

Castor Bean Allergy Among Workers in the Felt Industry

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We describe occupational allergy to castor bean in workers in a felt manufacturing plant. Twenty-six (37%) of the workers complained they were affected by the felt and were examined by us. Of these, 12 were considered to have occupational allergy. These 12 subjects had raised specific IgE levels to both felt and castor bean extracts. In addition, three subjects without occupational allergy had raised specific IgE. The presence of castor bean allergens in the felt was suggested by the correlation between the RAST scores to the felt and castor bean and confirmed by RAST inhibition experiments. The RAST results correlated well with the results of skin prick tests to felt and castor bean extracts. In addition it was found that atopy did not predispose the workers to castor bean allergy.

Key words: castor bean; immunoglobulin E; occupational medicine; radio-allergo-sorbent-test.

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Castor bean, *Ricinus communis*, is a very potent sensitiser and is able to provoke allergic symptoms at extremely low concentrations. As a result allergy to castor bean has been found in a number of cases where the subjects were unaware of their exposure to this agent. One of the first reports was by Frigley & Rawling (3) in 1950, who described sensitisation to castor bean in coffee workers handling sacks which had previously been used for the transport of castor beans. In a subsequent study (4) of coffee workers it was found that workers were sensitised to both castor and coffee beans. Sera from these subjects had raised IgE to both allergens, but RAST inhibition showed the allergens were immunologically distinct. More recently castor bean allergy has been demonstrated in upholstery workers in a furniture factory, who were handling felt made from sacks previously used for the transport of castor beans (7).

This paper describes a further occurrence of castor bean allergy in the felt industry. The

factory is involved in the production of felt from a mixture of waste natural fibres. Included in this mixture is old sacking. A number of employees complained of allergic symptoms, conjunctivitis, rhinitis and chest tightness on exposure to some batches of this material. Enquiries revealed that some of the sacking had previously been used for castor beans. Our immediate response that castor bean was the causative agent was confirmed by positive prick test and raised RAST scores to a castor bean extract.

MATERIAL AND METHODS

Subjects

Twenty-six of the 70 employees at the plant complained that they were affected by the felt. Blood was taken from 23 subjects, the serum separated and stored at -20°C. On a sub-

sequent visit to the factory 20 subjects were available for prick tests.

Preparation of extracts

Castor bean. A 20% w/v castor bean extract was prepared by mixing ground castor beans with phosphate buffered saline (PBS) overnight at 4°C. Insoluble material was removed by centrifugation and the supernatant dialysed against a five fold excess of PBS overnight at 4°C. The extract was then stored at -20°C.

Felt. A felt sample was obtained from the factory and a 1% w/v extract prepared. The felt was allowed to stand overnight in PBS at 4°C, then wrung out, and the resultant solution centrifuged. The supernatant was exhaustively dialysed against 0.02 M ammonium bicarbonate and then lyophilised. The lyophilised material was reconstituted in PBS to give a 50% w/v extract, which was stored at -20°C.

Radio-allergo-sorbent-test (RAST) assay

Felt extract and castor bean extract allergen discs were prepared by coupling the extracts to cyanogen bromide activated discs as described by Ceska et al. (1). The concentration of allergen specific IgE in serum samples was determined by incubating 200 µl of a 1:4 dilution of sera with an allergen disc for 18 h at room temperature. After washing the disc, bound IgE was detected by an overnight incubation with 50 µl of ¹²⁵I labelled anti-human IgE (Pharmacia, Uppsala, Sweden). After removal of unbound ¹²⁵I, the ¹²⁵I bound to the disc was measured with an LKB rackgamma II counter. All assays were performed in duplicate. The results were expressed as RAST scores, which are defined as:

$$\frac{\text{cpm bound to disc incubated with test serum}}{\text{cpm bound to disc incubated with known negative serum}}$$

A score of two or more has been found to be greater than three SD above the mean control serum count, and was, therefore, taken as indicating the presence of allergen specific IgE.

