
Ultraviolet A Radiation Suppresses an Established Immune Response: Implications for Sunscreen Design

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The ultraviolet radiation present in sunlight is the primary cause of nonmelanoma skin cancer and has been implicated in the development of cutaneous malignant melanoma. In addition, ultraviolet is immune suppressive and the suppression induced by ultraviolet radiation has been identified as a risk factor for skin cancer induction. Ultraviolet also suppresses the immune response to infectious agents. In most experimental models, ultraviolet is applied to immunologically naive animals prior to immunization. Of equal concern, however, is the ability of sunlight to suppress established immune reactions, such as the recall reaction in humans, which protects against microbial infections. Here we demonstrate that solar-simulated ultraviolet radiation, applied after immunization, suppresses immunologic memory and the elicitation of delayed-type hypersensitivity. Further, we found that wavelengths in the ultraviolet A region of the solar spectrum were critical for inducing immune suppression. Ultraviolet A

(320–400 nm) radiation was as effective as solar-simulated ultraviolet A + B (290–400 nm) in suppressing the elicitation of an established immune response. Irradiation with ultraviolet AI (340–400 nm) had no effect. Supporting a critical role for ultraviolet A in ultraviolet-induced immune suppression was the observation that applying a sunscreen that contained an ultraviolet B only filter had no protective effect, whereas, a sunscreen containing both ultraviolet A and ultraviolet B filters totally blocked ultraviolet-induced immune suppression. These data suggest that sunlight may depress the protective effect of prior vaccination. In addition, the observation that ultraviolet A is immunosuppressive indicates the need for ultraviolet A protection when designing sun protection strategies. *Key words: delayed type hypersensitivity/immune suppression/skin cancer/solar-simulated ultraviolet radiation. J Invest Dermatol 117:1193–1199, 2001*

The ultraviolet (UV) radiation present in sunlight is a complete carcinogen and the primary cause of nonmelanoma skin cancer (Urbach, 1997). UV radiation has also been implicated in the induction of cutaneous malignant melanoma (Setlow *et al*, 1993; Dooley, 1994). In addition to skin cancer formation, UV exposure causes cataract formation, sunburn, premature aging of the skin, activation of latent viruses, and immune suppression (reviewed by Ullrich, 2000). The immune suppressive effects of UV radiation contribute to skin cancer development by depressing cell-mediated immune reactions that normally serve to destroy the developing skin tumors. Epidemiologic studies with immune-suppressed renal transplant patients (Penn, 1984), experiments with laboratory mice (Fisher and Kripke, 1982), and immunologic studies with skin cancer patients (Yoshikawa *et al*, 1990), support the hypothesis that

the immune suppression induced by UV exposure is a major risk factor for skin cancer induction.

In addition, UV exposure suppresses immune responses to infectious organisms (Jeevan *et al*, 1992). In the majority of studies documenting UV-induced suppression of the immune response to microbial and viral agents, the UV was administered to naive animals prior to immunization (i.e., suppressing the induction of immunity). Of equal concern, however, is the ability of UV exposure to suppress established immune responses. Perhaps the most important medical advance of the twentieth century was the reduction, and in some cases the eradication (i.e., smallpox) of certain microbial infections through the widespread use of childhood vaccinations. Because UV radiation can suppress the elicitation of certain immune responses (Denkins *et al*, 1989; Magee *et al*, 1989; Moyal *et al*, 1997), sunlight exposure may compromise the ability of prior vaccination to control infectious disease. Unfortunately, little is known concerning the underlying immunologic mechanism(s) of UV-induced suppression of an established immune response.

Furthermore, little is known about the basic photobiologic mechanisms involved. Ambient UV radiation is divided into two major regions: (i) UVB (290–320 nm), which comprises less than 5% of the UV that reaches the biosphere, and (ii) UVA (320–400), which comprises at least 95% of the remaining UV radiation. Although the role of UVB in inducing skin cancer and immune

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Abbreviations: UVB, UV radiation between 290 and 320 nm; UVA II, UV radiation between 320 and 340 nm; UVA I, UV radiation between 340 and 400 nm.

suppression is well known, the contribution of UVA to the deleterious effects of sunlight are not as well defined. The action spectrum for melanoma induction in *Xiphophorus* implicates UVA in melanoma induction (Setlow *et al*, 1993). Whether this action spectrum also applies to melanoma induction in humans remains to be seen. Similarly, the scientific literature concerning the role of UVA in UV-induced immune suppression is contradictory. Examples of UVA suppressing the induction of immunity (Hersey *et al*, 1983, 1988; Bestak and Halliday, 1996; Damian *et al*, 1997; LeVee *et al*, 1997) are as numerous as examples where UVA fails to have an effect (Sjovall and Christensen, 1986; Skov *et al*, 1997, 1998; Reeve *et al*, 1998, 1999). Moreover, recent reports from Reeve and colleagues suggest that prior exposure to UVA radiation can protect against the immunosuppressive effects of UVB (Reeve *et al*, 1998; Shen *et al*, 1999).

