

of immunoglobulins and complement are simply trapped in fibrin deposits (13), although such a possibility seems to be rather unlikely because of the selectivity of immunoglobulins and of the specific site of deposition, viz. vessel walls and dermo-epidermal junction, the sites most frequently involved in the deposition of immune complexes in the skin.

Although there is much yet to be studied before a definite process of the pathogenesis for PPC is established, our findings seem to indicate at least a new direction of research in this disorder.

## REFERENCES

1. Baart de la Faille-Kuyper, E. M. & Baart de la Faille-Kuyper, H.: An immunofluorescence study of lichen planus. *Br J Dermatol* 90: 365, 1974.
2. Baden, H. P.: Familial Schamberg's disease. *Arch Dermatol* 90: 400, 1964.
3. Bjerke, J. R. & Krogh, H. K.: Identification of mononuclear cells in situ in skin lesions of lichen planus. *Br J Dermatol* 98: 605, 1978.
4. Davis, J. & Lawler, J. C.: Capillary alteration in pigmented purpuric disease of the skin. *Arch Dermatol* 78: 723, 1958.
5. Duncan, S. C. & Winkelmann, R. K.: Early histopathology of the cutaneous capillary fragility test (Rumpel-Leede). *J Cutan Pathol* 6: 1, 1979.
6. Gould, W. M. & Farber, E. M.: A familial pigmented purpuric eruption. *Dermatologica* 132: 400, 1966.
7. Greenwood, K.: Dermatitis with capillary fragility. *Arch Dermatol* 81: 113, 1960.
8. Illig, L. & Kalkoff, K. W.: Zum Formenkreis der Purpura pigmentosa progressiva (unter besonderer Berücksichtigung der Adalin-Purpura). *Hautarzt* 21: 497, 1970.
9. Kazmierowski, J. A. & Wuepper, K. D.: Erythema multiforme: immune complex vasculitis of the superficial cutaneous microvasculature. *J Invest Dermatol* 71: 366, 1978.
10. Lever, W. F. & Schaumburg-Lever, G.: Histopathology of the Skin, 5th edn., 164 pp. J. B. Lippincott Company, Philadelphia, 1975.
11. Pinkus, H. & Mehregan, A. H.: A Guide to Dermatohistopathology, 2nd edn., 201 pp. Appleton-Century-Crofts, New York, 1976.
12. Soter, N. A.: Two distinct cellular patterns in cutaneous necrotizing angitis. *J Invest Dermatol* 66: 344, 1976.
13. Weidner, E.: "Immune complex" in every cutaneous vasculitis with "immune deposits"? *Arch Dermatol Res* 264: 97, 1979.

## Autosensitization to DNA

F. Brauner, W. Brenner and F. Gschnait

Department of Dermatology I, University of Vienna,  
Vienna Austria

Received January 4, 1980

**Key words:** Gardner-Diamond syndrome; Autoerythrocyte sensitization syndrome; Painful bruising syndrome; Differential diagnosis

The spontaneous occurrence of widespread ecchymoses in a patient not exhibiting coagulopathy leads to a spectrum of differential diagnoses including amyloidosis, anaphylactoid purpura, pseudoxanthoma elasticum, Ehlers-Danlos syndrome, autoerythrocyte sensitization and autosensitization to DNA (11). This paper reports on a patient exhibiting the characteristic clinical findings and skin reactivity indicative of DNA autosensitization. The syndrome will be discussed briefly with particular reference to the differential diagnosis to autoerythrocyte sensitization.

## CASE REPORT

A 48-year-old female patient presented with widespread, well circumscribed ecchymoses on the buttocks and extensor and flexor sites of her extremities. The lesions, which erupt spontaneously, are not preceded by trauma and appear initially as red, tender, macular to slightly raised patches, 1–2 cm in size, which sometimes disappear quickly or more often spread peripherally within a few hours, becoming surrounded by a well defined blue ring-shaped area about 1–2 cm in width. Some hours later numerous ecchymoses develop in these sites, which become confluent and finally resemble widespread hematomas (Fig. 1 a, b).

The first attack leading to these skin changes occurred 5 months prior to admission and recurrent eruptions appeared at intervals of 2 to 3 weeks. There were no systemic symptoms such as fever, arthralgia or malaise.

The patient was strumectomized 18 years ago. Seven years ago vitiligo developed on the hands and knees. The patient is now in otherwise good health, she appears emotionally well balanced; the family history is unremarkable.

