Proc. Nat. Acad. Sci. USA
Vol. 71, No. 9, pp. 3363–3366, September 1974

The Wavelengths in Sunlight Effective in Producing Skin Cancer: A Theoretical Analysis

(DNA damage/action spectra/ozone)

R. B. SETLOW*

Carcinogenesis Program and The University of Tennessee-Oak Ridge Graduate School of Biomedical Sciences, Biology Division, Oak Ridge National Laboratory, Oak Ridge, Tenn. 37830

Contributed by R. B. Setlow, June 21, 1974

ABSTRACT **DNA is taken as the target for skin cancer induced by ultraviolet light, and the known data on the sensitivity of DNA as a function of wavelength are summarized. The sun's spectrum at the surface of the earth and the DNA action spectrum are used to calculate the carcinogenic effectiveness as a function of wavelength. The most effective wavelengths at 30°N latitude are <305 nm, and a 1% change in atmospheric ozone results in a 2% change in the effective dose of ultraviolet light. Since both the basic biological and physical data are reasonably precise, the major requirement for a quantitative evaluation of the dose response relation for ultraviolet-induced skin cancer in man is better epidemiological data to compare with data from animal models.**

Human skin cancers, especially basal and squamous cell carcinomas, are closely associated with exposure to sunlight. [See reviews by Blum (1), Epstein (2), and the volume edited by Urbach (3).] Three lines of evidence indicate that the most effective wavelengths are below 320 nm. (*i*) In mice, wavelengths longer than 320 nm are ineffective in inducing skin cancer although a recent report indicates that exposure to the longer wavelengths may accentuate the effects of shorter ones (4). (*ii*) The effective wavelengths for erythema production are below 320 nm. Skin cancer and erythema arise in the same tissue, and individuals who sunburn easily have a higher probability than average of developing skin cancer (5, 6). (*iii*) Ultraviolet light (UV)-induced skin cancer probably arises from photochemical changes in DNA, and the shorter wavelengths are much more effective than the longer ones in damaging this polymer (see below). Interest in wavelength dependence arises not only because of the inherent interest in the problem but also for the practical reason of estimating the effects of this model environmental hazard. The hazard could change. For example, the exhausts from a fleet of supersonic transports might result in a decrease in stratospheric ozone and the attendant increase in UV fluence at the earth's surface could result in an increase in the incidence rate of skin cancer. This particular problem has been succinctly stated (7). If we are to evaluate quantitatively such hazards, we must have good animal and epidemiological data as well as a theoretical framework to handle such data.

UV-induced skin cancer deserves more careful epidemiological study because (*i*) we know more about UV-induced lesions in DNA than any other physicochemical insult to the genetic material (8); (*ii*) a number of carcinogenic chemicals mimic UV damage (9); (*iii*) the incidence rate of skin cancer in the United States is increasing (10) and there is no indication that this large-scale, human cancer-induction "experiment" will stop; and (*iv*) an analysis of the data may yield a quantitative relation between DNA damage and neoplastic transformation in man.

CARCINOGENIC ACTION SPECTRUM

The effect of sunlight in cancer induction should be calculated from the product of the sun's spectrum at the earth's surface and the action spectrum for cancer induction. Since the latter is not accessible experimentally, one approach has been to assume that it is similar to that for the production of erythema (11, 12). There is little theoretical justification for this approach. It is used because similar wavelength regions seem to be involved (see above). However, the shape of the action spectrum for erythema depends on the erythemal end-point used to determine it, and hence is not unique (13).

I believe, for the reasons that follow, that the appropriate action spectrum to use is one that coincides with the action spectrum for affecting DNA. Although we do not know the detailed molecular mechanism by which the biological effects of UV are produced, the evidence is overwhelming that changes in DNA, such as formation of pyrimidine dimers and other photochemical products, have important biological consequences. In many cases, it has been possible to correlate the production of specific photoproducts in DNA with biological changes such as the inactivation of biologically active DNAs, the killing of cells, and the induction of mutations (8). There have been good arguments made that many chemical carcinogens are mutagens (14) (and hence affect DNA), and a number of screening techniques for chemical carcinogens actually measure the mutagenic action of such chemicals (15). Since UV is mutagenic, we also expect it to be carcinogenic by virtue of its action on DNA.

