

stools. We are now studying this possibility by means of a prospective case-control study, and also hope to establish whether the mortality in toxin positive patients is a result of the *C difficile* carriage, an associated premorbid marker, or an independent event.

The high level of carriage of *C difficile* among elderly patients is not entirely unsuspected. Many of the patients had received antibiotics in the previous month, and this is recognised as the major risk factor for acquiring the pathogen.^{10,12,24} The increased frequency may be due to other factors as well. Population-based studies in Sweden show that in adults the frequency of *C difficile* toxin recovery correlates directly with age.¹¹ This may reflect changes in local immunity,²⁶ decline in neutrophil function²⁷ or aspects of cellular immunity,²⁸ or changes in intestinal physiology.²⁹ Finally, these elderly patients are in a closed environment with the same enhanced potential for an outbreak as other individuals in similar settings.¹³⁻¹⁹

An important question raised by our findings is whether the results are applicable to other long-term care facilities. Since 22% of incoming patients are carriers of *C difficile* toxin, the phenomenon is unlikely to be an isolated situation. Detection of *C difficile* toxin requires tissue-culture facilities and is not routinely offered by many clinical microbiology laboratories; an earlier report of *C difficile* in a hospital ward for the elderly used culture for detection,¹⁸ a method not recommended by most investigators. We are frequently consulted in outbreaks of *C difficile* and are aware of several other long-term care facilities where *C difficile* is endemic. We fully expect that our experience will be replicated elsewhere.

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Preliminary Communication

DOES NATURAL SENSITISATION IN ECZEMA OCCUR THROUGH THE SKIN?

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Summary The specificities of serum IgE for the faecal and body antigens of the house-dust mite, *Dermatophagoides pteromyssinus*, were compared in 69 children with various combinations of asthma, eczema, and/or rhinitis. The concentration of IgE antibodies to the mite body was higher in those with eczema than in those with asthma, but the concentrations of IgE antibodies to the faecal allergen were not significantly different. The ratio of the serum concentration of anti-mite-body IgE to anti-faecal IgE was significantly greater in the children with eczema than in the children with asthma. The results in 4 subjects with rhinitis (2 with and 2 without eczema) support the view that IgE antibodies to the mite body are characteristic of eczema. Sensitisation to mite body and mite faecal particles may occur by different processes; the allergens of mite bodies may penetrate the skin, whereas faecal allergen may enter the body by other route(s).

INTRODUCTION

HOUSE-DUST mites are thought to aggravate eczema,¹ but the mechanism of sensitisation is unknown. This needs to be established, since 75% of children with atopic disease are sensitised to the house-dust mite by the age of 10.²

The faecal allergen has been suggested as the main allergen of the house-dust mite.³ Inhalation of faecal particles is likely to be minimal because of their size. Allergens of the mite body, which is 12 times greater in size

than the faecal particle,³ are even less likely to be inhaled. In atopic disease where sensitisation occurs by inhalation proportionately more anti-faecal IgE antibodies than anti-mite-body IgE would be expected. To test this hypothesis we have compared concentrations of IgE antibodies against the mite body and mite faeces in patients with asthma and eczema.

SUBJECTS AND METHODS

Serum samples were obtained from 69 children (aged 3–16 years, mean 8.5; 68% boys) attending Bristol Children's Hospital. Their clinical diagnoses were asthma, eczema, and rhinitis in various combinations (see table). Serum samples were also obtained from 10 healthy siblings of asthmatics whose blood was taken as part of another ethically approved study. The project was approved by the Bristol and Weston Health Authority ethical committee.

Allergen Preparation

Dermatophagoides pteronyssinus was grown in pure culture on a sterilised powdered extract of bovine liver (Oxoid Ltd.).⁵ The initial separation of mite bodies from faecal particles was by means of sieving and flotation. Crude faecal and body extracts were prepared with the use of slightly modified standard methods.⁶

A purified faecal protein, which migrated as a discrete band on sodium dodecyl sulphate/polyacrylamide gel electrophoresis, was obtained by means of gel filtration⁶ and chromatofocussing.⁷ This protein was identical to purified samples of faecal allergen from 2 other sources^{4,7} when compared by means of immunodiffusion, radioimmunoassay, and electrophoretic techniques.

