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INT. J. RADIAT. BIOL., 1980, VOL. 38, NO. 3, 323–334

Cerenkov ultraviolet radiation (^{137}Cs γ-rays) and direct excitation (^{137}Cs γ-rays and 50 kVp X-rays) produce photoreactivable damage in *Escherichia coli*

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(*Received 12 December 1979; accepted 18 March 1980*)

The mechanism of formation of photoreactivable damage in deoxyribonucleic acid (DNA) by ionizing radiation in a dark repair deficient strain of *Escherichia coli* (*uvrA recA*) has been investigated. By altering the ratio of damage produced directly (by ionization) to that formed indirectly (by Cerenkov ultraviolet (U.V.) radiation) by ^{137}Cs γ-rays, it has been demonstrated that the major portion of the photoreactivable damage is produced by Cerenkov U.V. radiation. The amount of photoreactivable damage produced by 50 kVp X-rays, which cannot generate Cerenkov radiation, is similar to that component of photoreactivable damage produced by ^{137}Cs γ-rays that is not attributed to Cerenkov radiation. It is suggested that the second mechanism of formation of photoreactivable damage in DNA by ionizing radiation is the direct excitation of DNA. The possible role of Cerenkov U.V. radiation in ionizing radiation mutagenesis is discussed.

1. Introduction

Photoreactivation, an enzymatic DNA repair process formerly thought to act only after ultraviolet (U.V.) irradiation, was observed by Myasnik and Morozov (1977) after inactivation of dark repair deficient strains of *Escherichia coli* with ionizing radiation. The proportion of damage susceptible to photoreactivation has been shown to be independent of the presence of oxygen during irradiation (Redpath and Tortorello 1977, Vinicombe, Moss and Davies 1978). The enzymatic nature of photoreactivation after ionizing irradiation has been demonstrated by Wang and Smith (1978) using a *phr* mutant of *E. coli*. These authors also correlated the yield of γ-ray-induced pyrimidine dimers (the substrate for the photoreactivating enzyme), as measured by chromatography, with the production of photoreactivable damage, as measured by biological recovery.

Two aspects of the phenomena of photoreactivation of ionizing radiation-induced damage, which at present have not been satisfactorily explained, are the variable amount of photoreactivable damage that has been observed for the different energies of radiation, and the mechanism of formation of this damage. The existing data are summarized in table 1 in terms of photoreactivable sectors (PRS), where PRS is defined as $(k_{irr}-k_{pr})/k_{irr}$, and k_{irr} and k_{pr} are the inactivation constants from the survival curves after irradiation, without and with maximum photoreactivation, respectively. Where k values have not been given in the literature they have been

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0020–7616/80/3803 0323 $02·00

Radiation	*E. coli* strain	PRS	Reference
50 kVp X-rays	AB2480 *uvrA recA*	0·07†	Wang and Smith (1978)
100 kVp X-rays	AB2480 *uvrA recA*	'little or none'	Redpath and Zabilansky (1979)
	B_{s-1} *uvrB exrA*	'little or none'	
300 kVp X-rays	AB2480 *uvrA recA*	0·05–0·10‡	Redpath and Zabilansky (1979)
	B_{s-1} *uvrB exrA*	0·07‡	
660 keV ^{137}Cs γ-rays	AB2480 *uvrA recA*	0·23§	Wang and Smith (1978)
1·2 MeV ^{60}Co γ-rays	B_{s-1} *uvrB exrA*	0·33§	Myasnik and Morozov (1977)
	WP2 *hcr exr*	0·32§	
1·2 MeV ^{60}Co γ-rays	AB2480 *uvrA recA*	0·37‖	Vinicombe, Moss and Davies (1978)
6 MVp X-rays	AB2480 *uvrA recA*	0·24–0·26‡	Redpath and Zabilansky (1979)
	B_{s-1} *uvrB exrA*	0·19–0·22‡	
25 MeV electrons	AB2480 *uvrA recA*	0·28§	Redpath and Tortorello (1977)
32 MeV electrons	AB2480 *uvrA recA*	0·42†	Moss and Davies (1979)

† Calculated from unpublished data referred to in the reference;
‡ estimated from a single point on a composite graph.
§ Estimated from full survival curves.
‖ Published value.

Table 1. Values from the literature for the photoreactivable sector (PRS; see text for definition) of damage after inactivation of strains of *E. coli* by ionizing radiation of various energies.

estimated from the published graphical data. It can be seen that there is a general trend for larger values of PRS after exposure to higher energy radiations, especially when different radiations have been used by the same workers using standardized conditions.

