

Risk of Damage Inside Wooden Cultural Heritage Sites Based on Temperature, Humidity, and Airborne Fungi in South Korea

Ik-Gyun Im ,^{a,b} and Gyu-Seong Han ,^{b,c,*}

The temperature, humidity, and indoor airborne fungi were evaluated at 24 wooden cultural heritage sites (WCHs) (5 from field surveys and 19 from previous studies). Surface contaminating fungi was present year-round in the indoor space of WCHs. Wood decay fungi float only in summer and fall when relative humidity is high. The internal conditions of WCHs, such as an average temperature of 20 °C and a relative humidity of 75% or more, persisted for at least 4 weeks and up to 8 weeks of the year. Accordingly, the growth of surface contaminating fungi and wood decay fungi on wood and Hanji (Korea paper) surfaces was evaluated for 12 weeks. There was no growth at relative humidity of 95% or less. The maximum average relative humidity of 24 WCHs nationwide was 90% or less, which makes it impossible for most wood decay fungi to germinate and grow, and the period when it remained above 95% was very short. Therefore, the indoor space of WCHs is unlikely to be damaged by surface contaminating fungi and wood decay fungi in all periods of the year, as the environment suitable for germination and growth is not created for a long period.

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Contact information: a: Conservation Laboratory, Curatorial Affairs Office, Gyeongju National Museum, Gyeongju 38171, Korea; b: Department of Cultural Heritage Science, Chungbuk National University, Cheongju, 28644, Korea; c: Department of Wood and Paper Science, Chungbuk National University, Cheongju, 28644, Korea; *Corresponding author: wood@chungbuk.ac.kr

INTRODUCTION

Due to their outdoor location, Wooden Cultural Heritage (WCH) sites in Korea are directly exposed to various weather events throughout the year, including torrential rains, heat waves, and heavy snowfall. As a result, the wooden components of the structure and the organic cultural heritage stored inside (wood, paper, fibers, *etc.*) are affected by the year-round climate and exposed to various deterioration factors. The main deterioration factors of organic cultural properties due to climate include meteorological factors (temperature, humidity, sunlight, rainfall, *etc.*), biological factors (plants, animals, microorganisms), and natural phenomena such as natural disasters (Sato 2019).

Among the deterioration factors of cultural properties, fungi, a biological factor, affects various materials with respect to odor, discoloration, and deterioration when damage occurs (Kosuke 2012). In particular, temperature and relative humidity, which are weather factors, have a significant impact on the germination and growth of various airborne fungi present inside. Especially during the summer rainy season, wooden cultural

properties are affected by the high temperature and relative humidity outside. In such an environment, if the air quality stagnates due to poor air circulation, the wood and organic cultural materials inside are subject to biological damage by microorganisms (Hong *et al.* 2018, 2019). For example, many cases of damage caused by fungi have been confirmed in wooden plates and large Buddhist paintings due to poor air circulation and high humidity during the annual rainy season (Park 2011; NRICH 2016, 2017, 2018, 2019, 2020, 2021, 2022, 2023). The germination and growth of these indoor airborne fungi on the surface of organic cultural assets can cause material weakening and discoloration, and secondary damage from insects that feed on the fungi and the weakened material. Over the past 106 years, from 1912 to 2017, the Korean Peninsula has experienced an earlier spring (13 days) and summer (10 days), and a later fall (9 days) and winter (5 days), with summer increasing by 19 days, from 98 to 117 days, and winter decreasing from 109 to 91 days (NIMS, 2018). This increase in the number of summer days can increase the risk of damage due to the germination and growth of fungi present in the interior and exterior of wooden cultural properties, so it is essential to periodically monitor the interior spaces of wooden cultural properties to identify the risk of damage and respond appropriately.

Therefore, since the 2010s, cultural heritage-related organizations such as the National Research Institute of Cultural Heritage (NRICH) and the Research Institute of Buddhist Cultural Heritage (RIBCH) have been conducting conservation environmental surveys such as year-round temperature and humidity and suspended microorganism surveys in the interior and exterior spaces of wooden cultural properties that house organic cultural properties such as Buddhist statues, large Buddhist painting and wooden tablets (NRICH 2016, 2017, 2018, 2019, 2020, 2021, 2022, 2023). However, the temperature and humidity surveys of the indoor spaces of WCHs have been conducted for less than a year for one or two WCHs, making it difficult to draw meaningful conclusions about changes in the internal environment. In addition, while the distribution and diversity of airborne fungi inside wooden cultural properties have been continuously investigated, damage risk studies linking fungal status data with climatic factors such as year-round internal temperature and relative humidity are lacking. The need for such a study has been mentioned as part of the assessment of the internal and external impacts of climate change on WCHs since 2010, but no conservation environmental management studies have been conducted except for the establishment of a disaster prevention facility management system (KRICH, 2010; CHA, 2020). In Europe, climate change impact assessments of cultural heritage have been conducted since the early 2000s, focusing on the prevention and mitigation of damage to cultural heritage, to identify the types of disasters and damage that are likely to occur to cultural heritage due to climate change and to develop strategic responses (Kim and Kim 2021). Among these early studies, NOAH's ARK, a pan-European research project on cultural heritage in the context of climate change from 2004 to 2007, prioritized biological damage to wood due to changes in relative humidity as an initial research topic for wooden cultural heritage (Sabbioni 2006). In addition, the recently revised UNESCO climate change policy document recommends that by 2030, adopting countries must develop risk assessment tools for cultural heritage and establish risk monitoring frameworks for climate change adaptation (UNESCO 2021). In line with this international response to climate change in cultural heritage, it is now necessary to move from identifying the diversity and impact characteristics of airborne fungi inside wooden cultural properties to understanding their relationship with climatic factors that affect their germination and growth.

This study analyzed 24 WCHs (5 WCHs from field surveys and 19 WCHs from previous studies) with year-round temperature and humidity surveys and indoor airborne fungus collection within the last 10 years. The goal was to understand the current status of relative humidity and indoor airborne fungi in WCHs in Korea during summer and fall. In addition, the growth evaluation experiment of surface contaminating fungi and wood decay fungi identified in the field survey was conducted to identify the actual germination and growth conditions to determine whether damage is possible.

EXPERIMENTAL

Investigating Temperature, Humidity, and Airborne Fungi Inside WCHs

To investigate seasonal temperature, humidity, and airborne fungi inside WCHs, field surveys were conducted at five WCHs (Yangju Cheongnyunsa, Cheongju Ansansa, Gimcheon Jikgiji, Gangjin Muwiisa, and Yeosu Heungguksa) (Table 1a). The data of 19 WCHs conducted by NRICH every year from 2017 to 2021 was used for comparative analysis (Table 1b).

