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# VASCULAR IMMUNE DEPOSITS IN A LYMPH NODE OF A PATIENT WITH SYSTEMIC LUPUS ERYTHEMATOSUS

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## Abstract

**A biopsy specimen of the lymph node of a 38-year-old woman with systemic lupus erythematosus was studied with immunofluorescent staining, and tissue bound IgG, IgM, C3, and plasminogen were demonstrated in vessel walls. The skin revealed no deposits of these elements at dermo-epidermal junction and in vessel walls. Circulating immune complexes in the serum were detected by the Raji cell test. This case indicates that lymph node biopsy gives us chance to observe those deposits in vessel walls of the patient with systemic lupus erythematosus when immunofluorescent staining of skin is negative.**

## INTRODUCTION

Deposits of immunoglobulins and complements have been demonstrated by direct immunofluorescence (IF) at dermoepidermal junction (D-E junction)[1,2] of the skin, and in renal glomeruli[3,4] and blood vessel walls of several organs[4,5] of the patient with systemic lupus erythematosus (SLE). The deposits are considered to be immune complexes,[6,7,8] but the mechanism of their deposition is not known so well enough. Svec et al.[9] referred to deposition of IgG and complement on the vessels of the lymph node obtained at necropsy of lupus patient, but its photograph and the comment were absent in their paper. As far as we know, the positive IF staining of vascular walls of the lymph node biopsy specimen has not been reported. We observed the deposits of immunoglobulins, complement, and plasminogen in vascular walls of the lymph node obtained from the patient at onset stage of SLE.

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## CASE REPORT

A 38-year-old woman experienced arthralgia on her both shoulders in April, 1978. A month later, pyrexia of 39.3°C developed with petechiae and she was admitted to Kawasaki Medical School Hospital on May 10. Since she was 20 years old, she has experienced Raynaud's sign in winter. Physical examination showed petechiae on the extremities, trunk, and hard palate. Generalized lymphadenopathy with tenderness was present. Most of the nodes enlarged to the diameter of approximately 2 cm.

Laboratory data showed the following : WBC count, 2900/cu mm, with 56% neutrophils, 36% lymphocytes, 5% monocyes, 2% eosinophils, and 1% basophils ; RBC count, 340 $\times$ $10^4$/cu mm ; hemoglobin level, 11.3 g/dl ; hematocrit reading, 33.4% ; platelet count, 1.2 $\times$ $10^4$/cu mm ; ESR, 68/hr ; urinalysis, 20 to 30 RBCs and 1 to 3 WBCs/high-power field, moderate hyaline casts, a few granular casts, 3 RBC casts and 2 WBC casts/whole field, urine protein 3 to 4 g/day ; serum protein, total 5.6 g/dl, A/G ratio 0.64, $\gamma$-globulin 35.4%, $\alpha_1$ 5.1%, $\alpha_2$ 11.2%, $\beta$ 8.9% : serum IgG, 1859 mg/dl(normal, 700 to 1600) ; IgA, 206 mg/dl (normal, 90 to 450) ; IgM, 112 mg/dl (normal, 60 to 280) ; IgE, 12990 units/ml (normal, 20 to 900) ; $\beta_1$A, 15 mg/dl (normal, 50 to 90) ; $CH_{50}$, 11.9 (normal, 30 to 50) ; C-reactive protein, negative ; rheumatoid factor, negative ; ASLO, 40 Todd units ; ANA, 1 : 1024 (peripheral pattern) ; Anti-DNA antibody, 1 : 1280 ; LE cell, positive ; anti-platelet antibody, positive ; direct and indirect Coombs' tests, negative ; STS, positive : TPHA, negative.

## HISTOLOGY

A skin biopsy specimen removed from lesion of petechia on the right forearm was stained with hematoxylin-eosin, which showed focal spongiosis of lower epidermis with exocytosis. The perivascular infiltration of lymphocytes and histiocytes and extravasation of erythrocytes were seen in the upper dermis. The vessel walls were edematous.

A lymph node biopsy specimen was obtained from the right inguinal region. The cortex lost its normal architecture and was characterized by capillary proliferation, mild fibrosis, and focal clustering of so-called immunoblasts. No evidence of vasculitis was observed in specimens of the skin and the lymph node.