Redpath and Zabilansky (1979), using the free-radical scavenger dithiothreitol, have presented data for 6 MVp photons, which suggest that photoreactivable damage may be produced via an excited state mechanism rather than via a free-radical mechanism. Two possible mechanisms by which ionizing radiation may produce excited species and thus produce pyrimidine dimers in DNA are by direct excitation (Smith 1976) or by Cerenkov radiation. The latter is electromagnetic radiation, including U.V. radiation, which is produced when a charged particle passes through any medium in which the phase velocity of light is less than the particle velocity. Cerenkov radiation is typically observed as the blue light emitted by radiation sources stored under water. Mathematical descriptions arising from classical physics and quantum mechanics, summarized by Jelly (1958), show that the relative and total energies of the wavelengths of light produced depend on the refractive index of the medium, the energy of the charged particles and their path length. One factor arising from the mathematical description is that Cerenkov U.V. radiation cannot be produced by radiations of energy less than 236 keV in aqueous solutions having a refractive index for U.V. radiation of $\sim 1{\cdot}4$ (see Appendix). It is of interest in this respect that Redpath and Zabilansky (1979) observed 'little or no' photoreactivation after exposure of *E. coli* to 100 kVp X-rays, but Wang and Smith (1978) did observe a small amount of photoreactivation after exposure to 50 kVp X-rays.

The goal of this work was to elucidate the method of formation of photoreactivable damage in *E. coli* by ionizing radiation. The experimental approach was to use conditions that vary the yield of Cerenkov radiation initially produced and/or subsequently absorbed by the bacteria, and then to observe what effect this has on the photoreactivable sector of the cellular damage induced.

2. Materials and methods

2.1. *Bacterial strains and culture condition*

SR18 is the *E. coli* K-12 derivative AB2480 *uvrA6 recA13* obtained from R. P. Boyce. SR362 is a derivative of *E. coli* K-12 (KH21) into which the *uvrA6*, *uvrB*, *recA56* and *phr* mutations have been transferred (Wang and Smith 1978). Stationary phase cultures were grown in a shaking water bath in 50 ml flasks containing 20 ml of nutrient broth (Difco) with yeast extract (Difco) added at 0·75 per cent. Innoculation was always with 0·2 ml of an overnight culture containing approximately 2×10^8 colony forming units (CFU) per ml, and cells were always harvested after 24 hours. This procedure was adhered to in order to minimize variation in sensitivity and photoreactivability, as previously demonstrated (Tyrrell, Moss and Davies 1972a, b). Cells were routinely harvested on membrane filters (Milipore Corp., HA, 0·45 μm pore size), and resuspended in 0·067 M $NaKPO_4$ buffer, pH 7·0, at a concentration of 10^7 CFU ml^{-1}. In those experiments using higher cell concentrations, harvesting and washing was done by centrifugation.

2.2. *Irradiation conditions*

Irradiations were performed on cell suspensions in equilibrium with the atmosphere without additional gassing, as it has been shown previously that the strains used, when grown as described, exhibit no enhancement of inactivation in the presence of oxygen (Vinicombe, Moss and Davies 1978).

X-irradiation was performed using a twin-tube 50 kVp X-ray unit (Loevinger and Huisman 1965). The X-ray tubes were operated at 48 and 50 mA in the vertical position, the top 50 mA tube with 0·3 mm Al added filtration, and the bottom tube blocked by 3·2 mm lead. A 4 ml volume of the cell suspension was irradiated in a 60 mm diameter black Bakelite dish. The dose-rate was determined by ferrous sulphate dosimetry, using a G value of 14·6 (Geisselsoder and Karzmark 1969), and a correction factor for the mass absorption coefficient of a biological sample as compared with the dosimeter solution (Loevinger and Huisman 1965). The dose-rate thus determined was 3·64 krad min^{-1}.

Gamma-irradiation was performed using an 8000 Ci ^{137}Cs source (J. L. Shepard and Associates), emitting a γ-photopeak with an energy of 660 keV. The irradiation vessel was either a quartz tube or, in one experiment a glass tube, of internal diameter 3·25 mm and external diameter 4·10 mm. Separate 0·8 ml cell suspensions were irradiated for each dose with the irradiation vessel held in position (a) in figure 1. This arrangement permitted irradiation with or without a large volume (800 ml) of distilled water in the surrounding glass beaker (b). Dosimetry was with the ferrous sulphate method described above, using a G value of 15·6 (Shalek and Smith 1969) with no mass absorption correction. The dose-rate was determined to be 1·54 krad min^{-1} with the surrounding 800 ml of water, and 1·68 krad min^{-1} without it.

