***Tribolium confusum* (Confused Flour Beetle, Rice Flour Beetle) – An Occupational Allergen in Bakers: Demonstration of IgE Antibodies**

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Abstract

Specific IgE to proteins from *Tribolium confusum* (TC), a flour beetle, was detected in 9/125 sera of subjects exposed to rye and wheat flour. TC RAST was not inhibited by *Dermatophagoides pteronyssinus*, rye or wheat flour. Immunoblot experiments showed specific binding to three proteins from adult TC or pupae, not present in rye or wheat flour. These findings suggest that TC might act as an occupational allergen in a proportion of bakers.

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Introduction

Baker's asthma is the most common type of occupational asthma in many countries. Common allergens are proteins from wheat or rye flour [1, 2]. Another increasingly important source of allergens is a number of enzymes, e.g. alpha-amylase from *Aspergillus oryzae* [3].

In a preliminary study we had identified a number of arthropode species in flour dust from bakeries. Subsequently, positive skin prick tests to a predominant beetle, *Tribolium confusum* (TC; confused flour beetle, rice flour beetle), were detected in 8.3% of 133 bakers, using a crude TC extract. Immediate type or dual reactions were seen after bronchial provocation tests with these TC extracts in TC skin prick test positive subjects, even if skin prick tests and bronchial provocation tests with flour extracts were negative [4]. Therefore, this study was performed to prove that TC is an occupational allergen in bakers and that this reaction is IgE mediated.

Material and Methods

We cultured a TC population (kindly supplied by the Beetle Lab., Hoechst AG, Frankfurt-Hoechst, FRG) without wheat or rye flour. After 4 months different stages of the beetle were separated: adult beetles (ATC), pupae (TCP), larvae (TCL), exuviae (TCH), and excrements (TCE). These materials were kept at -20°C, ground mechanically, defatted, extracted, and dialyzed. Proteins were coupled to cyanogen bromide-activated methylcellulose disks.

Sera from 125 bakers were investigated for specific IgE antibodies by enzyme immunoassay (Phadezym RAST, Pharmacia) and RAST inhibition. As controls we used a pooled serum from pollen-allergic subjects (IgE 450 kU/1), sera from 10 atopic subjects (IgE 120–1,000 kU/1) and 9 nonatopic subjects (IgE < 80 kU/1).

TC extracts, together with extracts from wheat and rye flour, and Dermatophagoides pteronyssinus (Dpt) were further investigated by SDS-PAGE and Immunoblot, using 7 TC IgE antibody-positive and 6 negative sera.

Results

Nine of 125 sera (7.2%) had IgE antibodies to ATC (binding rates 11–69%). None of the control sera showed any binding. RAST inhibition with 5 ATC IgE-positive sera and ATC disks resulted in a mean inhibition of 90% (ATC), 87.5% (TCP), 89% (TCL), 65% (TCH), and 76% (TCE), respectively (fig. 1). In 7/8 sera, wheat or rye flour-extracts were also inhibited significantly. Protein fractions of ATC, TCP, and TCE were detected by SDS-PAGE, with a MW between 10 and 180 kD. In the sera of bakers, but not in controls, 3 IgE-binding protein fractions (MW 67, 85, 132 kD) were seen with ATC and TCP. No corresponding IgE-binding fractions were detected in flour extracts.

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100

80-

60

40-

20

10 µl Allergen

0.01 Serum

Fig. 1. RAST

with IgE to

TCH (TC

mite

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G.M

inhibition experiment with serum from a subject

TC (RAST). Inhibition by ATC (adult TC),

exuviae), TCL (TC larvae), D.pt. (house dust

D.pteronyssinus), W.F1. (wheat flour), R.F1.

flour), TCP (TC pupae) and TCE (TC

excrements). No significant inhibition was

with flour extracts or D.pt.

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Conclusions

Specific immunoglobulin E to TC allergens is frequent in bakers. Extracts from adult TC or pupae contain at least three different proteins with allergenic activity. These proteins differ from flour proteins. In a proportion of bakers, beetles such as TC may act as occupational allergens.