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ULTRASTRUCTURE OF SYSTEMIC LUPUS ERYTHEMATOSUS SKIN

Dermo-epidermal Junction

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Abstract. Twenty biopsies of SLE were studied in the electron microscope. The changes in the dermo-epidermal junction were as follows. 1) Fourteen specimens were characterized by a multilayered thick basal lamina with numerous normal anchoring fibrils, covering a wide stretch of the outermost corium. The subepidermal space was narrow. 2) Four specimens showed multiplication of the basal lamina invading the outer dermis, and either thick meshy basal lamina structures and numerous normal anchoring fibrils, or bands and fragments with normal anchoring fibrils and underlying balls of abnormal anchoring fibrils. No basal cell protrusions were found in the area of the invading basal lamina. 3) Two specimens showed an irregularly thick basal lamina proper, a fragmented basal lamina with crystalline structures and degenerated anchoring fibrils; in addition, an irregularly widened subepidermal space crossed by blurred anchoring filaments. Virus particles were usually found in the junction area, mostly in the meshy basal lamina of the second type. Evidence is presented that these changes represent fibrinoid and possibly develop directly as a response to virus infection, or, indirectly, through an immune reaction.

Recently, electron microscopic studies of systemic lupus erythematosus (SLE) have revealed intracytoplasmic filiform inclusions resembling the nucleocapsid of paramyxovirus in involved as well as uninvolved skin (13, 19, 23). Extracellular virus particles have also been found in the dermo-epidermal junction of involved skin (19). Folding of the basal lamina and basal-cell necrosis in involved skin have been described (28, 33). Gamma-globulin precipitation on the basal lamina, anchoring fibrils, and collagen fibrils of the papillary layer of involved SLE-skin, has been demonstrated by electron microscopy (29, 34). The present paper gives a detailed report on ultrastructural changes of the dermo-epidermal junction in systemic lupus erythematosus.

MATERIAL AND METHODS

Twenty patients with SLE were studied, 19 females aged 23 to 58 years, and 1 male, aged 22. The patients had outbreaks of lupus erythematosus on the torso, overarms and thighs as well as the face, more or less accompanied by general symptoms and lesions of internal organs. Light microscopically, the tissues of all patients showed changes characteristic of lupus erythematosus.

Biopsies for electron microscopy were taken from the skin eruptions and immediately fixed in ice-cold 6% glutaraldehyde solution in Veronal-acetate buffer, pH 7.2, with 7.5% sucrose. After being washed in the same buffer, the specimens were osmicated, dehydrated and embedded in Epon 812. Ultrathin sections were cut by LKB and Reichert ultramicrotomes and stained by uranyl acetate and lead citrate. Sections of some specimens in which a meshy basal lamina or strong oedematous changes were found were picked up on gold grids and stained using the periodic-acid silver-proteinate technique of Thiérry (31) and ruthenium red (17). A Siemens electron microscope (Elmiskop 1A) was operated at 80 kV with double condensors.

OBSERVATIONS

The dermo-epidermal junction was the most severely changed area in SLE skin, although both normal and abnormal junction components were found.

The subepidermal space was mostly regular and narrow. In two specimens, however, it appeared irregular with infoldings of the basal-cell membrane and a distorted basal lamina (Fig. 1). Lattice-like material filled in the irregular subepidermal space. Indistinct anchoring filaments emerging from the basal lamina connected the latter with half-desmosomes. Distinct half-desmosomes were occasionally anchored to the basal lamina by homogeneous material (Fig. 2). Severe

