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GRANULES OF LANGERHANS CELLS IN LETTERER-SIWE'S DISEASE

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Abstract. Skin biopsies of two patients with Letterer-Siwe's disease were studied with the electron microscope. Langerhans cell granules were found in the infiltrating cells of the epidermis and corium. Beside typical Langerhans cell granules, some abnormally shaped granules were noticed. They showed thin, constricted tails, small bulges, large bulges containing glycogen particles, round double-membrane coated bodies, bent rods, and extruded granules. The finding of incomplete granules around the Golgi zone suggest a close relationship between these organelles. The presence of cells with structural evidence of activity in the infiltrates suggests a neoplastic nature of Letterer Siwe's disease.

Langerhans cells are dendritic cells showing a bright cytoplasm, characteristic granules, and lack of tonofilaments and desmosomes. Langerhans cell granules were first described in vitiliginous skin by Birbeck et al. (2) using ultrathin sections. Recently, identical granules have been described in inflammatory and neoplastic skin diseases (4, 6) as well as normal skin and mucous membrane of adults (3, 8, 10) and embryos (3). A granule is three-dimensionally of the shape of a disc or cup with a bulge (8, 11). In ultrathin sections, if hit at right angles to the granule surface, the bulge appears as a round bleb. The disc appears as a tail showing an outer single and an inner dotted membrane as well as a central band which is often double layered (8). In tangential sections, the disc has stripes at intervals of 90 Å either in one, or in two intersecting planes (2, 8). Another granule type has been found in normal adult human skin (14). These granules are round or oblong in shape with a diameter of about 100 to 200 nm and a dense matrix, occasionally containing grouped round bodies, and a triple-layered membrane. In previous publications Langerhans cell granules have been characterized by the above-mentioned structures, not only in normal, but also in diseased tissue, e.g. in Letterer-Siwe's disease.

Langerhans cell granules have been found in cytoplasm, lysosomes, or nuclei of infiltrating cells as well as in the extracellular space of the corium in Letterer-Siwe's disease (1, 6, 9, 12). As to the cytoplasmic figures, previous papers mention a similarity to macrophages (9) and giant cells of malignant lymphoma (5).

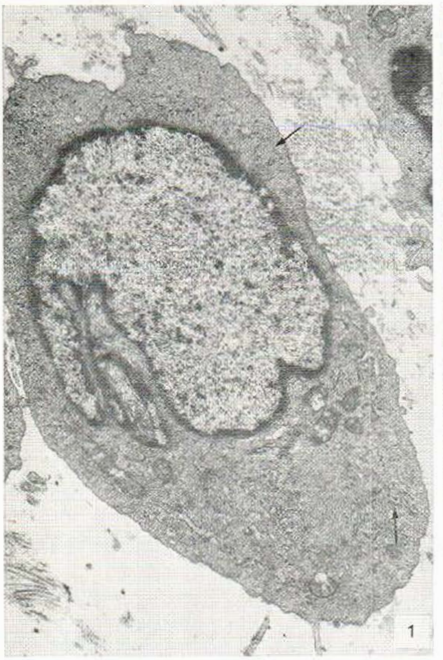
The present study revealed variations in number, distribution and ultrastructure of Langerhans cell granules in infiltrating cells of Letterer-Siwe's disease.

