

SUSPENSIONS OF COMPOST AND *ASPERGILLUS FUMIGATUS* CAN CAUSE HISTAMINE RELEASE FROM HUMAN BASOPHILS

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Abstract

During the last decade an increasing number of plants for reuse of refuse have been constructed in Europe and USA. During the same period several cases of occupational respiratory diseases among workers in the recycling industry have been reported. In an earlier study we have showed that isolated basophile from pig-farmers with asthma were sensitized against pig-protein. The aim with this study was to show if there was some individual differences by histamine release from basophile leucocytes. Nine formerly compost workers and five earlier unexposed persons have been examined. The histamine release was studied in isolated basophile leucocytes after incubation with suspensions of 35 days old compost and *Aspergillus fumigatus*. Compost caused histamine release from basophile leucocytes from both formerly compost workers and control persons. However, *Aspergillus fumigatus* caused histamine release in 3 of the workers and none of the control persons.

Introduction

During the last two decades it has been more common in the industrialized countries to reuse the refuse, therefore an increasing number of plants for reuse have been constructed in Europe and USA. However, several cases of occupational respiratory diseases have been reported. In 1986 a new plant which converted household waste to fuel-pellets started. During the first 1.5 years 8 out of 15 employees got ill with respiratory symptoms (Sigsgaard et. al., 1990). In Sweden as well as Denmark some investigations have shown an increased prevalence of toxic alveolitis, nausea and vomiting among compost and refuse workers compared to workers from water treatment plants (Lundholm and Rylander 1980; Sigsgaard et. al., 1994).

All the compost workers are exposed to organic dust. However, not all contracts symptoms of illness. In this project we focus on respiratory symptoms and the aim was to show individual differences in susceptibility as reflected by differences in histamine release from basophile leucocytes against suspensions of compost and *Aspergillus fumigatus*.

Materials and methods

Study population

Nine formerly compost workers, with respiratory symptoms at the time they stopped working with compost. And five control persons working in offices or laboratory.

Extracts

Compost from a plant which receives presorted organic household waste. After 24 hours in a rotating drum of the DANO type at a final temperature of 40°C, the rest of the composting process takes place in 5 aerated roofed piles. The compost stays in each pile for 10 days, before it is turned and moved to the next pile. The last pile is outdoors and the process takes 2-3 months. Extract have been made from compost aged 35 days after initial composting. The temperature of the compost at the time of sampling was approximately 70°C.

We have used a suspension of 35 days old compost or *Aspergillus fumigatus* (a gift from ALK-laboratory, Denmark) homogenized in sterile 0.9 % NaCl (30 mg/ml). For further dilution we have used the buffer from the assay.

Histamine release (Stahl Skov and Norn 1977)

Basophile leucocytes isolated by the percoll gradient method from 10 ml blood were used. A suspension of the isolated basophilocytes were incubated 40 min at 37°C with compost or *Aspergillus fumigatus* at different concentrations. The reaction was stopped and the cells were settled (2000 g, 10 min). The supernatant including the released histamine was thrown away to avoid interference. The cells were lysed and the rest histamine was measured by spectrofluometry after coupling to *o*-phthaldialdehyd. The results were found by subtraction of the rest histamine from the total histamine with a correction for spontaneous release (unexposed cells) and expressed as percent of the total. To show the IgE-mediated histamine release we analyzed in parallel intact cells and cells where we had removed cellbound IgE by washing for 55 sec in pH 3.8.

Results

The compost workers as well as the control persons were sensitive to compost. Three compost workers were sensitive against *Aspergillus fumigatus* in contrast to none of the control persons (table 1). When we studied the distribution of sensitivity with increasing concentration of the compost (figure 1) the control persons tended to react more at lower concentration of compost (1 mg/ml) compared to the compost workers. This difference was however only of border-line significance (P=0.06 Mann Whitney U-test). At the highest concentration there was no difference between the two groups.

Discussion

Composting is a destruction of organic material caused by microorganisms. It is known that some bacteria and peptidoglycan from there cell wall and some fungal spores

