15 Ultraviolet Radiation in Planetary Atmospheres and Biological Implications

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15.1 Solar UV Radiation

The extraterrestrial solar spectrum extends far into short wavelengths of UV-C (190-280 nm) and vacuum UV (<190 nm), wavelengths that no longer reach the surface of the Earth. The intensity of solar radiation reaching the Earth's atmosphere would probably be lethal to most living organisms without the shielding afforded by the atmosphere.

Solar UV undergoes absorption and scattering as it passes through the Earth's atmosphere with the absorption by carbon dioxide, molecular oxygen and ozone being the most important processes. Carbon dioxide has a peak absorbance at 190 nm, and so attenuates radiation below 200 nm. Ozone forms a layer in the stratosphere, thinnest in the tropics (around the equator) and denser towards the poles. The amount of ozone above a point on the Earth's surface is measured in Dobson units (DU) - typically ~260 DU near the tropics and higher elsewhere, though there are large seasonal fluctuations. It is created when ultraviolet radiation strikes the stratosphere, dissociating (or "splitting") oxygen molecules (O_2) to atomic oxygen (O). The atomic oxygen quickly combines with further oxygen molecules to form ozone (O_3). Figure 15.1-A shows the absorption cross section of ozone as a function of wavelength and Fig. 15.1-B a part of the extraterrestrial solar spectrum compared to terrestrial spectra calculated for different ozone concentrations. Increasing ozone concentrations result in lower irradiances in the UV-B range of the spectrum. Surface UV-B radiation levels are highly variable because of sun angle, cloud cover, and also because of local effects including pollutants and surface reflections.

Solar UV radiation affects life on Earth today, and probably even has had a stronger impact on early evolution [1]. The composition of the Earth's atmosphere at that time differed from that of today. Although its exact composition is not known, from model calculations it can be assumed that during the Archaean era, during which the diversification of early anaerobes took place and the first anaerobic photosynthetic bacteria appeared (about 3.5 Ga ago), the amount of free oxygen in the atmosphere was significantly lower than today (see Chap. 14, Cockell)]. There was very little or no absorption of solar UV radiation by ozone. The situation on the early Mars might have been comparable (see Chap. 13, Lammer et al. and Chap. 14, Cockell). Taking

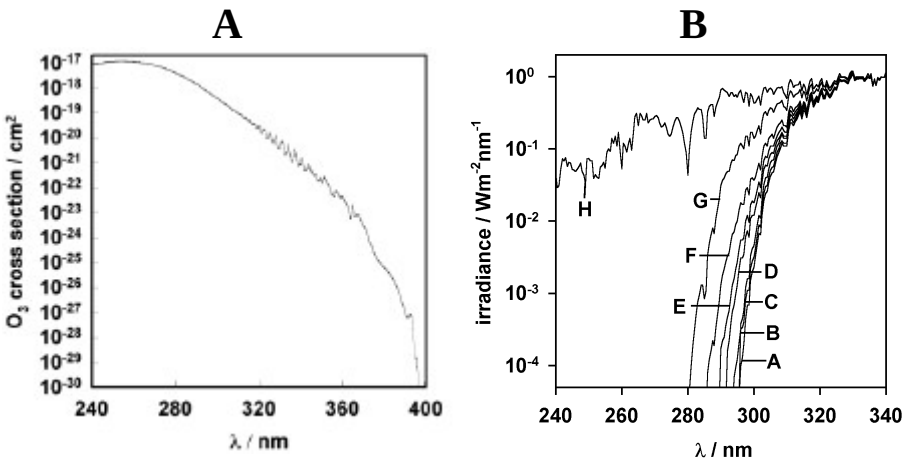

Fig. 15.1 Absorption cross section of O_3 (A) and solar irradiance calculated for different O_3 concentrations (B), A = 440 DU, B = 400 DU, C = 360 DU, D = 310 DU, E = 258 DU, F = 185 DU, G = 66 DU, H = extraterrestrial solar irradiance.

the presumed composition of the early Martian atmosphere and the lower solar luminosity into consideration radiative transfer calculations of the UV flux on the surface of Mars show a gradual increase of the irradiance including short-wavelength UV-B and UV-C over time until today. Thus, present-day solar UV irradiance on the surface of Mars may be similar to that on the surface of the Archaean Earth (see Chap. 14, Cockell).

