Acta Derm Venereol (Stockh) 1994; Suppl. 186: 49–51

Structural Alterations of Basal Keratinocytes and Capillary Loop in Psoriasis during Treatment with Topical Calcipotriol

G. M. PALLESCHI, A. GENTILI, M. CAPRONI, A. GIACOMELLI, D. FALCOS and P. FABBRI

Department of Dermatology, Florence, Italy

Recent research has demonstrated the activity of calcipotriol, effective as a potent inhibitor of cellular proliferation and known to increase differentiation in a number of cell lines in the topical treatment of psoriasis. Vit D3 receptors are expressed in keratinocytes and vascular endothelial cells. We studied the alterations in basal kertinocytes (stem cells or anchoring cells) and endothelial cell modification in 6 patients with psoriasis treated with calcipotriol ointment twice a day for 4 weeks. The samples were embedded in Epon resin for thin section and ultrathin section examination by electron microscopy. A normal pattern of distribution of the two different types of basal keratinocytes was observed before treatment. After treatment, only anchoring cells were detected. The alterations of endothelial cells in capillary loop disappeared after treatment, presenting normal aspects. Our morphological findings suggest that calcipotriol is therapeutically effective, due principally to an inhibition of cellular proliferation. ***Key words: calcipotriol; stem cells; serrated cells; non-serrated cells; psoriasis.***

Acta Derm Venereol (Stockh) 1994? Suppl. 186: 49–51.

G. M. Palleschi, Dept of Dermatology, Via Alfani 37, 50121 Florence, Italy.

Calcipotriol (MC 903), a new synthetic analogue of Vitamin D, was recently introduced in the topical treatment of psoriasis. This drug has a high affinity for the vitamin D receptor expressed in the skin by keratinocytes, fibroblasts, endothelial cells, monocytes, activated T-lymphocytes and B-lymphocytes.

In vivo, calcipotriol induces a significant reduction in epidermal proliferation and increased maturation of keratinocytes, and it modifies the inflammatory infiltrate by decreasing the numbers of polymorphonuclear leukocytes and, later, T-lymphocytes. These effects are probably due to an interference with some immunological mediators (IL-1, IL-2, IL-6) as observed in vitro (1) (2). Many studies suggest that this is achieved, after binding of the specific cytosolic receptor, both through transportation to the nucleus and transcription of those genes involved in hormonal response as steroids do, and through a more immediate mechanism not mediated via the genoma, which directly increases intracellular free calcium (3).

In normal skin there are two cell populations of keratinocytes in the basal layer: (i) stem cells with non-serrated morphology, generally localized at the tips of the deep rete ridges, and (ii) anchoring cells with serrated morphology, distributed mainly along the sides of rete ridges (4) (5).

In psoriasis, epidermal hyperproliferation involves not only the stem cells of the basal layer, but also the suprabasal population named transient amplifying cells.

Epidermal hyperplasia cannot occur without vascular proliferation. In psoriasis, capillary loops of the dermal papillae are abnormally dilated and convoluted, and they display the characteristics of venous capillaries (6).

The aim of this study was to evaluate the effect of calcipotriol in psoriasis, by studying the morphology of the basal layer and the capillary loops.

MATERIAL AND METHODS

Biopsy specimens were taken from 6 patients with nummular psoriasis before and after treatment with calcipotriol ointment twice a day for 4 weeks.

The samples were fixed by immersion in 2% formaldehyde and 2.5% glutaraldehyde 0.1 M cacodylate buffer, pH 7.4, followed by 1% OsO_4 in 0.1 M phosphate buffer, pH 7.4, dehydrated in graded ethanol, passed through propylene oxide and embedded in Epon 812. Semithin sections were stained with methylene-blue and fuchsin for light micros-

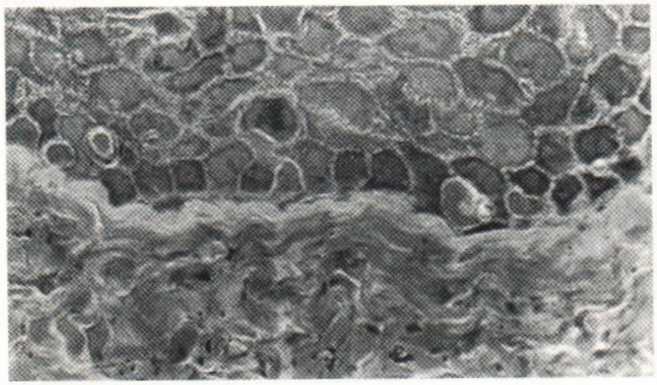


Fig. 1. Normal distribution of stem cells, localized at the tips of the deep rete ridges in the basal layer, before treatment. Semithin section, 2 μm, stained with methylene-blue and fuchsin (×1000).