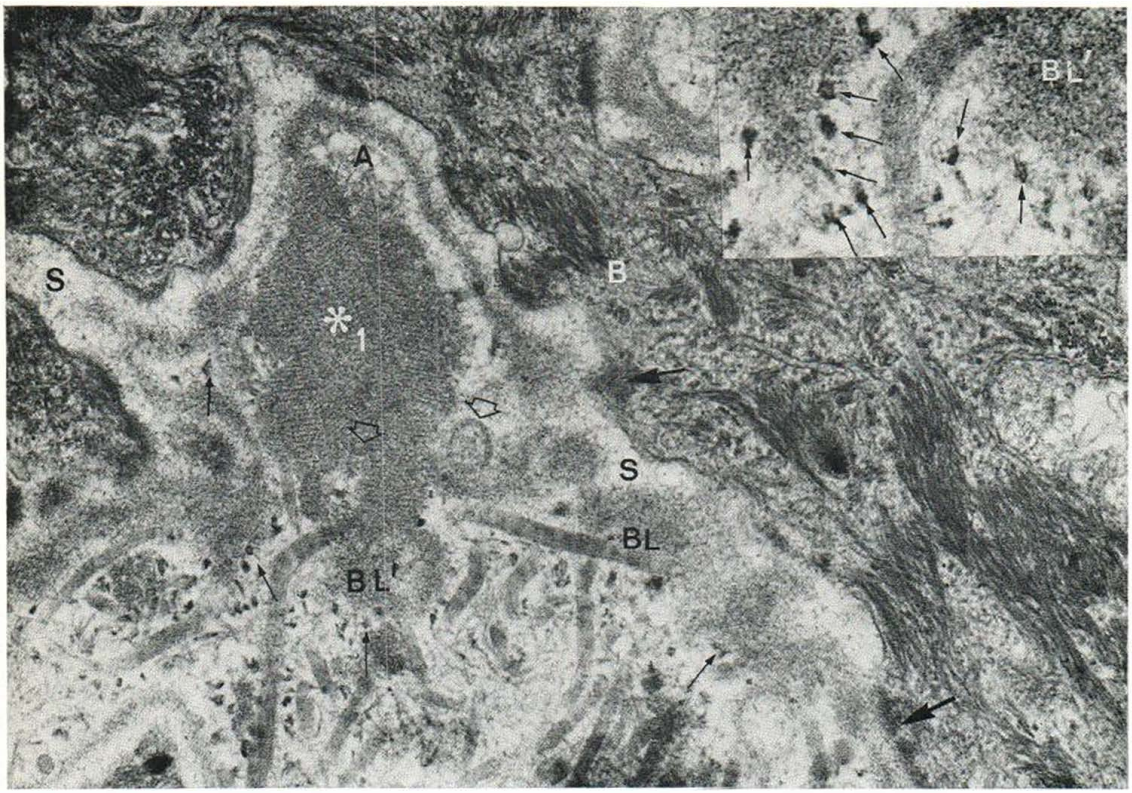

Fig. 1. An irregularly wide basal lamina (*BL*) is separated from a basal cell (*B*) by an irregular subepidermal space (*S*). Indistinct half-desmosomes are connected to the lamina by homogeneous masses of anchoring filaments (*thick arrows*). Lattice-like material is located in the subepidermal space (*S*). Two virus particles are seen in the junction area (*framed arrows*). Asterisk 1 indicates crystalline material with intervals of 8.3 nm covering fragmented basal lamina. Fragmented basal lamina (*BL'*) and anchoring fibrils (*A*) are seen in the periphery of the crystalline structure. Grainy anchoring fibrils (*thin arrows*) are seen on both normal and fragmented basal lamina. Distinct normal anchoring fibrils are sparse. × 45 000. Inset shows grainy anchoring fibrils (*thin arrows*). Fragmented basal lamina (*BL'*). × 90 000.

