

NOTES
PHOTOREVERSAL OF THE LENGTHENING EFFECT OF
ULTRAVIOLET RADIATIONS ON THE BACTERIAL
LAG PERIOD¹

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It is generally recognized that irradiation of bacterial cells with ultraviolet light of 2537 Å lengthens the lag period (Demerec and Latarjet: Cold Spring Harbor Symp. Quant. Biol., 11, 38, 1946). During the course of some preliminary experiments on another problem, it was necessary to determine the effects of various irradiation treatments on the lag period of *Escherichia coli*, strain B/r.

Cells from 24-hour aerated cultures, grown in either nutrient broth or a synthetic medium containing glucose, were washed, suspended in isotonic saline solution, and exposed to various doses of ultraviolet light from a 15-watt General Electric "germicidal" lamp. Immediately after this exposure they were diluted

TABLE 1
Effects of ultraviolet and reactivating light on the bacterial lag period

ULTRAVIOLET DOSE, ERGS/MM ²	LAG PERIOD (HR) IN NUTRIENT BROTH		LAG PERIOD (HR) ON NUTRIENT AGAR	
	Dark*	Light†	Dark*	Light†
0	1.6	1.7	1.5	—
500	3.4	—	3.8	1.8
1000	3.4	1.9	4.0	2.2

* Dark—Cells maintained in dark after exposure to ultraviolet.

† Light—Cells exposed to reactivating light for 1 hour after exposure to ultraviolet.

into nutrient broth at 37 C or diluted and spread on the surface of nutrient agar plates at 37 C. Aliquots suspended in saline were also exposed to the light of a 250-watt General Electric A-H5 lamp for one hour before inoculation into a nutrient medium, in order to determine the effect of reactivating light on the lag period. Viable cell counts were followed by means of plating at appropriate intervals. Cells growing on solid medium were washed from the agar surface, then plated. The lag periods obtained under these experimental conditions are listed in table 1.

For the purposes of this study, the lag period was arbitrarily defined as the time at which the logarithmic phase of the growth curve intercepted the starting concentration of cells when extended backward.

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Each value in table 1 represents the average of 3 to 8 independent observations. From these experiments it appears that the lag period is appreciably lengthened by as little as 500 ergs per mm² of ultraviolet, but that doubling this dose results in only a slight further increase for cells grown on solid medium and no increase for cells grown in nutrient broth. The most striking feature of these observations is the marked reduction in the lag period as the result of exposure of irradiated cells to reactivating light. The reactivation treatment appears to have no effect on cultures not previously exposed to ultraviolet light. None of the treatments used affected the generation time during the logarithmic growth period. In all cases this remained at 19 to 20 minutes.

VARIATION OF IRRADIATION EFFECTS ON MICROORGANISMS IN RELATION TO PHYSICAL CHANGES OF THEIR ENVIRONMENT

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It has been shown that slow cathode ray irradiation of bacteria *in vacuo* requires an unusually high dose compared to measurements made with soft X rays of comparable energies under normal air pressure (Moos, 1946, unpublished data; Moos, *Nucleonics*, **8**, 50, 1951; Hardung, *Helv. Phys. Acta*, **18**, 45, 1944; Haskins, *J. Applied Phys.*, **9**, 553, 1938). Experiments by C. G. Dunn *et al.* (*J. Applied Phys.*, **19**, 605, 1948) produced similar results with *Saccharomyces cerevisiae*. Hardung and Moos (*Experientia*, **5**, 155, 1949) pointed out that dehydration might be a cause of increased resistance of organisms irradiated *in vacuo* as compared with organisms irradiated in a liquid environment.

This hypothesis suggested that a more quantitative investigation might disclose some interesting information about the interaction of radiation with biological matter in different physical environments.

Pseudomonas aeruginosa and *Escherichia coli* were grown in horse meat infusion broth for 24 hours and 16 hours, respectively, at 36 C. For the experiments, 0.1 ml of these cultures was introduced into 2 ml "lusteroid" containers, then placed into pyrex test tubes and connected with an ordinary lyophilizing unit. After 2.5 hours of evacuation about 97.5 per cent of volatile material was removed from the broth leaving a residue of the bacteria, proteins, metabolic products, and salts; further drying did not change this result.

A 186 Kv X-ray unit served as radiation source. The beam was filtered by 0.25 mm copper and 0.55 mm aluminum. The samples were irradiated at a dose of approximately 250 rpm.

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