

orbital inclination of STS-9 (57°) compared with previous flights of the space shuttle (28.5°) is clearly seen. Even though STS-9 was at a somewhat lower altitude (241 km) than several previous flights (284 to 297 km), the low-LET dose rate is nearly double that previously recorded. The effect is even more dramatic when the dose-equivalents are compared: ~150 mrem for Spacelab 1 and ~50 mrem for the previous STS flights. This difference is the result of a substantial increase in the fluences of high-LET HZE particles.

The strong effect of altitude on dose rate can be observed (Table 2), with Skylab 4 (50°, 435 km) recording ~90 mrad/day. Since some of the future missions of Spacelab will have similar orbital trajectories, care will have to be taken to protect the parts and experimental equipment which may be sensitive to the radiation encountered. Equipment containing microprocessors, such as life-support systems and computers, is susceptible to single-event latchup and soft-error upset.

In the future, as spacecraft orbits are increased in altitude and inclination and the geomagnetic shielding is correspondingly reduced, the radiation hazard from large solar flare events will become significant (12). This is the case for orbits of inclination greater than ~50° and for polar and geosynchronous missions. Particularly during extravehicular activity, potentially lethal doses of protons can be encountered. Also, for these orbits, substantial fluxes of high-LET events from the HZE particles will be experienced. The radiobiological effects of these particles is not well understood, but there is evidence that they should be treated as single-event phenomena with high quality factors (13).

E. V. BENTON, J. ALMASI  
R. CASSOU, A. FRANK  
R. P. HENKE, V. ROWE

Physics Department,  
University of San Francisco,  
San Francisco, California 94117

T. A. PARNELL  
NASA-Marshall Space Flight Center,  
Huntsville, Alabama 35812

E. SCHOPPER  
Institut für Kernphysik,  
University of Frankfurt, 6000 Frankfurt,  
Federal Republic of Germany

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## Microorganisms in the Space Environment

**Abstract.** Preliminary results of the Spacelab 1 experiment on the response of *Bacillus subtilis* spores to conditions of free space are presented. Exposure to the vacuum of space on the Spacelab pallet reduced viability counts about 50 percent and increased mutation frequencies by a factor of about 10. Interpretation of apparent differences in the photobiological and photochemical data between flight and ground simulation experiments will require more statistical analyses and data from actual fluence measurements.

The objective of experiment IES029 on Spacelab 1 was to determine the response of a resistant microbial system to free space and to selected components of this hard environment. For this purpose 316 dry samples of *Bacillus subtilis* spores were exposed to the vacuum of space and/or to the full solar ultraviolet

spectrum (>170 nm) or selected ranges at peak wavelengths of 220, 240, 260, or 280 nm. Postflight analyses included studies of survival, mutation induction, reparability of ultraviolet damage, and photochemical changes in DNA and protein.

**Flight hardware.** An exposure tray partitioned in four square, quartz-covered compartments was mounted on a cold plate on the pallet of Spacelab 1. Two of the compartments were vented to the outside, allowing exposure of the samples to the vacuum of space. The other two compartments were hermetically sealed, with a constant pressure of 1 atm. Each compartment accommodated 79 dry samples of spores in thin layers, with 10<sup>5</sup> to 10<sup>7</sup> organisms per sample. Samples that were exposed to solar ultraviolet irradiation were placed beneath an optical filtering system composed of interference filters and neutral-density filters. A nontransparent shutter with optical windows was used to

Table 1. Survival (colony-formers) of *B. subtilis* spores exposed to space vacuum or laboratory-produced vacuum for 10 days. The following strains were used: HA 101 his<sup>-</sup>met<sup>-</sup>leu<sup>-</sup>; HA 101F polA<sup>-</sup>his<sup>-</sup>met<sup>-</sup>leu<sup>-</sup>; TKJ 6312 uvr<sup>-</sup>ssp<sup>-</sup>his<sup>-</sup>met<sup>-</sup>leu<sup>-</sup> (3, 4).

<i>B. subtilis</i> strain	Survival (%) after exposure to vacuum		
	Flight experiment	Simulation experiment	Simultaneous ground control
HA 101	62	100	100
HA 101F	50	98	100
TKJ 6312	46	94	97

