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IMMUNOFLUORESCENCE STUDIES IN HERPES GESTATIONIS

M. Kocsis, T. Larsen Eeg, G. Husby and G. Rajka

From the Department of Dermatology, and the Institute of General Immunology, Rikshospitalet, Oslo, Norway

Abstract. In 4 cases of H.G., direct junctional immunofluorescence findings showed an intense band of C3 present in the involved skin of the patients even after the active lesions had disappeared (i.e. 2 months later). Weak specific fluorescence for IgG and in one case also for IgA and IgM was seen in the involved skin. A less intense junctional band of C3 was also present in the uninvolved skin. Antibodies against basal membrane have not been detected in the sera of the patients.

Key words: Herpes gestationis; Immunofluorescence; Junctional Band of C3

Herpes gestationis (H.G.) is regarded as a separate and distinct entity within the group of bullous diseases (15, 16). The acute onset of H.G., its morphological features and its good therapeutic response to systemic steroids resemble those of bullous pemphigoid, but the pathology of H.G., including its ultrastructure, is different (11, 17). According to other views, however, the disease is closely related to dermatitis herpetiformis.

Since the demonstration of basement membrane direct immunofluorescence in pemphigus and bullous pemphigoid (3) the value of immunofluorescence studies for the diagnosis and classification of subepidermal blistering diseases has been proved (1, 4). Regarding H.G., however, there are only few and somewhat contradictory data available (12, 13, 14).

The present study describes immunoglobulin and C3 findings in four cases of H.G.

SHORT CASE REPORTS

Case 1. K.G., a 34-year-old woman with RH+. She had a normal pregnancy (male, weighing 2.6 kg and 42 cm long) at the age of 23, but afterwards she had eight spontaneous abortions between the second and fourth month of each pregnancy. In the sixth month of her latest pregnancy, after she was given Sulfapral® tablets for cystitis, erythematopapulous and vesicobullous itching lesions developed on the palms and soles and subsequently on the abdominal area too. Immediately after the delivery the lesions spread to the forearms and the legs and to other parts of the trunk, but not to the mucous membranes.

The baby, a girl weighing 1.75 kg and 41.5 cm long, was premature. In the obstretical protocol, dysfunction of the placenta was mentioned.

After 10 days the eruption was controlled by systemic steroids.

Findings: Rheuma factor tests (Waaler, Latex) negative, antinuclear test, LE-factor: negative. Paper electrophoresis: total proteins 6.0, albumen: 2.6, α-globulin: 0.5, β_1-globulin: 0.9, beta$_2$-globulin: 1.0, gamma-globulin: 1.0, g/dl. Immunoglobulins: IgG: 8.2, IgA: 1.5, IgM: 0.6 mg/ml. C3: 2.6 mg/ml.

Case 2. W.G., a 22-year-old woman with RH−. During her (first) pregnancy, hypertonia was discovered and an antihypertensive drug was given until delivery. Five days after delivery (the newborn girl weighed 2.13 kg and was 45 cm long) she experienced itching accompanied first by erythematopapulous lesions. After administration of 20 mg prednisolone daily, the eruption cleared up in 3 weeks.

Routine examination: normal values. Immunoglobulins: IgG: 16, IgA: 1.8 mg/ml, IgM: 2.7 mg/ml., i.e. highly elevated. C3/serum: 2.3 mg/ml.

Case 3. A.L.G. female, 35 years, RH+. This was her fourth pregnancy. Three days after delivery (normal female weighing 3 500 g, 50 cm long) vesicobullous eruption appeared on the limbs. The patient was given prednisolone for 3 weeks. The eruption showed good regression, but deteriorated during her first and second postnatal menstruation.

Routine examination: normal values. Immunoglobulins: IgG: 11.8, IgA: 2.3, IgM: 1.1 mg/ml.

Case 4. K.K., 26 years, RH+. After a normal pregnancy and immediately after an uncomplicated delivery (female 4 070 g, 51 cm long) a papulovesiculous, oedematous diffuse eruption appeared, followed by typical bullae, especially on the legs.