Determining the relative role of UVA in immune suppression may have broad implications besides being of interest to photo-immunologists. Oncologists and dermatologists have been promoting a campaign of 'safe sun exposure' to combat the dramatic rise in skin cancer incidence. Using sunscreens is an essential part of this campaign. Until very recently, most sunscreens available in the United States absorbed wavelengths in the UVB region of the solar spectrum, with little or no absorption in the UVA region. This appears to be sufficient to protect against sunlight-induced *p53* mutations and nonmelanoma skin cancer induction (Ananthaswamy *et al*, 1997, 1999); however, the action spectrum for melanoma induction in fish, and data suggesting that UVA induces immune suppression raise concerns about the ability of most sunscreens to provide adequate UVA protection. On the other hand, the provocative data presented by Reeve *et al*, 1998, questions whether it is even desirable to add UVA filters to sunscreens. It is extremely important, therefore, to clarify the role of solar UVA in immune suppression and other forms of photodamage.

MATERIALS AND METHODS

Animals Specific pathogen-free female C3H/HeNcr (MTV⁻) mice were obtained from the National Cancer Institute Frederick Cancer Research Facility Animal Production Area (Frederick, MD). The animals were maintained in facilities approved by the Association for Assessment and Accreditation of Laboratory Animal Care International, in accordance with current regulations and standards of the National Institutes of Health. All animal procedures were reviewed and approved by the Institutional Animal Care and Use Committee. Within each experiment all the mice were age matched. The mice were 8–10 wk old at the start of each experiment.

Radiation source A 1000 W Xenon UV solar simulator equipped with a Schott WG-320 atmospheric attenuation filter (1 mm thick), a visible/infrared bandpass blocking filter (Schott UG-11; 1 mm thick) and a dichroic mirror to further reduce visible and infrared energy (Oriol Corp. Stratford, CT) was used to provide solar-simulated UV radiation (Fig 1; WG-320). Replacing the WG-320 filter with a 3 mm thick WG-335 filter resulted in a UVA source deficient in UVB (Fig 1; WG-335). Replacing the WG-320 filter with a 2 mm thick WG-360 filter resulted in a UV source devoid of UVB and deficient in UVA II (Fig 1; WG-360). The WG-320 and WG-335 filters were purchased from Oriol Corp. The Schott WG-360 filter was generously provided to us by Dr F. J. Christiaens, L'Oréal, Clichy, France. The intensity and the spectral output of the solar simulator was measured with an Optronics model OL 754 scanning spectrophotometer interfaced to an Acer model 330 notebook computer (Optronics Laboratories, Orlando, FL). During irradiation, the mice were held individually in a specially constructed Plexiglas container with a quartz glass top, to prevent cage mates from climbing on top of each other and interfering with the UV dose applied. Spectrophotometer readings were taken through the quartz glass top. During the irradiation period (15–90 min in duration) the mice were conscious and had a full range of movement.

Suppression of immunologic memory and established immune responses by UV radiation Female C3H/HeN mice were immunized by subcutaneous injection of 10⁷ formalin-fixed *Candida albicans* into each flank (Moodycliffe *et al*, 2000). Seven days later the mice were

boosted with an identical dose of *C. albicans* into each flank. Thirty days later the mice were shaved and exposed to solar-simulated UV radiation as described previously (Ananthaswamy *et al*, 1997). The next day each hind footpad was measured with an engineer's micrometer (Mitutoyo, Tokyo, Japan) and then challenged by intrafootpad injection of 50 μ l of *Candida* antigen (Alerchek Inc, Portland, ME). Eighteen to 24 h later the thickness of each foot was re-measured and the mean footpad thickness for each mouse was calculated (left foot + right foot \div 2). Generally, there were five mice per group, the mean footpad thickness for the group \pm SEM was calculated. The background footpad swelling (negative control) was determined in a group of mice that were not immunized but were challenged. The specific footpad swelling response was calculated by subtracting the background response observed in the negative controls from the mean footpad swelling found in mice that were immunized and challenged. Each experiment was repeated at least three times. Statistical differences between the controls and experimental groups were determined by using the two-tailed Student's t test, where $p < 0.05$ was considered significant (Prism Statistical Software, GraphPad Inc, San Diego, CA). Percentage immune suppression was determined by the following formula: % immune suppression = $(1 - \{\text{specific footpad swelling of the UV-irradiated mice} \div \text{specific footpad swelling of the positive control}\}) \times 100$.