The general physical examination, ophthalmological examination, chest roentgenogram and thyroid scintigraphy showed no abnormalities. Platelet count, fibrinogen, prothrombin time and partial thromboplastin time were within normal ranges, as were ESR, urinalysis, complete blood count, serum analysis for glucose, blood urea nitrogen, creatinine, sodium, potassium, chloride, calcium, phosphate, uric acid, iron, cholesterol, triglycerides, alkaline phosphatase, SGOT, SGPT and LDH.



Fig. 1 a, b. Eight-hour-old lesions forming well defined blue ring-shaped borders while spreading peripherally on the extensor surfaces of the thighs.

Serum electrophoresis and immunoelectrophoresis, quantitative determination of T and B cells and of serum IgG, IgM, IgA, C<sub>3</sub>, C<sub>4</sub> and C<sub>5</sub> showed no abnormalities. VDRL, ANA, latex test, C-reactive protein and repeated LE cell preparations were negative. The patient exhibited IgG microsomal antithyroid antibodies; antiparietal cell antibodies were not detected.

Biopsies taken from the central red portions of the lesions revealed slight hyperkeratosis, spotty acanthosis of the epidermis and thickening of the basement membrane. There was marked perivascular edema in the upper dermis and a pronounced perivascular cell infiltrate in the upper and lower dermis composed mainly of mononuclear cells and also of eosinophils which were found interspersed between collagen bundles. There was slight extravasation of erythrocytes. Biopsies taken from the bluish borders of the lesions revealed a similar picture but the extravasation of red blood cells was far more pronounced. Direct immunofluorescence of lesional and perilesional skin using anti IgA, IgG, IgM and C<sub>3</sub> antisera proved negative.

#### SKIN TESTING METHODS

Skin testing was performed in the groin areas.

1. *Autologous lymphocytes* were obtained under sterile conditions from heparinized blood using a mixture of sodium metrizoate/ficoll (Lymphoprep<sup>®</sup>) according to Bøyum (1) and the modifications of Thorsby et al. (13). The lymphocytes were washed three times in saline and suspended at  $10^6$  cells per ml. In order to lyse these cells a part of the suspension was subjected to five cycles of alternate freezing in carbon dioxide snow and thawing at 37°C. Both the lysed and the non-lysed lymphocyte sus-

pension (0.1 ml respectively) were injected intracutaneously (i.c.).

2. *Autologous red blood cells* (RBC) (0.1 ml), obtained from heparinized blood, were washed five times in sterile saline, adjusted to a hematocrit of 80% and injected i.c.

*Autologous RBC ghosts* were obtained by hemolysing 10 ml of packed RBC with 100 ml of 0.98 M ammonium chloride (10 min), washed five times in sterile saline and suspended to a hematocrit of 80%. 0.1 ml of this suspension was injected i.c.

4. *Calf thymus DNA* (Boehringer-Mannheim) (0.1 ml) in suspension was passed through a 0.45  $\mu$ m pore-size micropore filter and injected i.c.

5. *Controls*. Autologous lymphocytes, autologous RBC, RBC ghosts and calf thymus DNA were tested i.c. in the groin area of a normal, healthy volunteer. Sterile saline (0.1 ml) was used as a control in the patient.

#### SKIN TESTING RESULTS

Wheal and flare responses occurred in the patient's skin after injection of lymphocytes, lysed lymphocytes and calf thymus DNA. 15 min after injection there appeared an itchy pink infiltrate, about 1 cm in diameter which increased in size to 6 cm after about 6 hours. The control person (normal age-matched, healthy, male volunteer) did not show abnormal reactions. There were no abnormal responses after injection of autologous RBC, RBC ghosts and saline in either the patient or the control individual.

## DISCUSSION

Levin & Pinkus (7) reported in 1961 on a 40-year-old woman who presented clinically with severe ecchymoses in the skin on the extremities, developing rapidly from an erythematous wheal. The reproducibility of the lesions after intracutaneous administration of calf-thymus DNA led to the term autosensitization to deoxyribonucleic acid (7).

