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Differential effects of UVA1 and UVB radiation on Langerhans cell migration in mice

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Abstract

The UVB (280–315 nm)- and UVA1 (340–400 nm)-induced migration of Langerhans cells (LC) from the epidermis and accumulation of dendritic cells (DC) in the lymph nodes draining the exposed skin site of C3H/HeN mice have been investigated. One minimum erythemal dose (MED) of UVB (1.5 kJ/m^2) and of UVA1 (500 kJ/m^2) were chosen, which have been shown previously to suppress delayed hypersensitivity (DTH). UVB irradiation resulted in a reduction in epidermal LC numbers, local to the site of the exposure, which was most apparent 12 h after exposure, but, in contrast, UVA1 had no significant effect even at 72 h after exposure. UVA1 did not exert any protection against the UVB-mediated depletion in LC numbers. The reduction in local LC following UVB exposure was prevented by systemic (intraperitoneal) treatment of mice with neutralising antibodies to either tumor necrosis factor (TNF)-α or interleukin (IL)-β 2 h prior to the irradiation. It has been reported previously that UVB exposure caused an increase in the number of dendritic cells (DC) in the lymph nodes draining the irradiated skin site. In the present study we have shown that UVA1 had a similar effect. Pretreatment of the mice with neutralising antibodies to IL-1β (by intraperitoneal injection) substantially inhibited DC accumulation induced by both UV regimens. However, anti-TNF-α antibodies affected only the UVB-induced increase, and did not alter the elevation in DC numbers observed following UVA1 exposure. These results indicate that UVB causes the migration of LC from the epidermis and an accumulation of DC in the draining lymph nodes by a mechanism that requires both TNF-α and IL-1β. In contrast, UVA1 does not cause LC migration from the epidermis and the accumulation of DC in the draining lymph nodes observed following UVA1 exposure requires IL-1β, but not TNF-α. It is likely therefore that UVA1 acts through a different mechanism from UVB and may target a cutaneous antigen presenting cell other than LC, such as the dermal DC.

Keywords: UVA1; UVB; Langerhans cells; TNF-α; IL-1β

1. Introduction

In the course of a normal cutaneous immune response, Langerhans cells (LC), the antigen presenting cells of the epidermis, internalise and process antigen, and, in response to changes in the local cytokine milieu, migrate to the draining lymph nodes. During migration through the afferent lymphatics, LC mature, as revealed by the altered expression of various adhesion and co-stimulatory molecules. They localise in the paracortical region of the lymph node by which time they have acquired the properties of immunostimulatory dendritic cells (DC). They present antigen peptides to specific T cells, which are then stimulated to proliferate and to produce a particular cytokine profile. The key signals for mobilisation and migration are believed to be provided in an autocrine fashion by interleukin (IL)-1β, and in a paracrine fashion by tumor necrosis factor (TNF)-α [1,2]. LC express only the type 2 (p75) TNF-α receptor, and mice deficient in expression of this receptor demonstrate significantly impaired LC migration [3]. Blocking of TNF-α activity by neutralising antibodies revealed that TNF-α acts early in the response, as injection of the antibodies prior to contact sensitisation, but not 18 h after, inhibited the sensitisation phase and the subsequent contact hypersensitivity (CH) on challenge [4]. IL-1β is produced solely by LC in the normal mouse epidermis, and its synthesis is enhanced by topical exposure to chemical allergens [5]. Intradermal injection of IL-1β stimulates LC migration from the epidermis, but at a slower rate than that induced by the

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identical administration of TNF-α [6]. It is believed that IL-1β provides one signal for the upregulation of TNF-α production by keratinocytes, and also delivers an autocrine stimulus for LC migration [1].

UV radiation is divided into three wavebands: UVC (200–280 nm), UVB (280–315 nm) and UVA (315–400 nm), further divided into UVA1 (340–400 nm) and UVA2 (315–340 nm). It is recognised that UVB exposure can suppress cell-mediated immune responses to a variety of antigens, including chemical contact sensitisers [7]. In previous studies we have shown that irradiation of C3H/HeN mice with either UVB or UVA1 can suppress both the CH response, using oxazolone as the skin sensitiser, and the delayed type hypersensitivity (DTH) response, using an infectious model with herpes simplex virus [8]. However, the doses required to exert such immunomodulatory effects were found to differ markedly. For CH, significant downregulation occurred with a dose of 500 kJ/m^2 UVA1 (equivalent to 1 minimum erythemal dose, MED) or higher, and a dose of 5 kJ/m^2 UVB (equivalent to 3.3 MED) or higher. For DTH, significant downregulation occurred with a dose of 1 kJ/m^2 UVA1 or higher, and a dose of 0.1 kJ/m^2 UVB or higher. These results indicate that different mediators and pathways may be involved in the immunomodulatory cascade, initiated by UV absorbance in the skin, which depend on the waveband of the irradiation as well as on the immune parameter monitored. In addition, some recent evidence demonstrates that waveband interactions may occur. For example irradiation with total UVA before UVB was shown to partially protect mice from the suppression of CH responses induced by UVB exposure alone [9,10].