RAST inhibition

RAST inhibition was performed by a method adapted from that of Yman et al. (8). Briefly this was as follows, aliquots of a dilution of a serum with high IgE concentration to the allergen under test were mixed with increasing amounts of soluble phase allergen, then a solid phase allergen disc added, and the IgE bound to the disc measured in the same way as for the RAST assay. The results were expressed as a percentage of the counts bound to a disc incubated with serum in the absence of added soluble allergen.

Total IgE

Total IgE was determined using a Phadebas PRIST kit (Pharmacia, Uppsala, Sweden) according to the manufacturer's instructions.

Skin prick tests

Castor bean and felt extracts were prepared for prick testing by dialysing an aliquot against Coca's solution (2) and then filtering 2.5 ml through a Millex 0.22 µm filter (Millipore, Ltd.) into 2.5 ml of sterile glycerol containing 0.4% phenol. Serial dilutions were made, under sterile conditions, into Coca's solution containing 50% glycerol to give prick test solution of 0.001% for castor bean and 0.1% for felt. Prick test solutions to B2 grass pollens, cat fur, and *D. pteronyssinus* were purchased from Bencard (Brentford, England). Prick tests were performed as described by Pepys et al. (6) and a wheal of 2 mm or greater regarded as positive. Subjects with a positive prick test to one or more of grass pollen, cat fur or house dust mite were defined as atopic.

RESULTS

Clinical features

The 26 subjects were examined by two of us (R.H.) and (A.W.) and 12 gave clinical histories suggesting work-related allergy. All 12 had symptoms of rhinitis and conjunctivitis. In addi-

Table 1
Summary of clinical history, RAST and prick test responses in workers handling felt

Subjects with history of work-related allergy and +ve RAST scores	Total IgE iu/ml	RAST score to castor bean	RAST score to felt	Prick test to castor bean	Prick test to felt
1	190	24.5	22.5	+	+
2	520	16.5	24.7	+	+
3	380	12.2	13.7	+	+
4	ND	15.8	16.6	+	+
5	133	9.5	7.0	+	+
6	64	8.8	4.4	ND	ND
7	68	8.5	13.0	+	+
8	176	8.1	9.4	+	+
9	320	7.7	2.6	-	-
10	80	7.3	4.8	+	+
11	24	5.6	2.4	+	+
12	51	5.1	4.2	+	+
Subjects without history of work-related allergy and +ve RAST scores					
13	37	9.2	12.3	+	+
14	210	8.2	5.8	+	+
15	22	4.7	2.2	-	-
Subjects without history of work-related allergy and -ve RAST scores					
16	21	0.9	0.8	-	-
17	21	1.1	0.9	-	-
18	17	0.9	0.8	-	-
19	11	0.8	0.7	ND	ND
20	28	0.8	0.9	-	-
21	2	1.0	0.9	-	-
22	4	0.9	0.7	ND	ND
23	160	2.0	1.0	-	-

tion seven complained of wheezing: in four this occurred within a short time of exposure to the felt, while in the other three it occurred on waking the morning following exposure. The symptoms commenced approximately 6 months prior to our investigations and followed the introduction of the waste natural fibre into the factory a few months previously. Of the 20 subjects who were available for prick testing, eight (40%) gave a positive response to one or more of grass pollen, cat fur or house dust mite, and were, therefore classified as atopic. Of the 11 in this group with work-related allergy five (45%) were atopic. Statistical analysis, using the

Chi-squared test, showed that prevalence of work-related allergy in the atopic group was not significantly different from the non-atopic group.

