

Fungal index in dwelling environments

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ABSTRACT

Microclimates in moisture chambers and environment in houses were evaluated using a fungal index. The index was calculated from the growth rate of a sensor fungus in a test piece, fungal detector, during an exposure period to the test environment. In the constant climates in the moisture chambers, higher indices were obtained at higher relative humidity. In the rooms with higher fungal indices, the densities of airborne fungi were higher, indicating a relationship between the index and fungal contamination. On beddings with positive fungal indices, which were folded and kept in closets, the densities of mite allergens increased during the exposure period of fungal detectors, and on those with negative indices the densities decreased. The houses in which fungal indices above 4 were detected in north rooms were regarded as chronically damp. The index must be a useful tool for detecting chronic dampness that brings on both contaminations by fungi and by mites.

INDEX TERMS

Fungal index; Fungi; Dampness; Contamination; Mite allergen

INTRODUCTION

Dampness induces fungal growth and mite proliferation in houses. These fungi and dust mites become sources of allergic diseases. A large quantity of spores spread in the air, and faecal pellets and body debris of dust mites cause respiratory symptoms like asthma or atopic dermatitis. The contamination by fungi and mites in houses are deleterious to the health of those who dwell there. Also, fungi cause damage to wood, paper, textiles, paint and other organic materials in houses. However, these contaminations are usually not noticed until the symptoms have developed or damage has occurred.

If it can be determined whether the environment within a house is damp or not, thus suitable for the growth of fungi and mite or not, measures can be taken. We, therefore, proposed the fungal index as an indicator of environment based on the growth of fungi as biosensors (Abe, 1993; Abe *et al.*, 1996). This index represents the capacity of a given environment to support the growth of fungi according to their responses (growth) to the environment.

In this paper, the fungal index was measured by using three fungi, two xerophilic fungi, *Eurotium herbariorum* and *Aspergillus penicilloides*, and a hydrophilic fungus, *Alternaria alternata*. This paper describes the relationships between the index and relative humidity (RH) in constant climates, between the index and densities of airborne fungi in a house, and between the index and the densities of dust mite allergens in houses.

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METHODS

Measurement of Fungal Index

A fungal index (Abe, 1993; Abe *et al.*, 1996), which represents a climate, was determined by using the response of a sensor fungus in a fungal detector. Figure 1 shows the detector exposed to a test environment, and Figure 2 shows the inner contents of the fungal detector. Spore suspensions of three sensor fungi, *Aspergillus penicillioides*, *Eurotium herbariorum* and *Alternaria alternata* are spotted on a plastic plate, dried, and covered with water vapour, but not spore permeable, transparent film so as not

to leak into the environment. This test piece was enclosed by non-woven fabric and sealed with heat (Figure 1).

The fungal index was determined as follows: (i) a fungal detector was exposed to a test environment; (ii) the sensor fungi in the detector were photographed under a microscope after the exposure; (iii) the hyphal length of sensor fungus that showed the highest response among the sensor fungi was measured, and the response to environment, response unit (Abe, 2001), was determined; (iv) fungal index was determined by the calculation:

$$\text{Fungal index} = X/Y$$

where X is the response unit and Y the exposure weeks.

Fungal Index and Relative Humidity in Constant Climate

A moisture chamber, in which the microclimate can be adjusted, was prepared using a disposable Petri dish 9 cm in diameter. The RH in each chamber was adjusted by the concentration of a glycerine solution. A square case 3 cm × 3 cm was placed in the Petri dish, and a glycerine solution was placed inside and outside of the square case in the dish. The RH in the moisture chamber was determined by calculation from the refractive index of the glycerine solution (ASTM E 104-51, re-approved 1971), which was determined using a refractometer (NAR3T, Atago, Co., Ltd.). To prevent changes in humidity in the chamber, the margin of the dish was sealed with a vinyl tape, and the dish was placed in an air-tight box adjusted to the same humidity using the same glycerine solution. For measuring the fungal index, a test piece in a fungal detector (Figure 2) was placed on a glass slide, which was placed on the square case in the Petri dish chamber, and incubated (exposed) 1, 2, 4, 7, 14, 28, 56 or 120 days at 25°C. The exposure period was varied depending on the climate so that the hyphal length fell into the measurable range between 10 and 2000 µm. After the exposure, the fungal index in each chamber was calculated.

Fungal Index and Airborne Fungi

A chemical sensitivity (CS) patient house built for care of CS patients at Asahikawa in Japan



Figure 1 Fungal detector on a wall.