One mechanism of UV-induced immunomodulation may be through effects on LC at the irradiated sites. It is known that UVB irradiation leads to a reduction in LC at the exposed site, with a loss of dendritic processes in the cells which remain in the skin [11–13]. We have demonstrated previously that UVB irradiation results in the accumulation of DC in lymph nodes draining the irradiated sites [14]. TNF-α was found to be a key cytokine in this response as systemic treatment of mice with an anti-TNF-α antibody before irradiation blocked the UVB-induced increase in DC numbers [15]. A similar accumulation of DC in local lymph nodes has been reported following contact sensitisation, with inhibition by the anti-TNF-α antibody [4,16].

The aim of the present investigation was to examine whether UVA1 irradiation induced a loss of LC from the epidermis of C3H/HeN mice and an accumulation of DC in the lymph nodes draining the exposed skin sites, as has been reported for UVB. Doses of UVA1 and UVB were chosen which are equivalent in terms of the induction of erythema at 1 MED each. The roles of TNF-α and IL-1β in these processes were examined in experiments where neutralising anti-TNF-α or anti-IL-1β antibodies were administered systemically prior to UVA1 or UVB irradiation.

2. Materials and methods

2.1. Mice

Female C3H/HeN mice, aged 6–8 weeks, were obtained from the specific pathogen-free animal breeding facility in the Medical Microbiology Transgenic Unit, University of Edinburgh. Mice were age-matched to within 1 week in individual experiments.

2.2. UV sources and irradiation of mice

Two Philips TL-12 lamps with an output range of 270–350 nm, peak 305 nm, emitting 2.5 W/m^2 (120 $J/m^2/min$) were used as the broadband UVB source. One MED for the C3H/HeN mice was 5 kJ/m^2. The UVA1 exposure was from a Doctor Hoenle Bluelight lamp, emitting 38.58 W/m^2 (2315 $J/m^2/min$). Wavelengths below 340 nm were filtered out by passage through a 1 cm depth of cation X (2,7-dimethyl-diaza-(3,6)-cycloheptadiene-(1,6)-perchlorate: Provided by Doctor F. de Gruijl, University of Utrecht) in distilled water at a concentration of 0.1 mg/ml, contained in a perspex tray. One MED for the C3H/HeN mice was 500 kJ/m^2. The output of both sources was determined using a filtered photodiode meter, calibrated against measurements made with a UV–visible spectroradiometer (model 742, Optronic Laboratories) across the spectral range 250–450 nm, with a tube to target distance of 16 cm. The spectroradiometer was calibrated against tungsten halogen and deuterium standard sources which are calibrated by the UK National Physics Laboratory (Teddington, UK).

Mice were UVB-irradiated in a perspex box with a maximum of 4 per box. Due to the length of exposure needed for the UVA1, mice were first anaesthetised by intraperitoneal injection of 150 μl sterile distilled water containing 1.67 mg/ml Hyponorm (Janssen Pharmaceutical, Oxford, UK) and 0.83 mg/ml Hyponovel (Roche, Welwyn Garden City, UK). They were then spread out in perspex boxes for irradiation to reduce shielding by littermates, and were cooled with an adjacent fan. Control mice for the UVA1 experiments were anaesthetised but not irradiated.

2.3. Epidermal LC numbers

Mice were exposed to UVB or UVA1 or not irradiated, as detailed above. In experiments examining the neutralising effects of TNF-α and IL-1β antibodies, 20 μg of each antibody (both goat immunoglobulins prepared after immunisation with mouse recombinant proteins, R&D Systems, Abingdon, UK) was injected intraperitoneally in a 100 μl volume 2 h before irradiation. Control mice, both irradiated and unirradiated, were injected with the same volume of normal goat serum at the same time. Mouse ears were collected into phosphate buffered saline at various

times after irradiation and split with focerps. Epidermal sheets from the dorsal surface only were prepared and stained for ADPase [17]. Sheets were mounted in 50% glyerol under a glass cover slip and the number of ADPase$^+$ cells counted in 10 fields per sheet (1 field=0.1 mm^2), with a minimum of four sheets per group. Results are shown as the mean±S.E.M. Significant differences between the relevant control and the experimental groups were calculated by Student's *t*-test.

