ORIGINAL ARTICLE

Assessment of the decay risk of airborne wood-decay fungi III: decay risks at different sampling sites

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Abstract The decay risk of airborne wood-decay fungi in the same volume of air was investigated by using an air sampler over the course of a year at three different sampling sites. Japanese cedar disks measuring 7.8 cm in diameter and about 3 mm in thickness, and with a moisture content of about 100 % were placed in a "BIOSAMP" air sampler and then exposed to 1000 l of air in the northern, central, and southwest parts of Japan. The exposed disks were incubated for 20 weeks in a damp container maintained at 26 ± 2 °C and degraded by fungi trapped on the disks. The decay risk was calculated from the mass loss during incubation, and the factors affecting the said risk were explored. The results showed that sampling sites apparently do not affect decay risk, even though the Scheffer's climate indexes of the sites were quite different. The relation between the sampling month and decay risk reveals that decay risk remains virtually the same year-round. Relative humidity on a sampling day is one of the key factors affecting decay risk in sampling conducted at the central or southwest site. In contrast, no weather factors influenced decay risk at the northern sampling site.

Introduction

Since harvested wood products have been defined as a carbon sink [1, 2], it is very important to prolong the service life of wooden products and delay their emissions of carbon dioxide gas. One possible method of retarding gas emissions is through wood protection [3], and many investigative studies have been conducted on preservatives and chemical treatments [4–6].

Research on the factors that accelerate carbon dioxide gas emissions from wooden structures is also important and has revealed that one of the most important biological factors in accelerating emissions is wood-decay fungi [7, 8]. The decay risk of wooden products was consequently investigated through various conventional testing methods, such as field tests [9–11], the lap joint test, L-joint test and double-layer test [12], and is thought to be predicted by Sheffer's climate index [13] in the case of outdoor aboveground wood products. In addition to these conventional studies and new study using potato dextrose agar plates [14], we developed a novel method of assessing the decay risk of airborne wood-decay fungi in a certain amount of air by using an air sampler and wood disks [15, 16].

In our previous paper [15], we employed a novel method of assessing decay risk in the same volume of air and concluded that relative humidity on a given sampling day influenced the decay risk. The study was not complete, however, as the previous study lacked data on the winter season. Moreover, the study was only conducted at one sampling site located in central Japan. Therefore, whether

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Table 1 Location of the three sampling sites

Sampling site	Abbreviation of site	Latitude (°C)	Longitude (°C)	Sea level (m)	Mean temp. (°C)[a]	Precipitation (mm)[a]
Hokkaido Research Center, FFPRI[b]	N-Site	43.0 N	141.4 E	140	8.6	72.2
Head Office of FFPRI[b]	C-Site	36.0 N	140.1 E	20	14.8	96.7
Kyushu Research Center, FFPRI[b]	SW-Site	32.8 N	131.7 E	45	16.6	129.8

[a] Mean temperature and total precipitation from May 2010 to May 2011

[b] *FFPRI* Forestry and Forest Products Research Institute

the above relation applies to other sampling sites remains unclear.

To confirm the question posed above, we conducted similar experiments at different sites whose climate indexes are quite different. This paper examines the decay risk of airborne wood-decay fungi at these sites during the course of an entire year. It also discusses the weather factors affecting decay risk.

Materials and methods

Preparation of wood specimens

Wood specimens were prepared using the method employed in the previous study [15]. Japanese cedar (*Cryptomeria japonica*) disks about 3 mm thick were dried at 60 °C for 1 day and then weighed to determine their initial mass (M_0). Disks were randomly placed in individual Petri dishes and then into individual sealed bags for gas sterilization. Gas sterilization was carried out in ethylene oxide gas sterilizers (Model YS-A-C64E, Yuyama Co. Ltd., Osaka). Each disk in a sterilized sealed bag was randomly sent to three sampling sites.

Sampling and incubation procedure

Air sampling was conducted at the three sampling sites listed in Table 1. The sampling points at the sampling sites were chosen to have similar surrounding environments, and air at each site was sampled on a sunny or a cloudy day, but not on a rainy day.

