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EFFECTS OF SHORT-WAVE ULTRAVIOLET LIGHT (UVB) ON DELAYED HYPERSENSITIVITY IN THE GUINEA PIG

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Abstract. Guinea pigs, previously sensitized to dinitrochlorobenzene (DNCB), were exposed to varying doses of UVB radiation on the right flank for a period of 12 days. The response to an elicitation dose of DNCB was diminished in the irradiated skin immediately after UV treatment. This effect was dose-dependent. No reduction in the response could be demonstrated in unexposed skin. One week after UVB treatment the response to DNCB was increased, and after 2 weeks there was a normal eczematous reaction after application of DNCB.

Key words: Short-wave ultraviolet light (UVB); Delayed hypersensitivity; Dinitrochlorobenzene; Guinea pig

During recent years several reports have been published on the influence of ultraviolet (UV) radiation on the immune system. In 1963 Haniszko (4) reported that ultraviolet light, 280–320 nm (UVB), inhibits the cutaneous sensitization reaction in guinea pigs. It has also been shown that UVB radiation diminishes the allergic contact dermatitis reaction, when guinea pigs are exposed to UVB during the period of sensitization (8). The extensive use of photochemotherapy (PUVA) combining psoralen and long-wave ultraviolet light in the treatment of psoriasis, has initiated numerous studies on the effect of this treatment in other conditions. PUVA seems to inhibit induction of contact sensitivity in guinea pigs (9) and we have found that animals, previously sensitized to dinitrochlorobenzene (DNCB), have a decreased response to the allergen when it is applied after PUVA treatment (2).

The purpose of this investigation was to examine the effect of UVB radiation on established delayed hypersensitivity in the guinea pig. Furthermore we wanted to study the duration of the effect of the UV radiation which, to our knowledge, has not previously been investigated.

MATERIALS AND METHODS

Sensitization. Female, albino, Hartley strain guinea pigs weighing 350–400 g were used. Contact sensitivity to dinitrochlorobenzene (DNCB) was induced by a combination of intradermal and epidermal sensitization. This method gives a very high rate of sensitization (6). 0.1 ml of 0.01% DNCB in propyleneglycol was injected intradermally. One week later, closed patch topical induction was performed. A 2×2 cm filter paper, saturated with 0.15 ml 0.1% DNCB in 70% ethanol, was applied to the clipped and shaved dorsal thorax. This was covered by overlapping impermeable plastic tape, and held in place for 48 hours by elastic adhesive bandage, encircling the trunk. Challenge was performed 10 days later by closed patch with 0.05 ml 0.1% DNCB in 70% ethanol. This was applied to the dorsal thorax in a Finn Chamber (Epitest Ltd., Finland) and fixed with porous adhesive tape and elastic bandage. The patch was removed after 24 hours, and the reactions were evaluated the following day. The reactions were graded: 0=negative, 1+=slight erythema, 2+=strong erythema, 3+=intense erythema and swelling. All animals responded with either 2+ or 3+ reactions.

UVB treatment. The light source was 10 Sylvania F20 T 12 fluorescent light bulbs emitting a continuous spectrum from 280–320 mm with a peak at 310 mm. The irradiance was 1 mW/cm² at a distance of 20 cm. The minimal erythemal dose (MED) was 1 J/cm².

Preliminary studies showed that repeated exposures to 1 and 2 MED produced a homogeneous pink colour of the skin in the whole irradiated area. The erythema caused by the allergic reaction to DNCB was confined to the area covered by a Finn Chamber, was generally stronger, and could be evaluated in the presence of an UV-induced erythema. Very weak reactions to DNCB were, however, more difficult to evaluate.

Repeated exposures to 4 MED (4 J/cm²) or higher doses induced strong erythema, hyperkeratosis and scaling, making it impossible to evaluate any allergic response to DNCB.

To keep the animals in position during light exposure, they were given a combination of 3 mg fluanisone and 0.1 mg fentanyl citrate (Hypnorm®, Mekos) subcutaneously.

Experimental design. 20 animals previously sensitizied to DNCB were divided into three groups. Group A consisted of 5, group B, 5 and group C, 10 animals. An area of 6×6 cm on the right flank was clipped and shaved and then

Table I. *Allergic response to an elicitation dose of DNCB immediately after a 12-day period of UVB radiation*

Animal number	Group A, 0.5 J/cm²		Group B, 1 J/cm²		Group C, 2 J/cm²	
	Unexp. skin	Irrad. skin	Unexp. skin	Irrad. skin	Unexp. skin	Irrad. skin
1	+++	+++	+++	+	++	−
2	+++	++	++	−	+++	+
3	++	++	+++	++	++	−
4	++	++	++	+	++	+
5	+++	++	++	+	+++	+
6					++	−
7					++	−
8					+++	+
9					++	−
10					+++	+
Mean score	2.6	2.2	2.4	1.0	2.4	0.5

exposed to UVB radiation alternate days for 12 days. At each exposure, group A were given ½ MED (½ J/cm²); group B, 1 MED (1 J/cm²) and group C, 2 MED (2 J/cm²). A control group of 5 animals were not exposed to UV light.

