Mycotoxins in Crude Building Materials from Water-Damaged Buildings

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We analyzed 79 bulk samples of moldy interior finishes from Finnish buildings with moisture problems for 17 mycotoxins, as well as for fungi that could be isolated using one medium and one set of growth conditions. We found the aflatoxin precursor, sterigmatocystin, in 24% of the samples and trichothecenes in 19% of the samples. Trichothecenes found included satratoxin G or H in five samples; diacetoxyscirpenol in five samples; and 3-acetyl-deoxynivalenol, deoxynivalenol, verrucarol, or T-2-tetraol in an additional five samples. Citrinine was found in three samples. *Aspergillus versicolor* was present in most sterigmatocystin-containing samples, and *Stachybotrys* spp. were present in the samples where satratoxins were found. In many cases, however, the presence of fungi thought to produce the mycotoxins was not correlated with the presence of the expected compounds. However, when mycotoxins were found, some toxigenic fungi usually were present, even if the species originally responsible for producing the mycotoxin was not isolated. We conclude that the identification and enumeration of fungal species present in bulk materials are important to verify the severity of mold damage but that chemical analyses are necessary if the goal is to establish the presence of mycotoxins in moldy materials.

Mycotoxins are "natural products produced by fungi that evoke a toxic response when introduced in low concentrations to higher vertebrates by a natural route" (J. W. Bennett, Editorial, Mycopathologia 100:3–5, 1987). These compounds can cause a wide range of acute and chronic systemic effects in humans and animals that cannot be attributed to fungal growth within the host or allergic reactions to foreign proteins (22). The over 400 known mycotoxins are all complex organic compounds, most with molecular masses between 200 and 800 kDa (40), that are not volatile at ambient temperatures. Inhalant exposure to mycotoxins can occur by inhaling airborne particulates containing mycotoxins, including dust and fungal components. In agricultural settings, mycotoxicoses in both farm animals and humans can result from oral, dermal, or inhalant exposure of mycotoxin-contaminated grain or dust (for reviews, see references 4, 11, 12, 23, 36, 38, and 41). In laboratory mammals, symptoms can be induced by systemic, oral, dermal, subcutaneous, or inhalant exposure (25, 44), with inhalant exposure in some cases being several orders of magnitude more toxic than dermal or even systemic administration (13, 32, 34).

Toxigenic fungi have been isolated from building materials and air samples in buildings with moisture problems, where the residents have suffered from nonspecific symptoms possibly related to mycotoxin production, such as cough; irritation of eyes, skin, and respiratory tract; joint ache; headache; and fatigue (3, 8–10, 24, 27, 29, 37, 39). In some cases involving *Stachybotrys chartarum* (Ehrenberg ex Link) Hughes, exposure has resulted in pulmonary hemorrhage (8–10), and *S. charta-*

rum isolates from such sites have been shown to produce a number of mycotoxins, including satratoxins (26). Very few studies have, however, established a causal relationship between mycotoxin exposure and building-related illnesses (reviewed in reference 19).

All known mycotoxins are fungal secondary metabolites, which means that mycotoxin production need not be correlated with the growth and proliferation of the producing species and that factors such as induction, end product inhibition, catabolite repression, and phosphate regulation will determine production (6, 7). Therefore, even though some fungi can grow on almost any natural or synthetic construction material, mycotoxin production occurs preferentially on materials that both allow these fungi to grow and provide the conditions for mycotoxin production. From the many studies of the production of mycotoxins by fungal isolates derived from agricultural environments, a great deal is known about the fungal species that are capable of producing known mycotoxins and about the growth media and conditions that induce production (5, 14, 25, 28). It is known that some species include strains that produce mycotoxins and others that lack this ability (6, 7). It also has been established that many of the known mycotoxin producers are frequent colonizers in indoor environments (30, 35, 37). Less is known, however, about the presence of mycotoxins in indoor environments, and it is only in recent years that the presence of some mycotoxins has been verified in crude building materials (1, 14, 21, 27, 40, 41a). In fact, most mycotoxins have yet to be extracted from either air samples or bulk material derived from indoor environments.