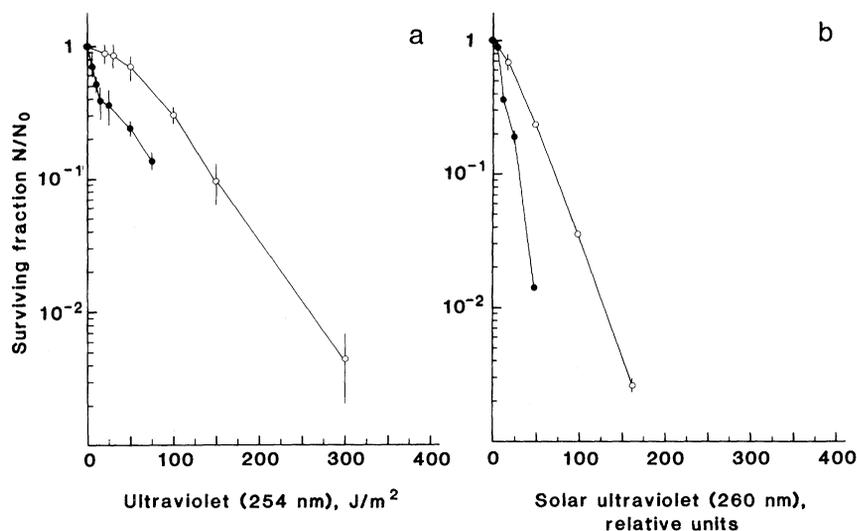


Fig. 1. Survival curves after ultraviolet irradiation of dry spores of *B. subtilis* HA 101 (○) at 1 atm and (●) in vacuo. (a) Simulation experiment and (b) flight experiment. Error bars represent standard deviations.

Table 2. Ultraviolet fluences for 10 percent survival of spores of *B. subtilis* HA 101 and its repair-deficient mutants. The ultraviolet fluence in the simulation experiment was measured by potassium ferrioxalate actinometry (5); that in the flight experiment was calculated from data in (1).

<i>B. subtilis</i> strain	F <sub>10</sub> values								F <sub>10</sub> simulation	
	Flight experiment				Simulation experiment				F <sub>10</sub> flight	
	λ (nm)	1 atm	Vacuum	F <sub>10</sub> (atm)/F <sub>10</sub> (vac)	λ (nm)	1 atm	Vacuum	F <sub>10</sub> (atm)/F <sub>10</sub> (vac)	1 atm	Vacuum
HA 101	260	71.0	29.0	2.5	254	151.3	36.7	4.1	2.1	1.3
HAF polA <sup>-</sup>		23.7	2.6	9.1		30.8	6.6	4.7	1.3	2.5
TKJ 6312 uvr <sup>-</sup> ssp <sup>-</sup>		7.7	3.1	2.5		6.6	5.3	1.2	0.9	1.7
HA 101	>170	74.7	52.5	1.4	>170	431.9	64.9	6.7	3.1	1.2

achieve precise irradiation intervals during the hot phase of the seventh day of the mission. Each compartment was equipped with a recording thermometer.

**Flight experiment protocol.** The flight experiment was loaded with the microbial samples in the laboratories of the Deutsche Forschungs- und Versuchsanstalt für Luft- und Raumfahrt (DFVLR) in the last week of August 1983. After transport to Kennedy Space Center, the exposure tray was installed on the cold plate of the Spacelab 1 pallet on 12 September 1983. Environmental data on the space transportation system (STS) payload bay in the Orbiter Processing Facility were given by NASA as 21° ± 3°C air temperature, 30 to 50 ± 5 percent relative humidity.

The experiment was activated in orbit 12 hours after launch. Temperature data were transferred at a rate of 1 sec<sup>-1</sup>. The temperature remained in the noncritical range during the whole mission. On mission day 7, the shutter was moved to positions allowing access of solar ultraviolet radiation to the four compartments for 19 minutes, 23 minutes, 43 minutes, or 5 hours and 17.5 minutes, respectively. The experiment was deactivated on hour 10 of mission day 9. It was dismantled 12 days after the return of Spacelab 1 to the Orbiter Processing Facility at Kennedy.

**Ground controls.** Two kinds of ground controls were used: a simulation experiment before the flight, and an identical experimental setup in parallel with the flight but with a 1-day phase shift. The latter was kept at a simulated mission temperature profile and exposed to vacuum of 10<sup>-4</sup> Pa for 10 days.

**Response to the vacuum of space.** Exposure to the vacuum of space for 10 days reduced viability counts (colony-formers) to about 50 percent of those in samples kept at 1 atm. Similar vacuum treatment in the laboratory either did not affect or only slightly inactivated the spores (Table 1). Furthermore, a tenfold higher frequency of histidine prototropic mutations was observed in spores exposed to the vacuum (1.2 × 10<sup>-4</sup>) than

in samples kept at 1 atm (1.0 × 10<sup>-5</sup>).

