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Bioastronomy 2007: Molecules, Microbes, and Extraterrestrial Life
ASP Conference Series, Vol. 420, 2009
K. J. Meech, J. V. Keane, M. J. Mumma, J. L. Siefert, and D. J. Werthimer, eds.

Exobiology at Southern Brazil: Spore Dosimetry and the UV Solar Radiation

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Abstract. The ultraviolet - UV is considered the range of solar radiation most immediately lethal to the life organisms on the Earth's surface. In this context, since 2000, the monitoring of the biologically-effective solar radiation using spore dosimeter at the Southern Space Observatory (29.4°S, 53.8°W), South of Brazil, has been performed. The biological dosimeter is based in the spore inactivation doses of *Bacillus subtilis* strain TKJ6312, who is sensitive to the UV solar radiation. Monthly expositions of biological dosimeter have been compared with solar irradiance obtained by Brewer spectrophotometer. Correlations indices about $r > 0.86$ shows the potential applicability of the biosensor in the monitoring of biologically-effective solar radiation. Since spores are stabile microorganisms, considering extreme environment variations, the biosensor may be used for studies of the effects of the solar radiation in others planetary environments for future work.

1. Introduction

The study of solar ultraviolet radiation (UVR) at different latitudes of the planetary surfaces has been discussed in the last decades, because the UV radiation plays a decisive role on the course of the biological evolution. The understanding of photobiological processes under different planetary climates like the early Earth, Mars and the comprehension of the role of the ozone leer protecting the biosphere for the UVB radiation is a very important aspects for better understanding the life evolution (Rontó et al. 2004).

Considering the exobiology context, a special attention has been given to Mars, due mainly the similarities between the present Mars and the early Earth, when considering the life appearance on the Earth (Jakosky 1998). Although the climatology effects of the UV on the early Earth is still relatively speculative, the study of the biological effects of UVR on Mars can help us to understand a possible biological evolution on Mars, as well as the transference mechanism of organisms from Mars to spacecraft and still this reasoning could be applied to understand the potential UV stress generated on the molecules of essential microorganisms presents in the early Earth's surface (Rontó et al. 2003). Therefore, studies have been focused in understanding of how it is possible to adapt the biological UV dosimetry's concepts and technologies developed under terrestrial conditions for the Mars environment; temperatures from -80°C to +20°C, atmospheric pressure about 7 mbar and a mixture atmospheric gases about 95.3 % CO_2, 2.7% N_2, 1.6% Ar, 0.2% O_2, 0.03% H_2O (Fajardo-Cavazos et al. 2007). In this work, the potential applicability of spores as biosensors for biologically-effective solar radiation on Earth is presented, aiming to open the discussion in terms the potential applicability of the spore dosimetry for and in Mars environmental conditions.

2. Methodology

Bacillus subtilis TKJ 6312. Due the deficiency of both DNA repair mechanisms, Nucleotide Excision Repair (NER) and Spore Photoproduct Lyase (SP lyase), the spores were genetically modified to be sensible to UVR, but maintaining the resistance of extreme environment conditions (Munakata et al. 2000). In this method, four spots of about 10^6 spores of *B. subtilis* strain TKJ 6312 are spotted on a membrane filter, which two spots were covered with cardboard to serve as unexposed controls. After, the samples are covered and dually wrapped with blue polyethylene sheets (Umeya Sangyo, Tokyo) and placed in a plastic slide holder (Munakata et. al. 2006). The samples are characteristically stable, when stored in dark either before or after exposure. After monthly exposures, the SID (Spore Inactivation Dose) is calculated from the natural logarithm of surviving fraction; SID = -ln (Ne/Nc), where Ne and Nc represent the average number of colony-formers recovered from exposed and control spots, respectively. The spore's action spectrum is obtained by "*Okazaki Large Spectrograph*" comprising from 254 nm to 400 nm (Munakata et. al. 1996). This is used in the calculation of SID by the Brewer spectrophotometer. The SID's values were compared with the integrals of monthly UVB as well the SID calculated by the Brewer Spectrophotometer (Schuch et. al. 2006). All exposures were performed at the National Institute For Space Research – INPE's, Southern Space Observatory (SSO) (29.4°S, 53.8°W), in Southern Brazil.

3. Results

A seasonal variation of Biologically-Effective Solar Radiation (BESR), in terms of SID and UV-B is presented in **Figure 1**. As expected, the temporal variation of BESR and UV-B presents a seasonal profile associated to the annual cycle of the zenithal solar angle. The data profiles shows higher correlation indices, 0.86 (2000), 0.86 (2001), 0.96 (2002), 0.88 (2003), 0.98 (2004) and 0.90 (2005). Considering the SID calculated, pondered by the Brewer and the SID observed, obtained by the spores (**Figure 2**), an elevated correlation index about $r \sim 0.91$ ($p < 0.0001$) is also observed.