Six years earlier, Gardner & Diamond (5) described a form of purpura producing painful bruising in four women, characterized by local pain, swelling and extension of bleeding into adjacent areas. Skin tests performed in these cases indicated an abnormal tissue response of sensibility to red blood cells, the responsible factor being present in the red blood cell stroma. Therefore, this peculiar syndrome has been named autoerythrocyte sensitization (5), which was re-evaluated by Ratnoff & Agle (10) in 1968: since most of their patients had had bizarre behaviour and since hypnosis could reproduce the lesions in some, the designation "psychogenic purpura" has been suggested. These authors, however, failed to reproduce the clinical lesions by intracutaneous injection of autologous whole blood in one-third of their patients and this is comparable to the studies of McDuffie & McGuire (4). The question therefore remains whether skin testing for autoerythrocyte sensitization is indeed erratic in nature (10) or whether some cases reported as autoerythrocyte sensitization with negative skin tests to red blood cells, in fact represented autosensitization to DNA. The present case represents clinically a typical example of autosensitization to DNA. This was confirmed by positive skin tests reactions to DNA and lymphocytes but not to RBC or RBC ghosts.

The skin changes of DNA autosensitization begin as warm, red, raised sometimes tender lesions (7, 8, 11) progressing to ecchymoses after several hours, which seem to be present predominantly at the periphery of the lesion, sometimes appearing as an ecchymotic ring. In contrast, in autoerythrocyte sensitization, as Gardner & Diamond described in their original paper (5), bleeding is the initial sign which in turn is followed by redness, swelling and tenderness (3, 6, 10).

Histologically, in both autoerythrocyte sensitization and DNA autosensitization the most striking feature of the lesions is the paucity of biological changes despite their gross appearance (10): there may be edema of the upper dermis, a mild perivas-

cular mononuclear cell infiltrate and some extravasation of red blood cells. In early lesions of DNA autosensitization, however, an amorphous material may be found between collagen bundles, which stains blue with hematoxylin and gives a strong Feulgen reaction, thus resembling the hematoxylin bodies commonly associated with disseminated lupus erythematosus (7, 8, 11).

Recently, Pinnas et al. (9) described a patient with typical autoimmunization to DNA in whom immunofluorescence studies of the spontaneous lesions (but not of the normal skin) revealed deposits of IgM, C<sub>3</sub>, factor B and properdin at the dermo-epidermal junction. In addition there was a decreased number of T cells and a large null cell population and a defective repair mechanism of UV-damaged DNA in lymphocytes. Since these features are common findings in systemic lupus erythematosus (SLE) the authors concluded that autosensitization to DNA may represent a variant of SLE. Our patient, who presents with the classical clinical features and skin test results of DNA autosensitization, had negative direct immunofluorescence and a normal distribution of lymphocyte subgroups, but exhibited antithyroid antibodies, which, in turn, may be of immunological importance in the development of the patient's vitiligo (2). Despite these immunological findings, obviously a larger number of patients needs to be investigated before there is firm evidence of an immunological basis for the skin lesions in autosensitization to DNA.

## ACKNOWLEDGEMENT

We thank Mr Gamil Eltawil for excellent technical assistance and Professor Franz Greiter, Greiter AG, Weidling, Austria, for financial support.

## REFERENCES

1. Bøyum, A.: Separation of white blood cells. *Nature* 204: 793, 1964.
2. Brenner, W., Diem, E. & Gschnait, F.: Coincidence of vitiligo, alopecia areata, onychodystrophy, localized scleroderma and lichen planus. *Dermatologica* 159: 356, 1979.
3. Davidson, E.: Factitious purpura presenting as autoerythrocyte sensitization. *Br Med J*: 104, 1964.
4. McDuffie, F. C. & McGuire, F. L.: Clinical and psychological patterns in autoerythrocyte sensitization. *Ann Intern Med* 63: 255, 1965.
5. Gardner, F. M. & Diamond, L. K.: Autoerythrocyte sensitization. A form of purpura producing painful