**Response to solar ultraviolet irradiation.** Survival curves after solar ultraviolet irradiation were compared with those obtained in the laboratory simulation experiment (Fig. 1). Ultraviolet fluences that reduced survival to 10 percent (F<sub>10</sub> values) were taken as a measure of ultra-

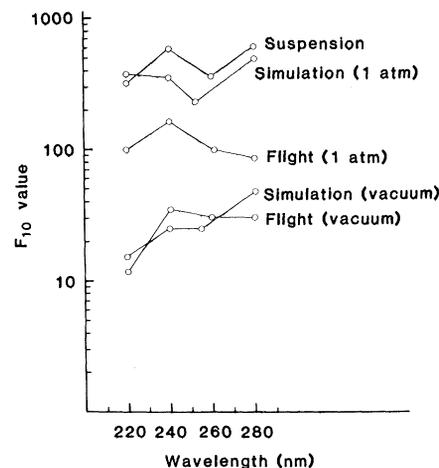
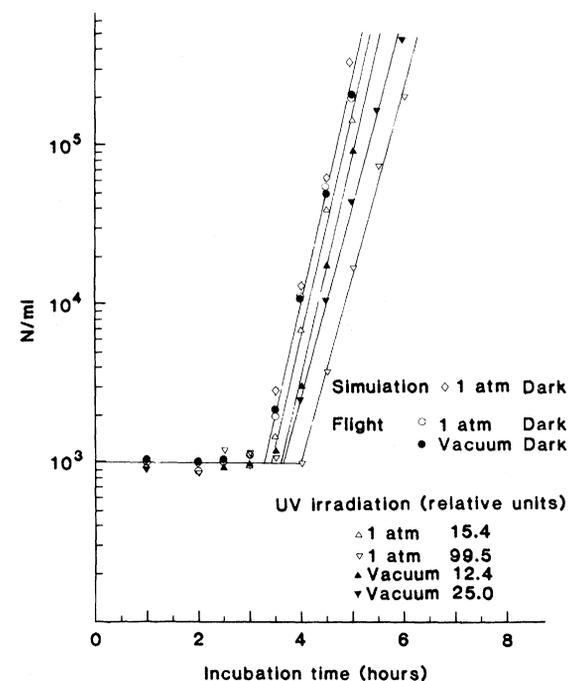


Fig. 2. Action spectrum of inactivation of *B. subtilis* 168 spores.

violet sensitivity (Table 2). The major results so far are as follows: (i) All strains tested showed a higher ultraviolet sensitivity in vacuo than at 1 atm (by a factor of 1.2 to 9.1). (ii) Under all experimental conditions, the repair-deficient strains (HA 101F polA<sup>-</sup>; TKJ 6312 uvr<sup>-</sup> ssp<sup>-</sup>) showed a higher ultraviolet sensitivity than the wild-type strain (HA 101). (iii) In vacuo, the ultraviolet sensitivities of the two repair-deficient strains were about the same. (iv) The samples in the flight experiment showed higher ultraviolet sensitivities than those in the simulation experiment in all cases but one. (v) Compared to the simulation experiment, the wild-type strain HA 101 exposed to the full spectrum of solar ultraviolet radiation (>170 nm) showed the largest increase in sensitization, by a factor of 3.1 after irradiation at 1 atm. In the flight experiment, fluence data were calculated from (1). More precise data on the actual solar ultraviolet fluence are expected from experiment 1ES016 (2). The F<sub>10</sub> values corrected with these actual data may differ from those for the flight experiment given in Table 2.

Fig. 3. Growth of *B. subtilis* HA 101 after incubation of spores in Bacto Penassay Broth (Difco).



From survival curves for spores of *B. subtilis* 168 after irradiation with solar ultraviolet at peak wavelengths of 220, 240, 260, and 280 nm at 1 atm or in the space vacuum, action spectra for ultraviolet inactivation were obtained and compared with those of the simulation experiment and of spores in aqueous suspension (Fig. 2). In this case, too, the  $F_{10}$  values for the flight experiment are based on calculations as above and may have to be corrected with actual data. Concerning ultraviolet effectiveness in vacuo, there is reasonable agreement between the flight and simulation experiments. In the flight experiment, for all wavelengths tested, the ultraviolet effectiveness at 1 atm was much higher than in the simulation experiment.

During the incubation of samples of strain HA 101 in the flight and simulation experiments, after exposure to the vacuum and to 1 atm, respectively, identical growth curves were obtained (Fig. 3). Solar ultraviolet (260 nm) irradiation induced a division delay which, for similar ultraviolet fluences, was slightly higher for samples in vacuo than for samples kept at atmospheric pressure. Ultraviolet fluences for which there were similar surviving fractions in vacuo and at 1 atm caused a much higher division delay at 1 atm than in vacuo.