can cause histamine release from basophilocytes from both allergic and non-allergic persons (Norn et. al., 1985; Norn et. al., 1987; Larsen et. al., 1996). LPS enhances the histamine release caused by other allergens (Norn et. al., 1986). Compost contains at least one component which was able to release histamine from basophile leucocytes both from formerly compost workers and our control persons. However, it is not all types of organic dust which are able to induce histamine release from basophile leucocytes in our control persons. In another study we used suspensions of organic dust from swine confinement buildings and tested young farmers and the same control persons (not published). However, we found no histamine release from the control persons against swine dust. It seems like one has to be sensitized against a component to have a positive histamine release, and the ubiquitous exposure to moulds in the environment seems to be enough to become sensitized to compost. There was a borderline significant ($P=0.06$) trend for a raised release of histamine at concentration 1 mg/ml compost among the control persons in contrast to the compost workers (figure 1). Among the ex-compost workers histamine release was only seen at concentrations of 3.0 mg/ml compost. Only few of the compost workers still got symptoms at the time the blood sample was taken. Three of the workers were sensitized against *Aspergillus fumigatus* (table 1), however, non of them showed a positive skin reaction against this mold at the time the symptoms arose. None of the control persons reacted against *Aspergillus fumigatus*. In an earlier study with elderly farmers (Larsen et. al., 1997) we have found that the farmers with asthma showed an IgE-mediated histamine release against pig-protein. However, in the same study we were not able to show specific IgE against this allergen in traditional methods as RAST, immunoblotting etc.. In the light of this there seems to be a history of first developing an unspecific histamine reactivity against the complex organic dust, which might develop into specific allergy toward components in the dust.

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Table 1. Histamine release from basophile leucocytes exposed with suspensions of compost or *Aspergillus fumigatus*.

Histamine release	N	exposure	no re-lease	IgE-mediated	Non-immunology
Controle persons	5	Compost	-	2	3
		<i>Aspergillus</i>	5	-	-
Compost workers	9	Compost	1	4	4
		<i>Aspergillus</i>	6	2	1

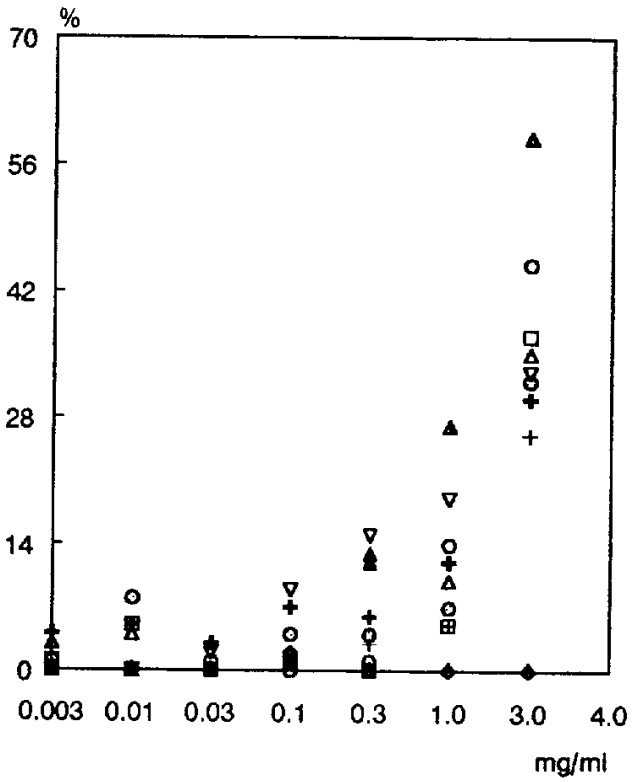
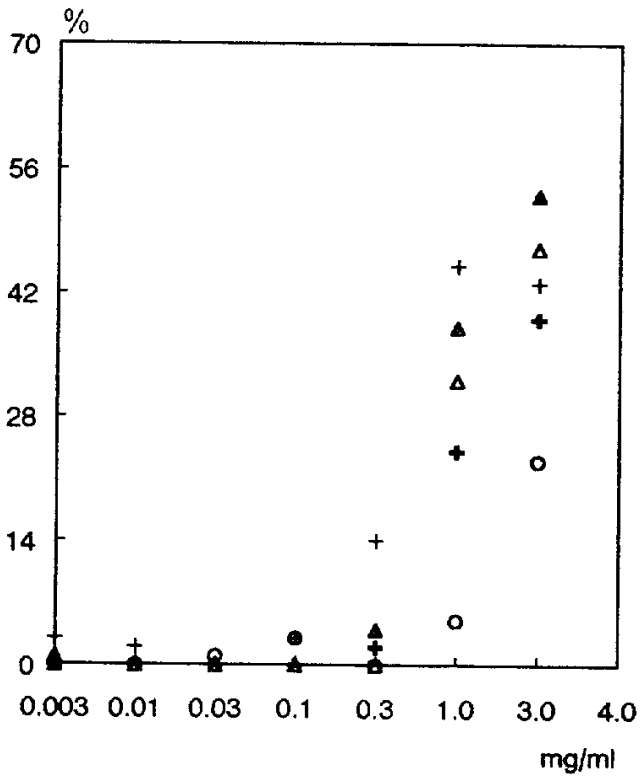


Figure 1. The distribution of sensitivity against compost. Top: the control persons. Bottom: The formerly compost workers.