

Effects of Prolonged Irradiation of Mice with Fluorescent Light.*

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The carcinogenic property of ultraviolet irradiation is now well established. In fact, such radiant energy is one of the most effective of the many known carcinogens and the repeated exposure of mice¹⁻⁴ or rats⁵ to ultraviolet rays of the region 2,800-3,341 Å^{6,7} has been shown to result in a high incidence of skin cancer in several months. Furthermore, the production of neoplasms with ultraviolet light is not restricted to laboratory experimentation for a great majority of all the cancers of the skin in humans have also been attributed to prolonged exposures of excessive sunlight.^{1,8} It therefore becomes apparent that all sources of ultraviolet irradiation to which man is exposed for long periods of time should be investigated for possible carcinogenic activity. One type of lamp that deserves such attention is the fluorescent type that has become so widely used in recent years. The light from this source approximates that of sunlight closely, and analysis of the emission spectrum of the daylight fluorescent lamp reveals that a very small amount of the wavelengths 3125 and 3341 Å are also present.⁹

Although the intensity of the radiation of fluorescent lamps in the region below 3650 Å is negligible, it is possible that the total dose of these rays may approximate a carcinogenic amount in special cases where such lights are used repeatedly at short distances. The present report describes the effect of repeated and prolonged exposure of albino mice to the rays of a Westinghouse "Daylight" Mazda F fluorescent lamp.

Methods. Two groups of 48 strain C male albino mice 2 to 3 months old were housed in metal boxes on shavings and were fed a grain diet and water *ad libitum*. Six days each week they were put in screen cubicles³ and placed under 2 parallel Westinghouse "Daylight" fluorescent lamps at a distance of 4 inches. They were irradiated for 6 hours a day the first 8 months and 4 hours a day the last 4 months for a total of 12 months irradiation. In order to facilitate the penetration of the light, the ears of the mice in one group were painted with purified mineral oil (U.S.P.) daily before exposure.²

The intensity of the light was measured with a thermopile, and the amount of energy emitted by the 3125 Å band was calculated from data obtained from a curve of the emission spectrum.⁹ The intensity of the fluorescent lamp at the distance employed was 9.6×10^3 ergs/cm²/sec., so that the mice were exposed to a total of 57.6×10^9 erg/cm² over the one year period. The mice were weighed and examined closely at regular intervals for any pathological changes.

Results. According to the emission spectrum, radiant energy ranging from 3000 Å to 10,500 Å is emitted by the "Daylight" fluorescent lamps,⁹ and the 3125 Å band accounts for approximately 0.5% of the total intensity. Therefore, the amount of this wavelength received by the mice over a period of one year was 28×10^7 ergs/cm². This is slightly more than the minimum of 26.4×10^7

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¹ Rusch, H. P., and Baumann, C. A., *Am. J. Cancer*, 1939, **35**, 55.

² Rusch, H. P., Baumann, C. A., and Kline, B. E., *Proc. Soc. Exp. Biol. and Med.*, 1939, **42**, 508.

³ Rusch, H. P., Kline, B. E., and Baumann, C. A., *Arch. Path.*, 1941, **31**, 135.

⁴ Blum, H. F., Grady, H. G., and Kirby-Smith, J. S., *J. Nat. Cancer Inst.*, 1941, **2**, 259; 1942, **3**, 91.

⁵ Roffo, A. H., *Centralbl. f. allg. Path. u. path. Anat.*, 1935, **62**, 324.

⁶ Bain, J. A., and Rusch, H. P., *Cancer Res.*, 1943, **3**, 425.

⁷ Rusch, H. P., Kline, B. E., and Baumann, C. A., *Arch. Path.*, 1941, **31**, 135.

⁸ Blum, H. F., *J. Nat. Cancer Inst.*, 1940, **1**, 397.

⁹ Zimmerman, W. W., Report on Fluorescent Light, Lockheed Aircraft Corp., 1943, personal communication.

ergs/cm² reported to be carcinogenic when the mercury burner was the source of radiant energy.³ The comparisons are not valid, however, since the calculations involving the mercury vapor lamp included all the wavelengths from 2900-3341 Å. Furthermore, the radiations from the mercury lamp were given over a short period and were considerably more intense than those received from the fluorescent light. This is important since the intensity must not fall below a certain minimum level if cancer is to be induced.⁴

Actually there is no direct evidence that the 3125 Å band will induce cancer. The carcinogenic portion of the spectrum has been shown to lie above 2500 Å and below 3341 Å as nearly as can be determined with the aid of special filters,^{6,7} but with the methods now available it is not practical to measure the effects of individual bands within this range. However, since the 3125 Å band constitutes a fair portion of the total intensity between the region 2800-3341 Å emitted by the mercury vapor lamp, there is good reason to believe that it should be included in the carcinogenic range.

