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An ultraviolet simulator for the incident Martian surface radiation and its applications

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Abstract: Ultraviolet (UV) radiation can act on putative organic/biological matter at the Martian surface in several ways. Only absorbed, but not transmitted or reflected, radiation energy can be photo-chemically effective. The most important biological UV effects are due to photochemical reactions in nucleic acids, DNA or RNA, which constitute the genetic material of all cellular organisms and viruses. Protein or lipid effects generally play a minor role, but they are also relevant in some cases. UV radiation can induce wavelengths-specific types of DNA damage. At the same time it can also induce the photo-reversion reaction of a UV induced DNA photoproduct of nucleic acid bases, the pyrimidine dimers. Intense UVB and UVC radiation, experienced on early Earth and present-day Mars, has been revealed to be harmful to all organisms, including extremophile bacteria and spores. Moreover, the formation of oxidants, catalytically produced in the Martian environment through UV irradiation, may be responsible for the destruction of organic matter on Mars. Following this, more laboratory simulations are vital in order to investigate and understand UV effects on organic matter in the case of Mars. We have designed a radiation apparatus that simulates the anticipated Martian UV surface spectrum between 200 and 400 nm (UVC–UVA). The system comprises a UV enhanced xenon arc lamp, special filter-sets and mirrors to simulate the effects of the Martian atmospheric column and dust loading. We describe the technical setup and performance of the system and discuss its uses for different applications. The design is focused on portability, therefore, the Mars-UV simulator represents a device for several different Mars simulation facilities with specific emphasis on Mars research topics.

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Key words: astrobiology, biological matter, Mars, organic matter, photobiology, UV climate, UV simulator.

Introduction

In order to investigate the effects on organic or biological matter exposed to ultraviolet (UV) radiation at the Martian surface, a rigorous simulation of the Martian environment is necessary. In this context, the ambient Martian UV radiation plays an important role along with the Martian soil substrate and the Martian atmosphere, with ambient pressure and temperature altered to fit the conditions found on Mars.

UV radiation and the formation of oxidants at the Martian surface

Several analyses were performed by the Viking landers in the course of the search for life on Mars. No biological signals were found, but instead, the surface and subsurface soil samples gave evidence for chemical reactivity (e.g., Klein *et al.* 1976; Zent & McKay 1994). A release of oxygen was observed upon the humidification of soil samples (Oyama & Berdahl 1977). Decomposition of induced isotopically

labelled organic nutrient solutions due to contact with the soil samples gave further (debatable) evidence for oxidative reactions (Levin & Straat 1977; Levin *et al*. 2001). Further, adsorbed superoxide ions, such as O_2^-, are thought to be responsible for the chemical reactivity of the Martian soil (Yen *et al*. 2000). It was shown by means of experimental studies under Martian atmospheric and pressure conditions that UV irradiation, low atmospheric oxygen, very low water-concentrations and mineral grain-surfaces are the main elements in the formation process of these adsorbed superoxide ions (Yen *et al*. 2000). Yen *et al*. (2000) showed that these ions are formed progressively under intense UV irradiation (low-pressure Hg vapour lamp with a peak flux at 254 nm). Thus, it is likely that UV photons are not a limiting factor in this oxidant production process – as is the case for H_2O_2. UV is involved in the formation of H_2O_2, but also acts to destroy H_2O_2 under the UV photon flux throughout the day (Zent & McKay 1994). Möhlmann (2004) showed experimentally that radiation-induced hydroxyl formation in the presence of dissolved iron might take place in the presence of adsorbed water at the Martian surface ('Photo-Fenton reaction'; Spacek *et al*. 1995). The importance of UV radiation in Martian photochemical processes was not only shown by the experiments of Yen *et al*. (2000), but is also implied from the investigations of Martian surface samples by the Viking Landers. One should note that the UV radiation dose tends to be more intense at the Viking 1 landing site in Chryse Planitia than at the Viking 2 landing site in Utopia Planitia (Patel *et al*. 2003), consistent with the observed greater release of oxygen from the Chryse samples (Oyama & Berdahl 1977).

