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BIOPAN experiment LICHENS on the Foton M2 mission Pre-flight verification tests of the *Rhizocarpon geographicum*-granite ecosystem

R. de la Torre Noetzel [a,*], L.G. Sancho [b], A. Pintado [b], P. Rettberg [c], E. Rabbow [c], C. Panitz [c], U. Deutschmann [d], M. Reina [a], G. Horneck [c]

[a] *Department of Earth Observation, Area of Atmospheric Research and Instrumentation, INTA, 28850 Torrejón de Ardoz, Madrid, Spain*

[b] *Department of Plant Biology-II, Faculty of Pharmacy, University Complutense of Madrid, 28040 Madrid, Spain*

[c] *Institute of Aerospace Medicine, German Aerospace Center DLR, 51170 Köln, Germany*

[d] *A.C. Hispano-Alemana, Alcobendas, Madrid, Spain*

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Abstract

The objective of the LICHENS space experiment within the BIOPAN facility of ESA on board of the Russian Foton satellite is to test the limits of life in the hostile environment of space, and the effects of selected parameters, such as space vacuum and specific wavelength bands of extraterrestrial UV-radiation, on the viability of lichens. In preparation of this space experiment, pre-flight verification tests were performed with the lichen system *Rhizocarpon geographicum* on its natural granite substrate using the Planetary and Space Simulation Facilities at the DLR in Cologne, Germany. The test parameters (high temperature, UV-radiation, vacuum) were adjusted to the conditions expected to be experienced during the 15 days long space mission of LICHENS. After exposure, the maximum quantum yield of photosynthetic activity was determined as a measure of survivability of the lichen. The lichen *R. geographicum* was capable to fully recover from exposures to vacuum (up to 3×10^{-6} hPa for 1 week), to the full spectrum of UV-radiation (>200 nm for up to 20 h giving a final fluence of 10.89×10^6 J/m^2), as well as to a combined treatment with both simulated space parameters. It was less resistant to heating: after 1 week at 40 °C the photosynthetic activity was reduced by about 20%. The results demonstrate the high resistance of the *R. geographicum*-granite ecosystem to simulated space conditions and justify its use in the LICHENS space experiment. It might even be capable of coping with the intense influx of extraterrestrial solar UV-radiation, which so far no biological system was able to withstand.

Keywords: Lithopanspermia; Lichens; Space experiment; Photosynthetic activity; Survival

1. Introduction

Astrobiology aims at identifying the pathways through which life arose, evolved, and may have been distributed throughout the universe. With the ability to expose micro-organisms to the space environment in a controlled manner (reviewed in Horneck, 1999; Horneck, in press) the opportunity has arisen to explore questions related to the hypothesis of interplanetary transfer of life (Arrhenius, 1903). Possible mechanisms of interplanetary transfer include spacecraft, or natural mechanisms such as meteorites, comets or interplanetary dust particles. Interplanetary transfer of life is particularly relevant with regard to Mars and Earth because it is thought that the SNC meteorites traveled from Mars to Earth, and possibly meteorites from Earth traveled to Mars, providing a pathway for the transport of biological material between these two planets (Melosh, 1988; Mileikowsky et al., 2000). This transport process with the aid of meteorites has been called "Lithopanspermia". It requires that microbial communi-

* Corresponding author. Tel.: +34 91 520 1589; fax: +34 91 520 1317.
E-mail addresses: torrenr@inta.es (R. de la Torre Noetzel), sancholg@farm.ucm.es (L.G. Sancho), petra.rettberg@dlr.de (P. Rettberg), elke.rabbow@dlr.de (E. Rabbow), corinna.panitz@dlr.de (C. Panitz), gerda.horneck@dlr.de (G. Horneck).

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ties colonizing the interior of rocks survive three consecutive steps: (1) escape from the planet of origin; (2) travel through space; and (3) capture by and landing on another planet.

Whereas previous space experiments on the survival of microorganisms in outer space mainly dealt with bacterial endospores (Horneck, 1993), we have selected a microbial rock colonizer, the lichen *Rhizocarpon geographicum* for those studies. This bipolar epilithic lichen grows at high mountain regions with continental climate (e.g., Sierra de Gredos, Central Spain) as well as in the Arctic and Antarctica. *Rhizocarpon geographicum* is the most abundant lichen species in the Sierra de Gredos in zones with long periods of snow cover.