Immunological results

The RAST scores obtained to the felt extract discs and castor bean extract discs are shown in Table 1. All the 12 subjects with histories of work-related allergy gave positive RAST scores to the felt and castor bean extract discs. In addition three subjects with no history of work-related symptoms had positive RAST to both

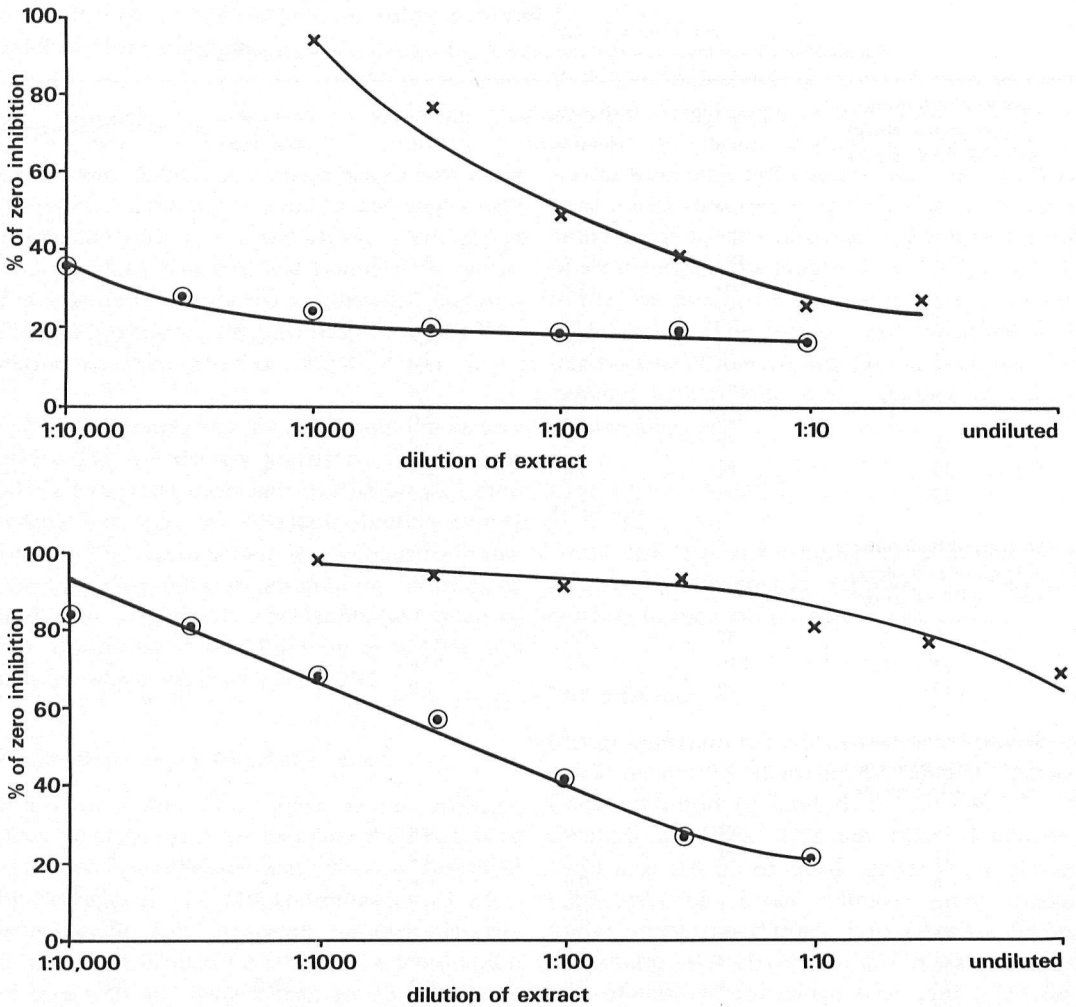


Fig. 1. RAST inhibition of (upper) felt allergen discs and (lower) castor bean allergen discs by $\times$ — $\times$ felt extract and $\circ$ — $\circ$ castor bean extract.

castor bean and felt. Comparison of the RAST scores to felt and castor bean discs in the individual subjects showed that the two were very closely correlated (correlation coefficient $r = 0.96$, $P < 0.001$) Table 1. This suggested that the castor bean and felt extracts had common allergenic determinants. In an attempt to verify this suggestion RAST inhibition experiments were performed.