A plate containing a Japanese cedar disk kept in a sterilized sealed bag was removed from the bag at each site just before air sampling. Six mL of sterilized deionized water was also added aseptically to each disk. The "BIOSAMP" air sampler (Midori Anzen Co. Ltd., Tokyo) was used to conduct air sampling. The sampling procedure followed the product manual except for replacing the agar medium for damp Japanese cedar disks. Each disk exposed to 1000 l of air at about 100 cm in height was kept in a
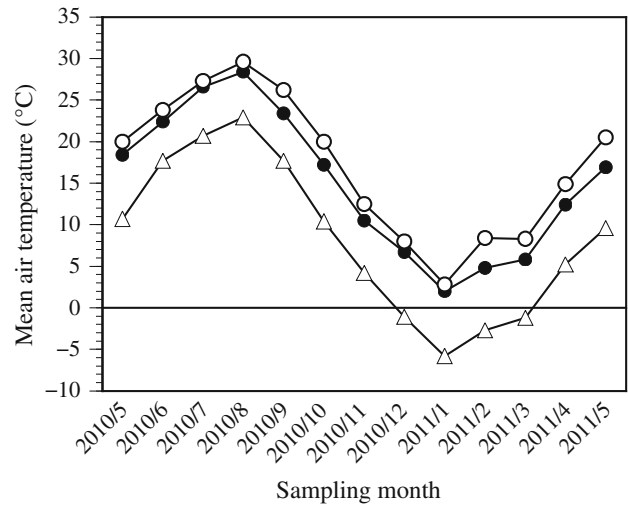

Fig. 1 Mean monthly air temperature at three sampling sites from May 2010 to May 2011. The *open triangle*, *closed circle*, and *open circle* denote the mean air temperature at the N-site, C-site, and SW-site, respectively

plastic plate and incubated in a damp container at 26 ± 2 °C. After incubation for 20 weeks, each disk was dried at 60 °C for 2 days. Then the disk mass after incubation (M_1) was measured. The mass loss during the incubation period was calculated from M_0 and M_1.

Collection of weather data

Weather data for the N-site and that for the C-site were collected by the MC-2100 weather monitoring system (Koshin Denki Kogyo co. Ltd., Tokyo) and the C-CR1000 weather monitoring system (CLIMATEC Inc., Tokyo), respectively. Both systems were set at about a few hundred meters away from each sampling site. Weather data for the SW-site was obtained from Japan's Meteorological Agency (as monitored at about 3.5 km west of the SW-site).

Scheffer's climate index [13] at each site was calculated according to the following equation:

$$\text{Climate index} = \sum_{\text{Jan.}}^{\text{Dec.}} [(T - 35)(D - 3)]/30 \qquad (1)$$

where T denotes the mean monthly temperature (°F) and D the mean number of days in the month with 0.01 in. or more of precipitation.
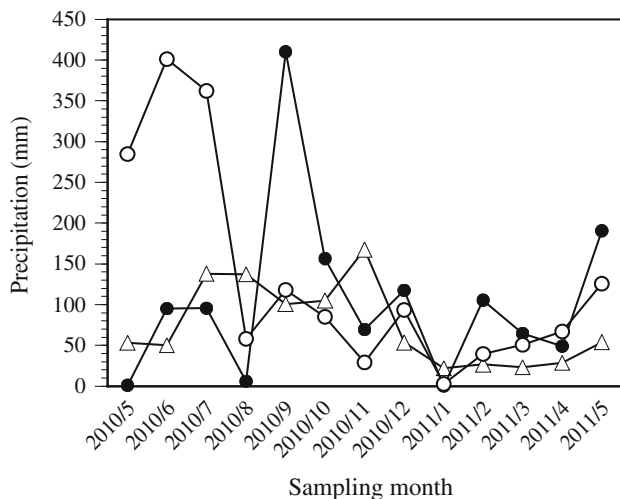

Fig. 2 Amount of monthly precipitation at three sampling sites from May 2010 to May 2011. The *open triangle*, *closed circle*, and *open circle* denote the amount of precipitation at the N-site, C-site and SW-site, respectively

Table 2 Scheffer's climate index at the three sampling sites

Sampling site	N-site	C-site	SW-site
Climate index	44	68	103

Statistical analysis

All statistical analyses were conducted using JMP 9.0 software (SAS Institute, Cary, North Carolina, USA) with a significance level set at 0.05 [17].

Results and discussion

Decay risk and sampling sites

Figure 1 shows the mean monthly air temperature at the three sampling sites during the sampling period from May 2010 to May 2011. As expected from the latitude of each site listed in Table 1, the air temperature of each sampling site decreases in line with higher site latitude. For instance, the mean monthly air temperature at the N-site was about 10 °C lower than at the other sites. Figure 2 plots the amount of precipitation at each site. Although the monthly data varied widely, particularly at the C-site and SW-site, the total precipitation at each site also decreased in line with the sampling site latitude.

