

## Letter to the Editor

**Detection of novel occupational wood allergens in locust wood dust (*Robinia pseudoacacia* L.)**

To the Editor:

IgE-mediated allergy to wood dust is a known occupational health problem. Among the main sources of wood dust allergy, such as obeche,<sup>1-3</sup> case reports indicate the potential of many different woods, especially tropical, to induce IgE-mediated allergy among exposed workers.<sup>4,5</sup> The recently identified obeche wood allergen shows homology to latex hevein and other plant class I chitinases.<sup>2</sup> Over the last few years, tropical woods have been increasingly replaced by cultured wood species. Locust wood (*Robinia pseudoacacia* L.) is mostly used as an exchange wood for teak or makore, especially for outdoor constructions such as garden furniture or children's playgrounds. However, locust wood is used to a much lower extent in comparison with hardwoods such as oak or beech. The 2 cases we report here are the first descriptions of IgE-mediated occupational asthma triggered by the nontropical locust wood dust (*R pseudoacacia* L.).

Patient 1, a 42-year-old nonsmoker mechanic employed for 10 years in a company constructing toys for children's playgrounds, had shortness of breath, sneezing, and burning eyes after grinding of locust wood. As a mechanic, he had no contact with wood dust, and asthma occurred after 1 high-exposure incident to locust wood dust. He had hay fever during childhood, without any allergic symptoms during adulthood. Spirometry showed mild airways obstruction, and bronchial hyperresponsiveness was demonstrated (Table I). Locust wood extract was prepared freshly for skin prick test (SPT) and bronchial challenge as described recently.<sup>2</sup> SPT reactions with wheal diameter  $\geq 3$  mm were considered positive. Patient 1 showed positive SPT reactions to locust wood, house dust mites (*Dermatophagoides farinae*, *D pteronyssinus*; ALK-SCHERAX, Hamburg, Germany), grass pollen mix (ALK-SCHERAX), and tree pollen mix (HAL Allergie, Düsseldorf, Germany). SPT with wood extracts of obeche, maple, ash, oak, and teak were negative, as were intradermal tests with commercial wood extracts (obeche, beech, oak, limba, mahogany, makore, walnut, ramin, elm, fir, teak, spruce and pine; Allergopharma, Reinbek, Germany). A bronchial challenge test was

performed with locust wood extract in doses between 2 ng and 20  $\mu$ g protein in 10-fold concentration steps by an APSpro dosimeter and a DeVilbiss 646 nebulizer (output, 900  $\mu$ L/min; Viasys, Wuerzburg, Germany).<sup>6</sup> Although the inhalation of saline control induced only an 8% reduction in FEV<sub>1</sub>, a significant immediate reaction to locust wood extract was observed (Table I), and the specific airway resistance increased from 1.7 to 3.5 kPa·s.

Patient 2, a 57-year-old carpenter (exsmoker), produced toys for children's playgrounds for 11 years and was exposed to a number of different wood dusts. He reported work-related shortness of breath without eye or nose symptoms for 3 years, but the symptoms persisted while off work. Spirometry showed severe airways obstruction (Table I). SPT reactions were positive to house dust mite (*D farinae*, *D pteronyssinus*; Allergopharma). Intradermal tests were performed with the commercial wood extracts as in patient 1 with completely negative results. Patient 2 did not undergo specific challenge testing with locust wood extract because of poor baseline lung function.

Atopic controls with bronchial hyperresponsiveness showed negative SPT with all woods including locust wood, and challenges with locust wood extract (method as in patient 1) were negative (Table I).

*In vitro* tests with serum of patient 1 showed specific IgE to *D pteronyssinus* ( $>100$  kU/L), *D farinae* (74 kU/L), *Acarus siro* (4 kU/L), *Lepidoglyphus destructor* (8 kU/L), *Tyrophagus putrescentiae* (6 kU/L), grass pollen (42 kU/L), and tree pollen (50 kU/L). Patient 2 was sensitized to *D pteronyssinus* (5 kU/L) and *D farinae* (4 kU/L; ImmunoCAP; Phadias, Uppsala, Sweden). Locust wood extract was coupled by streptavidin/biotin affinity to ImmunoCAP solid phase as described earlier.<sup>7</sup> Both patients displayed elevated IgE concentrations to locust wood allergens (patient 1, 0.9 kU/L; patient 2, 1.4 kU/L). Wood dust extracts of obeche, oak, teak, ash, maple, and mahogany were tested with the same method for specific IgE with negative results ( $\leq 0.35$  kU/L). Thus, both patients were sensitized to locust wood but not to other woods. All controls showed no specific IgE response to locust wood ( $\leq 0.35$  kU/L).