Contaminating faecal allergen was removed from the crude mite-body extract by means of immunoabsorption. Antibodies to faecal allergen were raised in rabbits⁸ and purified with ion-exchange chromatography.⁹ Short immunoabsorbent columns (2 ml disposable syringes, Gillette surgical) were packed with cyanogen-bromide activated sepharose 4B (Pharmacia) coupled to the anti-faecal antibodies. The crude body extract was applied to the column and elution carried out with sodium citrate (50 mmol/l) buffered to pH 6. The eluate was re-applied 3 times to an immunoabsorbent column to obtain the final extract. The body fraction so prepared was clearly distinct from the faecal extract since results of radioallergosorbent (RAST) tests for anti-mite-body IgE, with the use of pooled sera from 6 mite-allergic subjects, were reduced by only 6% with a gross excess of faecal allergen.

RAST and RAST Inhibition Tests

These were carried out by means of standard techniques¹⁰ with the use of Phadebas RAST ¹²⁵I-anti IgE (Pharmacia Diagnostics) and an LKB rack gamma counter. The results are expressed as counts per minute (cpm) and as RAST scores, where the counts are made against International Standard Samples and given in arbitrary units.

The faecal and body fractions were coupled to activated paper discs. For the RAST inhibition test serum was incubated with a

gross excess of faecal allergen, and the percentage inhibition of the previous standard RAST result calculated as:

$$\frac{\text{cpm (standard)} - \text{cpm (inhibited)}}{\text{cpm (standard)}} \times 100\%$$

All RAST tests on patients' sera were carried out in 1 batch, and the analyst (S. T.) was unaware of the clinical diagnosis.

Mann-Whitney tests were used to compare diagnostic groups.¹¹

RESULTS

The results of estimation of IgE antibody in the patients' sera are given in the table. Only 1 of the 10 control subjects had a weak positive (RAST 1) reaction to the faecal allergen, and none had a positive RAST reaction to body allergen.

2 of the 4 patients with rhinitis had eczema as well. In these 2 the mean mite-body RAST was 2.0, whereas in the 2 patients with rhinitis without eczema both mite-body RASTs were 0.

The ratio of body:faecal IgE antibody counts for each diagnostic group is shown in the table. The ratios for the 4 rhinitis subjects, 2 with eczema and 2 without, were 0.93 and 0.77, respectively.

DISCUSSION

Our results show that serum concentrations of IgE antibodies to the body of the house-dust mite were greater in children with eczema than in those with asthma. The difference is likely to be real since the ratio of body/faecal IgE antibody (see table) is greater when eczema is the primary diagnosis. Asthmatic children with eczema apparently had a higher ratio than those without eczema, and children with rhinitis apparently had more anti-mite-body IgE if they had eczema. The faecal IgE concentrations were similar in asthmatics whether or not they had anti-mite-body IgE which implies that anti-faecal antibody can be synthesised independently of anti-mite-body IgE. The production of IgE to the mite body and to the faecal allergen may arise from different processes.

Given that the IgE antibody production to mite body is greater in eczema than in asthma the mechanism of synthesis is important. IgE antibody production is directly proportional to mite allergen exposure in vitro.¹² We have confirmed (unpublished results) that there are multiple body allergens,⁴ and this suggests that sensitisation to body allergens is not a consequence of a unique chemical structure.

The most obvious explanation for the raised mite-body IgE concentration in children with eczema is increased exposure to mite-body allergens as a result of eczema. Less than 5% of mite faecal particles—about 18 µm in diameter—impact in artificial airways of the same size as the respiratory tract.³ The mite body is about 250 µm in size

CLINICAL DIAGNOSIS AND THE SITE SPECIFICITY OF ANTI-MITE IgE

Diagnostic group	No patients	Specific IgE antibodies†				Ratio of body/ faecal IgE antibody counts‡
		To faecal allergen		To body allergens		
		cpm	RAST score	cpm	RAST score	
I Asthma only	34	88 ± 87	2.1 ± 1.4	71 ± 69	1.9 ± 1.3	0.96
II Asthma + eczema	23	192 ± 73	3.5 ± 0.9	190 ± 63	3.5 ± 0.9	1.02
III Eczema	8	131 ± 69	2.9 ± 1.4	172 ± 72	3.8 ± 0.7	1.32
IV Rhinitis	4	41 ± 53	0.8 ± 1.5	37 ± 46	1.0 ± 1.4	..