## IMMUNOFLUORESCENT STUDIES

The biopsy specimen obtained from the lesions described above were also submitted for the IF studies according to the method described before.[10] Antisera

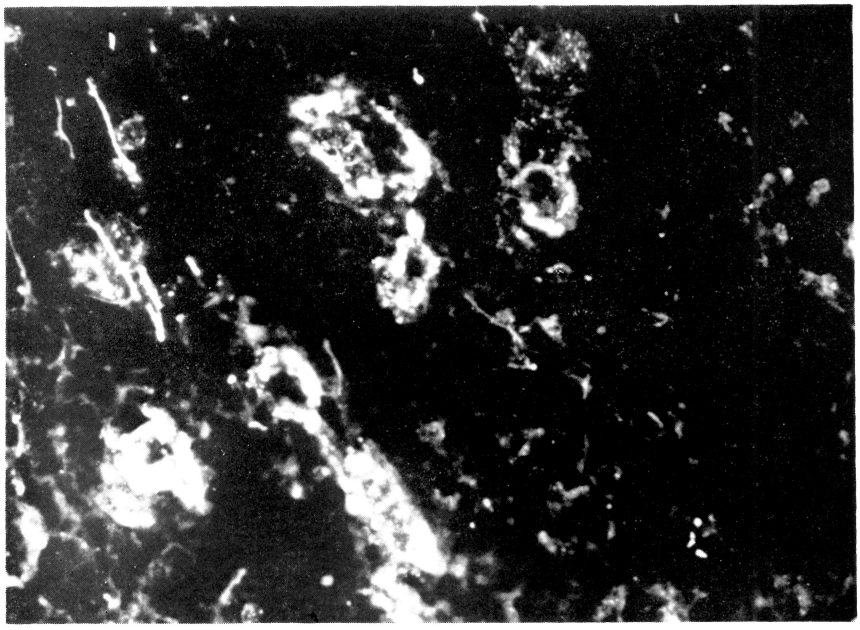

Fig. 1.

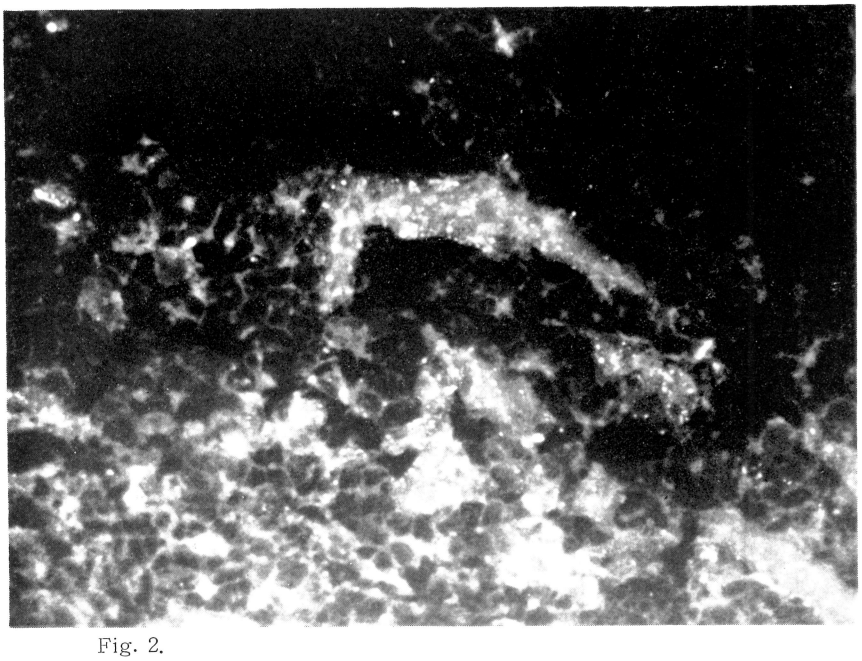

Fig. 2.

