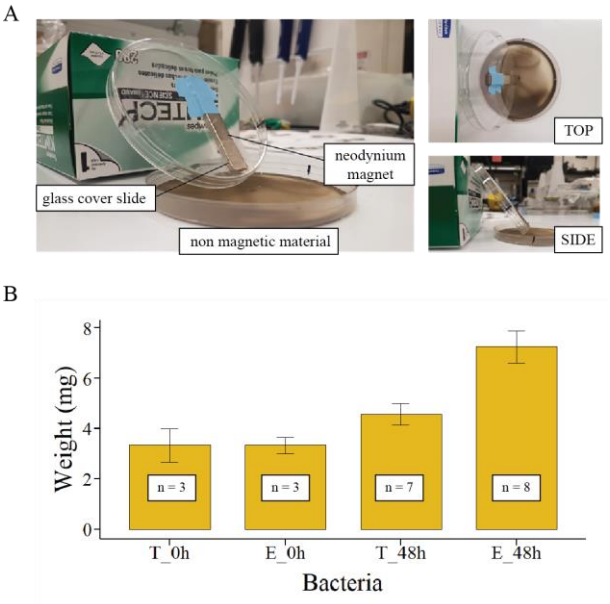

Fig 6: Bacterial modification of the regolith allows for an improved magnetic extraction rate than without treatment (A) Laboratory set-up for the magnetic extraction. The neodymium magnet was mounted in a 55° angle, which allows for extraction of only strongly-magnetically attracted material (the rest will be washed away). The material is deposited on cover glass slides to quantify and compare the total amount of extracted material. (B) Weight measurements of the magnetically extracted regolith. Bacterial samples: E...*E .coli* and non-bacterial controls: T...TSB were performed after 0h and 48 h (Errors are the standard deviation of the mean).

3.2.3 Gas extraction

During any metabolically active process, bacteria produce technical useful gases (CH_4, CO_2, O_2 H_2), which are collected into the emptied fuel tanks. The amount of these gases is measured after each iteration (see section 3.3). Over time, this arrangement stores a significant amount of gas-by-products useful for rocket fuel, life-support system, and similar applications.

3.2.4 Other extraction types

Bacteria can also be used for other types of extraction. It was shown that they could help to recycle copper from electrical circuits[35], leach the ores for further mechanical methodologies[36] and even produce biofuel or medicine[37] on demand. One bioreactor can host several different operational modes and might be used for a variety of different purposes depending on the current need. Resupply missions of the bioreactor can supply not only fresh medium and water but also different organisms.

3.2.5 Potential difficulties regarding bacterial growth in a closed containment system on the Moon

Mutation rate: After materials extraction, the same bacterial culture can be used to re-inoculate a new batch. This process is limited due to the increased mutation rate in comparison to earth, caused by higher cosmic radiation and the mutagenic nanoparticles expected in lunar regolith. If the yield or health of the bacterial culture declines or the extraction methodology is switched, fresh bacteria can be used from a frozen stock (-80°C). Due to the inactive metabolism of the bacteria, the frozen stock is barely affected by the radiation.

There is a low chance of having a strong disturbing mutation during a single batch if the extraction process takes longer than a few days. However, the core extraction processes suggested here are performed in 48 hours, which makes this issue neglectable.

Biofilm formation: Bacteria tend to form biofilms, which can cloak tubing and cause other mechanochemical and biochemical issues leading to a decreased yield or even failure of our bioreactor. Several studies showed the enhanced biofilm formation of different organisms under stress[38] and in the case of low gravitational fields[39]. Our reactor design induces a turbulent flow to counteract this effect, but a pretest performed with the exact reactor on earth will be necessary to better understand the potential problems for the mission due to biofilm formation.

Slicing of cells: Lunar regolith is composed of sharp-edged nanoparticles which can pose a severe threat for the microorganisms[40]. A better understanding of the interaction of moon dust with cellular systems will be of utmost importance for any human endeavor to the moon. Our proposed mission can be seen as a first scientific testbed for this interaction.