Since the decay risk of outdoor aboveground wood products is known to be in accordance with Scheffer's climate index [18], the climate index of each sampling site is calculated (Table 2). According to Scheffer's climate index, the decay risk must increase in line with lower sampling site latitude.

To compare our results with the decay risk predicted by Scheffer's climate index, we first investigated the relation between the sampling sites and the mass loss caused by airborne wood-decay fungi. Figure 3 shows the distributions
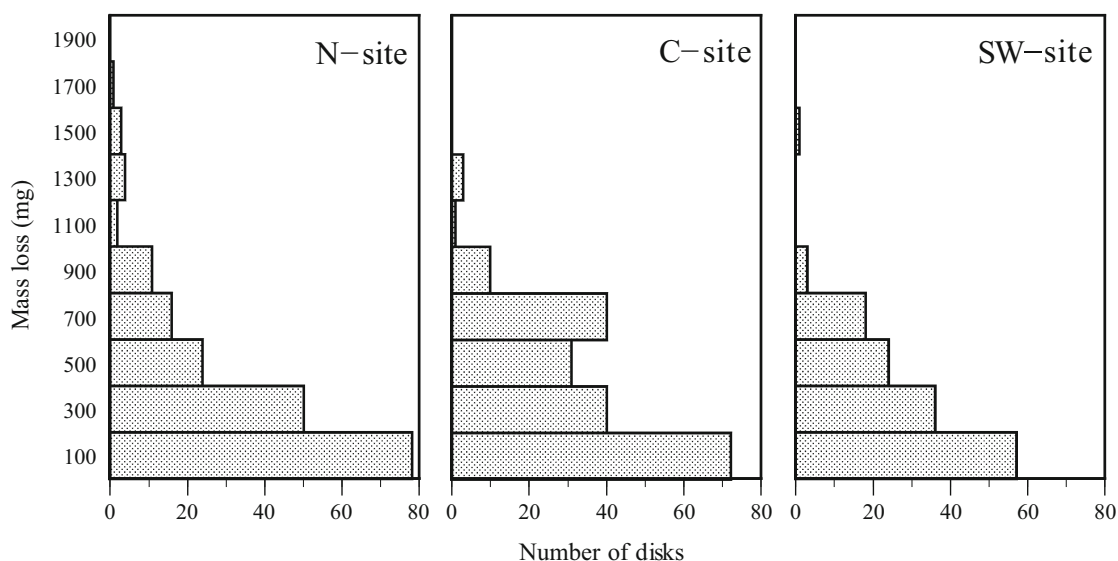

Fig. 3 Distributions of the mass loss of specimens exposed to three different sites

of mass loss at the three sampling sites. The figure suggests no differences among the mass loss at each sampling site, even though the climate indexes of the three sites are quite different. The non-parametric Steel–Dwass test revealed no significant differences among the mass loss at each sampling site. Our experimental results clearly indicated about the same level of decay risk caused by airborne fungi at all three sampling sites.

Perhaps, the reason why the experimental results of this study differ from those predicted by Scheffer's climate indexes may be the temperatures at which the wood specimens were incubated. Scheffer's index is an equation used to estimate the decay risk of outdoor aboveground wood products. Therefore, the index must include the effect of outdoor temperature in the equation. In contrast, we set the temperature to 26 °C during the incubation period because we intended to estimate the decay risk of airborne wood-decay fungi in 1000 l of air. Thus, Scheffer's climate index was considered unsuitable for predicting the decay risk of wood products placed in an environment where the products' ambient temperature did not change with the outdoor temperature. Timber frames covered with insulating materials may be one wood product whose temperature is not affected by outdoor temperature. Scheffer's climate index must preferably be used to define the decay risk of building materials outside of insulators, but not inside them.

Sampling month and decay risk

In the previous paper [15], we reported that decay risk was not affected by the sampling month. However, we reached this conclusion based on sampling data collected from June to December in 2008. To confirm whether the conclusion can be applied to the winter season, air sampling in this study was conducted over the course of an entire year. Figure 4 shows the relation between the sampling month and the mass loss at each site. The months of winter showed relatively low decay risk, but no differences were observed in the months of other seasons. Nonparametric comparisons for all pairs using the Steel–Dwass test was applied to investigate whether any significant difference existed in terms of the decay risk in each month. Figure 4 also shows the results. At all three sampling sites, significantly lower decay risk ($p < 0.05$) was observed in 1 month during winter, but no significant differences were observed among the other months. We therefore conclude that at present, the decay risk is virtually the same in each month at each area, even though the risk drops during 1 month in winter.