15.2 Biological Effects of Solar UV Radiation

In biological systems, UV radiation causes photochemical reactions with different biological target molecules, the so-called chromophores. These interactions result in temporary or permanent alterations. The most important UV target in cells is the DNA because of its unique role as genetic material and its high UV sensitivity. The absorbing parts of DNA are the bases, the purine derivatives adenine and guanine, and the pyrimidine derivatives thymine and cytosine. Although the base composition of DNA is different 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 (Fig. 15.2). Absorption of proteins between 240 and 300 nm is much lower than that of nucleic acids of equal concentration in weight per volume. 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 like 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.

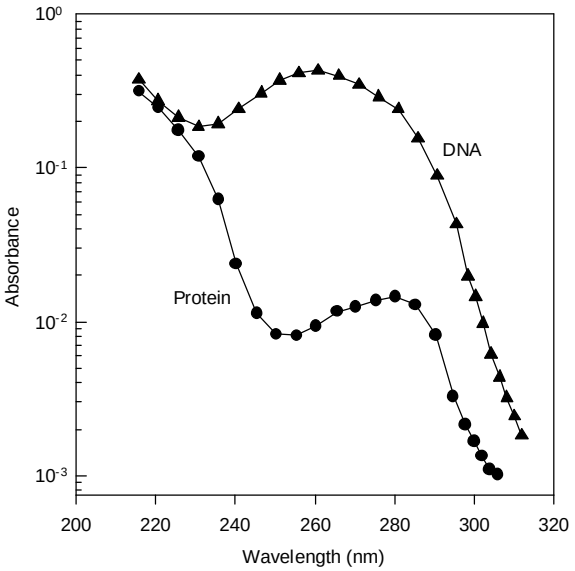

Fig. 15.2 Absorption spectra of DNA (calf thymus DNA) and a protein (bovine serum albumin) at identical concentrations.

The spectrum of UV radiation from the time that it 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, the chromophores. Therefore the action spectrum describing the wavelength dependence of a biological UV effect is often not identical to the absorption spectrum of a chromophore. In Fig.15.3 examples for normalized biological action spectra obtained with monochromatic radiation are given. These action spectra show a remarkable similarity of the slopes of the curves in the UV-B range. However, the curves differ from each other significantly in the UV-A range. This is caused by different photochemical reaction mechanisms. UV-B radiation is directly absorbed by the DNA molecules and causes photodamages. In contrast, UV-A radiation mainly excites so called photosensitizer molecules in the cell, which can either react with the DNA or with oxygen to give reactive oxygen species, which themselves can cause DNA damages.

Due to the wavelength specificity of biological action spectra, especially in the UV-B range, and the highly wavelength-specific absorption characteristics of components of the atmosphere like ozone, the assessment of the influence of environmental (polychromatic) UV radiation on critical biological processes requires a biological weighting of the solar UV irradiance according to the biological responses under consideration. The biological effectiveness of solar UV radiation is determined by the shape of the action spectrum of the biological endpoint and the spectral irradiance [2, 3] using equation 15.1

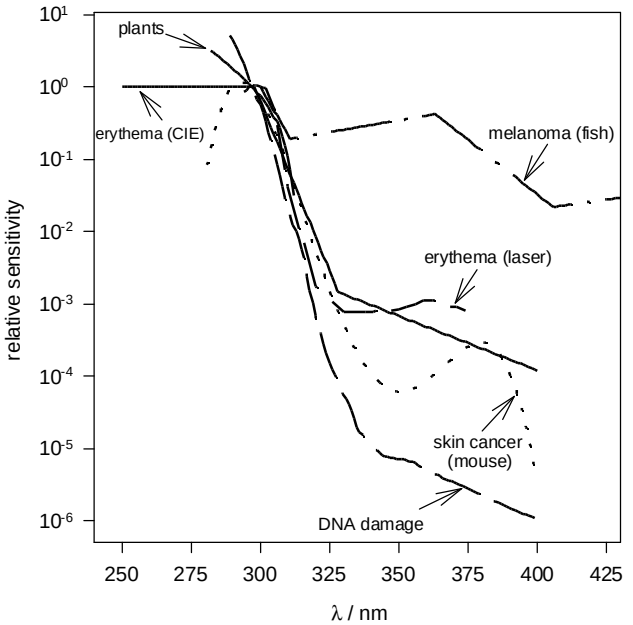

Fig. 15.3 Examples for different action spectra (from [2]).