Satratoxins belong to the macrocyclic trichothecene class of mycotoxins. Over 100 trichothecenes with irritatory and immunosuppressive effects are known (43). Most trichothecenes were originally isolated from species of *Fusarium*, but they also

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may be produced by other fungi, such as species of *Stachybotrys*, *Trichothecium*, *Cylindrocarpon*, *Myrothecium*, *Trichoderma*, *Vertinosporum*, and *Acremonium* (5, 14, 28, 43). Other mycotoxins potentially present in indoor environments include the carcinogenic aflatoxins and their precursor, sterigmatocystin, which has immunosuppressive and carcinogenic properties. Fumonisins, ochratoxins (nephrotoxic and carcinogenic), zearalenone (estrogenic), gliotoxin (immunosuppressive), patulin (carcinogenic and neurotoxic), and citrinine (nephrotoxic) also may be present (reviewed in reference 22). *Penicillium* and *Aspergillus* species also may produce mycotoxins, commonly found in association with indoor air problems (18). *Aspergillus ochraceus* Wilhelm (ochratoxin A), *Aspergillus fumigatus* Fresenius (fumitremorgins, gliotoxin, and verrucologen), *Aspergillus versicolor* (Vuillemin) Tiraboschi (sterigmatocystin), *Aspergillus flavus* Link (aflatoxins), *Aspergillus parasiticus* Speare (aflatoxins), and *Penicillium citrinum* Thom (citrinine) are among those of particular concern (16–18, 22). Sterigmatocystin also may be produced by *A. flavus*, *Aspergillus nidulans*, *Aspergillus rugulosus*, *Aspergillus unguis*, *Bipolaris* spp., and *Chaetomium* spp., while *Penicillium verrucosum* and *Penicillium viridicatum* may produce citrinine (5, 14, 25, 28).

In the present study, over a period of 4 months, we collected samples for mycotoxin analysis from four major environmental laboratories in southern Finland that are collectively responsible for over 90% of the mycological analyses performed on moisture problem sites in this area. Samples were selected based on mycological analyses down to genus level. A group of 17 mycotoxins likely to be encountered in indoor environments were analyzed, including 4 macrocyclic trichothecenes, 10 nonmacrocyclic trichothecenes, citrinine, sterigmatocystin, and ochratoxin A. As only one set of growth conditions was used to isolate fungi growing on one particular medium, we did not attempt to identify the fungi responsible for producing the mycotoxins in each case. Rather, our objectives were to establish (i) whether these mycotoxins occur in moisture problem sites, (ii) in what materials individual mycotoxins occur, and (iii) which fungal species are associated with mycotoxin-containing samples.

MATERIALS AND METHODS

Sample composition. We analyzed 79 bulk samples of moldy interior finishes, including samples of wallpaper, cardboard, wood, plywood, plasterboard, paper-covered gypsum board, mineral wool, plaster, sand, soil, linoleum, polyurethane insulation, pipe insulation, and paint. The samples were collected from buildings where a moisture problem had been detected either by a municipal inspector or by an occupational hygienist. Additionally, in all these buildings, the examining inspector, hygienist, or physician had recorded the presence of symptomatic individuals, or possibly a mold-induced disease. The 79-sample subset was selected from a larger group based on two criteria: (i) selected samples were usually covered with visible fungal growth, and (ii) one or more of the following species dominated in CFU measurements: *Fusarium* spp., *Stachybotrys* spp., *Trichothecium* spp., *Cylindrocarpon* spp., *Myrothecium* spp., *Trichoderma* spp., *Verticinosporum* spp., *Acremonium* spp., *Bipolaris* spp., *Chaetomium* spp., *A. fumigatus*, *A. ochraceus*, *A. nidulans*, *A. flavus*, *A. unguis*, *A. versicolor*, *A. rugulosus*, *P. verrucosum*, *P. citrinum*, and *P. viridicatum*. Samples were collected over a period of 4 months by health inspectors, occupational hygienists, or environmental inspectors and made available to us by the City of Helsinki Environment Center, Helsinki, Finland; the City of Vantaa Environment Center, Vantaa, Finland; the Finnish Institute of Occupational Health (FIOH), Uusimaa Regional Institute, Helsinki, Finland; HUCH Diagnostics, Mycological Laboratory, Helsinki University Central Hospital, Helsinki, Finland; and the Department of Dermatology and Allergic Diseases, Helsinki University Central Hospital.