200 μ l aliquots of 1:10 dilution of a serum pool from subjects with high RAST scores to felt and castor bean discs were incubated with a felt allergen disc and various dilutions of either

felt or castor bean extract. In the absence of added inhibitor binding to the felt disc was 3785 ± 85 cpm. Fig. 1a shows that the castor bean extract gave considerable inhibition of the binding of specific IgE in the sera to the felt disc at the lowest dilution tested 1:10,000 (1082 ± 112 cpm bound), whereas with the felt extract similar inhibition was not observed until a dilution of 1:30 was reached. This result confirms that the felt and castor bean have common allergenic determinants, and suggests that the only allergen in the felt to which the subjects have responded are those derived

from castor bean. A second experiment in which felt and castor bean were used to inhibit binding to castor bean discs demonstrated that felt could inhibit castor bean discs, but only at high concentrations (Fig. 1b). In this experiment the serum was used at a 1:40 dilution, binding to an uninhibited disc was 5662 ± 192 cpm and addition of undiluted felt extract reduced the binding by 2575 ± 239 cpm.

Recent reports (5) that total IgE levels can influence the RAST scores to castor bean discs prompted us to investigate the effect of total IgE in our RAST system. Using sera from unexposed subjects with total IgE values ranging from 90–1650 iu/ml, we determined RAST scores to castor bean. The results showed that significant false positive RAST scores to castor bean did not occur below total IgE levels of 1,000 iu/ml. The total IgE was determined for the test serum samples, and the highest value found was 520 iu/ml (Table 1). Thus the RAST scores obtained with these sera are not influenced by total IgE levels.

Prick tests

Castor bean extract for prick tests was not used at a concentration higher than 0.001% owing to the toxicity of this material. Since RAST inhibition had shown considerable differences in allergenicity between castor bean and felt, the felt extract was used at 0.1%. Twenty subjects (11 with work-related allergy) were prick tested. Ten of the 11 subjects with allergy gave positive prick tests to both felt and castor bean extracts, and one gave a flare only to castor bean. The subject would probably have given a positive response if a lower dilution of castor bean had been used. Of the three subjects with positive RAST, but without work-related allergic symptoms, two also had positive prick tests; the remaining six subjects were negative. These results gave an agreement between RAST and skin prick test of 90% (Table 1).

DISCUSSION

The results described in this paper further emphasise the potent nature of castor bean as a

sensitiser, and the need for the occupational physician to be aware of the possibility of sensitisation to castor bean in subjects handling waste natural fibres. In this survey all the subjects examined had complained of symptoms on exposure to the felt. However, in a number of them the symptoms were vague and not compatible with those associated with Type I hypersensitivity. In the 12 subjects who gave a clear history of work-related allergy RAST was positive to both felt and castor bean, and 10 had positive prick tests to felt and castor bean. A further two subjects gave a positive RAST and prick tests to felt and castor bean and another positive RAST only; in these cases it is likely that, although the clinical history did not suggest work-related allergy, they were in fact sensitised.

The material used to manufacture the felt consists of waste carpet, rags and wool as well as hessian sacks. Thus the mixture contains a large number of proteins, many of which would be allergenic, as well as contaminants such as moulds and mites. However, the close correlation between the RAST scores to the felt and castor bean strongly suggests that allergen derived from castor bean are the only allergens to which the subjects have responded. The inhibitions of the binding of antibody to the felt disc by the castor bean confirms this suggestion.

Since castor bean is highly toxic, the use of provocation tests to confirm diagnosis is not feasible, and prick tests need to be done at very low concentrations of extract. Thus where the clinical history raises the possibility of castor bean allergy, we suggest that RAST is a suitable method to confirm the diagnosis.

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