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Supporting Mars exploration: BIOMEX in Low Earth Orbit and further astrobiological studies on the Moon using Raman and PanCam technology

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The Low Earth Orbit (LEO) experiment *Bio*logy and *M*ars *E*xperiment (BIOMEX) is an interdisciplinary and international space research project selected by ESA. The experiment will be accommodated on the space exposure facility EXPOSE-R2 on the International Space Station (ISS) and is foreseen to be launched in 2013. The prime objective of BIOMEX is to measure to what extent biomolecules, such as pigments and cellular components, are resistant to and able to maintain their stability under space and Mars-like conditions. The results of BIOMEX will be relevant for space proven biosignature definition and for building a biosignature data base (*e.g.* the proposed creation of an international Raman library). The library will be highly relevant for future space missions such as the search for life on Mars. The secondary scientific objective is to analyze to what extent terrestrial extremophiles are able to survive in space and to determine which interactions between biological samples and selected minerals (including terrestrial, Moon- and Mars analogs) can be observed under space and Mars-like conditions. In this context, the Moon will be an additional platform for performing similar experiments with negligible magnetic shielding and higher solar and galactic irradiation compared to LEO. Using the

Moon as an additional astrobiological exposure platform to complement ongoing astrobiological LEO investigations could thus enhance the chances of detecting organic traces of life on Mars. We present a lunar lander mission with two related objectives: a lunar lander equipped with Raman and PanCam instruments which can analyze the lunar surface and survey an astrobiological exposure platform. This dual use of testing mission technology together with geo- and astrobiological analyses will significantly increase the science return, and support the human preparation objectives. It will provide knowledge about the Moon's surface itself and, in addition, monitor the stability of life-markers, such as cells, cell components and pigments, in an extraterrestrial environment with much closer radiation properties to the surface of Mars. The combination of a Raman data base of these data together with data from LEO and space simulation experiments, will lead to further progress on the analysis and interpretation of data that we will obtain from future Moon and Mars exploration missions.

1. Introduction

The search for life or fossil remnants of life on other planets is a prime goal of future space missions due to its high scientific and philosophical relevance. The possibility that organisms could survive in harsh environments outside the Earth was investigated in previous experiments (Horneck et al., 2010), see also (Table 1). Spaceflight and ground based experiments have shown that especially micro-colonies of bacteria, meristematic black fungi and symbiotic associations of microorganisms such as lichens are able to survive and to be reactivated after space experiments (de la Torre et al., 2007, 2010; de Vera et al., 2003, 2004a,b, 2007, 2008, 2009; de Vera, 2005; Horneck et al., 1994; Kordyum et al., 1983; Olsson-Francis et al., 2009; Onofri et al., 2008, 2009; Sancho et al., 2007; Tarasenko et al., 1990). Bacteria strains, such as *Bacillus subtilis* and *Deinococcus radiodurans*, are radiation and vacuum tolerant (Horneck, 1993; Horneck et al., 1994, 2001; Moeller et al., 2007a,b,c). Gram-negative endophytic bacteria and cyanobacteria survived during a 14 day-shuttle flight and their co-existence with plants exposed to microgravity showed enhanced plant colonizing activity (Tarasenko et al., 1990). During the BIOPAN 5 and 6 experiments the lichens *Rhizocarpon geographicum*, *Xanthoria elegans* and *Aspicilia fruticulosa* (renamed as *Circinaria gyrosa*) were analyzed after an exposure time to space conditions of about 10–16 day and parallel ground based tests were performed. The results after flight and simulation experiments lead to the conclusion that the tested symbiotic eukaryotic associations of alga and fungi in the lichen were not seriously damaged and nearly 70%–90% of the tested lichens have survived. They were physiologically active and able to germinate and grow. Some investigations on the mutation rate expressed by photoproducts produced in the DNA have also shown that the fungus was practically not affected by UV radiation but the algal symbiont was more sensitive (de la Torre et al., 2007, 2010; de Vera et al., 2003, 2004a,b, 2007, 2008, 2009, 2010b; de Vera, 2005; de Vera and Ott, 2010a; Sancho et al., 2007). Cyanobacteria were also able to survive Low Earth Orbit (LEO) environment and simulated extraterrestrial conditions, as shown by analysis of the akinetes (resting-state cells of cyanobacteria, Olsson-Francis et al., 2009). Vegetative cells of *Chroococcidiopsis* sp. CCMEE 029 survived prolonged desiccation (Billi, 2009) and a few minutes of exposure to unattenuated martian UV flux (Cockell et al., 2005). The cells also survived, shielded by 3 mm of sandstone, in real space as well as simulated martian conditions representing 1.5 years exposure in LEO (Billi et al., 2011). Moreover, these same vegetative cells associated with an epilithic microbial community survived 548 day on the EXPOSE-E facility (Cockell et al., 2011). Numerous species mentioned here are even able to survive simulated asteroid impacts (Horneck et al., 2001, 2008; Meyer et al., 2011; Stöffler et al., 2007). Mars simulation tests on methanogenic archaea have also shown a remarkable level of survival (Morozova and Wagner 2007; Morozova et al., 2007). The same has been observed for meristematic black fungi during a ground based experiment in the space simulation facilities at DLR Cologne named EVT (Experiment Verification Test), performed in preparation for the recent and completed EXPOSE experiments (Onofri et al., 2008). They survived also further exposure to real space conditions on the ISS with a total exposure time of 1.5 years (Onofri et al., 2012). In other ground-based experiments, we were able to demonstrate that *Paenibacillus* sp. IMBG221 caused bio-corrosion of an anorthosite rock—a Moon analog (Lytvynenko et al., 2006). This brief overview shows that a wide variety of different microorganisms, including evolved ones, is able to resist and to survive space conditions for certain periods of time.