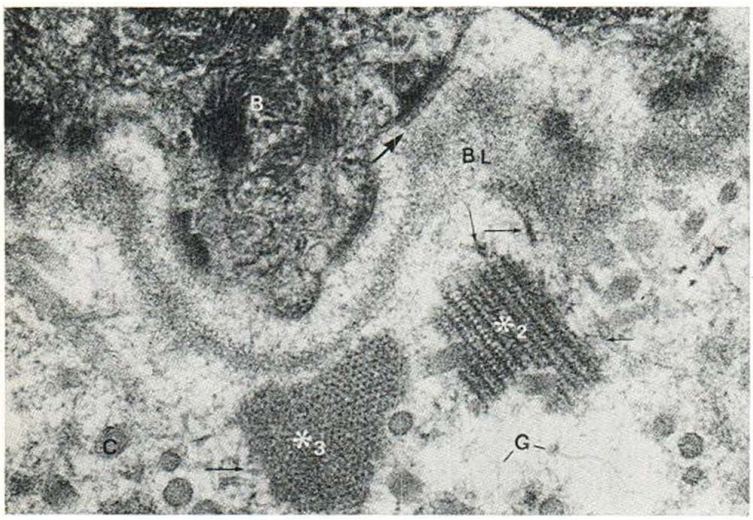

Fig. 2. Crystalline figure with 16.6 nm wide intervals (*asterisk 2*). Pointed crystalline figure (*asterisk 3*) with anchoring fibrils (*thin arrows*) close to the basal lamina (*BL*). In some areas the lamina is widened. Thick arrow indicates homogeneous material connecting a distinct half-desmosome with the basal lamina. The subepidermal space contains lattice-like material. Collagen fibrils (*C*). Acid glycosaminoglycan microfibrils (*G*) × 60 000.

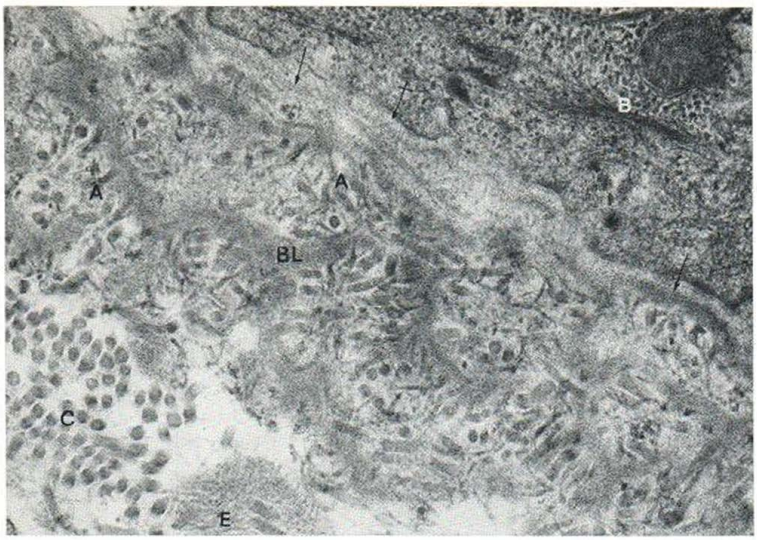

Fig. 3. Multilayered, wide basal lamina (*BL*) in the outermost corium, a narrow subepidermal space, and normal anchoring fibrils (*A*). Thin arrows indicate the basal lamina proper. Elastic fibril bundle (*E*). Collagen fibrils (*C*). Basal epidermal cell (*B*). × 30 000.

changes of both the subepidermal space and the anchoring filaments were often combined with widened intercellular spaces and cell hypertrophy in the basal epidermal cell layer.

The basal lamina showed either diffuse or irregular thickening reaching a maximum thickness of about 10 nm. In the majority of specimens the basal lamina was multilayered in the outermost dermis, appearing as bands, fragments, multiple lamellae (Fig. 3) or net-works (Fig. 4). Numerous normal anchoring fibrils protruded into the corium. Four biopsy specimens showed a basal lamina invading the outer and middle layers of the corium (Figs. 4, 5). Two showed crystalline patterns covering fragments of basal lamina (Figs. 1, 2). Most of these patterns showed parallel lines, 4.16 nm in width, with intervals of 8.3 nm (Fig. 1). Others showed 8.3 nm wide bands separated by 16.6 nm wide spaces through which 4.6 nm wide lines with 8.3 nm intervals were crossing (Fig. 2). Here and there a pointed pattern could be seen with distances between the points of 8.3 nm (Fig. 2). Some abnormal anchoring fibrils with dense grains were seen emerging from the crystalline patterns. Crystalline structures ocurred in the mucinous areas. The original basal lamina in this area was single, irregularly thick and separated from the basal cells by an irregular sub-