2.4. DC enrichment and counting

The method described by Moodycliffe et al. [14] was used. In brief, draining auricular lymph nodes were collected from groups of 8–12 mice 24 h after irradiation. In some experiments the mice were pretreated with TNF-α or IL-1β antibodies 2 h before the exposure, as detailed above. The lymph nodes from each group were pooled and single cell suspension prepared by gentle disaggregation through a 200-mesh steel gauze (J. Staniar and Co., Manchester, UK). The cells were washed and suspended in 8 ml RPMI-1640 containing 100 I.U./ml penicillin, 200 mg/ml streptomycin, 2 mM L-glutamine, 100 μg/ml gentamicin, 10 μg/ml fungizone, 25 mM HEPES buffer and 10% heat-inactivated foetal calf serum (all Flow Laboratories, Irvine, UK) [RPMI–FCS]. The cell suspension was gently underlaid with 2 ml 14.5% w/v metrizamide (Nygaard, Oslo, Norway) and centrifuged at 600 *g* for 20 min at room temperature. The DC-enriched population was removed from the interface and washed twice in RPMI–FCS. The pellet was resuspended in RPMI–FCS and the DC counted by morphological analysis using light microscopy. DC were counted in multiple fields and the results shown are the means of these counts.

3. Results

3.1. Irradiation with 1 MED UVA1 does not deplete epidermal LC, whereas UVB causes a dose-dependent depletion

The frequency of epidermal LC was assessed as a function of ADPase$^+$ cells in epidermal sheets prepared from the dorsal ear skin of C3H/HeN mice. Data pooled from 17 independent experiments revealed that the average number of LC in untreated (naive) skin was 384±11.5 per mm^2 (mean±S.E.M.). Twenty-four h after exposure to 1 MED UVA1, there was no change in the number or the morphology of epidermal LC. In contrast, irradiation with 1 MED UVB reduced LC numbers significantly. The results of four individual experiments are shown in Table 1, and demonstrate a mean drop in epidermal LC numbers of approximately 28%. The LC remaining in the epidermis after UVB exposure were rounded and had fewer dendritic processes than in the unirradiated controls (data not presented). Two smaller doses of UVB were used in comparison with the 1 MED dose. When the LC were counted 24 h later, the numbers fell by 39.4% after 1 MED ($P<0.01$ v. unirradiated control), by 23% after 0.5 MED ($P<0.05$ v. unirradiated control) and by 7.6% after 0.25 MED (no significant change), demonstrating that LC migration induced by UVB exposure is dose-dependent.

It has been reported that irradiating mice with UVA immediately before UVB led to a partial reversal of the immunosuppressive effects of UVB on CH [10]. To determine whether such a combination influenced the ability of UVB to mobilise LC, mice were irradiated with 1 MED UVA1 followed by 1 MED UVB. At 24 h the LC numbers were depleted to levels comparable with those achieved by UVB alone (UVA+UVB 192.5 per mm^2±19.8, UVB 200±15.9, unirradiated control 329±17.6 in one experiment; and UVA+UVB 219±16.5, UVB 256±23.6, unirradiated control 329±18.8 in a second independent experiment).

3.2. The timecourse for LC depletion following UVB irradiation

The numbers of LC in the epidermis at various times after exposure to 1 MED UVB were examined and the results are shown in Fig. 1a. There was maximal depletion at 12 h, followed by a steady recovery back to the resting levels. The LC were rounded and had fewer dendrites at 12 h, but the morphology recovered to normal as the numbers of LC increased. In a second experiment, a 6 h time point was included and a significant reduction in LC numbers was demonstrated. The fall was 32% at 6 h ($P<0.05$ v.

Table 1
UVB induces a reduction in LC, but UVA1 does not. Mice were either unirradiated, or received either 1 MED UVB or 1 MED UVA1. Ears were collected and epidermal sheets stained for ADPase activity 24 h later. In each of four individual experiments, four epidermal sheets were prepared for each group. Ten fields were counted per individual sheet. Results from four individual experiments are shown as mean±S.E.M., and percentage change from control values

Experiment	unirradiated	UVB	% change	UVA1	% change
1	374±16.6	263±8.7	−29.7[a]	351±11.6	−6.1
2	329±17.6	200±15.9	−39.2[a]	301±21.4	−8.5
3	329±18.8	256±23.6	−22.2[a]	355±53.3	+7.9
4	430±9.9	346±4.3	−19.5[a]	432±19.5	+0.5

[a] $P<0.05$, versus control.