UV radiation and the interaction with organic and biological matter

Since short-wavelength UV radiation in the UVB and UVC range can penetrate to the surface of an oxygen-free planetary atmosphere, damaging consequences on molecules essential for life should be expected. Because solar UV radiation was a driving force for the organic chemical evolution on Earth, UV may have played a similar role on other planets such as Mars (Walker *et al*. 1983; Lowe 1994; Holland 1999; Cockell & Horneck 2001). Polycrystalline uracil, a pyrimidin base as a component of RNA can serve for models of nucleic acid damage due to UV radiation. The double stranded DNA in the bacteriophage T7 is also suitable to measure the degradation of nucleic acids due to UV radiation as demonstrated in Bérces *et al*. (1999). In both systems the radiation can cause two main processes: covalent-bond formation between two pyrimidine molecules (either uracil or thymine and cytosine) to produce a so-called dimer (dimerization), or breaking apart a dimer into monomers (monomerization). Under the influence of short-wavelength solar UV radiation (UVC) photo-products are induced, whereby reversion can be stimulated by photons with similar energies (Rontó *et al*. 1967; Rontó *et al*. 2004; Fekete *et al*. 2004). Irradiation experiments on uracil thin layers gave evidence of a higher monomerization efficiency of a deuterium lamp (short UVC) and that of a higher dimerization efficiency of a low-pressure Hg lamp (254 nm) as presented in Bérces *et al*. (2002). Thus at a given polychromatic spectral composition of the UV source, the dimerization and monomerization processes could find an equilibrium, where the level depends on the ratio of the longer to the shorter wavelengths. Until now the existence of dimer–monomer photoreactions has been obtained mostly in indirect experiments with more or less monochromatic UV sources (e.g., Setlow & Setlow 1965; Rontó *et al*. 2004). In terms of DNA stability there is a certain chance for a piece of DNA such as bacteriophage T7-DNA to remain intact under short-wavelength UV radiation, as the equilibrium in the radiation field provides stability for the DNA. The effect of photoreversion can be studied in detail with the new Mars UV simulator because of the short-wavelength UV components of the simulated Martian solar UV radiation. The possibility of the photoreversion reaction of pyrimidine dimers might have been an important protective characteristic of the genetic material during evolution of life on Earth, but perhaps also on other planetary surfaces, e.g. on Mars.

In biological systems UV radiation causes temporary or permanent alterations, which result from photochemical reactions of UV with different biological target molecules, the so-called chromophores. The most important UV target in cells is the DNA because of its unique role as genetic material and its high UV sensitivity. In the case of wavelengths longer than 200 nm, absorption of DNA is restricted to the bases, the purine derivatives adenine and guanine, the pyrimidine derivatives thymine and cytosine. Although the base composition of DNA is not the same in different genes and organisms, there are the common features of an absorption maximum in the 260 nm region and a rapid decline toward longer wavelengths. Absorption by proteins between 240 and 300 nm is much lower than that of nucleic acids of equal concentration. Most proteins are present in cells in higher numbers of identical copies. Therefore, photochemical alterations in only a fraction of them do not disturb their biological function significantly. The same is true for molecules such as unsaturated fatty acids, flavins, steroids, chinones, porphyrins or carotenoids, which serve as components of the cell membrane, as coenzymes, hormones, or electron donor transport molecules. The spectrum of UV that first hits the surface of a biological object changes while it passes the outer parts of the cell or tissue to reach the sensitive targets in the cells. Therefore the action spectrum, which describes the wavelength dependent UV sensitivity of a biological effect, is often not identical to the absorption spectrum of a chromophore.

In Fig. 1 examples of biological action spectra obtained with monochromatic radiation are given for spores of the bacterium *Bacillus subtilis*. In this graph the UV sensitivity is shown for two strains differing in their capability to repair UV induced DNA damage. The strain TKJ6312 with the higher sensitivity possesses a mutation in an important enzymatic DNA repair pathway, whereas the strain HA101 is a DNA repair wild-type strain (Rettberg & Horneck 2000;

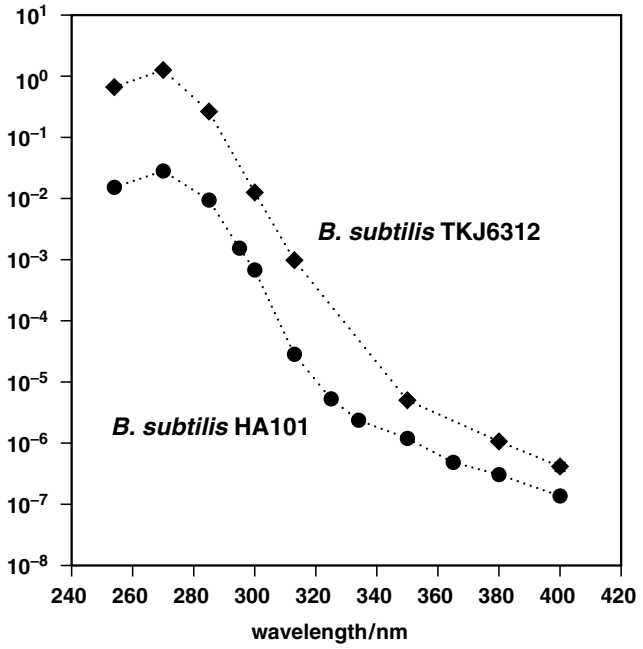

Fig. 1. Wavelength-dependent UV sensitivity of dry *B. subtilis* spores of two strains differing in their DNA repair capability.