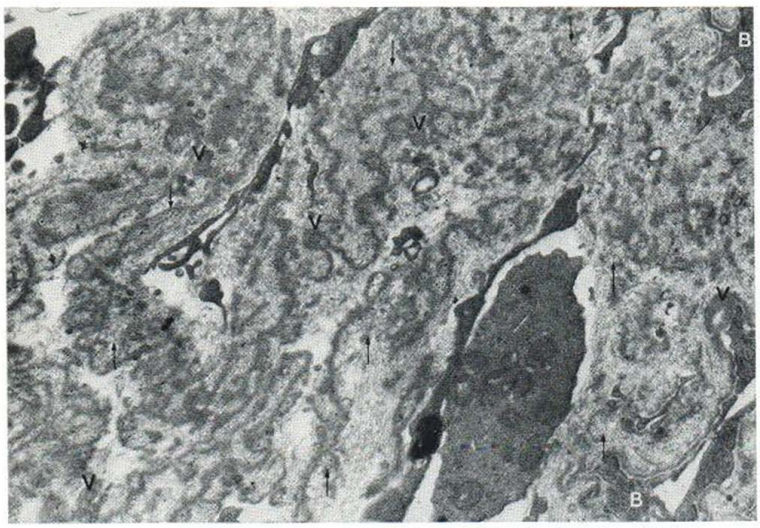

Fig. 4. Meshy basal lamina in the outer dermis. In the meshes, numerous virus particles (*V*) are seen. Anchoring fibrils (*arrows*) are numerous and normal. Basal epidermal cells (*B*). × 7 500.

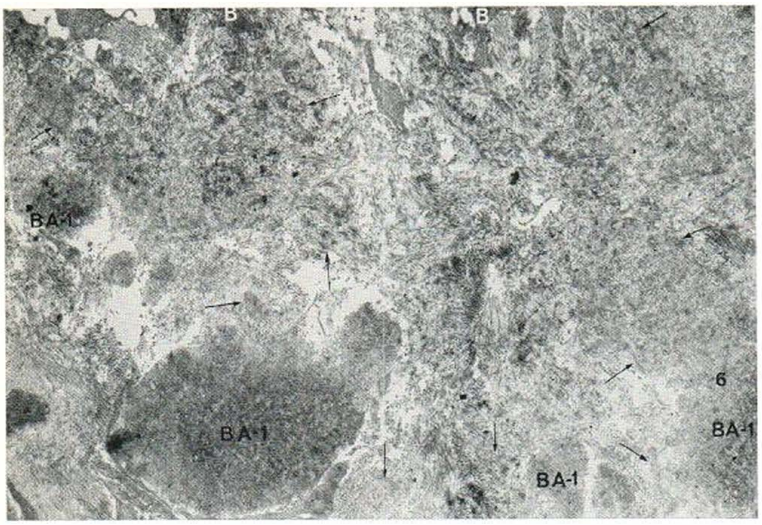

Fig. 5. Fragmented and band-shaped basal lamina with normal anchoring fibrils in the outer corium. Thin arrows indicate some examples. Balls of abnormal anchoring fibrils (*BA-1*). Basal epidermal cells (*B*). *6* indicates the area of Fig. 6. × 6 000.

epidermal space. Acid glycosaminoglycan microfibrils stained by ruthenium red were found beneath a fragmented basal lamina. Virus particles were demonstrated in the junction area (Figs. 1, 4, 5).

