

of glucose (6) did not prolong the life of the bacteria; however, the authors found that highly dilute (0.003%) Tryptose Phosphate Broth, although unable to support growth, protected the bacteria from death for about 2 weeks (6). It is conceivable that many bacteria, given adequate nutrients, could survive the stress of being suspended in Triton X-100 at the strength used.

A word on the *P. acnes* and *S. epidermidis* data is in order. No simple comparison between the experimental and the control data can be made. Fewer bacteria and possibly less "debris" were removed from the skin with the detergent-free solution than with the detergent solution. The difference in the numbers of bacteria removed was probably not such as to make any difference in the survival rate. High densities of bacteria protect each other in "starvation" solutions, presumably because those bacteria that die first lyse out nutrients to protect the remaining bacteria (4). Of greater importance perhaps is the "debris" removed from the skin; it may have protected the bacteria either by supplying nutrients or, more likely, by adsorbing Triton X-100 and lowering its activity (effective concentration). We were not primarily concerned with measuring the theoretical contribution of Triton X-100 to the death of the bacteria. Our main concern was the *in use* situation in order to help investigators know how many bacteria are lost in the wash-solution if delay is introduced between sampling and plating. The organisms died at the same rate whether observed in the first 2 hours of incubation or observed over a 10–12 hour incubation period.

In conclusion, except for *S. pyogenes* which seems to be particularly sensitive to the detergent, the species of bacteria tested can be left in the wash-solution for one half-hour with only about a 20% loss of the *P. acnes*—less for other species. Due to the variability of the data, our estimated half-lives cannot be used as correction factors when longer delays occur between sampling and plating.

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## Linear Dermo-Epidermal IgA Deposition in Bullous Pemphigoid

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Received April 2, 1979

**Abstract.** A case is reported of a patient with a bullous eruption with *in vivo* linear deposition of IgA and complement (C3) along the basement membrane zone of perilesional skin (DIF method). Some data presented here are in favour of the concept that cases of linear *in vivo* deposition of IgA alone, or in combination with other IF findings, might be classified as bullous pemphigoid (BP). Clearing of the lesions due to oral prednisone therapy was accompanied by disappearance of complement deposition, while IgA deposition remained unchanged. Some aspects of tissue injury in this case are briefly discussed.

From several reports in the literature it appears that, even with the aid of immunofluorescence (IF) techniques, in some so-called indeterminable cases of bullous pemphigoid (BP) and dermatitis herpetiformis (DH) a definite classification may be difficult or impossible (1, 3, 6). In view of the characteristic occurrence of IgA deposition in DH, several authors stated that even cases of strictly linear deposition of this class of immunoglobulin along the basement membrane (BM) of the skin and in the absence of circulating BM antibodies should be included in the diagnostic category DH (3,9).

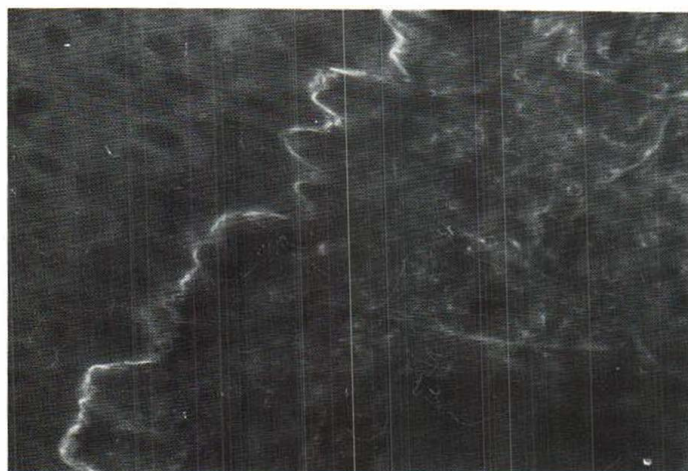


Fig. 1. Pure linear IgA deposition along the BM (DIF method).

However, it has been argued by a few authors that cases of "linear in vivo deposition of IgA" alone or in combination with IgG should rather be classified as BP (7, 2). Recently supporting evidence for this concept has been reported in cases in which linear in vivo staining of IgA was concomitantly present with circulating BM antibodies of the IgA class (8, 6, 10). In the present case of a subepidermal bullous eruption the findings with respect to classification and tissue injury are briefly discussed.

#### CASE REPORT

A man, age 56, developed small, partly grouped haemorrhagic blisters predominantly on the lower abdomen, on the extensor sides of the arms and on the upper legs.

Histologically, subepidermal cleavage at the site of the dermo-epidermal junction and an infiltrate consisting predominantly of eosinophils and also to some extent of neutrophilic leukocytes were demonstrable. Micro-abscesses were not clearly evident.

