

VIRUS-LIKE STRUCTURES IN LUPUS ERYTHEMATOSUS DISCOIDES

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Abstract. Biopsy specimens from typical skin lesions of DLE patients were studied by means of electron microscopy. Virus-like structures were discovered in the cytoplasm of fibrocytes, endothelial cells and keratinocytes. After prolonged chloroquine treatment a decrease in the frequency and size of these structures was observed.

Key words: Virus-like structures; Tubuloreticular structures; Discoid lupus erythematosus, Chloroquine treatment

On the basis of light and electron microscopic investigations Melczer et al. (23) proposed as early as 1962 a viral etiology for lupus erythematosus (LE). Fresco (5) observed the presence of tubular structures in lupus nephritis and they have since been found in the skin of LE patients too (7, 8, 10, 32). These structures were localized in different types of cells in SLE as well as in DLE (1, 12, 13, 14, 19, 21, 29, 34). Some of the investigators have found them only in active skin lesions (13, 14) while others also found them in clinically symptom-free skin (12, 20). Most of the above publications concerned the intracellular localization and morphological characteristics of the tubuloreticular structures, of which several variations were described by Kerl & Auböck (19).

There are some controversial data in the literature concerning the effect of disease duration on the tubuloreticular structures (13, 15, 24). According to Hausteiner (15) and Metz & Metz (24) previous treatment has no influence on these structures.

The aim of our study was to investigate the tubuloreticular structures in DLE skin lesions of different duration before and after prolonged chloroquine treatment.

MATERIAL AND METHOD

Our material was obtained from 10 patients (4 male, 6 female) suffering from DLE. The duration of the symptoms was 2-8 months. The skin lesions selected for biopsy were localized on the face. The diagnosis has been confirmed histologically. The patients had not received any therapy prior to the excision. In 3 cases the biopsy was repeated after 3 months of chloroquine treatment, and in 3 cases after 6 months (daily dose 0.25 g).

The biopsy material was fixed in 2.5% glutaraldehyde in 0.1 M phosphate buffer (pH 7.2) for 4 hours at 4°C, washed overnight in the same phosphate buffer solution, post-fixed in 1.5% osmium tetroxide buffered with 0.2 M *s*-collidine (pH 7.2) for 2 hours at 4°C.

Dehydration was carried out with ethanol and propylene oxide, embedding in Durcupan ACM. Sections from several blocks from each patient were cut on a LKB III ultratome, and contrasted with uranyl acetate and lead citrate. Micrographs were taken with a JEOL JEM 100 B electron microscope.

RESULTS

In biopsy specimens taken from untreated lesions we have observed the tubuloreticular structures in fibroblasts (Fig. 1) and endothelial and epidermal cells. Although we did not count their number, they were conspicuous, i.e. they could be found easily in every preparation without searching for them. In our material there was no difference between the fresh and older lesions as to the presence of the tubuloreticular structures. Their morphological characteristics and intracytoplasmic localization were the same as have been described by previous investigators.

The material of the same patients differed from the above group after prolonged chloroquine treatment. Tubuloreticular structures were rarely found; they had to be searched for very carefully. Not only