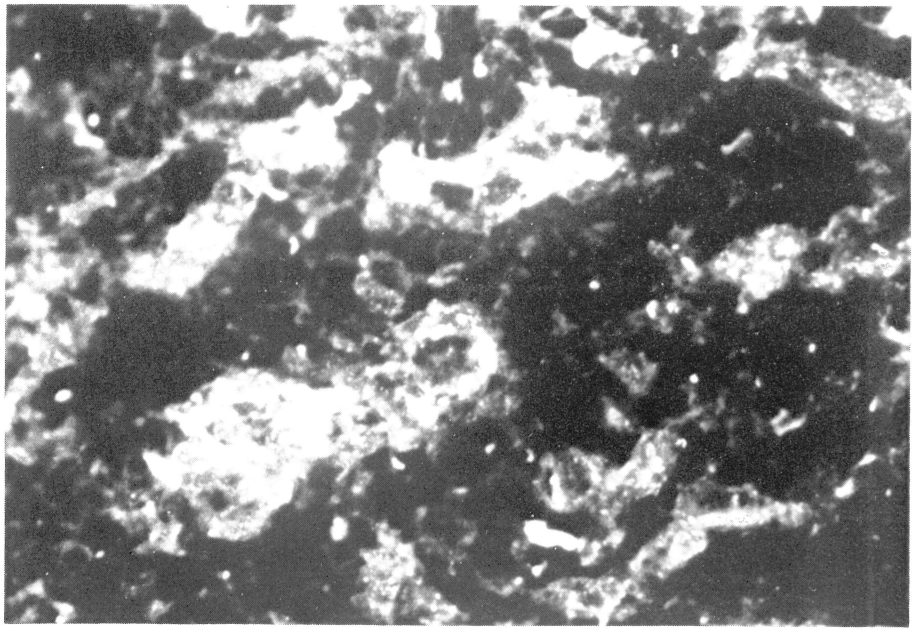

Fig. 3.

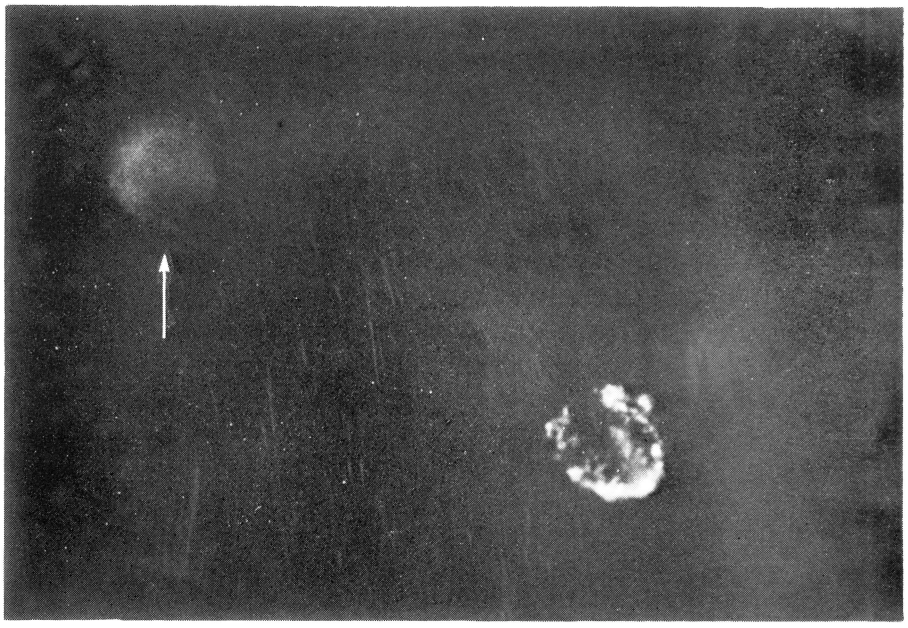

Fig. 4.

and FITC-labeled conjugates were treated with rabbit liver acetone powder before use. The direct IF method was applied for the detection of IgG, IgM, IgA, and C3, using fluorescein-isothiocyanate (FITC)-conjugated rabbit antisera (MBL, use dilution 1 : 10) and the indirect method was performed to detect plasminogen and fibrin, using rabbit antisera (Dakopatts) and FITC labeled anti-rabbit $\gamma$-globulin goat serum (MBL). The molar fluorescein-to-protein ratio of the conjugates ranged 1.2 to 1.9.