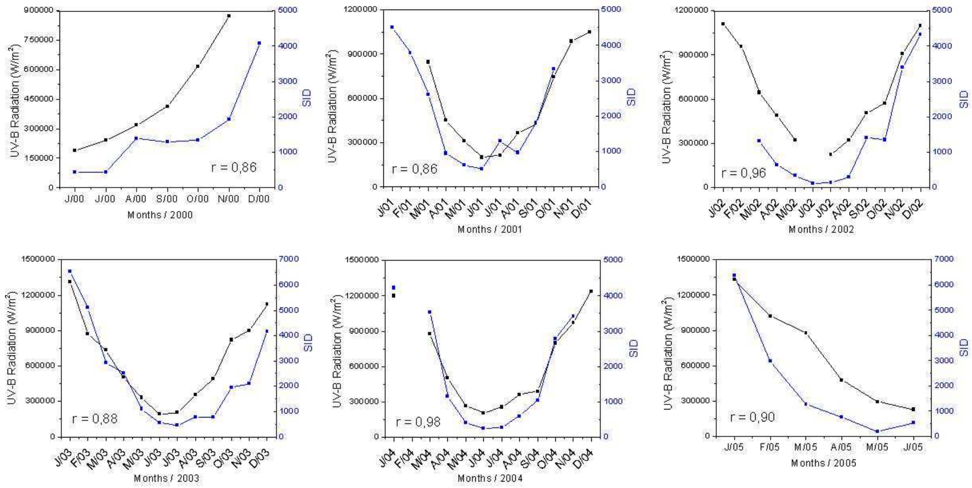

Figure 1. Seasonal profile of BESR obtained by Spores (blue line) and UV-B radiation, by Brewer (black line) at the INPE's Southern Space Observatory (SSO) for: 2000 - 2005

4. Discussion

Since the 70's the biological dosimetry has been developed, which studies have tried to apply it irrefutably as UV-biosensor and obeying the criterions established by BIODOS project from the European Commission (Horneck, 2000). The simplicity, facility of use and transport, long term storage, and well defined action spectrum is the principal requisites when field experiments are wished. The exposures have been made under natural sunlight by several minutes, days or weeks. However, for long term application, some problems need be circumvented including the reduction of the dose and the protection of the samples from adverses climatic conditions. This new stage, long term exposures, began in 2000 in our station at the SSO. The results observed in Figure 1 and 2 indicate the potential applicability of spores as biosensors of biologically-effective solar radiation on the Earth surfaces. To adapt this biosensor technology for Mars UV environments some considerations need to be considered. Mars receives characteristically 44% less of the total solar radiation compared with the Earth. However, the UVC and UVB presents in comparison higher levels, due the similarity lack between the Earth's and the Mars atmosphere (Mancinelli & Klovstad 2000). In the spores, NER and SP lyase appear sufficiently to explain the great majority of repair of damage by UVC and UVB (Nicholson et. al. 2005). As they are deficient of both repair mechanisms mentioned and considering a well defined action spectrum, they could be an alternative to study the biological implication of UVR on Mars surface. Although the UVR on Mars surface is higher in comparison with the observed values on the Earth, the dusts, rocks and ice shows that Mars surface presents physical substrates adequate to protect biomolecules against the harsh radiation regimen and allow the long-term survival of microbes under these substrates (Rontó et al. 2003). To ratify

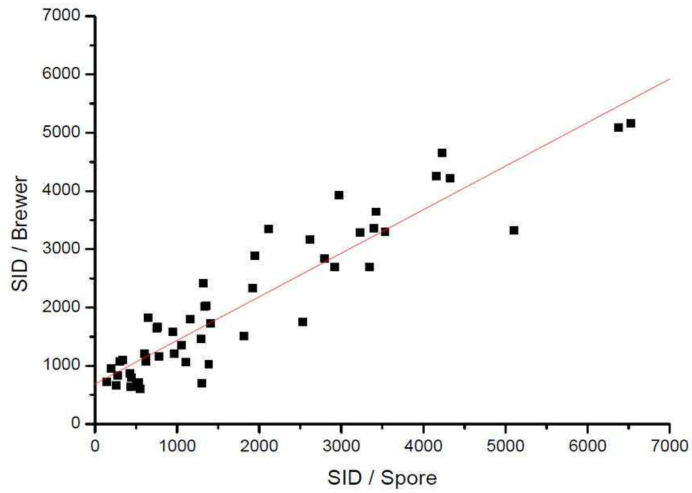

Figure 2. SID observed (by the Spores) versus SID calculated (by Brewer) for 2000-2005

the possible application of biosensor under others planetary environmental, simulations of Mars radiation conditions have been performed, where a layer of dust of 1 mm thickness shows to be completely effective in the prevention of spore inactivation, when submitted a exposure about 12 kJ (Mancinelli & Klovstad 2000).

5. Conclusions

The results presented in this paper indicate the potential applicability of spores as biosensors of biologically-effective solar radiation on the Earth surfaces. This biosensor technology may be adapted for application on the Mars surface but with some considerations for the difference of the UV environments.

Considering the interests of the international exobiology groups to study the spatial solar radiation under planetary environments, the application of *Bacillus subtilis* TKJ 6312 has been also considered. However, some problems are recognized to applications under extreme environments and further studies to elucidate these questions are still necessary. The current interests have been to find appropriate filter materials with good optical properties, reasonable and uniform transmittance for the UVC-UVB bands (Mars condition) with a same comparatively performance with the used under Earth's scenarios.

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