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The Mandatory Role of IL-10–Producing and OX40 Ligand-Expressing Mature Langerhans Cells in Local UVB-Induced Immunosuppression

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The mechanism underlying the local UVB-induced immunosuppression is a central issue to be clarified in photoimmunology. There have been reported a considerable number of cells and factors that participate in the sensitization phase-dependent suppression, including Langerhans cells (LCs), regulatory T cells, IL-10, and TNF-α. The recent important finding that LC-depleted mice rather exhibit enhanced contact hypersensitivity responses urged us to re-evaluate the role of LCs along with dermal dendritic cells (dDCs) in the mechanism of UVB-induced immunosuppression. We studied the surface expression of OX40 ligand (OX40L) and the intracellular expression of IL-10 in LCs and dDCs from UVB-irradiated (300 mJ/cm²) skin of BALB/c mice and those migrating to the regional lymph nodes from UVB-irradiated, hapten-painted mice. In epidermal and dermal cell suspensions prepared from the UVB-irradiated skin, LCs expressed OX40L as well as CD86 and produced IL-10 at a higher level than Langerin⁺ dDCs. The UVB-induced immunosuppression was attenuated by the administration of IL-10–neutralizing or OX40L-blocking Abs. In mice whose UVB-irradiated, hapten-painted skin was dissected 1 d after hapten application, the contact hypersensitivity response was restored, because this treatment allowed dDCs but not LCs to migrate to the draining lymph nodes. Moreover, LC-depleted mice by using Langerin-diphtheria toxin receptor–knocked-in mice showed impaired UVB-induced immunosuppression. These results suggest that IL-10–producing and OX40L-expressing LCs in the UVB-exposed skin are mandatory for the induction of Ag-specific regulatory T cells. *The Journal of Immunology, 2010, 184: 5670–5677.

Ultraviolet radiation is one of the significant environmental factors affecting humans or other animals. It is well known that UV, in particular the middle wavelength range (290–320 nm, UVB), can be hazardous to human skin by acutely evoking sunburn and epidermal cell death and by chronically inducing skin cancers and skin aging (1–4). UVB radiation also exerts an immunomodulating effect on cutaneous contact hypersensitivity (CHS) by affecting various skin-constituent cells and factors (5). Preirradiation of sensitizing area with low-dose UVB suppresses the development of CHS to hapten in mice (6). In addition to the failure to generate hapten sensitization, mice develop tolerance, because animals treated in this way cannot be resensitized with the same hapten at a later time point. The UVB-induced immunosuppression appears to be hapten-specific, because the sensitization with other nonrelated haptens is not affected (6). Moreover, this hapten-specific immunosuppression can be transferred as an injection of lymph node cells or splenocytes from UVB-tolerized mice into naive mice inhibits the sensitization with the relevant hapten in the recipients (7). It was once considered that the UVB-induced immunosuppression was mediated by hapten-specific suppressor T cells (5, 8, 9). Now, this suppressor T cell is renamed regulatory T cell (Treg) (10–12). Therefore, a suppressive signal that causes UVB-induced tolerance is hypothesized to exist in the draining lymph node (DLN) of UVB-irradiated skin, where Tregs are induced and suppress the generation or function of effector T cells. However, it remains unclear how the suppressive signal is transmitted from the skin to the DLNs. On one hand, Tregs act in part through the induction of IL-10 production (13). On the other hand, IL-10 is a key cytokine to induce Tregs, and keratinocytes have been considered to be the source of IL-10. However, all the mechanisms underlying UVB-induced immunosuppression are not attributed to keratinocyte-derived IL-10, because human keratinocytes are incapable of producing IL-10 (14). More fundamentally, keratinocytes are unable to migrate to the DLN. Therefore, alternative cells with a migrating ability are likely responsible for mediation of suppressive signals.

Recent studies have revealed the involvement of OX40 (CD134) and its ligand (OX40L) in T cell–APC interaction (15–18). OX40 is expressed on activated CD4⁺ T cells and on certain populations of CD8⁺ T cells (15, 17, 19), whereas OX40L is expressed on APCs, such as activated B cells (20), dendritic cells (DCs) (21, 22), microglia (23), and endothelial cells (24). Ligation of OX40L on human DCs enhances their maturation and production of cytokines (22), and blockade of OX40L during naive T cell–DC interaction suppresses the development of IL-4–producing T cells (25). It is thus suggested that OX40 and OX40L play an important role in the interaction of DCs with T cells to induce, in particular, Th2 cells. Moreover, CD4⁺CD25⁺ Tregs express OX40 at a high level compared with CD4⁺CD25⁻ T cells (26, 27). Considering that UVB-induced suppressor T cells were historically identified...
as Th2 cells (28), these findings provide an implication that OX40–OX40L interaction participates in the development of UVB-mediated CD4+CD25+ Tregs.