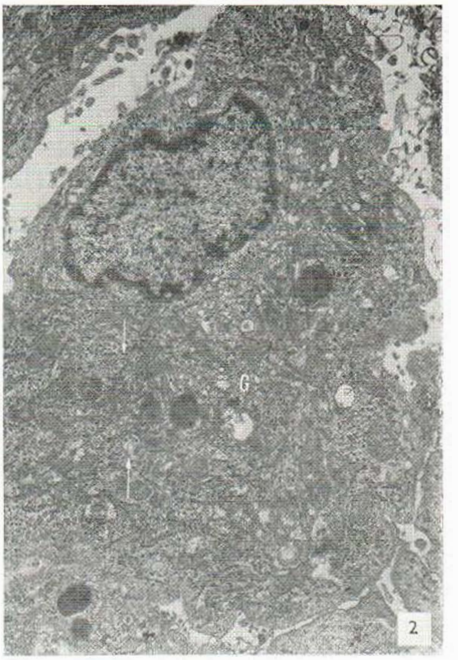
MATERIAL AND METHODS

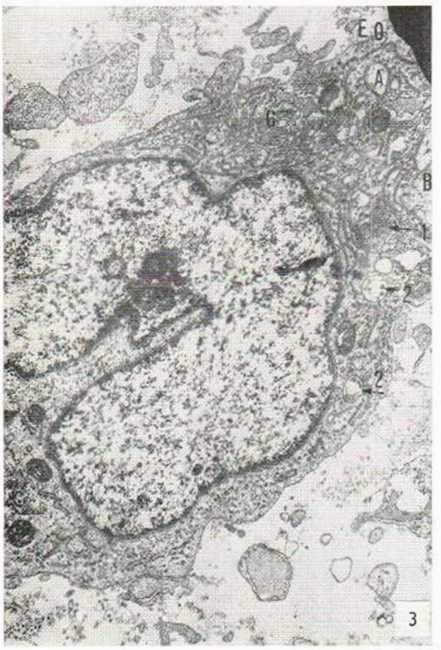
Two children suffering from Letterer-Siwe's disease were studied with the electron microscope. Biopsies were taken from reddish-yellowish papules of the upper abdomen and fixed in a 6% glutaraldehyde solution of Veronal acetate buffer, pH 7.2, with 7.5% sucrose at 4°C for about 3 hours. After glutaraldehyde fixation, the specimens were washed in the same buffer overnight in a refrigerator and fixed in 1% osmic acid solution of Veronal acetate buffer, pH 7.2, at 4°C for 1 hour. After dehydration in a series of alcohol solutions of increasing concentration, the specimens were embedded in Epon 812. Ultrathin sections were stained with uranyl acetate and lead citrate and studied with a Siemens electron microscope (Elmiskop 1A).

OBSERVATIONS

Both specimens showed pronounced infiltration of Langerhans cells in the epidermis as well as in the corium. The shapes of the infiltrating cells were various, with irregular cytoplasmic protru-







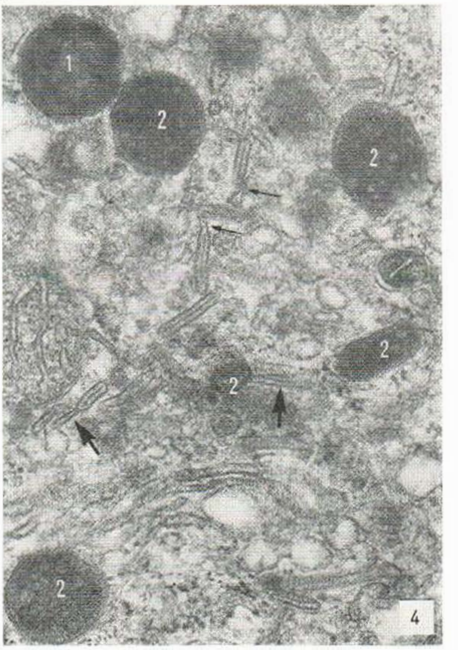
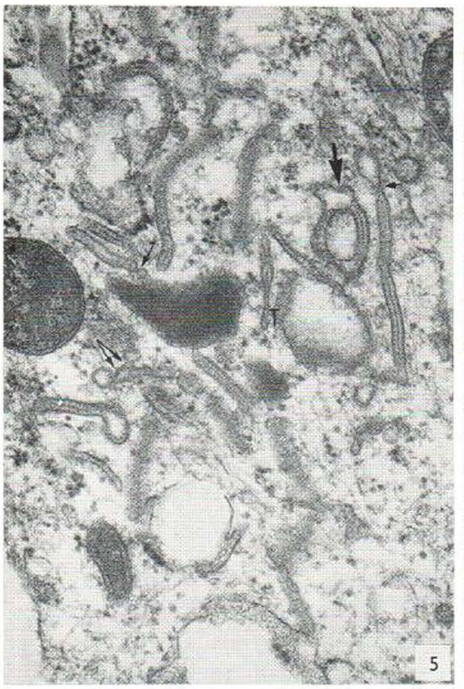