To determine if UV radiation suppresses the elicitation of delayed type hypersensitivity (DTH) mice were immunized by the subcutaneous injection of 10⁷ formalin-fixed *C. albicans*, as described above. Nine days later the immunized mice were shaved and their dorsal skin was exposed to solar-simulated UV radiation. The next day each hind footpad was measured and challenged by intrafootpad injection of 50 μ l of *Candida* antigen. Footpad swelling was read 18–24 h later.

Sunscreen formulations Two SPF 15 sunscreens were used in these studies, a predominantly UVB absorber (P532) and a sunscreen that equally absorbs both UVA and UVB radiation (P533). The UV absorption spectra of the sunscreens and their chemical composition were reported previously (Ananthaswamy *et al*, 1999). The vehicle, an oil-in-water emulsion, was the same for both sunscreen preparations. The sunscreens and the vehicle were applied to the shaved dorsal skin of the mice (100 μ l per mouse: \approx 2 mg per cm²) 30 min prior to irradiation, as described previously (Ananthaswamy *et al*, 1999).

RESULTS

Solar-simulated UV radiation suppresses established immune responses First we measured the effect of UV radiation on immunologic memory. Formalin-fixed *C. albicans* was injected on days 0 and 7 as described above. Thirty days after the second immunization, the mice were exposed to 120 kJ per m² of UVA + B radiation (WG 320-filtered solar simulator). The mice were challenged with antigen on the next day, and DTH was measured 18 h later. As shown in Fig 2A, exposing mice to UV radiation 30 d postimmunization, significantly suppressed immunologic memory (60% immune suppression; $p < 0.05$ vs the positive control).

Next we measured the effect that UV radiation had on the elicitation of DTH. In these experiments, the mice were immunized with *C. albicans* on day 0, exposed to UV on day 9, challenged with antigen on day 10. DTH was read 18 h later. Results obtained when mice were exposed to UVA + B radiation (WG-320 filtered solar simulator) are shown in Fig 2(B). Significant immune suppression ($p < 0.01$) was observed with doses equal to, or in excess of 60 kJ per m² of UV radiation (290–400 nm). Similar results were obtained when mice were irradiated with a solar simulator equipped with a Schott WG-335 filter that effectively removes UVB radiation (Fig 2C). Compared with the positive control, significant immune suppression ($p < 0.01$) was observed with doses equal to, or in excess of 60 kJ per m² of UVA radiation (320–400 nm). These findings indicate that UV radiation suppresses established immune reactions and that the critical wavelengths reside within the UVA region of the solar spectrum.

For the sake of comparison the Δ footpad swelling data presented in Fig 2(B, C) were converted into percentage immune suppression and plotted vs the dose of UV applied. These data are found in Fig 3. There was a highly significant correlation ($p < 0.001$; Pearson correlation test) between the suppression induced by the

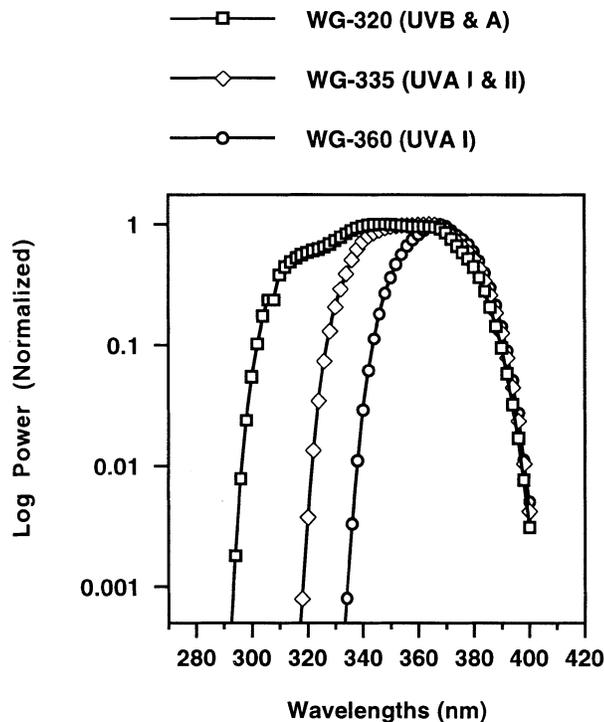


Figure 1. Spectral output of the light source used in this study. The Xenon UV source was equipped with: a WG 320/1 mm filter to provide UVB + UVA radiation (WG 320); a WG 335/3 mm filter to provide UVA I and II radiation (WG 335); or a WG 360/2 mm filter to provide UVA I radiation (WG 360).