Cells that are unable to repair DNA damage by excision repair are killed more readily by UV and by many chemicals, and repair-deficient microorganisms are more mutable per unit of UV fluence than repair-proficient ones (8). Individuals with the disease xeroderma pigmentosum are very susceptible to sunlight-induced skin cancer (16). Most of these individuals are also defective in excision repair (17, 18), and the severity of the disease is related to the magnitude of the defect (18). They are the analogs of UV-sensitive bacteria and we argue by analogy to bacteria that damage to DNA is the important photochemical damage to human cells, and thus make the

Abbreviation: UV, ultraviolet light.

* Present address: Biology Department, Brookhaven National Laboratory, Upton, N.Y. 11973.

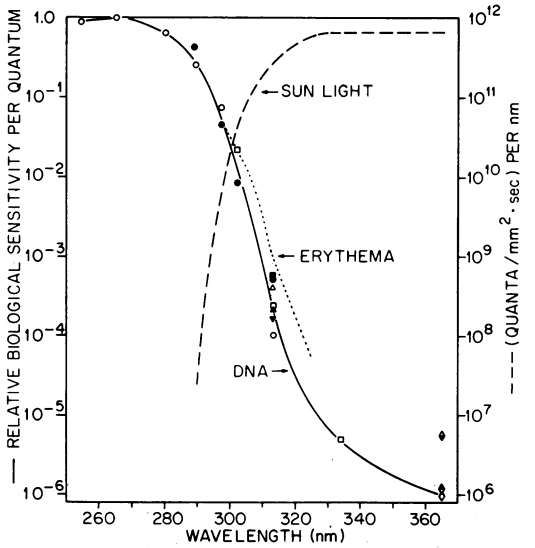

FIG. 1. The solid curve is an average action spectrum for affecting DNA. The data are normalized to the value 1.0 at 265 nm. The individual data points represent experiments on the following systems. Killing *Escherichia coli* (20, 21) (○, ■), mutations in *E. coli* (22) (△), killing T4 and T6 phages (23, 24) (●, □), photoproducts in DNA (25) (▲), endonuclease-sensitive sites in DNA (W. L. Carrier and R. B. Setlow, unpublished) (▼), O_2-dependent photoproducts in DNA (26) (◇), O_2-dependent killing of host cell reactivation deficient (Hcr^-) *E. coli* (26) (◆), O_2-dependent killing of Hcr^- recombination deficient *E. coli* (27) (◆). The broken curve represents the sun's spectrum at the earth's surface calculated by Green *et al.* (28) for Gainesville, Florida (12 noon, 8 Sept. 1972), 2.3 mm O_3, zenith angle 25°. The dotted curve is a recent erythemal action spectrum (29).

strong inference that damage to DNA that is not repaired effectively results in mutation and the transformation of cells.

THEORY AND DATA

We assume that DNA is the target for UV-induced carcinogenesis and calculate in a standard way (1, 19) the effects of sunlight and changes in sunlight on this target. At any given wavelength, λ, the probability, $p(\lambda)$, of a reaction taking place in DNA will depend upon the spectral irradiance, $I(\lambda)$, at the earth's surface, the transmission of the skin, $T(\lambda)$, and the cross section for the reaction $\sigma(\lambda)$.

$$p(\lambda)d\lambda = \sigma(\lambda)\cdot I(\lambda)\cdot T(\lambda)\cdot d\lambda. \quad [1]$$

The total probability of a change in DNA (the biological dose) arising from such reactions depends on the integral P,

$$P = \int p(\lambda)d\lambda. \quad [2]$$

But the function, f, relating skin cancer to P,

$$\text{skin cancer} = f(P), \quad [3]$$

is an unknown, complicated one that depends on the genetic background of the individual, the repetitive nature of the stimulus, the accumulated exposure, the time between exposures, the peak exposures, average exposure, the lifestyle of the individual, etc. All the terms involved in P are strong functions of λ.

Sunlight (I_o at the top of the atmosphere), entering at a zenith angle θ, is attenuated as a result of absorption by ozone and by scattering and absorption by other components, so

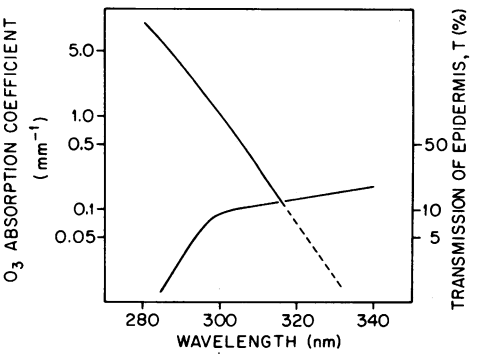

FIG. 2. Representative values for the transmission of white epidermis [from Kirby-Smith *et al.* (34)] and the linear absorption coefficient for ozone for amounts of ozone in millimeters at standard temperature and pressure (35).