Langerhans cells (LCs) are capable of migrating from the epidermis into the DLNs on sensitization (29). Several investigator groups have suggested that LCs are responsible for induction of Treg in DCs in the UVB-irradiated skin remains unclear in major parts. Recent immunological studies have demonstrated that there are dermal DCs (dDCs), including Langerin+ dDCs and Langerin+ dDCs in the murine skin (32–36). This raises the possibility that not only LCs but also dDCs have an ability to induce Tregs by UVB irradiation of the skin.

In this study, we demonstrate that UVB irradiation of the skin leads to IL-10 production and OX40L expression by LCs. Our study using Langerin-diphtheria toxin receptor (DTR)–knocked-in mice shows that the IL-10–producing and OX40L-expressing LCs play a mandatory role in the induction of Tregs.

Materials and Methods

Animals and reagents

Six- to 10-wk-old BALB/c female mice were purchased from Kyudo (Kamamoto, Japan). Mice were maintained on a 12-h light/dark cycle under specific-pathogen-free conditions. Langerin-DTR–knocked-in mice was generated (37). To deplete Langerin+ cells, mice were injected i.p. with diphtheria toxin (DT) (100 ng each; Sigma-Aldrich, St. Louis, MO). Protocols were approved by the Institutional Animal Care and Use Committee of the University of Occupational and Environmental Health, Fukuoka, Japan.

Contact hypersensitivity

Mice were sensitized with dinitrofluorobenzene (DNFB) by applying 50 μl 0.5% DNFB in acetone:olive oil (4:1) to the shaved abdomen on day 0. On day 5, 20 μl 0.2% DNFB was applied to both ears for elicitation. Ear swelling was measured with a micrometer 24 h post elicitation.

UVB irradiation

The shaved abdomen was exposed to UV with a bank of four UVB lamps (Toshiba FL 20S, Toshiba Medical Supply, Tokyo, Japan) (5, 8, 9) that emit mostly of their energy within the UVB range (290–320 nm), with an emission peak at 313 nm. The irradiance was measured with a UVR-305/365 digital radiometer (Tokyo Kogaku Kikai KK, Tokyo, Japan). Mice were exposed to UVB irradiation (300 mJ/cm2), whole skin suspensions were stained with APC-Cy-7–conjugated anti-MHC class II or PE-conjugated EpCAM and FITC-conjugated CD103 (BD Biosciences) for 30 min and stained with Alexa Fluor 647-conjugated Annexin V (Invitrogen, Carlsbad, CA) and 7-aminoactinomycin D (7-AAD; BD Biosciences), according to the manufacturer’s protocol. Apoptosis in keratinocytes or DCs was analyzed by FACSCanto (BD Biosciences) using FlowJo software (Tree Star) as previously described (39).

In vitro promotion of LC IL-10 production by RANK ligand and its blockade with neutralizing Ab against RANK

Freshly isolated epidermal cell suspensions (5 × 106/well) were cultured with or without 1 μg/ml rRANK ligand (RANKL) (R&D Systems, Minneapolis, MN) for 24 h. For RANK-neutralizing assay, 1 μg/ml anti-RANK Ab or isotype-matched control Ab (R&D Systems) was added to the culture 3 h before the addition of rRANKL. Intraacellular IL-10 of LCs was measured by FACS.

In vivo neutralization of IL-10 and blocking of OX40L

Mice received i.p. injections of 25 μg anti-mouse IL-10 Ab (R&D Systems) or 10 μg anti-mouse OX40L Ab (Biologend) for 4 consecutive days (on days 1–4) after UVB irradiation (on day −2) and DNFB sensitization (on day 0). They were challenged with DNFB on day 5, and the ear swelling responses were measured. For control, mice received the same volume of PBS and were sensitized and challenged with DNFB.

Statistical analysis

All data were statistically analyzed using the Student t test. A p value of <0.05 was considered to be significant. Bar graphs were presented as mean ± SD of the mean value.