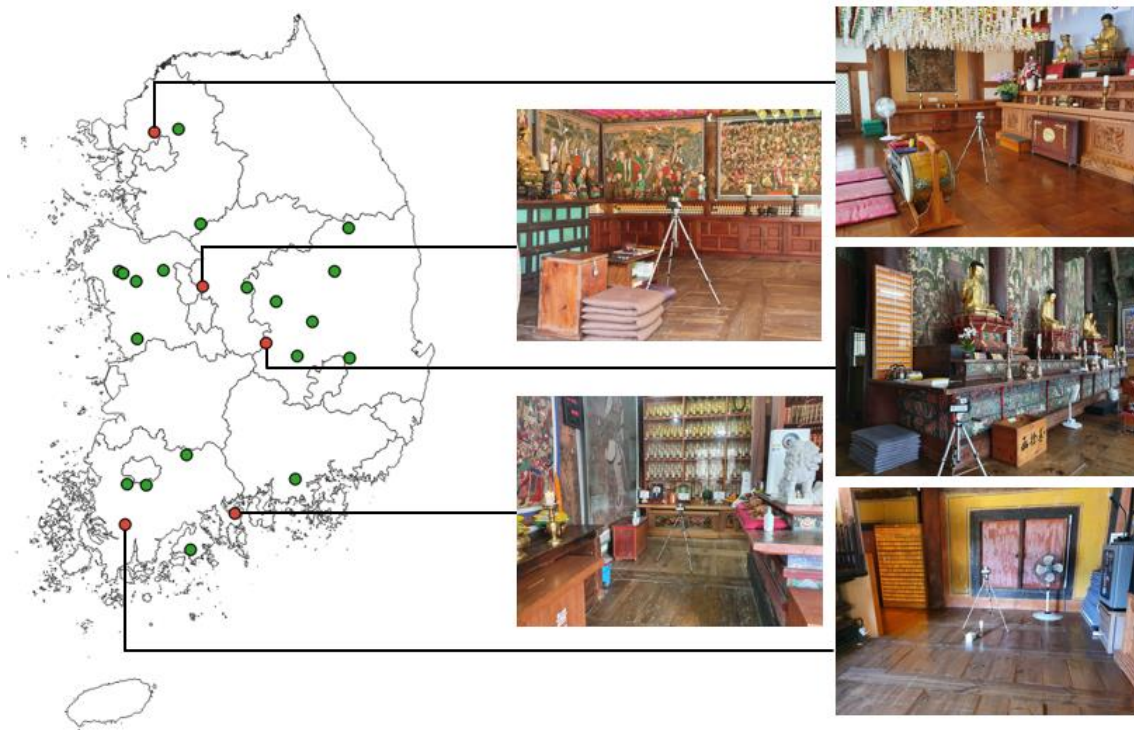


Fig. 1. WCHs indoor temperature and humidity and airborne fungi survey locations (Red dot: Fieldwork, Green dot: WCHs already researched in prior studies)

For the temperature and humidity surveys of the five WCHs with field surveys, Real-Time Monitoring (RTM), a system for real-time temperature and humidity verification and data collection introduced in the study by Im *et al.* (2021), was installed in the field and data were collected at 10-min intervals for 18 months from July 2021 to January 2023. As the risk period of xerophilic fungi damage due to increased internal humidity in WCHs has been identified as the summer season (June-September) (Im and Han 2023), the temperature and

humidity data of a total of 24 WCHs, including 19 from previous studies, were divided into two periods, summer (June-August) and fall (September-November), which are the highest risk periods for fungi damage throughout the year, to check the internal temperature and humidity status of WCHs at the national level.

In addition, the June-September internal temperature and humidity data of the five WCHs in the field survey (Table 1a) were organized into monthly periods, and hourly averages were calculated to understand the trend of temperature and humidity changes in the internal space by time of day. The seasonal average temperature and humidity data of the 24 WCHs identified in the field survey and previous studies (Table 1b) were used to select the evaluation strains in '2. Identification of the growth characteristics of the airborne fungi inside the WCHs'. The indoor airborne fungi of the five WCHs under field investigation were collected four times (July 21, October 21, January 22, and April 22) from June 2021 to April 2022, and molecular biological identification was performed to identify seasonal differences (Fig. 1).

Table 1. Mortality Over Time in Each Termiticide-treated Group using the Petri Dish Method

No.	Region		Temple	latitude & longitude		Wooden Cultural Property
1	Gyeong-gi	Anseong	Chiljangsa ^b	37.0265	127.3975	Daeungjeon Hall
2		Namyangju	Bongseonsa ^b	37.7473	127.1838	Main Buddha Hall
3		Yangju	Cheongnyeongsas ^a	37.7299	126.9480	Daeungjeon Hall
4	Chung-buk	Boeun	Beopjusa ^b	36.5420	127.8330	Daeungjeon Hall
5		Cheongju	Ansimsa ^a	36.5522	127.4130	Daeungjeon Hall
6	Chung-nam	Buyeo	Odeoksa ^b	36.1557	126.7968	Daeungjeon Hall
7		Cheonan	Gwangdeoksa ^b	36.6760	127.0428	Wooden Buildings
8		Hongseong	Yongbongsas ^b	36.6502	126.6556	Jijangjeon Hall
9		Yesan	Daeryeonsas ^b	36.5874	126.7820	Geungnakjeon Hall
10		Yesan	Sudeoksas ^b	36.6630	126.6227	Daeungjeon Hall
11	DaeGu	Gunwi	Beopjusa ^b	36.2728	128.4379	Bogwangjeon Hall
12	Gyeong-buk	Andong	Bongjeongsas ^b	36.6533	128.6624	Daeungjeon Hall
13		Bonghwa	Chukseosas ^b	36.9813	128.7998	Daeungjeon Hall
14		Gimcheon	Jikjisas ^a	36.1164	128.0038	Daeungjeon Hall
15		Sangju	Namjangsas ^b	36.4309	128.1056	Geungnakbojeon Hall
16		Seongju	Seonseoksas ^b	36.0160	128.2989	Wooden Buildings
17		Yeongcheon	Eunhaesas ^b	35.9920	128.7896	Geungnakbojeon Hall
18	Gyeong-nam	Goseong	Okcheonsas ^b	35.0802	128.2623	Daeungjeon Hall
19	Jeon-nam	Gangjin	Muwisas ^a	34.7384	126.6868	Geungnakbojeon Hall
20		Goheung	Geumtapsas ^b	34.5429	127.2871	Geungnakjeon Hall
21		Gokseong	Dorimsas ^b	35.2670	127.2575	Bogwangjeon Hall
22		Naju	Dabosas ^b	35.0472	126.6983	Cheonbulbojeon Hall
23			Jungnimsas ^b	35.0404	126.8836	Wooden Buildings
24		Yeosu	Heongguksas ^a	34.8211	127.7003	Daeungjeon Hall

The airborne fungi from WCHs were sequenced from the ITS1(5'-TCCGTAGGTGAACCTGCGG-3' as the forward primer) and ITS4 (5'-TCCTCCGCTTATTGATATGC-3' as the reverse primer) region (600~800 bp), and the analysis was performed by Solgent (Daejeon, Korea). PCR and sequencer were performed using a Veriti™ 96-Well Thermal Cycler (Applied Biosystems, Foster City, CA, USA) and an ABI 3730XL DNA Analyzer (Applied Biosystems, USA), respectively. The gene sequences of the analyzed ITS1/4 region were compared for homology with previously reported ITS1/4 region sequences of microorganisms using the Blast search program of the National Center for Biotechnology Information (NCBI, USA) (<http://www.ncbi.nlm.nih.gov/pubmed/>) to identify the strain. The results of the internal airborne fungal surveys of 19 WCHs from the NRICH survey were only used to compare fungal status on an annual basis, as the exact timing of the collections could not be determined.