Table 1
Survival in Mars and space environment

Tested organisms	Mars atmosphere	Vacuum	UV/space radiation	Shock experiments
Archaea				
Methanosarcina	+	+	+/−	n.d.
Bacteria				
Bacillus subtilis	+	+	+	+
Deinococcus radiodurans	+	+	+	n.d.
Endophytic gram⁻ bacteria	+	+	+	n.d.
Cyanobacteria (*e.g. Chroococcidiopsis*)	+/−	+/−	+/−	+/−
Lichens				
Aspicilia fruticulosa (*renamed*: *Circinaria gyrosa*)	+	+	+	n.d.
Rhizocarpon geographicum	+	+	+	n.d.
Xanthoria elegans	+	+	+	+
Merismatic black fungi				
Cryomyces antarcticus	+	+	+	n.d.

(+) Positive survival; (−) No survival; (n.d.) Not determined.

2. Experimental approach of BIOMEX—a step further

BIOMEX is an exposure experiment which will be performed on the exposure facility EXPOSE-R2 attached to the Russian Zveszda module of the International Space Station (ISS). The Launch is foreseen for mid-2013. The planned exposure duration is expected to be in maximum 1.5 years. EXPOSE-R2 is a hardware payload built by Kayser-Threde, where the samples can be placed into sample holders and exposed outside the module (Fig. 1). The concept of this experiment is an improvement on the previously executed experiments in Section 1.

2.1. First scientific objective of BIOMEX

Its primary objective is to pay more attention to the structure and stability of most of the biological components, especially to the protecting surface coats, membranes, proteins, pigments or secondary metabolite deposits on cell surfaces. In previous exposure experiments, these investigations were incomplete with respect to the identification of space-resistant biosignatures, although the