Results

Langerin+ dDCs are decreased in number and become apoptotic in UVB-irradiated skin

It is a long-held concept that LCs play a critical role in CHS, as they serve as APC and migrate to the DLNs (40). However, recent immunological studies have demonstrated that not only LCs but also Langerin+ dDCs and Langerin− dDCs exist in the skin and may differentially function as APCs. We first investigated the numerical change of LCs, Langerin+ dDCs, and Langerin− dDCs in UVB-irradiated skin. Whole skin suspensions were prepared from the UVB-irradiated and nonirradiated skin as control 24 h after UVB exposure and analyzed by flow cytometry. Using anti-MHC class II, anti-CD11c, anti-Langerin (CD207), and EpCAM Abs, skin-resident DCs were clearly sorted out of the suspensions (Fig. 1A, 1B). As assessed by the percentage analysis, the populations of LCs (Langerin+ EpCAM+) and Langerin− dDCs (Langerin− EpCAM−) showed no substantial change after UVB irradiation (Fig. 1C versus 1D), although UVB-irradiated skin-derived DCs had a slightly broader MHC class II expression (Fig. 1B). However, the percentage of Langerin+ dDCs was dramatically decreased in UVB-irradiated skin (Fig. 1C versus 1D). When the absolute number of each LC/DC subset per skin specimen was calculated, UVB irradiation reduced dramatically the number of Langerin+ dDCs and moderately that of LCs and did not affect that of Langerin− dDCs.
UVB-induced immunosuppression

With the use of anti-EpCAM and anti-Langerin Abs, DCs from nonirradiated skin were clearly sorted out into the three categories: LCs (Langerin+ EpCAM-), Langerin+ dDCs (Langerin+ EpCAM+), and Langerin- dDCs (Langerin- EpCAM+). A, These single-cell suspensions were stained with anti-CD11c, anti-EpCAM, and anti-Langerin Abs. By flow cytometry, LC subsets (Langerin+ EpCAM+) and Langerin+ dDC subsets (Langerin+ EpCAM+) of CD11c+ FITC+ cells were detected in the DLNs. LCs were gradually increased in number in both UVB-irradiated and nonirradiated groups (Fig. 2A). In contrast, the number of Langerin+ dDCs peaked on day 3 in nonirradiated mice, but their number in UVB-irradiated mice was very low (Fig. 2B). The number of Langerin+ dDCs in UVB-nonirradiated skin was increased until day 2 and gradually declined, whereas in UVB-irradiated skin peaked on day 1 and rapidly decreased (Fig. 2C). Thus, UVB irradiation allowed LCs and Langerin+ dDCs to migrate into the DLNs, but Langerin+ dDCs in the irradiated skin did not migrate to the DLNs. There was no significant difference in the number of FITC+ DCs of each subset (data not shown). Therefore, the numerical reduction of Langerin+ dDCs in the UVB-irradiated skin did not result from their emigration from the skin. It is assumed that when a hapten is applied to the UVB-preirradiated skin, there are few Langerin+ dDCs capable of migrating to the DLNs and priming Tregs or effector T cells.

UVB upregulates LC maturation and promotes IL-10 production and OX40L expression

It has long been thought that LCs represent one of the most likely targets for UVB in immunosuppression because of their location in the skin and their importance as APCs. Recent studies using LC-depleted mice have shown that LCs are dispensable for CHS (37) and rather downregulate the CHS response (41). In this line of thinking, dDCs may play an essential role for the development of CHS (42). To address the regulatory functions of UVB-irradiated DC populations, we examined the expression of intracellular IL-10 and surface OX40L, as well as CD86 in LCs and Langerin+ dDCs.

Epidermal suspensions were prepared from UVB-irradiated and nonirradiated skin and subjected to flow cytometric analysis. Compared to the nonirradiated control skin, LCs from UVB-irradiated skin showed high expression levels of CD86, OX40L, and intracellular IL-10 (Fig. 3A). However, such elevations were not observed in Langerin+ dDCs. This suggests that UVB irradiation upregulates the maturation (CD86 expression) of LCs and promotes the production of IL-10 and the expression of OX40L by LCs, but Langerin- dDCs are not susceptible to UVB.

To examine these IL-10–producing and OX40L-expressing mature LCs in the UVB-irradiated skin, we analyzed apoptosis of LCs and dDCs in the UVB-irradiated mice. Six hours after UVB irradiation, we assessed apoptotic cells by flow cytometry and defined them as Annexin V+ and 7-AAD- cells. Langerin+ dDCs and LCs became apoptotic after UVB irradiation (Fig. 1F). There was no selectivity for UVB-induced apoptosis in these two subsets, but when they were compared in the apoptotic cell percentage, Langerin+ DCs were more sensitive to UVB. In contrast to these cells, Langerin- dDCs were resistant to UVB.