Isolation and identification of fungal species. Fungal propagules were isolated from a suspension of 10 g of material in 90 ml of buffer solution (0.3 mM KH_2PO_4, 2.1 mM $MgSO_4$, 2 mM NaOH, 0.02% Tween 80). Dilutions from 10^{-2} to 10^{-5} were spread on 2% malt extract agar (Difco, Detroit, Mich.). Plates were incubated, in the dark, at 25°C for 7 days prior to enumeration and identification. Fungi were identified morphologically to species or genus level.

Preparation and analysis of mycotoxin samples. Mycotoxins were extracted with aqueous 95% methanol, purified by a hexane wash and solid-phase extrac-

tion, separated by reverse-phase high-pressure liquid chromatography (HPLC), identified by tandem mass spectrometry, and quantified using electrospray ionization (ESI) on a quadrupole ion trap mass analyzer, as described previously (42).

The analytes were introduced to the mass spectrometry detector by injecting 10 μl of sample through an HPLC system consisting of an Alliance 2690 separations module (Waters Associated, Milford, Mass.) connected to a Lichrocart 250-3 Purospher RP18 column (Merck, Darmstadt, Germany) online with a four-by-four Purospher precolumn (Merck), both operated at 30°C (Jones chromatography column oven model 7981, HPLC Technology Company Ltd.). A methanol-aqueous buffer (10 mM ammonium acetate) solvent system was used. Sodium acetate (20 μM) was added to solvents for enhancement of cationization in ESI-mass spectrometry. An initial methanol concentration of 20% was held for 4 min, after which the concentration of methanol was raised linearly to 70% at 8 min. This concentration was held for 11.5 min, after which the concentration was raised linearly within 1 min to 90%. The final concentration was held for 15.5 min. The flow rate was 400 μl/min. Between samples, 10 μl of pure methanol was injected into the column and the column was eluted for 4 min with 90% methanol before lowering the methanol concentration to 20% in 1 min and conditioning for 4 min with this solvent. This protocol minimized cross contamination of samples.

Mass spectral analysis was performed on a Finnigan LCQ (Finnigan Corp., San Jose, Calif.) fitted with an ESI probe. The operating conditions were optimized using T2 toxin, roridin A (RDRA), and T2-tetraol. These conditions were as follows. The ESI probe was operated in the positive ion mode and set at a voltage of 1.10 kV. Pressurized nitrogen (690 kPa) was used as auxiliary and sheath gas with a flow rate of 2.5 and 47 dm^3/min, respectively. Helium was used for collision-induced dissociation at a pressure of 275 kPa. Capillary temperature was 260°C, and capillary voltage was 46 V with a tube lens offset of 55 V. The system includes two octapole ion guides with an interoctapole lens in between. The first octapole direct current offset potential was −3.24 V, and the second was −6.5 V, with the interoctapole lens voltage set at −16 V and the octapole RF amplitude at 400. The electron multiplier voltage was set to −800 V. For collision-induced dissociation experiments, the relative collision intensity in the ion trap varied from 12.6 (verrucarol) to 25.0 (satratoxin H [SATH] and RDRA). Maximum injection time was 200 ms, and total microscans were set to 3. Samples were not analyzed in replicates. To each sample, 2 μg of the alkaloid reserpine was added as an internal standard prior to the extraction procedure. Each sample series of six samples contained one or more blank samples to exclude the possibility of false positives. Blank samples were analyzed prior to injecting the actual samples and once more after the last sample had been analyzed. The ion trap, particularly when used as a tandem mass spectrometric device as in the present study, is qualitatively reliable. However, the accuracy of the quantitative analysis was limited by the characteristics of the ion trap, which is a semiquantitative rather than a precise quantitative instrument (42).

Yields of the extraction and purification procedure ranged from 7 to 92%, and detection limits ranged from 0.02 to 200 ng (Table 1). Irrespective of the compound, the intensity of at least two major fragments was used for quantitation purposes (Table 1).

RESULTS

Thirty-four of the 79 samples analyzed (43%) contained one or more of the mycotoxins (Table 2). Mycotoxins were found in most of the material categories tested, with most (82%) of the mycotoxin-positive samples containing cellulosic matter, such as paper, board, wood, or paper-covered gypsum board (Table 3).

