

New Rules for AAAS-Newcomb Cleveland Prize

The AAAS-Newcomb Cleveland Prize, which previously honored research papers presented at AAAS annual meetings, will henceforth be awarded annually to the author of an outstanding paper published from September through August in the Reports section of *Science*. The first competition year under the new rules starts with the 3 September 1976 issue of *Science* and ends with that of 26 August 1977. The value of the prize has been raised from \$2000 to \$5000; the winner also receives a bronze medal.

To be eligible, a paper must be a first-time presentation (other than to a departmental seminar or colloquium) of previously unpublished results of the author's own research. Reference to pertinent earlier work by the author may be included to give perspective.

Throughout the year, readers are invited to nominate papers

appearing in the Reports section. Nominations must be typed, and the following information provided: the title of the paper, issue in which it is published, author's name, and a brief statement of justification for nomination. Nominations should be submitted to the AAAS-Newcomb Cleveland Prize, AAAS, 1515 Massachusetts Avenue, NW, Washington, D.C. 20005. Final selection will rest with a panel of scientists appointed by the Board of Directors.

The award will be presented at a session of the annual meeting at which the winner will be invited to present a scientific paper reviewing the field related to the prize-winning research. The review paper will subsequently be published in *Science*. In cases of multiple authorship, the prize will be divided equally between or among the authors; the senior author will be invited to speak at the annual meeting.

Reports

Irradiation of Bacterial Spores in Water: Three Classes of Oxygen-Dependent Damage

Abstract. *Studies of irradiated bacterial spores in aqueous suspension indicate that the sensitization of spores by oxygen can depend on three chemical processes. One of these processes involves reactions of hydroxyl radicals; the other two apparently do not.*

The response of dry *Bacillus megaterium* spores to ionizing radiation involves at least three major classes of lethal damage (1). Class I is oxygen-independent. The times necessary for the exchange of oxygen and nitrogen during the experiments (1) allowed two classes of oxygen-dependent damage to be recognized. One of these, class II, occurs only when O₂ is present during irradiation; additional damage, called class III, can occur if spores are held in dry oxygen after irradiation. Little is known of the chemical nature of class II damage. Class III, which may develop very slowly in dry spores if the O₂ concentration is low, is the consequence of a reaction between O₂ and radiation-induced radicals (2). In the very dry spore, class II and class III damage occur in a ratio of about 40 : 60, respectively.

The effects of spore water content on these three classes of radiation-induced damage (with 50-kv-peak x-rays) have also been described (3). As the amount of intracellular water increases, both

classes of oxygen-dependent damage decrease in magnitude and, in water or saturated water vapor (22 torr at 23°C), the two classes of oxygen-dependent damage could not be separated with the techniques that had allowed their recognition in the dried state. Class III has not been observed in spores in water suspension by introducing O₂ after anoxic irradiation, an operation that requires at least several seconds with the techniques used thus far. It was suggested, however, that both these classes of oxygen-dependent damage may still operate in the fully wet, irradiated spore (3).

Recently, Tallentire *et al.* (4) measured the sensitivity of *B. megaterium* spores (suspended in phosphate buffer) to ⁶⁰Co γ-rays over a wide range of O₂ concentrations. Their results supported the proposal that there are two oxygen-dependent, sensitizing processes in spores irradiated in suspension. Our analysis of their data shows that sensitization begins when the dissolved O₂ reaches a concentration of about 10⁻⁶M, and that

an initial plateau is reached with O₂ concentrations between about 1 × 10⁻⁵M and 3 × 10⁻⁵M. The level of the response at this first plateau accounts for about 27 percent of the full sensitization from oxygen. As [O₂] is raised further, the response to γ-rays increases very rapidly. At about 10⁻⁴M dissolved O₂ (approximately 7 percent O₂ in the equilibrating gas) γ-rays cause the maximum amount of oxygen-dependent damage to spores in suspension (4).

t-Butanol, which presumably acts only to scavenge hydroxyl radicals (·OH), does not reduce the radiation sensitivity of spores (to 50-kv-peak x-rays) in water under anoxic conditions or in air (5, 6). Thus, although ·OH reactions are clearly involved in some sensitization processes in spores [for a review, see (7)], there seems to be no ·OH component of damage demonstrable by adding *t*-butanol when irradiation is in nitrogen or air. However, with 0.8 percent O₂ (~ 10⁻⁵M), 0.1M *t*-butanol has a strong protective effect (8). This result prompted the systematic study with low concentrations of O₂ reported here.