LCs but not Langerin+ dDCs migrate from UVB-irradiated skin to DLNs

We examined the numbers and migration timings of LCs, Langerin+ dDCs, and Langerin- dDCs in the DLNs after FITC application of UVB-irradiated or nonirradiated skin. UVB-irradiated (day -2) and nonirradiated control mice were painted with FITC (day 0). On days 1–4, single-cell suspensions were prepared from the DLNs and stained with anti-CD11c, anti-EpCAM, and anti-Langerin Abs. By flow cytometry, LC subsets (Langerin+ EpCAM+) and Langerin+ dDC subsets (Langerin+ EpCAM+) of CD11c+ FITC+ cells were detected in the DLNs. LCs were gradually increased in number in both UVB-irradiated and nonirradiated groups (Fig. 2A). In contrast, the number of Langerin+ dDCs peaked on day 3 in nonirradiated mice, but their number in UVB-irradiated mice was very low (Fig. 2B). The number of Langerin+ dDCs in UVB-nonirradiated skin was increased until day 2 and gradually declined, whereas in UVB-irradiated skin peaked on day 1 and rapidly decreased (Fig. 2C). Thus, UVB irradiation allowed LCs and Langerin+ dDCs to migrate into the DLNs, but Langerin+ dDCs in the irradiated skin did not migrate to the DLNs. There was no significant difference in the number of FITC+ DCs of each subset (data not shown). Therefore, the numerical reduction of Langerin+ dDCs in the UVB-irradiated skin did not result from their emigration from the skin. It is assumed that when a hapten is applied to the UVB-preirradiated skin, there are few Langerin+ dDCs capable of migrating to the DLNs and priming Tregs or effector T cells.

It has recently been reported that LCs express RANK, and UVB irradiation upregulates cutaneous RANKL, which modulates the
functions of DCs to induce Tregs (43). We have previously reported that when rRANKL was added to LC culture, the RANKL-exposed LCs produce a high amount of IL-10 (44). In contrast, UVB radiation is known to induce apoptosis of epidermal cells. To examine whether epidermal keratinocytes produce RANKL upon UVB exposure in relation to the apoptotic state, epidermal suspensions were prepared from the UVB-irradiated skin 24 h post-exposure and stained to see apoptosis and RANKL expression. By flow cytometry (Fig. 4A), keratinocytes were divided into live (Fig. 4Aa; 7-AAD−, Annexin−), apoptotic (Fig. 4Ab; 7-AAD−, Annexin+), and dead (Fig. 4Ac; 7-AAD+, Annexin+) populations. The apoptotic keratinocyte expressed RANKL at a higher degree than did the live and dead keratinocytes (Fig. 4B). Thus, UVB-irradiated apoptotic keratinocytes are capable of producing RANKL and subsequently stimulate LCs to produce IL-10 (44). Next, we performed a RANK-blocking study. The production of IL-10 by LCs was promoted by the addition of rRANKL to the culture of epidermal cells, and this increased IL-10 production was increased sharply at day 3 in nonirradiated mice but not increased in UVB-irradiated mice. The number of Langerin+ dDCs peaked at day 1 in the UVB-irradiated mice.

**FIGURE 2.** Numbers of LCs, Langerin+ dDCs, and Langerin− dDCs in DLNs after FITC application to UVB-irradiated or nonirradiated skin. Mice were irradiated with UVB (300 mJ/cm2) on day −2 or nonirradiated and sensitized with FITC on day 0. On days 1–4, DLNs were collected and stained for CD11c, Langerin, and EpCAM. We gated on the FITC+CD11c+ population and counted the EpCAM+ Langerin+ (LCs), EpCAM+ Langerin+ (Langerin+ dDCs), and EpCAM− Langerin− (Langerin− dDCs) cells. A, The number of LCs was gradually increased after FITC application in the UVB-irradiated and nonirradiated skin. B, The number of Langerin+ dDCs was increased sharply at day 3 in nonirradiated mice but not increased in UVB-irradiated mice. C, The number of Langerin− dDCs peaked at day 1 in the UVB-irradiated mice.