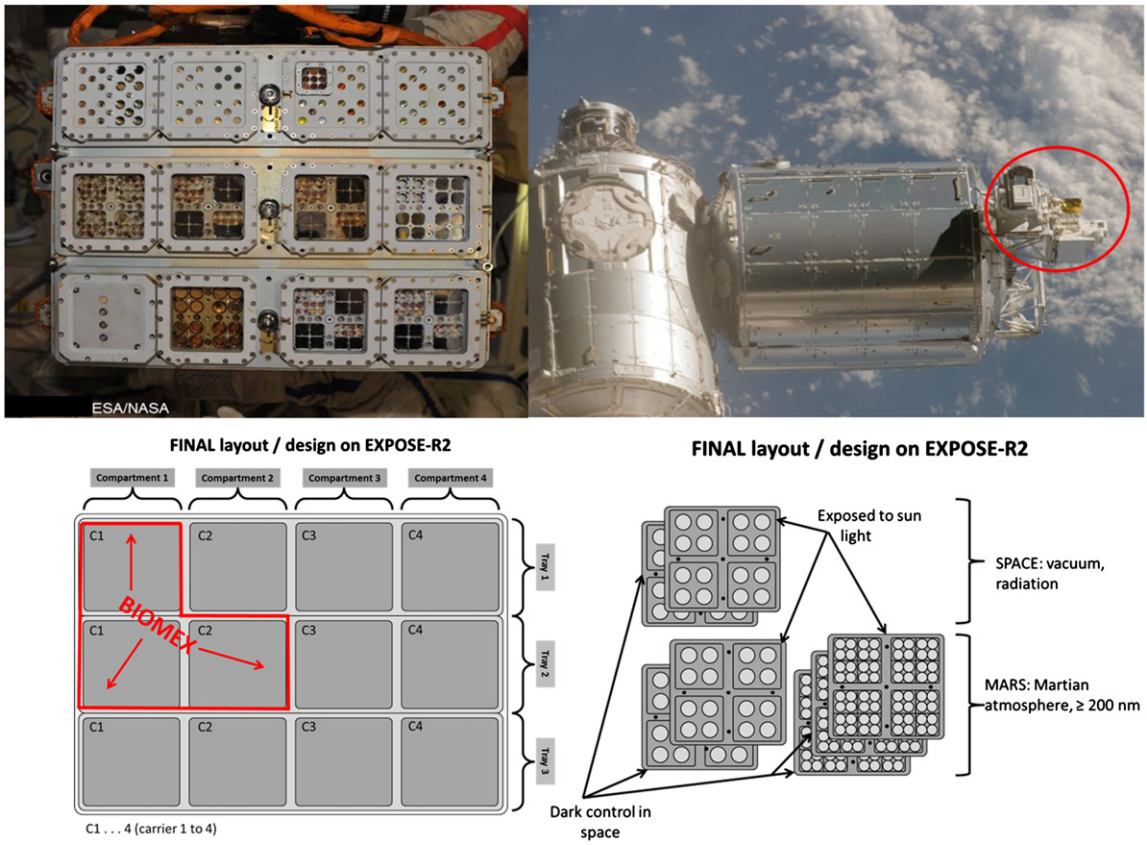


Fig. 1. EXPOSE R (Zvezda module/ top left) and EXPOSE-E on racks of Columbus module (top right, red circle); position of BIOMEX experiment in the EXPOSE-R2 hardware (bottom left) and sample holders (bottom right).

results we might get by such a systematic analysis are of high relevance for the characterization and detection of potential biosignatures of recent/extant microorganisms on future missions to Mars. Another reason for using the BIOMEX experiment is to deepen these investigations and to analyze the effect of space environment by using the biological samples in contact with Lunar Regolith analog rocks, such as anorthosite (Mytrokhyn et al., 2003) and two Mars regolith analog mixtures with the characteristics of phyllosilicatic Mars Regolith soils (P-MRS) and sulfatic Mars Regolith soils (S-MRS) reflecting alteration minerals formed during different environmental conditions of the Noachian and Hesperian/ Amazonian, respectively (Böttger et al., 2012). These two Mars analog mineral mixtures have been developed by the Naturkunde-museum Berlin in the framework of the Helmholtz-Alliance "Planetary Evolution and Life". The production of the analogs is based on recent Mars data and publications (Bibring et al., 2005, 2006; Chevrier and Mathé, 2007; Poulet et al., 2005). The effects of space conditions, together with simulated lunar conditions and Mars-like environment, on rock and mineral analogs is an important parameter for optimizing exploration procedures focusing on the detection of mineral composition as a context for biological investigations. Hence space exposure experiments, such as presented by the concept of BIOMEX, might help to avoid possible pitfalls which might occur due to possible overlapping of signals during spectroscopic detection procedures. The influence of interfering mineralogical features or changes in the molecular conformation of biological substances after exposure to the space environment on the ISS may be identified. Such experiments will greatly aid to identify if signatures of extinct or extant life are present on another planet.

As mentioned above, a number of pigments and membrane components, representing bio-relevant substances extracted from cells and embedded in the Mars and Lunar regolith analog mineral pellets are also part of the experiment. This will provide valuable information about the stability of the pigments before and after space exposure and might also help to determine the location of protected bio-signatures in Mars-like soils.