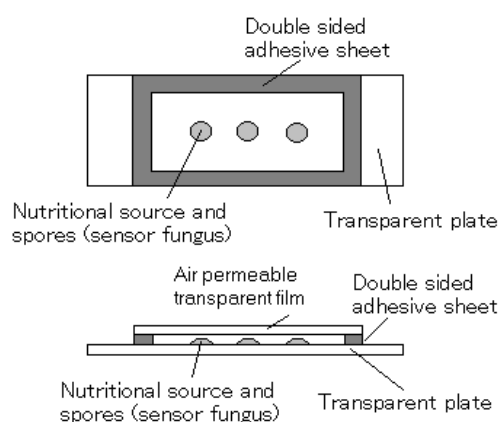


Figure 2 Inner contents of a fungal detector.

(Nakai *et al.*, 2001) was surveyed. Indoor chemical pollutant levels in this house were very low. However, the patients detected mould odours in the summer of 2001. So, fungal index and density of airborne fungi were examined in September and October of this year. For examination of fungal index, fungal detectors were placed at the lower site on the north wall in each room and exposed for 1 month. At the start and the end of the exposure, density of airborne fungi in each room was measured in the centre of the room using an RCS air sampler (Biotest Co., Ltd.).

Fungal Index and Mite Allergens

The fungal index in houses and mite allergens on beddings, which were folded and kept in closets in the houses, were determined in 16 houses in Isogo, Japan, in the summer of 2000. Fungal detectors were set at three sites in each house, on the north corner floor in a north bedroom, on the corner in a closet, and between the beddings that were folded and kept in that closet. The exposure period of the fungal detector was 8 weeks. Dusts on beddings were collected from ca. 1 m² surface of one side at the start of the fungal detector exposure, and again collected from the other side at the end of the exposure. The densities of mite allergens Der 1 and Der 2 per mg of dust were measured with ELISA method by SRL, Inc. (Tachikawa-City Tokyo, Japan). Der 1 is the sum of Der p1 and Der f1, derived from faecal pellets of *Dermatophagoides pteronyssinus* and *D. farinae*, respectively, and Der 2 is the sum of Der p2 and Der f2, derived from the bodies of *D. pteronyssinus* and *D. farinae*, respectively. The densities determined at the start and the end of fungal detector exposure were compared to detect mite proliferation during the exposure period.

RESULTS AND DISCUSSION

Fungal Index and Relative Humidity in Constant Climate

Figure 3 shows fungal indices in constant climates between 74 and 94% RH at 25°C. The indices shown in Figure 3 were determined by *Eurotium herbariorum* after 2–7 days of exposure. In these climates, this fungus showed the highest response among the three sensor fungi. A fungal index of about 180 was obtained at 94% RH, and a fungal index of about 8 was obtained at 75% RH. Higher indices were obtained at a higher RH in the climates between 74 and 94%RH.

Figure 4 shows fungal indices in constant climates below 75% RH at 25°C. The exposure period of the sensor fungi to the test climate was 14–120 days. Below 73% RH, *Aspergillus penicillioides* showed the

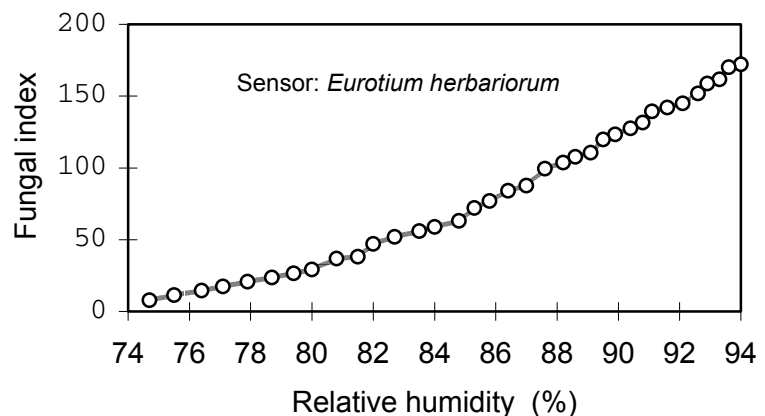


Figure 3 Fungal indices on constant climates with 74–94% RH at 25°C.