$$E_{eff} = \int E_l(l) \cdot S_l(l) dl \tag{15.1}$$

with $E_l(l)$ = solar spectral irradiance (W/m² nm),
$S_l(l)$ = action spectrum (relative units), and
λ = wavelength (nm).

The resulting biological effectiveness spectrum is shown exemplary for a terrestrial UV spectrum in Fig.15. 4. Integration of the biologically effective irradiance E_{eff} over time (e.g., a full day, one year) gives the biologically effective dose H_{eff} $(J/m^2)_{eff}$ (e.g., daily dose, annual dose).

The significance of solar UV radiation as an environmental driving force for the early evolution of life on Earth is reflected by the development of different protection mechanisms against the deleterious biological effects of UV radiation [1]. The most important one is the development of several partly redundant enzymatic pathways for the repair (see Chap. 17, Baumstark-Khan and Facius) of UV-induced DNA damages very early in evolution [4]. Examples are (i) the photoreactivation (PHR), that is the removal of cyclobutane pyrimidine dimers and (6-4)pyrimidine-pyrimidone found in bacteria, Archaea and eukaryotes as a direct repair reaction in a single-step process, (ii) the nucleotide excision repair (NER) for the removal of bulky DNA lesions in bacteria, Archaea and eukaryotes, e.g., the UvrABCD pathway in *E. coli*, (iii) the

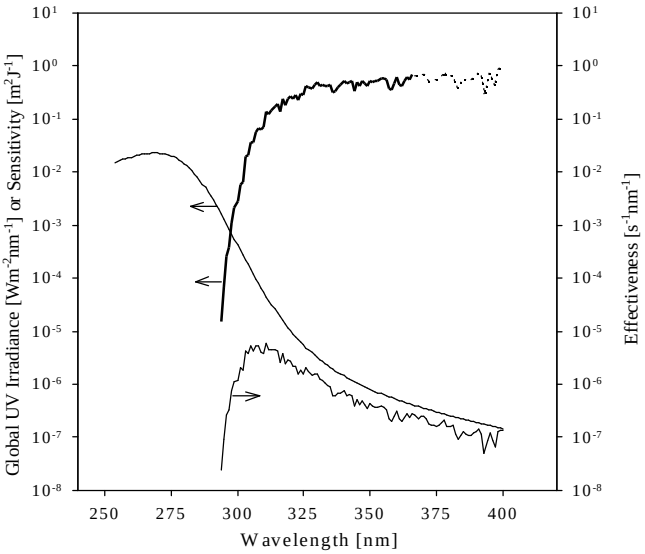

Fig. 15.4 Solar spectral irradiance at the Earth's surface (left axis, measured by A. Bais), a biological weighting function (the absolute DLR-Biofilm action spectrum, left axis) and the resulting solar effectiveness spectrum (right axis).

recently discovered alternative excision repair of UV-induced photoproducts in bacteria and eukaryotes (UV-DE pathway), (iv) the base excision repair (BER) for the removal of damaged or altered bases from the DNA backbone by DNA glycosylases in bacteria, Archaea and eukaryotes (several pathways exist for the removal of different types of oxidative DNA damages indirectly induced by UV-A, e.g., MutM in *E. coli*, Ogg 1 and 2 in yeast) and (v) recombinational repair with several pathways for homologous recombination in bacteria, Archaea and eukaryotes (for review of phylogenomic comparisons, see [4]). In addition to these essential enzymatic reactions for the protection of the genetic material against the deleterious effects of UV radiation, other mechanisms for protection against and avoidance of UV radiation have been developed, e.g., the formation of highly resistant metabolically inactive forms like spores, the synthesis of UV absorbing pigments, the trapping and binding of sediments to form microbial mats, the behavioral adaptation of motile organisms (see Chap. 16, Wynn-Williams and Edwards).

