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Topical Calcipotriol for Psoriasis – An immunohistologic study

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The aim of the present study was to investigate the distribution of Langerhans cells and T cells in the lesions and also the phenotypic expression of markers of activation on lesional T cells and keratinocytes, before and after 2 weeks of topical treatment of 7 psoriatic patients with calcipotriol. Before treatment, the infiltrate was composed mainly of T cells and there was decreased expression of CD1 on the intra-epidermal Langerhans cells. ICAM-1 and EGF receptor were present throughout the epidermis, but keratinocytes expressing Transferrin receptor were detected only in the basal layer. After 14 days of calcipotriol therapy, there were significantly fewer CD4T cells in the dermis and an increased number of intra-epidermal CD1 + Langerhans cells. ICAM-1 expression on lesional keratinocytes was reduced in all patients, but the expression of EGF receptor was decreased in 3 patients only, and Transferrin receptor expression on keratinocytes had not changed. All these changes were concurrent with moderate clinical improvement of the lesions. The results suggest that in the early stages of the clinical response to calcipotriol there is an immunomodulating effect of the drug associated with variable decreases in keratinocyte expression of markers of activation.

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The clinical antipsoriatic efficacy of Calcipotriol (C) is well established (1, 2), but its mode of action is not. C has antiproliferative effects and induces differentiation in cultured keratinocytes (Ks) (3, 4). Immunomodulating effects of C have also been demonstrated. Like calcitriol (the natural bioactive form of Vitamin D3), C inhibits the thymocyte proliferative response to IL1 (5) and it is conceivable that C has analogous inhibitory effects on T Lymphocyte (TL) proliferation, on IL2 production, on Transferrin-receptor (TFr) expression.

The aim of the present immunohistologic study was to determine the distribution of lesional TL and Langerhans cells (LCs) and the phenotypic expression of markers of activation on lesional TL and Ks, before and during topical C treatment, to obtain additional informations pertinent to the mechanism of action of C in Psoriasis.

PATIENTS AND METHODS

Seven psoriatic patients (36 to 50 years) were treated with C ointment (50 μg/g) (Formenti, Milan, Italy) twice daily for 6 weeks. The clinical severity of psoriasis was assessed by PASI scores, evaluated before and every 2 weeks during therapy, and analysed by Student's *t*-test.

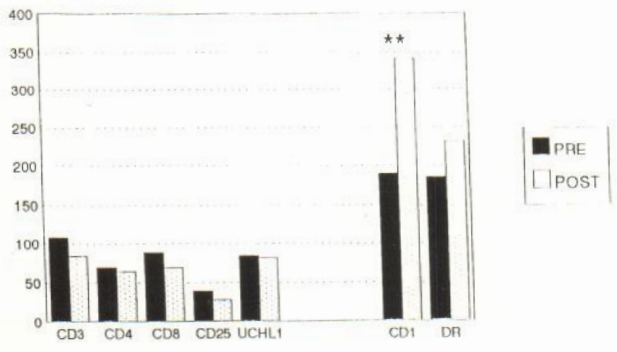
Punch biopsies, taken from patients before and after 2 weeks of treatment were embedded in OCT compound, snap frozen and serial cryostat sections prepared. A panel of monoclonal antibodies (MoAbs) was used to detect TL (Leu4), T cell subpopulations (Leu3a and Leu2a), memory TL (UCHL1) and LCs (OKT6 and HLA-DR). MoAbs against HLA-DR, TFr and IL2r (TAC) were markers of activated TL. To assess metabolic activation of Ks, MoAbs detecting Epidermal Growth Factor-receptor (EGFr), TFr and ICAM-1 were used.

A standard immunohistochemical technique, using APAAP (6), was applied to pathologic and normal skin samples. The APAAP processed slides were viewed with an ocular square grid and the absolute numbers of positive staining cells for each MoAb were counted. Twenty adjacent grid fields of epidermal sections, representing a surface area of 1.68 mm^2, were examined. Positive cells in papillary and reticular dermis were counted as described for epidermis. K expression of markers of activation was evaluated in 4 normal controls and psoriatic patients. The intensity of immunostaining (absent, faint, moderate, strong) and the extension of the epidermal distribution were assessed.

RESULTS

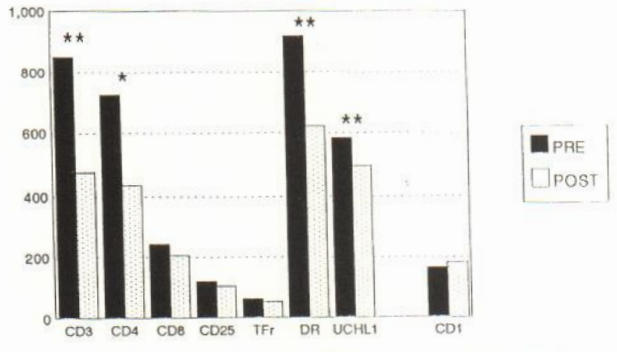
C improved psoriasis in all patients after 2 weeks of therapy, as confirmed by a significant decrease in the PASI score (mean values, 9.8 ± 4.0 before vs. 4.6 ± 1.6 after therapy, $p < 0.05$).

The pretreatment immunohistologic study, showed a cellular infiltrate consisting mainly of primed/memory TL (CD3+, UCHL1+), with a prevalence of CD4T cells in the dermis.



**, p< 0.01 PRE, before C therapy POST, after C therapy

Fig. 1. Total number of positively staining cells in 1.68 mm^2 of continuous psoriatic epidermis (mean values).



*, p< 0.02 **,p<0.01 PRE, before C therapy POST, after C therapy

Fig. 2. Total number of positively staining cells in 1.68 mm^2 of continuous psoriatic dermis (mean values).

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Infiltrating TL expressed markers of activation. There was strong positivity for HLA-DR in dermal TL, and a small number of TL were positive for IL2r and for TFr (Figs 1 and 2). Intra-epidermal CD1 + LCs were fewer and irregularly distributed. After treatment, there was a decrease in dermal memory and CD3TL: Only the dermal CD4TL were decreased, with no changes in the CD8T cell subset. There were no changes in the numbers of IL2r + cells and TFr + cells, but there were fewer dermal HLA-DR + cells. In the epidermis we observed normalization of the distribution of LCs, associated with increases in CD1 + LCs (Figs 1 and 2).

Before treatment, ICAM-1 and EGFr expression in the Ks of the lesional epidermis was moderate in the basal layer, and in the upper squamous layers was faint. Immunoreactivity for TFr was similar to that observed in normal skin, being limited to basal layer. After C treatment, the expression of ICAM-1 was reduced in intensity and extent in all patients, whereas decreased expression of EGRr on Ks was observed in only 3 patients. No significant changes were observed in the expression of TFr on Ks.

DISCUSSION

The results of the present study indicate that in the early stages of the clinical response to C there is an immunomodulating effect of the drug on cutaneous immunocytes, associated with variable decrease in K expression of markers of activation.

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