Figure 4 also shows another fact worthy of mention. As listed in Table 1 and shown in Fig. 1, the N-site is located in northern Japan, and the mean air temperature during the

winter season drops below 0 °C. Therefore, any rainfall at this site in winter becomes snow, which remains on the ground until spring. During the sampling period from January to March 2011, the snow was about 30–90 cm deep at this site. On the other hand, decay risk at the N-site does not remain zero throughout the year. Moreover, higher mass loss was observed in several wood disks exposed to air in winter. It is reported that many wood-decay fungi preferentially produce spores between zero to 10 °C [19] and release them to the environment [20, 21].
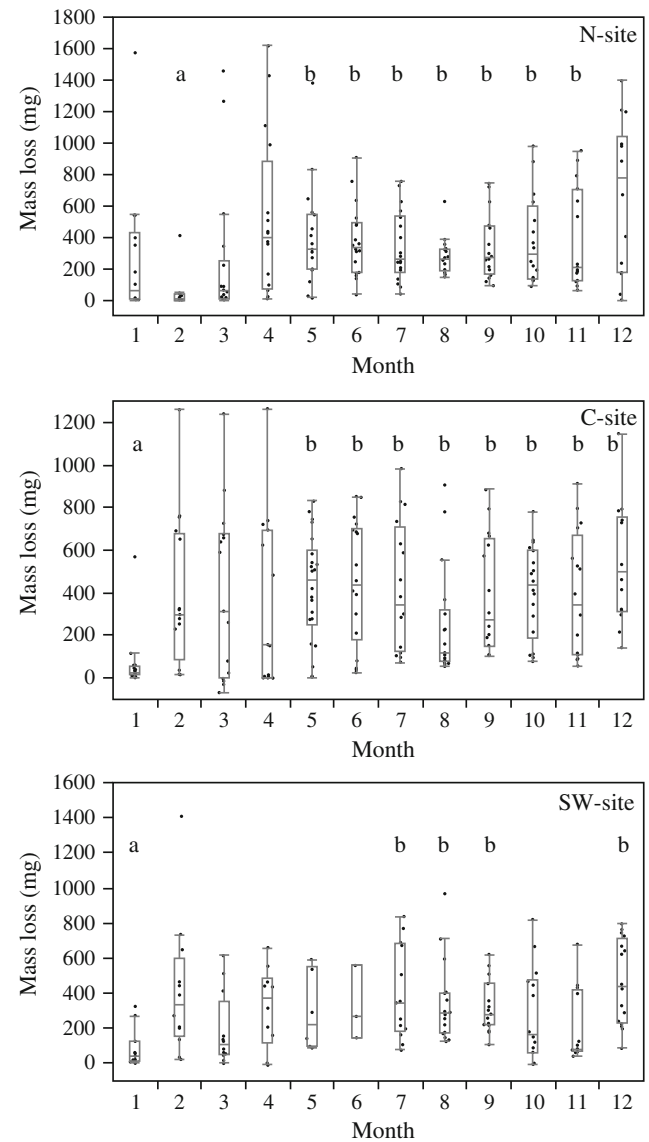