No tumors resulted from irradiation and no pathological changes of the eyes or other tissues were observed. The mice remained in relatively good health throughout the experiment and by 8 months 67% were still alive. During the last 4 months of the investigation, there was a slight weight loss and more deaths; but 32% of the mice were still alive when the experiment was terminated at

the end of a year. The group of mice that received the applications of mineral oil to the ears were not essentially different from those not getting this treatment.

The results of the present experiment do not rule out the 3125 Å band as lacking carcinogenic activity but they do demonstrate that such activity does not exist with the doses applied. These negative findings are all the more significant because the thin skin of albino mice is especially susceptible to the carcinogenic effect of ultraviolet irradiation. In fact, tumors did not even occur on the ears of mice that had been painted with mineral oil just prior to raying, a procedure that accelerates the genesis of tumors by permitting a better penetration of the rays.¹ Thus it is safe to assume that the radiations at 3125 and 3341 Å as emitted by the fluorescent "Daylight" lamps have no carcinogenic effect on albino mice.

Summary. Two groups of 48 albino strain C mice were placed directly under the light of two Westinghouse "Daylight" fluorescent lamps for periods of 4 or 6 hours a day, 6 days a week, for one year. The ears of one of the groups were painted with mineral oil before raying to facilitate the penetration of the rays into the skin. Although the total amount of radiant energy applied over the total period was considerable, the intensity of ultraviolet radiations in the 3125 Å band was very low. There was no evidence of erythema, or neoplastic changes of the skin, or of irritation of the cornea.

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Comparison of the Golden Hamster to the Guinea Pig Following Inoculations of Virulent Tubercle Bacilli.

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This is a brief note on comparative studies of the reactions of the golden hamster (*Cricetus auratus*) and the guinea pig to inoculations with virulent human tubercle bacilli. The work was carried out for the following reasons: (1)

The hamster has been used in the study of another Mycobacterial infection, *i.e.*, leprosy;¹ (2) It is reputedly susceptible to both the human and the bovine types of tubercle

¹ Burnet, E., *C. R. Acad. d. Sc.*, 1938, **207**, 690.

TABLE I.

A Quantitative Comparison (Expressed Numerically) of the Extensiveness of Disease Developing in Various Organs of the Hamster and Guinea Pig 120 Days After Individual Inoculations of 30,000 and 60,000 Virulent Human Type Tubercle Bacilli, H₃₇R_v.

Animals used	No. of animals in group	Dosage of micro-organisms	Average degree of tuberculosis				
			Lungs	Liver	Spleen	Lymph nodes	Total
Hamsters	10	30,000	0.7	1.0	0.9	1.5	4.1
Guinea pigs	10	30,000	2.5	2.6	3.3	3.4	11.8
Hamsters	10	60,000	0.9	0.9	1.1	1.9	4.8
Guinea pigs	10	60,000	3.0	3.0	4.0	4.0	14.0

bacilli;² (3) Its ability to thrive and breed in the laboratory environment³ makes it a desirable animal for experimental and diagnostic work.

Twenty guinea pigs and 20 hamsters* were used. They were divided into two groups, each group consisting of 10 hamsters and 10 guinea pigs. Each animal in the first group was inoculated subcutaneously with 30,000 virulent tubercle bacilli of human type, H₃₇R_v. Each member of the second group received similarly 60,000 bacilli of the same strain. The number of bacilli was calculated from the actual dry weight of the inocula according to a technic previously described.⁴ Twenty-four days after the infection each animal was skin tested with 0.1 cc of 5% and 10% Old Tuberculin. At the end of 48 hours, the guinea pigs showed areas of marked erythema, induration, and early necrosis. The skin of the hamsters exhibited no macroscopic reactions. At the end of 120 days the animals were sacrificed. The degree of macroscopic tuberculosis in each animal was evaluated by the method described by Steenken and Gardner.⁵ The involvement of the spleen, liver, lungs, and lymph nodes was expressed in values proportionate to the extent and severity of the disease. The maximum rating of 4 in any organ

was used to indicate widespread caseous disease. The maximum value of 16 for the animal as a whole signified advanced generalized tuberculosis.

The guinea pigs in both groups had extensive tuberculosis, but the hamsters showed relatively little disease. In the spleen of a few of the hamsters tubercle bacilli were demonstrated on smear. Microscopic sections of the lungs and spleen revealed only a few scattered small clusters of mononuclear cells.