It is likely that the production of IL-10 and the expression of OX40L in LCs contribute to the UVB suppression of CHS. To test the significance of IL-10 and OX40L in the suppression, UVB-preirradiated mice (on day −2) were injected i.p. with anti–IL-10 or anti-OX40L Ab for 4 consecutive days (days 0–3), whereas mice were sensitized (day 0) and challenged (day 5) with DNFB. Preirradiation of sensitizing sites to UVB suppressed CHS in mice (Fig. 5). The administration of anti–IL-10 Ab completely restored the CHS response. In contrast, UVB-induced CHS suppression was partially but significantly abrogated by anti-OX40L Ab. We cannot negate the possibility that not only LCs but also other cells are the targets of this blocking procedure, but it seems that IL-10 is profoundly involved in UVB-induced suppression, and OX40L expression is required for the full-blown suppression of CHS.

**CHS is successfully induced by dissection of UVB-irradiated and hapten-applied skin at early phase of sensitization**

To determine whether LCs and dDCs serve as inducers of Tregs, a skin dissection study was performed for prevention of LC migration at the sensitizing phase. Mice were sensitized with FITC on day 0. When the sensitized skin was dissected on day 1, the total number of migrating LCs was significantly decreased particularly in mice preirradiated with UVB before FITC application (Fig. 6A). We therefore examined the CHS response to FITC in mice whose UVB-irradiated and hapten-applied skin was dissected on day 1. This treatment is considered to allow Langerin+/− dDCs to migrate to the DLNs, but most LCs cannot emigrate there. Mice receiving dissection of the sensitizing site did not exhibit UVB-induced immunosuppression of CHS compared with the nondissected and

**FIGURE 3.** Expression of surface CD86, intracellular IL-10, and surface OX40L in LCs and Langerin− dDCs from the skin and DLNs. A, Epidermal cell suspensions were obtained from UVB-irradiated skin 24 h after UVB exposure or nonirradiated skin. Solid line, UVB-irradiated skin; dotted line, nonirradiated skin; and closed shadow, isotype-matched control. B, Cell suspensions were obtained from the DLNs of mice receiving UVB irradiation (day −2) and FITC painting (day 0) or mice receiving FITC painting without UVB irradiation. Lymph nodes were taken on day 1, and migrating LCs were identified as FITC+CD11c+EpCAM−Langerin− cells and migrating Langerin− dDCs as FITC+CD11c+EpCAM−Langerin− cells. Solid line, UVB-irradiated mice; dotted line, nonirradiated mice; and closed shadow, isotype-matched control.
mice were irradiated with UVB on shaved skin (day 2), sensitized with DNFB on day 0, and challenged with DNFB on day 5. IL-10-neutralizing Ab (25 μg per mouse) or PBS (for control) was injected i.p. on days 0–3. Positive control mice were sensitized and challenged, and negative control mice were challenged without sensitization; *p < 0.05.

**Discussion**

This study addressed the immunological mechanism underlying the impaired sensitization through UVB-irradiated skin. We found that the UVB-induced immunosuppression of CHS is mediated by IL-10-producing, OX40L-expressing, and CD86 highly expressing mature LCs, which are induced by exposure to RANKL released from UVB-irradiated, apoptotic keratinocytes. The mandatory role of LCs for the UVB-induced suppression was confirmed by the two types of studies, the dissection of sensitizing site and the use of LC-depleted mice. In addition, the recently identified Langerin+ dDCs as well as Langerin+ dDCs (36) seem to play no suppressive role.