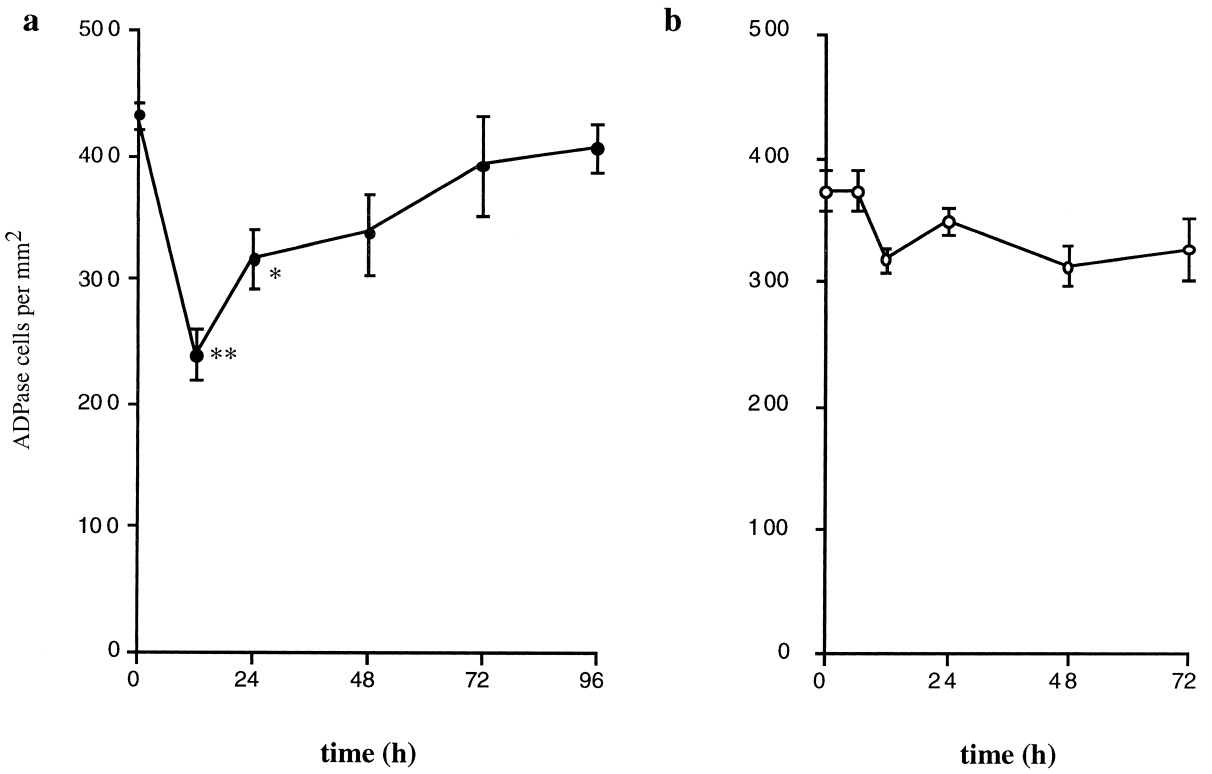

Fig. 1. The timecourse for the depletion of ADPase$^+$ cells in murine epidermis following irradiation with 1 MED UVB (a) or 1 MED UVA1 (b). Four epidermal sheets were prepared for each group and stained for ADPase. Ten fields were counted per individual sheet. Results are shown as the mean±S.E.M. for each timepoint, with 0 h representing the number in unirradiated animals. *=$P<0.05$ and **=$P<0.01$ v. unirradiated.

unirradiated) and 45.3% ($P<0.01$ v. unirradiated control) at 12 h.

LC were also monitored between 6 and 72 h following exposure to 1 MED UVA1 but there was no significant reduction at any time examined (Fig. 1b). The morphology of the LC was unaltered throughout with long dendritic processes remaining clearly visible.

3.3. Neutralisation of TNF-α and IL-1β prevents LC depletion after UVB irradiation

The available evidence indicates important roles for TNF-α and IL-1β in the induction of LC migration from the epidermis following contact sensitisation [1]. Therefore, experiments were performed to investigate the contribution of these cytokines to the UVB-induced reduction in LC numbers. Neutralising goat antisera against either murine TNF-α or IL-1β were injected intraperitoneally into mice 2 h prior to UVB irradiation. Control animals were injected with normal goat serum before exposure. The results of two independent experiments are shown in Fig. 2a for anti-TNF-α antibody, and in Fig. 2b for anti-IL-1β antibody. First, the antibodies themselves did not alter LC numbers significantly in unirradiated animals compared with those injected with normal goat serum. Second, the UVB-irradiated group injected with the normal goat serum showed a significant reduction in LC numbers compared with the unirradiated group also injected with the normal goat serum (a decrease of 32% and 41% for the anti-TNF-α antibody experiments in Fig. 2a, and 31% and 26% for the anti-IL-1β antibody experiments in Fig. 2b). Third, there was no significant reduction in LC numbers in the mice pretreated with the anti-cytokine antibodies before irradiation compared with unirradiated animals pretreated with the same antibody (7.3% and 9% for the TNF-α experiments in Fig. 2a, and 2% and 0% for the IL-1β experiments in Fig. 2b), and the morphology of the LC was no different from that in the unirradiated animals (data not shown). On the basis of these results, it is concluded that TNF-α and IL-1β are both critical cytokines in the migration of LC from the epidermis in response to UVB exposure.