To assess possible cross-reactivities between locust wood and environmental allergens or cross-reactive carbohydrate determinants (CCD), ImmunoCAP inhibition

**TABLE I.** Spirometry and bronchial challenges with methacholine and locust wood extract of patients and controls

|                                                                                              | Patient 1 | Patient 2 | Control 1 | Control 2 | Control 3 | Control 4 | Control 5 |
|----------------------------------------------------------------------------------------------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|
| Sex (M/F)                                                                                    | M         | M         | F         | M         | M         | F         | M         |
| Age (y)                                                                                      | 41        | 56        | 22        | 27        | 27        | 25        | 25        |
| FEV <sub>1</sub> % predicted                                                                 | 72        | 26        | 96        | 94        | 81        | 95        | 99        |
| PD <sub>20</sub> FEV <sub>1</sub> methacholine ( $\mu$ g)                                    | 61        | ND        | 34        | 4         | 46        | 149       | 41        |
| Bronchial challenge with locust wood<br>(maximum fall of FEV <sub>1</sub> ; % from baseline) | 26        | ND        | 7         | 0         | 2         | 1         | 0         |

M, Male; F, female; ND, not done.

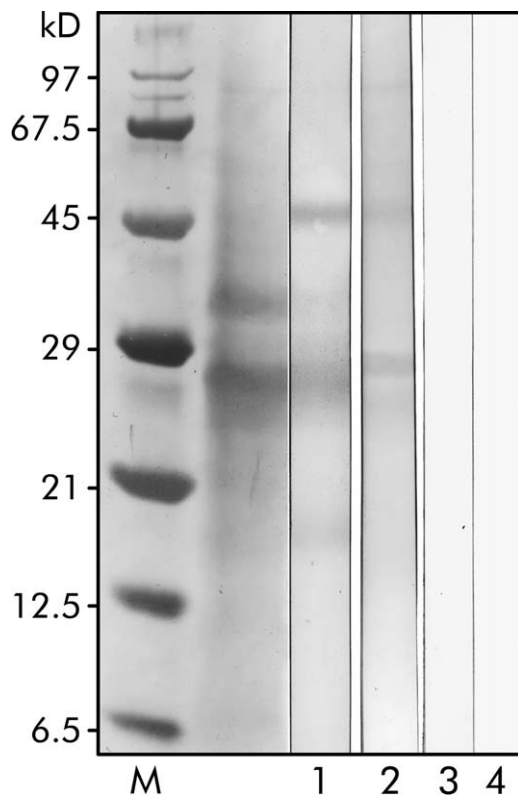


**FIG 1.** IgE immunoblots of locust wood extract were probed with serum of patient 1 (lane 1), serum of patient 2 (lane 2), pooled sera of 5 carpenters without sensitization to locust wood (lane 3), and buffer (lane 4).

experiments were performed. Serum of patient 1 was preincubated with extracts of storage mites (*A siro*, *L destructor*, and *T putrescentiae*) and grass pollen (10  $\mu$ g/inhibition) and subsequently, IgE binding to locust wood ImmunoCAP was measured. IgE binding was completely unaffected by preabsorption of serum with mites or pollen allergens. In addition, preincubation with locust wood extract (10  $\mu$ g) did not reduce IgE binding to grass pollen or mites. Serum of patient 2 was tested with locust wood extract as an inhibitor on house dust mite ImmunoCAPs without any inhibitory effect. In contrast, in both patients' serum IgE reactivity to locust wood extract was totally abolished by preincubation with locust wood extract (autoinhibition). Thus, cross-reactivities between locust wood and other environmental allergens could not be demonstrated. To study the relevance of nonspecific IgE binding to CCD,<sup>8,9</sup> ImmunoCAP inhibition with horseradish peroxidase (HRP) as an inhibitor and locust wood as the solid phase was performed. Specific serum IgE to HRP was low (0.40 kU/L) in patient 1 and <0.35 kU/L in patient 2. In both cases, preincubation of the sera with HRP (10  $\mu$ g) did not reduce the IgE binding to locust

wood. A cross-reaction based on nonspecific CCD could be excluded.