2.2. *Second scientific objective of BIOMEX*

The BIOMEX space experimental platform provides an ideal opportunity to pursue a second objective: the samples used can also test the viability and space resistance of microorganisms in the context of (Litho-)Panspermia (Arrhenius, 1903, 1908, 1918; Chyba and Sagan, 1992; Martins et al., 2008). This concept was tested in space experiments on FOTON/BIOPAN (Sancho et al., 2007; de la Torre et al., 2010) and more recently on the latest EXPOSE missions on the ISS. Replicates are needed, as well as data during different phases of the sun's activity for a re-evaluation of the Lithopanspermia theory. In addition to replicates, a new set of microorganisms, representing species of all three main branches of the tree of life have been chosen (Fig. 2), in particular microorganisms which are known to be relevant for Mars (*e.g.* methane producing archaea, cyanobacteria and iron bacteria/ Fig. 3).

Currently, a number of space simulation experiments, the Experiment Verification Tests (EVTs) and Scientific Verification Tests (SVTs), are being performed and are in continuation at the space simulation facilities at DLR Cologne to aid sample preparation and the selection processes of the best samples to be sent to space. The preliminary results indicate a high survival rate of the investigated organisms and the stability of their pigments. Therefore the final arrangement of the sample distribution in the space hardware will not differ much from the layout presented in Figs. 2 and 3.

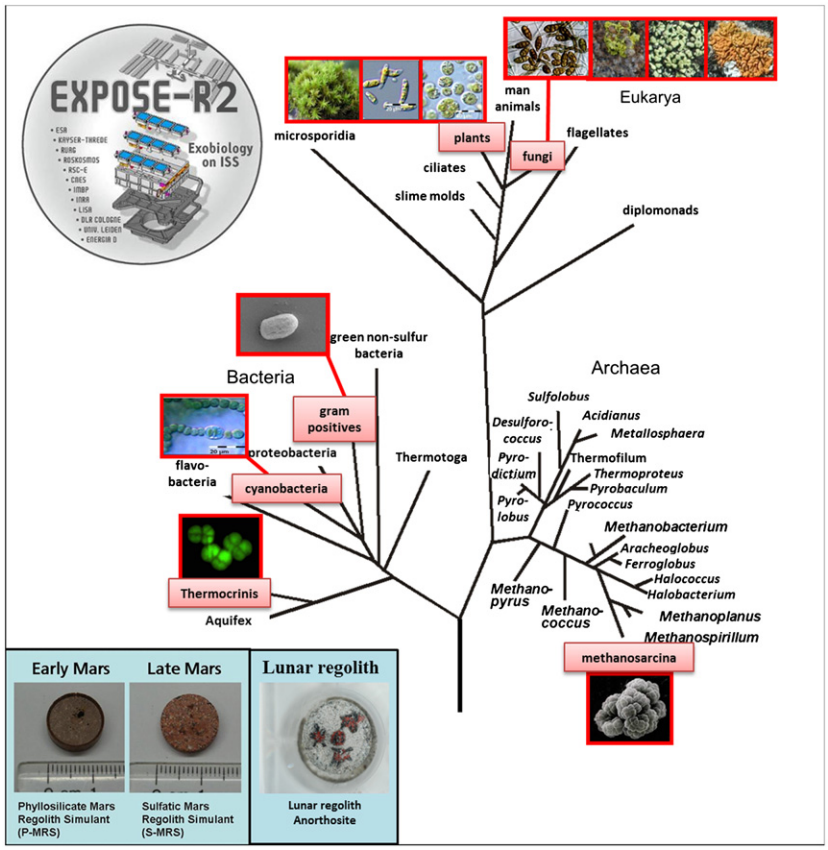


Fig. 2. The rRNA analysis-based phylogenetic tree of terrestrial life including the selected organisms for the BIOMEX space experiments onboard the ISS. Bottom left: mineral pellets selected for BIOMEX.