Figure 1 shows the radiation sensitivity of spores irradiated in water, or in water with 0.1M *t*-butanol, in the presence of various concentrations of O₂. The results with different [O₂] in water alone (Fig. 1) are qualitatively similar to the γ-ray data (for spores suspended in buffer) of Tallentire *et al.* (4) that were discussed above; both sets of data indicate at least two oxygen-dependent damaging processes. An important quantitative difference, however, is apparent. With γ-rays, the smaller component of damage (about 27 percent of the total O₂ enhancement) occurs at low O₂ concentrations (4); with 50-kv-peak x-rays (Fig. 1) the level of the initial response plateau accounts for about 73 percent of the full oxygen effect. It is not clear why approxi-

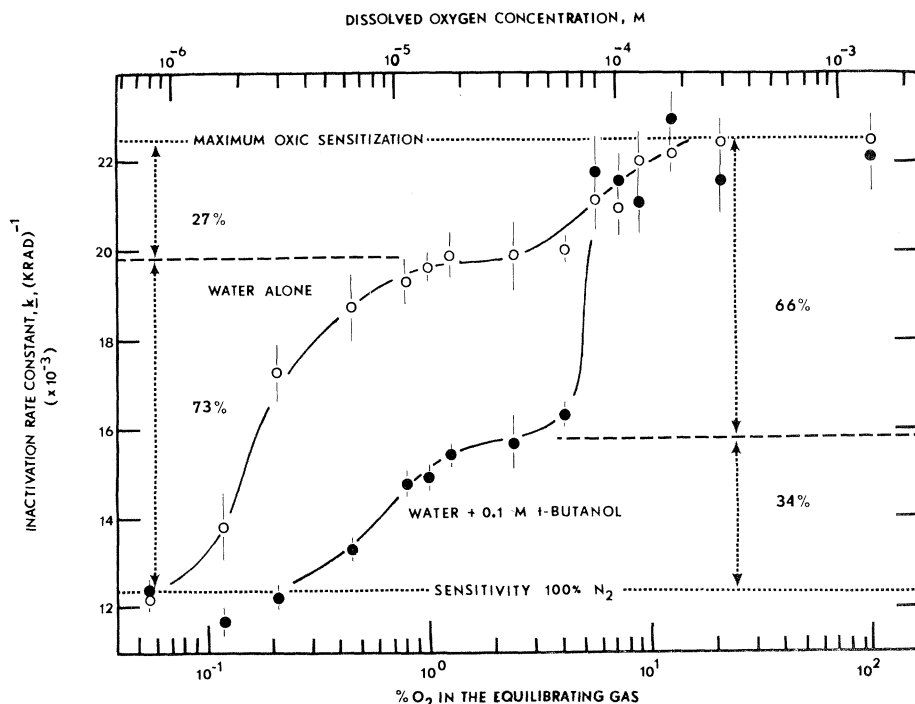


Fig. 1. The radiation sensitivity of *B. megaterium* (American Type Culture Collection No. 8245) spores as a function of O_2 concentration. Different $[O_2]$ were prepared by adding measured amounts of N_2 to a cylinder containing O_2 . The resulting $[O_2]$ was measured with a gas chromatograph (sensitivity ~ 20 parts per million). The suspending water or water with $0.1M$ *t*-butanol (having 2×10^7 spores per milliliter) was brought into equilibrium with the selected $[O_2]$ by flowing the gas over the liquid surface for 20 minutes prior to irradiation and during the entire irradiation period. The suspension was less than 5 mm deep, and it was stirred continuously to ensure equilibrium between the concentration of dissolved O_2 and the proportional O_2 concentration in the gas. The volume of the spore suspension was 1 ml. Irradiation was with 50-kv-peak x-rays at 14.05 krad/min, determined by ferrous sulfate dosimetry. The survival data were fitted to the equation $S = n [\exp(-kD)]$, where S is the fractional survival after dose D . The extrapolation number, n , and the inactivation rate constant, k , vary with the conditions during irradiation. [See (6) and (10) for complete procedural details.] The lower scale on the abscissa shows the percentage of O_2 (in N_2) in the equilibrating gas. The upper scale on the abscissa shows the dissolved O_2 concentration, a linear relationship between solubility and the percentage of O_2 in the gas phase being assumed (100 percent O_2 is equivalent to $1.41 \times 10^{-3}M$ dissolved O_2). The ordinate shows the inactivation rate constant, along with the standard errors, obtained at each $[O_2]$.