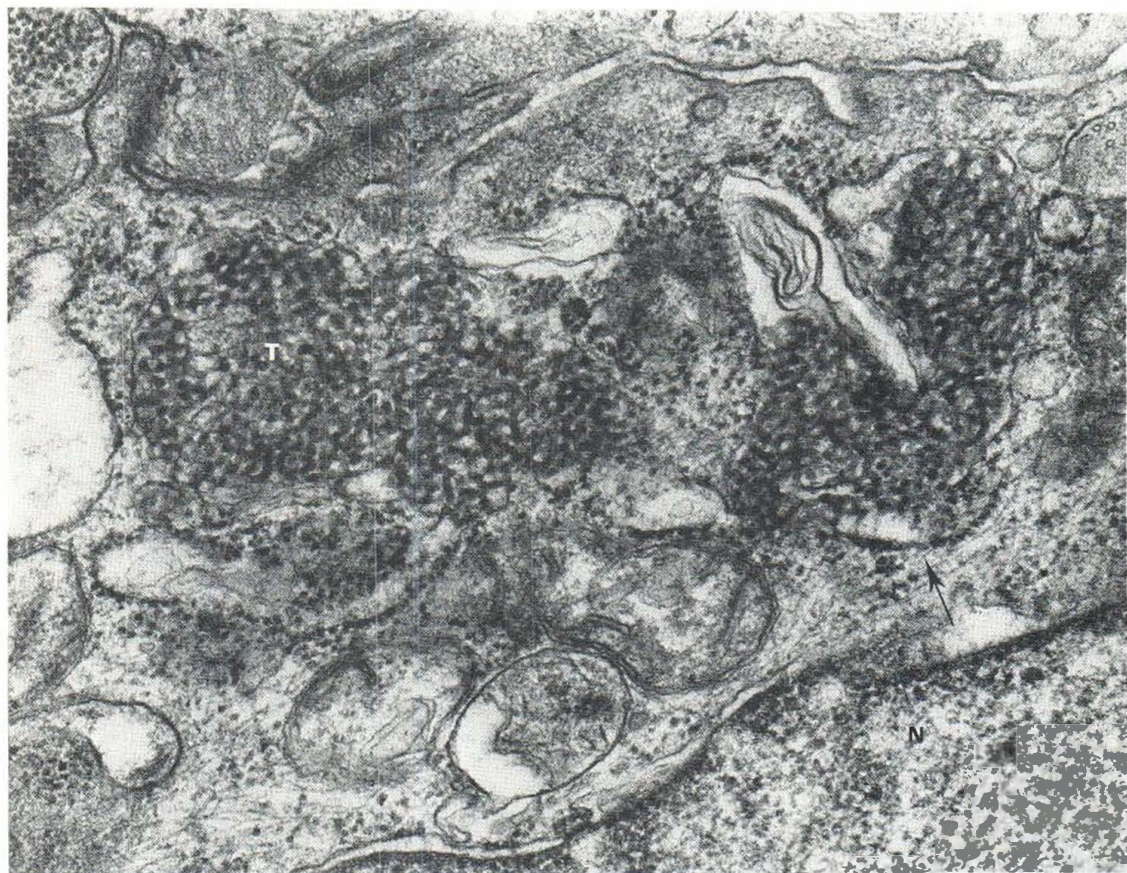


Fig. 1. Tubuloreticular structure localized within the dilated, rough-surfaced endoplasmic reticulum of a fibroblast. Skin lesions of an untreated DLE patient. N:

nucleus, arrow: membrane of the rough endoplasmic reticulum, T: tubuloreticular structure. $\times 132\,500$.

their relative incidence but also their size seemed to be reduced (Fig. 2). Their limiting membrane was expanded in several places and the tubuloreticular structure was not so well developed, the electron density decreased and the structure seemed to be amorphous (Fig. 3).

DISCUSSION

These virus-like structures, the nature of which is not clear even today (2, 4, 6, 9, 12, 13, 16, 17, 18, 20, 21, 26, 31, 32, 33), are morphologically similar to paramyxoviruses. Their role is unknown and all attempts at isolation of the virus have failed—even animal experiments for inoculation of the supposed infective agent were unsuccessful (19). Moolten *et al.* (26) reported the isolation of the virus from the

blood, but they did not characterize it electron microscopically.

Hurd *et al.* (18) consider that the virus-like bodies in the skin are a sign of a regenerative or reactive process following cell damage, rather than a phagocytic phenomenon of endothelial cells. The fact that these virus-like bodies show a topographical connection with the endoplasmic reticulum or are localized beside the nucleus, supports the view that they could be a special cell product. It is probable that the membrane surrounding the tubular structures originates from the endoplasmic reticulum.

Although virus-like structures have been found in several diseases (11, 19, 21, 22, 25), it is uncertain whether these structures, which are morphologically identical, have the same biological function.



Fig. 2. Tubuloreticular structure in a fibroblast after chloroquine treatment. Arrow: the remnants of the structure, m: mitochondrion, M: the limiting membrane, $\times 236\,700$.

A: amorphous material surrounded partly by the membrane. $\times 236\,700$.

In our material we have found no connection between the duration of the disease and the frequency of the tubuloreticular structures. According to Hashimoto & Thompson (13) the number of these structures decreases with the duration of the disease. Haustein (15) and Metz & Metz (24) are of the opinion that their numbers reflect the acuteness of the disease. In the material of these latter authors, previous therapy had had no effect on the tubuloreticular structures. We cannot compare our results with those of Haustein (15) and Metz & Metz (24), because their observations were made on the glomeruli of the kidney and in circulating leukocytes, after the administration of immunosuppressive drugs, steroid preparations and chloroquine, alone or combined. The duration of the treatment is

not mentioned. The chloroquine therapy in our material seemed to be effective in two ways: the skin symptoms showed an improvement and the relative incidence and size of the tubuloreticular structures diminished.