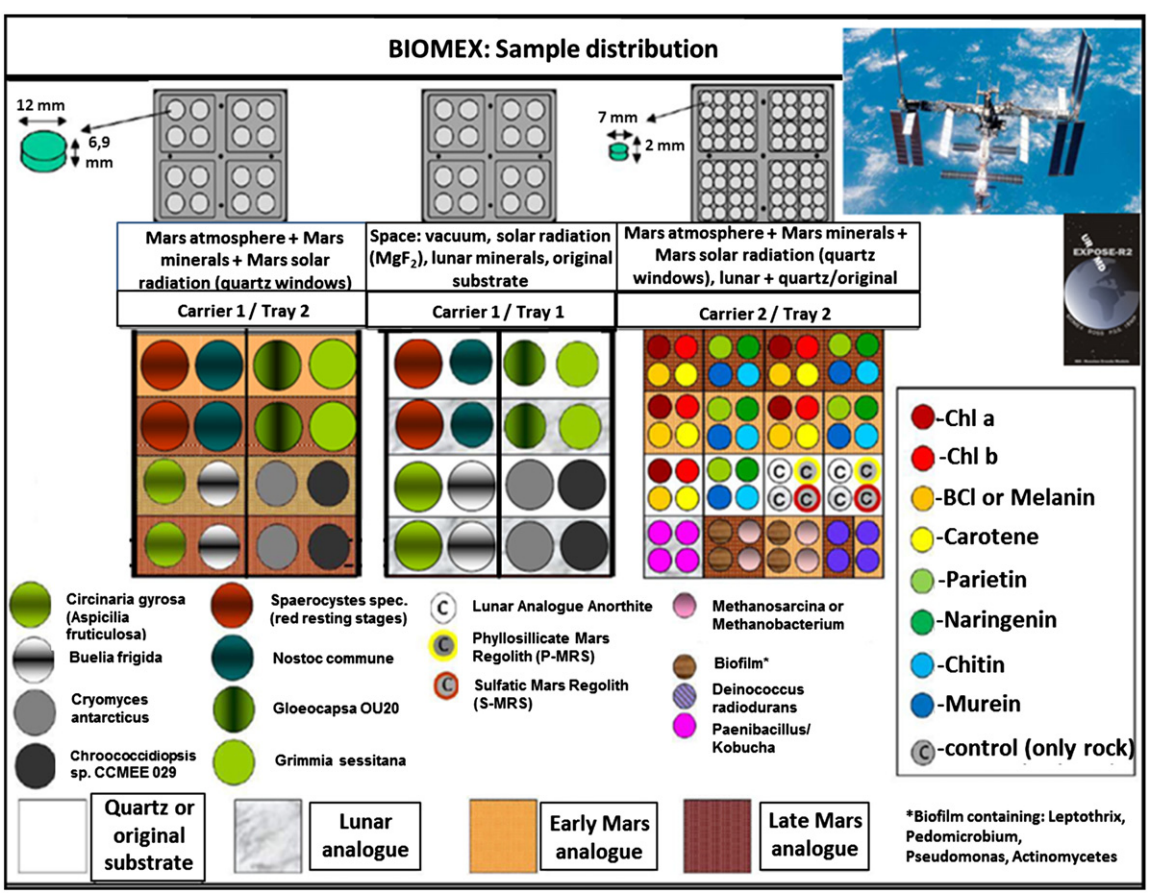


Fig. 3. Sample positions in hardware and parameter description.

3. The relevance of exposure experiments on the Moon

BIOMEX is designed as a precursor experiment for follow-on geo-biological studies on the Moon and as preparation and support for future Mars exploration missions as described in the original proposal (ILSRA-2009-0834).

In response to ESA's call for letters of interest and proposals for a Lunar Lander, the national and international HGF-Alliance "Planetary Evolution and Life" and its cooperating teams have submitted a proposal, named LOGOS (Lunar-Organisms and Geology Operation Sciences), emphasizing research on space weathering on organisms and rocks.

The LOGOS-Cube is designed as a package which has a closely connected context to the present BIOMEX experiment in LEO. It is intended to serve as an exposure platform on the Moon for organisms, organics and mineral surfaces of different rocks with the same scientific focus as described for the BIOMEX experiment. The LOGOS-Cube will obtain information on the effect of space weathering on organisms and rocks and the degradation or resistance of organics produced by life forms, thus contributing to our understanding of relevant life markers/bio-signatures as a reference to the search for life on other planets. The Moon is a natural vantage platform with an enhanced radiation exposure due to the absence of both, an own magnetic field and an atmosphere. The data expected from the present BIOMEX project on the ISS will facilitate identification of the most suitable samples for lunar orbiter or lander experiments and will provide reference data for future Moon experiments. These data will be extremely useful in support of future missions to Mars to search for biosignatures as evidence for remnant or fossil and therefore extinct life and for the search of extant life on other Solar System bodies.