Immediately after the last treatment all animals were exposed to an elicitation dose of DNCB by a Finn Chamber closed patch technique for 24 hours, as described above. This was performed both on the treated right flank and on the untreated left flank. The responses were evaluated 24 hours after removal of the patches. The animals in groups B and C were re-challenged with DNCB first after 7, then after 14 days, following exactly the same procedure.

To evaluate the effect of a non-specific inflammation on the contact allergic reaction, 3 sensitized animals were treated every other day for 12 days with 5% lauryl sulphate on the right flank. This induced an erythematous reaction comparable to the response of repeated exposures of 2 MED of UVB radiation. The animals were then patch-tested with DNCB on both flanks.

Statistical analysis. The statistical significance of the results obtained was calculated by using Student's *t*-test.

RESULTS

The results are summarized in Tables I, II, III and IV. The animals in group A received 0.5 J/cm² of UVB radiation at each exposure. The response to an elicitation dose of DNCB was not significantly less on exposed skin than on unexposed skin ($p>0.2$).

The group B animals exposed to 1 J/cm² of UVB radiation showed a significant inhibition of allergic

Table II. *Allergic response to an elicitation dose of DNCB when re-challenged one week after UVB radiation*

Animal number	Group B, 1 J/cm²		Group C, 2 J/cm²	
	Unexp. skin	Irrad. skin	Unexp. skin	Irrad. skin
1	++	++	++	+
2	++	++	++	++
3	+++	++	++	−
4	++	++	++	+
5	++	+	+++	+
6			++	−
7			++	+
8			++	+
9			++	−
10			+++	+
Mean score	2.2	1.8	2.2	0.8

Table III. *Allergic response to an elicitation dose of DNCB when re-challenged 2 weeks after UVB radiation*

Animal number	Group B, 1 J/cm²		Group C, 2 J/cm²	
	Unexp. skin	Irradi. skin	Unexp. skin	Irrad. skin
1	++	++	++	++
2	++	++	+++	++
3	++	+++	++	++
4	++	++	++	++
5	++	++	++	+++
6			++	+
7			++	++
8			+++	++
9			++	++
10			+++	++
Mean score	2.0	2.2	2.3	2.0

Table IV. *Allergic response to an elicitation dose of DNCB*

Re-challenge was performed one and then 2 weeks later

Animal number	Control animals unexposed to UVB radiation		
	First challenge	1 week later	2 weeks later
1	++	++	++
2	+++	+++	++
3	+++	++	+++
4	++	+++	+++
5	+++	++	+++
Mean score	2.6	2.4	2.6

response on exposed skin ($p<0.01$). After one week there was no significant difference between exposed and unexposed skin ($p>0.2$), and the same was found after 2 weeks.

In group C the animals were exposed to 2 J/cm² of UVB radiation, with a total dose of 12 J/cm². There was a marked reduction in the response to DNCB on exposed skin and in half the animals there was no reaction when tested immediately after radiation. When re-challenged one week later there was a slightly increased response, but there was still a significant difference between exposed and unexposed skin ($p<0.001$). After 2 weeks there were no longer any significant differences in the response to DNCB ($p>0.1$). In all three groups there was no change in allergic response in unirradiated skin compared with the animals in the control group, which were patch-tested the same days as the animals in groups A, B and C (Table IV).

The animals treated with lauryl sulphate on the right flank responded with equal 2+ or 3+ reactions on both flanks when challenged with DNCB.

DISCUSSION

This study demonstrates that UVB radiation diminishes the contact allergic reaction to DNCB in the guinea pig. The effect is dose-dependent. Radiation doses of 0.5 MED were shown to have no effect, while repeated exposure to 4 MED led to a strong local reaction, making it difficult to evaluate the response to DNCB. The suppression of cell-mediated reactivity was confined to UV exposed skin. After the period of UV exposure the reactivity to DNCB gradually increased, reaching the pre-treatment level after 1–2 weeks.

The mechanisms of action are only partly understood. Inflammation caused by lauryl sulphate did not reduce the allergic response. This indicates that the reduction of allergic response in the irradiated animals is a specific UVB effect. UV light has been shown to affect immunocompetent cells in several ways. Langerhans cells seem to play a central role in the afferent phase of the immune response by presenting the antigen to immunocompetent lymphocytes. Small doses of ultraviolet light (both UVA and UVB) alter and damage the surface markers of these cells (1). Furthermore, it has been shown that epidermal cells, in UVB irradiated skin, have an impaired antigen-presenting function (3, 11).

The intensity of the contact dermatitis reaction reflects the number of effector cells (10). Suppressor T-lymphocytes play a central role in the immune response by regulating the number of effector cells. T-lymphocytes are more sensitive to UV light than are B-lymphocytes (5), and it is probable that UV radiation influences both the T-effector cells and the regulation mechanism.

About 10% of UVB radiation penetrates the epidermis. The effect of this radiation on dermal structures may also be of importance in the suppression of contact dermatitis.

The results of this study express the total effect of UVB radiation on allergic contact dermatitis and do not provide a basis for evaluating the relative importance of the different mechanisms.

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