

Topical tacrolimus (FK506) leads to profound phenotypic and functional alterations of epidermal antigen-presenting dendritic cells in atopic dermatitis

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Background: Atopic dermatitis (AD) is a chronic inflammatory skin disease in which antigen-presenting epidermal dendritic cells (DCs), ie, Langerhans cells and the so-called inflammatory dendritic epidermal cells (IDECs) expressing the high-affinity receptor for IgE (FcεRI) may play a significant pathophysiologic role. Therapeutic efficacy of the immunosuppressive macrolide tacrolimus (FK506) in AD has been demonstrated in clinical trials, but little is known of its mode of action.

Objective: The present study focused on the effects of topical tacrolimus treatment on epidermal CD1a⁺/FcεRI⁺ DC populations in lesional AD.

Methods: Immunohistological analysis, epidermal DC phenotyping, and functional studies were performed on skin biopsy specimens from treated and untreated lesional skin of 10 patients with AD participating in a clinical trial with tacrolimus.

Results: Untreated lesional skin was characterized by a high proportion of CD1a⁺ cells, which was largely due to a high proportion of IDECs strongly expressing FcεRI. Epidermal DCs isolated from untreated lesional skin exhibited high stimulatory activity toward autologous T cells, which was strongly reduced while clinical improvement was seen during application of tacrolimus. Concomitantly, a decreased FcεRI expression was observed in both Langerhans cells and IDECs. Finally, topical tacrolimus led to a progressive decrease in the IDEC population within the pool of CD1a⁺ epidermal DCs and also to a decrease in their CD36 expression, which is indicative of lower local inflammation.

Conclusion: Epidermal CD1a⁺ DCs may represent a target for topical tacrolimus in the treatment of AD. (*J Allergy Clin Immunol* 2001;107:519-25.)

Key words: Atopy, Langerhans cells, inflammatory dendritic epidermal cells, dendritic cells, IgE receptor

Atopic dermatitis (AD) is a chronic inflammatory skin disease that is associated with an increased systemic production of IgE and local infiltration of mononuclear cells, mainly T cells and antigen-presenting cells.¹⁻³ Because all existing therapeutic approaches involve significant risks and side effects, much effort has been made to develop new therapeutic alternatives.⁴ Topical tacrolimus therapy has recently been shown to be highly effective in moderate-to-severe AD, with minimal systemic absorption and an acceptable safety profile.⁵⁻⁸ Tacrolimus appears not to cause skin atrophy, which is found with topical steroid therapy.⁹

Little is known about the *in vivo* mode of action of tacrolimus on the cutaneous immune system in normal subjects or in patients with AD. It is known that IgE molecules are found on the surface of epidermal antigen-presenting Langerhans cells (LCs),^{10,11} especially in AD lesions, which may contribute to the amplification of the IgE response toward environmental allergens.¹² This hypothesis is strengthened by the recent demonstration of high-affinity receptors for IgE (FcεRI) on LCs,¹³⁻¹⁵ which is strongly and specifically upregulated on epidermal dendritic cells (DCs) in AD.^{16,17} Thus AD may involve an IgE-mediated delayed-type hypersensitivity reaction in which FcεRI-bearing, antigen-presenting LCs act as the pivotal link between aeroallergens and antigen-specific T cells infiltrating the skin lesions.^{3,12} In addition, AD has been shown to be associated with phenotypic and functional alterations of CD1a⁺ epidermal DCs.^{16,18-20} Two distinct CD1a⁺ epidermal DC populations have been identified: (1) LCs that contain Birbeck granules and (2) inflammatory dendritic epidermal cells (IDECs) that do not contain Birbeck granules. The latter also have higher FcεRI expression and may therefore be the key DCs in AD.¹⁶

The present study focused on investigating the effects of topical tacrolimus on the 2 distinct epidermal DC populations in lesional AD skin by using flow cytometric methods, immunohistochemistry, and functional studies. A subset of 10 patients from a recent multicenter, randomized, double-blind clinical trial, which compared

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TABLE I. Synopsis of individual clinical data of the respective response groups