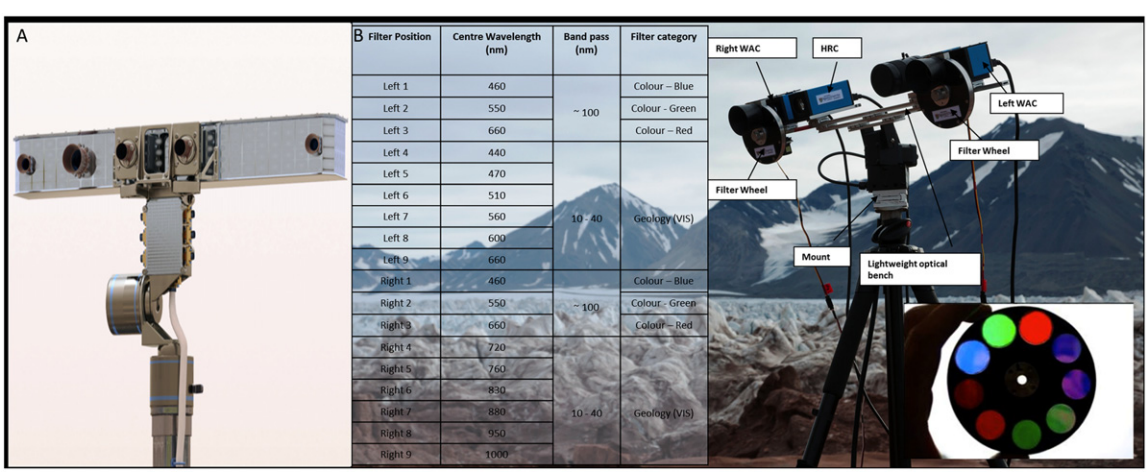
4. Our proposal: biosignature analysis combining PanCam and Raman in situ measurement devices on one exposure-platform

For a new lander with multifunctional technology tests and additional scientific outcome, our suggestion is to combine existing and space proved exposure platforms and technologies with a Technical Readiness Level (TRL) of about 6–9. We propose to use a small version of the EXPOSE platform, as used on the ISS, with a maximum of 3 carriers in a compartment which could serve as sample holder for geo-biological samples as shown in Fig. 1. One additional sensor carrier will be used to collect temperature and radiation data. This particular exposure device is space proven and its TRL level is almost 9 (Rabbow et al., 2009). In addition, *in situ* measurements in the visible and multispectral wavelength are required. Therefore, instruments such as a multispectral camera, sensitive in the visible (VIS) and near infrared (NIR) range, as well as fluorescence, Raman and IR spectrometers are needed for detection of the spectral characteristics of life forms and biosignatures and for the observation of their behavior during the exposure time (*e.g.* position and possible changes of specific detected peaks in different spectral ranges previously determined as specific bio-relevant finger prints of, for example, unaffected pigments can indicate changes and destruction of the molecular structure during space exposure (Dartnell et al., 2012))".

4.1. Panoramic wide angle multispectral Stereo Camera (PanCam) combined with IR and Raman spectroscopy

One of the proposed instruments which can be helpful to support this task is a panoramic wide-angle multispectral stereo camera (PanCam, Coates et al., 2012; Griffiths et al., 2006) with TRL 6–7 that has been selected as part of the ExoMars rover (Vago and Kminek, 2007; Cousins et al., 2010). A similar device is also already a part of the model payload of ESA's Lunar Lander (Carpenter, 2011).

This camera system (1) delivers the geological context of the landing site (cf. Bell et al., 2004; Smith et al., 1997) and (2) helps to identify the best locations for further study via multi-level analyses (Vago, 2005). Further capabilities of this system include multispectral analysis in the VIS/NIR (400–1000 nm) range that provides compositional information. The final optical characterization is done by a close-up imager inspection of surfaces at mm- or sub mm-scale (such as ExoMars PanCam HRC) or microscopically (Clupi on ExoMars (Josset et al., 2012), MAHLI (Edgett et al., 2005) on MSL). For ExoMars, the PanCam (Fig. 4A and B) consists of two wide angle cameras (WACs), which form a stereo pair and a narrow angle high resolution camera (HRC). Its first and foremost task in any payload is the delivery of geological context information. Geological information that can be gathered from greyscale WAC and HRC images includes outcrop context, structure, morphology, and brightness. Such knowledge can be used to make a preliminary classification of rock type and its weathered state. Additionally, the WACs are equipped with 3 wideband RGB filters and 11 narrow band (30–40 nm) geological filters to provide color and visible and near infrared spectral data, respectively. This provides both mineralogical and compositional information, which, in combination with morphology, structure and context helps for interpretation of the lithology of the landing site. In the context of life detection missions,



