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Quantification of mold contamination in multi-level buildings using the Environmental Relative Moldiness Index

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ABSTRACT

The goal of this study was to evaluate the possible use of the Environmental Relative Moldiness Index (ERMI) to quantify mold contamination in multi-level, office buildings. Settled-dust samples were collected in multi-level, office buildings and the ERMI value for each sample determined. In the first study, a comparison was made between two identical four-story buildings. There were health complaints in one building but none in the other building. In the second study, mold contamination was evaluated on levels 6–19 of an office building with a history of water problems and health complaints. In the first study, the average ERMI value in the building with health complaints was 5.33 which was significantly greater than the average ERMI value, 0.55, in the non-complaint building. In the second study, the average ERMI values ranged from a low of –0.58 on level 8 to a high of 5.66 on level 17, one of the top five ranked levels for medical symptoms or medication use. The mold populations of ten (six Group 1 and four Group 2) of the 36-ERMI molds were in significantly greater concentrations in the higher compared to lower ERMI environments. The ERMI metric may be useful in the quantification of water-damage and mold growth in multi-level buildings.

KEYWORDS

Dust; ERMI; mold; office building; water damage

Introduction

Building managers are sometimes called upon to investigate health complaints in the workplace. One of the possibilities to consider in such an investigation is dampness and mold growth. Exposure to damp, moldy buildings has previously been linked to respiratory health problems.^[1,2] In addition, dampness in buildings has also been linked to other health effects, like tiredness and headaches.^[3] Therefore, it would be of value for building managers to have a standardized metric for quantifying mold contamination.

In order to quantify mold contamination in homes, the U.S. EPA, in conjunction with the U.S. Department of Housing and Urban Development, developed the Environmental Relative Moldiness Index (ERMI) scale.^[4] The ERMI value is based on the mold specific quantitative PCR (MSQPCR) analysis of 36 indicator-molds: 26 Group 1 molds which are associated with water-damaged environments and 10 Group 2 molds which are commonly found indoors, independent of water-damage.^[4] The ERMI scale ranges from about –10 to about 30, i.e., lowest to highest mold contamination.

The ERMI metric has primarily been used to quantify water-damage and mold contamination in homes for studies of occupant asthma. In six epidemiological studies of asthma, higher ERMI values were associated with asthma development and/or exacerbation.^[5] For example, infants exposed to homes with ERMI values greater than 5.2 nearly doubled their risk of developing asthma by age seven.^[6] The goal of this study was to evaluate the potential use of the ERMI metric to quantify mold contamination in multi-level, office buildings.

Materials and methods

Building descriptions

In the first study, two identical four-level office buildings, within 100 m of each other in the southeastern U.S., were the subjects of study. The buildings were masonry and concrete structures with flat roofs. Each level of each building was served by a separate heating, ventilation and air-conditioning system and the levels were each approximately 3,000 m²; subdivided into multiple, separate offices. Five years after building occupancy, some

of the employees on two levels, 2 and 4, of one building complained of respiratory problems when at work. There were no health complaints in the other building. Seven samples were obtained from each of levels 2 and 4 in the complaint building and from the same levels in the non-complaint building for a total of 28 samples. The locations for sampling (Figure 1) were selected to represent the entire level.

In the second study, a previously sampled office-building in the northeastern U.S. was the subject.^[7,8] The building had a long history of water problems since construction in 1985. The major sources of water intrusion were previously traced to leaks through exterior walls, terraces, and windows on levels 17, 18, 19 and from the roof.^[9] Earlier publications about this building showed that the epidemiologically defined respiratory cases and post-occupancy asthma were significantly associated with the fungal/mold populations in floor-dust samples.^[8,10] Therefore, for this study, three frozen (-80°C) dust-samples from each of the levels 6–19 (there was no level 13) were randomly selected for ERMI analysis from the 338 dust samples obtained in 2002.

ERMI analysis of dust

In each study, the analyst was blinded to the source of the dust samples or location of origin in the buildings. Each settled-dust sample was sieved through 300 μm pore mesh and 5.0 \pm 0.1 mg of dust from each sample was analyzed. The dust sample was added to an extraction tube, along with 200 μL of the DNA-EZ kit extraction fluid (GeneRite, Inc., Monmouth Junction, NJ) and then spiked with 1×10^6 conidia of *Geotrichum candidum* as an external reference.^[11] Each extraction tube was shaken in a bead beater (Biospec Products, Bartlesville, OK) at 5,000 rpm for one min and the DNA purified using the DNA-EZ kit (GeneRite, Inc.).