A direct IF study, by means of highly purified FTC-

labelled antisera and an optimal microscopic system (1), of the erythematous zone of a fresh blister revealed a conspicuous pure linear IgA staining along the BM (Fig. 1). Deposition of IgG, IgM, IgD, IgE was not found. With an anti-complement (C3) serum a strong linear and partly granular staining could be visualized at the BM zone (Table I).

An indirect IF study by means of several heterologous substrates (rat tongue and guinea pig lip) and also a homologous substrate failed to disclose any circulating anti-BM antibodies or other skin-reactive antibodies. However, by using patient's own clinically healthy skin (at a great distance from the lesions and with weak to dubious IgA staining in vivo) as the substrate and low dilution of the patient's serum (indirect IF method) an intense linear fluorescent staining of IgA at the BM zone was demonstrable. The iodine patch test was negative. The xylose absorption test was normal.

Tentatively the patient was administered 100 mg diaminodisulfone (DDS) daily. Although after the first 3 weeks of treatment the clinical activity of the disease had improved, new small blisters developed subsequently. For this reason and also due to the development of methaemoglobinemia and leukopenia it was decided to discontinue this treatment. Instead of sulfone therapy a low dose of prednisone (40 mg daily) was administered orally. This dose proved to be sufficient to clear all lesions. Subse-

Table I. *Relation between direct immunofluorescence (DIF) findings and clinical activity of disease*

|                                          | Untreated                   | After 3 weeks of sulfone treatment | After 3 weeks of prednisone treatment |
|------------------------------------------|-----------------------------|------------------------------------|---------------------------------------|
| Clinical activity                        | Several acute lesions       | Discrete (new) lesions             | No lesions                            |
| Site of biopsy                           | Perilesional skin           | Perilesional skin                  | Site of healed lesions                |
| IgA deposition                           | ++ linear                   | ++ linear                          | ++ linear                             |
| Complement (C3) deposition               | +++ linear, partly granular | ++ linear, partly granular         | —                                     |
| Other immunoglobulins IgG, IgM, IgD, IgE | —                           | —                                  | —                                     |

quently during the various stages of the disease. DIF investigations were performed at comparable sites of the skin (Table I).

Immediately after withdrawing sulfone therapy, the DIF test on newly appearing lesions revealed unchanged IgA fluorescence. The complement staining at the BM was also still clearly visible, though possibly a little weaker. Other classes of immunoglobulins were still absent.

DIF examination performed during the stage of complete remission (3 weeks after oral prednisone therapy) at a comparable site on the skin (healed lesions) revealed an unchanged linear IgA staining, but the complement staining had disappeared completely at this stage.

## DISCUSSION

In the present case, clearing of the skin lesions was immunologically accompanied by the complete disappearance of complement (C3), while IgA deposition remained apparently unchanged. This might indicate a primary pathogenic role for complement in tissue injury, leading to blister formation in this case. It can be speculated that IgA deposition might, however, play an indirect pathogenic role in the induction of alternative complement activation. It is of interest that evidence in favour of both classical and alternative pathway activation of complement in BP has been reported (5). In the present case the indirect IF observation indicates the presence of free circulating BM antibodies, as in BP. The antibody appears to be of the IgA class and seems to be highly specific for patient's own (autologous) skin.

It should be noted that apart from the existence of microabscesses, the exact cellular composition of the infiltrate in the papillary regions might be of importance for differences between BP and DH. It has been reported that, in contrast to BP, the number of eosinophils in cases of DH can be relatively large, though never predominant (7). From this point of view the histopathology in the patient described might be better interpreted as BP. Increased incidence of HLA-B8 has been reported in patients with DH (4). In neither of two cases with linear *in vivo* staining of IgA, recently described (6), could HLA-B8 be demonstrated.

Although it can be said generally that classification based on clinical and laboratory finding and on therapeutic response is hazardous as long as the aetiology of the disease is unknown, the data in the present case with linear IgA deposition might well favour the diagnosis Bullous Pemphigoid.

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## Treatment of Generalized Scleroderma: Updated Results

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Received April 2, 1979

**Abstract.** Long-term treatment of patients with generalized progressive scleroderma by means of inhibitors of connective-tissue biosynthesis brings about total or subtotal regression of dermal sclerosis in 40.8%, partial regression in 33.1%, arrest of progression without regression in 14.8%, while in 11.3% it had no effect whatsoever. The drugs used were D-penicillamine, benzylpenicillin-diethyl-aminoethyl-esterhydro-iodide, glutamine, hydralazine, chlorpromazine, L-dopa, diphenylhydantoin, and corticosteroids. Disease activity before, during and after treatment was indicated by the urinary frac-