Fifteen samples (19%) contained trichothecenes (Table 2), 5 containing the macrocyclic trichothecene satratoxin G or SATH, and 10 containing one of the nonmacrocyclic trichothecenes, diacetoxyscirpenol (DAS), deoxynivalenol (DON), 3-acetyl-DON (3-Ace-DON), T2-tetraol, or verrucarol. The most prevalent toxin was sterigmatocystin, which was detected in 19 samples (24%), while three samples (4%) contained citrinine (Table 2).

Fungi associated with mycotoxin-containing samples. Eighteen of 63 samples contaminated with *Aspergillus* spp. contained sterigmatocystin (Table 3), with *A. versicolor* occurring most frequently (13 samples). Three sterigmatocystin-containing samples did not yield any *Aspergillus* isolates. Species of *Penicillium* were isolated in two of the three cases where sterigmatocystin was found in the absence of *Aspergillus* spp. In addition to 14 samples containing sterigmatocystin, toxin-containing samples contaminated with *Penicillium* spp. included two of the three citrinine-containing samples. The majority of

TABLE 1. Compound-specific properties of separation and detection

Compound	Retention time (min)	Charge	Parent peak (m/z)	CI[a] (%)	Major fragments (m/z)	LOD[b] (ng)	Yield[d] (%)
Nivalenol	6.9	−	371.0	10.5	311.0, 280.0	200	7
T2-tetraol	6.1	+	321.1	15.8	291.1, 263.1, 260.0	20	31
DON	10.4	−	265.1	15.0	217.2, 138.2, 247.2[c]	200	9
Verrucarol	12.6	+	289.1	14.6	245.1, 259.1, 274.0	20	46
3-Ace-DON	13.1	−	337.3	19.0	307.1, 295.2, 277.1	200	10
DAS	14.4	+	389.2	19.0	329.1, 247.2	2	52
Citrinine	14.5	+	251.5	13.5	265.1, 233.2	20	10
T2-triol	14.9	+	405.3	19.0	333.9, 303.1	2	63
Ochratoxin A	15.2	+	426.2	17.2	279.0, 261.1	20	45
Satratoxin G (SATG)	15.9	+	567.2	24.0	523.2, 493.0, 263.1,[c] 231.1[c]	0.2	88
HT2	16.2	+	447.2	17.7	345.1, 387.2,[c] 285.1	0.2	76
SATH	16.6	+	551.5	25.0	523.1, 507.2,[c] 321.1,[c] 303.0[c]	0.2	39
Verrucarine A	17.6	+	525.2	20.8	497.2, 295.1, 249.1,[c] 231.2[c]	0.02	47
T2 toxin	18.1	+	489.1	19.7	387.1, 327.2, 245.1	0.02	82
RDRA	18.2	+	555.2	25.0	527.2, 325.1, 279.1, 231.4[c]	0.02	92
Acetyl-T2 toxin	22.3	+	531.6	20.3	429.1, 369.4, 287.5	0.02	85
Sterigmatocystin	25.7	+	325.5	18.5	310.1, 297.3	2	67
Reserpine (internal standard)	16.4	+	609.3	24.0	609.3, 448.4, 397.1	0.02	76

[a] CI, relative collision intensity.
[b] LOD, limit of detection.
[c] Fragment not used for quantitation.
[d] Yield of extraction procedure.

the 56 samples that contained *Penicillium* spp., however, were negative for both citrinine and sterigmatocystin (Tables 2 and 3). Species of *Fusarium* were detected in 12 samples, only two of which were associated with the production of nonmacrocyclic trichothecenes characteristic of *Fusarium* spp. (Tables 2 and 3). Satratoxins, with one exception, were found only in association with *Stachybotrys* species (Table 2).

Some species were more frequently associated with mycotoxin-containing materials, even when the toxins found were not characteristic of these species (Table 2). For example, *A. ochraceus* was found on eight occasions, all of which were associated with the production of mycotoxin. Yet, ochratoxin A, which is characteristic of this species, was not detected in any of the analyzed samples. On the other hand, all six samples containing *Aspergillus niger* were free from mycotoxin.