Rettberg & Rothschild 2001). In general, the sensitivity of biological objects increases with decreasing wavelengths in the UV range. In the case of *B. subtilis* spores the maximal sensitivity was found at a wavelength of approximately 270 nm, which is not exactly identical with the DNA absorption maximum.

Further, space experiments as well as Mars simulation studies with terrestrial bacterial soil communities and pure cultures have revealed that UV radiation is a primary detrimental factor on bacterial survival under Martian conditions (Rettberg *et al.* 2004). The solar radiation reaching the surface of Mars is found to be extremely harmful to bacteria and only 15 min of Mars equivalent UV exposure is shown to degrade the endospores of the bacterium *Bacillus subtilis* (Schuerger *et al.* 2003). This direct UV effect on bacterial survival is restricted to bacteria at the very surface directly exposed to the UV radiation. Bacterial cells covered with soil particles, cell layers or dust are efficiently protected against the harmful UV radiation (Green *et al.* 1971; Mancinelli & Klovstad 2000; Rettberg *et al.* 2002; Schuerger *et al.* 2003). Nonetheless, the protected subsurface bacteria are metabolically affected by indirect UV effects probably from photochemically generated free oxygen species (Hansen *et al.* 2005). Pigmented bacteria and Gram-positive bacteria are found to have a significantly higher UV resistance compared to other bacteria (Arrage *et al.* 1993; Jacobs & Sundin 2001). However, the absorption of UV radiation by bacterial pigments can be restricted to specific wavelengths and generally UVC radiation is bacteriologically more harmful than UVB radiation, which is, in turn, more harmful than UVA radiation (Moeller *et al.* 2005). The UV screening compounds scytonemin and mycosporine-like amino acids are known to be synthesized by some phototrophic cyanobacteria. This passive UV screening strategy allows the cyanobacteria to inhabit UV exposed environments without high energy costs to UV damage repair (reviewed in Cockell & Knowland 1999).

Halobacteria (also called haloarchaea) are markedly more resistant to UV radiation than many common bacteria such as *Escherichia coli*. Differences expressed as D_{37} (radiation dose that leaves 37 % of surviving cells) following irradiation with UVC were 10 J m^{-2} for *E. coli* and 212 J m^{-2} for *Halobacterium salinarum* (Shahmohammadi *et al.* 1997). UV induced photoproducts are, as in other organisms, mainly cyclobutane pyrimidine dimers, where the C4 and C5 carbon atoms of any two adjacent pyrimidines are connected such that a four-membered ring is produced, and 6–4 photoproducts, where the bond is formed between the C4 carbon of the first and the C6 carbon of the second of two adjacent pyrimidines (see McCready & Marcello 2003). The DNA repair systems of haloarchaea, which have been identified so far, include several photolyases and are very efficient in the light (McCready & Marcello 2003); there is some evidence that additional repair systems are present that work in the dark (Baliga *et al.* 2004). Most haloarchaea, which live in salty brines and lagoons, experience intense exposure to sunlight; their striking pigmentation, due to carotenoids and bacterioruberins, is thought to afford protection against UV radiation (Shahmohammadi *et al.* 1997; McCready & Marcello 2003), although some data are not compatible with this latter notion (Baliga *et al.* 2004). Several viable halobacteria have been isolated from salt deposits of the Permo-Triassic age and were identified as novel species (Stan-Lotter *et al.* 2004), which suggested a potentially extreme longevity of these micro-organisms. Together with the discovery of extraterrestrial halite in meteorites (Zolensky *et al.* 1999), in the ocean of the Jovian moon Europa (McCord *et al.* 1998) and recently, on the surface of Mars by the NASA rovers Spirit (http://www.msnbc.msn.com/id/5166705/) and Opportunity (Rieder *et al.* 2004; Christensen *et al.* 2004; http://www.missionspace.info/news/merupdate/saltwater.html), the possibility of surviving halophilic life in the Martian subsurface has been considered by various authors. Therefore, more experiments that study the responses of haloarchaea (Stan-Lotter *et al.* 2003) towards Martian atmospheric and UV conditions need to be carried out during the near future.

