Lack of Lysozyme, $\alpha_1$-Antitrypsin and $\alpha_1$-Antichymotrypsin in Normal Langerhans' Cells: Differentiation from Cells of the Monocyte-Histiocyte-Macrophage Series

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Abstract. Immunohistochemical studies for the presence of lysozyme, $\alpha_1$-antitrypsin and $\alpha_1$-antichymotrypsin in Langerhans' cells were carried out in situ on paraffin sections of normal skin. These cell markers which are routinely employed in the histological demonstration of cells of the monocyte-histiocyte-macrophage series were found to be absent from normal Langerhans' cells.

The Langerhans' cell is assuming increasing importance in the pathogenesis of many cutaneous diseases, for example in contact dermatitis (10) where it is considered to be the "epidermal macrophage" processing antigen for presentation to the T lymphocyte, and in mycosis fungoides where the population of epidermal Langerhans' cells appears to be increased in active disease (2, 7) and decreased by PUVA therapy (2).

At present, the histochemical properties (1, 14) and immunological properties (9, 11, 12) of the Langerhans' cell are frequently used for their identification. Many of these properties appear to be shared by cells of the mononuclear phagocyte (MNP) series (13).

Lysozyme, $\alpha_1$-antitrypsin (AT) and $\alpha_1$-antichymotrypsin (ACT) have been successfully employed as histiocyte markers in dermatopathology (4, 5). Langerhans' cells have not been investigated for the presence of these markers, although reactivity has been found to be absent in interdigitating reticulum cells (8).

MATERIALS AND METHODS

Tissue

Biopsy samples of normal skin were divided into two parts, the first of which was fixed with buffered formal saline and routinely embedded in paraffin wax. The second piece was snap-frozen in liquid nitrogen and stored at $-70^\circ$C, for histochemical and immunohistochemical studies for the presence of Langerhans' cells. Biopsies of positive Kveim tests were also fixed in buffered formal saline for paraffin embedding to provide controls for the lysozyme, AT and ACT.

Methods

Identification of intracellular lysozyme, AT and ACT. These intracellular constituents of MNPs were localized on paraffin-embedded tissue using a peroxidase-antiperoxidase technique as previously described (5). The sections were initially incubated with the rabbit anti-lysozyme or rabbit anti-A T or rabbit anti-A CT. Subsequent application of swine anti-rabbit immunoglobulins and the rabbit peroxidase-antiperoxidase complex allowed the location of the peroxidase to be revealed using 3,3 diaminobenzidine and hydrogen peroxide. The sections were counterstained, dehydrated, cleaned with xylene and mounted in Depex for light microscopical examination.

Controls employed were replacement of the primary antibody by normal rabbit serum, the use of swine antimouse immunoglobulins as the secondary antibody, and incubation with 3,3 diaminobenzidine and hydrogen peroxide alone.

RESULTS

No positive staining for lysozyme, AT or ACT could be detected in the epidermis, but Kveim test biopsies, used as positive controls, were consistently labelled with all three antibodies.

DISCUSSION

Although Langerhans' cells are considered to be components of the mononuclear phagocyte system, we have demonstrated that they are negative for lysozyme, AT and ACT, even though these markers are easily identified in tissue histiocytes. This may reflect the relative lack of phagocytic function of epidermal Langerhans' cells.

Increased numbers of lysozyme, AT and ACT positive cells have been described in the epidermis.
of mycosis fungoides (MF) (6). Our findings suggest that these cells are not Langerhans' cells, which are known to be numerically increased in MF (2, 7), and that indeterminate cells which are related to Langerhans' cells also show absence of lysozyme, AT and ACT. It is possible, however, that the epidermal histiocytes identified in mycosis fungoides may be derived from Langerhans' cells and indeterminate cells by progressive acquisition of "histiocytic features". Immunoelectron microscopic study, simultaneously to label the membrane of Langerhans' cells with OKT6 (3) and the cytoplasm of histiocytes with lysozyme will help to resolve this question.

REFERENCES