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Advances in Brief

Ultraviolet Radiation A-induced Precursors of Cutaneous Melanoma in *Monodelphis domestica*[1]

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Abstract

Two groups of 30 dorsally shaved opossums (*Monodelphis domestica*) were exposed three times per week for 81 weeks to 250 J/m^2 of UV radiation from FS40 sunlamps (~150 J/m^2 of UV radiation B; UV-B), or to 2.5 × 10^4 J/m^2 of UV radiation A (UV-A) from filtered F40BLB fluorescent lamps (black lights). Animals were monitored for the appearance of nonmelanoma skin tumors (NMSTs) and melanocytic hyperplasia (MH). After 81 weeks of exposures, the prevalence of NMSTs was 71% and 4% for animals exposed to UV-B and UV-A, respectively. The difference between the treatment groups was statistically significant ($P < 0.001$). However, the prevalence of MH in the treatment groups, 31% for UV-B-exposed animals and 22% for UV-A-exposed animals, was not significantly different ($P > 0.05$). Thus, a dose of UV-A that was relatively ineffective in producing NMSTs, compared to UV-B, was as effective as UV-B in the induction of MH. If, as shown previously, MH is the precursor lesion for melanoma in this model, these results suggest that the action spectra for the induction of melanoma and NMSTs in the opossum are different.

Introduction

The incidence of malignant melanoma in the white population has increased at a rate of ~5% per year over the last 30 years (1). This dramatic increase in melanoma cannot be attributed to an increase in UV-B[3] (290–320 nm) striking the Earth's surface as a result of stratospheric ozone depletion. Although some decrease in stratospheric ozone has been measured, most notably over Antarctica, the amount of UV-B observed in populated areas actually decreased over the period from 1974 to 1985 (2). Garland *et al.* (3) proposed recently that increased use of sunscreens that protect efficiently against UV-B, but not UV-A (320–400 nm), may be responsible for the increased incidence of melanoma. Garland and coworkers reason that the use of sunscreens that are very effective in preventing sunburn results in increased exposure to UV-A during extended hours spent outdoors. This hypothesis would require that the action spectrum for melanoma induction deviate from the action spectrum for sunburn formation. (An action spectrum is the relative response of a system to different wavelengths of radiation.) If these action spectra were the same, sunscreens protective against sunburn would be equally protective against melanoma (4).

Setlow and coworkers (5) have determined an action spectrum for the induction of melanoma in a fish model. They reported that melanomas were induced readily at 365, 405, and probably 436 nm, which fall within the UV-A and visible radiation spectrum. The relative sensitivity for melanoma induction in fish at 365 nm was several orders of magnitude greater than the sensitivity for erythema induction in humans (6) or nonmelanoma skin cancer in mice (7). Setlow *et al.* (5) concluded that the general shape of the action spectrum for transforming fish melanocytes should be the same for transforming mammalian melanocytes. On the basis of this action spectrum, Setlow and colleagues (5) have calculated that 90–95% of human melanoma induction by natural sunlight may be caused by wavelengths >320 nm. Thus, stratospheric ozone depletion would have little effect on the melanoma incidence, because only those wavelengths below 320 nm would be affected. In addition, the use of conventional chemical sunscreens could be less effective in preventing melanoma than in preventing sunburn and may result in an increased exposure to melanoma-inducing wavelengths (8). Diffey (9) calculated that the use of a sunscreen preparation containing currently used UV-B and UV-A absorbers to achieve a sunburn protection factor of 8 would provide a protection factor of only 2–3 for the UV-A wavelengths. Therefore, extending the time an individual could stay outdoors without getting sunburned could result in enhanced exposure to UV-A.

We have used an opossum, *Monodelphis domestica,* as an animal model to determine the capacity of UV-A to induce MH, a melanoma precursor, in a mammal. *M. domestica* has been shown by our group (10) and by others (11–13) to be susceptible to the induction of melanoma upon exposure to UVR, primarily UV-B, alone.