Evaluation of Airborne Fungal Growth Characteristics Inside WCHs

Most fungi inside buildings germinate from 0 to 40 °C, with 22 to 35 °C being the optimal temperature (Barghman and Arens 1996). Nine fungi (five fungi and four wood-decaying fungi) were selected from five WCHs in July and October, when the internal temperature was maintained above 20 °C; their growth characteristics were determined under different relative humidity conditions (Table 2). Each of the nine fungi was inoculated on PDA (Potato dextrose agar, Difco, US) solid medium and incubated at room temperature for 2 weeks. A sample of the fungi was placed in a 1.5 mL vial tube and used for the experiment within 1 week. The medium for characterizing the growth of airborne fungi was selected as Hanji and wood, which are the main materials of WCHs. Hanji was weighed at 30 g/m² and measured 50 mm x 20 mm. Pine wood (*Pinus densiflora*), which was used for more than 90% of WCHs in Korea according to NRICH (2015, 2016), was cut into pieces with dimensions of 50 mm (C) x 20 mm (R) x 5 mm (T). The cut hanji and wood were autoclaved at 121 °C for 15 min.

Table 2. Airborne Fungi Identified inside five WCHs during Summer Field Surveys

No.	Fungal identity	Species	Microbe Types
1	<i>Alternaria sp.</i>	<i>Alternaria alternata</i>	Fungi
2	<i>Aspergillus sp.</i>	<i>Aspergillus niger</i>	Fungi
3	<i>Aspergillus sp.</i>	<i>Aspergillus fumigatus</i>	Fungi
4	<i>Fusarium sp.</i>	<i>Fusarium incarnatum</i>	Fungi
5	<i>Penicillium sp.</i>	<i>Penicillium oxalicum</i>	Fungi
6	<i>Coprinellus sp.</i>	<i>Coprinellus radians</i>	Wood-decay fungi
7	<i>Irpex sp.</i>	<i>Irpex lacteus</i>	Wood-decay fungi
8	<i>Trametes sp.</i>	<i>Trametes hirsuta</i>	Wood-decay fungi
9	<i>Trametes sp.</i>	<i>Trametes versicolor</i>	Wood-decay fungi

The experimental design for the evaluation of growth characteristics was as follows. A plastic mesh was placed inside a 90 mm diameter Petri dish for inoculation and growth of the fungus. Sterilized agar and wood were placed on top of the inserted mesh and 200 µL of each of the nine selected strains were inoculated on the surface and covered with a lid (Fig 2A). Inside the plastic sealed box [(230 mm x 170 mm x 100 mm (h))], 100 ml of saturated salt aqueous solution and distilled water per condition were added to create four relative humidity conditions (NaBr: 60-65%, NaCl: 70-75%, KCl: 80-85%, D.W.:

95% or more). A breathable netting for the Petri dish was set up and the Petri dish was placed on it. A real-time temperature and humidity sensor was placed inside the plastic-sealed box to measure the temperature, and relative humidity changes every 10 min during the experiment and to sound an alarm when a change is detected. The box was covered with a lid and sealed with parafilm to ensure that the set relative humidity of the indoor space remained constant (Fig. 3B). The temperature condition was set at 23 °C, and the condition-specific RH was maintained with a constant temperature and humidity hygostat. Each evaluation was performed in triplicate with three replicates of nine strains. The growth rate of the fungi was assessed using the Fungi index (Viitanen and Salonvaara 2001), starting from the third stage when the growth of the fungus was visually observed, and the growth was evaluated weekly for 12 weeks (Table 3).

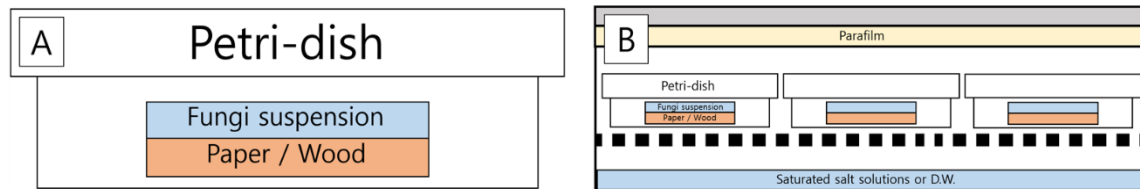


Fig. 2. Experimental design to observe airborne fungal growth in summer by relative humidity and material conditions

Table 3. Fungi Index Values and their Description for Fungi Growth

Index	Descriptive Meaning
0	No growth
1	Growth starts, some growth detected only with microscopy
2	Moderate growth detected with microscopy
3	Some growth detected visually, new spore formation
4	Visually detected coverage more than 10%
5	Visually detected coverage more than 50%
6	Visually detected coverage more than 100%

RESULTS AND DISCUSSION

Investigate WCHs Internal Temperature and Humidity

The average internal temperature and humidity of the 24 WCHs were 24.7 ± 0.9 °C and $76.8 \pm 4.2\%$ in summer (June-August) and 16.2 ± 1.3 °C and $70.8 \pm 5.6\%$ in fall (September-November), with the fall season being about 8 °C and 6% lower than the summer season on average (Fig. 3A). This is because the Korean Peninsula has a monsoon climate with four distinct seasons and 70% of the average annual precipitation is concentrated from June to September (Lee *et al.* 2011). This weather pattern is consistent with the results that the five WCHs in the field study are affected by external climatological changes that drop sharply in October, resulting in internal average temperatures below 16 °C and relative humidity below 75% (Im and Han, 2023). Also contributing to these seasonal temperature and humidity variations, Han (2003) mentioned that the geographical setting of the WCHs, where they are located in narrow valleys or on flat areas cut into the slopes of mountainsides, also plays a role.

The average internal temperature and humidity of the 24 WCHs during the summer months were within the zone of growth and germination of xerophilic fungi identified by Lee (2016) (Fig. 3B), and 14 of the WCHs met the conditions for the growth of surface contaminating fungi proposed by Viitanen and Salonvaara (2001), with an internal average temperature of 20 °C and relative humidity above 75% (Fig. 3b). Regarding the temperature and humidity environment inside the WCHs during summer, Kim *et al.* (2015) also predicted that the relative humidity inside the WCHs connected to the outside would rise above 75% regardless of whether the WCHs were open or closed during summer, which could lead to wood deterioration by fungi. In addition, most of the 24 WCHs had internal relative humidity in the 85% to 95% range for about two to three weeks in August. This is believed to be due to the increase in the number of precipitation days due to heavy rains caused by the rainy season and typhoons (Shim and Jeong 2002; Kim *et al.* 2019; Korea Meteorological Administration 2023, 2024). However, several previous studies have reported that germination and growth of fungi require a temperature in the range of 21 to 32 °C and a relative humidity of 75% or higher for a certain period, and that short-term intermittent high relative humidity does not have a significant effect (Carll and Highley 1999; Viitanen and Salonvaara 2001; Kim *et al.* 2015).