- bruising following autosensitization to red blood cells in certain women. *Blood* 10: 675, 1955.
6. Groch, G. S., Finch, S. C., Dogoway, W. & Fischer, D. S.: Studies in the pathogenesis of autoerythrocyte sensitization syndrome. *Blood* 28: 19, 1966.
  7. Levin, M. B. & Pinkus, H.: Autosensitivity to deoxyribonucleic acid (DNA). Report of a case with inflammatory skin lesions controlled by chloroquine. *N Engl J Med* 264: 533, 1961.
  8. Little, A. S. & Bell, H. E.: Painful subcutaneous hemorrhages of the extremities with unusual reaction to injected DNA. *Ann Intern Med* 60: 886, 1964.
  9. Pinnas, J. L., Tan, E. M., Teplitz, R. L. & Boyer, J. T.: Autosensitization to DNA: evidence for an immunologic basis. *J Invest Dermatol* 72: 157, 1979.
  10. Ratnoff, O. D. & Agle, D. P.: Psychogenic purpura. A re-evaluation of the syndrome of autoerythrocyte sensitization. *Medicine* 47: 475, 1968.
  11. Schwartz, R. S., Lewis, F. B. & Dameshek, W.: Hemorrhagic cutaneous anaphylaxis due to autosensitization to deoxyribonucleic acid. *N Engl J Med* 267: 1105, 1962.
  12. Spiera, H. & Schwartz, R. S.: Autoerythrocyte sensitization reproducible by both autologous red cells and heterologous DNA. *Mt Sinai J Med NY* 37: 108, 1970.
  13. Thorsby, E. & Bratlie, A.: A rapid method for preparation of pure lymphocyte suspensions. In *Histocompatibility Testing* (ed. P. I. Terasaki), 655 pp. Munksgaard, Copenhagen, 1970.

## Antireticulin Antibodies in Psoriasis

Ana Siri, Winston Lozano and Raúl A. Vignale

*Department of Dermatology, Hospital de Clínicas "Dr. Manuel Quintela", Faculty of Medicine, University of the Republic, Montevideo, Uruguay*

Received July 18, 1979

**Abstract.** Antireticulin antibodies were studied in the sera of 32 patients affected by various clinical forms of psoriasis. The indirect immunofluorescence method was used on histological sections of human kidney and mouse liver and kidney. This method was also used with sera from children with coeliac disease and from healthy controls. In 10 these antibodies were positive, while in 22 they were negative, the intensity of the titres varying with the extent of the disease. These results support the notion that immunological phenomena may be involved in the physiopathological mechanisms of psoriasis.

**Key words:** Antireticulin antibodies; Psoriasis; Coeliac disease; Dermatitis herpetiformis; Malabsorption disease

Immune mechanisms may be involved in the pathology of psoriasis. Antibodies to IgG, anti IgG 7, 9, 13, 19), as well as some other factors have also been found in blood (18, 19) and in psoriatic skin (IgG, IgA, IgM) and C<sub>3</sub> (13). Anti-stratum corneum antibodies and anti-IgG were found in the epidermal antigen-antibody complexes (13). The presence of auto-antibodies anti-stratum corneum and complement could be an initial auto-immune phenomenon (3, 12). The formation of antibodies antibasal cells nucleus provokes immune reactions (5).

No signs of delayed hypersensitivity reactions were detected by other methods (4, 10, 16, 24). We have not found in the literature any paper on antireticulin antibodies (ARA) in psoriasis.

## MATERIAL AND METHODS

The sera of 32 patients, 18 male and 14 female, whose ages were between 12 and 61, were studied. The disease had an evolution ranging in time from 5 to 40 years, except for the guttate form which was of only 3 months' duration. ARA was investigated by indirect immunofluorescence methods on cryostatic sections of human kidney and mouse liver and kidney.

As positive controls the sera of 8 children with coeliac disease (CD) were used and as a negative control, the sera of 10 healthy controls. The sera were diluted to 1/10 in PBS, pH 7.2. These diluted positive sera were re-examined in order to determine the antibodies titres. We used goat antihuman IgG, FITC conjugated (Lab. Kallesstad; fluorescein-conjugated antisera). Titre 1/16, F/P weight ratio 6.46 µg/mg, F/P molar ratio 2/65. Dilution 1/10: specificity control, healthy controls, coeliac disease. Observation was performed with a Leitz Ortholux microscope, HBO 200 W/4 Hg lamp, dark camera D:1.20, with the corresponding filters: UG 1 (1 mm), BG 38, barriers K 460.

## RESULTS

According to Seah et al. (20) those sera are positive which leave in the liver a fibrillar fluorescence around the portal space, and a linear one around the sinusoids (Fig. 1), sometimes showing a granular fluorescence corresponding to the transversely cut fibres, while in the kidney they were periglomerular and peritubular, in both human and animal tissues (Fig. 2). Of the 32 patients ARA was positive in 10, and negative in 22. As to the clinical forms, ARA had higher titres in the 5 erythroderma, except for cases 18 and 31 which had low titres because they had been treated with corticoids and cytostatics over a long time. Of the 11 patients with generalized psoriasis, with large plaques in the scalp, chest,