WG 320-filtered solar simulator and the WG 335-filtered solar simulator. In addition, the amount of UV radiation required to generate 50% immune suppression was identical when a combination of UVB and UVA (WG 320), or UVA deficient in UVB (WG 335), was used to suppress the elicitation of DTH (80 kJ per m² UVA + B; $r^2 = 0.976$ vs 82 kJ per m² UVA; $r^2 = 0.983$).

In the next series of experiments mice were exposed to UV radiation from a solar simulator equipped with a WG-360 filter (Table I). This filter removes all the UVB radiation and most of the UVA II (320–340) radiation. Unlike the results presented above, irradiation with a source devoid of UVB, deficient in UVA II and rich in UVA I radiation, failed to suppress the elicitation of DTH. These data (Fig 2B,C and Table I) indicate that wavelengths in the UVA II region of the solar spectrum (320–340 nm) are responsible for suppressing established immune reactions.

These data imply that the UVA in solar-simulated radiation is solely responsible for suppressing the elicitation of DTH to the fungal antigen, *C. albicans*. We decided to test this hypothesis further by measuring the protective effect of two sunscreens in our experiments. The first P533, absorbs both UVA and UVB radiation, whereas the second, P532 absorbs wavelengths primarily in the UVB region of the solar spectrum. Both sunscreens had a sun protection factor of 15; as measured in humans (Ananthaswamy *et al*, 1999) and hairless mice (Anny Fourtanier, L'Oreal, personal communication). Data from this experiment are presented in Fig 4. In Fig 4(A), the sunscreen that absorbed both UVA and UVB (P533) was applied. The positive control in this experiment consisted of a group of mice that were immunized with *C. albicans* on day 0, painted with the vehicle on day 9, and then challenged with antigen on day 10. The DTH reaction was measured 18–24 h later. The negative control mice were handled in an identical manner, but they were not immunized. Applying the sunscreen to another group of positive and negative control mice demonstrated that the sunscreen by itself did not have any effect on the immune