Many studies have shown that IL-10 is an essential cytokine in the depression of CHS (45–47). The administration of rIL-10 suppresses CHS and induces Ag-specific tolerance (48). IL-10 has also been reported to be a key cytokine in the mechanism of UVB-induced tolerance, as anti–IL-10 Ab treatment before UVB exposure prevents UVB-induced tolerance (49). The neutralizing study using anti–IL-10 Ab further confirmed that IL-10 is essential for the UVB-induced immunosuppression of CHS. Concerning the source of IL-10, a number of studies have demonstrated keratinocytes to be the producer. However, our present study showed that IL-10 is efficiently produced by LCs when the skin is exposed to UVB. The earlier studies on the production of IL-10 by keratinocytes were performed by determining IL-10 mRNA induction and IL-10 protein release in murine keratinocytes shortly postirradiation with UVB (50) or poststimulation with hapten coupling (51). Because cultured keratinocytes were used in those studies, the conclusion may not correctly reflect the in vivo UVB exposure to the skin. In addition, the mechanism of human UVB-induced immunosuppression cannot be explained with the finding obtained from murine keratinocytes. Whereas murine keratinocytes are capable of releasing IL-10 (50, 52), human keratinocytes are an unlikely source of IL-10 following in vivo UVB exposure, as they express little mRNA for IL-10 and secrete no IL-10 protein (14). We have previously reported that IL-10–producing LCs in the grafted skin have a crucial role in the induction of Ag-specific Tregs (44). Together with the present finding, it is suggested that the LCs that migrate from the skin to the DLNs are the important source of IL-10 under the condition of UVB irradiation or skin grafting. Such a finding of
DC production of IL-10 has also been reported in pulmonary DCs critical for the induction of tolerance (53). A group of investigators have found that RANKL, which is expressed in keratinocytes of the UVB-irradiated skin, regulates Treg numbers via activation of DCs (43). In another line of studies, i.v. injection of photopheresis-induced apoptotic cells inhibited an immune response to hapten, and this was caused by CD11c+ cells that induce Ag-specific Tregs (54). Likewise, Tregs have been shown to be generated following APC engagement of apoptotic cells (55). Thus, ingestion of apoptotic cells is not merely a scavenging event but also an active process of immune tolerance induction. Teleologically, this process has been described as one of the peripheral tolerance mechanisms (56). We have previously shown that when LCs are exposed to RANKL, they produce IL-10 (44). In this report, we found that apoptotic keratinocytes express RANKL at a higher degree than live keratinocytes and dead keratinocytes. Besides the phagocytosis of apoptotic cells by APCs, RANKL is another tolerogenic signal from apoptotic cells, and the resultant change of DCs to regulatory cells is one of the mechanisms by which apoptosis is related to tolerance.

The blockade of OX40–OX40L interaction by neutralizing OX40 Ab ameliorates experimental allergic encephalomyelitis and experimental colitis, which are Th1-mediated inflammatory diseases (23, 57). OX40 signaling is thus required for the optimal evolution of the Th2 response (58). Moreover, OX40 signaling is involved in the generation of Tregs, and the delivery of OX40 signals can override Treg activity (59). In our data, the blockade of OX40–OX40L interactions partially abrogated the UVB-induced immunosuppression in a comparison with IL-10 neutralization, suggesting that OX40–OX40L interaction is partially responsible for the Treg induction.
We confirmed the crucial role of LCs in the UVB-induced immunosuppression by two strategies. One is that UVB-irradiated, hapten-painted skin was dissected 1 d after hapten application. By this treatment, a considerable number of dDCs could migrate to the DLNs, but LC migration was inhibited, and as a result, the CHS response was restored. In the other study, LCs were more effectively depleted by DT injection to Langerin-DRTR-knock-in mice, and the UVB-induced suppression was markedly abolished in the mice, clearly demonstrating the necessity of LCs for the suppression. Recently, Wang et al. (60) have reported that LCs play no critical role for the UVB-induced immunosuppression.

There are major differences in UVB-irradiation doses and mouse strains between their and our studies. Wang et al. (60) irradiated C57BL/6 with UVB at 45 mj/cm² for 3 consecutive days. We irradiated BALB/c mice with UVB at 300 mj/cm² once. Although BALB/c mice are usually not very susceptible to UVB, we found that a single exposure of BALB/c mice to UVB at 300 mj/cm² induces UVB immunosuppression with an elevated percentage of Foxp3+ CD25+ cells in the DLN cells. Because single irradiation of the skin to UVB and following FITC painting is convenient for the study of DC migration to the DLN, we used this protocol and this strain of mice in our study. These differences in the UVB dose and mouse strain possibly give rise to the different results. Alternatively, because their study includes no adoptive transfer experiment, it is unclear whether Tregs are induced in their experimental system. As shown in our study, Langerin+ dDCs become apoptotic after UVB irradiation and cannot migrate to the DLN following hapten application. Given the CHS-inductive role of Langerin+ dermal DCs (37), the abrogation of them by UVB may attenuate the sensitization process of CHS to hapten even without the induction of Tregs. The suppression observed by Wang et al. (60) might be related to this phenomenon. Moreover, the study using the mice deprived of LCs and repopulated with Langerin+ dDCs showed that Langerin+ dDCs are not a requirement for the suppression. Therefore, the UVB immunosuppression in our system is not merely caused by the attenuation of inductive role of Langerin+ dDCs.

The UVB-induced immune tolerance is mediated by Ag-specific Tregs, as the suppression can be adoptively transferred into naive recipients (61). According to the recent observations, UVB-induced Tregs have the CD4+CD25+ phenotype (62), express CTLA-4 (13), and bind to the lectin dectin-2 (61). The Tregs are modulated by T cell function and amelioration of experimental allergic encephalomyelitis. They accumulate in B follicles.

References