Patient No.	Age (y)	Severity grade*	Therapy	Treatment time	SMLR	Clinical group
101	22	6	0.1% FK	14 d	ND	Response
107	34	7	0.03% FK	7 d	Done	Response
116	30	8	0.03% FK	7 d	ND	Response
109	28	5	0.03% FK	7 d	ND	Response
114	18	7	0.3% FK	7 d	Done	Rebound
118	20	8	0.03% FK	14 d	Done	Rebound
110	17	7	Placebo	7 d	ND	Placebo
113	19	8	Placebo	21 d	ND	Placebo
108	31	7	0.3% FK	2 d	Done	Short
119	30	8	Placebo	21 d	ND	Self healing

FK, Topical tacrolimus; ND, not done.

*Severity grades as in *Acta Derm Venereol (Stockh) Suppl.* 1989;144:13-4.**Abbreviations used**

AD: Atopic dermatitis
 DC: Dendritic cell
 IDEC: Inflammatory dendritic epidermal cell
 LC: Langerhans cell
 MFI: Mean fluorescence intensity
 rFI: Relative fluorescence index
 SMLR: Skin mixed lymphocyte reaction

tacrolimus ointment with a vehicle control,⁵ volunteered to provide skin biopsy specimens for these analyses.

METHODS**Patients and therapeutic protocol**

This study was conducted at the Department of Dermatology, University of Munich. Ten patients with moderate-to-severe AD who were participating in a multicenter clinical trial⁵ were randomized to receive, in a double-blind fashion, 0.03%, 0.1%, or 0.3% tacrolimus ointment or a vehicle control (the ointment base without tacrolimus) for a period of 3 weeks and maximum treatment area of 1000 cm². Patients in this substudy had atopic dermatitis lesions of similar severity on both of their cubital fossae and were treated on one side only. The primary endpoint was a summary score for erythema, edema, and pruritus (maximum score, 9). Table I presents the summary score for each patient. This trial was conducted according to the principles expressed in the Declaration of Helsinki and was approved by the local ethics committee. Written informed consent was obtained from all the patients.

Biopsy specimens

On days 2, 7, 14, or 21, shave biopsy specimens were obtained from lesional and nonlesional skin after achievement of local anesthesia from the patients' cubital fossae. For technical reasons, the biopsy specimens of the treated area of 2 patients were taken immediately before and after treatment (patients No. 119 and 116).

Preparation of epidermal cell suspensions for flow cytometric and functional analysis

Crude epidermal cell suspensions were obtained by means of trypsinization of skin specimens, as previously described,²¹ and processed for immunolabeling and flow cytometric analysis as follows.

Flow cytometric analysis of epidermal DCs

For the flow cytometric analysis, a previously described triple-

staining method was used.²¹ The following primary antibodies were used: mAb 22E7 detects the IgE-binding α chain of Fc ϵ RI (IgG1; generous gift of Dr J. Kochan, Hoffmann—La Roche Co, Nutley, NJ) and does not interfere with the binding site for IgE on the receptor²²; mAb IV.3 (IgG2b; Medarex, West Lebanon, NH) labels the human low-affinity IgG receptor Fc γ RII/CD32; mAb IOP36 (IgG2b; Immunotech, Marseille, France) is directed against the thrombospondin receptor CD36; mAb BEAR1 (IgG1, Immunotech) reacts with the integrin chain CD11b, which is highly expressed on IDECs but not on LCs²¹; and MOPC (IgG1; Sigma, Deisenhofen, Germany) and UPC10 (IgG2b, Sigma) were used at the same concentration as the isotype control mAb. Further labeling and blocking were performed with FITC-conjugated goat anti-mouse antibody (Jackson Laboratories, West Grove, Penn); normal mouse serum (Sigma, 1:10); and a mixture of phycoerythrin-labeled T6/RD1 antibody (Coulter, Krefeld, Germany), which stains CD1a⁺ DCs (ie, LCs and IDECs) and 7-amino-actinomycin-D (Sigma) to detect dead cells.

