

Occupational Immunoglobulin E-Mediated Asthma due to *Penicillium camemberti* in a Dry-Sausage Packer

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Key Words

Immunoglobulin E-mediated asthma · *Penicillium camemberti* moulds · Occupational allergy

Abstract

A 62-year-old female sausage packer reported progressive work-related asthma, rhinitis and contact urticaria after contact to dry sausages refined by moulds. Whereas skin prick tests with commercial mould extracts were negative, the patient showed weak skin sensitization to a cultivated mould sample from a dry sausage. Specific immunoglobulin E antibodies to various moulds were demonstrated by ImmunoCAP and antibodies to the cultivated mould sample were demonstrated by enzyme allergosorbent test. The mould was identified by pheno- and genotyping as *Penicillium camemberti*. Five atopic controls did not show sensitization with the same tests. Crossreactivity of *P. camemberti* and *Penicillium notatum* was shown by enzyme allergosorbent inhibition tests. Although no challenge tests were considered due to the patient's airway obstruction, a diagnosis of allergic occupational asthma was made. We conclude that immunoglobulin E-mediated allergic occupational asthma due to moulds may occur in dry sausage packers.

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Introduction

Although a recent report from Finland indicated that moulds from moisture-damaged buildings were among the most frequent causative agents of occupational asthma notified to the Finnish Register of Occupational Diseases [1], only few case reports of immunoglobulin E (IgE)-mediated occupational allergies related to mould exposure were published. This is in contrast to a higher number of reports about occupational hypersensitivity pneumonitis, which has been described in various occupations with high mould exposure [2–4]. For example, *Penicillium casei* has been demonstrated as the cause of hypersensitivity pneumonitis in cheese washers in numerous publications, but only one case history of occupational asthma (positive intradermal test and immediate reaction after challenge with a commercial *Penicillium* mix) in these workers has been reported [5].

There is epidemiologic evidence of an increased prevalence of work-related airway symptoms in workers exposed to dry sausage moulds which are used for the refinement of some dry sausages [6]. However, symptoms were considered to be due to hypersensitivity pneumonitis. We report the case of a dry-sausage packer with work-related shortness of breath.

Table 1. Results of total IgE, SPT and specific IgE to *P. camemberti* and *P. notatum* of the patient and 5 atopic controls

	Patient	Subject 1	Subject 2	Subject 3	Subject 4	Subject 5
Gender	F	M	M	M	F	M
Age, years	62	25	35	38	34	26
Total IgE, kU/l	1,360	66	133	18	207	58
SPT						
Environmental allergens (wheal diameter ≥ 4 mm)	–	cat, mites	trees	cat, trees, grasses	horse, trees, grasses	trees
Maximum wheal diameter of environmental allergens, mm	–	4	15	7	15	5
Wheal diameter of <i>P. camemberti</i> , mm	4	0	0	0	0	0
Wheal diameter of <i>P. notatum</i> ¹ , mm	3	0	0	0	0	0
Specific IgE (EAST)						
<i>P. camemberti</i> , kU/l	0.37	0.02	0.05	0.07	0.05	0.03

¹ Obtained from Allergon.

Case Report

The 62-year-old patient, examined in 2004, had worked in a sausage producing plant from 1979 to 2000. She was exposed to moulds used for sausage refinement only in the packing area where she worked rarely until 1998. From 1999 to 2000, she started to work in the packing area on most days of the week. While work-related cough was reported after contact to moulds since the mid 1980s, work-related asthma occurred since the mid 1990s. Asthma deteriorated, and runny nose, sneezing and itchy red wheals on both hands and forearms appeared shortly after the beginning of the shift during the last 2 years. The patient ascribed worsening to an increased frequency of work in the packing area. Symptoms improved on weekends and holidays. Finally, she quit her job for medical reasons (lateral epicondylitis and allergic symptoms). After exposure cessation, nose and skin symptoms disappeared completely and shortness of breath improved. Asthma attacks ceased completely, but dyspnea on exertion persisted. The patient had smoked 10–15 cigarettes a day from 1969 to 2000 (about 20 pack-years). She was on regular medication with long-acting β -agonists and topical steroids. Medication was withheld 15 h before lung function testing.