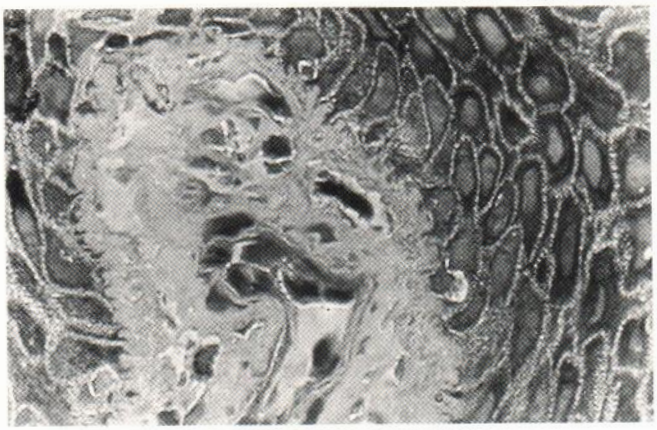
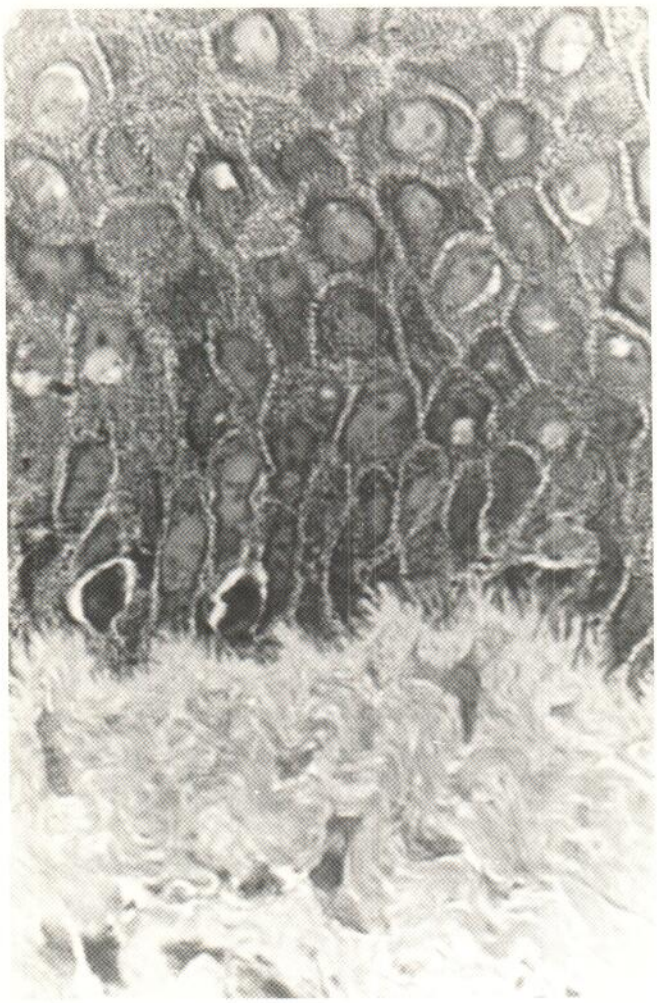
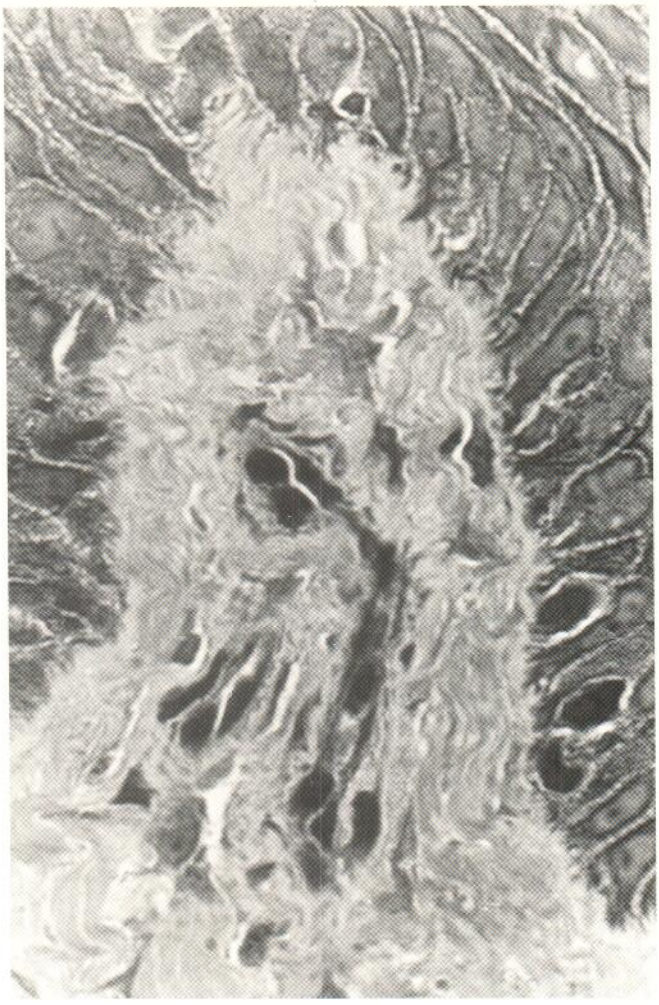