For quantitative evaluation, the vital CD1a⁺ populations were gated out manually. Their proportion in the CD1a⁺ cell pool, as well as the mean fluorescence intensity (MFI) for each surface molecule, were determined for each population of interest by using Cell-Quest software.²¹ The relative fluorescence indices (rFIs) of all surface receptors were determined as follows:

$$\text{rFI} = (\text{MFI} [\text{receptor}] - \text{MFI} [\text{control}]) / \text{MFI} (\text{control}).$$

In situ immunolabeling on cryosections

Immunohistochemistry was carried out on the biopsy specimens of one responder patient. A portion of the skin specimens taken at day 7 was snap-frozen in liquid nitrogen and stored at -80°C. Cryostat sections (6 μ m) were prepared for immunohistochemistry by using the mouse alkaline phosphatase-antialkaline phosphatase complex technique, as described in detail elsewhere.¹¹ As primary monoclonal mouse antibodies 22E7, IOP36, BEAR1, BL6 (anti-CD1a, Immunotech), or the relevant isotype control (see above) were used at 10 μ g/mL. Sections were evaluated by 2 independent investigators. Scoring of cells in the dermis was performed at 400 $\times$ magnification and evaluated per square micrometer of area section by using digitalized pictures obtained with the Diskus software (Hilgers, Königswinter, Germany).

Functional analysis of epidermal DCs

In 4 of the patients (Table I) selected on the availability of a sufficient number of epidermal cells, the evaluation of the functional capacity of CD1a⁺ epidermal DCs was done by using an autologous skin mixed lymphocyte reaction (SMLR). Freshly isolated epidermal cell suspensions of the treated and untreated lesions were simultaneously prepared to minimize any influence of preparation

variability and washed twice in 50 mL of supplemented RPMI-1640 medium containing 10% FCS, 1% antibiotics-antimycotics, and 2 mmol/L L-glutamine (Life Technologies, Karlsruhe, Germany). Autologous responder T cells were prepared from heparinized whole blood of the patients by means of density gradient centrifugation on Lymphoprep (Nycomed, Oslo, Norway), followed by purification (>95% T cells) over nylon wool and washing twice in supplemented RPMI-1640 medium. Finally, 1×10^5 T cells were cocultured for 5 days with epidermal cells containing 2×10^3 CD1a⁺ cells per well in round-bottom 96-well microtiter plates (Nunc, Roskilde, Denmark) in a total volume of 200 μ L. In addition, 1×10^5 T cells were cultured without epidermal cells in a total volume of 200 μ L to control for spontaneous cell proliferation. Cell multiplication was assessed by adding tritiated thymidine (1 mCi per well; Amersham International, Amersham, United Kingdom) to each well 18 hours before harvesting. Culture plates were shock frozen at -80°C , and incorporation of the isotope was measured by means of liquid scintillation and expressed as the average counts per minute per well of triplicate cultures. The relative stimulation indices were calculated from the counts per minute obtained for the SMLR and T-cell conditions as follows:

$$\text{Relative stimulation index} = (\text{cpm [SMLR]} - \text{cpm [T cells]}) / \text{cpm [T cells]}.$$

For statistical evaluation of significance, the Mann-Whitney *U* test was performed. Results are given as mean rFI or mean percentage of positive cells $\pm$ SEM.

RESULTS

Clinical response of AD lesions to tacrolimus treatment

The 10 patients were assigned to one of 3 clinical response groups (Table I) according to their treatment and local response as follows: the *responder* group consisted of 4 patients treated with tacrolimus who experienced clinical improvement at the time of biopsy; the *placebo* group consisted of 2 vehicle control-treated patients; and the *rebound* group consisted of 2 tacrolimus-treated patients whose symptoms temporarily improved but who then experienced a flare-up around the time of the biopsy. The clinical courses for a further 2 patients did not meet any of these definitions, and the data from these patients were analyzed separately. One of these patients had a tacrolimus treatment time of only 2 days at the time of biopsy and was classified as having a short treatment course. The second patient underwent biopsy before and 3 weeks after treatment with the vehicle control and healed completely. This patient was classified as self-healing.