Information about the quality of the moulds used for sausage refinement was not available. Measurements of airborne moulds at the patient's workplace were reported within reference values by the technical division of the accident insurance.

Methods and Results

The patient's spirometry was of moderate quality. Formally, an obstructive ventilation pattern with hyperinflation was measured (FEV₁ 49% predicted, total lung capacity 93% predicted, residual volume 140% predicted, FEV₁/

FVC ratio 79% predicted). Although more chronic obstructive pulmonary disease-like symptoms were reported during the previous years, partial reversibility was demonstrated by an increase in FEV₁ from 1.21 to 1.53 liters (26.5% increase) after inhalation of 200 μ g salbutamol. Skin prick tests (SPT) showed 2-mm wheals without erythema with both the negative control and environmental allergens including a panel of commercial mould extracts, among them *Penicillium* species from four manufacturers (table 1). Total IgE was 1,360 kU/l. Specific IgE antibodies to house dust mites (*Dermatophagoides pteronyssinus* 0.64 kU/l, *Dermatophagoides farinae* 0.98 kU/l) as well as to various moulds were measured by UniCAP 100 (Sweden Diagnostics, Freiburg, Germany): *Aspergillus fumigatus* (m3) 2.5 kU/l, mould mix (mx1) 1.9 kU/l, *Candida albicans* (m5) <0.35 kU/l, *Penicillium notatum* (m1) 1.21 kU/l, and *Aureobasidium pullulans* (m12) 0.58 kU/l. IgG concentrations to *A. fumigatus* (Gm3), *Alternaria alternata* (Gm6), *P. notatum* (m1), *A. pullulans* (m12) and *Micropolyspora faeni* (Gm22) were within reference values.

Cultivation of four sausage skin samples on Kimmig agar yielded two different moulds with preponderance of one mould phenotyped as *Penicillium camemberti* (the other mould phenotyped as *Candida albicans* was considered a contamination). Four samples were taken from the cultured fungi for DNA preparation using the PrepMan (Ultra) method from Applied Biosystems (Darmstadt, Germany). For genotyping, the MicroSeq™ D2 LSU rDNA Fungal Sequencing Kit from the same company was chosen. The obtained sequences of all analyzed

samples were identical and showed 100% identity to a not further specified *Penicillium* species (EMBL database); the next best homology of 99.7% was to *P. notatum* (syn. *Penicillium chrysogenum*). These findings combined with the phenotyping led to the conclusion that a modified *P. camemberti* was used for sausage refinement.

SPT with extracts of the cultivated moulds showed a 2-mm wheal (no erythema) with *C. albicans* (4.4 µg protein/ml), but a 4-mm wheal (20-mm erythema) with *P. camemberti* (0.74 mg protein/ml). A 3-mm wheal (10-mm erythema) was observed with a self-made extract of *P. notatum* (0.34 mg/ml) from Allergon (Ängelholm, Sweden). Enzyme allergosorbent tests (EAST; Allergopharma, Reinbek, Germany) with the sausage mould coupled to cellulose disks indicated a weak sensitization (0.37 kU/l). Inhibition tests performed by UniCAP with *P. notatum* (m1) as solid phase yielded 100% inhibition when *P. camemberti* and *P. notatum* (Allergon) were used as inhibitors. When *A. fumigatus* (m3) was used as solid phase, inhibition was 78% with *P. camemberti* and 45% with *P. notatum*. Five atopic controls did not show positive SPT or EAST with *P. camemberti* (table 1).