15.3 Biological UV Dosimetry

The assessment of the biological effects of a changing UV climate like that on Earth and Mars requires monitoring methods and systems, which take the strong wavelength dependence of all UV effects into account. One possibility is to use biological UV dosimeters, which consist of biological objects as UV targets and which directly

weight the incident UV radiation [3, 5-7]. Up to now several different targets have been suggested and partially tested as UV dosimeters, e.g., biomolecules like provitamin D_3, uracil or DNA, different bacteriophages, whole cells like vegetative bacteria and bacterial spores, biochemical processes in eukaryotic cells like gene induction or DNA repair. Most biological UV dosimeters, such as uracil, DNA, and bacteria are simple systems based on the DNA damaging capacity of UV radiation, which is suggested as the initiating event in a variety of critical photobiological reactions in key ecological processes [2, 7, 8]. Spores of the bacterium *Bacillus subtilis* were found to be well suited for biological UV dosimetry and are applied in the UV dosimeter 'DLR-Biofilm'.

Depending on the dosimetric requirements of the individual measurement the appropriate strain of *B. subtilis* has to be chosen. DNA-repair wildtype strains are more UV-resistant than DNA-repair defective strains, which are not or only partially able to repair the different types of UV-induced DNA damages. Especially strains with mutations in the *uvr* operon show in a high UV sensitivity. They can be used in DLR-Biofilm dosimeters for short-term measurements with a high temporal resolution whereas DNA-repair wildtype strains are used in DLR-Biofilm dosimeters for longer-lasting or higher exposures. For DLR-Biofilm preparation spores of *B. subtilis* were suspended in a 0.5 % agarose solution at 65 °C and poured on the horizontally orientated surface of polyester sheets (FMC Biozym, Oldenburg, Germany) at a concentration of 5×10^5 per cm^2. After solidification at room temperature these biofilms were dried for 12 h at 60 °C without air circulation.

DLR-Biofilms are exposed in different types of exposure boxes depending on the requirements of the individual measurements. During exposure some areas on each biofilm remain unexposed and serve as dark controls for the analysis or for calibration, which is performed with each biofilm before development applying known doses of UV-C from a standard mercury low pressure lamp. For development each biofilm is incubated in nutrient medium (Tryptic Soy Broth, Difco, Augsburg, Germany) at 37 °C for 4.5 h with chaotic mixing. During this time the bacterial spores, which are not or only slightly damaged by the previous UV exposure are able to germinate and multiply inside the biofilm. After fixation and staining of the biomass formed inside the biofilm the quantitative analysis of the optical density of measurement and calibration areas on each biofilm is performed with an image analysis system using a Charge Coupled Device (CCD) camera and a data calculation unit. As result of the comparison with the constructed calibration curve biologically effective UV doses are obtained as equivalent doses of UV-C of 254 nm giving the same biological effect.

DLR-Biofilms are suited for the widespread application as wavelength- and time-integrating biological UV dosimeters [11-13]. They measure continuously even under unfavorable conditions e.g., cloudy weather. Their response is independent of temperature at least between −30 and +70 °C. DLR-Biofilms can be transported and stored in the dark at room temperature before or after exposure for more than one year without loss of sensitivity [14]. The biofilm response is additive without dose rate effects and it is very similar to an ideal cosine if no additional entrance optics is used. The normalized spectral sensitivity of the biofilm, the action spectrum for monochromatic radiation, is shown in Fig. 15.5. Below 300 nm the biofilm sensitivity increases strongly due to the increase in the absorption spectrum of DNA.