Topical tacrolimus leads to a depletion of IDECs from the epidermal DC population

Epidermal DCs in normal human skin are known to represent about 1% to 2% of all epidermal cells. From immunohistological studies, it is known that this percentage is increased in AD.¹¹ To better quantify the alterations of the epidermal DC infiltrate during treatment, enumeration of CD1a⁺ cells by means of FACS analysis was performed (Fig 1). After treatment with tacrolimus, the percentage of CD1a⁺ cells in the lesional epidermis was reduced in the responder group from $3.4\% \pm 0.6\%$ to $2.3\% \pm 0.1\%$, which is in the range for normal skin.¹¹ The

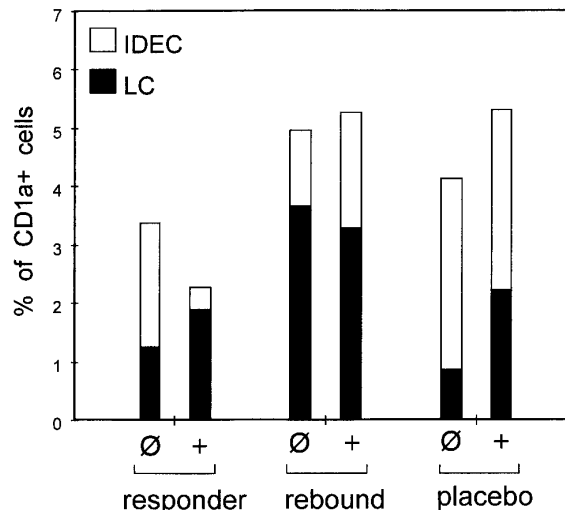


FIG 1. The proportion of CD1a⁺ epidermal DCs in cell suspensions prepared from biopsy specimens of treated and untreated skin and their subclassification into IDECs and LCs. Mean proportions of IDECs and LCs, as determined by flow cytometric analysis, in treated (+; 0.03%, 0.1%, 0.3% tacrolimus) and untreated ($\emptyset$) elbow lesions in patients with AD are shown. Included are 4 patients who responded to treatment (*responder*), 2 patients who initially improved but became worse at the time of biopsy (*rebound*), and 2 patients receiving placebo. The height of each column indicates the proportion of CD1a⁺ cells as a percentage of the total epidermal cells. Note that only the responder group showed a decrease in the mean CD1a⁺ cell proportion and a corresponding decrease in the relative proportion of IDECs.

placebo-treated patients did not show this effect ($4.1\% \pm 2.1\%$ vs $5.3\% \pm 0.8\%$), and therefore the decrease in the proportion of CD1a⁺ cells in the epidermis appears to be related to the topical tacrolimus treatment.

Using FACS analysis, we have previously shown that CD1a⁺ cells can be subclassified into CD11b⁺ LCs and CD11b⁺ IDECs.¹² During treatment with tacrolimus, the IDEC population was markedly depleted from the epidermis in all responders (Fig 2). A quantitative plot of the ratio of IDECs to all CD1a⁺ epidermal DCs showed that the proportion of IDECs was markedly reduced at day 7 and strongest at day 14, but not at day 2, suggesting that this reduction occurs between days 2 and 7 (not shown). The IDEC population of the patients in the placebo group remained unaltered (Fig 1). Therefore the main cause for the decrease of CD1a⁺ cells in AD lesions during topical tacrolimus treatment appears to be attributable to a depletion of the IDEC subpopulation, and this correlates with the clinical response and treatment time.

Taken together, these results suggest that the changes in the composition of the inflammatory infiltrate, namely of the CD1a⁺ epidermal DCs, underlie the clinical efficacy of tacrolimus ointment in AD.