Filter Position	Centre Wavelength (nm)	Band pass (nm)	Filter category
Left 1	460	~ 100	Colour – Blue
Left 2	550		Colour - Green
Left 3	660		Colour – Red
Left 4	440	10 - 40	Geology (VIS)
Left 5	470		
Left 6	510		
Left 7	560		
Left 8	600		
Left 9	660		
Right 1	460	~ 100	Colour – Blue
Right 2	550		Colour - Green
Right 3	660		Colour – Red
Right 4	720	10 - 40	Geology (VIS)
Right 5	760		
Right 6	830		
Right 7	880		
Right 8	950		
Right 9	1000		

Fig. 4. (A) Drawing of the ExoMars PanCam instrument accommodated on a Pan-Tilt Unit on top of the ExoMars rover for multiple use. (B) Field Model of the ExoMars Panoramic Camera, as used during the Arctic Mars Analog Svalbard Expeditions (AMASE), with a range of VIS/NIR filters for multispectral composition analysis.

such as ExoMars, the utilization of recorded VIS/NIR spectra is especially important to assess astrobiological objectives such as habitability condition like detection of hydrated minerals that might indicate past aqueous activity (Fig. 4A and B). In addition, the same instrument can be utilized for visible and multispectral monitoring of samples on an EXPOSE-like exposure platform.

Multiple spectrometers can be mounted on the mast in addition to PanCam including IR- and Raman-spectrometers; which could be used for (3) *in situ* measurements of the exposed geo-bio-samples (see Fig. 5), as well as geological investigations of the Moon's surface.

5. Operation scenarios

We have envisaged three different scenarios for operations during the lander mission (Fig. 5):

- Option (1) is a movable, *i.e.* in- and out-side gliding exposure platform which exposes the geo-biological samples to the lunar environment (vacuum, radiation) and upon which the samples can be analyzed by a PanCam (outside) and a Raman/LIBS-spectrometer accommodated inside the lander (TLR 6–9, ISS and ExoMars techniques).

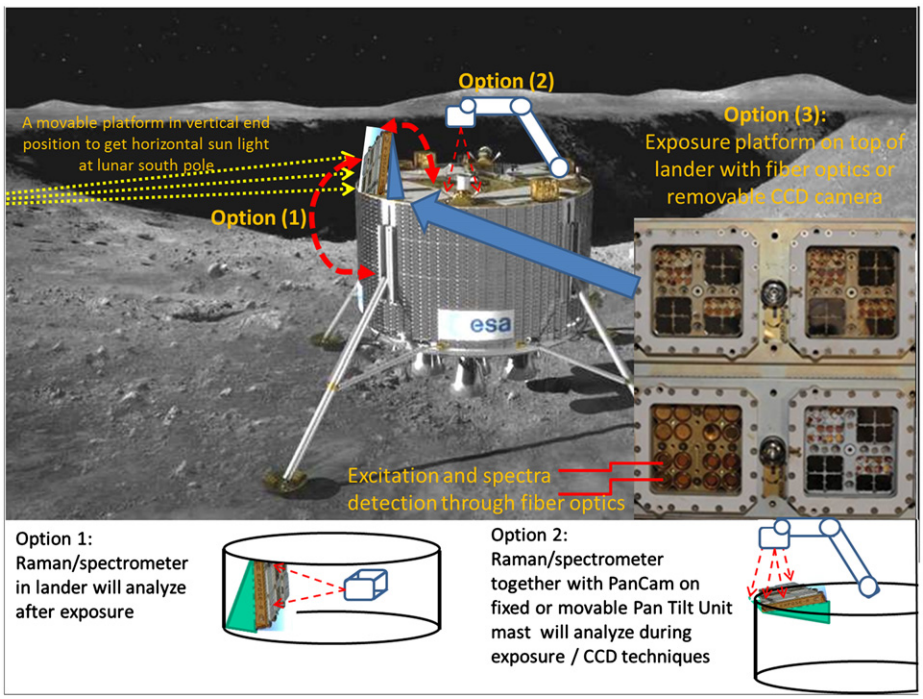


Fig. 5. The 3 options of operation scenarios to combine remote and *in situ* measurements.