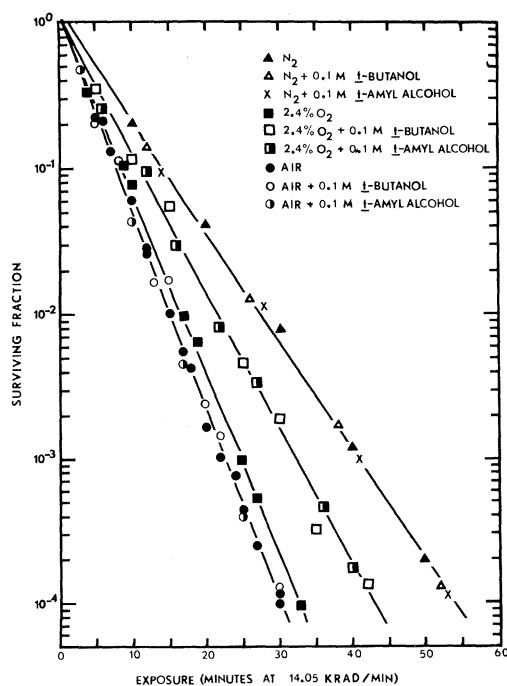


Fig. 2. Survival of *B. megaterium* spores under different conditions of irradiation. The identical effects of *t*-amyl alcohol and *t*-butanol in these experiments support the assumption that protection occurs because of hydroxyl radical scavenging.

mately the same low $[O_2]$ would have small sensitizing effects with 50-kv-peak x-rays but large effects with γ -rays. Also, it is not known whether *t*-butanol would protect the spores at low $[O_2]$ if γ -rays were used. Further work to answer these questions is required.

When $0.1M$ *t*-butanol is used to remove $\cdot OH$, protection is observed at low O_2 concentrations (Fig. 1). However, two components of oxygen-dependent damage not involving hydroxyl radicals are now apparent. These two damaging processes operate in a ratio of about 66 : 34, which is almost the same as the ratio for the two oxygen-dependent classes of damage in very dry spores (3). (The present experiments do not show, however, whether the oxygen-dependent damaging processes in wet and dry spores are mechanistically the same.)

t-Amyl alcohol [$CH_3CH_2(CH_2)_2COH$] is a structural analog of *t*-butanol. It reacts somewhat faster than *t*-butanol with $\cdot OH$ [$1.8 \times 10^9 M^{-1} \text{ sec}^{-1}$ and $5.2 \times 10^8 M^{-1} \text{ sec}^{-1}$ (9)], although this may be unimportant at high alcohol concentrations where $\cdot OH$ scavenging is assumed to be complete. Figure 2 shows that at $0.1M$ concentrations in water, the effects of these two alcohols are identical in nitrogen, in 2.4 percent O_2 , and in air. This similarity supports the assumption that *t*-butanol protects spores at low O_2 concentrations by removing an oxygen-dependent component of hydroxyl radical damage.

These results with bacterial spores irradiated in aqueous suspension indicate that, depending on the concentration of O_2 , oxygen-dependent damage can be separated into at least three chemical processes, one of which involves reactions of hydroxyl radicals and two that apparently do not. The exact chemical pathways through which O_2 functions as a radiation sensitizer are not known. Such information on mechanisms of oxygen-dependent sensitization and on general processes leading to radiation-induced cell death is urgently needed in radiation therapy. Truly anoxic regions in tumors are probably rare; it is more likely that gradients of O_2 concentrations are established across poorly vascularized tumors. If these results with spores apply to eukaryotic cells, when such a tumor is irradiated, both the amounts and kinds of damage may depend on the specific intracellular O_2 concentration.

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Comet West and the Scattering Function of Cometary Dust

Abstract. Observations of Comet West (1975n) at wavelengths from 0.5 to 18 microns and at a variety of scattering angles are used to infer the scattering phase function for the cometary dust. This function is strongly peaked in the forward direction. The form of the function indicates that the particles are dielectric grains with radii of approximately 1 micron. Abrupt increases in the intrinsic brightness of the coma (both in scattered sunlight and in thermal emission) are consistent with the projected times of comet fragmentation.

Observations of the comae, tails, and antitails of comets made at wavelengths λ from 0.5 to 20 μ have given new information about the constitution of the grains and limits on the sizes of grains

present in comets Bennett (1970II) (1), Kohoutek (1973f) (2), Bradfield (1974b) (3), and Kobayashi-Berger-Milon (1975h) (4). The discovery of the silicate feature at $\lambda = 10$ and 20 μ in the first three of

these comets demonstrated that silicate grains less than 5 μ in diameter were present. In addition, the antitail of Comet Kohoutek and the coma of Comet Kobayashi-Berger-Milon did not show the silicate signature, an indication that larger particles with diameters in excess of 20 μ were present, as predicted for the antitail of Comet Kohoutek by Sekanina (5).

Comet West afforded a unique opportunity to study another aspect of cometary dust which could be diagnostic. This is the light scattering or albedo of the dust as a function of the scattering angle. For the other comets mentioned the scattering angle θ observed was near 90°, but, because Comet West passed between the earth and the sun, forward scattering angles as small as 34° were observed.