Fig. 2. Normal distribution of anchoring cells localized along the sides of rete ridges in the basal layer, before treatment, Semithin section, 2 μm, stained with methylene-blue and fuchsin (×1000).

© 1994 Scandinavian University Press. ISSN 0365-8341





Figs 3, 4. After treatment with MC 903 the basal layer is composed only of serrated cells with amplified, highly convoluted dermal epidermal junction. Semithin section 2 μm, stained with methylene-blue and fuchsin (×1000).

copy. The samples were also cut in ultrathin sections, stained with uranyl acetate and bismuth tartrate and examined in a Siemens Elmiskop 102 electron microscope at 80 kW.

RESULTS

Examination by light microscopy showed a normal distribution of stem cells (Fig. 1) and of anchoring cells (Fig. 2) in all samples before treatment. After treatment with MC 903, we found only serrated cells with amplified highly convoluted dermal epidermal junctions (Figs 3, 4). These data were supported by the electron microscopic observations (Figs 5, 6) which, after therapy, showed an elongation of cytoplasmic projections of serrated cells extending deeply into the papillary dermis (Fig. 6) and an absence of non-serrated cells.

Before treatment in all samples, the capillary loop appeared typically dilated and tortuous, with endothelial swelling and nuclei protruding into the lumen. Electronmiscroscopic observation showed that the intrapapillary portion of the capillary loop had a venular-type multilaminated basement membrane. Furthermore, the endothelial cells presented a well defined rough endoplasmic reticulum. After treatment, light microscopy demonstrated a normal capillary loop. Both endothelial swelling and capillary lumen were reduced. Electronmicroscopic study demonstrated a return to arterial capillary features with the basement membrane characterized by a single electron-dense band and many cytoplasmatic free ribosomes.

Fig. 5. Transmission electronmicrography showing the non-serrated basal keratinocytes with the regular dermal-epidermal junction with short villi-like projections (×7,500).

Fig. 6. Transmission electronmicrography showing the serrated basal keratinocytes with the elongation of cytoplasmic projections of serrated cells extending deeply into the papillary dermis after treatment (×7,500).

DISCUSSION

Our data suggest that this drug has a therapeutic effect, due principally to an inhibition of cellular proliferation, supported by the disappearance of keratinocytes with a non-serrated morphology in the basal layer. Previous studies (4, 5, 7) have correlated basal layer cell morphology to its function, evidencing a stem activity for non-serrated cells that gives rise to suprabasally located, highly proliferative transient amplifying cells. In psoriasis the hyperplasia is supported by these cells. The other basal layer cells presenting a serrated morphology are post-mitotic cells, presumably with an anchoring function. The basal layer is composed mainly of serrated cells, which indicates that calcipotriol is effective in inhibiting cell proliferation and inducing cell differentiation. The normalization of the capillary loop could be correlated with the reduction in skin thickness or directly with an antiproliferative action of calcipotriol.

REFERENCES

1. Kragballe K, Beck H-J, Søgaard H. Improvement of psoriasis by a topical vitamin D3 analogue (MC 903) in a double-blind study. Br J Dermatol 1988; 119: 223–230.
2. Kragballe K, Wildfang I-L. Calcipotriol (MC 903) a novel vitamin analogue stimulates terminal differentiation and inhibits proliferation of cultured human keratinocytes. Arch Derm Res 1990; 282: 164–167.
3. Gerritsen MJP, Rulo HFC, Van Vlijmen-Willems I, et al. Topical treatment of psoriatic plaques with 1,25-dihydroxyvitamin D3: a cell biological study. Br J Dermatol 1993; 128: 666–673.
4. Lavker RM, Sun TT. Epidermal stem cells. J Invest Dermatol 1983; 81: 123 s.
5. Montagna W, Kligman AM, Carlisle KS. Epidermis. In: Atlas of normal human skin. New York: Springer-Verlag, 1992: 26–30.
6. Braverman IM, Yen A. Ultrastructure of the capillary loops in the dermal papillae of psoriasis. J Invest Dermatol 1977; 68: 53–60.
7. Zampetti M, Fattorossi A, Calvieri, Grico T. Basal keratinocyte subsets: ultrastructural and morphometric features. Acta Derm Venereol (Stockh) 1989; 69: 59–62.