Although others have reported that the hamster is susceptible to both the bovine and the human types of tubercle bacilli, their protocols do not bear out this impression. Balfour-Jones² infected 2 groups of hamsters intraperitoneally with 5 mg of "active" cultures of the human and the bovine types respectively. Two out of 3 hamsters in each group died in 5 weeks, and the third hamster showed marked evidence of infection when killed 8 weeks after inoculation. The author does not define his dosage of "5 mg" but if it was based upon wet weight the dose given to each animal was equivalent to *circum* 200,000,000 organisms;⁴ if he used a dry weight basis the dose for each animal was approximately 1,500,000,000 microorganisms. For an animal to live 5-8 weeks after such a tremendous inoculum speaks more for its resistance than for its susceptibility. This relative resistance appears ever more striking when, as shown here, guinea pigs develop extensive generalized disease within 6 weeks after an inoculation of only 30,000 virulent tubercle bacilli. Apropos to the hamsters' relative resistance to the bovine and human types, is the further observation that they show no macroscopic lesions after being given as much as 50 mg of avian tubercle bacilli.² Furthermore, Gardner

² Balfour-Jones, S. E. B., *Internat. J. Leprosy*, 1939, **7**, 77.

³ Laidlaw, Sir Patrick Playfair, *Internat. J. Leprosy*, 1939, **7**, 513.

* The hamsters were supplied to us through the courtesy of Merek & Company, Rahway, New Jersey.

⁴ Petroff, S. A., and Steenken, W., Jr., *Proc. Soc. Exp. Biol. and Med.*, 1927, **24**, 958.

⁵ Steenken, W., Jr., and Gardner, L. U., *Yale J. Biol. and Med.*, 1943, **15**, 393.

and Delahant⁶ have observed much less tuberculosis in hamsters than in guinea pigs after the inhalation of suspensions of tubercle bacilli (R₁ strain) in concentrations of 40-50 microorganisms per oil immersion field.

Conclusions. The hamster is highly re-

⁶ Gardner, L. U., and Delahant, A., personal communication.

sistant to doses of a virulent strain of the human type of tubercle bacilli that produces widespread tuberculosis in guinea pigs. Twenty-four days after infection 0.1 cc of 5% and 10% Old Tuberculin elicits no skin response. Therefore, in our opinion, we consider hamsters of little or no value in routine diagnostic procedures for tuberculosis.

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Relation of Plasma Level of Atabrine to Morphology and Motility of *Plasmodium vivax*.*

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(Introduced by Carl Ten Broeck.)

The effects of therapeutic doses of atabrine on the appearance of malaria parasites in stained films were noted soon after this drug had come into use. They have been described in detail for *Plasmodium vivax*,¹ for *P. falciparum*,² and for *P. lophurae*,³ a parasite of birds. The early changes consist essentially of the appearance of pale-staining wisps of cytoplasm stretching out from the cell, aggregation of the pigment in a well defined clump, and fragmentation of the chromatin. In these studies no attempt was made to correlate the morphological changes with the concentration of atabrine in the blood and plasma. It was, however, found for the monkey parasite *P. knowlesi* that the maximal blood level of atabrine occurred within one-half hour after intramuscular injection of the drug, and that the parasite count began to diminish shortly after the peak atabrine level had been reached.⁴

In the present note it is desired to present observations made with living *P. vivax*, and especially to call attention to the possibly reversible effect of otherwise lethal concentrations of atabrine if they act over only a short period of time. The work constituted a small portion of an extensive study of relapsing *vivax* malaria in the Southwest Pacific theater of the war.

Cases of *vivax* malaria were selected which, on admission to the hospital, showed a parasite count of about 500 or more parasites per 500 white blood cells, with most of the parasites in the late ameboid stage. Before treatment was begun, a sample of blood was taken and used for the preparation of a wet film and for the preparation of plasma for atabrine assay. The patient was then injected intramuscularly with 200 mg of atabrine dihydrochloride in water. Blood samples were taken at intervals after the injection and used for wet films and for atabrine determinations. The wet films were all examined on a warm stage within a few minutes after preparation. Plasma atabrine levels were determined by the double extraction method of Brodie and Udenfriend.⁵

The results for 7 patients are presented in Table I. Before treatment it was easy to find in the wet films large motile parasites with

* This work was done in 1943 in Australia at the 92nd Evacuation Hospital and the 6th Army Training Center. It is a pleasure to acknowledge the whole-hearted cooperation of the staffs of both of these organizations.

¹ James, S. P., *Trans. Roy. Soc. Trop. Med. and Hyg.*, 1934, **28**, 3.

² Hühne, W., *Deut. trop. Z.*, 1942, **46**, 385.

³ Hewitt, R. I., and Richardson, A. P., *J. Infect. Dis.*, 1943, **73**, 1.

⁴ Chopra, R. N., Ganguly, S. K., and Roy, A. C., *Indian Med. Gaz.*, 1936, **71**, 443.

⁵ Brodie, B. B., and Udenfriend, S., *J. Biol. Chem.*, 1943, **151**, 299.