# HAEMOLYTIC ANAEMIA IN DISSEMINATED LUPUS ERYTHEMATOSUS

## REPORT OF A CASE

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Anaemia is commonly associated with disseminated lupus erythematosus. Michael *et al.*[1] studied case records of 86 patients with lupus erythematosus, and investigated 25 cases of their own. Of the 111 patients, 87 were anaemic. Although the pathogenesis of the anaemia accompanying this disease is obscure in most cases, in some it is definitely due to abnormal haemolysis of the red blood-cells. Dacie[2] considers it likely that minor degrees of haemolysis would frequently be found if careful erythrocyte survival studies were carried out.

The patient may present clinically as a haemolytic anaemia, with or without other manifestations of lupus erythematosus. The haemolytic anaemia is usually of the auto-immune type, in that the erythrocytes show evidence of sensitization, and anti-erythrocyte antibodies are often demonstrable in the serum. Wiener[3] demonstrated the presence of auto-antibodies in the serum of such a patient by using trypsinized erythrocytes. Zoutendyk and Gear[4] mentioned that the direct anti-globulin test was positive in 4 out of 5 patients investigated by them. Michael *et al*[1] found that 25% of their cases showed erythrocyte sensitization, and 3 out of 25 had overt haemolytic anaemia. Pisciotta *et al.*[5] found 1 out of 7 cases complicated by haemolytic

anaemia associated with a positive direct antiglobulin test. These authors also showed that the anti-erythrocyte antibodies and the L.E. factor in the serum were not the same. Evans *et al.*[6] mentioned 4 cases of lupus erythematosus, all associated with positive direct antiglobulin tests. All 4 cases described by Baikie[7] had positive direct antiglobulin tests, and one was complicated by haemolytic anaemia. In this case warm auto-agglutinins were present in the serum. Dacie[2] carried out direct antiglobulin tests on 9 patients with lupus erythematosus. The reaction was strongly positive in 1 patient suffering from overt haemolytic anaemia, and weakly positive in 5 out of the remaining 8 patients. In each instance the reaction appeared to be of the 'cold' antibody type.

## CASE REPORT

The patient, a European female aged 46, was admitted to hospital for investigation. She complained of a rash on her hands and forearms, which had started 11 months previously. On the day preceding the onset of the rash she had washed clothes in washing-soda, which she considered to be the cause of the rash. The skin lesion became progressively worse, but she did not seek medical advice until 9 months after it had started, when she began to feel generally off-colour, with anorexia and bouts of nausea. She had also developed a persistent cough. These systemic symptoms became progressively worse, and 2 months later she was admitted to hospital.

### *Physical Examination*

The patient was obviously ill. Her skin had a sallow colour, and her temperature was 100·4°F. The conjunctivae were pale and jaundiced. A scaly erythematous rash was present on both hands and forearms, mainly on the dorsal aspects. There were irregular areas of brown pigmentation over the front of the chest and forehead. Examination of the respiratory system revealed a left-sided basal lobar pneumonia; rhonchi and fine crepitations were also audible in the upper lobe on the left side. Her blood pressure was 110/55 mm. Hg, and a soft systolic murmer could be heard over the whole praecordium. The spleen and liver were not palpable. No other significant abnormality was detected on clinical examination.

### *Laboratory Investigations*

1. *Haematological.* On admission there were 1,310,000 erythrocytes per c.mm., with 3·6 g. of haemoglobin per 100 ml. The PCV was 12·0%, and the MCHC 30%. There were 28,200 leucocytes per c.mm., with 68% neutrophils, 0·5% monocytes, 11·5% lymphocytes, 0·5% neutrophil metamyelocytes, 2·0% staff cells, 0·5 plasma cells, and 17% late normoblasts. The neutrophil leucocytes showed a shift to the left. The reticulocyte count was 24·4%. Examination of a blood film showed severe diffuse basophilia, anisochromia, anisocytosis and poikilocytosis.

2. *Biochemical.* The urine contained urobilin (4 plus); bilirubin and urobilinogen were absent. Microscopic examination showed no abnormality. The serum-bilirubin concentration was less than 0·5 mg. per 100 ml., and the total serum proteins 8·5 g. per 100 ml., with 3·7 g. albumin, 4·8 g. globulin and 1·82 g. gamma globulin. Liver function tests gave the following results: Thymol turbidity 6·0 units, thymol flocculation 4 plus, zinc-sulphate turbidity 30·0 units.