15.4 Experimental Determination of the Biological Effectiveness of Extraterrestrial Solar UV Radiation

The full unadulterated spectrum of solar UV radiation is experienced only in space without any absorption and scattering processes form the atmosphere. Therefore the biological effectiveness of pure extraterrestrial solar UV radiation can only be measured in space. In the experiment SURVIVAL II on the exposure facility BIOPAN [15, 16] the biological effectiveness of solar UV radiation was quantified for the first time with the DLR-Biofilm technique on a non-stabilized satellite (Fig. 15.6) in Earth orbit (FOTON 9 mission, June 14-30, 1994). One DLR-Biofilm was exposed to the whole solar spectrum in the space vacuum. The four parts of it were covered only by four different neutral density filters to enlarge the dynamic range of the dosimeter. The exposure was performed by the opening of a shutter system connected to a timer for 10 s. A second DLR-Biofilm mounted on BIOPAN remained unexposed as dark control to exclude the possible influence of other experimental parameters in space than UV. After post-flight calibration and development the biofilms were analyzed together with a ground control biofilm, which was exposed to the terrestrial solar spectrum in Köln on August 6, 1993, under clear sky conditions. In space, a biologically effective dose of 63 J_{eff} m^{-2} was obtained in 10 s (Fig. 15.7), whereas on Earth 1 J_{eff} m^{-2} was

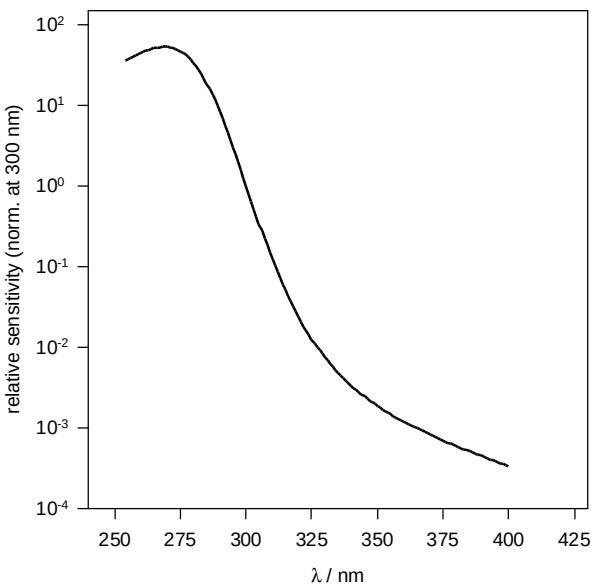

Fig. 15.5 The relative monochromatic action spectrum of the biological UV dosimeter 'DLR-Biofilm'.

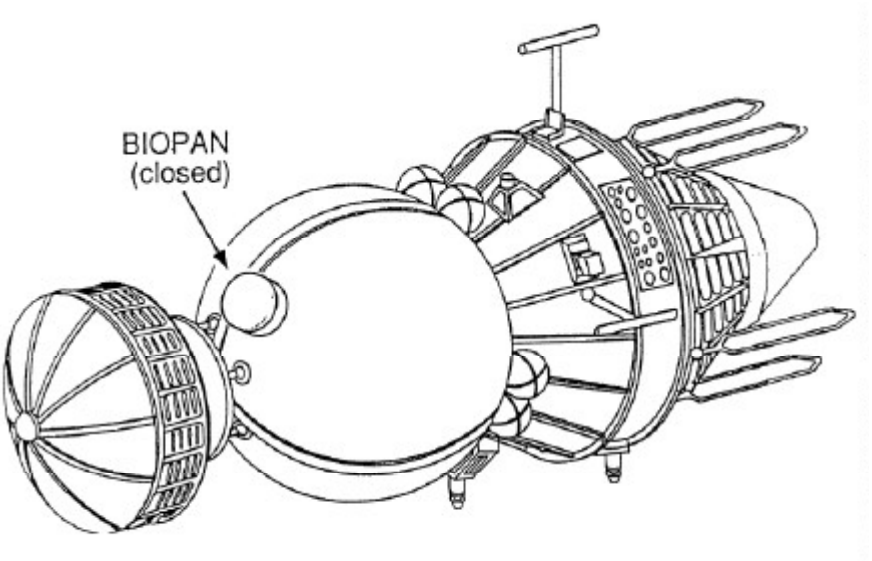

Fig. 15.6 The exposure facility BIOPAN mounted on the satellite.