Decrease of FcεRI expression on CD1a⁺ epidermal DCs by topical tacrolimus

Using FACS analysis of epidermal CD1a⁺ DCs, we found a clear reduction of FcεRI expression on CD1a⁺ DCs

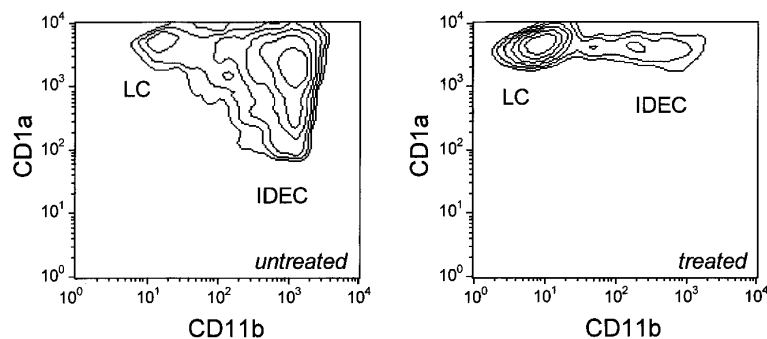


FIG 2. Qualitative changes of CD1a⁺ cell infiltrate after tacrolimus treatment. Contour plot analysis of lesional AD skin from a treated and untreated lichenified flexural lesion of a representative responder patient with severe chronic AD (patient No. 107) after 1 week of topical treatment with 0.03% tacrolimus ointment. Topical tacrolimus ointment therapy induced marked depletion of IDECs, as well as an increase of CD1a expression in the remaining IDECs.

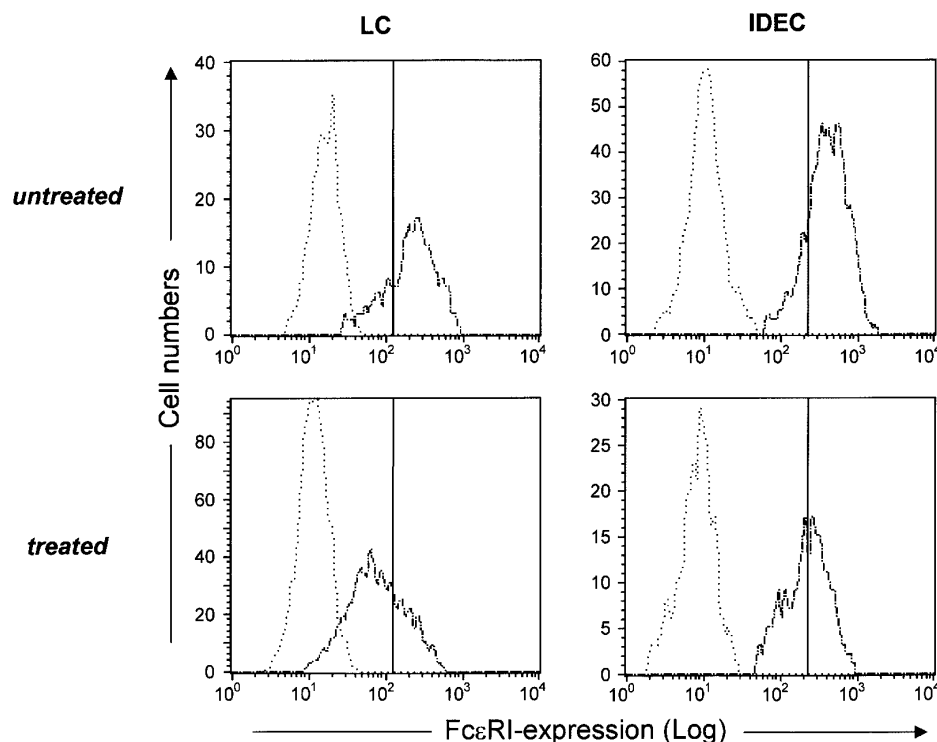


FIG 3. Changes in the FcεRI expression of CD1a⁺ cells after tacrolimus treatment. Histogram plot analysis of FcεRI expression on CD1a⁺ cells of a responder patient with severe chronic AD (patient No. 107). Epidermal cells were isolated from lesional AD skin from a treated and untreated lichenified flexural lesion after 1 week of topical treatment with 0.03% tacrolimus ointment. A reduction of FcεRI expression is observed in both CD1a⁺ cell populations.