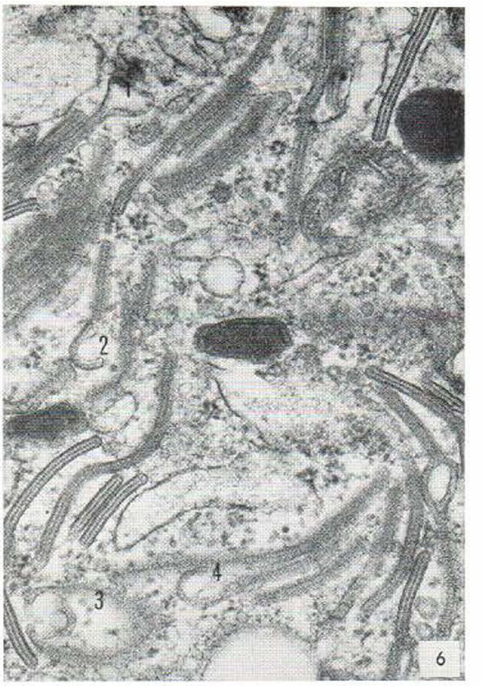
Fig. 1. A Langerhans cell with distinct cell-organelles in dark ground cytoplasm with a few characteristic granules (arrows). Notice cytoplasmic filaments and a peripheral, thick fibrous lamina of the indented nucleus. × 8 200.

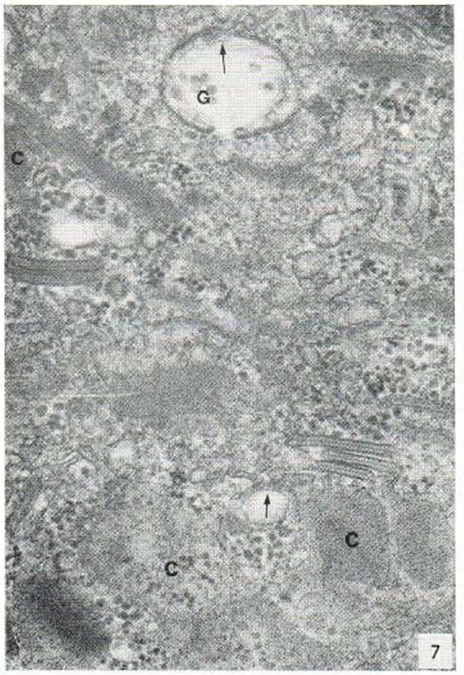
Fig. 2. Numerous normal granules appear intermingled with abnormal granules around the Golgi complex (*G*). Dense round bodies and distinct cell organelles are also seen. The area between two arrows appears in Fig. 10. × 8 200.

Fig. 3. Langerhans cell containing normal granules in bright cytoplasm. *A*, Circular tail. *B*, Branched tail. *O*, Opening for a granule to the extracellular space. *E*, Extracellular granules. *G*, Golgi complex. Arrow 1: Annular bodies within an enclosing membrane. Arrow 2: Round particles within membrane. × 8 200.

Fig. 4. Constriction of tails (thin arrows) and thin tails (thick arrows). The numbered dense round granules are so-called second type granules (Ref. 13) showing indistinct lamellae (1) and double membrane-bounded round bodies (2). × 35 200.







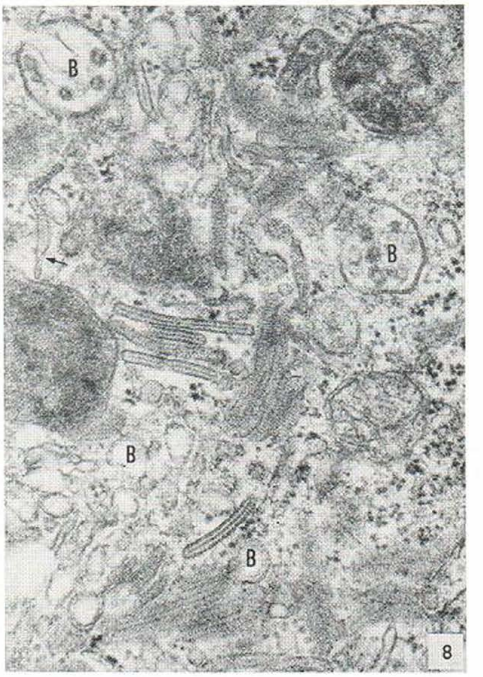


Fig. 5. Tails showing constrictions and small bulges (thin arrows). Thin tails (*T*). Annular bulge and tail (thick arrow). × 36 600.