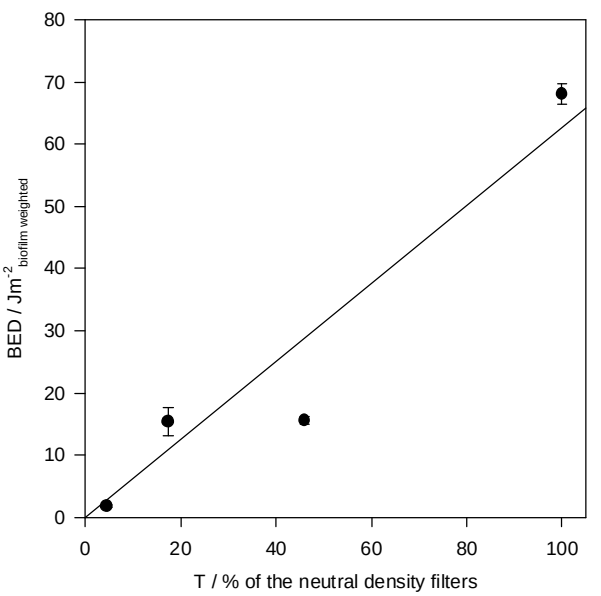

Fig. 15.7 Determination of the biologically effective dose (BED) of a 10 s exposure to extraterrestrial solar UV radiation on BIOPAN I.

measured in 2.6 min of solar exposure. Hence it can be concluded that the extraterrestrial solar radiation has a biological effectiveness, which is about thousand higher than the actual terrestrial solar UV radiation.

15.5 Experimental Determination of the Photobiological Effects of Different Ozone Concentrations

In another space experiment different atmospheric conditions were simulated to investigate the biological effects of an increase of ozone as it took place in the terrestrial atmosphere about 3.5 Ga ago. Conditions corresponding to an atmosphere without any ozone up to ozone concentrations higher than those experienced today on Earth were chosen in the simulation. During the Spacelab mission D-2 (April 26 to May 5, 1993) in the experiment RD-UVRAD DLR-Biofilms were exposed for defined intervals to extraterrestrial solar UV radiation filtered through an optical filtering system with a shutter mechanism. The filter combinations consisted of neutral density filters to enlarge the dynamic range of the DLR-Biofilm dosimeter and of short-wavelength cut-off filters to simulate different ozone column thicknesses in the stratosphere from 66 to 440 DU. After the predefined exposure period in space the DLR-Biofilms were transported back into the laboratory, calibrated, developed and analyzed together with a ground control biofilm, which was exposed to the terrestrial solar spectrum in Köln on August 6, 1993, under clear sky conditions. The results of the biological UV dosimetry are shown in Fig. 15.8-A with the measured biologically effective irradiance as a function of ozone concentration. At simulated present ozone concentrations (about 360 DU) the measured value is in good agreement with the actual ground measurement (GC = ground control). In Fig. 15.8-B the same data are normalized at 360 DU and compared to the solar UV irradiance in physical units. The results indicate that there is a strong decrease in biological effectiveness by increasing ozone concentrations (in contrast to a small increase in irradiance measured in physical units) and confirm the results of the BIOPAN experiment, that the unfiltered extraterrestrial solar radiation has a biological effectiveness nearly three orders of magnitude higher than the actual values on the Earth's surface at normal ozone concentrations [17, 18]. These results are in accordance with model calculations of the biological effective irradiance for the early Earth and allows the characterization of the conditions, under which life has evolved on Earth and might have evolved on other planets (see Chap. 14, Cockell).