that at the surface

$$I = I_o e^{-(\alpha c+\beta)\sec\theta}, \quad [4]$$

where α is the absorption coefficient per mm of ozone whose total vertical amount at standard temperature and pressure is c, and β represents all other sources of light attenuation. I_o, α, and β are functions of λ. It is apparent that

$$P = \int p d\lambda = \int (\sigma I T)d\lambda = \int (\sigma I_o e^{-(\alpha c+\beta)\sec\theta} T)d\lambda \quad [5]$$

and that

$$\frac{dP}{dc} = -\int(\alpha\sec\theta)(\sigma I T)d\lambda = -\int(\alpha\sec\theta)p d\lambda. \quad [6]$$

Hence both P and dP/dc depend critically on the value of α.

Fig. 1 shows action spectra, σ versus λ, for the effects of UV on a number of simple DNA-containing biological systems. (There exist many more DNA action spectra but most are at wavelengths below 300 nm, at which wavelength $I(\lambda)$ is very small.) The systems include inactivation of bacteria and bacterial viruses, mutagenesis in bacteria, formation of cyclobutane pyrimidine dimers in DNA, and the production of sites susceptible to the *in vitro* action of repair endonuclease. It is noteworthy that over a sensitivity range of 10^6 the data fit the same curve within a factor of about 2. Data on bacterial inactivation by 254–303 nm taken over 30 years ago (30, 31)—before there was a good appreciation of the various recovery factors influencing cell survival—agree with those in Fig. 1. It would be useful if there were equivalent action spectra on mammalian systems. At the longest wavelength shown, 365 nm, the biological effects depend on the presence of oxygen during irradiation, whereas there is no such dependence at shorter wavelengths. The high point at 365 nm is probably the result of enzymic photoreactivation during the long exposure times used (27). This repair system seems to be of little importance in placental mammals (32), although the enzyme has been detected in extracts of human leukocytes (33).

Fig. 2 shows the wavelength dependence of the parameters α and T. The irradiance at short wavelengths is critically dependent on the ozone concentration because of the high values of αc. Above 320 nm the absorption coefficient of the ozone is too small to influence the irradiance at the earth's surface and, as a result, the magnitude of any synergistic effects (4) between $\lambda > 320$ nm and short wavelengths will change only as a result of changes in the short-wavelength component.

TABLE 1. *Dosimetric parameters for different calculated sun spectra at the earth's surface*

Calculated by	ref. 28	ref. 37	ref. 38	ref. 37
c =	2.3 mm	3.2 mm	3.4 mm	3.2 mm
For θ =	25°	30°	30°	50°
A. For DNA				
$(1/P)(dP/dc)$	0.88 mm^{-1}	0.68 mm^{-1}	0.69 mm^{-1}	0.71 mm^{-1}
$(dP/P)/(dc/c)$	2.0	2.2	2.3	2.3
B. For erythema				
$(dP/P)/(dc/c)$	1.7	1.7	1.8	1.9

Such synergistic effects will not alter the biological dose, P, in Eq. **3** but would change the form of f. The precise data to use for the transmission through skin are not important. Recent measurements using somewhat improved techniques (36) give higher transmissions than shown in Fig. 2 but the shapes of the T versus λ curves are similar. Even if the transmission were 100%, there would be little effect on our conclusions (see below) because at short wavelengths the dominating effect is the fall-off of the sun's spectrum.

RESULTS AND DISCUSSION

Plots of Eqs. **5** and **6** are shown in Fig. 3. It is clear that wavelengths <305 nm in sunlight are more effective than those above, and that the exact position of the maximum of Eq. **1** is critically dependent on the shapes of both the action spectrum for DNA and on the sun's spectrum at the earth's surface. The area under the broken curve (Eq. **6**) has a value of 0.88 of that under the solid one. Hence $(1/P)(dP/dc)$ = 0.88 mm^{-1} and a change in ozone of 0.1 mm would result in an 8.8% change in the effective ultraviolet dose. Since, for the data in Fig. 3, a 0.1-mm change in ozone (2.3–2.2 mm) is a 4.3% change, $(dP/P) = 2.0\ (dc/c)$. If light were not attenuated in passing through skin, the change in effective dose resulting from a 0.1-mm decrease in ozone concentration would be 9.6%. Thus changes in transmission by white skin are of little importance compared to changes in the other quantities in Eqs. **5** and **6**. However, the very low transmission of black skin at the effective wavelengths (36) causes the integrals in Eqs. **5** and **6** to be an order of magnitude less than those for white skin, and hence the black population should be little affected by changes in solar UV.