Although steroids and immunosuppressive agents, the effect of which is more than those of chloroquine, had no influence on the virus-like structures, our observation can be explained in two ways.

1. It is known from the work of Tan Em & Stoughton (35) that ultraviolet light damages the deoxyribonucleic acid in human skin. Berk & Blank (3) have found that in the skin of SLE and DLE patients the numbers of tubuloreticular structures increased after 10 days' ultraviolet treatment,

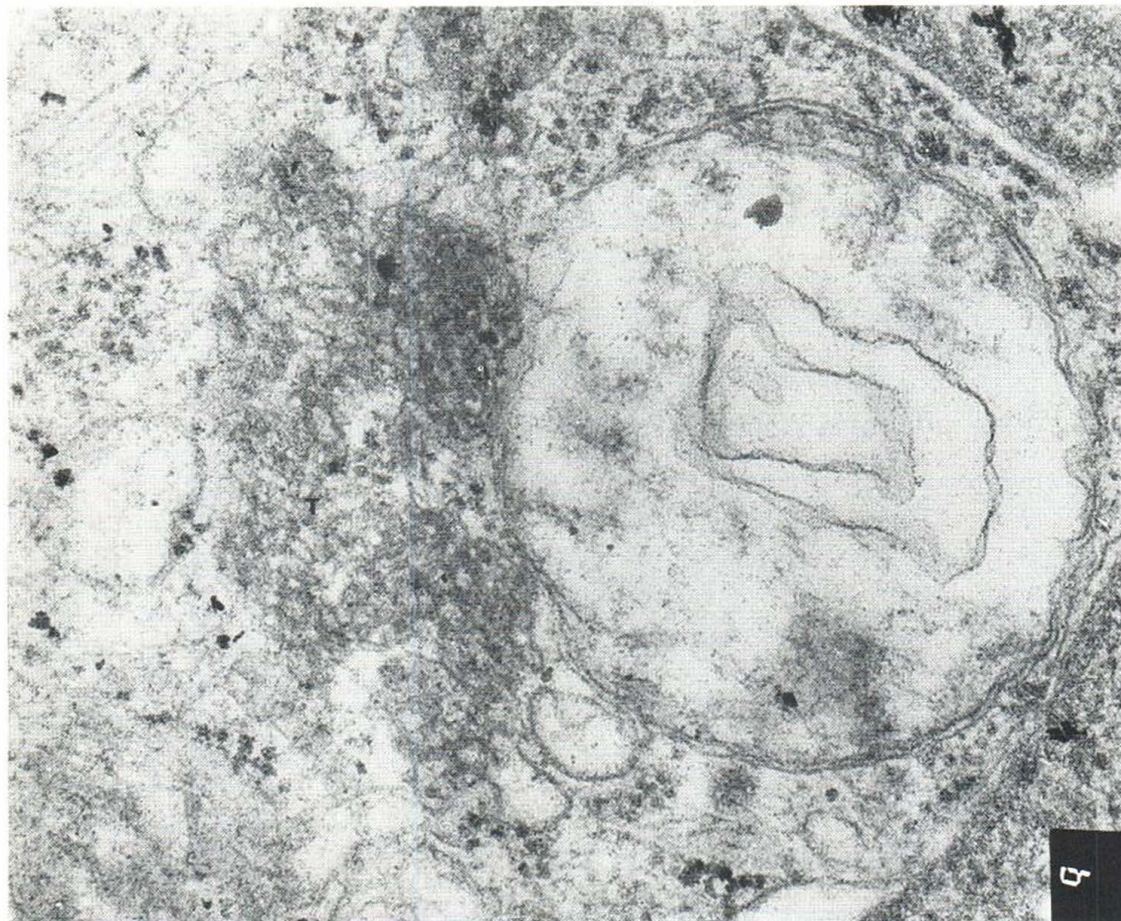


Fig. 3. Poorly defined tubuloreticular structure in a keratinocyte. Right: membrane limited space connected with the TRS. $\times 238\,500$.

which, as suggested, may be due to the fact that ultraviolet light causes an alteration of DNA of a neoantigenic character. The sun-screening effect of chloroquine may not be significant clinically, but its binding to the DNA (30) could well prevent the sun-induced transformation of DNA in the skin. It is also known that the chloroquine storage capacity of the skin is relatively small, compared with other organs (27), but in inflamed skin it is greater and remains unchanged for a longer period of time (28). This fact may account for our observation, provided that other known effects of chloroquine can play a part beside the DNA binding.

2. It is also possible that chloroquine accelerates these structures' natural involution, as has been described by Hashimoto & Thompson (13) in older skin lesions.

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