No specific deposits of IgG, IgA, IgM, C3, plasminogen, and fibrin were observed at the dermo-epidermal junction (D-E junction) and on blood vessels of the skin specimen. The vessel walls in the lymph node showed granular intense staining for IgG (Fig. 1), IgM, and plasminogen (Fig. 3), and less intense staining for C3 (Fig. 2). The involved vessels were capillaries and post-capillary venules.

Any sign suggesting the deposition of IgG, IgM, IgA, C3, and plasminogen was not found in vessel walls in the inguinal lymph node of three patients with psoriasis, one with wide-spread DLE and one with bullous pemphigoid.

## DETECTION OF CIRCULATING IMMUNE COMPLEXES

The serum was tested for circulating immune complexes by the Raji cell test according to Theofilopoulos' method.[11] Raji cells were cultured in RPMI 1640 supplemented with 20% fetal bovine serum for 3 days in 5% $CO_2$ incubator. FITC-labeled antiserum to IgG was purchased (MBL) and diluted 5 times before use. The serum from the patient was diluted 20 times. Blocking of receptors of Raji cells for IgG Fc was omitted, because IF staining of Raji cells by binding of monomeric IgG in the serum is so faint that it is negligible when the serum is diluted enough 20 times. As controls, aggregated human IgG[12] added with complement, and sera from patients with several diseases were tested. Raji cells were stained in granular pattern with the serum which contained immune complexes (Fig. 4). As shown in Table 1, circulating immune complexes were detected in the sera from patients with SLE and bullous diseases.

## DISCUSSION

The patient satisfied the criteria for diagnosis of SLE established by the American Rheumatism Association.[13] The petechiae may be due not to vasculitis but to thrombocytopenia, because no evidence of vasculitis was present and staining of immunoglobulin and complement was negative at D-E junction and in vessel walls of the skin specimen. Lymphadenopathy is not a diagnostic sign of SLE, but is a common sign next to arthralgia, exanthema,

and pyrexia in this disease.[14] Histologically, these nodes show follicular hyperplasia,[15,16] hematoxylin-stained bodies,[17] necrobiosis in different stages of development,[17] and infiltration of the medullary tissue with plasma cells.[15] Though hematoxylin-stained bodies and necrobiosis are regarded as diagnostic of SLE, they are not so frequently present.[16] Therefore, biopsy of lymph node seems much less popular than that of skin or kidney as a procedure for diagnosis.

Reports about IF studies of the lymph node of the patient with SLE are rare. Svec et al. referred to a lymph node obtained from a patient at autopsy, which demonstrated bound IgG and complement in vessel walls. In our case, the deposits were positive in vessel walls of the lymph node while they were negative both at D-E junction and in vessel walls of the skin. In general, the skin of SLE patient often demonstrates the immunoglobulins or complements at D-E junction even if it is the uninvolved skin, and seldom demonstrates the deposits in vessel walls. It is supposed that the components and the sites of predilection for binding of the deposits may vary with each other and the deposits in vessel walls of the lymph node may differ from those at D-E junction in some natures. Biopsy of the lymph node is easier than that of other internal organs. IF staining of the lymph node seems helpful to observe the bound immunoglobulins or complements in vascular walls of the patient with SLE when that of skin is negative at D-E junction and in vascular walls.

The deposits of immunoglobulin and complement in vessel walls as well as those at D-E junction and in renal glomeruli are considered to be immune complexes.[6,7,8] But it is still unknown whether the circulating immune complexes in the blood are deposited to the tissues or antibodies react to antigens bound to the tissues beforehand. In our patient the circulating immune complex was detected, so the former can be suggested.

Vascular deposits of plasminogen in the lymph node are reported for the first time in this paper. Plasminogen and plasmin were shown to be capable of activating the alternative pathway of complement and controlling its positive feed-back mechanism.[18] Though no evidence of vasculitis was seen in the lymph node of this case, the deposits including plasminogen suggest the possibility of exsistence or development of vasculitis.

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