At northerly latitudes, the thickness of the ozone layer is greater (see ref. 12) and, since the shorter wavelengths are attenuated more, the effective dose is smaller than indicated in Fig. 3. Moreover, the fractional change in effective dose per unit change in ozone, $(1/P)(dP/dc)$, is also smaller but the ratio of dP/P to dc/c is larger, as indicated in Table 1. Thus under most conditions the fractional change in biological dose is approximately twice the fractional change in ozone and increases with c and, because of the $\sec\theta$ term, with θ.

If equivalent analyses are carried out using the erythemal action spectrum† in Fig. 1, the maxima of $p(\lambda)$ are at longer wavelengths (see 11, 12, 19) and one obtains for the ratio of the relative change in dose to the relative change in ozone values that are about 20% less than for a DNA action spectrum (Table 1). Hence the use of erythemal action spectra for

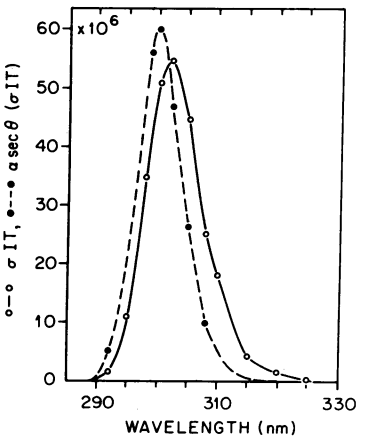

FIG. 3. The solid curve represents the effect of sunlight (of parameters given in Fig. 1) as a function of wavelength on DNA below a layer of skin. It is a plot of $p(\lambda)$ versus λ (Eq. **1**). The area under the curve is P (Eq. **2** or **5**). The broken curve shows the changes in the effect on DNA with changes in ozone. It is a plot of $\alpha\sec\theta\ p(\lambda)$ versus λ. The area under this curve is dP/dc (Eq. **6**). The ratio (dP/dc) to P is 0.88 mm^{-1}.

estimating changes in carcinogenic doses underestimates the effect. Similar calculations have been made by other authors using different erythemal action spectra (11, 12, 19) or earlier DNA action spectra (19). Although all the calculations indicate that the relative change in biological dose is 1.5 to 2.5 times the relative change in ozone, I believe the similarity between the action spectra for DNA damage and erythema production is fortuitous.

The relation between changes in UV dose (P in Eq. **3**) and changes in skin-cancer incidence depends critically on the form of the function f. If the assumptions in this analysis are correct, good epidemiological data‡ should permit the determination of the function for the exposures and exposure rates that man is subjected to, and hence also provide possible dose–response relations for chemical carcinogens. Dose–response relations are known at high UV levels for mice, but it is not clear how to extrapolate these data to man because of the different irradiance levels and because mouse cells in culture have a less efficient excision repair system than do normal human cells (40).

I appreciate the comments on the manuscript by M. L. Randolph and P. A. Swenson. This research was jointly sponsored by the National Cancer Institute and by the U.S. Atomic Energy Commission under contract with Union Carbide Corp.

1. Blum, H. F. (1959) *Carcinogenesis by Ultraviolet Light* (Princeton Univ. Press, Princeton, N.J.).
2. Epstein, J. H. (1970) in *Photophysiology*, ed. Giese, A. C. (Academic Press, New York), Vol. 5, pp. 235–273.
3. Urbach, F., ed. (1969) *The Biologic Effects of Ultraviolet Radiation* (Pergamon Press, New York).
4. Forbes, P. D. (1973) *Amer. Soc. Photobiol. Abstr.*, p. 136.
5. Silverstone, H. & Searle, J. H. A. (1970) *Brit. J. Cancer* **24**, 235–252.
6. Urbach, F., Rose, D. B. & Bonnem, M. (1972) in *Environment and Cancer* (Williams and Wilkins Co., Baltimore, Md.), pp. 355–371.

† Note that this action spectrum does not need a correction for skin transmission.

‡ The incidence data from a recent National Cancer Institute survey (10) were obtained from only four locations and these incidence figures, at a comparable latitude, are a factor of 2 to 3 higher than those from a recent survey in parts of Texas (39).