DNA protein cross-linking was induced by solar ultraviolet irradiation of spores in vacuo as well as at 1 atm. The amount of free DNA, which was a measure of DNA that was not cross-linked to protein, decreased with increasing ultraviolet fluence. More information is needed to obtain quantitative data of statistical significance.

Solar ultraviolet (>170 nm) irradiation induced the so-called spore type photoproduct 5-thymine-5,6-dihydrothymine (TDHT) in the DNA of spores of *B. subtilis* 168, irradiated at 1 atm as well as in the space vacuum. In vacuo, two additional photoproducts were induced and the amount of TDHT decreased.

G. HORNECK  
H. BÜCKER

G. REITZ  
H. REQUARDT

Division of Biophysics, Institut für Flugmedizin, DFVLR, 5000 Cologne 90, Federal Republic of Germany

K. DOSE  
K. D. MARTENS

Institute of Biochemistry, University of Mainz, 6500 Mainz, Federal Republic of Germany

H. D. MENNIGMANN, P. WEBER  
Institute of Microbiology, University of Frankfurt, 6000 Frankfurt am Main, Federal Republic of Germany

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## Cell Sensitivity to Gravity

**Abstract.** Cultures of human lymphocytes exposed in microgravity to the mitogen concanavalin A showed less than 3 percent of the activation of ground controls. This result supports the hypothesis, based on simulations at low g and experiments at high g, that microgravity depresses whereas high gravity enhances cell proliferation rates. The effects of gravity are particularly strong in cells undergoing differentiation.

An answer to the question of whether cells are sensitive to gravity was one of the objectives of experiment 1ES031, on the effect of weightlessness on lymphocyte proliferation, performed on board Spacelab 1. Another objective was to establish, by exposing cultures of human lymphocytes to a mitogen during spaceflight, whether functional changes occurred in the cells responsible for the immune response. Several investigators (1) reported that lymphocytes from the majority of crew members on U.S. and Soviet space missions had a decreased response to mitogen after flight. Stress may be one of the reasons for the reduction of lymphocyte response (2). A manifold increase of induced interferon pro-

duction was recently observed in cultures of human lymphocytes flown on the Soviet space station Salyut 6 (3). One of the few experiments dedicated so far (4) to the study of the effects of microgravity on cells was performed on Skylab with a strain of human embryonic lung cells, WI-38 (5). Although there were no significant changes in cell proliferation and motility, glucose consumption was significantly lower in the cells flown in space.

On Spacelab 1 we exposed cultures of human lymphocytes to concanavalin A (Con A), a mitogen capable of transforming resting T lymphocytes into activated (dividing) cells. This is a suitable model for the study of certain aspects of the immune system and of cell differentiation in vitro. In addition, in connection with the experiment on Spacelab, we studied the adaptation of animal cells to an altered gravitational environment and found that proliferation of five types of cells is remarkably enhanced at 10 g (6), whereas glucose consumption is the same at both high g and 1 g. However, cell motility is hindered at 10 g. In particular, lymphocyte activation is strongly gravity-dependent; the proliferation rate is almost doubled at 10 g. We suggest that, under gravitational stress, the cell is capable of switching to other metabolic pathways (6). Conversely, when lymphocytes are cultivated at simulated microgravity in a rapidly rotating clinostat, in which gravity is changed from a vector to a scalar in the presence of Con A, activation is depressed by 50 percent (7). In general, hypergravity promotes cell proliferation, whereas microgravity has a depressing effect. Our experiment on Spacelab 1 was designed to test this hypothesis. All preflight operations and the synchronous ground control experiment were performed at Kennedy Space

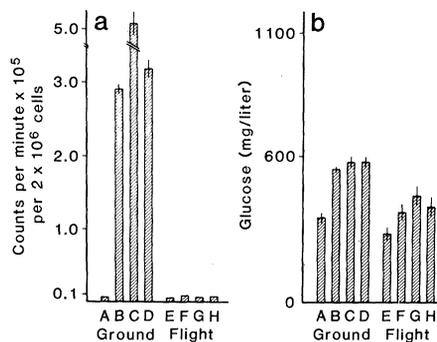


Fig. 1. Lymphocyte activation induced by Con A in microgravity. Cultures of human lymphocytes were exposed to mitogenic concentrations of Con A in ground samples B, C, and D and flight samples F, G, and H, respectively. Samples A (ground) and E (flight) were unstimulated controls. (a) Activation measured after 69 hours incubation at 37°C as [<sup>3</sup>H]thymidine incorporation into trichloroacetic acid-precipitable material. (b) Glucose remaining in the medium measured by the glucose dehydrogenase method (6). The initial concentration of glucose in the medium was 1100 mg/liter. The standard deviation of triplicate samples is given, except for samples A, E, F, G, and H in (a), for which it was too low to be shown here.