Materials and Methods

Animals. Animals were from the breeding colonies maintained at the Lovelace Respiratory Research Institute (Albuquerque, NM) and at the South West Foundation for Biomedical Research (San Antonio, TX). Animals were housed individually in polypropylene cages with paper towels and cellulose fiber (Cellu-Dri, Shepard Specialty Products, Kalamazoo, MI) used for bedding. Water and dry fox food (Milk Specialties Products, New Holstein, WI) were available to the opossums *ad libitum*. Opossums were maintained at 24–26°C with a relative humidity of ~40%. Animals were maintained on a 12-h light/12-h dark cycle with red fluorescent lighting (F40R; General Electric) to avoid exposure to photoreactivating wavelengths. Protocols used in this study were approved by the Lovelace Institute's Institutional Animal Care and Use Committee.

Radiation Sources and Exposure Conditions. A UVR spectrum rich in UV-B was obtained from a bank of FS-40 sunlamps (National Biological Corp., Twinsburg, OH). The emission spectrum and dose rate of these lamps were monitored with a calibrated Optronics model 742 spectroradiometer (Optronics Laboratories, Orlando, FL). The emission spectrum is presented in Fig. 1. The dose rate at the back of the exposed animals was 2.5 W/m^2. UV-A was obtained from a bank of F40BLB black lights (General Electric) filtered with 6 mm thick plate glass. The dose rate (7.0 W/m^2) and emission spectrum (Fig. 1) were determined with the spectroradiometer. Prior to initial exposure, animals were anesthetized in a Halothane/0_2 atmosphere, and dorsal hair was removed with animal clippers (Model A2; Oster Corp.) followed by shaving with a Remington Microscreen shaver (Remington Products, Inc., Bridgeport, CT). Prior to each subsequent exposure, any regrowth of hair was removed with the electric shaver. Two groups of 30 age- and sex-matched opossums were exposed to 250 J/m^2 (100-s exposure time and approximately one-half of an average opossum minimal erythemal dose) from the FS40 sunlamps or to

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[3] The abbreviations used are: UV-B, UV radiation B; UV-A, UV radiation A; MH, melanocytic hyperplasia; NMST, nonmelanoma skin tumor; UVR, UV radiation.

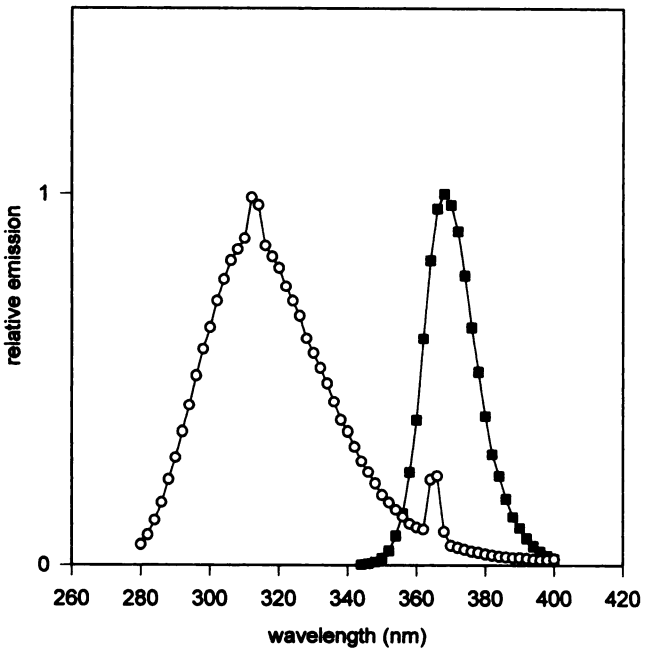

Fig. 1. Relative emission spectra from an FS40 sunlamp (○) and an F40BLB black light filtered with 6 mm thick plate glass (■). Emission spectra and dose rates were determined with a scanning spectroradiometer (model 72; Optronic Laboratories, Inc.). Dose rates were 2.5 W/m^2 and 7.0 W/m^2 for the FS40 sunlamps and filtered F40BLB lamps, respectively.