Numerous anchoring fibrils of varying length and width appeared on the dermal surface of the lamina. Most of the anchoring fibrils of a multilayered basal lamina had distinct bandings showing positive reaction with the periodic acid-silver proteinate method (Fig. 10). Three types of abnormal anchoring fibrils were considered. One was thin and blurred without bandings and often beset by dense rough grains. This type was seen in relation to basal laminae of irregular thicknesses and crystalline figures (Figs. 1, 2) as well as in the vicinity of balls of abnormal anchoring fibrils (Figs. 8, 9). Such balls, seen in three specimens showing multilayered basal lamina, usually contained one type in one ball, while a combination of two types was occasionally seen. Some were uniformly thick, straight, with a round cut-surface 7 nm across and a length of 200–300 nm (Figs. 7, 8), while the others were dense and short without bandings (Figs. 6, 9). The periodic acid-silver proteinate technique showed no normal bandings of anchoring fibrils, and, on the whole, the staining was faint (BA-1 in Fig. 10). The balls

Fig. 6. Band-shaped and fragmented basal lamina with normal anchoring fibrils is seen on the left-hand side of the photograph. Thin arrows indicate some normal anchoring fibrils; Virus particle (*V*). The lamina material continues into a ball of abnormal anchoring fibrils (*BA-1*). Abnormal anchoring fibrils in the ball are dense and short. Collagen fibrils are seen within the ball (*C*). × 15 000.

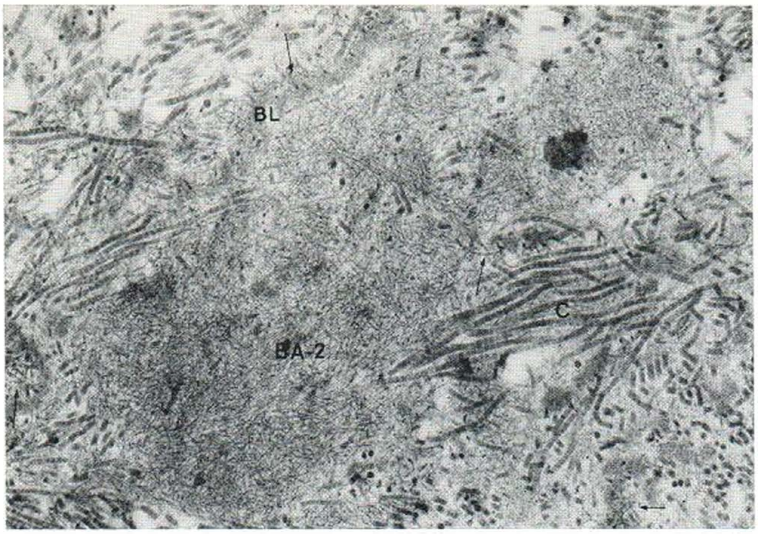

Fig. 7. Band-shaped invading basal lamina (*BL*) with normal anchoring fibrils (*thin arrow*) continuing into a ball of thin straight anchoring fibrils (*BA-2*). Collagen fibrils (*C*). × 15 000.

also contained more or less blurred fragments and bands of basal lamina (Figs. 5–9).

Elastic fibril anchorings to the basal lamina were rarely seen. Broad elastic fibres were occasionally connected with a multilayered basal lamina.

DISCUSSION

The first type of lesion in the dermo-epidermal junction suggests that, at the earliest stage, the disease process is located around the subepidermal space. The basal epidermal cells presumably play a role. The second type was proliferative with well-formed junctional components. The third was also proliferative, but the change extended deeper than the other types and often showed abnormal junctional components.

The above-mentioned changes are different from the junction lesions seen in chronic pemphigus (bullous pemphigoid) (14), light dermatosis (21), psoriasis (5), scleroderma (18), basal cell carcinoma (16), and squamous cell carcinoma (9, 15). It may be concluded that the junctional changes reported above are characteristic and specific of SLE.

