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THE EPIDERMAL CELL PROLIFERATION IN LICHEN PLANUS

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Abstract. The proliferative activity of epidermal structures was determined in 6 patients suffering from lichen planus (l.p.). By using the in vitro ^{3}H-thymidine labelling technique, semithin Epon sections of lesional and perilesional uninvolved skin were compared autoradiographically with normal controls. The following results were obtained.

The labelling index of epidermal cells was significantly increased within the l.p. papules. The mean values ranged from 41% in acanthotic l.p. to 23% in atrophic l.p., whereas in adjacent unaffected skin (15% and 11% respectively) as well as in normal controls (6%) a lower ^{3}H-index was observed.

As regards damage to basal cells within l.p. lesions, a factor—the relation between the number of basal cells in involved and uninvolved skin in reference to the epidermal length—was calculated in order to correct the autoradiographic results in involved skin. The differences between l.p. papules. adiacent skin and controls remained statistically significant even after correction.

The intradermal eccrine ducts revealed a significantly increased labelling index in l.p. lesions as compared with perilesional and normal skin. In this respect no difference could be observed between the acanthotic and the atrophic variant of l.p.

These findings indicate that replacement of damaged basal cells in l.p. is achieved by an increase in actively dividing keratinocytes in both epidermis and skin appendages within the lesion.

Key words: Lichen planus; In vitro labelling technique; Increased mitotic activity

As early as 1925, Kyrle considered damage to basal cells to be the primary event in lichen planus (l.p.). In recent years this assumption has been supported by modern evidence and is now generally accepted (1, 2, 4, 13). This alteration of the generative compartment of the epidermis is followed, in the opinion of various authors, by a cascade of reactions—depression of epidermal turnover, diminution or enlargement of prickle cells, hypergranulosis and hyperkeratosis—resulting in the characteristic histological picture of l.p. (1,6,7,10,11).

In order to gain further insight into the histogenesis of epidermal changes seen in this disease, autoradiographic investigations using the in vivo (9) and in vitro labelling technique (12,14) were also performed. Marks et al. (9) and Presbury & Marks (12) observed labelled, spindle-shaped keratinocytes migrating from the margins to the centre of l.p. lesions and thereby deduced a repopulation of the eroded l.p. areas by keratinocytes from the non-affected neighbourhood. Thus their results fitted into the framework of the concept described above.

We have performed studies using an in vitro technique with ^{3}H-thymidine in order to investigate the proliferative activity of the epidermis in involved and uninvolved (perilesional) skin of l.p.-patients in relationship to normal human skin.

METHODS

After local anaesthesia, punch biopsies were taken from involved as well as clinically unaffected, adjacent skin of 6 patients with generalized l.p. Normal skin was obtained under the same conditions from 2 control persons. Immediately after excision the tissue was sliced into small blocks and incubated in Trowell's T8 tissue culture medium, to which 20 μCi/ml ^{3}H-thymidine (Radiochemical Centre, Amersham, 5 mCi/ml) had been added. After an incubation time of one hour at 37°C the material was fixed in 3.5% glutaraldehyde for 2 hours and post-fixed in osmium tetroxide (Palade). Following dehydration in ethanol the blocks were embedded in Epon 812. Semithin sections were cut on a Reichert OMU 2 ultramicrotome, mounted on glass slides and coated with Kodak NTB 2