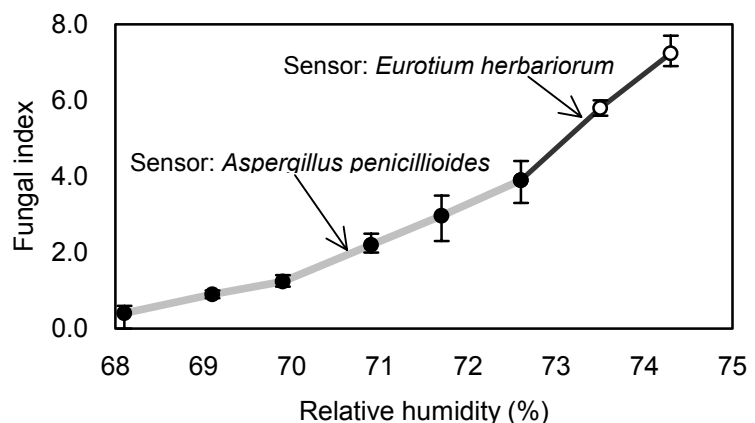


Figure 4 Fungal indices in constant climates below 75% RH at 25°C.

highest response, so that the fungal index in a humidity lower than 73% RH was determined using *Aspergillus penicillioides*. At about 73% RH, both *Aspergillus penicillioides* and *Eurotium herbariorum* showed the same response, and the fungal index was about 4.

At about 95% RH, *Alteraria alternata* and *Eurotium herbariorum* showed the same response. *Alteraria alternata* showed the highest response above 95% RH (data not show). In these climates, also, higher fungal indices were obtained at the higher RH.

Fungal Index and Airborne

Fungi

Figures 5 and 6 show the relationship between fungal indices and densities of airborne fungi in September and October, respectively. The densities of airborne fungi in these figures are the average of measurements at the start and the end of the fungal detector exposure. Higher densities of airborne fungi were detected in the rooms with higher fungal indices. In rooms with higher fungal indices, fungi might grow faster on the surface of the room walls during summer, and many fungal spores might spread from the extended hyphae of these fungi. Hamada has defined more than 1000 cfu/m³ as 'polluted' (Hamada and Yanada, 1994). Biological contamination must be a problem in the house for care of CS patients at Asahikawa, although chemical pollution was

not a concern at the house. The RH outside of the house was high in the summer (about 80%, data not shown). With the windows open all day long, wet outdoor air came into the house, making the indoor air humid. Few chemicals that are poisonous to living organisms were observed, so that this indoor air might be a suitable environment for the growth of fungi.

Fungal Index and Mite Allergens

Table 1 shows the fungal indices in houses. At five houses, Nos. 3, 7, 8, 10 and 16, fungal indices above 4 were detected in the north bedrooms. In these houses, fungal indices were detected at the other two sites. At five houses, Nos. 4, 5, 11, 13 and 14, fungal indices below 3 were detected in the north bedrooms. In these houses, fungal indices were not detected between beddings. At six houses, Nos. 1, 2, 6, 9, 12 and 15, negative fungal indices (fungal index below 0.9) were detected at each site. The highest index value was detected in the north bedrooms among the three surveyed sites in all houses. It was suggested that the north bedroom is a good survey site to detect dampness in a dwelling house.

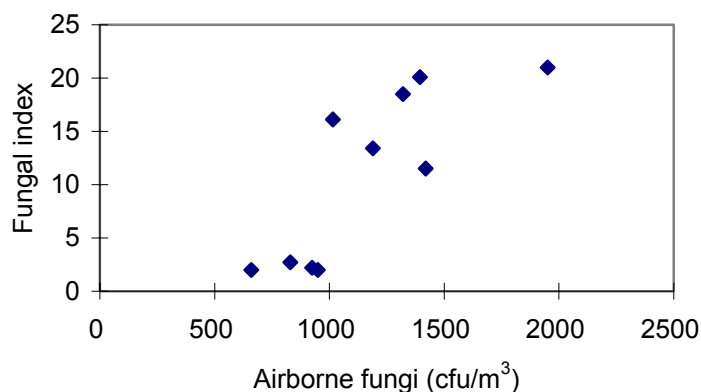


Figure 5 Comparison of fungal index and density of airborne fungi in September.