Discussion

We demonstrated weak IgE-mediated sensitization to the predominant mould at the patient's former workplace by SPT and EAST. The patient presented with high total IgE and differed in this respect from the controls; however, both in vivo and in vitro tests with the mould from the patient's workplace showed weak but clear sensitization to the mould. It is noteworthy that SPT with moulds

were positive only with extracts prepared in our laboratory, indicating a low sensitivity of commercial mould extracts. Sensitization to moulds is further substantiated by the presence of specific IgE antibodies to *P. notatum* in commercial CAP testing. Although no increased mould air concentrations at the former workplace were reported and underlying chronic obstructive pulmonary disease due to smoking cannot be excluded, a diagnosis of allergic occupational asthma was made. A bronchial challenge was not performed due to the patient's airway obstruction. The patient's workplace-related earlier symptoms were indicative of occupational rhinitis and contact urticaria and corroborate the diagnosis of occupational allergy.

Sausages are rarely refined by moulds, thus respective cases must be restricted to few producers. This may explain why such cases have not been reported in the literature earlier. Another explanation might be the low sensitivity of commercial SPT extracts. We have found only one case report in the literature of a butcher in sausage production with IgE-mediated allergy to *Monascus purpureus*, a mould that is used to prepare red fermented rice [7]. Hypersensitivity pneumonitis due to *P. camemberti* in a salami factory has been described [8]. Also, food allergy to sausage moulds has been reported, but is obviously rare [9, 10].

The mould was identified as *P. camemberti*. Inhibition experiments showed strong crossreactivity within *Penicillium* species, but also between *Penicillium* species and *A. fumigatus*, as described earlier [11]. We conclude that IgE-mediated occupational asthma may occur in dry sausage packers with exposure to moulds which are used for sausage refinement.

References

- Piipari R, Keskinen H: Agents causing occupational asthma in Finland in 1986–2002: cow epithelium bypassed by moulds from moisture-damaged buildings. *Clin Exp Allergy* 2005;35:1632–1637.
- Morell F, Roger A, Cruz MJ, Muñoz X, Rodrigo MJ: Suberosis. Clinical study and new etiologic agents in a series of eight patients. *Chest* 2003;124:1145–1152.
- Cormier Y, Israel-Assayag E, Bedard G, Duchaine C: Hypersensitivity pneumonitis in peat moss processing plant workers. *Am J Respir Crit Care Med* 1998;158:412–417.
- Campbell JA, Kryda MJ, Treuhaft MW, Marx JJ Jr, Roberts RC: Cheese worker's hypersensitivity pneumonitis. *Am Rev Respir Dis* 1983;127:495–496.
- Wuethrich B, Keiser G: Cheese washer's asthma, differentiation from cheese washer's disease (in German). *Schweiz Med Wochenschr* 1970;100:1108–1111.
- Rouzaud P, Soulat JM, Trela C, Frayse P, Recco P, Carles P, Lauque D: Symptoms and serum precipitins in workers exposed to dry sausage mould: consequences of exposure to sausage mould. *Int Arch Occup Environ Health* 2001;74:371–374.
- Hipler UC, Wigger-Alberti A, Bauer A, El-sner P: Case report. *Monascus purpureus* – a new fungus of allergologic relevance. *Mycoses* 2002;45:58–60.
- Marchisio VF, Sulotto F, Botta GC, Chiesa A, Airaudi D, Anastasi A: Aerobiological analysis in a salami factory: a possible case of extrinsic allergic alveolitis by *Penicillium camemberti*. *Med Mycol* 1999;37:285–289.
- Fiocchi A, Mirri GP, Santini I, Bernardo L, Ottoboni F, Riva E: Exercise-induced anaphylaxis after food contaminant ingestion in double-blinded, placebo-controlled, food-exercise challenge. *J Allergy Clin Immunol* 1997;100:424–425.
- Morisset M, Parisot L, Kanny G, Moneret-Vautrin DA: Food allergy to moulds: two cases observed after dry fermented sausage ingestion. *Allergy* 2003;58:1203–1204.
- Brouwer J: Cross-reactivity between *Aspergillus fumigatus* and *Penicillium*. *Int Arch Allergy Immunol* 1996;110:166–173.