DISCUSSION

The present samples are a subset selected from a large pool of buildings with moisture problems and were biased in their microbiology as examined on one particular universal growth medium. Therefore, we cannot draw any conclusions regarding the fungal frequency on moisture-damaged building materials in general. One in five samples of material from which species of *Aspergillus* were recovered contained detectable levels of sterigmatocystin, making it the single most prevalent toxin in this study and, perhaps, indicating that sterigmatocystin is more ubiquitous than previously thought. As in previous studies (17, 21, 33), most sterigmatocystin-producing strains appeared to be *A. versicolor*, but it also is possible that this toxin may have been produced by *Penicillium* spp.

Spread plating on malt extract agar favors the growth of rapidly growing species of *Aspergillus*, *Penicillium*, and *Alternaria* at the expense of the generally slower-growing species of *Stachybotrys*, *Acremonium*, and *Fusarium* (20). The isolation of *Fusarium* spp. might require direct plating on medium specific for this purpose (2). In the present study, 15% of materials were contaminated with *Fusarium* species, but 10 of 12 samples containing nonmacrocyclic trichothecenes characteristic of *Fusarium* spp. yielded no *Fusarium* cultures. Verrucarol has

been reported in *Stachybotrys* spp. (14), but judging from extensive reviews of the mycotoxins characteristic of different species of *Fusarium* and other fungi, it is highly unlikely that the other nonmacrocyclic trichothecenes present (DAS, DON, 3-Ace-DON, and T2-tetraol) originated from fungi other than *Fusarium* spp. (5, 28, 43). It seems that the procedure used to isolate the fungi left most of the *Fusarium* spp. undetected.

The mycology of the building materials did not correlate well with the toxin contents, although when a mycotoxin was found in a sample, representatives of a fungal genus known to contain toxigenic species were present. It is possible for toxigenic species with different growth requirements to be present in the same sample, as they may have proliferated during different stages of the water damage. For example, a surface may be overgrown by *S. chartarum*, which prefers cellulosic matter with a high water content, with nitrogen deficiency promoting satratoxin production, but at an earlier stage of the water damage, at a lower relative humidity, *A. versicolor* could have dominated.

Our findings agree with those of Gravesen et al. (21), in which sterigmatocystin was detected in 19 of 23 samples of building materials artificially contaminated with strains of *Aspergillus* sp. recovered from Danish buildings with moisture problems. They also found trichothecenes in six of eight natural samples tested. Previously, in Finnish water-damaged buildings, trichothecenes were detected in dust and construction material samples, as well as from samples of artificially enriched microbial media (41a). We hypothesize that sterigmatocystin and trichothecenes occur frequently in cellulosic construction materials of problem houses, where some of the fungi used to select the samples analyzed in the present study (*A. ochraceus*, *Stachybotrys*, *Fusarium*, *Trichoderma*, and *Acremonium*) have proliferated as a result of prolonged exposure to high water activities.

Risk assessment of the inhalation of mycotoxins cannot be made from the analysis of bulk samples of construction materials, even if dose responses of humans to airborne mycotoxins were known. However, as many of the fungi that we isolated can elicit allergenic reactions in addition to being toxic (15), it

TABLE 2. Frequency and concentration range of toxins and fungal species found in 79 samples of moldy building materials

