

Immediate-type hypersensitivity to reactive dyes

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Summary

Reactive dyes have gained extensive use in recent years. This paper reports four cases of immediate-type occupational allergy to these compounds. All the patients had had symptoms of asthma and allergic rhinitis, and had been weighing dyes for a minimum of 2 years. The skin prick tests were positive, and nasal and bronchial provocation challenges also produced positive immediate reactions. A high serum titre of specific IgE could be demonstrated for at least one dye by the radioallergosorbent test. The identification of specific IgE shows that the mechanism of the hypersensitivity is immunological, reactive dyes acting probably as haptens.

Introduction

The first reactive dye for cotton was marketed in 1956 by the Imperial Chemical Industries Ltd (ICI), and now more than 100 of them are available. They are used on a large scale in industry because of their bright colours and firm chemical fixation to cellulose, protein and polyamide fibres. The dye molecule contains a chromophobic system (usually an azo or anthrachinon group), hydrophilic groups to improve water solubility and reactive groups such as a heterocyclic halogen grouping or an aliphatic chain with reactive groups. The reactive groups are capable of forming covalent bonds with hydroxyl and amino groups in the fibre molecule.

The reaction of the dye with the fibre is presented in Fig. 1. The process is as follows: the material is first heated to 40°C; saline is then added, followed in the following order by a dye-water solution and a soda solution; finally the extra dye is washed away with water. About 1 hr is needed for the completion of the process.

We have observed four cases of immediate-type sensitization to different reactive dyes in work situations. These cases represent a new type of occupational asthma.

Materials and methods

Skin sensitivity to protein allergens was tested with the prick or scratch technique and the reactions were graded according to the method of Aas & Belin (1973). Approxi-

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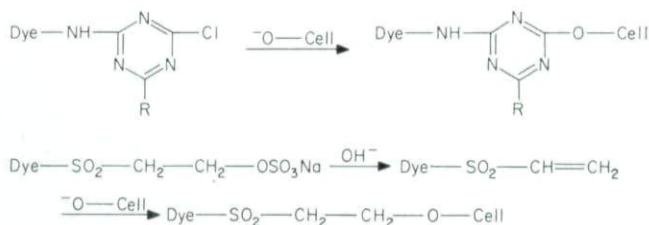


Fig. 1. Two reaction types in dyeing with reactive dyes.

mately seventeen antigen extracts, including animal epithelia, pollens, house dust and mites, were used.

Skin sensitivity to the suspected dyes, in powder form, was tested with the scratch technique. Each dye was also tested on a minimum of three control persons by adding it in powder form to a drop of sterile water with a wooden stick. All control tests with sterile water were negative, and a histamine control (1/1000) was also used.

Nasal challenges with the dye powders were made with the method used by Holopainen, Tarkiainen & Malmberg (1976), but without the use of rhinomanometry.

The bronchial provocation challenge tests were performed with lactose powder as the placebo and with 5–10% dye-lactose mixtures, according to the technique described by Pepys, Pickering & London (1972). Each challenge was made on a separate day and started about noon. The occupational exposure conditions were simulated in a 10 m³ challenge chamber.

A Wright peak-flow meter was used to measure ventilatory response immediately before the challenge, and then every 15 min up to 1 hr. Measurements were continued for as long as 8 hr, and then at 4 hr intervals until the next morning. A 20% drop in peak expiratory flow (PEF) was regarded as a significant positive response that clearly differed from the PEF values of the control day and those after the placebo challenge test.

Total serum immunoglobulin E (IgE) was determined with a Phadebas IgE kit (Pharmacia Diagnostics). The normal range for the assay is 0–160 u/ml. IgE antibodies specific to the dyes were determined with the radioallergosorbent test (RAST). For use in the test, filter paper discs were dyed at 40°C by a modification of the method suggested for textile dyeing by the manufacturers. In the process, the discs were first suspended in a 50 g/l sodium chloride solution. Discs were suspended in a 20 ml of the dye solution for every gramme of material to be dyed; thereafter the discs were transferred to a 3% dye solution using a 20 g/l sodium carbonate solution. All extra dye was finally washed from the discs with water. RAST reagents other than the discs were obtained from Phadebas RAST kits (Pharmacia Diagnostics). The results were graded with the semi-quantitative RAST scale of 0–4 recommended by the manufacturer. A score of 1 or higher is generally regarded as a positive response. Discs were prepared with four different dyes. Each type of disc was tested with sera from three atopic controls with no history of dye hypersensitivity. All control tests resulted in a RAST score of 0.