Fig. 6. Normal tails and abnormal bulges. *1*, Trigonal shape. *2*, Hook shape. *3*, Large bulge containing glycogen particles. *4*, Bulge containing membranous structures. × 36 600.

Fig. 7. Large bulge containing glycogen particles (*G*). The bulge shows indistinct single inner membranes (arrows). *C*, Crystalloid patterns on bulge surface. × 36 600.

Fig. 8. Two small and two large bulges (*B*) and thin tails (thin arrow). The large bulges contain round bodies coated by a double membrane. A bulge in the upper-left corner shows such round bodies located in the space between the outer double membrane and the inner single membrane. × 36 600.

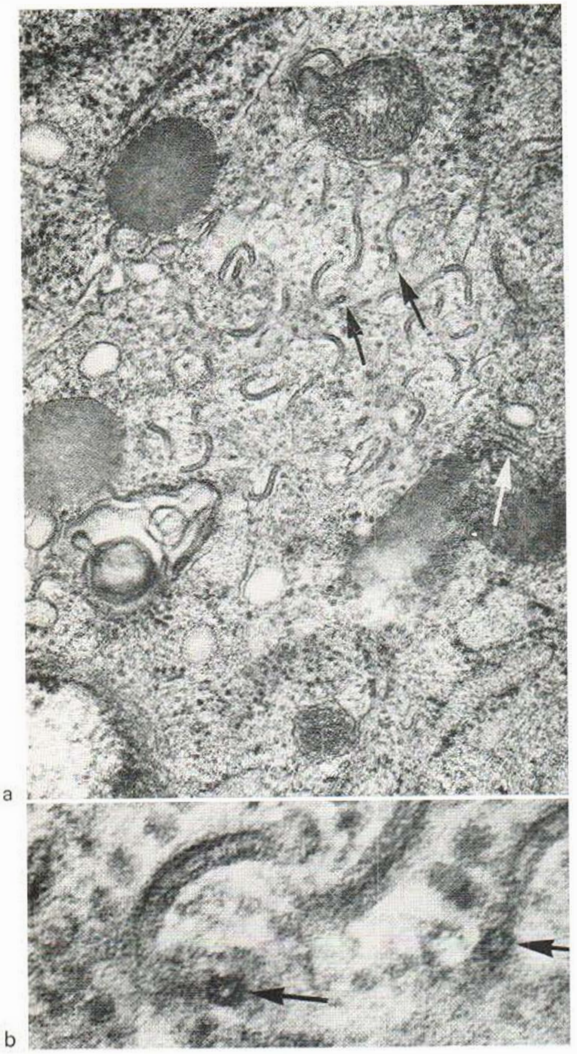


Fig. 9. (*a*) Bent thin rod-like granules. Thick arrow-pointed area is seen in the inset. A white arrow indicates one granule-tail. × 43 400. (*b*) Notice rosette-like cut-surfaces (arrows). × 192 000.

sions. The cells contained a nucleus and various amounts of characteristic Langerhans cell granules (Figs. 1, 2, 3). The nuclei were oval, highly convolute and lobate, and were surrounded by a distinct 300 to 500 Å thick fibrous lamina. Close to the inner surface of the fibrous lamina, fine chromatin granules were aggregated in thick masses, while coarse chromatin particles were scattered in the nucleoplasm. One, rarely two, nucleoli were seen. No mitotic figures could be demonstrated. A perinuclear cistern, although varying in width, did not exceed 500 Å.