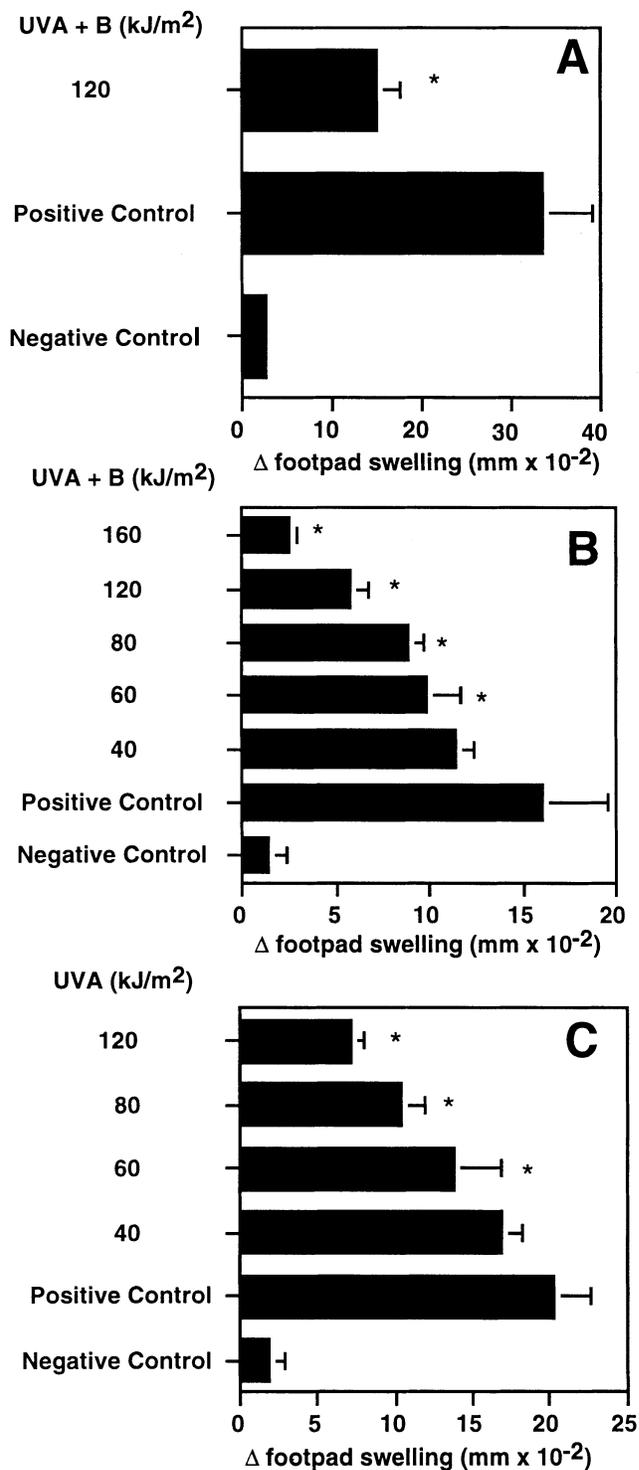


Figure 2. Solar-simulated UV radiation suppresses established immune responses. In (A), the effect of UV radiation on immunologic memory was measured. Mice were first immunized on day 0, again on day 7 and then exposed to UV radiation (WG 320) 30 d after the second immunization. In (B) and (C), the effect of UV radiation on the elicitation of DTH was measured. Mice were immunized and 9 d later treated with different amounts of UVA + B (B: WG 320) or UVA (C: WG 335) radiation. *Significant difference ($p < 0.01$; two-tailed Student's t test, $n = 5$) vs the positive control.

response ($p > 0.05$, Student's t test). We observed significant ($p < 0.01$) and substantial (60–80%) immune suppression when the vehicle-treated mice were irradiated with doses equal to, or greater

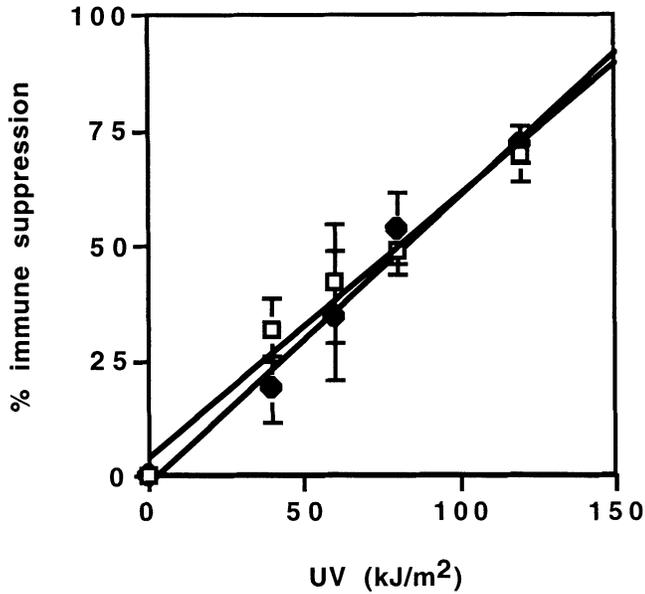


Figure 3. Dose-response curves for suppressing the elicitation of DTH to *C. albicans*. Mice were exposed to different doses of UV radiation supplied by the WG 320-filtered solar simulator (□) or the WG 335-filtered solar simulator (◆). The footpad swelling for each group was converted into percentage immune suppression and plotted vs the dose of UV applied. The regression curves shown here were generated using pooled data from three independent experiments.

Table I. WG 360-filtered SSR (UVA I) does not suppress the elicitation of DTH to *C. albicans*

Treatment ^a	Δ footpad swelling (mm × 10 ⁻² ± SD)	Specific swelling ^b	% suppression ^c	p ^d
Negative control	1.0 ± 0.9	–	–	–
Positive control	12.1 ± 1.6	11.1	0	–
WG 320 80 kJ per m ²	3.9 ± 2.0	2.9	73.8	0.0006
WG 360 80 kJ per m ²	11.8 ± 2.5	10.8	2.7	0.847
WG 360 160 kJ per m ²	12.0 ± 1.9	11	0.9	0.413

^aMice were immunized with *C. albicans*. Nine days later their shaved dorsal skin was exposed to different doses of UV radiation from a WG-320 or WG 360-filtered SSR. Ten days postimmunization they were challenged with *C. albicans* antigen and footpad swelling was read 24 h later. Negative control refers to mice that were not immunized but were challenged; positive control refers to mice that were immunized and challenged.

^bΔfootpad swelling of experimental groups minus the background swelling found in the negative control.

^c% immune suppression = (1 - {specific footpad swelling of the UV-irradiated group ÷ specific footpad swelling of the positive control} × 100).