Fig. 4 Relation between sampling month and mass loss at three different sites. The *vertical lines* within the *boxes* represent the median sample values; the ends of the *boxes* represent the 75th and 25th quantiles. The *upper* and the *bottom lines* indicate the values calculated as follows: *upper line*, 3rd quartile + 1.5 × (75th quantile − 25th quantile); *lower line*, 1st quartile − 1.5 × (75th quantile − 25th quantile). *Different letters* indicate significant difference at $p < 0.05$ by Steel–Dwass's test
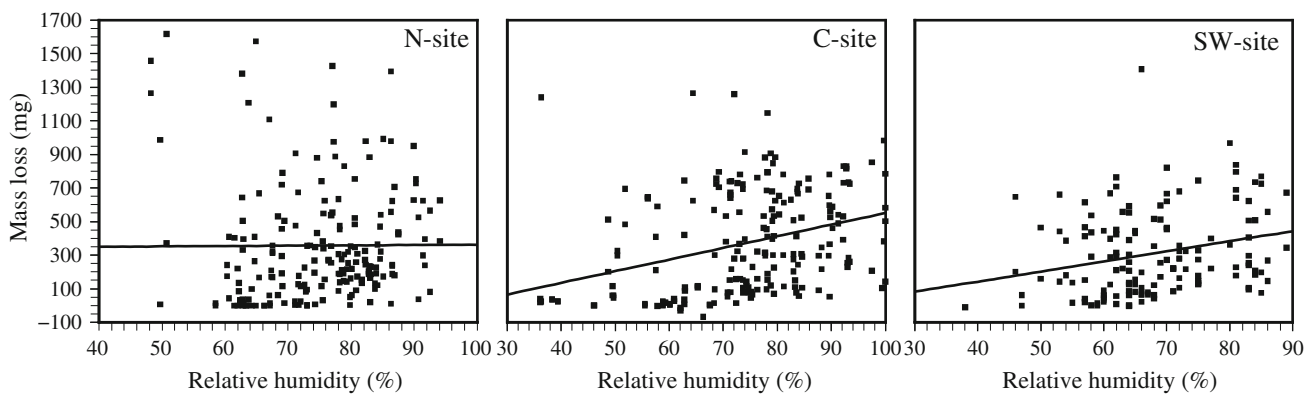
Fig. 5 Relation between relative humidity and decay risk. Each *solid line* indicates the linear least square fit of mass loss against relative humidity given by the following equation: N-site, mass loss = 0.207 × relative humidity + 342; C-site, mass loss = 6.93 × relative humidity − 142; SW-site, mass loss = 6.02 × relative humidity − 98.3

Some wood-decay fungi at N-site also release spores even under such conditions as ambient air temperature around the fruit body near 0 °C, though the ground is covered with thick snow.

Effect of weather factors

In the previous study, the decay risk of airborne wood-decay fungi was confirmed as being correlated with weather factors that indicated sampling was done on a cloudy day. Mean relative humidity and lowest relative humidity on the sampling day are among the most relevant factors in assessing decay risk. This fact looks acceptable because spore germinations are affected by the wood moisture content that becomes higher in line with higher relative humidity [22–24]. Therefore, fungi must release more spores on a cloudy day than a shiny day.

To confirm this relation, retests were done at all three sites as previously mentioned (Table 1). Figure 5 shows the relation between the mass loss of the disks and the mean relative humidity on the sampling day at each sampling site. Regression lines are also given in the figure. As shown in the figure, the relation between mass loss and relative humidity is not the same at all three sampling sites.

At the C-site and the SW-site, the relation is the same as that observed in the previous study, that is, a positive correlation exists between mass loss and mean relative humidity on the sampling day ($p < 0.05$). Contrary to this relation, the mass loss of disks exposed at the N-site was not affected by the mean relative humidity on the sampling day. This inconsistency may be partially due to fungi that caused mass loss above 1000 mg during the 20-week incubation period. Many such fungi appeared on a day of low relative humidity. The DNA sequences of these fungi are now being analyzed and will be discussed in our next paper.

Table 3 summarizes the relation between decay risk and weather factors. These results suggest that weather factors affecting decay risk are not equal among the different sampling sites.

The decay risk by airborne wood-decay fungi is affected by relative humidity, the amount of solar radiation, and duration of sunshine at the C-site, and by temperature, relative humidity, and atmospheric pressure at the SW-site. Positive and negative signs of the correlation coefficient of these factors suggest that decay risk increases on humid and cloudy days at the C-site, and on warm and humid days with low atmospheric pressure at the SW-site.

Table 3 Correlation between weather factors and mass loss of Japanese cedar disk

Weather factor	N-site		C-site		SW-site	
	Correlation coefficient	*p* value	Correlation coefficient	*p* value	Correlation coefficient	*p* value
Mean temperature (°C)	0.032	0.665	0.002	0.976	**0.174**	**0.039**
Relative humidity (%)	0.006	0.936	**0.327**	**<0.0001**	**0.250**	**0.003**
Precipitation (mm)	0.058	0.427	0.105	0.137	0.087	0.398
Atmospheric pressure (hPa)	−0.097	0.182	−0.073	0.304	**−0.334**	**<0.0001**
Amount of solar radiation (MJ/m^2)	0.002	0.980	**−0.167**	**0.018**	−0.020	0.811
Sunshine duration (h)	0.035	0.631	**−0.159**	**0.025**	−0.104	0.218

Significant correlations ($p < 0.05$) are indicated in boldface

In contrast to these warmer sites, the N-site shows that its decay risk is not influenced by any weather factors. It will be interesting to reveal whether any correlation exists at other high-latitude areas similar to the N-site.

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