Allergens were detected after SDS-PAGE of locust wood extract on an IgE Western blot as described earlier.<sup>2</sup> Serum of both patients were incubated with blot strips to identify IgE-binding proteins. As controls, a pool serum of patients with obeche wood allergy and buffer were used (Fig 1). IgE to allergens with molecular weights of 15, 27, and 47 kD were detected with the serum of patient 1. Patient 2 showed only weak IgE binding to a 27-kD and 47-kD protein, whereas both controls showed no binding. The locust wood proteins of 27 and 47 kD are good candidates as the major allergens of locust wood dust, but this has to be proven among a larger number of patients.

In conclusion, IgE-mediated allergy to locust wood dust is specific with no demonstrable cross-reactivity with other environmental and wood allergens. The potential risk to develop occupational asthma as result of IgE-mediated sensitization to locust wood dust should be considered clinically significant.

Sabine Kespohl, PhD<sup>a</sup>

Rolf Merget, MD<sup>a</sup>

Axel Overlack, MD<sup>b</sup>

Monika Raulf-Heimsoth, PhD<sup>a</sup>

From <sup>a</sup>the Research Institute for Occupational Medicine of the Berufsgenossenschaften (BGFA), Ruhr University Bochum; and <sup>b</sup>Gemeinschaftspraxis für Innere Medizin, Lungen und Bronchialheilkunde, Allergologie/Umweltmedizin, Bonn, Germany.

Supported by HVBG (Hauptverband der Berufsgenossenschaften), St Augustin, Germany. BGFA-project All/Med 9, St Augustin, Germany.

Disclosure of potential conflict of interest: The authors have declared that they have no conflict of interest.

## REFERENCES

- Ferrer A, Maranon F, Casanovas M, Fernandez-Caldas E. Asthma from inhalation of *Triplochiton scleroxylon* (Samba) wood dust. *J Invest Allergol Clin Immunol* 2001;11:199-203.
- Kespohl S, Sander I, Merget R, Petersen A, Meyer HE, Sickmann A, et al. Identification of an obeche (*Triplochiton scleroxylon*) wood allergen as a class I chitinase. *Allergy* 2005;60:808-14.
- Quirce S, Hinojosa M, Maranon F, Ferrer A, Fernandez-Caldas E, Sastre J. Identification of obeche wood (*Triplochiton scleroxylon*) allergens associated with occupational asthma. *J Allergy Clin Immunol* 2000;106:400-1.
- Algranti E, Mendonca EM, Ali SA, Kokron CM, Raile V. Occupational asthma caused by Ipe [Tabebuia spp] dust. *J Invest Allergol Clin Immunol* 2005;15:81-3.
- Alday E, Gómez M, Ojeda P, Caballero ML, Moneo I. IgE-mediated asthma associated with a unique allergen from Angelim pedra (*Hymenolobium petraeum*) wood. *J Allergy Clin Immunol* 2005;115:634-6.
- American Thoracic Society. Guidelines for methacholine and exercise challenge testing: 1999. *Am J Respir Crit Care Med* 2000;161:309-29.
- Sander I, Kespohl S, Merget R, Goldscheid N, Degens PO, Bruening T, et al. A new method to bind allergens for the measurement of specific IgE antibodies. *Int Arch Allergy Immunol* 2005;136:39-44.
- van Ree R. Carbohydrate epitopes and their relevance for the diagnosis and treatment of allergic diseases. *Int Arch Allergy Immunol* 2002;129:189-97.
- Vieths S, Scheurer S, Ballmer-Weber B. Current understanding of cross-reactivity of food allergens and pollen. *Ann N Y Acad Sci* 2002;964:47-68.

doi:10.1016/j.jaci.2006.03.042