^dp-values determined by two-tailed Student's t test vs the positive control.

than, 40 kJ per m². When, however, the mice were treated with the UVA + B absorbing sunscreen, no immune suppression was observed. In all cases, the response generated in the UV + P533 sunscreen-treated mice was not significantly different from the positive control.

An entirely different situation was observed when the mice were treated with the UVB only absorbing sunscreen (Fig 4B). Similar to the experiment described above, application of sunscreen P532 had no effect on the generation of the immune response (vehicle + positive control vs P523 + positive control). Here also, we observed significant (p < 0.01) immune suppression when the mice

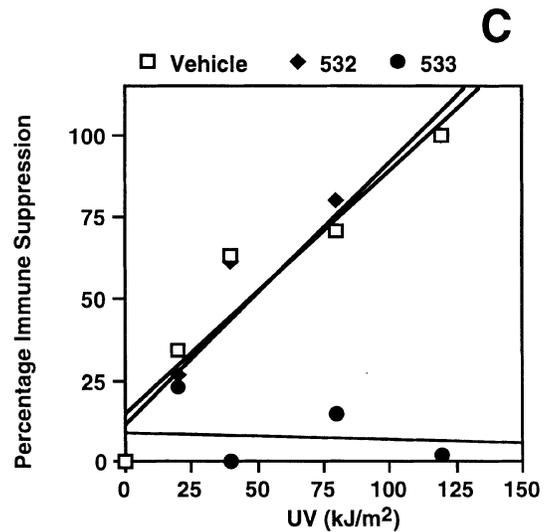
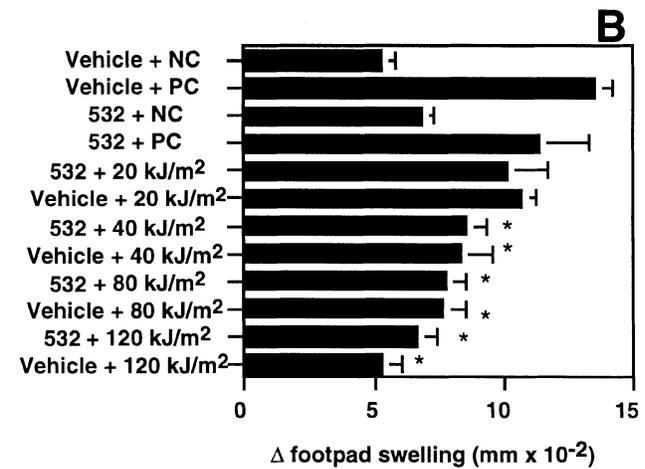
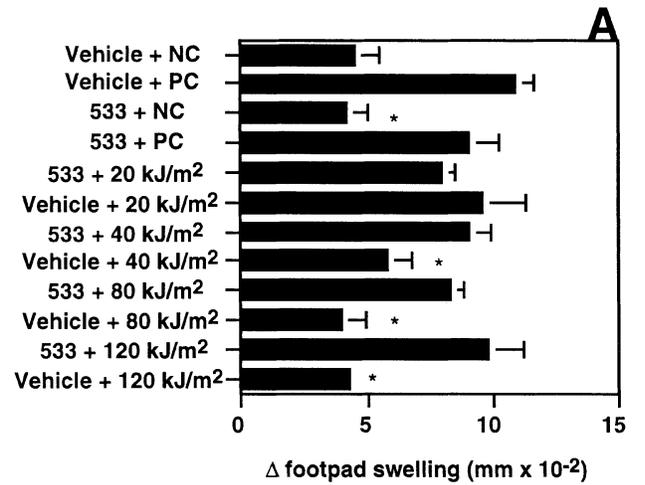


Figure 4. A UVA absorbing sunscreen affords immune protection. Two different sunscreen preparations were applied to mice 30 min prior to UV exposure. (A) Application of sunscreen P533, a UVA + B absorber blocks UV induced immune suppression (NC = negative control; PC = positive control). (B) Application of sunscreen P532, a UVB only absorber fails to block UV-induced immune suppression. (C) Comparison of immune protection afforded by UVB (P532) or UVA + B (P533) absorbing sunscreens. The percentage immune suppression generated in the presence of the vehicle (□); sunscreen P533 (◆); or sunscreen P532 (●) was plotted vs the dose of UV (WG-320) applied. *p ≤ 0.01; Student's t test vs the positive control. Individual experiments are shown, each experiment was repeated twice with similar results.

were exposed doses of ≥ 40 kJ per m² UV radiation; however, unlike the situation described above, the UVB only absorbing sunscreen had no protective effect. Significant immune suppression was generated regardless of whether the vehicle or sunscreen P532 was applied.