15.6 Conclusions

Solar UV radiation has been a driving force for the evolution of life on Earth. It acts as a mutagen by its DNA-damaging capacity and as a selective agent at the same time. The terrestrial organisms have developed very early several different and

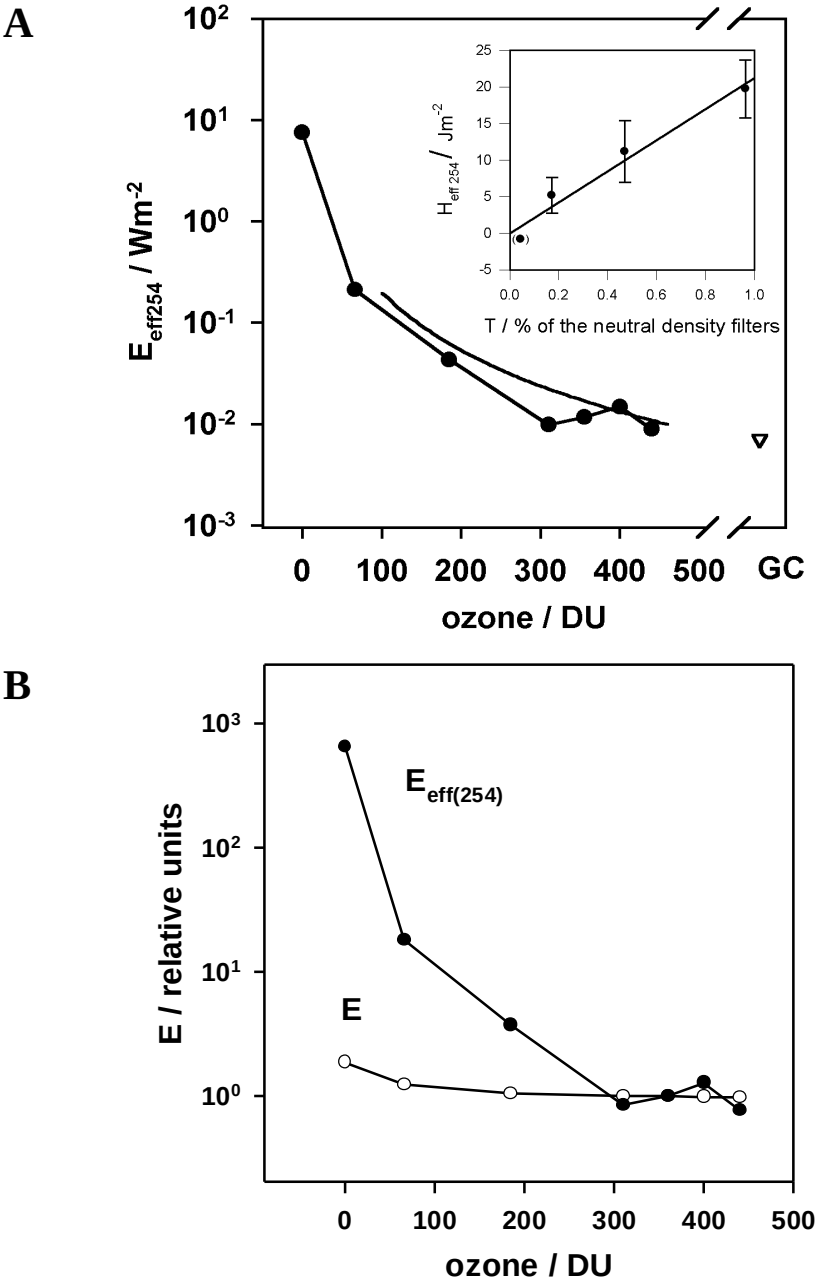

Fig. 15.8 (A) Biologically effective solar irradiance E_{eff} as a function of ozone concentrations (circles). Solid line: Calculated data under the assumption of a radiation amplification factor of 2, GC = ground control result, inset: biologically effective dose H_{eff} measured under different neutral densitiy filters; (B) The data from Fig. 15.8A are normalized at 360 DU (filled circles). Open circles: the solar irradiance in physical units, also normalized at 360 DU (from [17]).

complementary protection mechanisms against the deleterious effects of UV radiation. The most important one is the complex network of DNA repair enzyme activities. To assess the biological effectiveness of UV radiation biologically weighted measurements have been performed, which take the wavelength dependence of UV effects into account. The experimental data for the effectiveness of extraterrestrial solar UV radiation and its changes by an increasing ozone concentration in the atmosphere and the data from model calculations performed for the climatological conditions on the early Earth show a convincing agreement. This allows the characterization of the radiation conditions, under which life has successfully evolved on Earth and might have evolved also on other planets like Mars.

15.7 References

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