3. *Serological.* The following serological investigations were done, all with negative results (these are carried out by us on all cases of 'idiopathic' haemolytic anaemia): Agglutination reactions for typhoid and paratyphoid, rickettsial complement-fixation tests, virus complement-fixation tests, kolmer cardiolipin test for syphilis. Three weeks after admission to hospital L.E. cells were found in large numbers on specially prepared blood-smears, establishing the diagnosis as lupus erythematosus.

4. *Immuno-haematological.* The patient was group A Rh positive (CcDE). The direct antiglobulin test was strongly positive. The quantitative antiglobulin test (Dacie[2]) gave a reaction of the 'warm' antibody type (Table I). Abnormal antibodies were demonstrated in the patient's serum, with the indirect antiglobulin technique as well as with trypsinized and 'ficinated' cells. Although active at 37°C, the antibodies showed a marked increase in activity with a reduction in temperature (Table III). After inactivation of the serum by heating at 56°C for 30 minutes (to destroy the complement) the indirect antiglobulin test was negative on cells sensitized at 4°C for 1 hour, but still positive when sensitized at 37°C (Table IV). The inability of antibodies to sensitize red blood-cells in the absence of complement is considered to be characteristic of the 'cold' type. Incomplete cold antibodies were present to a titre of 64 with the indirect antiglobulin technique, after sensitization of the cells at 4°C (Table II). Agglutination of the enzyme-treated cells occurred despite the absence of com-

TABLE I. THE QUANTITATIVE ANTIGLOBULIN REACTION

| *Dilutions of antiglobulin serum* | | | | |
|---|---|---|---|---|
| 1 *in* 4 | 1 *in* 16 | 1 *in* 64 | 1 *in* 256 | *Control (saline)* |
| + | + | ++ | +++ | negative |

+ denotes weak agglutination; +++ denotes strong agglutination

TABLE II. TITRATION OF THE INCOMPLETE COLD ANTIBODY WITH THE INDIRECT ANTIGLOBULIN TECHNIQUE (SENSITIZATION AT 4°C)

| *Dilutions of the patient's serum* | | | | | | |
|---|---|---|---|---|---|---|
| 1 *in* 1 | 1 *in* 2 | 1 *in* 4 | 1 *in* 8 | 1 *in* 16 | 1 *in* 32 | 1 *in* 64 |
| ++++ | ++++ | ++++ | +++ | +++ | ++ | + |

++++ denotes strong agglutination; + denotes weak but definite agglutination

TABLE III. TITRATION OF THE ANTIBODIES AGAINST ENZYME-TREATED ERYTHROCYTES AT VARIOUS TEMPERATURES (SENSITIZATION FOR 1 HOUR)

| | | *Dilutions of the patient's serum* | | | | | |
|---|---|---|---|---|---|---|---|
| | | 1 *in* 1 | 1 *in* 2 | 1 *in* 4 | 1 *in* 8 | 1 *in* 16 | 1 *in* 32 |
| 4°C | Ficinated | +++ | ++ | ++ | + | + | − |
| | Trypsinized | ++ | ++ | + | + | (+) | − |
| 20°C | Ficinated | + | + | (+) | − | − | − |
| | Trypsinized | + | (+) | − | − | − | − |
| 37°C | Ficinated | + | (+) | − | − | − | − |
| | Trypsinized | (+) | − | − | − | − | − |

+++ denotes strong macroscopic agglutination; (+) denotes weak microscopic agglutination.

TABLE IV. THE INDIRECT ANTIGLOBULIN REACTION CARRIED OUT WITH ACIDIFIED AND INACTIVATED SERUM

| | *Temperature of erythrocyte sensitization* | | |
|---|---|---|---|
| | 4°C | 20°C | 37°C |
| Unacidified normal serum | ++++ | + | + |
| Acidified serum .. | ++++ | ++ | + |
| Inactivated serum .. | — | — | + |

++++ denotes strong agglutination; + denotes weak agglutination.

plement (Table V). Haemolysins could not be demonstrated at 37°C or at room temperature, even after acidification of the patient's serum. As is often the case in auto-immune haemolytic anaemia, group specificity of the antibodies could not be demonstrated; erythrocytes of a selected panel of donors were all agglutinated. Ham's acid-serum and the indirect Donath-Landsteiner tests were negative.

*Treatment and Progress*

During the initial period of hospitalisation, before the diagnosis had been established, the patient was treated with numerous antibiotics. Apart from some amelioration of the respiratory condition little improvement resulted. Treatment with Meticorten (prednisone) was started after 12 days because of the inadequate response to antibiotics. The patient also received a transfusion of 1,500 c.c. of whole blood, without any apparent reaction. The dermatitis responded dramatically to this therapy and after 10 days there was no evidence of abnormal haemolysis. This coincided with the disappearance of the warm auto-antibody, although the incomplete cold antibody was still present, and the direct antiglobulin test still positive.