These data (Fig 4A, B) were converted to percentage immune suppression and plotted vs the dose of UV applied (Fig 4C). From these data it is apparent that applying the UVB only absorbing sunscreen affords no immune protection, whereas substantial immune protection is afforded by applying a sunscreen that absorbs UVA radiation. These data, in conjunction with the findings presented in Fig 3, indicate that the UVA wavelengths present in solar-simulated radiation are responsible for suppressing an established immune response.

DISCUSSION

Although it is well recognized that UV exposure suppresses the induction of an immune response when applied before immunization, less is known about the effects of UV on established immune reactions (Denkins *et al*, 1989; Magee *et al*, 1989). Perhaps the most successful public health campaign of the twentieth century was using vaccination to reduce the morbidity and mortality due to infectious disease. If sunlight can suppress established immune reactions, it may suggest that UV exposure could increase susceptibility in immunized individuals. Because the best index of sunlight exposure, skin cancer incidence, is on the rise (Boring *et al*, 1992) we suggest that increased human UV exposure has the potential to depress vaccine efficacy. This hypothesis is supported by the data presented here demonstrating that exposure to UV radiation after immunization can suppress immunologic memory. These findings, along with studies on human volunteers demonstrating that solar-simulated UV radiation suppresses the immune response to recall antigens (Moyal *et al*, 1997), indicate that it is critically important to study this phenomena in detail, paying particular attention to the mechanisms involved.

In order to understand the mechanism(s) involved, it is necessary to first determine which wavelengths are responsible for immune suppression. Although the majority of UV radiation in sunlight is UVA, immunologists have historically concentrated on the immunosuppressive effects of UVB radiation. This is because wavelengths in the UVB region of the solar spectrum induce immune suppression and skin cancer (Black *et al*, 1997). In regard to immune suppression, UVA was considered to be benign because most studies in the literature suggested that UVA did not depress immunity (Kripke *et al*, 1983; Morison *et al*, 1985; Sjoval and Christensen, 1986; Baadsgaard *et al*, 1987; Granstein *et al*, 1987); however, data from more recent studies, including those presented here, indicate that UVA radiation can suppress the immune response (Bestak and Halliday, 1996; Damian *et al*, 1997, 1999; LeVee *et al*, 1997). We suggest three potential explanations for these divergent findings. First, for the most part, experiments showing no immune suppression by UVA used fluorescent sunlamps as the source of radiation. Although fluorescent sunlamps are excellent sources of UVB radiation, they are poor substitutes for sunlight and their emissions, particularly in the UVA region of the solar spectrum, differ significantly from sunlight (Kim *et al*, 1998). On the other hand, when solar-simulated radiation was used in the place of fluorescent sunlamps, UVA was immune suppressive (Bestak and Halliday, 1996; Damian *et al*, 1997, 1999; LeVee *et al*, 1997).

The second potential reason for the divergent findings in the literature may reflect the protocols used to measure immune function. El-Ghorr and Norval (1999) report that different amounts of UVA radiation are required to suppress different types of immune reactions. They noted that 500 times more UVA was needed to suppress contact allergic reactions vs DTH. Many of the reports mentioned above, showing that UVA radiation fails to induce immune suppression, used contact allergy as the endpoint.

Third, the different mechanisms involved in inducing local and systemic immune suppression by UV radiation may also contribute to the divergent findings reported in the literature. For example, when volunteers were exposed to low-dose UV radiation provided by an alternating array of UVA and UVB fluorescent bulbs, immune suppression was only observed when the antigen (purified protein derivative) was injected at the irradiated site (local suppression). No immune suppression was noted when noted when the PPD was injected at distant sites nonirradiated sites (systemic suppression) (Damian *et al*, 1998). In the experiments presented here, we observe systemic suppression of established immune reactions by UVA, and this may be a function of the higher doses of UV employed in our studies.

Regardless of the reasons for the conflicting data reported in the literature, our findings illustrate a number of important facts. First, we used UV radiation to suppress the elicitation of DTH and immunologic memory to a common opportunistic pathogen. This suggests that UV exposure may contribute to decreased vaccine efficacy. Second, substantial and significant suppression of an established immune reaction was achieved with physiologic doses of UV radiation. We achieve 50% immune suppression with 50–80 kJ per m² of solar-simulated UV radiation. Based on our measurements of the intensity of solar UV radiation present in sunlight (in September midday sun, Houston TX, 30°N latitude), we estimate that 30 min of sunlight exposure will provide an equivalent UV dose in humans. Because some have reported that rodent antigen-presenting cells are three to four times more susceptible to the immune suppressive effects of UV radiation than human antigen-presenting cells (Goettsch *et al*, 1998b), care must be employed when extrapolating results from animal models to humans. It is interesting to note, however, that the same authors subsequently reported that 90 min of sunlight exposure (in July, 40°N latitude) caused 50% suppression of the immune response to *Listeria monocytogenes* in humans (Goettsch *et al*, 1998a). These immunosuppressive doses of sunlight can easily be achieved during normal recreational or occupational exposure.