In specified experiments the cells were irradiated in the presence of varying concentrations of DNA. A stock solution of DNA was prepared by dissolving calf

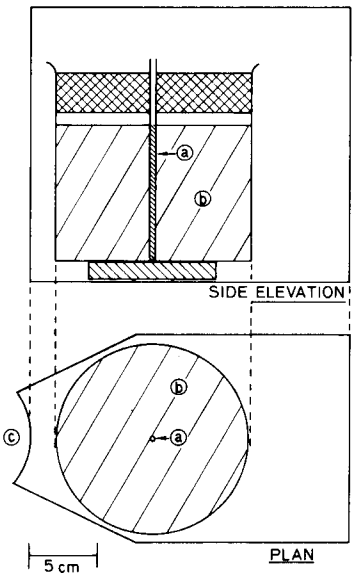

Figure 1. Diagrammatic representation of the irradiation vessel and chamber used for ^{137}Cs γ-ray irradiation; (*a*) quartz or glass vessel containing bacterial suspension, (*b*) glass beaker containing, when specified, 800 ml distilled water; and (*c*) the ^{137}Cs source.

thymus DNA (Worthington Biochemical Corp., N.J.) in 0·067 M $NaKPO_4$ buffer, pH 7·0, at a concentration of 0·5 per cent, and the required dilutions were prepared at the same pH.

2.3. *Survival curves*

For survival measurements, controls and irradiated cells were diluted in 0·067 M $NaKPO_4$ buffer, pH 7·0, and plated on nutrient agar containing 0·75 per cent added yeast extract. The plates were incubated for 48 hours at 37°C to allow colony formation. Inactivation constants (k) and y-axis intercepts (n) were estimated by least squares regression fitting of the equation: $S/S_0 = n \exp(-kd)$, where S/S_0 is the surviving fraction, and (d) is the dose.

2.4. *Photoreactivation*

Suspensions for photoreactivation were placed in 1 cm^2 spectrophotometer cuvettes. Between irradiation and photoreactivation a minimum period of 10 min was allowed to permit complexation of photoreactivating enzyme with DNA lesions. Photoreactivating light was administered to the suspensions in the cuvettes held at room temperature (25 ± 2°C). Illumination was supplied from a 500 W tungsten lamp (General Electric GE-DAK) in a slide projector (Argus 500 Model V). The light was filtered with a purple glass filter (Ealing 26-3384) that transmitted light of wavelengths between 350 and 425 nm. The time used for photoreactivation was 15 min, and the dose-rate was determined with a calibrated thermopile to be 100 Wm^{-2}. These conditions have previously been shown to achieve maximum photoreactivation of the strain SR18 (unpublished results).

Irradiated samples were collected under red light, and all other manipulations were performed under fluorescent yellow light.

3. Results and discussion

3.1. *The effect of increasing Cerenkov radiation*

The initial experiments were designed to differentiate between direct inactivation by the ionizing radiation, and any inactivation that might be due to Cerenkov U.V. radiation produced by passage of the ionizing radiation through the aqueous medium. The bacterial strain chosen for this and subsequent experiments was *E. coli* K-12 (AB2480) *uvrA recA* (SR18), which is sensitive to ionizing and U.V. radiation and, of importance for these experiments, is deficient in its ability to repair U.V. radiation-produced pyrimidine dimers, except by photoreactivation. A suspension of SR18, at 10^7 CFU ml^{-1}, was irradiated with ^{137}Cs γ-rays in a tube in position (a), as shown in figure 1. The viability was assessed after a range of doses, each with and without maximum photoreactivation. The conditions were varied by using either a glass or a quartz tube, and then by irradiating with or without 800 ml of distilled water in the surrounding beaker (*b*). Four representative survival curves are shown in figures 2 (*a*) and 2 (*b*), and the inactivation constants before (k_{irr}) and after maximum photoreactivation (k_{pr}) are included in table 2. It can be seen in figure 2 (a), using a quartz tube, that the surrounding water results in increased inactivation, and also in an increase in the PRS of damage from 0·29 to 0·42. When a glass tube, which does not transmit significantly below 300 nm, was used instead of quartz, the surrounding water did not increase the inactivation or the PRS (0·28 versus 0·23). It is inferred from these results that the increase in inactivation and PRS observed in figure 2 (a) with water in the surrounding volume is due to U.V. radiation produced as Cerenkov radiation in the surrounding water.