Case reports

H.S., a 54-year-old male, had had no previous diseases and had smoked for 35 years. His daughter had atopic dermatitis. The patient had worked preparing dye solutions for many years. In 1974 he got a new job in which 95% of the dyes were in powder

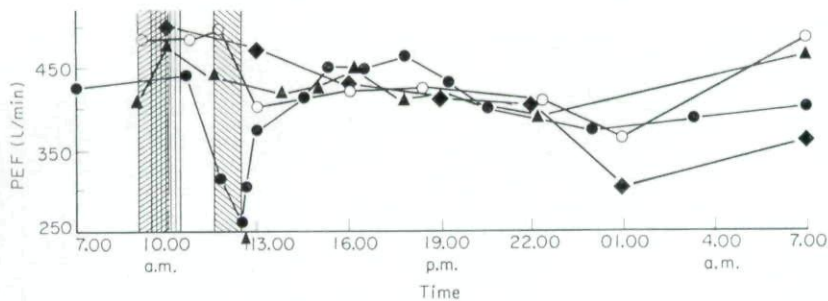


Fig. 2. Patient H.S. Immediate-type asthmatic reaction with Levafix brilliant yellow E-36 (dye 1). Test with the placebo and dye 2 were negative. Hatched bars show test period: (▨) placebo; (▩) dye 1; (▧) dye 2. For the curves: (◆) no test; (▲) placebo; (●) dye 1; (◻) dye 2.

form, including reactive dyes. The dye mixing soon produced rhinitis and sneezing. After 8 months, asthmatic symptoms developed that were relieved by bronchodilator therapy. Away from work he had no symptoms. In a local hospital, bronchial asthma was verified and an occupational origin was suspected. The patient was referred to the Institute of Occupational Health in Helsinki. Histamine produced a marked bronchial obstruction. The patient's diurnal PEF variation was 40%. There was eosinophilia in both his blood and sputum. He had no salicylate or strain-induced asthma. The usual protein skin tests were negative. Scratch tests were made with the twelve most suspect dyes. Two gave positive reactions. The IgE value was 2600 u/ml. The placebo bronchial provocation test was negative, but the test with Levafix brilliant yellow E-36 produced an immediate asthmatic reaction (Fig. 2). The nasal provocation test was positive, too. A high titre of specific IgE was also found for this dye.

V.T., a 58-year-old male, had worked in the textile industry since 1935, for 41 years, weighing reactive dyes for the last 6 years. For years he had suffered from lupus erythematosus disseminatus, which was symptomless at the time of the study. He had no atopy in his family or personal history. For 2 years he had experienced rhinitis and cough at work, but was well on Sundays and holidays. For the year before the investigation he had had asthmatic symptoms. He had no dyspnoea at rest and his cardiac status was compensated. There was mild blood eosinophilia. Histamine

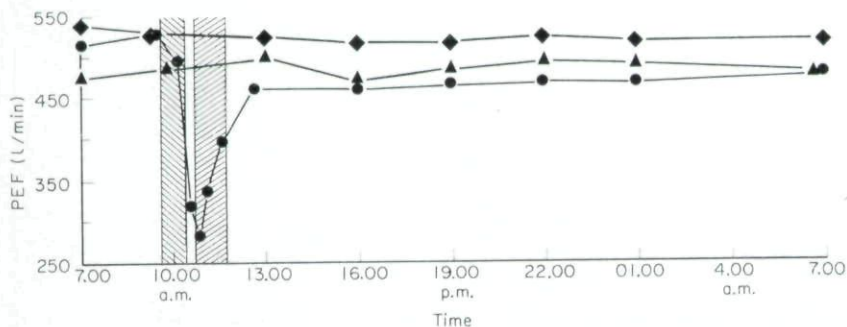


Fig. 3. Patient V.T. Immediate asthmatic reaction to Drimaren brilliant yellow K-3GL. The placebo test with lactose was negative. Hatched bars show test period: (▨) placebo; (▩) dye. For the curves: (◆) no test; (▲) placebo; (●) dye.