Basic requirements for Mars UV simulators

The demands on a UV simulator are constrained by its applications. In the design of this simulator prime focus was given to the spectral recreation of the anticipated UV spectrum at the Martian surface (Patel *et al.* 2002; Rontó *et al.* 2003). Portability represents a further requirement, since several cooperating Mars simulation facilities exist across Europe. Due to portability, constraints are imposed on the dimensions and mass of the system, which resulted in a compromise in illumination power. This is why the system aperture is restricted to a nominal diameter of about 5 cm.

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A further important demand on the system is the requirement for homogeneity of the intensity and spectrum across the beam spot, and its long-term stability. Homogeneity is constrained by optical parameters such as chromatic aberration, whereas stability is controlled by the power supply and operating temperatures. The infrared (IR) component of the beam causes the samples to heat, thus the IR component has to be blocked by means of filter devices beginning with the NIR range. Due to the collimated beam the distance of the sample position to the light source is an independent parameter. This provides for the flexible positioning of samples within a certain range and minimizes bias on the reproducibility of experiments and beam patterns. A considerable ozone accumulation in an irradiated gas volume may induce unfavourable absorption centred about 250 nm (Rontó *et al*. 2003), therefore the path of the beam has to be regularly vented during operation.

Table 1. *Technical data of Mars UV simulator*

Lamp	Xenon arc lamp, UV grade fused silica
Target spot	5 cm, round
Working distance	25–35 cm from aperture
Spectral range	≈180–900 nm
Maximum illumination power within 200–400 nm	55.6 W m^{-2}
Homogeneity within target spot	≈5% at 25–35 cm from aperture
Output intensity stability (30 min after turned on)	≈1%
Lenses	UV grade fused silica
Filters	Water filter, UV grade fused silica
Mirrors	Aluminium, UV enhanced coating
Weight	10 kg
Size	40 cm (W) × 33 cm (H) × 14 cm (D)

Radiative transfer modelling for UV fluxes at the Martian surface

The Martian atmosphere is predominantly CO_2 (95%) with a mean surface pressure of about 7 mbar. As such, below 190 nm no solar UV radiation reaches the surface (e.g., Kuhn & Atreya 1979), because the absorption cross-section of CO_2 rapidly increases at these wavelengths, effectively forming a cut-off point in all present-day Martian spectra. The only significant absorbing and/or reflecting constituents in the relevant UV range are CO_2, O_3, O_2, CO and NO_x molecules, along with significant amounts of suspended dust in the Martian atmosphere. The design of the UV simulator is based on model simulations of the Martian solar UV environment by Patel *et al*. (2002, 2004) and Rontó *et al*. (2003). For these simulations a radiative transfer model has been developed in a number of ways allowing for a more realistic representation of the actual Martian surface radiation environment. The Martian atmosphere was taken as 10 separate homogenous layers, each forming a mixture of gaseous particles as a function of altitude. The scattering and absorption effects of CO_2, N_2, Ar, O_2, O_3 and CO were accounted for.

In this model it is assumed that the atmospheric gas is distributed between the Martian surface and upper atmosphere, within layers placed at 0, 1, 5, 10, 15, 20, 21, 35, 50, 51 and 200 km altitudes. The column density of each atmospheric gas for each layer was determined through the knowledge of partial pressures at the layer interface and mean molecular mass. The column abundance of each individual species was then calculated from the total layer column abundance and mixing ratio of each species. Pressure values as a function of altitude used to determine the column abundance of each layer was taken from the Pathfinder entry data (Magalhaes *et al*. 1999). Aerosols were assumed to consist solely of dust particles and were confined to the lower 20 km of the atmosphere with a scale height equal to that of the atmospheric gas.

The Delta-Eddington approximation (Joseph *et al*. 1976) for radiative transfer was employed to determine the diffuse flux at the surface created by aerosol and molecular scattering, whereby the phase function typical for particulate scattering is approximated by consideration of a forward scatter peak and a two-term expansion of the phase function. The fraction of forward scattering is proportional to the square of the phase function asymmetry factor (Joseph *et al*. 1976). Direct solar flux was determined via Beer's law, exponential attenuation, again taking into account scattering and absorption from each species in each atmospheric layer (Patel *et al*. 2002, 2004). The direct flux was then added to the diffuse component to yield a total surface irradiance. This process was performed at 1 nm resolution between 180 and 400 nm.

System design and output measurements

The simulator consists of an electrical unit (power supply and ignition unit) and an optical system (inner lamp housing, optics). The electrical package is commercially available, whereby the optical part was custom made in order to meet the requirements. The technical specifications of the system are compiled in Table 1.