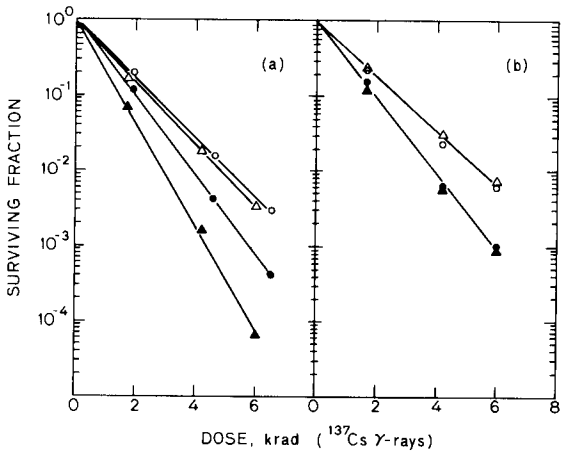

Figure 2. Survival of SR18 *uvrA recA* after ^{137}Cs γ-irradiation, (*a*) in quartz vessel, (*b*) in glass vessel, with (▲, △) and without (●, ○) 800 ml of water in the surrounding vessel. The open symbols represent survival after maximum photoreactivation.

3.2. *The effect of including U.V.-absorbing material in solution in the bacterial suspension*

A second series of experiments was designed to see if the sector of photoreactivable damage observed without any contribution from the surrounding water is also due to Cerenkov U.V. radiation; in this case generated within the bacterial suspension inside the small tube (*a* in figure 1). The approach adopted was to include

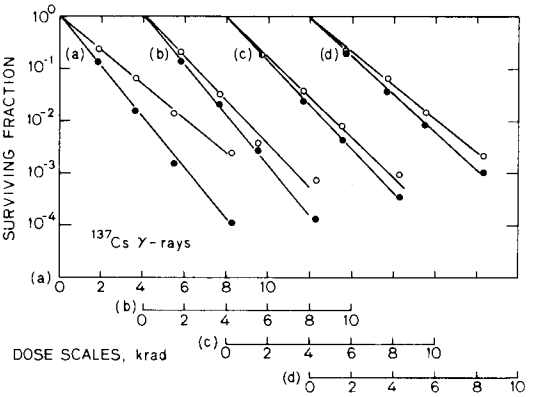

Figure 3. Survival of SR18 *uvrA recA* after ^{137}Cs γ-irradiation in the presence of various concentrations of DNA in solution; (*a*) 0·0% DNA, (*b*) 0·00475%, (*c*) 0·0475%, and (*d*) 0·475%. The closed symbols (●) are without photo-reactivation, and the open symbols (○) after maximum photoreactivation.

Experimental conditions	k_{irr}	k_{pr}	PRS
Effect of increased Cerenkov radiation—^{137}Cs γ-rays			
Quartz tube (†)	1·19	0·85	0·29
Quartz tube and surrounding water	1·55	0·89	0·42
Glass tube	1·09	0·78	0·28
Glass tube and surrounding water	1·08	0·83	0·23
Effect of including DNA in solution—^{137}Cs γ-rays			
No DNA (†)	1·19	0·85	0·29
0·00475% DNA	1·08	0·88	0·19
0·0475% DNA	0·95	0·81	0·15
0·475% DNA	0·81	0·71	0·12
Effect of increasing cell concentration—^{137}Cs γ-rays			
1×10^7 CFU ml^{-1} (†)	1·19	0·85	0·29
2×10^8 CFU ml^{-1}	1·03	0·74	0·28
2×10^9 CFU ml^{-1}	0·89	0·75	0·15
2×10^{10} CFU ml^{-1}	0·83	0·70	0·15
Photoreactivation after 50 kVp X-rays			
Control	1·07	0·97	0·09
0·00475% DNA	1·06	0·94	0·11
0·475% DNA	1·04	0·91	0·12
SR362*phr*	0·96	0·94	0·00

† A suspension of 1×10^7 CFU ml^{-1}, in a quartz tube, with no added DNA is the control condition and the values given are the average of five determinations.