Routine examination: normal values. IgG: 5.0 mg/ml, IgA: 2.6 mg/ml, IgM: 1.0 mg/ml, C3: 2.0 mg/ml.

PATHOLOGY

In all 4 cases a typical histological picture with subepidermal bullae and a perivascular infiltration was detected, consisting of lymphocytes, and also of neutrophils and eosinophil granulocytes. In addition, in case 2 a slight oedema of the endothelial vessels and in case 3 also plasma cells in the dermal infiltrate were observed.

Table 1. *Immunofluorescent findings in 4 patients with H. G.*

	Junctional DIF				
	C3	IgG	IgA	IgM	IIF
Case 1					
Involved skin	+++	+(+)	−	−	
Uninvolved skin	+	−	−	−	−
Involved skin 2 months later	+++	+(+)	−	−	
Case 2					
Involved skin	+++	(+)	−	−	
Uninvolved skin	+++	(+)	−	−	−
Involved skin 2 months later	− to ++	−	−	−	
Case 3					
Involved skin	++(+)	++	+	+	−
Uninvolved skin	+(+)	+(+)	(+)	(+)	−
Case 4					
Involved skin	++	−	−	−	−
Uninvolved skin	−	−	−	−	−

METHODS

(A) Direct Immunofluorescence tests (DIF) according to usual techniques (10) were used for the localization of immunoglobulins, and the complement fraction C3 using FITC-conjugated rabbit antisera against human IgG, IgA, IgM and C3 was employed. The specificity of the conjugates was checked by immunodiffusion and haemagglutination tests and by direct immunofluorescence on myeloma bone-marrow cells.

(B) The Indirect Immunofluorescence technique (IIF) using monkey oesophagus for detection in sera of antibodies against normal skin components was applied as previously described (3). The quantity of C3 in the serum was determined by means of radial immunodiffusion (9).

RESULTS

DIF findings are summarized in Table 1. Along the basal membrane a massive linear deposition of C3 classified comparatively as +++ (Fig. 1), as well as a moderate frequently "curled" deposition of immunoglobulins was observed. In the uninvolved skin a deposition of C3 was also detected in 3 of 4 cases (Fig. 2). Two months later, the areas of former lesions in patients K. G. and W. G. were tested by renewed DIF test and positive but less intense immunofluorescence was found (Fig. 3). In the sera of these patients no anti-basement membrane antibodies could be demonstrated. The C3 level was raised in case 1: 2.6 mg/ml and in case 2: 2.3 mg/ml, but was within normal limits in case 3: 1.8 mg/ml and in case 4: 2.0 mg/ml.

DISCUSSION

Kjartansson et al. (7) examined by means of immunoelectrophoresis sera from patients with H.G. as well as from those having pemphigus vulgaris and dermatitis herpetiformis. They found that IgA and IgM changes detectable in H.G. were similar to those found in dermatitis herpetiformis.

Rimbaud et al. (13) reported on a 22-year-old primipara with typical H.G. where intercellular antibodies against Malpighi cells were found in a titre of 1/50. In the same patient an examination of antibodies during her second pregnancy could not reveal such antibodies (14). Her first child, a girl, had skin lesions and IgG fixed to the basal membrane area was found in the skin. There were, however, no circulating antibodies (13).

Although negative findings were mentioned earlier in single cases of H.G. (2, 20), Provost & Tomasi (12) made immunofluoscence investigations in patients with H.G. In the first case basal membrane depositions of C3 and C5 were found in the absence of Clq, immunoglobulins and light chains. The second patient showed deposition of C3. Though no anti-basement membrane antibodies could be discovered in these patients, the authors found thermolabile humoral factors capable of depositing C3 (without Clq or C4) on normal skin basement membrane in the sera of these H.G. cases. They attempted an analysis of the characteristics of these serum factors too; what they found was that they corresponded to a non-dializable thermolabile pseudoglobulin (which is present only so long as the disease is active) rather than to immunoglobulin or properdin.