7. *Biological Impacts of Increased Intensities of Solar Ultraviolet Radiation* (1973) NAS-NAE, Washington.
8. Setlow, R. B. & Setlow, J. K. (1972) *Ann. Rev. Biophys. Bioeng.* **1**, 293–346.
9. Regan, J. D. & Setlow, R. B. (1973) in *Chemical Mutagens*, ed. Hollaender, A. (Plenum, New York), Vol. 3, pp. 151–170.
10. Scotto, J., Kopf, A. W. & Urbach, F. (1974) *Cancer*, in press.
11. Schulze, R. & Gräfe, K. (1969) in *The Biologic Effects of Ultraviolet Radiation*, ed. Urbach, F. (Pergamon Press, New York), pp. 359–373.
12. Cutchis, P. (1974) *Science* **184**, 13–19.
13. Johnson, B. E., Daniels, F., Jr. & Magnus, I. (1968) in *Photophysiology*, ed. Giese, A. C. (Academic Press, New York), Vol. 4, pp. 139–202.
14. Miller, E. C. & Miller, J. A. (1971) in *Chemical Mutagens*, ed. Hollaender, A. (Plenum Press, New York), pp. 83–119.
15. Ames, B. N., Durston, W. E., Yamasaki, E. & Lee, F. D. (1973) *Proc. Nat. Acad. Sci. USA* **70**, 2281–2285.
16. Cleaver, J. E. (1970) *J. Invest. Dermatol.* **54**, 181–195.
17. Cleaver, J. E. (1974) *Advan. Radiat. Biol.* **4**, 1–75.
18. Bootsma, D., Mulder, M. P., Pot, F. & Cohen, J. A. (1970) *Mutat. Res.* **9**, 507–516.
19. Chavaudra, J. & Latarjet, R. (1973) *C. R. H. Acad. Sci. Ser. D* **276**, 3481–3485.
20. Boyce, R. & Setlow, R. (1963) *Biochim. Biophys. Acta* **68**, 446–454.
21. Webb, R. B. & Lorenz, J. R. (1970) *Photochem. Photobiol.* **12**, 283–289.
22. Webb, R. B. & Malina, M. M. (1970) *Photochem. Photobiol.* **12**, 457–468.
23. Setlow, R. & Boyce, R. (1963) *Biochim. Biophys. Acta* **68**, 455–461.
24. Rapaport, S. A. (1964) *Virology* **22**, 125–130.
25. Setlow, R. B. & Carrier, W. L. (1966) *J. Mol. Biol.* **17**, 237–254.
26. Tyrrell, R. M. (1973) *Photochem. Photobiol.* **17**, 69–73.
27. Brown, M. S. & Webb, R. B. (1972) *Mutat. Res.* **15**, 348–352.
28. Green, A. E. S., Sawada, T. & Shettle, E. P. (1972) quoted by Grobecker, A. J. (1973) in *Proc. 2nd Conf. on Climatic Impact Assessment Program* (Nat. Tech. Info. Service, Springfield, Va.), pp. 2–12.
29. Berger, D., Urbach, F. & Davies, R. E. (1968) in *Proc. 13th Int. Dermatol. Cong.* (Springer-Verlag, New York, Heidelberg, Berlin), pp. 112–117.
30. Ehrismann, O. & Noethling, W. (1932) *Zeit. Hygiene Infectionskrank.* **113**, 597–628.
31. Gates, F. L. (1930) *J. Gen. Physiol.* **14**, 31–42.
32. Cook, J. S. (1970) in *Photophysiology*, ed. Giese, A. C. (Academic Press, New York), Vol. 5, pp. 191–233.
33. Sutherland, B. M. (1974) *Nature* **248**, 109–112.
34. Kirby-Smith, J. S., Blum, H. F. & Grady, H. G. (1942) *J. Nat. Cancer Inst.* **2**, 403–412.
35. Inn, E. C. Y. & Tanaka, T. (1959) in *Ozone Chemistry and Technology* (Amer. Chem. Soc., Washington), pp. 263–268.
36. Everett, M. A., Yeargers, E., Sayre, R. M. & Olson, R. L. (1966) *Photochem. Photobiol.* **5**, 533–542.
37. Green, A. E. S., Sawada, T. & Shettle, E. P. (1974) *Photochem. Photobiol.* **19**, 251–259.
38. Dave, J. V. & Furukawa, P. M. (1966) *Meteorol. Monographs* (Amer. Meteor. Soc., Boston), Vol. 7.
39. Macdonald, E. J. (1973) *Cancer of the Skin in Five Regions in Texas, 1962–1966*, Dept. of Epidemiol., Univ. Texas, Houston and M. D. Anderson Hosp. and Tumor Institute, unpublished.
40. Hart, R. W. & Setlow, R. B. (1974) *Proc. Nat. Acad. Sci. USA* **71**, 2169–2173.