Table 2. The inactivation constants for *E. coli* SR18 *uvrA recA* inactivated by ionizing radiation under various experimental conditions without (k_{irr}) and with (k_{pr}) maximum photoreactivation.

varying concentrations of calf thymus DNA in solution in the bacterial suspension. The choice of DNA was made as a material that will absorb the wavelengths of most interest, and also as a material that will remain outside the cell and be unlikely to influence inactivation by the ionizing component of the γ-rays. The final concentrations of DNA in the irradiated bacterial suspensions were 0·00475, 0·0475 and 0·475 per cent and had optical densities at 254 nm (1 cm) of 0·96, 9·6 and 96, respectively. The survival curves for 10^7 CFU ml^{-1} suspensions of strain SR18 in the absence and in the presence of DNA, and irradiated with ^{137}Cs γ-rays are shown in figure 3. The inactivation constants for these curves together with values for PRS are included in table 2. It can be seen that the PRS cannot be reduced below 0·12 even by a one hundred-fold increase in 254 nm absorption by the suspending medium. This is taken as evidence that when a 10^7 CFU ml^{-1} suspension of strain SR18 is irradiated under our control conditions (table 2), approximately 70 per cent of the photoreactivable damage observed is due to Cerenkov U.V. radiation generated in the suspension, the remaining 30 per cent is produced within the cells by a different process.

3.3. *Photoreactivation after 50 kVp X-rays*

Cerenkov U.V. radiation cannot be generated in aqueous solutions of refractive index close to that of water by radiation having energies less than approximately 236 keV (see Appendix). However, photoreactivation has been observed after 50 kVp X-rays (Wang and Smith 1978). In order to determine that this observation was not an artifact caused by radiation-induced luminescence of the plastic irradiation vessel, the experiment of Wang and Smith (1978) was repeated using an opaque black plastic vessel. The survival curves with and without photoreactivation are shown in figure 4 (*a*). Corresponding data for the *phr* strain SR362 are shown in figure 4 (*d*). It can be seen from the inactivation constants and PRS values included in table 2 that photoreactivation is observed after 50 kVp X-rays, and that it is due, by inference from the negative result with the *phr* strain, to the activity of the photoreactivating enzyme. It is of interest that the sector observed, 0·09, was very similar to the residual PRS observed when the effect of Cerenkov radiation was minimized during ^{137}Cs γ-irradiation (table 2). For confirmation that the photoreactivation observed after 50 kVp X-rays was not due to U.V. radiation generated in the aqueous medium, DNA was included in the solution at concentrations of 0·00475 and 0·475 per cent, which were shown to minimize the contribution of Cerenkov radiation in the experiments with ^{137}Cs γ-rays (figure 3). The survival curves are shown in figures 4 (*b*) and 4 (*c*). The inactivation constants and PRS values included in table 2 show no reduction in PRS from the value obtained in the absence of DNA.

3.4. *The effect of cell concentration on the photoreactivable sector*

Responses of cell populations to ionizing radiation are often investigated at concentrations higher than that used (10^7 CFU ml^{-1}) in the preceding experiments. Consideration of the nature of the photoreactivable component of ionizing radiation damage described in this paper suggests that the cell concentration should influence the contribution of Cerenkov U.V. radiation to cell inactivation.

Suspensions of strain SR18 in the quartz tube at position (a) (figure 1) without surrounding water in (b) were irradiated with ^{137}Cs γ-rays using cell concentrations of 10^7, 2×10^8, 2×10^9 and 2×10^{10} CFU ml^{-1}. The survival curves are shown in figure 5. From the inactivation constants and PRS values in table 2, it can be seen that

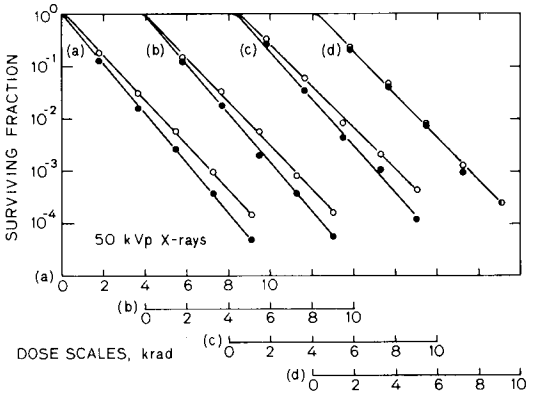

Figure 4. Survival of SR18 *uvrA recA* after 50 kVp X-irradiation in the presence of various concentrations of DNA in solution (*a*) 0·0% DNA, (*b*) 0·00475% DNA, and (*c*) 0·475% DNA. Survival of SR362 *uvrA uvrB recA phr* after 50 kVp X-irradiation (*d*). The closed symbols (●) are without photoreactivation, and open symbols (○) after maximum photoreactivation.