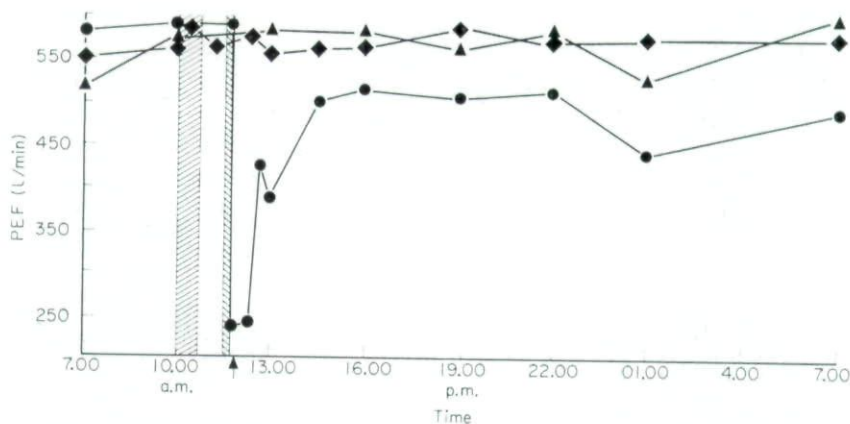


Fig. 4. Patient S.A. Immediate asthmatic reaction to test with Cibachrome brilliant Scarlet 3R. The placebo test with lactose was negative. Hatched bars show test period: (▨) placebo; (▩) dye. For the curves: (◆) no test; (▲) placebo; (●) dye. Arrow shows point of isoprenaline inhalation.

produced only mild bronchial obstruction. Bronchial provocative challenge tests with lactose and formaldehyde were negative. Drimaren brilliant yellow K-3GL, which gave a positive skin test, produced the immediate type of bronchial obstruction (Fig. 3) and a positive nasal test. A high titre of specific IgE was found for that dye, and also for Drimaren brilliant blue K-BL, which also produced a positive skin test.

A.S., a 44-year-old non-atopic male, had weighed dyes for 3 years. After 1 year he developed symptoms of rhinitis and then asthmatic attacks during work days. Asthmatic rhonchi were heard and he was sent for further examination. Histamine produced mild bronchial obstruction. Exercise induced mild bronchial obstruction. There was no salicylate intolerance. The IgE level was 20 u/ml. There was mild blood eosinophilia. Skin tests with usual allergens were negative, but three azo dye powders

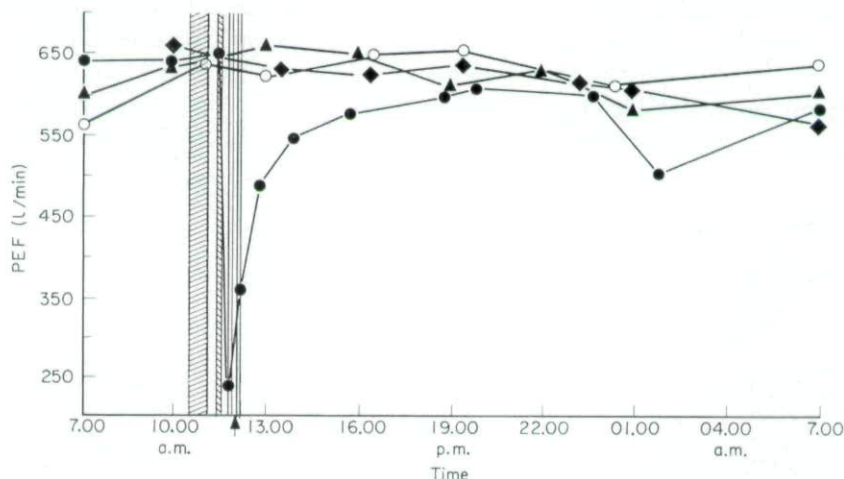


Fig. 5. Patient V.H. Immediate asthmatic reaction to test with 10 g Drimaren brilliant blue K-BL (dye 1) in 190 g lactose. The lactose control and the test with another reactive dye (dye 2) did not differ from normal values. Hatched bars show test period: (▨) placebo; (▩) dye 1; (▮) dye 2. For the curves: (◆) no test; (▲) placebo; (●) dye 1; (○) dye 2. Arrow shows point of isoprenaline inhalation.

produced positive scratch tests. The bronchial provocative challenge test with lactose was negative. The test with Cibachrome brilliant scarlet 3R produced immediate bronchial obstruction (Fig. 4). The test with another dye was negative, as was the skin test with it. Nasal provocation with the first dye was positive. The RAST technique produced negative results for this dye.

V.H., a 30-year-old male, had worked for 5 years in the dye industry and had weighed dyes for the 2 last years. There was no atopy in his personal or family history. He had smoked for 8 years. During his military service he had suffered from urticarial eczema of unknown origin. 1.5 years before the study he noted marked rhinitis at work, and asthmatic symptoms also occurred. At the time of the investigation, he had not been exposed to dyes for several months. Histamine caused no obstruction. There was no effort-induced asthma. Of the usual allergens only house dust produced a positive skin test. Scratch tests were done with eleven dyes, and two gave strong positive results and one a mild result. He had mild blood eosinophilia. His IgE level was 400 u/ml. The bronchial provocation test with lactose was negative. Drimaren brilliant blue K-BL gave a positive skin test and a positive nasal provocation test, and produced an immediate 62% bronchial obstruction 15 min after the beginning of the bronchial provocation test. No obstruction was caused by the provocation test with dyes that gave negative skin tests. There was a high titre of specific IgE to Drimaren brilliant blue K-BL in the serum of the patient.