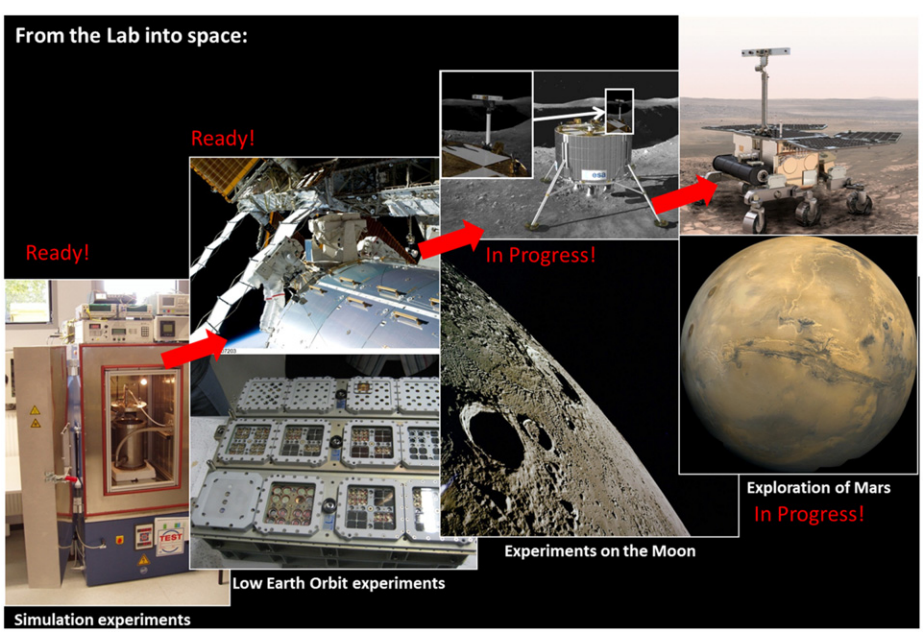


Fig. 6. The way to Mars exploration: the necessity and relevance of technology improvement and science tests in simulation facilities, in Low Earth Orbit and on the Moon.

- Option (2) is the use of the PanCam instrument combined with different spectrometers including Raman and IR on top of a tiltable mast (Fig. 4A and Fig. 5) that can analyze the landing site geology as well as monitor changes in the geo-biological samples on the exposure platform during the mission, as previously described (TLR 6–9, ISS and ExoMars techniques).
- Option (3) is an exposure platform (TRL 9) on the top of the lander which has fiber optics in the exposure sample holders or a removable CCD camera with a connection to an internal data logger. But this option needs further investigation and the present TRL is at 5–6.

Our preferred option is (2) because of its multi-functionality and the high expected scientific outcome. Another advantage is the possibility of using a similar but smaller version of the exposure platform, as used on the ISS, because the carriers with the samples are also easily removable. This might be relevant for future lunar manned space missions, where astronauts can remove or exchange the carriers from the lander to transfer the samples back to Earth for further investigations. Additional techniques that would not be possible to be conducted on the Moon during the lander mission because of technical and mass restrictions can be applied in ground-based laboratories.

6. Conclusion

The presented technology is available and the individual components have a high level of TRL. The new combination of existing and already applied techniques in LEO, as well as tested instruments for other space missions comparable to Mars missions, together with new innovative instrumentation, will support the ESA lunar lander objectives to prepare for human lunar exploration and will lead to high and valuable scientific outcome including astro-/exo-biology investigations.

In particular, data from the BIOMEX experiment obtained from spectrometers, such as Raman technology, will serve as reference for future exposure experiments on the Moon surface. By comparing data from LEO with data obtained on the surface of the Moon, we will improve our knowledge regarding the stability of selected biosignature spectra in high radiation environments similar to those expected on the surface of Mars. For such a proposed lunar lander mission we do not have to reinvent the wheel but can use resources, know-how and knowledge already available due to innovative enterprises in LEO and developed for Mars exploration studies (see Fig. 6).

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