Langerhans cell granules were constantly found in the cytoplasm and occasionally in the extracellular space. In this study, no granules were seen in the nuclei. Most granules were seen gathering around the Golgi complexes, while a few were connected to the inner surface of the cell membrane. The variability of the ultrastructure, number and distribution of the cytoplasmic granules and their presence in the extracellular compartment seemed to correspond with certain patterns of other cytoplasmic organelles (see below).

The granules often had either tails or bulges. Most of the tails were about 500 Å wide and showed straight, bent, branched or circular forms (Figs. 2, 3, 5, 6). Each tail was bounded by a double membrane and contained a central band. The surface of the tail showed a transverse banding with a periodicity of about 90 Å (Fig. 8). The longest tail found in this study was about 20 000 Å and the shortest about 800 Å. Thin and constricted tails (Figs. 4, 5) showing no distinct inner figures were seen here and there. The bulges were oval, triangular, spindle-shaped and hook-shaped (Figs. 6, 7) with various diameters ranging from 300 to 5 000 Å. The bulges were covered by a single membrane which continued from the tail. Another single membrane separated the central space from the bulge wall (Figs. 6, 7, 8). Glycogen particles were observed in the central space of bulges (Fig. 7) as well as round, double-membrane coated bodies in the space between the outer and inner single membranes (Fig. 8). The surface of the bulges showed a crystalloid structure (Fig. 7). Beside these granules, two other types of granule were seen in the cytoplasm of some infiltrating cells. One of these was represented by dense round and oval granules with approximately 3 500 Å as their largest diameter. These granules were surrounded by a double membrane and contained round, double-membrane coated bodies or lamellar structures in their dense matrix (Fig. 4). Cells containing such granules showed various abnormal shapes of tails and bulges. Finally, there were granules shaped as thin, bent rods grouping around and near the nucleus. The rods were 170 Å thick and 1 500 Å long with a rosette-like cut-surface. Few typical tails and bulges coexisted with such rods (Fig. 9 *a* and *b*). Cytoplasmic organelles such as mitochondria, lysosomes, ribosomes, intracytoplasmic

filaments, granular endoplasmic reticulum, Golgi complex, and centrioles, were noticed in the cytoplasm. The distribution patterns of these organelles varied from one cell to another suggesting possible relations with various granule figures. In cells containing few mitochondria, a narrow endoplasmic reticulum and an abundance of filaments, thin rods and few well-figured Langerhans cell granules were demonstrated (Figs. 1, 9). On the other hand, cells with dilated granular endoplasmic reticulum, Golgi complex, numerous ribosomes and mitochondria, and few filaments, showed numerous well-developed or abnormal tails and bulges as well as dense round bodies (Fig. 2). Cells showing a cytoplasmic pattern of the second type with well-developed granules, particularly often showed openings of the granules to the extracellular space and extracellular granules (Fig. 3).

DISCUSSION

The complete granules had long, branched or circular tails up to 1 μm (9). Similar shapes of complete granules have previously been noticed in Langerhans cells of normal human skin, which suggested that the figures were dependent on the level of sectioning (8). Hook-shaped bulges may come about in this way. Thin tails have been found in epidermoid epithelium of trachea and urinary bladder of Vitamin-A deficient Wistar rats (13). Glycogen particles and round, double-membrane coated bodies in bulges have not been mentioned in previous reports. The thin and constricted tails and small bulges seen around the Golgi zone probably represent incomplete forms. The thin rods are presumed to belong to these. The dense round granules probably represent lysosomes and are identical with those described by Zelickson (14). The thick fibrous lamina of the nucleus is an aid in identifying Langerhans cells, as is the indentation of the nucleus.

There are two theories on the formation of granules. According to one, the granule formation takes place in the Golgi zone (14) while the other considers infolding of the cell-membrane to be the first step of the formation process (7, 9). The present findings of incomplete granules around the Golgi zone suggest a close relationship between these organelles. Granules extruded into the extracellular space after cell disintegration were always complete (9, 12).

Though the function of the granules is obscure, coexistence of incomplete granules with other cell-organelles in patterns suggesting active cell function indicate a close relation between cell function and granule formation. The presence of such active cells in the cell infiltrates, together with previous findings of mitotic figures (7), confirms the neoplastic nature of Letterer-Siwe's disease.

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