Our measurements were made with a square diaphragm projecting a beam (20 by 20 arc seconds) centered on the coma, and the uniform background radiation from the sky was canceled by beam switching through an angle of 30 arc seconds. The same bolometer system was used at all wavelengths from 0.5 to 18 μ to guarantee that the comet colors were

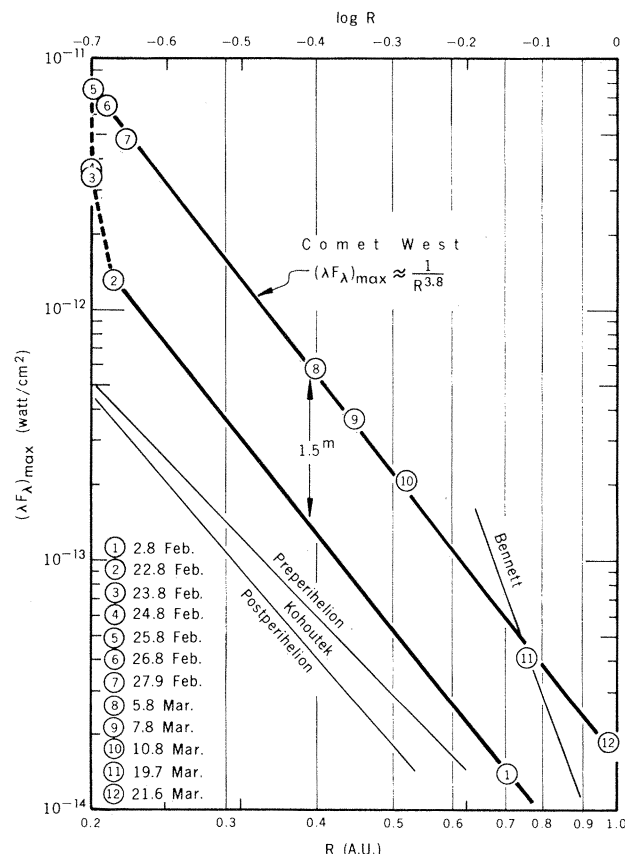
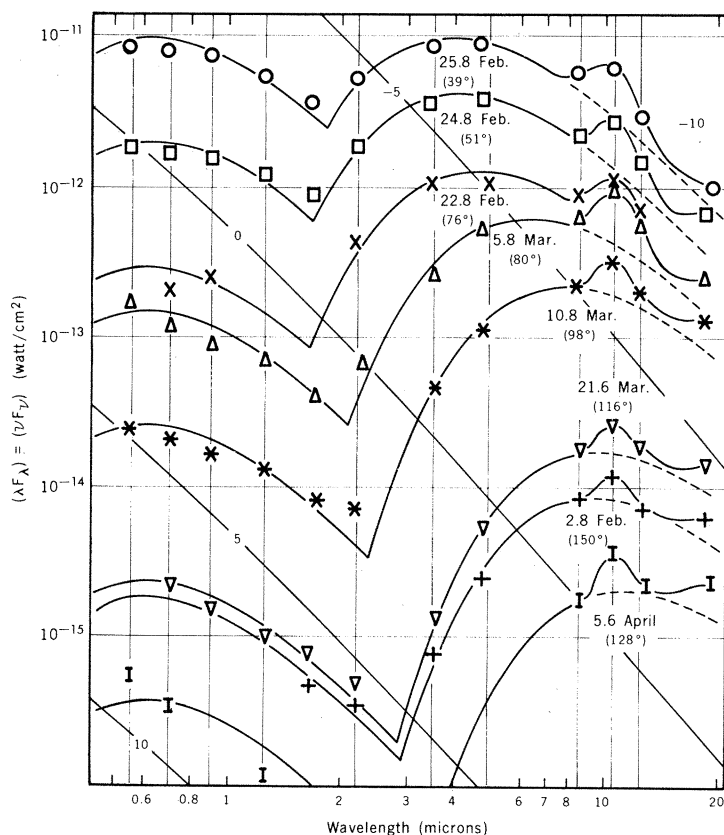


Fig. 1 (left). The energy distribution of Comet West (ν is frequency) between 2.8 February and 5.6 April. The silicate feature at 10 μ is always in evidence. The short-wavelength fluxes are due to scattered sunlight and the long-wavelength fluxes to thermal radiation. The forward scattering is shown by the increased brightness of the scattered light relative to the thermal emission at small scattering angles, for example, on 25.8 February. The scattering angles are given in parentheses below the dates. Fig. 2 (right). A plot of the thermal emission of the comet as a function of heliocentric distance. The effect of fragmentation is shown by the abrupt increase in brightness between observations 2 and 3 and observations 4 and 5.