from an rFI of 30.0 ± 6.6 to 7.6 ± 1.2 (mean $\pm$ SEM) in the responder group. This decrease was mainly caused by the disappearance of FcεRI-expressing IDECs from the epidermis (Fig 2) and a decrease in the expression of FcεRI at the level of each individual CD1a⁺ cell type (ie, LCs and IDECs; Fig 3). Both individual effects synergistically led to a marked overall reduction of FcεRI expression on CD1a⁺

cells in the treated epidermis. Furthermore, this phenomenon appeared to be related to the duration of treatment because the lowest expression was measured by day 14 of treatment (not shown). Consequently, the FcεRI/FcγRII ratio, a significant marker for chronic untreated AD skin lesions,¹⁷ was reduced in every responder patient during treatment but not in the other patient groups (not shown).

Taken together, the disease-specific cellular markers of AD were altered toward a milieu typical of normal, nonlesional skin.

Alterations of the inflammatory infiltrate in the dermis induced by topical tacrolimus treatment

Because it has been reported that AD lesions are composed of a complex infiltration of mainly mononuclear cells and T cells,^{1,23} we investigated putative alterations of this dermal infiltrate in biopsy specimens from untreated and treated involved skin and uninvolved skin in one patient. As seen in Table II, untreated involved skin was characterized by the presence of CD11b⁺/CD36⁺ cells. This was observed in the upper dermis, mainly around the vessels of the papillary dermis. These cells probably migrate into the epidermal compartment because they were absent in uninvolved skin. At the same time, there was an increase in CD1a and FcεRI-expressing DCs in the epidermis of untreated involved skin (not shown). Topical application of tacrolimus led to disappearance of CD11b⁺/CD36⁺ cells from the epidermis and the loss of these cells from the underlying papillary layer of the dermis. The immunohistochemistry results are further supported by a downregulation of CD36 surface expression in the epidermal DCs of treated lesions, as shown with FACS analysis (not shown).

Reduction of the stimulatory capacity of epidermal DCs by tacrolimus

One of the main functions of epidermal DCs is antigen presentation to lymphocytes. Therefore we investigated the stimulatory capacity of simultaneously prepared epidermal cell suspensions toward their autologous T cells. In all 4 patients investigated, there was a reduction in stimulatory activity of cells prepared from the tacrolimus-treated site compared with the untreated site varying from 64% to 90% (Fig 4).

In each biopsy specimen taken from inflammatory skin lesions, IDECs comprised a certain percentage of all epidermal DCs (see above). We have shown herein that treatment with tacrolimus is associated with a reduction of this IDEC percentage. The patient with the most striking clinical improvement (patient No. 107) showed the highest reduction of stimulatory capacity (90%) and the highest reduction of the IDEC percentage (97% to 31%). The patient who experienced a brief tacrolimus treatment of 2 days (patient No. 108) with visible improvement, mostly of erythema, had the lowest stimulatory capacity reduction (64%), suggesting that this effect may be related to treatment duration. Furthermore, he showed a slightly higher IDEC percentage in the treated lesion compared with the untreated lesion. However, his lesions were slightly different in size and severity, and he had (like most of our patients) chosen the worst lesion for treatment. The 2 rebound patients (patients No. 114 and 118) had a reduction of their stimulatory capacity from 88% and 80% and a reduction in their IDEC percentage

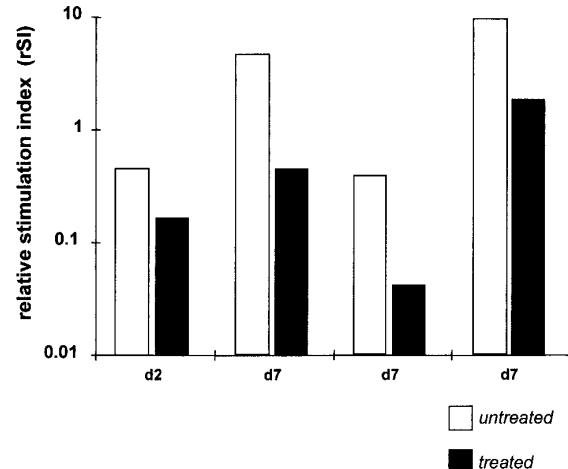


FIG 4. Reduction in the capacity of epidermal DCs to stimulate autologous T cells. The stimulatory capacity of epidermal cell suspensions, as prepared from untreated and tacrolimus-treated AD lesions of 4 individual patients, toward their autologous T cells is shown. There is considerable variation between the patients, but all of them show a reduction of the stimulatory capacity, which is already detectable at the second day of treatment and appears to be stronger at day 7. *rSI*, Relative stimulation indices.

