

Reaction to Intradermally Applied Phytohaemagglutinin in Asthma Patients in Relation to Corticosteroid Therapy

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Phytohaemagglutinin (PHA) skin test (diameter of induration 24 h following intradermal injection of 1.0 µg purified PHA) was carried out on 23 patients with exacerbated atopic asthma and 28 patients with exacerbated non-atopic asthma. Preselected adult patients had either not previously been treated with systemic corticosteroids or steroid therapy had been suspended for at least 3 months. Nearly all non-atopic asthma patients and patients with atopic asthma previously treated with corticosteroids showed increased reactions to PHA. Patients with atopic asthma not earlier treated with corticosteroids demonstrated normal responses. None of the asthmatics showed a negative PHA reaction. Administration of single depot doses of corticosteroids produced decreased reactivity to PHA in nearly all patients. These results suggest that neither atopic nor non-atopic asthma is in itself associated with impaired PHA skin reactivity but that changes in this reactivity are largely due to the corticosteroid therapy administered to these patients. In relation to PHA reactivity certain effects of this therapy may persist for as long as 3 months after its cessation.

Key words: bronchial asthma; corticosteroids; PHA-skin test.

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CLINICAL ASPECTS

After 24 h, intradermally injected PHA produces induration at the site of injection similar to that seen in skin tests of delayed hypersensitivity with antigens, and correlates with *in vitro* responses to PHA. This test, used to evaluate T cell function in asthma patients, failed to reveal any deficiencies. Administration of steroids almost uniformly decreased the response to PHA, which was increased in the majority of patients earlier treated with these drugs. These data indicate that at least some of the abnormalities in the immune response non-specifically related to asthma in patients with this disorder may be subsequent to the earlier or current steroid therapy. Therefore, it is important to take the possible effects of this therapy into consideration when evaluating the results of immune response tests carried out on these patients.

The role of disturbances in the immune system in the pathogenesis of bronchial asthma is widely recognized (4, 7, 11). However, it is difficult to find a common pattern for these disturbances which concern practically all parts and mechanisms of the immune response. In atopic asthma a major role is assigned to re-

ginnic antibodies and to the T cells responsible for the regulation of their production (7, 11, 14). Several studies have evidenced the problem of the reduced reactivity of asthmatics in various tests of cellular immunity (6, 11, 12). Recently, a simple screening test to measure at least some of the mechanisms of this immunity

has been introduced with the use of a common mitogen phytohaemagglutinin (PHA). In this test PHA is applied intradermally and the reaction is measured after 24 h. The results of this test correlate with both PHA-induced leucocyte migration inhibition, and PHA-induced lymphoblastic transformation (13). Moreover, the test does not require prior sensitization, and a positive response is observed in practically all healthy subjects (8).

The problems of studies of cellular immunity in asthma have been complicated by the fact that asthmatics routinely receive a multi-drug therapy, particularly systemic corticosteroids. Also, the majority of studies of cellular immunity in these patients were performed during corticosteroid therapy and in one recent study (6) such therapy was suspended 3 days prior to testing. Therefore, in these studies it is difficult to distinguish between abnormalities of cellular immunity related to asthma and the effects of corticosteroids, which are potent immunosuppressive agents (10).

Consequently, the aim of the present study was to evaluate the changes in the reaction to intradermally applied PHA in atopic and non-atopic asthma patients following administration of "depot" corticosteroids in relation to former corticosteroid therapy that was stopped at least 3 months prior to the study.

