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Astrobiology: Laboratory Experiments, Field Studies in Analogue Environments and Space Experiments in Low Earth Orbit (F3.2)

MARSBOX: FUNGAL SPORES SURVIVE MARS-LIKE CONDITIONS ABOARD STRATOSPHERIC BALLOON FLIGHT

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The ability of terrestrial life to survive in the Martian environment is of particular interest for both planetary protection measures and for future colonization endeavors. Many studies have examined bacterial spore survival and decontamination, however, little is known about fungal spore resistance properties. To understand the survival potential of fungal spores to Mars conditions, *Aspergillus niger* spores were sent on a 6.5-hour balloon flight to Earth's stratosphere (38 km), where UV radiation and temperature conditions were similar to levels typical for equatorial Mars. Spores were carried inside the TREX unit – a sealed aluminum container filled with a Mars gas mixture – and flown aboard the MARSBOx balloon payload. Two different spore concentrations were tested on the TREX, exposed as dried samples in small quartz discs (20 µL). Discs were set in two different layers: a top layer exposed to direct UV radiation [M(+)UV, in a total dose of 1500 J/m^2], and a bottom layer that was shielded from radiation [M(-)UV]. After the flight, fungal spore survival was determined by plating on agar and determining colony forming units (CFU/ml). A germination rate was calculated based on light microscopy analysis and revival metabolism assay was completed with resazurin dye. Results show that *A. niger* spores can survive Mars-like conditions [M(+)UV] for the 6.5 h

time period tested in the middle stratosphere with only a 2-log reduction and slight delays in germination and revival metabolism compared to unflown lab controls. When shielded from UV, but exposed to Mars

gas, pressure and temperature [M(-)UV] spore survival and germination were not affected. This study provides valuable insights on whether fungal spores could survive on Mars, and underscores the need for longer-duration exposure studies in Earth's stratosphere to better characterize microbial

resistance to space-related conditions.