Table 1. Occupational and clinical data

Patient	Exposure time (years)	Blood eosinophilia ($\times 10^{-9}/l$)*	Total IgE	Skin tests with usual allergens	Spirometric metric values	Histamine challenge
H.S.	2	0.48	2600	—	Normal	+
V.T.	6	0.37	220	—	Normal	+
A.S.	2	0.44	20	—	Normal	+
V.H.	5	0.49	400	±	Normal	—

* Absolute values.

Table 2. Results of skin, bronchial, nasal provocation and RAST tests

Patient	Reactive dye	Skin test (scratch)	Challenge tests		Specific IgE RAST score
			Nasal	Bronchial	
H.S.	Levafix brilliant yellow E-36	+++ ps	+	+	2
V.T.	Drimaren brilliant yellow K-3GL	+++	+	+	2
	Drimaren brilliant blue K-BL	++			1
A.S.	Cibachrome brilliant scarlet 3R	+++ ps	+	+	0
V.H.	Drimaren brilliant blue K-BL	+++	+	+	2

ps = pseudopodia.

Results

The mean laboratory and allergy results of the preliminary investigations are shown in Table 1. Three out of the four patients had both increased total serum levels for IgE and increased blood eosinophil values, which may be considered as an indication of immediate hypersensitivity. All skin tests with protein allergens were negative, with the exception of one positive to house dust. On the other hand, tests to detect immediate hypersensitivity to dyes were positive for the dyes listed in Table 2.

Discussion

Classical immunology of asthma and allergic rhinitis has been developed around protein allergens and the concept of atopy (Coca & Cooke, 1923). All classifications, diagnostic procedures and therapeutic measures have been influenced by this approach. However, industrial chemicals account for a significant proportion of occupational asthma and rhinitis. Compounds of low molecular weight (usually less than 500) are described as causing different types of hypersensitive reactions (Popa *et al.*, 1969). Usually, in such cases, one is able to characterize the mechanisms only clinically by provocation testing.

For many years skin tests and passive transfer were the only means of identifying the immediate reagin-mediated allergic reaction. Introduction of the RAST technique (Wide, Bennich & Johansson, 1967) offered a direct method for measuring specific IgE-class antibodies.

For some time now chemicals, like phthalic anhydride, sulphonechloramides and tannic acid, have been known to cause immediate hypersensitive reactions (Kern, 1939; Feinberg & Watrous, 1945; Johnston *et al.*, 1951). Classical methods were used. In aqueous solutions these chemicals produced passively transferable immediate weal and flare skin tests. Also reactions with ammonium hexachloroplatinate are much the same type (Pepys, Pickering & Hughes, 1972).

Recently, specific IgE-class antibodies to phthalic anhydride were demonstrated by means of the RAST method (Maccia *et al.*, 1976). In the present study we have described four patients with immediate hypersensitivity to reactive dyes. With the RAST technique we could demonstrate the presence of dye-specific IgE antibodies as well. Symptoms, skin test and challenge test results resembled those usually encountered in allergy to protein dusts. Thus these compounds, like proteins, penicillin and the before-mentioned chemicals, can cause IgE-mediated, type I allergy. It is known that penicillin metabolites act as haptens combining with body proteins, which become allergens. It seems probable that reactive dyes act through a similar mechanism.

Chemicals causing immediate reactions are exceptional in their behaviour. Most chemical agents do not produce direct immunological reactions, e.g. aluminium sulphate, ethanolamines (Sterling, 1967; Pepys & Pickering, 1972; Vallieres *et al.*, 1977) and many others, including isocyanates and formaldehyde. Usually the mechanisms can be characterized only clinically by provocative challenge testing. Even immediate obstructive bronchial reactions are not definitely indicative of the IgE-mediated mechanism if no other immunological information is available. Late reactions and an accentuation of the diurnal variation of ventilatory function are observed when provocative challenges are made with chemicals causing asthma. Placebo and control periods are needed to rule out cases due to non-specific or toxic irritative

effects. Histamine testing for bronchial hyper-reactivity is not as good as in atopic asthma caused by protein allergens (Alanko, Keskinen & Saarinen, 1977).

The use of various highly reactive chemicals in modern industry for 'high-speed' and 'low-cost' production is producing new health hazards.

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