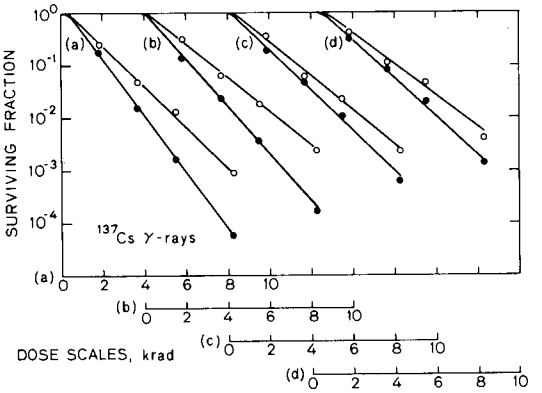

Figure 5. The survival of SR18 *uvrA recA* after ^{137}Cs γ-irradiation assessed using initial cell concentrations of: (*a*) 1×10^7 ml^{-1}, (*b*) 2×10^8 ml^{-1}, (*c*) 2×10^9 ml^{-1}, and (*d*) 2×10^{10} ml^{-1}. The closed symbols (●) are without photoreactivation, and the open symbols (○) are after maximum photoreactivation.

increasing the cell concentration from 10^7 to 2×10^8 CFU ml^{-1} did not significantly affect the results obtained, but an increase to 2×10^9 CFU ml^{-1} decreased the sensitivity without photoreactivation, and decreased the PRS value obtained. A further increase to 2×10^{10} CFU ml^{-1} did not result in any further decrease in sensitivity or PRS value. This result may be interpreted by considering two factors, the optical density of the cell suspension to the inactivating wavelength of U.V. radiation (254 nm as a first approximation), and the total number of cells present in the suspension. The optical densities at 254 nm (1 cm) of the cell suspensions used (10^7, 2×10^8, 2×10^9 and 2×10^{10} CFU ml^{-1}) were 0·17, 3·4, 34 and 340, respectively. If these values are adjusted for the path length of the Cerenkov U.V. radiation within the quartz irradiation tube (approximately 1·3 mm), then the effective optical densities of the four suspensions used are 0·22, 0·44, 4·4 and 44, respectively,

corresponding to values for per cent transmission of 95, 36, 4×10^{-3} and 1×10^{-46}, respectively. At a cell concentration where a significant proportion of the light is transmitted out of the suspension, the cells will receive approximately the same dose of U.V. radiation independent of the number of cells present. However, in the two cases when effectively all the U.V. radiation was absorbed (i.e., per cent $T = 4 \times 10^{-3}$ and 1×10^{-46}) the inactivation was limited by the ratio of the number of photons available (a constant) to the number of cells present. This is in agreement with the observation that an increase in concentration from 10^7 to 2×10^8 CFU ml^{-1} did not significantly affect the survival curves obtained, but that an increase to 2×10^9 CFU ml^{-1} resulted in a higher survival in the dark, and a lower PRS value, i.e., a reduction in the effect of the Cerenkov U.V. radiation. The observation that a further increase in cell concentration to 2×10^{10} CFU ml^{-1} did not eliminate the PRS is further evidence that this small residual sector of photoreactivable damage (PRS ~ 0·1) is not formed by Cerenkov radiation.

4. Conclusions

Evidence is presented that approximately 70 per cent of photoreactivable damage observed when the *uvr A rec A* strain of *E. coli* (SR18) was irradiated with ^{137}Cs γ-rays was due to Cerenkov U.V. radiation, and that the remaining 30 per cent was induced by a different mechanism. A small amount of photoreactivable damage was seen after irradiation with 50 kVp X-rays, which cannot generate Cerenkov radiation under the conditions used. It is suggested that this damage, and that portion of photoreactivable damage after ^{137}Cs γ-irradiation that is not due to Cerenkov radiation is formed by the same mechanism, perhaps by direct excitation of the DNA (Smith 1976).

Published results, summarized in table 1, where significantly larger PRS values have been observed with ^{60}Co γ-rays or 32 MeV electrons than with ^{137}Cs γ-rays, are in agreement with the conclusion that Cerenkov U.V. radiation is responsible for the production of photoreactivable damage. It can be seen from figure 6 in the Appendix that increasing the energy of the ionizing radiation results in an increased production of Cerenkov radiation. This increase is correlated in figure 6 with the increased PRS values observed. For the purpose of this correlation the residual PRS of 0·1, which is not attributed to Cerenkov radiation, has been assumed to be constant and removed from the values of PRS.

The formation of lesions induced by U.V. radiation during ionizing irradiation may have important consequences. Relative to their respective lethal efficiencies, U.V. radiation is much more efficient than ionizing radiation in producing mutations. The possibility should be considered that a portion of observed mutation induction by ionizing radiation may be due to the very small but relatively much more efficient U.V. radiation-type damage. This possibility is under investigation.