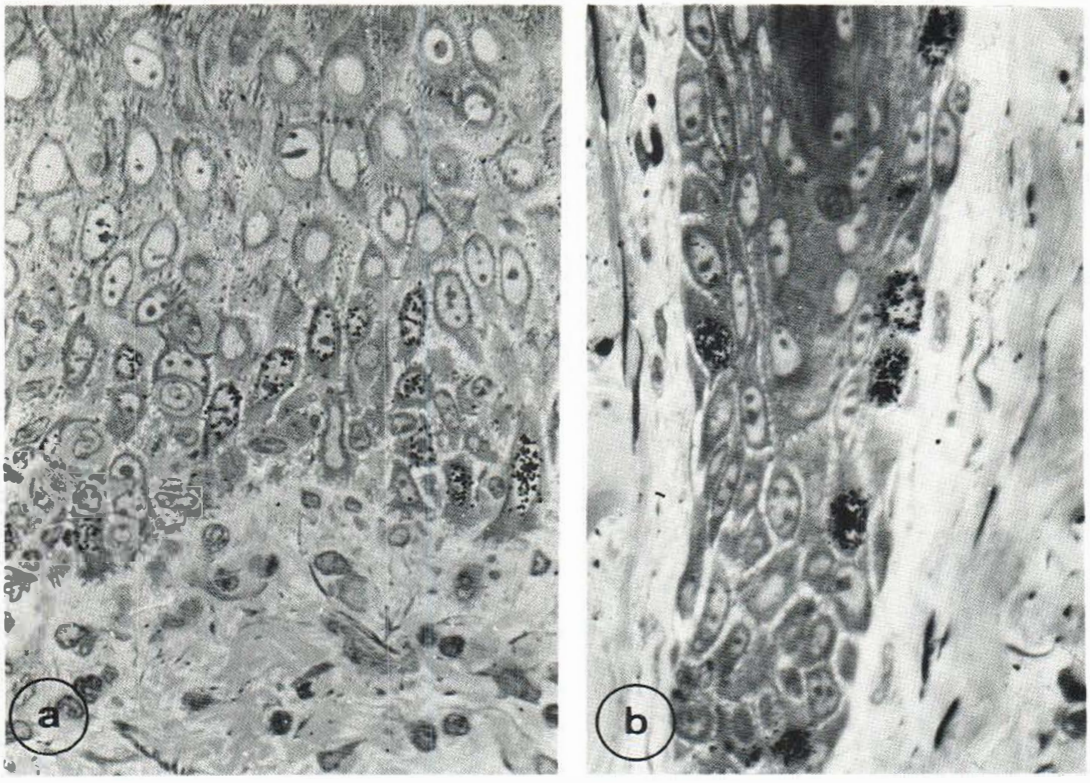

Fig. 1. (a) Extremely intense ^{3}H-thymidine labelling of basal and suprabasal keratinocytes in acanthotic l.p. Autoradiograph of semithin Epon section, toluidine blue staining. ×630. (*b*) Several labelled cells within the intradermal eccrine duct beneath the l.p. lesion. Autoradiograph of semithin Epon section, toluidine blue staining. ×650.

emulsion (dilution 1 : 1.5, dipping technique). Then they were allowed to dry in a vertical position for 5 min and stored in light-tight boxes at −20°C. After an exposure time of 3 to 4 weeks they were developed in Kodak D 19B, rinsed in water and fixed with Fixon Ultra Rapid. After careful washing in water they were stained with toluidine blue and examined in a light microscope. Labelled cells in the basal and lower squamous cell layer were counted in the lesions, in unchanged adjacent skin and in normal controls and correlated with the total number of basal cells in the corresponding areas. Since a decreased number of intact basal cells per length unit was found within the lesions, as compared with unaffected skin or controls, a factor was calculated to correct the counts. The statistical significance of the results was checked by the *z*-test (Fischer).

RESULTS

In all 6 cases of l.p. investigated, a great number of labelled basal and suprabasal cells could be observed (Fig. 1*a*). The labelling indices were determined by counting the number of labelled basal and suprabasal cells per 1 000 basal cells. The mean labelling index in the lesional skin of 5 patients suffering from "acanthotic" l.p. was 41%; in a case of "atrophic" l.p. this value was 23% (Fig. 2). In contrast, the labelling indices of the adjacent, uninvolved epidermis were 15% in acanthotic l.p. and 11% in atrophic l.p. respectively. The difference between involved and uninvolved skin was statistically significant. The mean labelling index in the control group was 5%. Because of the so-called liquefaction degeneration of basal cells in l.p. the ratio of labelled cells counted in basal and suprabasal localization and the number of intact basal cells is altered when involved and uninvolved skin are compared. In order to avoid this source of error the number of basal cells per length unit was determined in the affected and adjacent unaffected skin. In this way a factor (N) was elaborated to correct the labelling values in the lesional skin of l.p.