PATIENTS AND METHODS

Fifty-one patients comprising 23 with atopic and 28 with non-atopic bronchial asthma were studied. The atopic asthmatics comprised nine females and 14 males with a mean age of 31 years. The non-atopic asthmatics comprised seven females and 21 males with a mean age of 42 years. Duration of disease in both groups of patients varied between 1 and 15 years and the diagnosis was based on allergic history, allergen skin prick testing, RIST (IgE level) and RAST (presence of specific reaginic antibody). Complex therapy of patients included bronchodilators, expectorants, disodium cromoglycate and antibiotics. Additionally, 18 patients received systemic corticosteroids (prednisone, "depot" corticosteroids) for a period of more

than 3 years. Other patients were either not treated with these drugs or received them only occasionally. None of the patients received systemic corticosteroids during the 3 months prior to the study and this was achieved by widening the complex therapy (increase the dosage of other drugs received by patients, addition of specifics other than corticosteroids, e.g. disodium cromoglycate, not previously used in a given patient). After the study 16 out of 18 patients needed to continue with systemic corticosteroids.

The control group comprised 15 healthy subjects (medical personnel volunteers) in the same age range.

The PHA skin test was performed as described earlier (8, 13). Briefly, 1.0 µg of purified PHA (Wellcome Labs., Beckenham, England) in 0.1 ml of saline was injected intradermally in the right or left forearm and the diameter of induration at the site of injection was measured after 24 h.

Asthmatic patients were tested for PHA skin reactivity prior to and following a single dose of systemic corticosteroids. Twenty-five patients received i.m. 1.5 ml Kenalog 40® (Squibb, Princeton, N.J., USA) and were tested on day 7 following administration, while 10 patients received i.m. 1.0 ml of Celestone® chronodose (Schering, Kenilworth, N.J., USA) and were tested on day 3 following administration. As there were no differences in PHA reaction in relation to type of corticosteroid used, these patients were evaluated together.

RESULTS

The obtained results are shown in Fig. 1. The majority (67%) of patients showed markedly increased reactions to PHA prior treatment. Such reactions were demonstrated by 86% of patients with non-atopic asthma and 43% of patients with atopic asthma. Other patients with exacerbated asthma and prior corticosteroid therapy showed normal response, and none of the patients demonstrated negative response to intradermally injected PHA. Among atopic asthma patients, nearly all those showing increased reaction to PHA had previously been

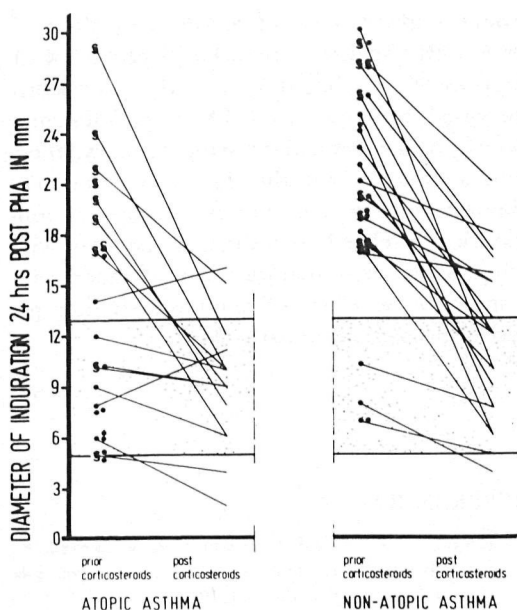


Fig. 1. PHA skin test in bronchial asthma patients in relation to corticosteroid therapy. Spotted area: range of reaction to PHA in healthy subjects. Prior and post corticosteroids: prior and post a single depot dose of corticosteroid. ●: reaction to PHA in patients that were not previously treated with systemic corticosteroids. *: reaction to PHA in patients treated with systemic corticosteroids for more than 3 years and suspended from this therapy for at least 3 months prior to testing.

treated with corticosteroids, while nearly all those showing normal reactions to PHA had not received systemic corticosteroid therapy. Among non-atopic asthma patients, all those previously treated with corticosteroids showed increased reactions to PHA, but similar increased reactions were demonstrated also by the majority of patients not previously treated with corticosteroids. Administration of single depot doses of corticosteroids reduced the elevated PHA responses in the majority of patients to normal values and in some patients, initially with normal responses, produced negative reactions. On the other hand, two patients with atopic asthma demonstrated an increased response to PHA following the administration of corticosteroids. Corticosteroids in the present study were administered because of clinical indications, i.e. exacerbation of bronchial asthma. All tested patients responded

well, with remission of the clinical signs of asthma, to the depot dose of corticosteroids, whose effect on the PHA skin test was under study. For the same reason, several patients, mainly with atopic asthma, did not require corticosteroids during this study. This applied both to patients previously treated with corticosteroids and patients who had not previously received systemic corticosteroids regularly.