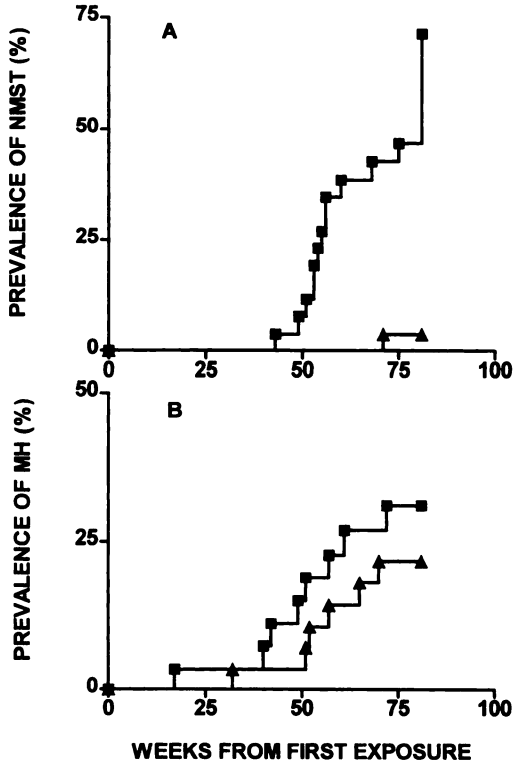

Fig. 2. Kaplan-Meier plots of the prevalence of NMSTs (*A*) and MH (*B*) as a function of weeks from first exposure to UV-B (■) or UV-A (▲). Treatment groups were exposed to 250 J/m^2 of UVR (~150 J/m^2 of UV-B) from the FS40 sunlamps or 2.5 × 10^4 J/m^2 of UV-A from the filtered F40BLB lamps three times per week for 81 weeks.

2.5 × 10^4 J/m^2 (1-h exposure) of UV-A from a bank of plate glass-filtered black lights three times per week (Monday, Wednesday, and Friday) for 81 weeks. Exposures were started when the animals were approximately 4 months of age. Opossums were monitored for the appearance of areas of focal MH and NMSTs.

Data Analysis. The appearance of MH and NMSTs was plotted as prevalence as a function of time from initial exposure (14). A log-rank test equivalent to the Mantel-Haenszel test was used to determine the statistical significance of difference in the time to appearance of NMSTs or MH between the two treatment groups (15).

Results and Discussion

Plotted in Fig. 2*A* is the prevalence of NMSTs as a function of time from first exposure for the two treatment groups. The time to appearance of NMSTs was significantly ($P < 0.001$) shorter in those animals exposed to UV-B as compared to animals exposed to UV-A. Only one animal in the UV-A group was scored as having a NMST. (This daily UV-A dose is equivalent to a 10–15-min exposure to midday sun in Albuquerque, NM,[4] and is ~10% of a UV-A dose, delivered 12 h/day 7 days/week, that resulted in a median NMST induction time of 265 days in mice; Ref. 16). In contrast, although the induction of MH in the UV-A exposure group was somewhat delayed in comparison to UV-B-exposed animals, the difference in time to appearance of MH was not statistically significant ($P = 0.36$; Fig. 2*B*). Thus, if these areas of MH are, as previously reported, precursor lesions for the formation of cutaneous melanoma, both treatments were equal in their capacity to induce melanoma. The responses observed in this study with UV-B-exposed animals are similar to those reported previously for NMST (17) and MH (10) formation in opossums.

Although limited in scope, these data suggest the following: (*a*) UV-B radiation is equally effective in the induction of both NMSTs and MH. The different slopes of the prevalence curves as a function of time for the formation of NMSTs and MH in UV-B-exposed animals may reflect differences in the underlying mechanisms of induction of these two end points; and (*b*) at a dose 100 times greater than the UV-B dose, UV-A is more effective at inducing MH than at inducing NMSTs. Thus, these data imply that in the UV-A region of the UVR spectrum, the relative efficiencies for the induction of melanoma precursors in the opossum are significantly greater than the efficiencies for the induction of NMSTs. Measuring the magnitude of this difference will require additional studies to generate dose-response curves using various wavebands within the UV-A spectrum.

[4] Unpublished data.

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