Appendix

When a charged particle, having a velocity (V) approaching the velocity of light in vacuum (c), passes into a medium in which the phase velocity of light is less than the velocity of the particle, then electromagnetic radiation is emitted. This radiation is called after P. A. Cerenkov who provided the first explanation of its origin. Cerenkov's original papers published between 1934 and 1938 together with related

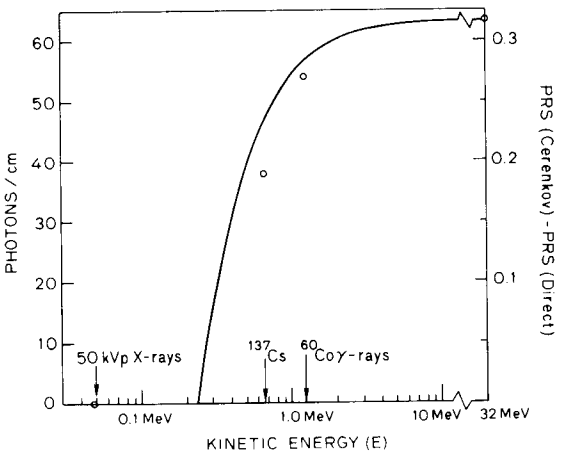

Figure 6. The continuous curve represents the relative amount of Cerenkov radiation (between wavelengths of 250 and 270 nm) produced by ionizing radiations of different kinetic energies passing through water. (The refractive index at 20°C for this band of wavelengths is ~1·37.) The symbols (○) are experimental data for the amount of photoreactivable damage induced by 50 kVp X-rays and ^{137}Cs γ-rays (this paper), ^{60}Co γ-rays (Vinicombe, Moss and Davies 1978), and 32 MeV electrons (Moss and Davies 1979), corrected for the contribution by direct excitation. The scale for PRS has been chosen so that the corrected value after 32 MeV electron irradiation corresponds with the plateau level of Cerenkov radiation. Other existing data, summarized in table 1, have not been included in this graph due to the lack of information about the conditions of irradiation, which might affect the production of Cerenkov radiation, and also about the optimization of conditions for photoreactivation.

and subsequent work have been reviewed by Jelley (1958). Briefly, the explanation for this phenomenon is that upon entering the medium the velocity of the particle is unaltered, except by subsequent ionization and radiative losses, but the electric field associated with the particle's charge and the magnetic field associated with the motion of this charge are propagated with a phase velocity of only c/n, where n is the refractive index of the medium. Expressing the velocity of the particle V as βc, where β = velocity of particle/velocity of light in vacuum, then the resultant electromagnetic radiation is cancelled by destructive interference in all directions if $\beta c < c/n$, i.e., if $\beta n < 1$. If, however, $\beta n > 1$ then there will be one direction in which constructive interference takes place, and electromagnetic radiation can be emitted. Hence, for a medium of given refractive index there is a minimum value for β for the emission of Cerenkov radiation. The value of β for a charged particle is related to its kinetic energy and its rest mass by the relationship.

$$m_0c^2 + T = \frac{m_0c^2}{\sqrt{1-\beta^2}} \tag{1}$$

solving for β

$$\beta = \frac{\sqrt{[(m_0c^2 + T)^2 - (m_0c^2)^2]}}{m_0c^2 + T} \tag{2}$$

where m_0 = rest mass of an electron

c = speed of light in vacuum

m_0c^2 will have a value of 511 keV

β may be solved by substitution $T=kE$ where kE is the kinetic energy of the electron in keV.

The theoretical explanation of Cerenkov radiation, developed by Frank and Tamm (1937), also provides for calculation of the energy emitted as Cerenkov radiation. The total energy, dT_{Cer}, radiated in a short element of the particle's path, ds, is

$$\left(\frac{dT}{ds}\right)_{Cer}=\frac{4\pi^2z^2e^2}{c^2}\int\left(1-\frac{1}{\beta^2n^2}\right)vdv \tag{3}$$

where ze is the charge on the moving particle and v is the frequency of the emitted radiation. The integration is to be carried out over all frequencies for which $\beta n>1$. Consider the Cerenkov radiation emitted between two frequencies, v_1 and v_2, as being composed of quanta whose average energy is

$$h\bar{v}=\frac{h(v_1+v_2)}{2} \tag{4}$$

then from equation (3) the average number of quanta emitted per centimetre is

$$\begin{aligned}\frac{1}{h\bar{v}}\left(\frac{dT}{ds}\right)_{Cer}&=\frac{4\pi^2z^2e^2}{hc^2}(v_2-v_1)\left(1-\frac{1}{\beta^2n^2}\right)\\&=\frac{2\pi z^2}{137}\left(\frac{1}{\lambda_2}-\frac{1}{\lambda_1}\right)\left(1-\frac{1}{\beta^2n^2}\right).\end{aligned} \tag{5}$$