These results were also analysed regarding the possible role played by such differences between patients as forms and dosage of received therapy, age, sex, and duration of disease. No consistent differences in the PHA response related to these factors were observed, except for the earlier corticosteroid treatment.

DISCUSSION

In this study it was observed that the majority of patients with exacerbated asthma exhibited increased reaction to intradermal PHA. This concerned nearly all non-atopic patients and patients with atopic asthma previously treated with corticosteroids. Only those patients with atopic asthma who had not previously been treated with corticosteroids showed predominantly normal responses. These observations are in contrast to the results of the most reported studies on the immune status of asthma patients. These studies have shown a decrease in the delayed type hypersensitivity reactions in asthma patients when using recall antigens (candidin, tetanus toxoid, streptokinase-streptodornase) (3, 6, 9). This was associated with decreased *in vitro* reactivity of peripheral blood lymphocytes to T cell mitogens (6, 11). The discrepancies between these results and ours may be secondary to the differences in the physiological significance of the tests used as well as to the selection of different groups of asthmatics for the study, and to differences in the advancement of the diseases and administered therapy. Although the PHA skin test has many similarities to the classic type IV hypersensitivity reactions (1), it differs in that it does not require prior sensitization and utilizes mitogen and not antigen. Asthmatics form a heterogenous group of subjects both as regards characteristics of

their disease and treatment. Additionally, altered reaction to PHA in exacerbated asthma, i.e. prior to the administration of depot corticosteroid, might have been due to the effect of mediators of allergic reactions. However, as no studies are available on the relationships between the PHA skin response and the plasma levels of substances such as histamine, slow reacting substance of anaphylaxis, etc., this problem will require future consideration. In the present study, a prior history of prolonged corticosteroid therapy was of particular significance. In our opinion corticosteroids were the most potent immunomodulators administered to these patients and their effects regarding imbalance in the immune system may persist even months after intake has ceased. Results of the initial PHA skin testing in patients with atopic asthma largely confirmed the significance of such thinking. Additional confirmation of the importance of considering the effects of corticosteroids came from the results of PHA skin testing following the administration of single depot doses of corticosteroids. An almost uniform decrease in the skin reactivity to PHA in asthma patients was observed, which suggests that at least a part of the decreased immune reactivity in these patients observed in earlier studies might have been related to the effect of therapy and not to the disease itself. The results may be partly explained by the observations on T suppressor/T helper cell imbalance produced by both the asthma itself and by corticosteroids. As it was observed, particularly atopic asthma is associated with decreased activity of T suppressor cells (2, 12, 14). It is known that these cells regulate both B and T cell responses. Therefore, it is possible that their defect may on the one hand allow for increased IgE level and, on the other, permit the mounting of an increased reaction to intradermal PHA.

Corticosteroids preferentially decrease numbers of T helper cells and only slightly affect the numbers of T suppressor cells (5). The direct action of corticosteroids on T helper cells would, according to this concept, be responsible for the decrease in the skin PHA response shortly after administration of these drugs. On the other hand, the recovery of T helper cells

from the direct action of corticosteroids in subjects with persisting reduced function of T suppressor cells might lead to the prevalently increased responses to PHA in patients previously treated with these drugs. Finally, these results indicate that the data concerning the abnormalities in the immune response non-specifically related to asthma in patients with this disorder may require reconsideration with respect to the effect of administered therapy, and corticosteroids in particular.

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