3.3 Scientific analysis

The understanding of the interaction between biological systems and the lunar environment will be critical for any future moon mission. It is possible (Vis-spectroscopy and flow-cytometry) for an autonomous system to observe the health of microorganisms or even cell cultures. The proposed Vis-spectroscopy uses a laser with a wavelength of 600 nm to measure the optical density (OD) of four 5 µL samples every two hours. The result shows the number of grown bacteria in the sample and can be used to understand the kinetics of the microorganisms grown. The flow-cytometer complements this measurement with the total number of life/dead stained cells in four additional 5 µL samples at the same time points[41].

Gas sensors (O_2, N_2, CH_4, CO_2, H_2) will observe the composition and production of gases with different organisms and environmental conditions. An electromagnet together with a scale will specifically help in understanding how much iron/magnetic material can be extracted from different lunar soil samples with and without treatment. The integrated ChemCam enables, meanwhile, the measurement and visualization of ore compositions and enables the comparison of ores before and after bacterial treatment as well as in different ground depths and areas (shovel & drill)[30, 42].
The same bioreactor will work simultaneously in LUNA[16] a lunar training hall currently under construction at the European Astronaut Centre (EAC) on earth. The terrestrial bioreactor allows for a better understanding of the biotoxicity caused by lunar radiation and lunar dust as well as an exact comparison of lunar regolith with its simulant.

This mission will acquire critical data to understand the effect of lunar dust particles and the high radiation field on the moon towards biological systems. Furthermore, it will measure the production yields of microorganisms under lunar conditions in comparison to earth conditions.

4. The gather rover

4.1 Rover design

The main task of the rover is to gather and transport lunar regolith to the bioreactor. It consists of a front shovel and a vertical drill to gather the material from different positions as well as a chamber for internal storage. Next to the drill are transfer-ports to drop the material into the lander. The rover's navigation is partly telerobotic and partly autonomous, its navigation is mainly done via optical cameras including infrared sensors at the head camera and the back camera. The solar cells are mounted on a rotating frame to always face the sun (Fig. 7A). The rover dimension will be very similar to resource prospector (approximately 300 kg, 1.4 m x 1.4 m x 2 m)[9] and the rover will reuse originally for resource prospector designed instrumentation. The main difference is the design with six legs instead of four which allows for redundancy and driving in terrains with higher slopes.

4.2 Rover positions and operations

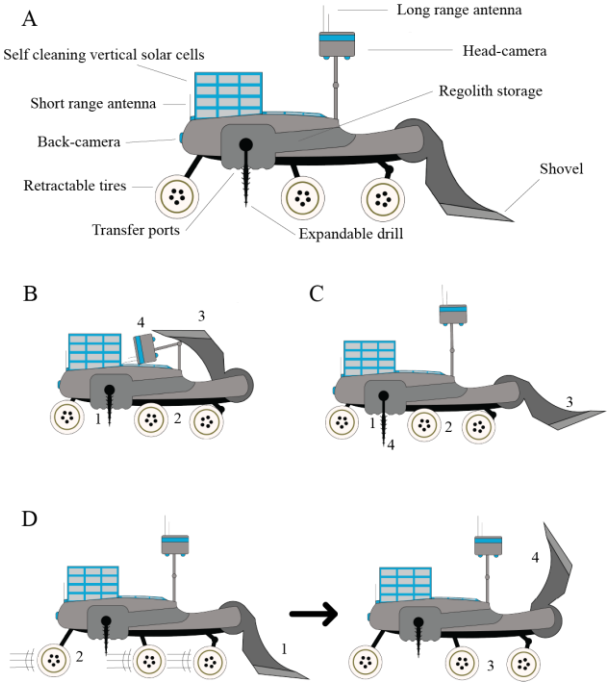

Fig.7. Rover positions and tasks. A: Overview of the instruments and parts of the rover. B: Rover in transport-position with open transfer ports (1), retracted legs (2), folded shovel (3) and head camera (4). C: Rover in drilling position with closed transfer ports (1) retracted legs (2), half-folded shovel (3) and active drill (4). D: Rover in shoveling position starting with a grounded shovel (1) and a forward movement (2). Ending in a standing position (3) with a raised shovel (4)

