Induction of DNA Strand Breaks in Plasmid by VUV Photons in Aqueous Solution

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Introduction

Radiobiological effects in aqueous system are known to be mediated mainly by water radicals such as OH. Studies on the radical-mediated yields of DNA strand break induction in aqueous solution are fundamentally important in understanding the biological efficiency of radiation. We already reported those radicals are produced by vacuum ultraviolet(VUV) photons using Fricke dosimeter [1]. As the next step, we measured the yields of strand breaks in plasmid by VUV photons in aqueous condition..

Materials and Methods

We developed a new apparatus, which consists of a MgF2 window and a rotating quartz disc. MgF2 window protects the vacuum of the beamline, but can transmit VUV photon above 130 nm. The gap between the window and the disc is adjustable as small as 0.05 mm. Since VUV photons are absorbed in a very thin layer attached to the window in the sample, we rotate the disc to make absorption of VUV photons in the sample homogeneous as time average.

We put 5 μl of plasmid solution on the disc, which contains 0.5 μg of DNA in TE buffer (10 mM Tris, 1 mM EDTA). The gap between the window and the disc was adjusted to be 0.15 mm in order to cover the VUV beam area, typically 3 mm in diameter. The rotation speed of the disc was about 10 rpm. The VUV photon flux was measured with batch-calibrated photodiode (International Radiation Detectors, USA), which was set in vacuum, and the transmission of MgF2 window was taken into account in the calculation of dose. Irradiation was done at BL-20A. Photon flux was around 10^{12} photons per sec. Irradiated samples were analyzed with electrophoresis and strand breaks were quantified.

Results and Discussions

Induced number of strand breaks was plotted against the absorbed energy in the sample, and the yield was calculated from the slopes of the linear fitting, although small quadratic components were observed. Typical dose-yield relationships are shown in the figure and obtained yields are tabulated in Table 1. Interestingly, wavelength dependence of the single strand break yield was different from that of double strand break, indicating that different mechanisms contribute to the induction of strand breaks.

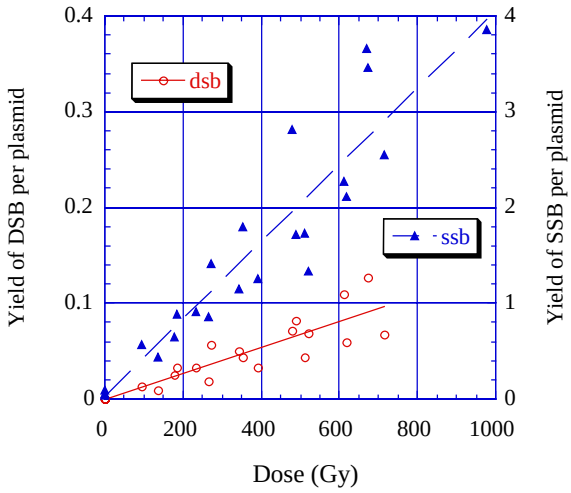

Table 1: Yields (Gy^{-1}) of single strand break (SSB) and double strand breaks (DSB) by VUV photons

Wavelength (nm)	SSB	DSB
150	1.3E-3	1.5E-5
160	1.9E-3	3.1E-5
170	4.0E-3	1.3E-4
180	6.2E-3	7.9E-5

References

[1] Watanabe et al, Radiat. Res., (1997) **148**, 489-490
[2] Kobayashi et al, Photon Factory Activity Report (1999) 258

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