Technical realization of the Mars-UV simulator (optical component)

Generally, every design to apply radiation to a defined sample area needs the combination of a ray source and focusing techniques. Further attenuation optics are necessary to adjust the spectral characteristics of the ray source to the desired target spectrum of incident light. Focusing techniques comprise the range of mirrors and lens systems as well as aperture devices to collect and focus the beam onto the sample. Attenuation optics are used to remove IR radiation from the target spectrum and to adjust the spectral shape, especially in the UV range. The whole appliance is continually vented with forced air to prevent ozone accumulation in the path of the beam. All optical devices are made of materials meeting high UV transmission/reflection requirements.

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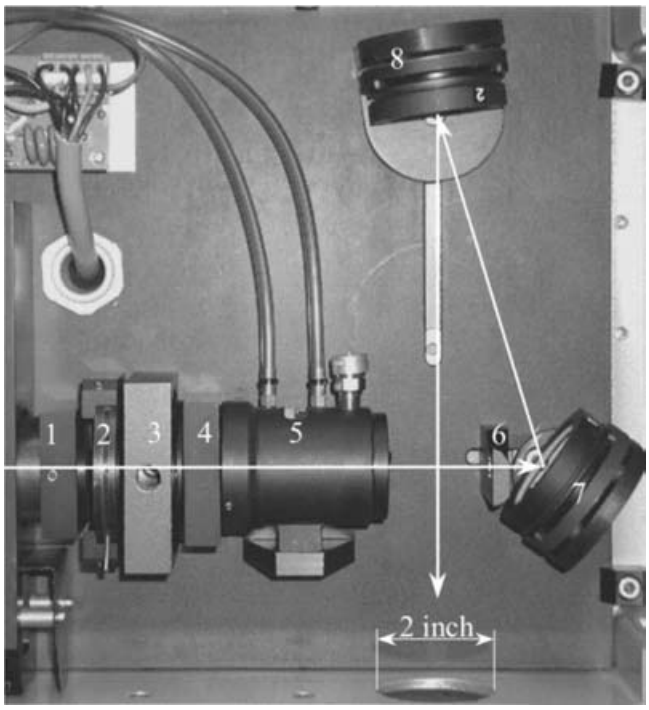

Fig. 2. The optical component of the simulator, including: (1) condensing lens assembly, (2) aperture iris, (3) filter set to carry optional band pass, interference and neutral density filters, (4) plano-convex lens, (5) liquid filter, (6) internal aperture, (7) deflection mirror and (8) spherical mirror.

Ray source

With a black-body temperature of 5600 K, the emission spectrum of the xenon arc is very close to that of the Sun. The spectral UVC output of the lamp is highly dependent upon the material of the glass bulbs and on other product-specific design parameters. In order to meet the basic requirements for the system, a 150 W xenon arc lamp made of UV grade fused silica bulbs was employed. The bulb was installed in an inner lamp housing, which was vented with air to keep the operating temperature of the lamp socket and bulb within tolerable levels and to remove ozone around the bulb. An adjustable rear reflector collects the rear illumination of the lamp and focuses it near the light arc, for subsequent collection and collimation by the condensing lens assembly.

Focusing techniques

A condensing lens system (F/1) was mounted on a guidance slot in order to provide adjustments of the intensity output. After filtering the light with specialized filters, the beam is focused onto the deflection mirror by means of a plano-convex lens (Fig. 2). A spherical mirror collects and focuses the light onto the target spot perpendicular to the horizontal mounting plate. The mirror system is designed to assure a collimated beam with negligible divergence ($\approx 1^{\circ}$) and the system was calibrated to provide spectral homogeniety at a working distance of 25–35 cm. The resulting beam is 5 cm in diameter.

Attenuation optics

UV grade fused silica materials were used throughout, which provides a wavelength cut-off at about 180–190 nm. Further attenuation components are used to remove unavoidable IR radiation and consequently prevent unfavourable heating of the samples. A liquid filter (5 cm path length) filled with distilled water was used, due to its high effectiveness in filtering IR (Fig. 2, spectral range from ≈180–900 nm).

Experimental determination of the spectral irradiance

A portable spectroradiometer (Optronic Laboratories, Inc., OL 754-O and Bentham, Inc., DMc150 double monochromator) was used to measure the spectral irradiance of the UV simulator in the wavelength range of 200–400 nm. To collect the light beam of the UV simulator a PTFE integrating sphere was employed.

Results of spectroradiometric and sensoric measurements of the Mars-UV simulator output

The spectral intensity output of the simulator in the range between 200 and 400 nm as measured by the spectroradiometer is given in Fig. 3. The experimental results are very close to that of theoretical surface flux models (e.g. Patel *et al.* 2002; Rontó *et al.* 2003, Fig. 1, curve a).