4.2.1 Transport & night position

The rover is designed to be transported inside the lander and is, therefore, foldable (Fig 7B). In this position, the legs are retracted which leads to a blocking of the tires. The drill is retracted not to scratch the lander nor to get damaged throughout the journey. The head camera is inactive and folded together with the shovel. In this position, the rover is unlikely to slide around and can withstand the launch and landing (with additional support). After the landing, the legs are partly extended to enable the rover to move out of the lander. The back camera can be used for this maneuver.
To avoid damaged electronics and to add new lunar regolith to the lander the rover approaches its indoor position during lunar nights as well. The rover is moved backward towards the gateway to enter the lander, the head camera is deactivated and folded together with the shovel. The legs are retracted to the minimal moving position, and the rover is navigated autonomously into the lander. Its regolith transfer ports are opened, and the gathered material drops into the landers storage.

4.2.2 Drilling position

The head camera is used to move the rover to a previously screened and for drilling suitable location. As soon as the drill is aligned the rover retracts its legs, blocks the tires and the shovel gets half-way folded. After the rover is in a stable position, the drill starts spinning and is extended to engage the ground. The material is moved throughout the drilling process into the storage area of the rover. The so-gathered material will be transported back to the lander (Fig. 7C). The drill concept is mainly to acquire regolith and, therefore, the drilling depth is limited to a few centimeters.

4.2.3 Shovel position

The head camera is used to move the rover to a previously screened position and for shoveling suitable location. The shovel is moved downwards to touch the ground, and the rover is moved slowly forward to gather material in the shovel.

5. Power supply and thermal shielding

The design of the electrical power supply unit for the mission is crucially dependent on diverse mission drivers; the most important of which is the selection of the landing site. In fact, a selection of a permanently illuminated site would probably push the selection of the power source as mainly solar. However, given the considerations in section 2.2, it is best to strive for a design which is as robust and flexible as possible, that is, which could be re-adapted for different environments. So, the main power source would be based on RTG technology, with additional support coming from solar panels for the rover.

The main components of the system are:

- The bioreactor: running at 30-40 °C with short peaks of 100 °C.
- The water tank: high volume of water, which shall be reused. Heating the bioreactor to 100 degrees and refilling the tank after each cycle.
- Pumps:
  (2) Airflow pumps (< 5 L) to move the gas through the reactor into the inflatable module.
  (4) Waterflow pumps (< 5 L) to move the liquid to the reactor and mix it with the medium and regolith.
- Extraction mechanism: magnetic or weight dependent extraction (i.e. electromagnet), and/or small centrifuge.
- Freezer for the different cell types and scientific/analytical instruments
- Communications system.
- UV lamps for sterilization.
- Controlling electronics: automatic controlling of the valves, pumps, heating system with potential human interference from Earth.

5.1 Lander/Bioreactor power supply

The power requirement for the bioreactor and attached systems is estimated at 2.4 kW (including all previously mentioned systems). The power will be mainly supplied via radioisotope thermoelectric generator (RTG) technology. In fact, as per Section 2.2, even on a semi-permanent illuminated area, it is too optimistic to rely on solar panels only, with a rover that would move dust and then potentially cover the solar panels partially with Lunar dust. The need for a high-efficiency thermoelectric transformation technology appears to be still crucial. Currently, the most realistic method is to utilize the RTG, which is based on the Seebeck principle to transform the heat energy into the electric energy. When generating electric power, the RTG can also supply great heat energy to regulate temperature. Generally, Pu-238 is used as the RTG source[43].