Multiplication and thickening of the basal lamina is usually accompanied by basal cell pro-

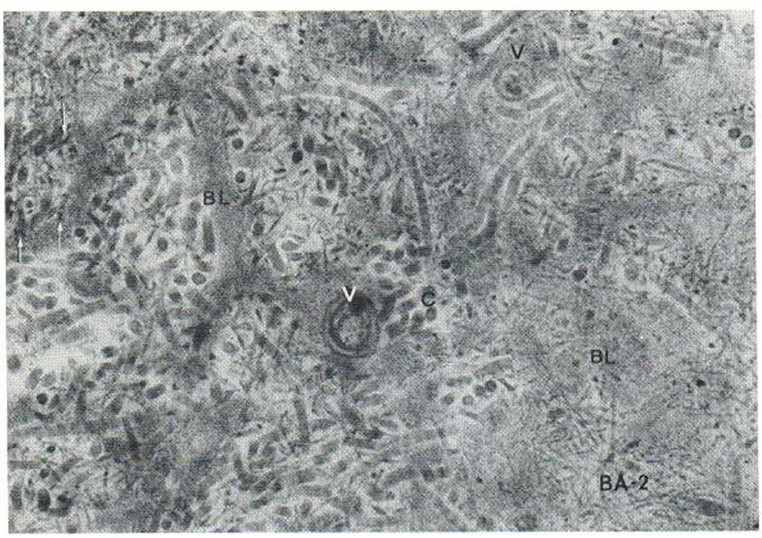

Fig. 8. Band-shaped meshy basal lamina (*BL*) shows normal anchoring fibrils (*thin arrows*) and thin straight anchoring fibrils (*BA-2*). The lamina is diffuse and less dense in the ball (*BA-2*). Virus particle (*V*). Collagen fibrils (*C*). × 30 000.

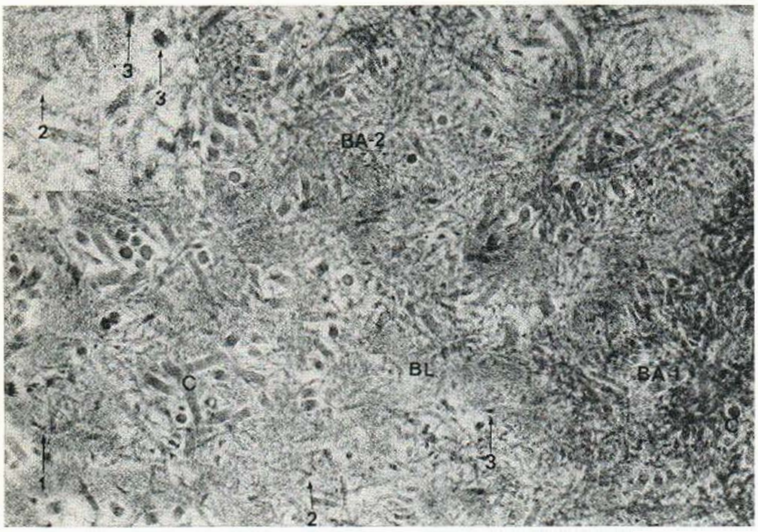

Fig. 9. Masses of dense thick (*BA-1*) and straight thin (*BA-2*) anchoring fibrils are continuous with band-shaped basal lamina material (*BL*). The lamina appears blurred. Normal (*arrow 1*), non-banded (*arrow 2*; *left inset*) and grainy (*arrow 3*; *right inset*) anchoring fibrils are also found. × 30 000 and × 60 000 (*inset*).

trusions. This phenomenon was not found in the study of SLE. Virus particles were always located in pathological junction areas and were exceptionally numerous in the meshes of the junctional lesion of type 3 (Fig. 4). It is known that virus envelopes originate from host-cell membranes and have host-cell specificity (1). The virus-like particles in the junction areas were probably extruded by budding from the basal epidermal cells (19), and their envelopes had the character of basal-cell membrane. Our findings represent evidence that the particles found in the junction area of SLE skin are real virions (19). They are presumed to cause the proliferation of the junctional structures.