All 4 specific IgE antibody results are significantly ($p < 0.0005$) lower in group I than in group II. The mite body cpm and RASTs are significantly ($p < 0.0025$) lower in group I than in group III whereas the mite faecal counts are not. The mite body counts and RAST scores and the mite faecal counts are significantly lower ($p < 0.05$) in group IV than in group III. There were no significant differences between groups II and III.

†Mean ± SD. ‡Mean value is shown. The ratios are significantly lower in groups I and II than in group III ($p < 0.02$), but the difference between groups I and II is not significant.

which makes it even less likely that it could be inhaled.¹³ Inhalation of faecal and body fragments, as happens with pollen grains, which are of a similar size,¹⁴ could explain why some of the asthmatic children without eczema had antibodies to mite body if sensitisation in asthma occurs by way of the respiratory tract. However, the chitinous covering of the body is less likely than the membranes of the faecal particles to fragment,³ and therefore any force disrupting mites and their particles should accentuate the differences we observed. Since about 0.1% of ingested protein is absorbed antigenically intact,¹⁵ it is unlikely that the swallowing of mites will produce the increased sensitisation to the body allergen that occurs in eczema. House-dust-mite allergen has been shown to penetrate gently abraded skin,¹⁶ but both our own and other unpublished studies failed to show delayed hypersensitivity to crude mite extracts with patch tests on unabraded skin.

It may be that allergen penetration occurs in active excoriated eczema because of the reduced skin thickness. Reitamo et al¹⁷ found that delayed reactions occurred in eczematous subjects on application of high concentrations of allergens (birch pollen and house-dust mite) to the skin for 48 h. 3 of the 4 patients who deteriorated in the birch pollen season also had the delayed skin reaction characteristic of eczema on patch testing.¹⁷

The presence of IgE specific for the mite body is probably a consequence of sensitisation through the skin.¹⁸ These results are the first direct indication that natural sensitisation in eczema can occur through the skin. There is no doubt that eczema is associated with increased cutaneous reactivity to allergens and to mites in particular.^{17,19} Evidence suggests there is a temporal relation between allergen exposure and exacerbations of eczema,^{1,17,20} and allergens can be shown experimentally to produce lesions histologically identical to eczema.^{16,17} Cutaneously presented allergens probably, therefore, cause exacerbations of eczema. The present results do not address the question of whether allergen exposure sets up the atopic state which predisposes to eczema.²¹⁻²⁴

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References continued at foot of next column

Hypothesis

CAMPYLOBACTER PYLORIDIS, UREASE, HYDROGEN ION BACK DIFFUSION, AND GASTRIC ULCERS

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Summary *Campylobacter pyloridis*, a bacterium implicated as the aetiological agent of gastritis and possibly gastric ulcers, has a very high urease activity. The rapid hydrolysis of urea at intercellular junctions results in alterations in the milieu of the gastric epithelium preventing the normal passage of hydrogen ions (H⁺) from the gastric glands through the mucus to the lumen and permits back diffusion. A consequence of H⁺ back diffusion is hypochlorhydria and a predisposition to ulcer formation. Several conflicting reports on the physiology of normal, gastritic, and ulcerated stomachs are reconciled by this hypothesis.

INTRODUCTION

IN 1925 Luck and Seth¹ proposed several roles for the then newly discovered "gastric urease" in animals. One suggestion was that gastric urease countered gastric hyperacidity by promoting secretion of alkali; another was that the enzyme was part of an intracellular mechanism of mucosal-cell protection against gastric acid. When Glick² found urease in the human stomach he noted an association between urease and parietal-cell distribution, and the notion of a protective role for urease was supported by the observation of Fitzgerald and Murphy³ that the enzyme activity was concentrated at the surface of the mucosa and in the vicinity of functioning oxyntic cells. In the early 1950s, however, the demonstration that gastric urease was of bacterial

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