However, to strive for a more sustainable form of energy supply in the framework of a Lunar habitat, the potential solar panels will be designed to satisfy most of the power requirements. It is important to remark that the initial demonstration mission will rely on RTG, which is more reliable at the moment. Since the Earth-Moon system is in heliocentric orbit, they share a similar solar constant of about 1.36 kW/m^2 [44]. Given the current Solar panel technology and considering solar cells arrangement, an efficiency of about 30% can be considered, which gives roughly 330 W/m2 on state-of-the-art spacecraft. So, producing 2.4 kW of power would require approximately 7.2 m2 of solar panels with a weight of 40 kg. Considering the environmental conditions of a Lunar south sole landing site [see section 2.2], focus shall be set on Sun illumination angles: while Azimuth varies 0-360 degrees in a Moon cycle (~27 days), elevation angles features much lower variations during the same period. The power system design features the selection of a "Power Generation Area", which shall be as flat as possible to allow for Sun tracking during the entire mission lifetime and avoid possible panel shadings due to orography. The solar panel will track the Sun only in Azimuth, neglecting the small variations in Elevation.

In order to support this sun tracking feature, the base of the panel will be equipped with a DC motor [45], the rotation speed needed is affordable for state of the art components (360 deg in 27 days is a very slow rotation). The solar panels will gather sun power and store it in rechargeable batteries; this power will be used for Lunar nights and in case of contingency (e.g. low/no power available due to malfunctions, dust on cells, etc.).

5.2 Rover power supply

The rover will be designed to be a smart and simple machine, with the purpose in mind to achieve a mobility

item that is agile, rapid, and simple. It is crucial to reduce at most its complexity and its weight: the rover will be powered by a combination of solar panel and rechargeable batteries, which are used to maintain system survivability during times without sun illumination (e.g. shadowed areas). In such cases, the rover will activate a series of heaters to allow it to resist to the low temperatures and possibly move back to an illuminated area.

6. Discussion & conclusion

Space agencies all over the globe are proposing return missions to the moon and the lunar vicinity with the ultimate goal of human exploration and habitation. Our understanding of how biological systems interact with the very hostile environmental factors on the lunar surface is insufficient and only based on the short visits during the Apollo missions. This data, however, is essential to understand the behavior of life support systems, biomining or bioleaching approaches and of course the health risks posed to astronauts throughout a prolonged visit. The mission proposed here will act as a test-bed to gather such data and findings for a variety of biological systems as will also act as a technical demonstration in a closed environment on the lunar surface.

The summit of the Malapert Mountain, near the South Pole, was considered the best location for this mission. Being designed as precursor research, it makes sense for our mission to take place on a site that is already evaluated for a future settlement. With its exceptional environment in term of illumination, temperature, and topography, this site does not only provide excellent conditions for operations, but it is also close to numerous areas of scientific interest.

The rover and the lander are well-equipped to test critical ISRU processes such as the transport of lunar regolith from the surface into a closed environment, shoveling as well as low-depth drilling operations to gather regolith, controlled liquid as well as gas-flow under low gravity and the mixture of telerobotic with autonomous operations of the rover.

Direct extraction and production experiments done via microbial processes will build on prior tests with regolith simulant here on Earth. There are several proposals to use microorganisms as nano-factories and to produce, for example, on-demand medication, extract materials from the regolith or produce gases for life-support systems and as rocket fuel.

Our successful tests to bind silicon from lunar regolith simulant onto our genetically engineered bacteria showed that 13.4 ± 0.9% of the total cells are covered with silicon in comparison to 1.3 ± 0.2% in control with no modified bacteria. Currently, we are investigating methods to extract the silicon-covered bacteria from the rest of the regolith. Also, experiments with magnetic iron extraction were improved from 3.34 ± 0.58 mg to 7.25 ± 1.83 mg after a 48-hour treatment with bacteria using a differently modified *E .coli*.

The experimental set-up and scientific instrumentation allow for an analysis of the biotoxicity caused by lunar dust and lunar regolith particles in general. The simultaneous running experiment in LUNA at the EAC in Cologne will directly compare these results to bacteria grown with regolith simulant. We expect to have higher toxicity due to the sharp edges and small sizes occurring in lunar regolith. Experiments with eukaryotic cells or even cell cultures might enable even further insights into their toxicity on humans.

Different approaches for the power supply of the system and its communication with earth are discussed. To our knowledge, this is the first mission architecture designing a bioreactor for the lunar environment and enabling biological tests in the sectors life-support-systems, biotoxicity, biological ISRU and technological demonstrations. These are critical for a human presence on another planet or moon.

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