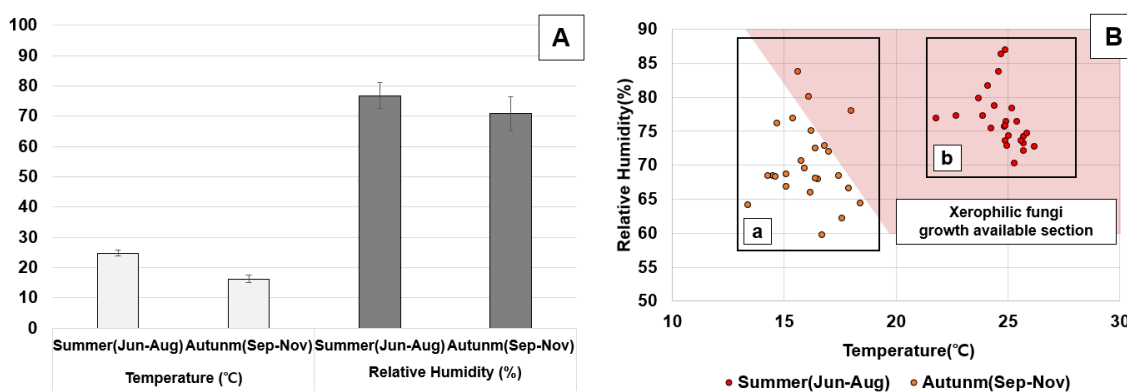


Fig. 3. Average temperature and relative humidity inside 24 WCHs in summer and Autumn (a: Internal temperature and relative humidity of 24 WCHs in autumn, b: Internal temperature and relative humidity of 24 WCHs in summer)

For the four months from June to September, the five WCHs in the field study had a common pattern of internal relative humidity by the time of day, with a decrease from 8-9 am to 3-5 pm and a rise again from 8-9 am the next day (Fig. 4). This is believed to be due to the continuous influx of fresh air and the operation of internal air conditioning units after 9:00 am due to religious ceremonies, visitors, *etc.* The average relative humidity by month was examined from June to September, when the internal temperature of WCHs starts to rise above 20 °C, and found that in June and July, the internal relative humidity of the five WCHs surveyed in the field did not rise above 75% in all time zones or rose within 6 hours during the day and then fell again (Fig. 4). In August, the average RH of 75% to 83% was maintained at all times of the day, and in September, three WCHs, except for the trainer and the tenderizer, maintained an internal RH of 75% at all times of the day following August (Fig. 4). Therefore, it was confirmed that the internal RH of the WCHs was maintained at an average temperature of 20 °C and a RH of 75% or higher, which are the conditions for the growth of fungi, for 4 to 8 weeks in August and September of the year, depending on the location and internal conditions. In addition, it is considered that a

growth evaluation over time should be conducted over the maximum period (8 weeks) when the relative humidity is above 75% on the Hanji and wood surfaces to determine the damage of surface contaminating fungi to the airborne fungi inside the WCHs.

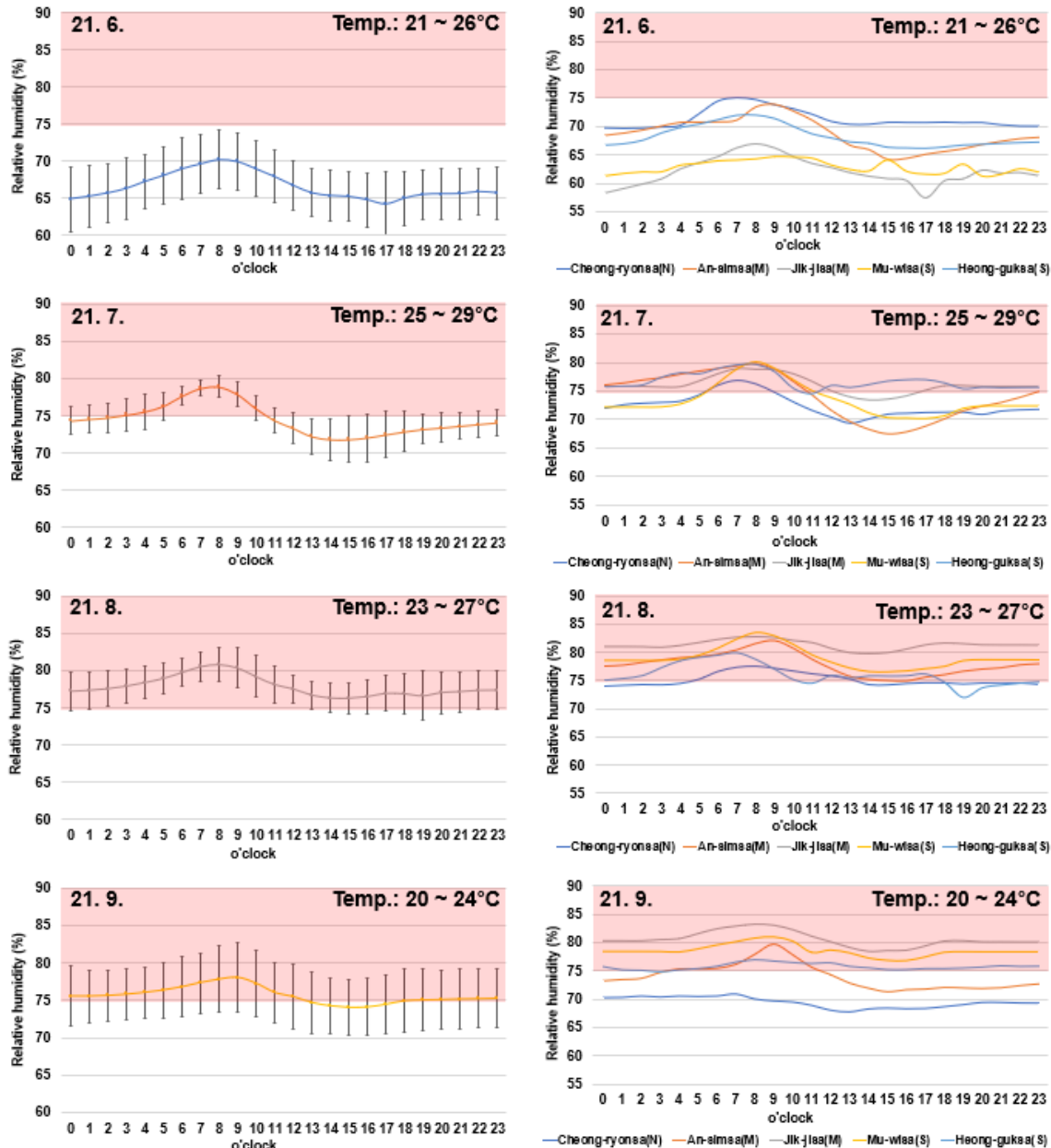


Fig. 4. Internal average temperature and relative humidity changes by month and time of day for the five WCHs surveyed in the field (left: mean and deviation for the five temples; right: each of the five temples individually)