3.4. Both UVA1 and UVB exposure cause the accumulation of DC in the lymph nodes draining the irradiated sites

We have shown previously that UVB irradiation causes the accumulation of DC in lymph nodes draining the exposed skin site [14]. Similar experiments were conducted here to corroborate this finding and to investigate whether UVA1 similarly induces such an increase in DC in the draining lymph nodes (DLN). Mice were exposed to 1

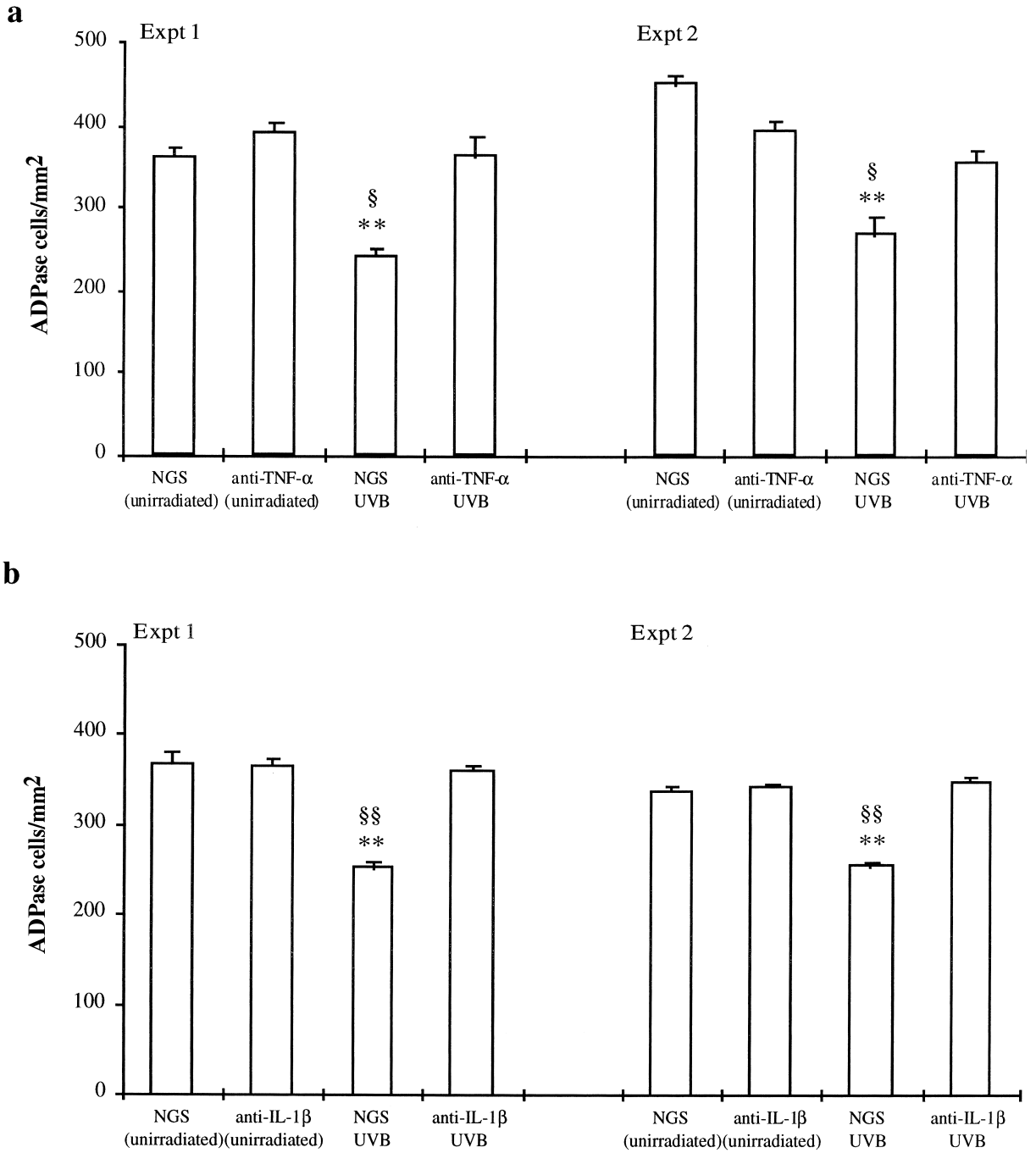

Fig. 2. The roles of TNF-α and IL-1β in UVB-mediated LC depletion from the epidermis. Mice were injected intraperitoneally with normal goat serum (NGS) or anti-TNF-α antibody (a) or anti-IL-1β antibody (b) 2 h before 1 MED UVB irradiation. Four epidermal sheets were prepared for each group and stained for ADPase activity. Ten fields were counted per individual sheet. Results are shown as the mean±S.E.M. for each treatment group. **=$P<0.01$ v. unirradiated, §=$P<0.05$ and §§=$P<0.01$ v. the same treatment with the anti-cytokine antibody.