In all these cases of H.G. we have found an intense deposition of C3 on the basement membrane in the involved and in 3 of 4 in the uninvolved skin. The amount of C3 was somewhat smaller in the uninvolved skin, especially in case 1 where skin lesions showed a "macroscopic healing". Though to a smaller degree than C3 and in the uninvolved skin only in case 3, immunoglobulins, primarily of the type IgG could also be demonstrated in our cases. These findings confirm those of Provost & Tomasi (12) as far as the role of the complement along the basement membrane in H.G. is concerned. The

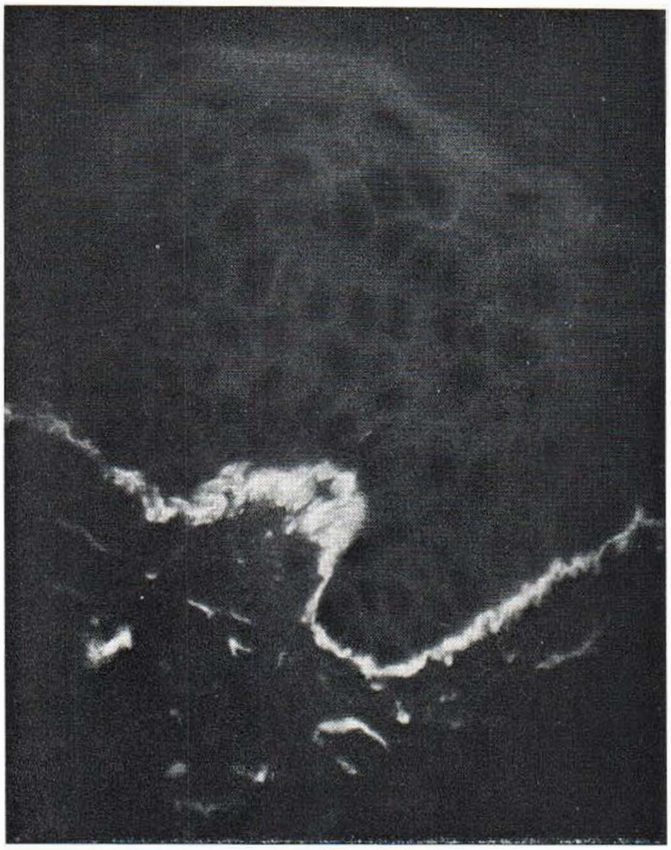

Fig. 1. Direct immunofluorescence. Section from case 1, stained with FITC-labelled anti-C3 (× 780).