Third, we show that UVA radiation, deficient in UVB, suppresses established immune reactions. The data presented in Fig 3 demonstrate that the dose–response curves for suppressing the elicitation of DTH by UVA + B and UVA are identical. This is an important observation impacting on sunscreen design. There has been a debate in the photobiologic community in regard to the need for UVA protection to prevent immune suppression (Ullrich *et al*, 1999). The reports indicating no immune suppression by UVA suggest there is little need for UVA protection (Kripke *et al*, 1983; Morison *et al*, 1985; Sjoval and Christensen, 1986; Baadsgaard *et al*, 1987; Granstein *et al*, 1987). On the other hand, some have shown immune protection when broad-spectrum sunscreens were applied before UV exposure, indicating a need for UVA protection (Moyal *et al*, 1997; Fourtanier *et al*, 2000). Determining the critical wavelengths required for immune suppression based on differential protection with different sunscreen formulations can be somewhat problematic. Although efforts are made to control the amount of sunscreen applied, the area of coverage, the skin type of the volunteers, and the sunburn protection factor of the sunscreen, some degree of uncertainty, especially in regard to the exact wavelengths and amount of UV radiation that actually reach the skin remains. In our study we used optical filters to achieve a three to four log reduction in the amount of UVB in the solar-simulated radiation. This removes any uncertainty about which wavelengths are involved. Moreover, the requirement for UVA protection is clearly supported by our sunscreen experiments, in which we found immune protection only when the applied sunscreen absorbed UVA radiation. These data provide compelling evidence for the need to protect against UVA exposure, particularly UVA II, in order to block UV-induced suppression of established immune reactions.

Data published by others indicate that exposure to UVA prior to UVB can prevent immune suppression (Reeve *et al*, 1998, 1999; Reeve and Tyrrell, 1999; Garssen *et al* 2001). Based on these

findings some have suggested that a short course of UVA adaptation before sunlight exposure can be used as a strategy for photoprotection. For this approach to be successful, UVA radiation by itself cannot be immune suppressive. Although we have not tried to reproduce Reeve's data directly (Reeve *et al*, 1998), our results demonstrating that solar-simulated UVA is not benign and can induce immune suppression, cautions against the use of UVA as a natural photoprotective agent. Our data imply that UVA protection is an absolute requirement to block immune suppression.

Melanoma is the most dangerous of all skin cancers. Although considerable evidence exists demonstrating that melanomas are immunogenic (Donawho *et al*, 1996; Wang *et al*, 1999) they clearly are capable of escaping immune destruction. This may be due in part to the production of immune suppressive cytokines, such as interleukin-10, by the melanoma cells (Dummer *et al*, 1996; Huang *et al*, 1996). The data presented here provide new insight into the mechanism behind the ability of melanoma to escape immune surveillance. Setlow's melanoma action spectrum suggests a role for UVA radiation in the induction of this disease (Setlow *et al*, 1993). Furthermore, studies by Donawho *et al* demonstrate that UV-induced suppression of the elicitation of the immune response is involved in the progressive growth of transplanted melanoma cells (Donawho and Kripke, 1991; Donawho *et al*, 1996). We suggest that the UVA may be playing a dual role in melanoma development; induction, as suggested by Setlow *et al* (1993) and promotion by suppressing the immune response, as demonstrated here. If this is true it may explain previous findings indicating that UVB-absorbing sunscreens were incapable of interfering with UV-induced enhancement of melanoma growth in mice (Wolf *et al*, 1994). Whether these findings (i.e., suppression of established immune responses by UVA) may also help to explain the confusing positive risk ratio between melanoma development in humans and the use of sunscreens that primarily absorb UVB radiation (Wolf *et al*, 1998), remains to be seen.

In summary, the data presented here demonstrate that UV exposure suppresses immunologic memory and the elicitation of the immune response to a common opportunistic pathogen. In addition, we found that UVA radiation, deficient in UVB was effective in suppressing the elicitation of immunity. These data suggest that sunlight-induced suppression of established immune reactions may serve as a risk factor for increased susceptibility to infectious agents. Moreover, they clearly indicate the need for UVA protection in blocking sunlight-induced immune suppression.

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