Status of Airborne Fungi inside WCHs

Fungi such as *Alternaria* sp., *Aspergillus* sp., *Cladosporium* sp., *Epicoccum* sp., *Fusarium* sp., *Penicillium* sp., and *Trichoderma* sp. were identified inside the 5 WCHs of the field study that were collected four times per season and 19 WCHs that were collected once per year (Figs. 5, 6). These results are consistent with Kim *et al.* (2004), who mentioned that the types of fungi that can invade and grow on wood surfaces include *Alternaria* sp. (black), *Aspergillus* sp. (yellow, green, black), *Trichoderma* sp. (green),

Penicillium sp. (light blue-green), *Cladosporium* sp. (blue-green, black-green), and *Fusarium* sp. (red, purple). In addition, the results of seasonal sampling inside the five WCHs confirmed fungi was not affected by the season and was found to be present throughout the year (Figs. 5, 7). Wood decay fungi such as *Coprinellus* sp., *Irpex* sp., and *Trametes* sp. were suspended inside the WCHs during summer and fall (Fig 5, 7). The fungi identified as floating inside the 24 WCHs in the field survey and previous studies are the same as the fungi and wood decay fungi that have been identified in many previous diversity surveys of airborne fungi in WCHs (Hong *et al.* 2009, 2011, 2018, 2019; Jo *et al.* 2009; Kim *et al.* 2017; Kim *et al.* 2022). Therefore, it is concluded that if there is damage caused by fungi and wood decay fungi inside WCHs, it is likely to be fungi from the 11 genera identified in Figs. 5 and 6.

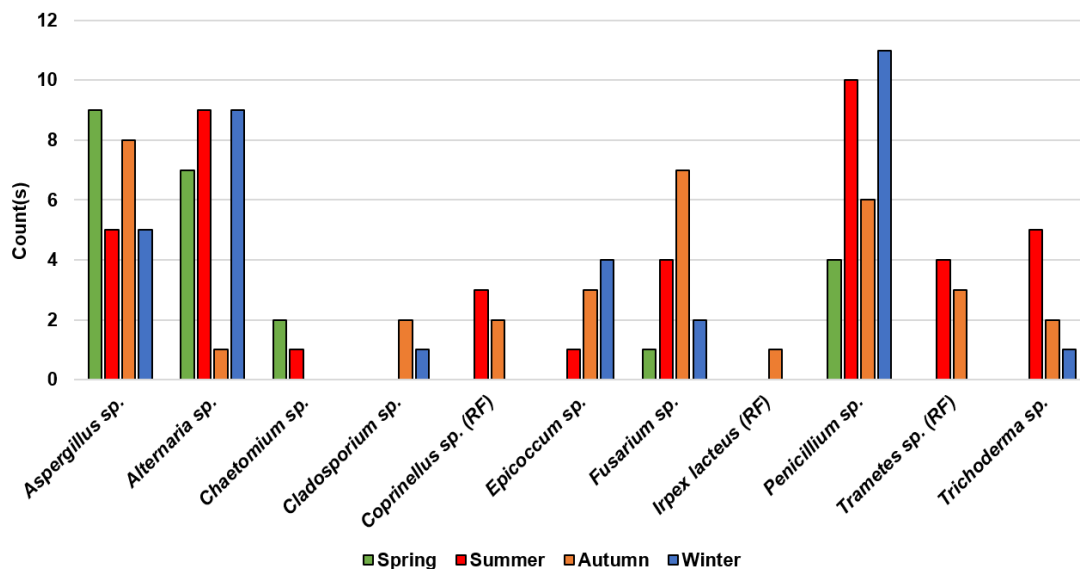


Fig. 5. Seasonal suspended fungi counts inside five WCHs surveyed in the field (RF: Wood-decay fungi)

Based on the comparison of the field survey and previous studies, eight genera of fungi were identified, and most of them are aerobic, meaning that spores from the air attach to the surface of the material, and then gradually grow and discolor the surface of the material when the environmental conditions required for germination are met (Kosuke 2012). In addition, Viitanen and Salonvaara (2001) reported that *Alternaria* sp., *Aspergillus* sp., *Cladosporium* sp., *Penicillium* sp., and *Penicillium* sp. are typical fungi found in building materials. Damage caused by surface contaminants such as stains and discoloration was confirmed in several large Buddhist painting made of hanji stored in boxes inside WCHs (NRICH 2016; 2017; 2018; 2019; 2020; 2021; 2022; 2023). In this study, *Alternaria* sp., *Aspergillus* sp., and *Penicillium* sp. were present year-round in the interiors of the five WCHs surveyed, regardless of the season (Fig. 5).

Alternaria sp., *Aspergillus* sp., *Cladosporium* sp., and *Penicillium* sp. were also identified as the most frequently counted fungal genera inside the 24 WCHs, except *Fusarium* sp. (Fig. 6). Among them, *Alternaria* sp. is a fungus that grows on plants and plant debris at a high-water activity (a_w) (Magan and Lacey 1984; Andersen and Frisvad 2002) and has been reported to be isolated from the surface of church murals (Pepe *et al.* 2010). *Aspergillus* sp. and *Penicillium* sp. are typical xerophilic fungi that grow in low humidity conditions and are surface contaminants that grow in a wide range of habitats (Magan and Lacey 1984; Andersen and Frisvad 2002; Seo 2012). Most xerophilic fungi can survive and grow in dry conditions without dying from changes in the external environment by accumulating internal solutes that allow the cells to maintain a high osmotic pressure (Pettersson and Leong 2011; Irwin 2020). These two species produce a variety of enzymes and organic acids, as well as mycotoxins, which can pose a threat to visitors and restoration and conservationists working with fungal materials (Moon *et al.* 2009; Frisvad 2015; Skóra *et al.* 2015; Savković *et al.* 2019). *Alternaria* sp., *Aspergillus* sp., *Penicillium* sp., and *Cladosporium* sp. are highly dispersive fungi with dry spores that send countless spores into the air at the slightest impact and have been found suspended in wood and various organic artifacts and spaces (Choi *et al.* 1998; NRICH 2016, 2019, 2021, 2023; Hong *et al.* 2009, 2011, 2018, 2019; Zalar *et al.* 2023).