When used with Mars simulation chambers, the light beam of the UV simulator is directed onto the sample by passing through UV grade fused silica windows. To assess the spectral content, a quartz fibre optic probe with a PTFE diffusing unit on top of the sensor head was placed into a simulation recipient (DLR, Cologne). The loss of intensity, at a given working distance, due to the use of UV grade fused silica windows (10 mm thickness) was found to be around 14% in the range of 200–400 nm with the test chamber evacuated. Filling the chamber with about 7 mbar of Martian atmosphere resulted in an additional loss of about 2%. The attenuation effect on the light beam is given in Fig. 4 and shows maintenance of the spectral shape.

The spatial homogeneity of the output of the simulator was examined with a photodiode sensor having band-pass filters at 254.3 nm (full width at half maximum (FWHM) = 10.2 nm) and 221.3 nm (FWHM = 9.2 nm). Ten measurements were taken across the diameter of the beam (Table 2).

Applications

To date, UV radiation has only been included in a few Martian simulation studies. Additionally, the UV lamps applied have been mercury, mercury–xenon, deuterium and xenon-arc lamps all with different spectra making it difficult to compare the different results (reviewed in Schuerger *et al.* 2003). Only two studies have simulated the exact Martian UV radiation with respect to both wavelengths and fluence rates (Green *et al.* 1971; Schuerger *et al.* 2003). Therefore, more UV related experiments under Martian conditions with various microbial strains, spores and uracil thin layers,

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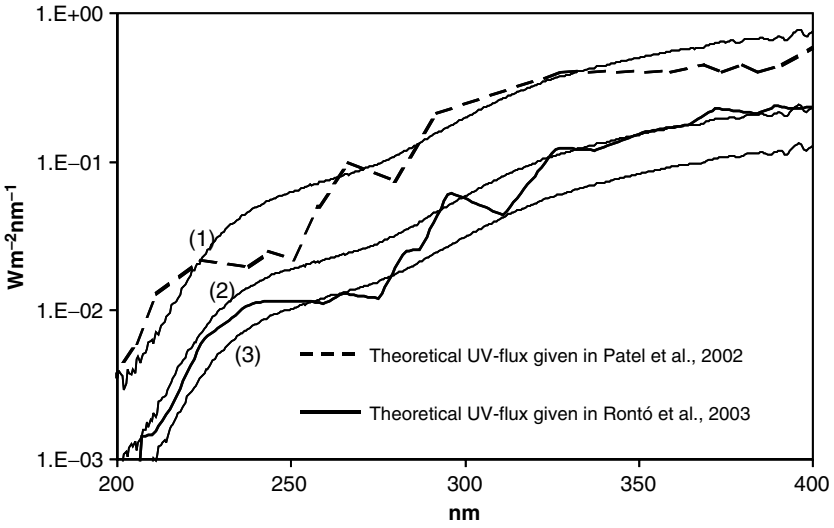

Fig. 3. Spectral intensity output of the Mars UV simulator in comparison with theoretical, total UV fluxes at the recent Martian surface, based on atmospheric column and low dust loading (from Patel *et al.* 2002 and Rontó *et al.* 2003, Fig. 1, curve a). Curve (1) maximum spectral intensity output (optical density = 0.0); curve (2) attenuated output (optical density = 0.5) and curve (3) attenuated output (optical density = 0.8).

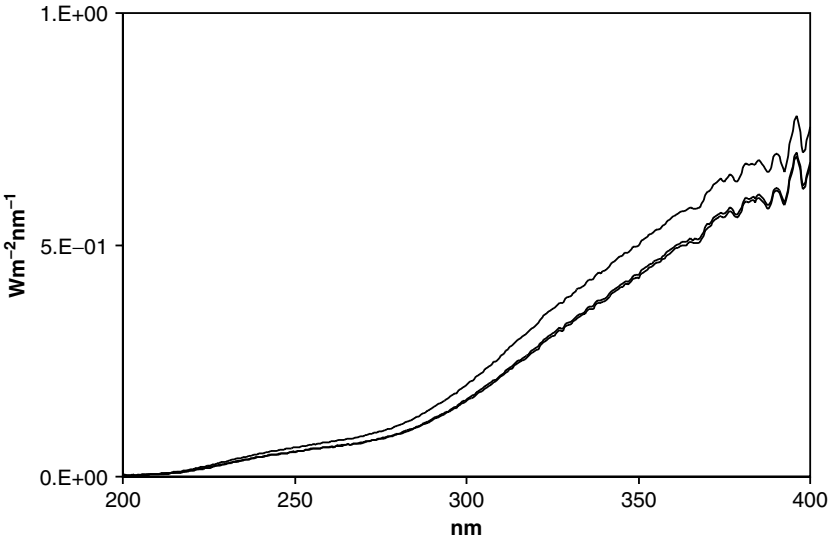

Fig. 4. Attenuation effects due to windows of simulation chambers and application of Martian atmospheres. Upper curve, spectral intensity output without attenuation; middle curve, intensity output after passing through UV grade fused silica windows with the chamber evacuated; lower curve, intensity output after filling with 7 mbar of Martian atmosphere.

which will use the UV simulator discussed in this paper are planned for the near future.