MED UVB or UVA1, and the auricular lymph nodes draining the ears collected 24 h later. Control mice were not irradiated. The DC in the lymph nodes were counted following enrichment on a metrizamide gradient and the results of four independent experiments are shown in Table 2. The DC numbers per lymph node were increased by both UV regimens by approximately two-fold.

3.5. Neutralisation of TNF-α prevents the UVB-induced, but not the UVA1-induced, accumulation of DC in DLN

An important role for TNF-α in DC accumulation induced by UVB exposure had been revealed in a previous study [15]. The results in Fig. 3a for one representative experiment substantiate this finding. There was a 123%

Table 2
Irradiation with either UVA1 or UVB induces the accumulation of DC in DLN. Mice were given a 1 MED exposure to either UVB or UVA1, and DLN collected 24 h later. Control mice were unirradiated. DC were enriched on a 14.5% metrizamide gradient before counting. Results from four individual experiments are shown as the number of DC per DLN, and the percentage change from their respective control (unirradiated) mice, within individual experiments

Experiment	DC/resting LN	UVB	% change	UVA1	% change
1	1067	2186	+105	1857	+74
2	997	2136	+114	1929	+93
3	992	2214	+123	3450	+248
4	1923	4149	+116	3606	+88

increase in DC numbers in UVB-irradiated mice compared with unirradiated controls, and this was reduced to 36% in animals pretreated with anti-TNF-α antibody compared with unirradiated controls given the same antibodies.

However, for UVA1 the findings were quite different and these are also shown in Fig. 3a. There was a 248% increase in DC numbers in UVA1-irradiated mice, and a 236% increase in animals pretreated with anti-TNF-α antibody compared with unirradiated controls, also given the anti-TNF-α antibody. Thus, a major role for this cytokine in UVA1-induced DC accumulation is not indicated.