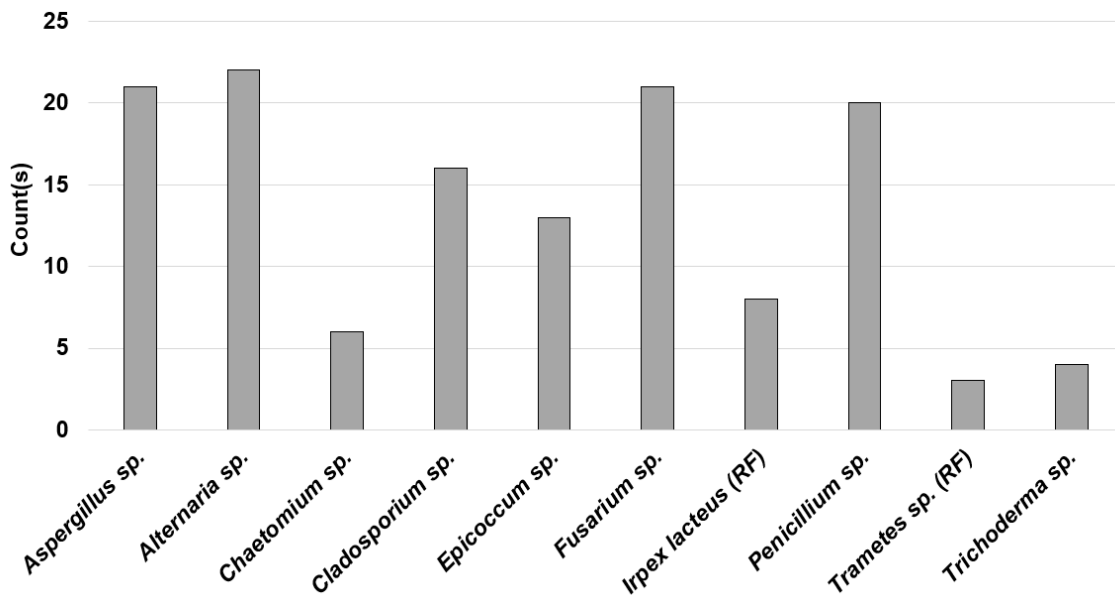


Fig. 6. Year-round suspended fungi count inside 24 WCHs (RF: Wood-decay fungi)

Epicoccum sp., *Fusarium* sp., and *Trichoderma* sp. were found to float inside WCHs during summer and fall seasons, except for some seasons in spring and winter, although their capture counts were relatively lower than the aforementioned four fungi (Figs. 5, 6). In response to this, Viegas *et al.* (2015) noted that the spores of *Fusarium* sp. and *Trichoderma* sp. are agglutinated in viscous substances and are relatively heavy, so diffusion by air is relatively more difficult than in the form of dry spores. They also noted that spores in a moistened state, rather than in dry spore form, generally rely on water or other vectors such as insects for dispersal (Abbott 2002; Trovão *et al.* 2013). Because of these dispersal characteristics, *Epicoccum* sp., *Fusarium* sp., and *Trichoderma* sp. are predominantly found on the soil floor of WCHs located in forested areas, which may explain the increased counts of *Fusarium* sp. and *Trichoderma* sp. in the summer and fall

months, when visitors visit WCHs the most during the year, due to increased external soil introduction by visitors. Lavin *et al.* (2015) reported that *Fusarium* sp. isolated from historic properties can pose a risk to historic properties by producing reddish-brown spots and biofilms on surfaces and decreasing pH, resulting in aesthetic and structural damage. In addition, *Trichoderma* sp. a blue fungi that grows on soil, dead leaves, stumps, and rotting wood, produces a variety of enzymes, including cellulase and hemicellulase, and has been identified in the air in the storage of paper and fibrous cultural heritage (Min and Ahn 1981; Verma *et al.* 2007; Hidangmayum and Dwivedi 2018). *Chaetomium* sp. was captured in some WCHs during the spring and summer months in the five WCHs where field surveys were conducted, but in combination with the results from the 24 WCHs, it does not appear to be a dominant species suspended inside. As a result, the eight fungi identified in this study live on extractive components, dirt, dust, and sugars on the surface of wood, but do not utilize cellulose and lignin, the components of wood, as a nutrient source, so they do not cause intense damage, but since they float in the indoor space throughout the year, if an environment suitable for their growth is created, they can cause damage such as discoloration, stains, and decomposition of adhesives on the surface of the material, which can damage the aesthetic and structural value of cultural properties (Ciferri 1999; Kim *et al.* 2004; Sterflinger 2010; Yoon *et al.* 2012; Jeong 2014; Zalar *et al.* 2023).

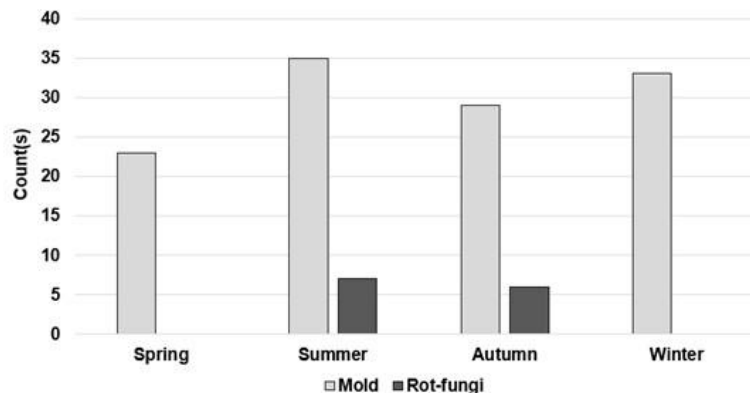


Fig. 7. Number of fungi and wood-decay fungi captured by season inside five WCHs surveyed in the field

Irpex lacteus was the most frequently counted among the 24 WCHs with 8 counts (Fig. 6), while *Coprinellus* sp. and *Trametes* sp. were counted less than 5 times (Figs. 5, 6). In addition, wood decay fungi were found to float inside the WCHs mainly in summer and autumn in the five WCHs surveyed in the field (Figs. 5, 7). This indicates that wood decay fungi can grow in wood moisture content of at least 30%. Because it can germinate and grow in high-humidity conditions with relative humidity above 95% (Morris 1998; Viitanen and Salonvaara 2001; Jo *et al.* 2012), it is likely that it germinated and grew in the high-humidity environment outside during summer and fall and entered the WCHs by way of visitors and outside air. Choi *et al.* (1998) noted that spaces with frequent visitor access have a higher distribution of airborne fungi, suggesting that the continuous influx of air from human traffic affects the introduction and distribution of fungi in space. Wood decay fungi are classified into brown decay fungi, white decay fungi, and soft decay fungi based on the physical and chemical changes that occur in wood and the resulting changes in the color and appearance of the wood (Carll and Highley 1999). The interior of a building, where the relative humidity is consistently high, creates conditions suitable for

the germination and growth of decay fungi, which rapidly degrade the color, shape, and strength of wood (Viitanen and Salonvaara 2001).

However, several fungi, including *Alternaria* sp., *Aspergillus* sp., *Cladosporium* sp., and *Penicillium* sp. are dominant species among fungi to which humans are routinely exposed, accounting for more than 95% of all fungi collected in WCHs as well as in various multi-use facilities such as hospitals, early childhood facilities, elderly care facilities, and maternity homes (Park *et al.* 2006). In addition, there is no significant difference in the species diversity between indoor and outdoor in general because most of the internally suspended fungi, including wood decay fungi, are introduced into the interior of buildings by air movement due to ventilation (Park *et al.* 2006; Shin 2014). Therefore, it is necessary to identify the growth patterns of fungi and wood decay fungi that are mainly suspended inside WCHs from June to September (summer and early fall), when the relative humidity is the highest during the year, to check the possibility of damage to organic cultural properties.