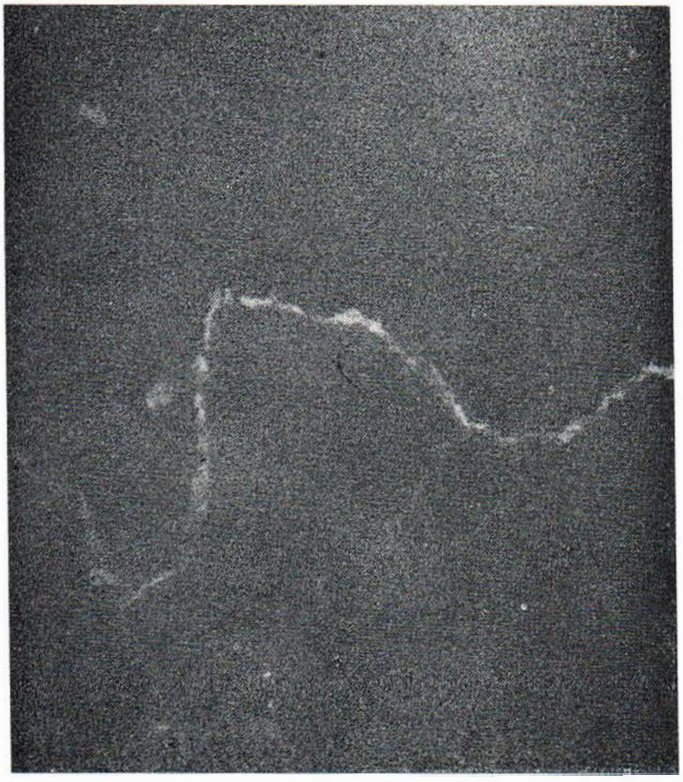

Fig. 2. Direct immunofluorescence. Section from case 3, uninvolved skin, stained with FITC-labelled anti-C3 (× 780).

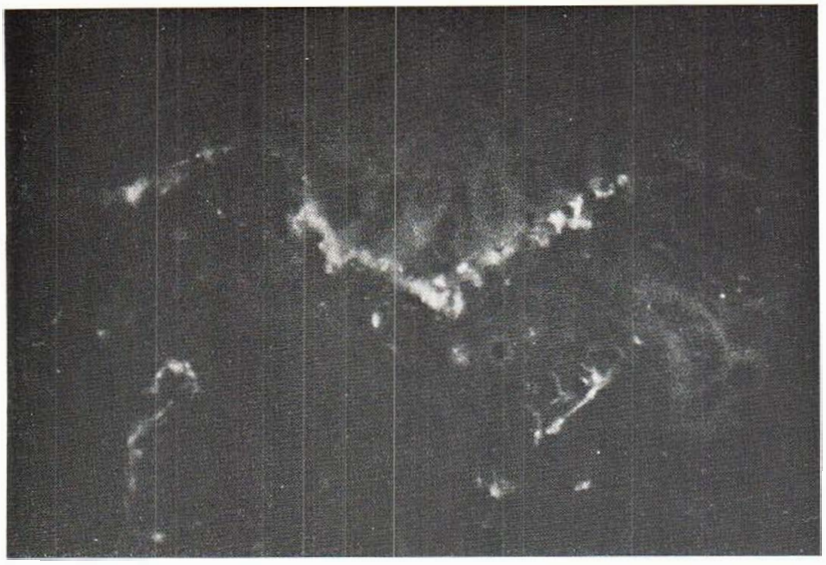

Fig. 3. Direct immunofluorescence. Section from case 2, involved skin investigated 2 months later. Staining with FITC-labelled anti-C3 (× 780).

mechanism of these event is, however, but little understood. It may well be that C3 is bound to the basement membrane and subsequently to an antigen (progesterone?, 18)—antibody reaction, as also postulated in dermatitis herpetiformis (20), bullous pemphigoid (6), lupus erythematosus (19) and pustulosis palmoplantaris (5), though the reason why immunoglobulins can only be demonstrated in smaller amounts remains unclear (probably temporal or technical reasons).

The higher levels of C3 during active skin disease found in 2 of our 4 cases of H.G. may also be referred to as a finding, speaking possibly in favour of the role of the complement (C3) in this disease. It should be mentioned, however, that serum C3 levels were unrelated to the severity of pemphigus and pemhigoid, or to the treatment applied. It was emphasized, however, that these findings cannot exclude pathogenic involvement of the complement in these diseases, due to quantitative factors or the possible role of other complement components (8). In order to gain a better understanding of the significance and mechanism of the intense basal membrane deposition of C3 in H.G., further studies are required.

Present as well as earlier immunofluorescence findings (12) in H.G. differ from those observed in dermatitis herpetiformis or in bullous pemphigoid.

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G. Rajka, M.D.
Department of Dermatology
Rikshospitalet
Oslo
Norway

Addendum. Two works are recently published on this field. Heid et al. (Bull Soc Derm Syph *80:* 110, 1973) found IgA deposition on the junctional area. Bushkell et al. (Arch Dermatol *110:* 65, 1974) describe in their case deposition of IgG, C1q, C3 and properdin at the basement membrane. Furthermore, a circulating antibody against the basement membrane was detected.