Lichens are poikilohydric organisms, i.e. they are capable of adopting a latent state, if environmental conditions become extreme. Examples are extreme desiccation, high influx of solar UV-radiation, and extreme high or low temperatures. In this latent state, lichens become much more resistant to the environmental stress than in the active state, even if this stress lasts for quite long periods. So far, most studies on the resistance of lichens have focused on their responses to very low temperatures, concentrating on the extreme adaptability of bipolar lichen species to the harsh Antarctica and Arctic regions (Ott and Sancho, 1993; Schroeter and Scheidegger, 1995). The bright colors of alpine lichens are caused by phenolic acids, which act as UV screening pigments shielding the thalli from the intense UV-radiation prevailing in high altitude habitats (Gauslaa and Ustvedt, 2003; Solhaug et al., 2003).

The lichen system *R. geographicum* has been selected for the LICHENS space experiment within the BIOPAN facility of ESA (Demets et al., 2005) on board of the Russian Foton satellite. The objectives of this space experiment are to test the limits of life in the hostile environment of space, and the effects of selected parameters, such as space vacuum and specific wavelength bands of extraterrestrial UV-radiation, on the viability of the lichens (de la Torre et al., 2004). Foton M1, which carried the experiment LICHENS, exploded during the launch in October 2002. The reflight of the LICHENS experiment was successfully performed on board of FOTON M2 in May–June 2005.

It is a prerequisite for every space experiment to test before flight the responses of the biological systems to simulated space parameters. We report here on the results of tests performed at the Planetary and Space Simulation Facilities of the DLR (de la Torre et al., 2002, 2004; Rabbow et al., 2005). *Rhizocarpon geographicum* was used on its natural rock habitat. Its resistance to simulated space parameters, applied separately or in specific combinations, was tested with regard to survivability, measured as preservation of its photosynthetic activity. These tests demonstrated the enormously high capability of the lichen system to cope with the parameters imposed on them.

2. Materials and methods

2.1. Biological samples

The epilithic lichen species *R. geographicum* (DC.) L. was used in the pre-flight verification tests. It is a crustose bipolar lichen species that grows in both poles and is widespread in mountains of both hemispheres (Feuerer, 1978). The color is yellow to green. Their growth rate is very low (about 0.5 mm/year, Sancho and Pintado, 2004) and the thalli may reach a final diameters up to 200 mm or more. Small rocks with homogeneous distribution of lichens were collected from the Plataforma de Gredos (Central Spain, 1.819 m, N40°17′, W5°14′). The lithology of this sector is predominately granite with phenocrysts. The lichen-colonized rock samples were stored at −20 °C in the dark for up to 6 months until use for the experiments. Cylindric samples of 10 mm in diameter and 6.9 mm in height were cut with a diamond point machine under continuous cooling with running water. Only those areas of the rock were selected that were completely covered by the lichen thallus. Before the start of the experiment, the samples were pre-activated at 10 °C and high humidity for 2–3 days at a 12 h light/dark cycle to simulate day/night cycles. The same pre-activation treatment was performed before each measurement of the photosynthetic activity of the lichens.

For the studies of photosynthetic activity of *R. geographicum* in its natural environment (Plataforma de Gredos), samples with and without cortex were prepared. In this paper, we report results of samples with cortex only.

2.2. Tests using planetary and space simulation facilities

The interplanetary space environment is characterized by a high vacuum, intense radiation of galactic and solar origin, low gravity, and extreme temperatures (Table 1). Testbeds for interplanetary space conditions should provide: (i) UV and vacuum-UV-radiation, (ii) low and high LET radiation, (iii) a high or ultrahigh vacuum of controlled residual gas composition, (iv) a reduced gravity if appropriate, and (v) defined temperature at the sample site. These parameters must be controlled, and according to the experiment requirements, applied separately or in selected combinations. For the pre-flight verification tests of the LICHENS experiment, the Planetary and Space Simulation Facilities PSI at the Institute for Aerospace Medicine at the DLR, Cologne (http://www.me.kp.dlr.de/) provided the required test parameters that were applied individually or in selected combinations.

An experimental aluminum hardware was used that was identical with the hardware of the space flight experiment (Fig. 1) (Sancho et al., in press). It consisted of two layers, each of them with 12 sample holes (an upper layer for samples exposed to UV and a bottom layer for samples kept in the dark). For each test run, one cylindrical rock sample with *R. geographicum* was accommodated in one of four