Evaluation of Airborne Fungal Growth Characteristics inside WCHs

During the 12 weeks, none of the nine fungal species were observed to grow above Fungi index stage 3 on wood and Hanji surfaces at any of the four relative humidity conditions (60-65%, 70-75%, 80-85%, and 95% or higher) except for 95% or higher. These results are consistent with Viitanen and Salonvaara's (2001) findings that wood decay fungi germinate and grow at relative humidity above 95%. The results also support the results of the experiments with fungi, as shown in Table 4, where the minimum water activity (a_w) values for germination and growth of each species were all above 0.85 (Ayerst 1969; Hocking and Pitt 1979; Northolt and Bullerman 1982; Magan and Lacey 1984). Water activity refers to the moisture content of a substance and is the most important factor for microbial growth along with temperature and pH, with fungi starting to develop depending on the moisture content of an object's surface (McMeekin and Ross 1996; Lavin *et al.* 2015). In this regard, Viitanen and Salonvaara (2001) stated that the moisture content of the surface is the most important factor in the development of fungi on wood, rather than the moisture content inside the wood, and it has a mutual influence with the relative humidity of the indoor space that affects the surface. Therefore, given the characteristics of surface fungi that germinates and grows in a thin layer on the surface of the wood that is directly exposed to the relative humidity of the interior space, it is not unreasonable to equate the relative humidity of the indoor space for a long period with the minimum moisture activity. As shown in Figs. 3 and 4, the maximum average relative humidity in the 24 WCHs across the country was below 90% for most of the year, with very short periods above 95%. In addition, for these short periods of intermittent high humidity conditions, Viitanen and Bjurman (1995) reported that *Aspergillus* sp., *Cladosporium* sp., *Penicillium* sp., and *Trichoderma* sp. had very slow growth rates when subjected to rapid changes in humidity between 75% and 97% relative humidity or when drying periods were longer than 1 week, which they attributed to the slow increase in moisture on the wood surface. Kim *et al.* (2015) noted that wood deterioration caused by fungi and wood decay fungi is unlikely to occur in the interior space of WCHs throughout the year because the period of time after the relative humidity rises to 95% or higher is very short, less than 24 h. Therefore, in all periods of the year, including summer, the indoor space of the WCHs is not considered to be a suitable environment for the germination and growth of fungi and wood decay fungi for a long enough period to cause damage to the wood and Hanji surface.

Table 4. Minimum Water Activity (a_w) Reported for Growth of Fungi

a_w	Species with this a_w as minimum for growth	Citation
0.76-0.84	<i>Aspergillus nidulans</i> , <i>Aspergillus versicolor</i> , <i>Penicillium citrinum</i>	Ayerst (1969), Galloway(1935), Griffin (1963), Magan and Lacey (1984), Mannaa and Kim (2017), Northolt and Bullerman (1982), Pitt (2002), Pettersson and Leong (2011)
0.85-0.90	<i>Alternaria tenuissima</i> , <i>Alternaria alternata</i> , <i>Aspergillus fumigatus</i> , <i>Aspergillus niger</i> , <i>Cladosporium cladosporioides</i> , <i>Epicoccum nigrum</i> , <i>Fusarium solani</i> , <i>Penicillium glabrum</i> , <i>Penicillium oxalicum</i>	Griffin (1963), Hocking and Pitt (1979), Kozakiewicz and Smith (1994), Magan and Lacey(1984), Sautour (2001), Sempere and Santamarina (2010), Sinigaglia (1998)
0.91-0.95	<i>Aspergillus fumigatus</i> , <i>Fusarium graminearum</i> , <i>Fusarium incarnatum</i> , <i>Penicillium oxalicum</i> , <i>Trichoderma harzianum</i>	Griffin (1963), Lahouar <i>et al.</i> (2017), Magan and Lacey (1984), Mannaa and Kim (2017), Santamarina and Roselló (2006)

At 95% relative humidity, *Alternaria alternata*, *Aspergillus niger*, and *Aspergillus fumigatus* took 3, 3, and 5 weeks from the beginning of the experiment to reach fungi index stage 3 on the wood specimens, but all three species grew to stage 3 on the Hanji at the end of 2 weeks (Figs. 8, 9). *Aspergillus niger* spread to 10% of the total area of both wood specimens and Hanji within one week of progressing to stage 3 and remained at fungi index stage 4 until week 12. *Alternaria alternata* and *Aspergillus fumigatus* grew to Fungi index stage 3 on wood specimens at weeks 3 and 5 and did not change significantly until week 12. However, on Hanji, *Alternaria alternata* was found to spread over the entire surface very quickly, reaching fungi index 5 at 3 weeks and 6 at 4 weeks. Viitanen and Salonvaara (2001) noted that if the material is wet, the microclimate humidity on the surface can be high for a long time, which can promote the growth of fungi on the surface. *Aspergillus fumigatus* also grew from 5 weeks to stage 4 and was found to grow better on agar than on wood specimens. *Penicillium oxalicum* and *Fusarium incarnatum* grew to Fungi index stage 3 at 10 and 11 weeks on wood specimens and 8 and 10 weeks on agarwood, respectively. The results of the growth rate of the surface contaminants showed that *Alternaria alternata* and *Aspergillus niger* can germinate and grow on the wood surface and cause surface contamination in the shortest period among the surface contaminants. In addition, *Penicillium oxalicum* and *Fusarium incarnatum* can germinate and grow under high humidity conditions of 95% or more for at least 2 months (8 weeks) compared to *Alternaria alternata*, *Aspergillus niger*, and *Aspergillus fumigatus*. Based on this, *Penicillium oxalicum* and *Fusarium incarnatum* can germinate and grow within 5 weeks and are relatively unfavorable in competition with *Alternaria alternata* and *Aspergillus niger*, which are present in most spaces, so the risk of damage is considered to be relatively low. In addition, differences in the growth of fungi were identified based on the material difference between wood specimens and Hanji. Viitanen and Salonvaara (2001) explained that the relative humidity required for the germination and growth of fungi varies depending on the material, and the most important requirement is the surface moisture content rather than the moisture content of the material. Therefore, if high humidity conditions of 95% or more are created for more than 3 weeks for wooden surfaces and

more than 2 weeks for organic artifacts such as paintings and electrolyzed water due to rainy season, typhoons, and water leakage, damage caused by the growth of surface contaminating fungi such as *Alternaria alternata* and *Aspergillus niger* is expected to appear.