Methods and assays have been reported previously for performing the MSQPCR analyses.^[11] The standard reaction assay contained 12.5 μL of “Universal Master Mix” (Applied Biosystems Inc., Foster City, CA), 1 μL of a mixture of forward and reverse primers at 25 μM each, 2.5 μL of a 400 nM TaqMan probe (Applied Biosystems Inc.), 2.5 μL of 2 mg/mL fraction V bovine serum albumin (Sigma Chemical, St. Louis, MO) and 2.5 μL of DNA-free water (Cepheid, Sunnyvale, CA). To this mix was added 5 μL of the DNA extract from the sample. All primer and probe sequences used in the assays, as well as known species comprising the assay groups, are at the website: <https://www.google.com/patents/US6387652>. Primers and probes were synthesized commercially (Applied Biosystems, Inc.).

Statistical analyses

In Studies 1 and 2, the statistical differences between the average ERMI values and the average sums of the logs of the Group 1 and Group 2 molds were investigated using the Student's T-test. In the Study 2, multiple comparisons of the average ERMI values on each level in the water-damaged building were performed using Dunnett's method to adjust for multiple testing. Also, in each study, the concentration differences for each of the 36-ERMI molds in the dust samples from high and low ERMI environments were evaluated using the Wilcoxon rank-sum test and adjusting for multiple testing using the Holm–Bonferroni test. Analyses were performed in SAS version 9.3 (SAS Institute, Cary NC) and R version 2.14 (R Foundation for Statistical Computing, Vienna, Austria).

Results

In the Study 1, the average ERMI value for the samples ($n = 14$) in the building with health complaints was 5.33 which was significantly ($p = 0.006$) greater than the average ERMI value, 0.55, for the samples ($n = 14$) from the non-complaint building (Table 1). In addition to higher ERMI values, both the average sum of the logs of the Group 1 and Group 2 molds were also significantly ($p < 0.006$) greater in the complaint building (Table 1).

In Study 2, the average ERMI values ranged from a low of -0.58 on level 8 to a high of 5.66 on level 17 (Figure 2). Multiple comparison using Dunnett's method showed that the average ERMI value on level 17 was significantly greater than the average ERMI value on all other levels except for level 9 (ERMI 3.20) (Figure 2). Therefore, the sum of the logs of the Group 1 and Group 2 molds for dust samples ($n = 6$) from levels 9 and 17 were combined and compared to the average sum of the logs of the Group 1 and Group 2 molds for the samples ($n = 33$) from all other levels to further examine the two different groups of mold between those floors (Table 1). In addition to higher ERMI values on levels 9 and 17 than that of other levels ($p < 0.001$), the sum of the logs of the Group 1 molds were also significantly greater ($p < 0.001$) on levels 9 plus 17 compared to the other levels whereas the sum of the logs of the Group 2 molds were not different between two groups of floors (Table 1).

The concentrations of each of the 36-ERMI mold species were compared in each of the studies. In the first study, there were five Group 1 and three Group 2 molds in significantly ($p < 0.001$) greater concentrations in the complaint building compared to the non-complaint building (Table 2). In the second study, there were three Group 1 molds and one Group 2

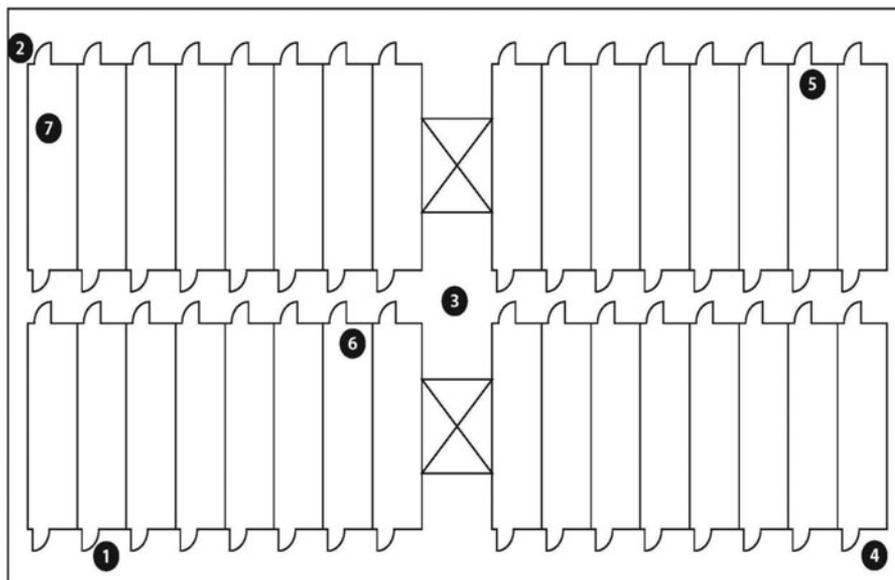


Figure 1. General layout of each level of each building and the seven sampled locations on each level in Study 1.