In histochemical studies of SLE, the periodic acid-Schiff positive basement membrane has shown diffuse and granular thickening (26). A similar phenomenon has been obtained by fluorescence microscopy using antibodies against human gamma-globulin (30) and human fibrin (27). Using the Thiérry technique, we found the reaction to be strong for anchoring fibrils and faint for basal lamina. Earlier immuno-electron microscopical studies showed ferritin-tagged (34) and peroxidase-conjugated anti-human gamma-globulin antibody (29) on the basal lamina and the anchoring fibrils. Thus, there is evidence that the pathological junction components of SLE contain gamma-globulin and fibrin.

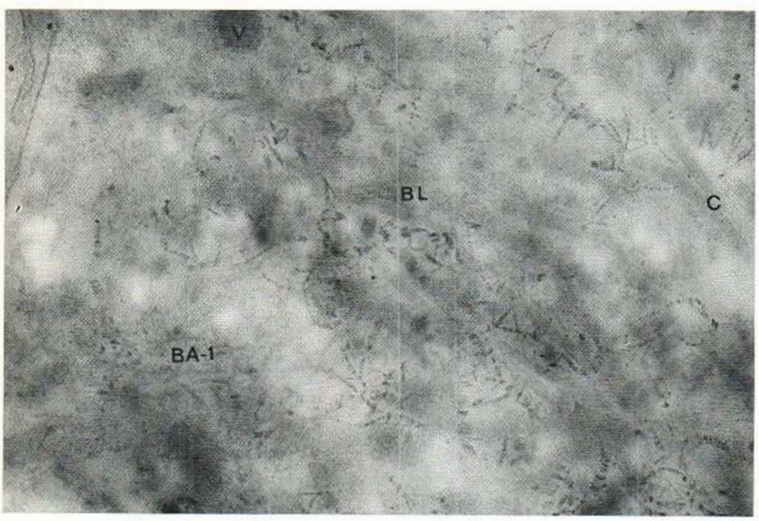

Fig. 10. Invading basal lamina stained after Thiérry. Transverse bands of normal anchoring fibrils on band-shaped basal lamina (*BL*) show positive reaction. Basal lamina and pathological anchoring fibrils (*BA-1*) react indistinctly. Collagen fibrils (*C*). Virus particle (*V*). × 30 000.

The glomerular basal lamina is sandwiched between epithelial and endothelial cells and is connected to both cells by fine filaments (20), very much like the anchoring filaments in skin. However, no anchoring fibrils exist. In lupus nephritis, the basal lamina is thickened with precipitation of gamma-globulin, and dense material with "organized bodies" is deposited in the lamina as well as in the subepithelial space (8, 12, 32). Thickenings and dense deposits with precipitation of gamma-globulin and fibrin have also been found in allergic glomerulonephritis (22), equine infectious anemia, virus glomerulonephritis (2, 3), and experimental lupus nephritis in New Zealand black mice (7, 24). The findings suggest that the above-mentioned glomerular changes were produced by an immunological process. Some authors found evidence that similar changes in the glomerular basal lamina of SLE represent fibrinoid precipitation (4, 10, 12). Although the term fibrinoid undoubtedly covers several different entities of diverse pathogenesis (6, 7, 25), it may be established that fibrinoid represents connective-tissue components with fibrin and immune globulins. The ultrastructural changes of junctional components found in this study correspond well to the fibrinoid material of glomeruli found in lupus erythematosus.

Grishman & Churg (11, 12) found "organized bodies" in renal glomeruli, in dermal vessels and under the dermo-epidermal basal lamina in SLE and presumed that these remarkable crystalline structures were deposits of immune globulin. However, according to other immuno-morphological studies, gamma-globulin precipitated diffusely over a wide stretch of the changed basal lamina and no immuno-electron microscopical studies have demonstrated immune globulins in "organized bodies". The detailed structure of the crystalline bodies found in this study was not identical with that described by the above-mentioned authors. We presume the presence of a precipitated immune reactant in the crystalline structures because of the regular patterns. However, the exact nature of the structures remains obscure.

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