UV exposure of pyrimidine molecules

Polycrystalline uracil thin layers have been used in the phage and uracil response (PUR) experiment of the response of organisms to the space environment (ROSE) consortium (Horneck *et al.* 1999), assigned to study the biological dosimetry of extraterrestrial solar radiation on the exterior of the International Space Station (ISS). In recent ground-based UV exposure experiments with deuterium and germicidal lamps (UVC, down to 200 nm) in vacuum chambers over a temperature range of $\pm 40\,^{\circ}$C, the kinetics of UV driven formation (dimerization) and reversion (monomerization) of uracil dimers have been measured (Rontó *et al.* 2004). It was found that the monomerization efficiency of the output of the polychromatic deuterium lamp is higher than that of the Germicidal lamp. To study the impact of photo-reversion on the Martian surface, future experiments are planned that will use special interference filters, which block the UV outside wavelength regions that are not relevant for dimerization or monomerization reactions.

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Table 2. *Representative homogeneity test of target spot. Photodiodes with band-pass filters (221.3 and 254.3 nm) were placed at 10 equidistant points along the target spot. Data are given in nA and indicate an intensity variation of less than ±5%*

Filter	1	2	3	4	5	6	7	8	9	10
221.3 nm	1.22	1.27	1.31	1.33	1.35	1.34	1.32	1.3	1.26	1.23
254.3 nm	5.96	6.01	6.05	6.08	6.1	6.09	6.07	6.05	6.02	5.97

UV exposure of amino acids

Meteoritic input to Mars is expected, with caveats about the variable rate of surface gardening, to lead to an areal flux at the surface of the order of 2×10^{-3} to 8×10^{-5} g m^{-2} yr^{-1}. A fraction of this material will take the form of organic matter and the potential for this material to act as a feedstock for ancient or present prebiotic processes depends on its longevity in the Martian environment. The photolytic and photochemical stability of biologically relevant molecules such as amino acids has also been studied with the use of solar simulators in past studies (Oró & Holzer 1979; Stoker & Bullock 1997; tenKate *et al.* 2005). Such studies often require the use of relatively immobile precision analytical hardware such as mass-spectrometers or infrared spectrometers. The existence of a portable solar simulator is a great advantage to those researchers in the astrobiological community, allowing the light source described here to be brought to the samples and their associated measurement systems. So as to perform cross-calibration of existing data sets it is planned that the UV simulator described here will be used to illuminate organic samples as studied by tenKate *et al.* (2005) in experiments at various temperatures and under varied atmospheric conditions.

UV simulation for catalytic experiments

Photochemical processes are assumed to be active in oxidant formation at the recent Martian surface, however little is known about their nature and interplay. Since UV radiation represents an important prerequisite in photochemical reactions at the Martian surface, care has to be taken on the choice of UV sources for rigorous simulations of the Martian environment. Several photochemical reactions and specific interactions may be induced by a broad range of irradiation energies in the UV–VIS region (Spacek *et al.* 1995; Yen *et al.* 2000; Möhlmann 2004). This is why UV simulators, dedicated for photo catalytic Mars research, should be capable of emitting light in the full range of anticipated Martian surface fluxes.