Table 1. Comparison of the mean and standard deviation (SD) of the Environmental Relative Moldiness Index (ERMI) values, sum of the logs of Group 1, and sum of the logs Group 2 values: Study 1- comparison of a building with health complaints vs. an identical building with no health complaints; Study 2- comparison of water-damaged building, levels 9 plus 17 vs. all other levels (there was no level 13 in this building).

Study 1	Complaint building		No-complaint building		T-test p-value
	Levels- 2 + 4 mean	SD	Levels- 2 + 4 mean	SD	
Sum of the logs Group 1	23.52	8.59	12.40	4.59	< 0.001
Sum of the logs Group 2	18.19	4.43	11.55	5.34	0.002
ERMI Study 2	5.33	4.72	0.55	2.87	0.006
	Water-damaged Levels - 9 + 17 mean	SD	Water-damaged Levels - others mean	SD	T-test p-value
Sum of the logs Group 1	15.77	2.42	10.80	2.82	< 0.001
Sum of the logs Group 2	11.34	1.05	10.15	2.16	0.20
ERMI	4.43	1.80	0.65	1.45	<0.001

mold in significantly ($p < 0.001$) greater concentrations in dust samples from levels 9 plus 17 compared to the other levels in this water-damaged building (Table 2).

Discussion

These are the first studies to apply the ERMI metric to multi-story, office buildings. In Study 1, occupant health complaints were associated with higher ERMI values than in the comparable building where there were no health complaints. As a result of the ERMI findings, a more intense investigation of the complaint building led to the discovery of a leaky roof and mold growth.

In Study 2, the highest average ERMI value was for level 17. At the time these dust samples were taken in

2002, level 17 had the largest number of epidemiologically defined respiratory cases.^[8,9] Also, dust samples from the upper levels of this building had been previously cultured and hydrophilic molds were shown to be significantly more common on the upper levels.^[9,10,12]

Since 2000, there had been many efforts at remediation of levels 16–19 but no remediation on levels 6–15 prior to 2002. Despite these many efforts, some water problems and health complaints remained in 2002, when these floor-dust samples around workers' workstations were collected.^[10] Relatively high ERMI values on levels 9, and to a lesser extent on level 10, may reflect the lack of remediation on these lower levels. The high ERMI value on level 17 and lower ERMI values on levels 16, 18, and 19 might demonstrate where remediation was not successful compared to where it was more successful. The

Table 2. Comparison of the mean and standard deviation (SD) of each of the 36 molds in the Environmental Relative Moldiness Index measured in dust samples obtained in the two studies. Study 1 – Complaint building levels 2 + 4 vs. No-complaint building levels 2 + 4; Study 2 – Water-damaged building (BLDG) levels 9 plus 17 vs. other all levels (i.e., 6–19, there was no level 13 in this building). The molds in significantly greater concentrations, based on the Wilcoxon rank-sum test and after adjustment for multiple comparison using the Holm–Bonferroni test, are bolded.

Group 1 molds	STUDY 1			STUDY 2		
	BLDGs- Complaint vs. Non-complaint			Water-damaged BLDG		
	(Mean no. cells/mg dust)			(Mean no. cells/mg dust)		
	Levels 2 + 4 (complaint)	Levels 2 + 4 (non-complaint)	Wilcoxon p-value	Levels 9 + 17	Levels – all others	Wilcoxon p-value
<i>Aspergillus flavus</i>	9	3	0.003	0	0	1.00
<i>Aspergillus fumigatus</i>	20	4	0.015	1	0	0.03
<i>Aspergillus niger</i>	404	368	0.03	6	5	0.14
<i>Aspergillus ochraceus</i>	56	9	0.003	2	1	0.33
<i>Aspergillus penicillioides</i>	10	285	0.11	84	44	0.007
<i>Aspergillus restrictus</i>	88	0	0.002	3	2	0.06
<i>Aspergillus sclerotiorum</i>	1	0	0.35	0	0	0.59
<i>Aspergillus sydowii</i>	6	2	0.02	2	0	0.005
<i>Aspergillus unguis</i>	1	0	0.04	0	0	0.59
<i>Aspergillus versicolor</i>	2	7	0.11	51	14	< 0.001
<i>Aureobasidium pullulans</i>	4769	1060	< 0.001	148	126	0.13
<i>Chaetomium globosum</i>	13	15	0.61	6	1	0.09
<i>Cladosporium sphaerospermum</i>	550	130	0.17	24	12	0.01
<i>Eurotium group</i>	22	15	0.83	52	84	0.02
<i>Paecilomyces variotii</i>	9	4	0.03	1	0	0.04
<i>Penicillium brevicompactum</i>	46	5	< 0.001	8	2	< 0.001
<i>Penicillium corylophilum</i>	1	0	0.47	1	1	0.39
<i>Penicillium crustosum</i>	25	1	< 0.001	9	6	0.03
<i>Penicillium purpurogenum</i>	1	0	0.48	0	0	1.00
<i>Penicillium spinulosum</i>	1	1	0.49	0	0	1.00
<i>Penicillium variabile</i>	18	1	< 0.001	30	2	< 0.001
<i>Scopulariopsis brevicaulis</i>	3	0	0.02	0	0	0.72
<i>Scopulariopsis chartarum</i>	2	1	0.04	1	1	1.00
<i>Stachybotrys chartarum</i>	40	22	0.04	0	0	0.73
<i>Trichoderma viride</i>	3	1	0.004	1	1	0.14
<i>Wallemia sebi</i>	226	9	< 0.001	220	85	0.03
Group 2 molds						
<i>Acremonium strictum</i>	444	120	0.08	2	2	0.11
<i>Alternaria alternata</i>	44	71	0.02	13	17	0.54
<i>Aspergillus ustus</i>	1869	1037	0.61	2	0	< 0.001
<i>Cladosporium cladosporioides 1</i>	19	3	0.04	223	234	1.00
<i>Cladosporium cladosporioides 2</i>	1523	98	< 0.001	24	17	0.04
<i>Cladosporium herbarum</i>	2003	967	< 0.001	84	96	0.30
<i>Epicoccum nigrum</i>	181	36	0.12	235	214	0.30
<i>Mucor group</i>	1780	51	0.06	6	3	0.02
<i>Penicillium chrysogenum type 2</i>	5	7	< 0.001	11	17	0.10
<i>Rhizopus stolonifer</i>	18	12	0.8	3	3	0.22