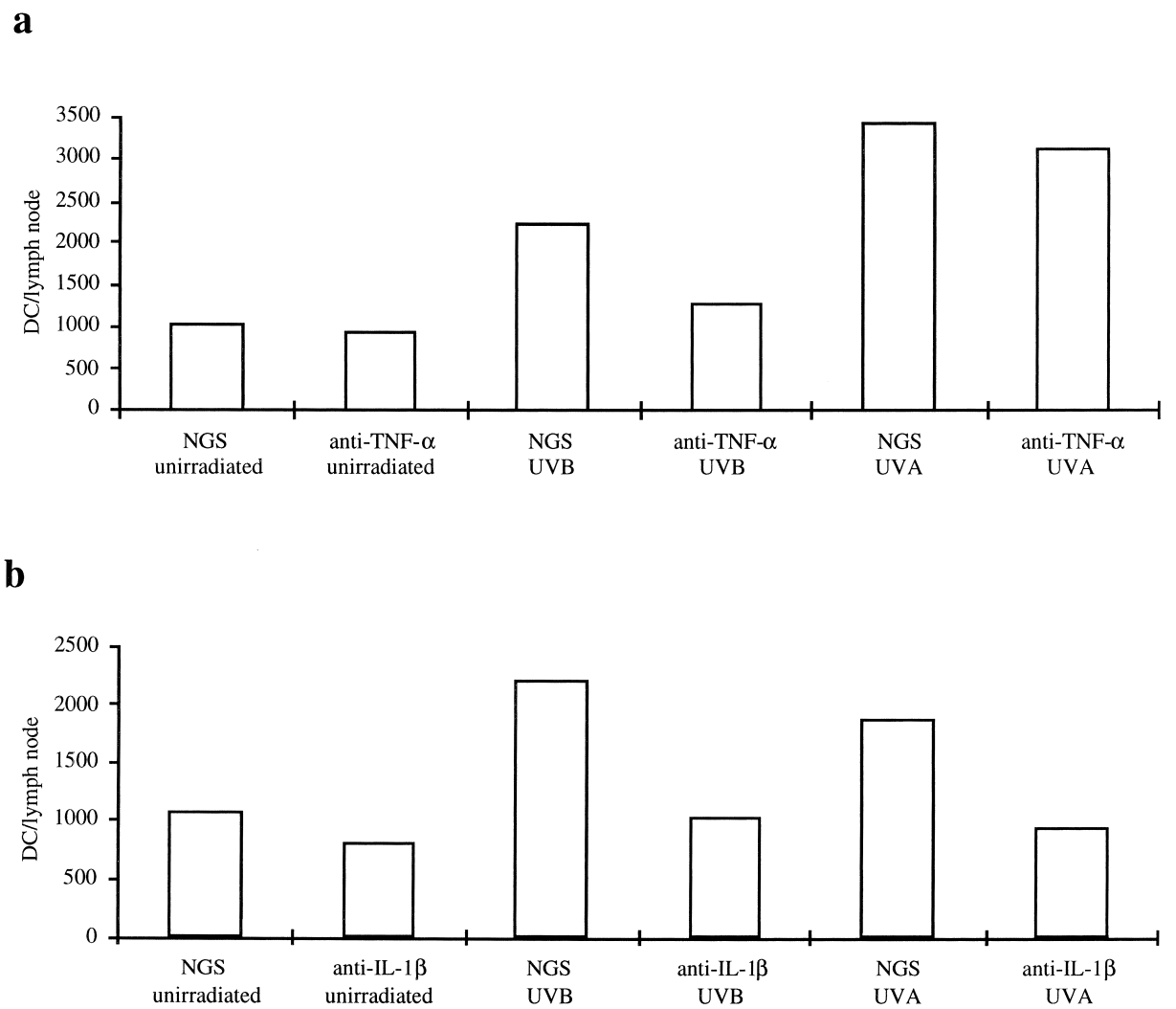

Fig. 3. The roles of TNF-α (a) and IL-1β (b) in DC accumulation in lymph nodes following 1 MED UVB or UVA1 irradiation. Mice were injected with normal goat serum (NGS) or anti-TNF-α antibody (a) or anti-IL-1β antibody (b) 2 h before irradiating. Auricular lymph nodes were collected 24 h later, and the DC counted following enrichment.

3.6. Neutralisation of IL-1β prevents both the UVB and the UVA1-induced accumulation of DC in DLN

As IL-1β was found to have a significant effect on the UVB-induced migration of LC from the skin, a similar approach to that just described for TNF-α was employed but using anti-IL-1β antibody [16]. It was revealed that the anti-IL-1β antibody, injected 2 h prior to either UVB or UVA1 exposure, inhibited substantially the accumulation of DC in the DLN (Fig. 3b). The antibody itself reduced the number of DC slightly compared with control mice injected with normal goat serum. UVB exposure induced a 103% increase in DC numbers, and UVA1 a 74% increase. In both cases, the anti-IL-1β antibody prevented this accumulation, and the DC numbers remained normal.

4. Discussion

Under normal circumstances, LC represent the major antigen presenting cells of the epidermis and their numbers and morphology are assessed most frequently by Ia or ADPase staining of epidermal sheets. Both detection methods give the same LC frequency in the C3H/HeN strain of mouse [18]. One mechanism whereby UV radiation results in a modulation of immune responses is thought to be by trafficking of a proportion of LC from the epidermis. The cells that remain show abnormal morphology and have a reduced capacity for antigen presentation. Most studies in this area have used broadband UVB sources or solar simulated irradiation, and the contribution of the UVA waveband to this process is not well defined. Limited information indicates that the effect on LC may be wavelength dependent. Noonan et al. [19] reported that irradiation at 270 or at 290 nm resulted in a decrease in epidermal LC numbers with associated changes in dendrites, but this did not happen at 320 nm. Similarly, using monochromatic light, Obata and Tagami [20] found that UVA from 320 to 400 nm was almost ineffective in reducing LC numbers: Wavelengths shorter than 300 nm were the most efficient. Previously we reported that a single dose of 40 kJ/m^2 UVA did not result in a significant reduction in LC numbers 24 h later [21]. However Aubin et al. [18] demonstrated that chronic UVA exposure (10 kJ/m^2, 3 times weekly for 4 weeks) reduced the number of $ATPase^+$ cells in the epidermis of C3H/HeN mice by approximately 45% compared with unirradiated controls.

In the present study UVB was found to reduce epidermal LC numbers, while UVA1 did not (Table 1 and Fig. 1). The doses from the two lamps were equivalent in terms of erythema. This parameter was chosen as it has been suggested that the action spectrum for LC depletion from the epidermis is similar to that for the induction of erythema [20]. We had shown previously that DTH responses could be suppressed by either of the UV doses used, and CH responses by the UVA1 dose but a 3.3-fold higher dose of UVB was required [8]. Therefore, on the basis of these data, there does not appear to be a direct correlation between the migration of LC from the epidermis and the UV-induced modulation of DTH or CH. This has also been indicated elsewhere. For example, Noonan et al. [19] found no loss in epidermal LC following exposure at 320 nm, even at doses which caused 50% suppression of CH, and the chronic UVA irradiation protocol used by Aubin et al. [18] which resulted in a considerable loss of LC did not alter CH responses, despite the sensitisation occurring through the irradiated site. It is interesting to note that tolerance to subsensitising doses of allergens can be occur without LC migration [22].