UV exposure of spores and micro-organisms

During three space missions, using the BIOPAN facility of the European Space Agency ESA on board a Russian FOTON satellite, we investigated whether or not, and to what extent, natural soil, rock or meteorite material may protect bacterial spores against the harsh environment of space, especially against solar UV radiation, in the SURVIVAL experiments. For this purpose spores of *B. subtilis* HA101 were exposed to space either unprotected, or under a filter of clay, or mixed with different soil, rock or meteorite powders. Details about the BIOPAN facility and the flight hardware for SURVIVAL are described in Burger (1995). The results of the three space experiments together with laboratory experiments in space simulation chambers confirmed the deleterious effects of extraterrestrial solar UV radiation which, in contrast to the UV radiation reaching the surface of the Earth, also contains the very energetic, short-wavelength UVB and UVC radiation (Rettberg *et al.* 2004). A thin layer of clay did not protect the spores, if it was placed at some distance (about 5 mm) from the spore layer. Probably, tiny cracks in the clay layer allowed solar UV to reach the spore layer. A certain degree of protection could be reached by mixing the spores in the layer directly with clay or other rock or meteorite material (survival rates ranged between 10^{-3} and 10^{-4}). These results are consistent with observations during the PERSEUS mission on the MIR space station, where spores mixed with similar meteorite material were exposed to space for more than 3 months (Rettberg *et al.* 2002). Probably, soil or rock grains served as a shield against UV for those spores that were located beneath them. Maximum protection was achieved, if the spores were exposed to solar UV within a mixture of clay, rock or meteorite powder in a similar ratio as occurring in terrestrial soil (10^8–10^9 cm^{-3}). Within a column of 5 mm, sunlight was attenuated so efficiently that the same high survival rates were observed for both dark and sun-exposed flight samples.

Future experiments are planned again for the ESA BIOPAN facility, which will test the survivability of bacterial spores of *B. subtilis* embedded in minerals under the influence of UV radiation in space. Related laboratory studies are also planned. As biological endpoints in these investigations, the survival, and UV induced DNA-photoproducts and gene expression in *B. subtilis* after germination will be analysed. The results of these experiments will give new insights into the survivability of terrestrial organisms on Mars and will help to define adequate planetary protection measures.

UV experiments on bacterial communities

The indirect effects of UV radiation on different bacterial communities at present are not clear, because only two studies have simulated the exact Martian UV radiation with respect to both wavelengths and fluence rates (Green *et al.* 1971; Schuerger *et al.* 2003). Therefore, knowledge about the biological effects of true Martian UV radiation is still very sparse. Short-term simulation studies have revealed that Martian conditions negatively affect bacterial community activity (Hansen *et al.* 2005). However, more long-term simulation experiments in laboratories and space are necessary in order to get a better understanding of the bacterial community parameters that are permanently affected by indirect UV effects, e.g. by reactive oxygen radicals. Future

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simulation experiments that use exact Martian UV radiation are crucial for our understanding of how complex bacterial communities evolve and survive under simulated Martian conditions.

UV experiments with halobacteria

All resistance measurements of haloarchaea up to now were done on semisolid agar plates or in liquid suspensions, sometimes with stirring while irradiating the cells. However, it is unlikely that liquid salty pools exist on the surface of Mars, and thus, data from these experiments are to be interpreted with caution. It is more likely that in putative Martian sediments fluid inclusions are present, which could be considered as targets for the search for life. In addition, the transport of micro-organisms from Mars to Earth via meteorites is a possibility that emerged from the analysis of ALH 84001 (McKay *et al*. 1996). Therefore, the UV resistance of haloarchaea embedded in salt crystals (artificial halite) will be tested with the UV simulator. Recently a staining procedure for haloarchaea using the LIVE/DEAD kit was developed, which allows a rapid estimation of viable and non-viable cells in a sample by epifluorescence microscopy (Leuko *et al*. 2004). The application of this procedure greatly facilitates the observation of haloarchaeal cells under a wide range of experimental conditions, including entrapment in fluid inclusions of halite (Fendrihan & Stan-Lotter 2004). Besides using transparent halite as the embedding material for haloarchaea, small amounts of iron oxides, as they are present on Mars, and in terrestrial halite, could be added and their potential efficiency as a shielding material against simulated Martian UV light could be examined. The design of a parallel beam of UV light for the simulator will allow irradiation of multiple samples and thus the establishment of an experimental concentration course or the measurement of effects on different microbial strains under identical conditions.

Conclusions

Exposure to intense UV radiation usually results in the destruction of organic matter and deleterious effects at cellular level. In order to understand the various effects of UV irradiation, occurring at the recent Martian surface, a UV simulator was designed. The system posseses a spectral intensity output on the order of 55.6 W m^{-2} (at maximum level) between 200 and 400 nm, adjusted to theoretical models of UV fluxes at the recent Martian surface between 200 and 400 nm. Further demands on the system design are given by long-term stability, spectral and illumination homogeneity and portability. The UV simulator is dedicated to experiments in multiple fields of European Mars research: (i) UV induced photoreversion of pyrimidine dimmers; (ii) UV exposure of amino acids; (iii) heterogeneous catalysis and the formation of UV induced oxidants; (iv) the deleterious effects of UV on bacterial spores and halobacteria; and (v) UV radiation as a detrimental factor on the prosperation of bacterial soil communities.

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