The patient was discharged about 2 months after admission, with 11·6 g. of haemoglobin per 100 ml. and no evidence of abnormal haemolysis. The direct antiglobulin test was still positive. The persistence of a positive direct antiglobulin reaction is frequently found in treated and clinically cured cases of auto-immune haemolytic anaemia.

## DISCUSSION

In lupus erythematosus the appearance of the skin lesion after exposure to an irritant is not uncommon. King-Smith[8] reported 18 cases in which dermatitis followed external irritation. Development of the skin lesion after exposure to sunlight has also been described.

The immuno-haematological picture was interesting in that the antibody which was active at 37°C was not affected by inactivation of the complement (Table IV), suggesting the presence of two separate antibodies, and not an incomplete cold antibody with a high thermal amplitude. It was this 'warm' antibody which was sensitizing the patient's cells, as evidenced by the 'warm' type of reaction with the quantitative antiglobulin test. The incomplete cold antibody appeared to play no part in the causation of the abnormal haemolysis.

It is interesting to note that inactivation of the complement has little, if any, effect on the ability of the incomplete cold antibody to agglutinate enzyme-treated erythrocytes, and yet completely inhibits its ability to sensitize a saline suspension of erythrocytes to antiglobulin. This indicates that the antibody is thermostabile (as is the case with incomplete warm antibodies) and that only the complement is destroyed by heating at 56°C for 30 minutes. Why complement should be necessary in the one instance and not the other is uncertain.

The haemolytic process in lupus erythematosus responds well to steroid therapy. Michael *et al.*[1] reported good response to such therapy, although the haematological improvement was not quite so dramatic as the clinical improvement. Baikie *et al.*[7] reported amelioration of the haemolytic process, with disappearance of the auto-agglutinin. Pisciotta *et al.*[5] also found that cortisone therapy resulted in improvement of the symptoms, and a gradual decline in the rate of blood destruction.

In this patient treatment with Meticorten resulted in fairly rapid cessation of the haemolytic process, with disappearance of the 'warm' antibody from the plasma. The skin lesion and constitutional symptoms also improved dramatically.

*Comment*

The aetiology and pathogenesis of lupus erythematosus has not been elucidated. That it has an auto-immunologic basis is suggested by: (1) The abnormal facility with which patients produce antibodies. (2) Abnormal response to drugs and sunlight. (3) The frequency with which false positive serological reactions occur. (4) The occurrence of hyperglobulinaemia. (5) The high incidence of cases which show erythrocyte sensitization with multiple antibody production. The relative frequency with which auto-immune haemolytic anaemia complicates the disease is in keeping with this concept.

TABLE V. TITRATION OF THE ANTIBODY ACTIVITY AGAINST TRYPSINIZED AND FICINATED CELLS, BEFORE AND AFTER INACTIVATION OF THE COMPLEMENT

| | | | *Dilutions of the patient's serum* | | | | | |
|---|---|---|---|---|---|---|---|---|
| | | | 1/1 | 1/2 | 1/4 | 1/8 | 1/16 | 1/32 |
| Before Inactivation | 4°C | Ficinated .. .. .. | +++ | ++ | ++ | + | + | — |
| | | Trypsinized .. .. | ++ | ++ | + | — | — | — |
| | 20°C | Ficinated .. .. .. | + | + | (+) | — | — | — |
| | | Trypsinized .. .. | + | (+) | — | — | — | — |
| After Inactivation W | 4°C | Ficinated .. .. .. | ++ | ++ | + | + | (+) | — |
| | | Trypsinized .. .. | + | + | (+) | — | — | — |
| | 20°C | Ficinated .. .. .. | + | + | (+) | — | — | — |
| | | Trypsinized .. .. | + | (±) | — | — | — | — |

+++ denotes strong agglutination, + weak macroscopic agglutination, and (+) agglutination which is only visible microscopically.

As the disease process may precede its clinical manifestations by several years, it must be excluded in every case of 'idiopathic' auto-immune haemolytic anaemia.

Since the haemolytic process usually responds well to steroid therapy, it should always be given a full clinical trial before splenectomy is considered.

SUMMARY

A case of auto-immune haemolytic anaemia complicating lupus erythematosus is presented. The immuno-haematological findings and the treatment are discussed.

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