Previously, the importance of TNF-α in mediating UVB-induced LC migration from the epidermis was demonstrated [15]. This was corroborated in the present study (Fig. 2). By pretreatment of mice with an anti-IL-1β antibody prior to UVB exposure which restored the LC numbers to the level found in unirradiated controls (Fig. 2), a role for IL-1β has now been also established. These two cytokines are critical in LC movement during the early phase of contact sensitisation [1], together with the down-regulation of E-cadherin, expressed on the LC surface [23].

The induction of cutaneous TNF-α may be wavelength dependent. Skov et al. [24] found that irradiating human skin with 3 MED UVB led to a rapid increase in TNF-α protein in suction blister fluid, the response being maximal at 6 h after exposure. However, UVA1, at an equivalent erythematous dose, resulted in a slight decrease at 6 h compared with the pre-irradiation value. Furthermore, in mice carrying a chloramphenicol acetyl transferase reporter transgene bearing the entire TNF-α promoter, UVA1 did not induce the expression in skin, in contrast to UVB [25]. DNA damage, occurring predominantly at dipyrimidine sites and including the formation of cyclobutane pyrimidine dimers, may be the initiating factor for TNF-α gene expression following UVB irradiation [26]. Other types of DNA damage, mainly oxidative, are found at longer wavelengths which may not be appropriate for the induction of TNF-α. Such a situation has been described recently for another UV-induced cytokine, IL-6. The wavelength dependence for the induction of IL-6 expression in keratinocytes was found to match the spectra for both DNA absorption and cyclobutane pyrimidine dimer formation [27]. For example at 365 nm, approximately one-thousandth of the quantity of IL-6 was produced compared with the same dose but at 302 nm. Thus it is possible that different wavelengths may have a variable capacity to induce one or more of the cytokines required for LC migration.

Although IL-1β is known to be essential for contact sensitisation in the mouse and is required for allergen-induced LC migration [16], little information is available regarding the wavelength dependence and the timing of its induction by UV. In one study of human subjects, IL-1β

was detected at elevated concentrations in suction blister exudates at 15 h after 3 MED solar simulated radiation, an increase which was sustained for at least the next 72 h [28].

In agreement with previous studies [14,15], we detected an accumulation of DC in the lymph nodes draining the irradiated skin site following exposure to UVB (Table 2). This was dependent on both TNF-α and IL-1β as neutralisation of either cytokine inhibited the increase in numbers (Fig. 3). Perhaps surprisingly, given the lack of effect of UVA1 on epidermal LC numbers, exposure to this waveband also resulted in an accumulation of DC in the DLN (Table 2). A similar role for IL-1β in UVA1- as in UVB-induced accumulation was indicated. In contrast to the situation with UVB, TNF-α appeared not to be involved after UVA1 exposure as the anti-TNF-α antibody failed to alter substantially the increase in DC (Fig. 3).

It is possible, therefore, that UVA1 irradiation may not lead to a significant upregulation in TNF-α expression in the epidermis, with the lack of LC migration being a consequence. However, as DC still accumulate in the DLN after UVA1, the source of DC must be other than the LC. It is possible that they originate in the dermis. DC are found in the papillary dermis of normal skin and are considered to be migrating LC, either precursors on their way to the epidermis or mature cells on their way to the DLN. In addition the dermis contains other subsets of DC, with one such in mice showing a LC-like phenotype [29]. An inflammatory infiltrate consisting of lymphocytes and macrophages is found around the vessels of the upper and mid-dermis following UVA1 irradiation [30], indicating that cellular changes are induced by this waveband in the dermis. TNF-α is neither chemotactic nor chemokinetic for macrophages [31], so blocking of its activity is unlikely to alter this response. The same may be true for the dermal DC moving from the skin to the DLN, in response to UVA1. In addition DC from different sites may express different adhesion molecules and the ones on dermal DC may render the cells susceptible to IL-1β-induced, but not to TNF-α-induced migration. To distinguish the source of the DC in the DLN following UVA1 exposure, it may be possible to use a range of phenotypic markers, as has been described recently in studies by Randolph et al. [32] and Anjuere et al. [33].

In summary, we have shown that 1 MED UVB irradiation causes the migration of epidermal LC and the accumulation of DC in lymph nodes draining the exposed skin site, both processes being dependent on the availablity of TNF-α and IL-1β. One MED UVA1 does not cause the migration of epidermal LC but DC do still accumulate in the DLN, an effect which requires IL-1β but not TNF-α.

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