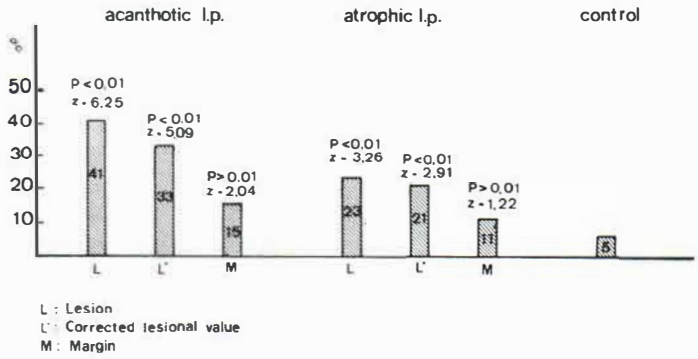

Fig. 2. Epidermal labelling index in l.p.

$$N = \frac{\text{Number of basal cells per length unit area involved skin}}{\text{Number of basal cells per length unit area uninvolved skin}}$$

Even after applying this correction the l.p. papules exhibited a significantly higher labelling index than the unaffected marginal and normal skin (Fig. 2).

The intradermal, eccrine ducts also revealed a significantly increased labelling index in l.p. lesions when compared with adjacent uninvolved and normal skin (Fig. 1*b*). In this respect no difference between the acanthotic and atrophic variant of l.p. could be found (Fig. 3).

DISCUSSION

The present investigations show the ^{3}H-thymidine labelling index in l.p. to be significantly higher within the lesions than in the clinically unaffected adjacent epidermis. Nevertheless, even in the perilesional skin an increased proliferative activity can be observed when compared with normal human skin.

These findings do not fit in with the concept suggested by Marks et al. (9) and Presbury & Marks (12) that the eroded l.p. areas are repopulated by keratinocytes originating from the margins of the lesions. In agreement with Walker & Dolby (14) we too were unable to observe spindle-shaped, fusiform keratinocytes as described by Marks et al. (**9**). The different embedding technique used in the present study could be responsible for this fact.

Like Marks et al. (9), we too found a high labelling index in the dermal eccrine ducts. According to Lobitz et al. (8) and Christophers & Braun-Falco (3) this phenomenon occurs during the healing phase after injury to the epidermis and is not due to an inflammatory process (8).

Our results indicate that the response of basal cell damage in l.p., the origin of which is not known to date, is a massive increase in actively dividing keratinocytes in the epidermis. In skin lesions of l.p., the eccrine ducts also seem to play an important role in the replacement of necrotic basal cells. Perhaps this mechanism can explain the fact that l.p. lesions localized on the skin have a better prognosis than those occurring on the mucous membranes. In this connection it should be emphasised that not all cells labelled by ^{3}H-thymidine complete mitosis and undergo cell division. Therefore, the possibility of a disturbance of

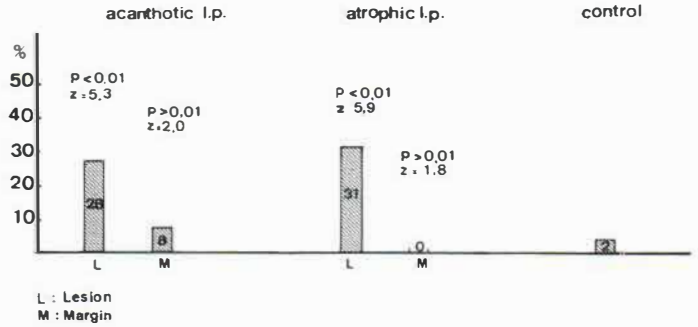

Fig. 3. Labelling index of eccrine ducts in l.p.

the epidermal mitotic cycle activity in l.p.—as pointed out by El-Labban & Kramer (5) on the basis of electron microscopical investigations—has yet to be excluded.

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