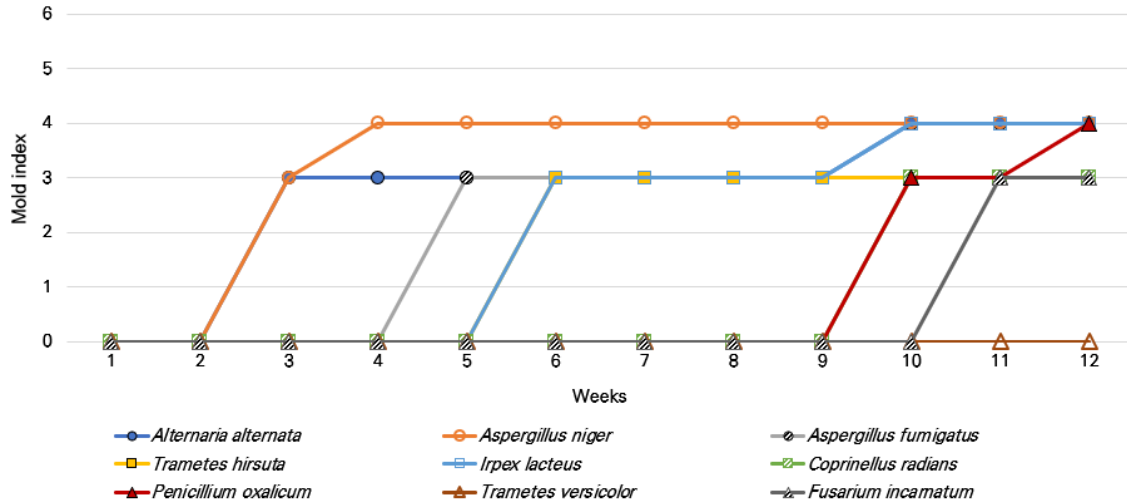


Fig. 8. Fungi index of major airborne fungi in summer at 95% relative humidity (Wood)

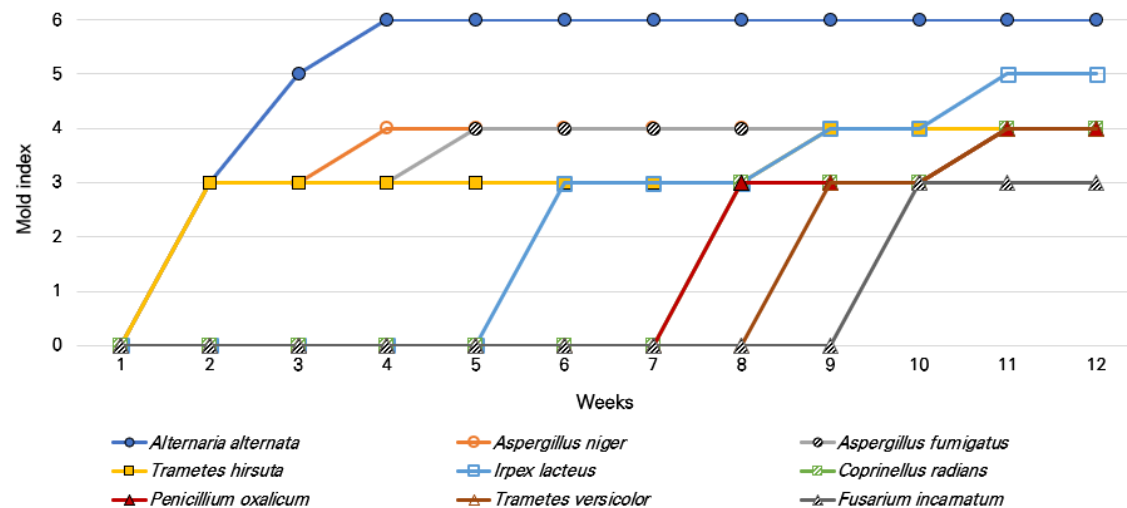


Fig. 9. Fungi index of major airborne fungi in summer at 95% relative humidity (Hanji)

The growth behavior of wood decay fungi at 95% relative humidity for 12 weeks is as follows. *Irpex lacteus* took 6 weeks to reach Fungi index stage 3 on both wood and Hanji. However, *Trametes hirsuta* was not significantly different from *Irpex lacteus* with 6 weeks on wood to stage 3, but on Hanji, it was found to cover the surface to stage 3 two weeks after the start of the experiment and stage 4 at 9 weeks (Figs. 8, 9). *Coprinellus radians* took 10 weeks to reach stage 3 on wood, 8 weeks to reach stage 3 on Hanji, and 11 weeks for stage 4. *Trametes versicolor* showed no growth to stage 3 in 12 weeks on wood. On Hanji, it grew to stage 3 in 9 weeks and stage 4 in 11 weeks. Most wood decay fungi grew more rapidly on Hanji than on wood. This is likely due to the smaller thickness

of the paper, which allowed it to reach a water activity of 0.95 or higher, which is suitable for growth at 95% relative humidity, more quickly. Therefore, if the relative humidity above 95% is maintained for more than 2 weeks on wood and Hanji due to partial leakage in the internal space or micro-high humidity environment inside the storage case, the damage will be accelerated, first by *Trametes hirsuta* on Hanji, and then by the growth of various saprophytic fungi after 6 weeks.

As confirmed by this study, WCHs are inevitably exposed to a variety of suspended fungi from the outside world due to regular viewing and religious rituals. However, so far, species diversity surveys have only reported on the presence of fungi in the interior spaces, not on whether the interior spaces of WCHs create a favorable environment for fungal damage. Therefore, in the future, it is essential that WCHs be surveyed for fungal diversity in their interior spaces, as well as the range and duration of temperature and relative humidity increases throughout the year. In addition, WCHs' wooden timbers and internal collections of artifacts are exposed to various biohazardous factors due to the external conditions of being located mostly in forests and the internal environmental changes that occur through various viewing and religious rituals, so continuous management is needed to prevent damage.

CONCLUSIONS

1. The same surface contaminants such as *Alternaria* sp., *Aspergillus* sp., and *Penicillium* sp. were present in the interior spaces of WCHs throughout the year as in the interior of multi-use facilities frequented by humans. Wood decay fungi, which require a relative humidity of more than 95% to germinate and grow, had spores that germinated and grew outside only during the summer and fall seasons, and floated in the interior spaces due to visitors and fresh air inflow.
2. Depending on the location and internal environment, the internal environmental conditions for the growth of fungi, an average temperature of 20 °C and relative humidity of 75% or more, were continuously created for at least 4 weeks and up to 8 weeks of the year.
3. The maximum average relative humidity of the 24 WCHs nationwide was below 90%, which is the level at which most wood decay fungi cannot germinate and grow, and the period above 95% was very short. Therefore, the internal space of WCHs is unlikely to be damaged by surface contaminants and wood decay fungi in all periods of the year, including summer because the environment suitable for germination and growth is not created for a long period to cause damage to wood and paper surfaces.
4. All surface contaminating fungi and wood decay fungi germinated and grew on wood and Hanji surfaces at 23 °C temperature and 95% relative humidity or higher. In particular, *Alternaria alternata*, *Aspergillus niger*, and *Aspergillus fumigatus* were confirmed to grow on the surface of Hanji. The influx of moisture caused by various factors into cultural properties consisting of wood and hanji throughout the year can cause sufficient damage if conditions are created for the germination and growth of fungi and wood-decay fungi.

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