high ERMI value on level 17 suggests that water intrusion was still occurring on that level when these samples were obtained. Therefore, the analysis of dust samples for specific fungi (e.g., hydrophilic fungi) with culture method or ERMI with MSQPCR may be useful in locating and quantifying mold contamination in multi-level buildings.

Most previous studies of mold contamination in multi-level buildings have utilized short air-samples. However, Burge et al.^[13] concluded that “even relatively extensive

air sampling protocols may not sufficiently document the microbial status of buildings.” The Institute of Medicine also noted that air sampling methods have major limitations such as large temporal and spatial variability.^[1] Park et al.^[7] noted that air sampling for microbial agents in indoor environments have many pitfalls and used settled-dust sampling for the study. As a result of analyzing dust samples from this building, mold contamination was linked to health complaints.

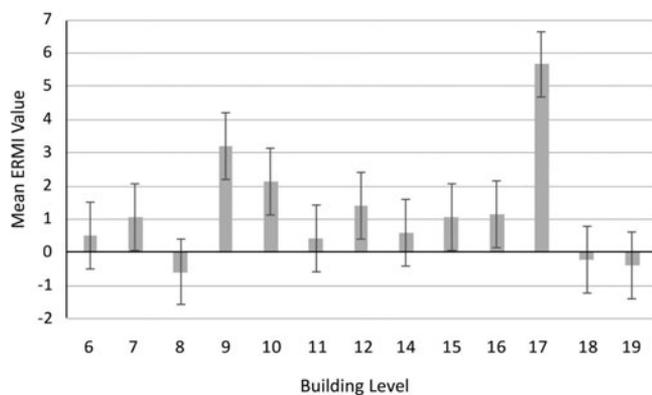


Figure 2. In the second study of the water-damaged building, the mean Environmental Relative Moldiness Index (ERMI) values plus standard deviations (bars) are shown for each level, starting at level 6 (there was no level 13 in the building). The mean ERMI value for all 39 samples was $1.23 \pm$ standard deviation of 2.02.

Although six Group 1 molds and four Group 2 molds of the 36-ERMI molds were in significantly greater concentration in samples from locations associated with health complaints, the results from these studies do not prove that molds caused the health complaints. However, these results are consistent with many earlier reviews of the scientific literature linking dampness in buildings to respiratory health complaints^[1,2,14,15] but there are many other indoor exposures, including pesticides, particulates, volatile organic compounds, etc. which may also be sources of health complaints.^[16] Therefore, having standardized and highly quantitative methods for measuring indoor exposures, including for mold contamination, might help to identify the relevant exposures and directly link them to health effects.^[17]

Conclusion

The ERMI metric may be useful in the quantification of water damage and mold growth in multi-level buildings. However, more studies using the ERMI in the evaluation of water damage and mold growth in large buildings will be needed.

Disclaimer

The findings and conclusions in this report are those of the authors and do not necessarily represent the views of the National Institute for Occupational Safety and Health. The U.S. Environmental Protection Agency (EPA) through its Office of Research and Development collaborated in the research described here. Although this work was reviewed by EPA and approved for publication it may not necessarily reflect official EPA policy. Mention of trade names or commercial products does not constitute endorsement or recommendation by the

EPA for use. Since MSQPCR technology is patented by the US EPA, the Agency has a financial interest in its commercial use.

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