Langerhans cells increase in the dermal lesions of adult T cell leukaemia in Japan

M SHAMOTO

From the Department of Pathology, Fujita-Gakuen University School of Medicine, Toyoake, Aichi, Japan

SUMMARY In cases of adult T cell leukaemia neoplastic T cell infiltration in the skin was accompanied by an increase in Langerhans cells. This is in keeping with the view that Langerhans cells may induce antigen-specific and allogenic T cell activation.

Langerhans cells were first discovered in the epidermis of the skin in 1868¹; subsequently these cells were also found in the lymph nodes,²⁻⁴ thymus,⁵ and dermis.⁶ Langerhans cell granules have also been observed in histiocytic cells of histiocytosis X^{7,8} which thus appears to be a pathological condition of Langerhans cells. It is generally recognised that Langerhans cells belong to the mononuclear phagocyte system⁹ and function as antigen presenting cells.¹⁰ It is thought that Langerhans cells could stimulate T lymphocytes, and this could be one of the reasons why Langerhans cells are markedly increased in the paracortical areas of dermatopathic lymphadenitis.^{2,3}

Recently, immunological examinations have confirmed that Langerhans cells bear Fc-IgG, C3 receptors,¹¹ express Ia¹² and the monoclonal antibody, OKT6.¹³ Takahashi *et al*¹⁴ have proved that the monospecific antibody which is formed against nervous system specific S-100 protein is positive in the interdigitating reticulum cells of the human lymph nodes, and S-100 protein is also positive in the epidermal Langerhans cells.¹⁵ Rausch *et al*³ have stressed a close relation between Langerhans and interdigitating reticulum cells. Morphologically and functionally, it is difficult to differentiate between these cells, except by the presence of granules in Langerhans cells.

Adult T cell leukaemia in Japan is a specific T cell leukaemia which frequently has skin involvement.¹⁶ In the dermis of adult T cell leukaemia relatively large and clear cells often mingle with neoplastic cells with markedly convoluted nuclei. We have clarified that these clear cells are Langerhans cells by the

peroxidase antiperoxidase (PAP) method using S-100 protein and by electron microscopy.

Material and methods

Ten skin biopsy specimens, the diagnosis of which was confirmed as adult T cell leukaemia were examined. For light microscopy, tissues of the skin were fixed in buffered formalin and carefully embedded in paraffin in such a way that the temperature did not rise above 60°C. The peroxidase antiperoxidase (PAP) method was used. Namely, paraffin sections were dewaxed and treated with 0.3% H₂O₂ in methanol (15 min) to block endogenous peroxidase activity, and were then rinsed in 70% ethanol. After washing with phosphate-buffered saline (PBS, pH 7.4), they were treated with normal swine serum (1/20, 30 min), and then washed with PBS, and incubated with monospecific anti S-100 protein antibody (20 µg/ml) overnight at 4°C. The sections were rinsed in PBS next day, and incubated with swine antirabbit IgG (Dako, 1/20, 30 min). They were rinsed in PBS and incubated with rabbit antihorseradish peroxidase complex (PAP) (Dako, 1/50, 30 min). After washing, an enzyme reaction was developed with a DAB-H₂O₂ solution according to Takahashi *et al*.¹⁴ Sections were counterstained with methyl green pyronin. Paraffin sections were also stained with haematoxylin and eosin for standard histological study.

Ultrastructurally, small pieces of the tissues were fixed in cold 2.5% glutaraldehyde and 2% paraformaldehyde buffered at pH 7.2 with a 0.05 M cacodylate buffer. They were then postfixed in 1% osmium tetroxide buffered with cacodylate. After dehydration in graded-series of ethanol, these were embedded in epoxy resin. Thin sections were cut with glass knives on an ultramicrotome, and stained with

Fig. 1 *There are a few S-100 protein-positive cells in the epidermis, and many in a Pautrier's microabscess (arrow) and in the dermis. × 250*

Fig. 2 *Many S-100 protein-positive cells mingle with leukaemic cells in the dermis. × 250*

uranyl acetate and lead compounds, and examined on a Jeolco 100 CX electron microscope.

Results

Cells resembling Sézary cells and/or the small cell variant form of Sézary cells (Sézary-type cells)

infiltrated the dermis, especially the upper dermis, although the extent and degree of the infiltration differed in each case. Pautrier's microabscesses were also characteristic in adult T cell leukaemia. Relatively large cells with clear and broad cytoplasm and irregularly shaped nuclei mingled with Sézary-type cells in the dermis. A few cells were positive for

Fig. 3 A *Langerhans cell* is seen in the dermis. $\times 4800$

Fig. 4 Several *Langerhans cell granules* are found in the cytoplasm. $\times 48\,000$

S-100 protein in the epidermis (Fig. 1). However, the ratio of the appearance of the S-100 protein positive cells was higher in the dermis than in the epidermis, although the number of the positive cells corresponded to that of the clear cells when stained with haematoxylin and eosin, but differed slightly according to case (Figs. 1 and 2). S-100 protein was devoid of the reaction products in some nuclei, and was positive only in the cytoplasm.

Ultrastructurally, a few Langerhans cells were observed in the epidermis, but in the dermis were often observed mixed with neoplastic cells (Fig. 3). The nuclei of the Langerhans cells were irregular in outline. The chromatin appeared finely dispersed. The broad cytoplasm contained Golgi apparatus, free ribosomes, and rough endoplasmic reticulum. Mitochondria were moderate in number. Filamentous structures were often found and Langerhans cell granules were characteristically observed in the cytoplasm (Fig. 4).

Discussion

In adult T cell leukaemia in Japan, it is one of the characteristic features that neoplastic T cells with markedly convoluted nuclei infiltrate the dermis.¹⁶ We have reported in an ultrastructural study¹⁷ that the greater part of the cells of adult T cell leukaemia is very similar to the small cell variant form of Sézary syndrome. In the present study, we have shown by the PAP method and electron microscopy that Langerhans cells also increase in the dermal lesions. Langerhans cells normally comprise only 4–5% of the epidermal cells and are rarely found in the dermis.¹⁸ In adult T cell leukaemia, an increase in the number of Langerhans cells was observed in the dermis, although not in the epidermis. Langerhans cells can be found in the lymph nodes of several other conditions.^{2–4} In particular, an extreme enlargement of the paracortical, thymus-dependent areas which is due to a large number of interdigitating reticulum cells and Langerhans cells is the main morphological feature of dermatopathic lymphadenitis.²³ It has been suggested that Langerhans cells may arrive in the paracortical areas of the lymph nodes from the skin, because Langerhans cells can be found in dermal lymphatics, in the marginal sinuses and in the small lymphatics of the lymph nodes. Langerhans cells and interdigitating reticulum cells closely connect with T lymphocytes in the lymph nodes. Stingl *et al*¹⁰ have suggested that Langerhans cells have a macrophage-like function as antigen-presenting cells and allogenic cells to T lymphocytes. The present evidence indicates a close relation between Langerhans cells and T lymphocytes. Loo *et al*¹⁹ have found C-type virus-like particles in the Langerhans

cells of skin biopsies and lymph nodes from patients with mycosis fungoides and Sézary's syndrome. It has been suggested that the aetiology of adult T cell leukaemia may be retrovirus particles with type C morphology.²⁰

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Requests for reprints to: Mikihiro Shamoto, Department of Pathology, Fujita-Gakuen University School of Medicine, Kutsukake-cho, Toyoake-city, Aichi-prefecture 470-11, Japan.