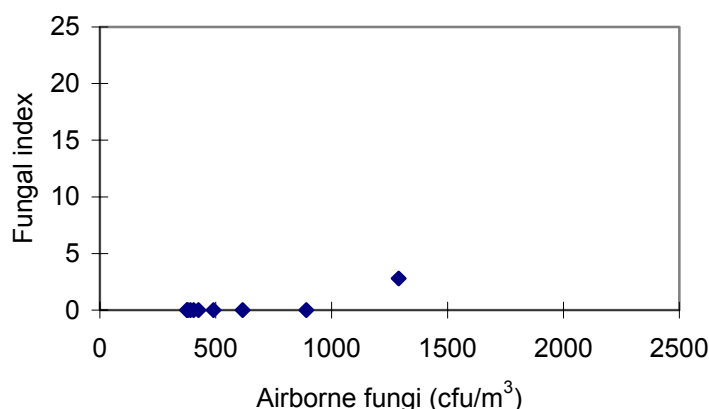


Figure 6 Comparison of fungal index and density of airborne fungi in October.

Table 1 Fungal indices in 16 houses

House no.	Fungal index		
	North bedroom	Closet	Beddings in closet
1	<0.9	<0.9	<0.9
2	<0.9	<0.9	<0.9
3	4.5	2.5	3.2
4	2.0	<0.9	<0.9
5	2.6	<0.9	<0.9
6	<0.9	<0.9	<0.9
7	4.5	3.4	4.6
8	4.2	4.2	1.7
9	<0.9	<0.9	<0.9
10	4.9	2.0	1.4
11	2.3	2.5	<0.9
12	<0.9	<0.9	<0.9
13	2.6	<0.9	<0.9
14	2.9	<0.9	<0.9
15	<0.9	<0.9	<0.9
16	4.5	3.4	2.2

<0.9: Fungal index was not detected. No germination in sensor fungi after 8 weeks of exposure.

Table 2 Mite allergen in 16 houses

House no.	Density of mite allergens in dusts on beddings ($\mu\text{g/g}$ dust)					
	Der 1			Der 2		
	First	Second	Difference	First	Second	Difference
1	1.5	0.4	-1.1	2.6	0.9	-1.7
2	11.7	0.2	-11.5	15.4	0.9	-14.5
3	21.6	40.9	19.3	44.8	35.0	-9.8
4	32.0	9.4	-22.6	41.7	10.4	-31.3
5	75.8	58.4	-17.4	58.1	33.5	-24.6
6	5.8	4.2	-1.6	4.3	1.8	-2.5
7	21.1	31.5	10.4	23.5	35.4	11.9
8	5.9	19.5	13.6	3.6	17.0	13.4
9	4.4	1.3	-3.1	4.1	2.3	-1.8
10	6.6	10.0	3.4	6.6	13.0	6.4
11	16.9	16.0	-0.9	12.2	13.7	1.5
12	47.5	27.0	-20.5	52.4	17.8	-34.6
13	11.9	2.0	-9.9	12.5	3.5	-9.4
14	14.4	6.4	-8.0	17.9	10.7	-7.2
15	0.4	0.1	-0.3	0.0	0.0	0.0
16	34.1	36.6	2.5	36.6	29.1	-7.5

First: the time when fungal indices were set to the test sites.

Second: the time when fungal indices were removed from the test sites.

Difference: the density of allergen determined at the second time minus that determined the first time. The minus values indicate decrease of the densities.

Table 2 shows the densities of mite allergens Der 1 and Der 2 measured at the beginning and the end of fungal detector exposure to survey sites, and the differences between the values

of those two measures. The densities of Der 1 increased in the dusts on beddings that showed positive fungal indices in the five houses, Nos. 3, 7, 8, 10 and 16, where fungal indices in the north room exceeded 4, and did not increase in beddings that showed negative fungal indices. The densities of Der 2 increased in three beddings among the five. The densities of Der 2 in the dusts on beddings decreased at all six houses in which negative fungal indices were detected at every site. In houses where fungal indices above 4 were detected in the north bedrooms, mite must also proliferate on the beddings kept in the closet during the exposure period of fungal detectors. At the second measurement (the end of fungal detector exposure), the densities of Der 1 were above 10 µg/g dust in all the beddings in these houses. A level associated with increased risk of asthma is reported to 10 µg/g dust (Platts-Mills *et al.*, 1987). Measures should be taken in the houses where fungal indices above 4 were detected. We would like to categorize these houses as chronically damp, in which fungal indices above 4 were detected in north bedrooms.

CONCLUSION

1. In constant climates higher fungal index values were obtained at higher RH.
2. Fungal index correlated with the densities of airborne fungi.
3. House dust mite allergens increased on beddings stored in closets that showed a fungal index above 4 at the floor of the north room.
4. We propose that houses in which fungal indices above 4 were detected on the floor in the north room be categorized as chronic damp homes.
5. Fungal index is a useful tool for detecting chronic dampness that induces fungal and mite proliferation.

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