

TABLE I. Clinical and demographic characteristics and results of inhalation challenge

Patients	1	2	3	4	5	6	7	8	9	10	11	12
Age (y)	39	29	48	21	22	41	29	23	24	26	41	25
Perennial asthma symptoms	Yes	No	Yes	Yes	Yes	Yes	Yes	Yes	No	Yes	Yes	Yes
Treatment	ICS + B2	B2 prn	ICS + B2	ICS	ICS + B2	No	B2 prn	B2 prn	B2 prn	ICS + B2	ICS + B2	B2 prn
Specific IgE to <i>B germanica</i> (kU/L)	0.71	ND	2.98	1.39	<0.35	1.07	<0.35	0.36	<0.35	1.95	1.64	1.76
Sensitization to other allergens	Mites	Pollen, cat, dog, molds	Pollen	Pollen, mites, legumes	Pollen, mites, dog, cat, molds	Mites, dog	Molds	Pollen, mites, cat, dog	Pollen, mites, cat, dog, molds	Pollen, cat, dog	Mites	Pollen, mites, shrimp
PC ₂₀ methacholine (mg/mL)	0.5	0.16	2.46	0.44	0.77	0.36	0.40	0.125	0.3	0.0314	2.40	0.250
PC ₂₀ <i>B germanica</i> (mg/mL)	5	0.01	0.156	0.625	1.25	0.625	5	0.156	2.5	0.2	Negative	0.156
Early asthmatic reaction	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	No	Yes
Late asthmatic reaction	No	No	No	No	No	Yes	No	No	No	No	No	No
Sputum cell count (%) at baseline/24 h after cockroach SIC												
Bronchial epithelial cells	0/0	20/0	0/0	0/0	0/0	20/20	5/0	0/0	0/0	0/0	ND	0/3
Macrophages	60/40	70/30	40/60	80/40	60/40	60/10	75/50	60/40	60/70	40/70		60/67
Neutrophils	38/6	8/70	42/8	20/54	40/24	20/50	20/50	30/12	25/18	18/18		30/6
Eosinophils	2/54	2/0	18/32	0/6	0/36	0/20	0/0	10/48	15/12	42/12		10/24

ICS, Inhaled corticosteroids; B2, inhaled β_2 -agonist; prn, as needed; ND, not done.

Eosinophilic and neutrophilic sputum response to bronchial challenge with cockroach

To the Editor:

Previous studies have established that cockroach allergens are important sensitizing agents in the induction of asthma,¹ although their prevalence varies in different parts of the world. Specific inhalation challenge (SIC) studies^{2,3} revealed that most subjects with asthma sensitized to cockroach allergens have early or dual asthmatic reactions on inhalation of cockroach extract, with a significant increase of peripheral blood eosinophilia 24 hours after the challenge.³ There are some data on animal models^{4,5} on the possible pathophysiologic mechanisms of cockroach-induced airway disease. In guinea pigs Zhou et al⁴ demonstrated that inhalation of cockroach extract by sensitized animals provoked leukocytosis and eosinophilia in the bronchoalveolar lavage fluid. In a murine model Campbell et al⁵ found a peribronchial

inflammatory response characterized by eosinophil accumulation and airway hyperresponsiveness. To the best of our knowledge, there are no data on bronchial inflammation after SIC in patients sensitized to cockroach. We analyzed the induced sputum samples obtained before and 24 hours after SIC with a cockroach extract in a group of patients with asthma who were sensitized to cockroach.

Twelve asthmatic patients with positive skin prick test responses to *Blattella germanica* were recruited from our outpatient clinic. All study subjects completed a questionnaire. Skin prick tests were performed with a panel of common inhalant allergens (CBF LETI SA). Histamine phosphate, 10 mg/mL, and normal saline were used as positive and negative controls, respectively. The response was read 15 minutes after puncture, and the results were expressed as the mean wheal diameter (in millimeters). A wheal diameter of 3 mm or greater with erythema compared with that elicited by the saline control was defined as a positive reaction. Specific IgE was measured by the Pharmacia CAP system; values greater than 0.35 kU/L were considered positive.

B germanica (Biopol) extract was prepared at a concentration of 10 mg/mL in PBS. The allergen samples were filtered through a 0.22- μ m membrane (Millipore) and placed in aliquots. Several 2-fold dilutions of cockroach extract in PBS were used for skin and inhalation tests. Endotoxins (Limulus assay, limit of detection <0.5 EU/mL) were not detected in this extract.

SIC to cockroach extract was carried out in 12 patients with asthma who gave informed consent, according to the

tidal volume method, as previously described.⁶ A control challenge with PBS was carried out before allergen provocation. Increasing concentrations of cockroach extract were given by means of inhalation, starting with a concentration that induced a 2-mm wheal on skin prick testing. The same test was performed in 2 asthmatic patients who were not sensitized to cockroach, with negative results.

Induction of sputum was performed before and 24 hours after SIC, as previously described.⁶ The cell pellets were resuspended in saline solution and stained with the Papanicolaou and Giemsa methods for differential cell counts. Two investigators counted at least 400 nonsquamous cells on each sputum slide.

Characteristics of patients with positive SIC responses, demographic data, and results of SIC and sputum analysis are presented in Table I. Eleven patients had positive SIC responses. An early asthmatic reaction was observed in 10 patients, and a dual asthmatic reaction was seen in 1 patient. Baseline sputum eosinophilia ($\geq 2\%$) was detected in 7 of 11 patients. Twenty-four hours after a positive SIC response, sputum eosinophilia was observed in 9 of 11 patients, and neutrophils predominated in the sputum samples of the remaining 2 patients. In 3 patients sputum eosinophilia was observed only after SIC. Sputum neutrophil counts increased in 4 patients, eosinophils in 7 patients, and both in 2 patients. The mean percentage of sputum neutrophils before and after SIC were $26.5\% \pm 10.1\%$ and $28.7\% \pm 21.8\%$, respectively ($P =$ not significant, Wilcoxon test), and values for eosinophils were $9\% \pm 12.1\%$ and $22.18\% \pm 17.6\%$, respectively. This increase approached statistical significance ($P = .07$, Wilcoxon test).

This study confirms that cockroach allergens are strong inducers of bronchial inflammation in sensitized patients. The inflammatory infiltrate after SIC consisted of eosinophils and neutrophils, with an increase of sputum eosinophils from baseline in most patients. However, neutrophils were also observed, and their percentages increased after SIC in 6 patients. These findings are consistent with those of previously published studies showing an increase of eosinophils and neutrophils after asthmatic reactions induced by an IgE-dependent mechanism with both occupational and nonoccupational agents.⁷⁻⁹

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