TABLE II. Alterations of the inflammatory infiltrate in the dermis induced by topical tacrolimus treatment

	Uninvolved skin	Involved untreated skin	Treated skin
CD1a	10.7 ± 4.16	10.7 ± 1.6	16.3 ± 2.02*
CD11b	13.67 ± 1.6	20.67 ± 5.03*	12.3 ± 1.5
CD36	3.67 ± 0.58	21.3 ± 2.53*	13.3 ± 2.0

Biopsy specimens were taken from uninvolved, involved and untreated, and involved and treated skin areas (patient No. 114). Cryosections were stained for CD1a, CD11b, and CD36 and scored as described in the "Methods" section. Results are given as positive cells per square micrometer area section.

**P* < .05, significantly different from both other values.

of 64% to 49% and of 22% to 16%, respectively, in spite of ongoing flare-ups.

All patients treated with tacrolimus exhibited a reduction of their epidermal DC stimulatory activity toward their autologous T cells that was essentially independent of their clinical course but corresponded to the composition of the epidermal DC pool of the respective lesion.

DISCUSSION

This study was designed to investigate the effects of topical tacrolimus treatment on the epidermal DC populations in lesional AD skin with respect to the clinical improvement of the lesions. We have demonstrated that the clinical improvement was accompanied by an overall decrease of CD1a⁺ DCs in the epidermal compartment to within the normal range of about 2%. An analysis of the relative proportion of IDECs and LCs revealed that this

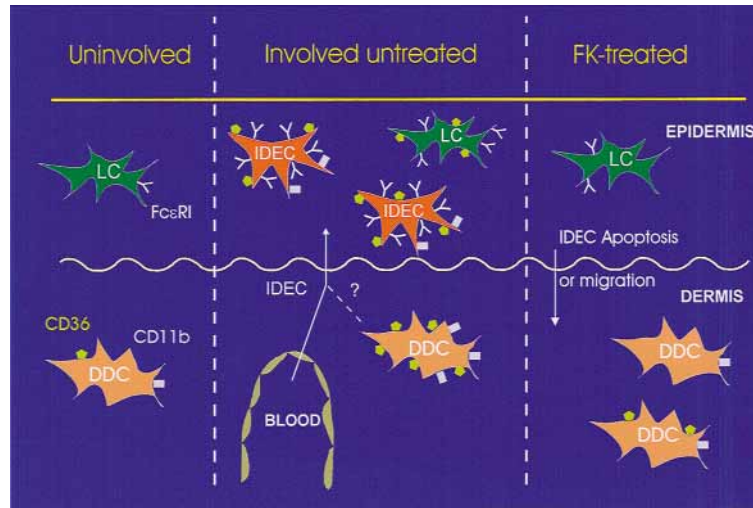


FIG 5. Distribution and phenotype of DCs in the epidermis and dermis in uninvolved skin, involved but untreated skin, and tacrolimus-treated atopic skin. *FK*, Topical tacrolimus; *DDC*, dermal dendritic cell.

decrease was largely the result of a decrease in IDECs. In addition, tacrolimus induced quantitative changes in receptor expression of both CD36 and FcεRI, as well as functional alterations of the epidermal DCs.