where $\lambda=c/v$ the wavelength in a vacuum

and n = average refractive index over the wavelength interval from λ_2 to λ_1

If numerical substitution is made into this equation, for an electron ($z=-1$), for wavelengths between 250 and 270 nm, the mean refractive index of water at 20°C is 1·37, and β will vary for the energy of the radiation. For electrons of a few MeV or more, β is approximately equal to unity. For electrons produced by the radiations referred to in the present study, that is by 50 kVp X-rays, ^{137}Cs γ-rays, ^{60}Co γ-rays and by 32 MeV electrons, the respective values of β are 0·413, 0·900, 0·957, and 0·9988. The value for 50 kVp X-rays gives a value for βn of 0·56, which is less than unity and so no Cerenkov radiation will be emitted. The yields of Cerenkov radiation between 250 and 270 nm produced by ^{137}Cs γ-rays, ^{60}Co γ-rays and 32 MeV electrons will be 46·5, 56·8 and 63·3 photons per centimetre, respectively. Yields for other radiations may be taken from figure 6, which shows yields of 250 to 270 nm U.V. radiation produced as Cerenkov radiation as a function of ionizing radiation energy in water. For these conditions it can be seen that the threshold for Cerenkov radiation emission is 236 keV.

Acknowledgments

This work was supported by U.S. Public Health Service research grants CA-02896 and CA-06437, and programme project grant CA-10372 from the National Cancer Institute, DHEW.

On a étudiè le mécanisme de formation des lésions photo-réactivables de l'acide désoxyribo-nucléique (ADN) par radiation ionisante, chez *Escherichia coli* (*uvrA recA*), espèce déficiente en réparation obscure. En modifiant le rapport entre les lésions produites directement (par l'ionisation) et celles qui sont formées indirectement (par la radiation UV de Cerenkov), au moyen des rayons γ de ^{137}Cs, on a démontré que la majeure partie des lésions photoréactivables est produite par la radiation U.V. Cerenkov. La quantité de lésions photo-réactivables produites par les rayons X de 50 kVp, lesquels ne peuvent émettre de radiation Cerenkov, est semblable à la composante photo-réactivable des lésions produites par les rayons γ de ^{137}Cs et qui ne sont pas dues à la radiation Cerenkov. On suggère que le second mécanisme de formation des lésions photo-réactivables produites dans l'ADN par les radiations ionisantes est l'excitation directe de l'ADN. On examine le rôle possible de la radiation U.V. Cerenkov dans la mutagénèse par les radiations ionisantes.

Eine Untersuchung des Mechanismus der Bildung von photo-reaktivierbarem Schaden in DNS durch ionisierende Strahlen in einer dunkel-Reparatur defizienter Art von *Escherichia coli* (*uvrA recA*) wurde unternommen. Durch Ändern des Verhältnisses des direkt en Schadens (durch Ionization) zu dem des indirekten (mit Cerenkov ultra-violet (U.V.) Bestrahlung) mit ^{137}Cs γ-Strahlen erzeugten Schadens, wurde bewiesen, daß der Hauptanteil des photo-reaktivierbaren Schadens von der Cerenkov U.V. Bestrahlung erzeugt wurde. Der Anteil des photoreaktivierbaren Schadens, der von 50 kVp Roentgen Strahlen erzeugt wurde, und n ‘t durch Cerenkov Strahlen erzeugt werden kann, ist vergleichbar mit dem photor ‹tivierbaren Schaden, der von ^{137}Cs γ-Strahlen erzeugt wird und der nicht auf Cerenk ‹ Strahlen zurückzuführen ist. Es wurde vorgeschlagen, daß der zweite Mecha..smus zur Bildung von photoreaktivierbarem Schaden in DNS mit ionisierenden Strahlen eine direkte Anregung von DNS ist, Die mögliche Rolle der Cerenkov UV Strahlung in der Mutagenese ionisierender Strahlen wird besprochen.

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Note added in proof

The role of Cerenkov radiation in the inactivation of *E. coli* by ionizing radiation has been independently described by Myasnik, M. N. and Morozov, I. I., 1980, *Radiat. Res.*, **82,** 336, by Michaels, B. D., Harrop, H. A., and Held, K. D., (submitted to *Int. J. Radiat. Biol.*), and by Redpath, J. L., Zabilansky, E., Morgan, T., and Ward, J. F. (submitted to *Int. J. Radiat. Biol.*).