On the basis of clinical and histopathologic evidence, AD should be classified as a delayed-type hypersensitivity reaction. In this type of IgE-mediated reaction, the role of antigen-presenting cells and T cells seems to be critical,^{2,3,23} and FcεRI α -expressing DCs apparently represent the pivotal element.¹² Whereas normal LCs express low amounts of FcεRI,¹³⁻¹⁵ the expression is strongly and specifically increased in AD.²¹ Moreover, phenotypic and functional alterations of the epidermal CD1a⁺ DCs of AD skin have been demonstrated,^{16,18-20} leading to the identification of 2 distinct CD1a⁺ epidermal DC populations (ie, LCs and IDECs). Consequently, FcεRI-expressing epidermal DCs represent potential targets for new therapeutic approaches, possibly based on drug interference with the receptor itself or the stimulatory function of these antigen-presenting cells.¹² To test this hypothesis, we examined the effect of topical tacrolimus treatment on cell numbers, immunophenotype, and function of the CD1a⁺ cell populations in AD skin. Our findings showed that tacrolimus leads to qualitative, quantitative, and functional alterations of the epidermal CD1a⁺ cell infiltrate.

From a clinical point of view, tacrolimus led to a rapid improvement of the AD skin lesions. This clinical improvement was accompanied by (1) a decrease in the IDEC percentage from all CD1a⁺ cells in the inflammatory infiltrate and (2) a decrease in CD1a⁺ cells in the epidermal compartment to within the normal range of about 2%. Taken together, these changes indicate a depletion of IDECs from the epidermal compartment attributable to either a migration of IDECs out of the epidermal compartment or to an in situ cell death. The exact nature of this depletion remains to be determined.

Another important alteration from topical tacrolimus was the reduction of FcεRI expression on both epidermal CD1a⁺ DC populations. This effect might be one of the clinically relevant effects of tacrolimus at a cellular level because (1) the upregulation of FcεRI is highly disease specific²¹ and (2) the FcεRI-dependent antigen capture and subsequent presentation of aeroallergens is assumed to be a central pathogenetic event in AD.^{12,24} In vitro studies are currently in progress to further investigate the mechanisms controlling receptor expression on DCs and the extent to which tacrolimus contributes to this phenomenon. Preliminary data suggest a direct downregulation of FcεRI expression rather than an indirect effect mediated through soluble or membrane-bound signals from other types of cells in the inflammatory infiltrate (Panhans-Groß et al, unpublished data).

The expression of CD36 on CD1a⁺ cells has previously been shown to be related to the inflammatory activity inside the epidermal compartment.²¹ The presence of CD36 indicates that IFN- γ , which is known to induce this molecule,²⁵ acts in the epidermis of AD lesions. The presence of IFN- γ has been linked to the clinical course of the disease.²⁶ As expected from the clinical improvement seen in tacrolimus-treated patients, CD36 expression in epidermal CD1a⁺ cells was downregulated. This was not only due to the depletion of IDECs and the consequent loss of CD36 expression in lesional epidermis but to a downregulation of CD36 in both the LC and IDEC populations. Topical tacrolimus inhibits the effluent phase of allergic contact dermatitis in animals and human subjects,^{27,28} and there is increasing evidence that the stimulatory capacity of antigen-presenting cells is altered. Because epidermal CD1a⁺ DCs exert a strong stimulatory activity toward autologous T cells,²⁰ the dramatic decrease in the stimulatory activity of epidermal CD1a⁺ DCs on topical tacrolimus reported here is critical for understanding how this compound acts on the

skin's immune system. This may be in part because of the decrease of the total CD1a⁺ cells in the epidermis (ie, the disappearance of the hyperstimulatory fraction of these DCs; Fig 5).²⁰ However, this effect was seen after only 2 days of treatment. Similar to cyclosporin A,^{29,30} tacrolimus seems to impair the stimulatory capacity of LCs and related DCs by mechanisms that involve the modulation of costimulatory molecules.³¹

In conclusion, the clinical improvement of AD lesions treated with tacrolimus ointment was accompanied by a reduction in the stimulatory activity of epidermal DCs, phenotypic changes in both the LC and IDEC populations of the epidermal DC infiltrate, and a decrease of the IDEC population within the pool of CD1a⁺ epidermal DCs. Thus the present study suggests that epidermal antigen-presenting DCs represent a target for topical tacrolimus in AD.

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