Species[a]	Citrinine	DAS	DON	3-Ace-DON	T2-tetraol	Satratoxin G	SATH	Verrucarol	Sterigmatocystin	No toxin found
Acremonium sp.	2 (20–35 × 10³)	4 (14–3,300)		1 (19)		2 (2.5–5.5)			5 (0.2–98)	15
Aspergillus sp.		1 (411)			1 (11 × 10⁴)	2 (170–770)	2 (80–93)	1 (14)	3 (12–31 × 10³)	6
Aspergillus flavipes									2 (0.2–0.8)	
A. ochraceus	1 (320)		1 (920)						5 (0.8–43)	
Aspergillus sydowii									5 (0.8–5)	5
Aspergillus ustus		1 (310)								7
A. versicolor	3 (20–35 × 10³)			1 (22)					13 (0.8–320)	14
Aureobasidium sp.				1 (22)					2 (5.0–24)	
Chaetomium sp.			1 (920)		1 (11 × 10⁴)			1 (14)	2 (43–44)	5
Cladosporium sp.		1 (3,300)							3 (3.9–98)	9
Cunninghamella sp.									2 (0.8–4.2)	
Exophiala sp.	1 (320)								3 (5.0–8.7)	3
Fusarium sp.	1 (320)	1 (14)	1 (920)						5 (4.2–43)	7
Mucor sp.		1 (3,300)								3
Order *Mucorales* sp.		1 (970)								
Oidiodendron sp.		1 (3,300)								
Paecilomyces lilanicus									2 (0.8–24)	4
Penicillium sp.	2 (20–320)	4 (310–3,300)	1 (920)			3 (2.5–35)			14 (0.8–920)	35
Phoma sp.	1 (320)	1 (14)							5 (3.9–120)	5
Rhinocladiella sp.		1 (14)								
Rhizopus sp.		1 (970)								2
Rhodotorula sp.	1 (320)								4 (3.9–8.7)	6
Scopulariopsis sp.	1 (35 × 10³)								3 (4.8–170)	2
Stachybotrys sp.	1 (35 × 10³)					3 (35–770)	2 (80–93)		5 (4.7–31 × 10³)	
S. chartarum						1 (2.5)			1 (3.9)	7
Trichoderma viride		1 (3,300)							2 (2.4–120)	9
Ulocladium sp.		1 (310)								5
Verticillium sp.	1 (20)	1 (3,300)				2 (170–770)	2 (80–93)		5 (0.8–31 × 10³)	2
Aspergillus niger										6
Chaetomium globosum										3
Eurotium sp.										3
Mycelia Sterilia										2
Paecilomyces sp.										3
Paecilomyces variotii										8
Unidentified yeast										4
Total no. of samples	3	5	1	2	1	5	2	1	19	47

[a] CFU measurement was used to determine the presence of each species.
[b] Toxin incidence is shown as number of samples containing the given fungal species which also contained the given toxin. The concentration range, in parentheses, is shown as nanograms of toxin per gram (fresh weight) of sample.

seems that care should be exercised when moisture-damaged sites are torn down or renovated. Sterigmatocystin is an International Agency for Research on Cancer class 2B carcinogen (25) and also has immunotoxic properties (5), while satratoxin G and SATH are probably the chemical agents responsible for stachybotryotoxicosis in mammals (41). In a recent study (24), *S. chartarum* and *A. versicolor* were implicated as causes of building-associated pulmonary disease in workers in three adjacent office buildings. *A. versicolor* predominated in the indoor air, and *S. chartarum* was isolated from bulk samples containing parts-per-million levels of satratoxins. Unfortunately, sterigmatocystin could not be isolated in that study, due to peak interference in UV-HPLC. In addition to the work of Hodgson et al. (24) (2 to 5 μg/g), satratoxins have previously been found in building materials by Johanning et al. (27) (16 μg/g), Croft et al. (14) (not quantified), and Anderson et al. (1) (17 μg/g). To our knowledge, sterigmatocystin has not previously been extracted from building materials naturally contaminated by fungi. The levels of satratoxins in our present study (≤0.77 μg/g of extracted material) were lower than those previously found in building materials naturally contaminated by *S. chartarum* (1, 24, 27) but as high as those found in building materials artificially inoculated with *S. chartarum* and incubated to enrich toxins (31) and almost as high as those encountered with *Stachybotrys*-contaminated rice or fodder (31).

Mycological analyses of air and crude building materials are routinely performed in environmental laboratories to evaluate the extent and spread of damage in buildings with moisture problems and to assess the risk to residents. The isolation of toxigenic species does not substantiate the presence of mycotoxins. However, the present study demonstrates that when mycotoxins are found in bulk materials, some genus known to include toxigenic species usually is present, even if strains from the fungal species probably responsible for producing the mycotoxin are not recovered. In this context, we suggest that the sources of mycotoxic fungal contamination should be removed and necessary precautions should be taken to prevent exposure to potentially harmful aerosolized particles during renovation of buildings with moisture problems.

As the techniques to collect and analyze airborne propagules develop, mycotoxins can be analyzed from indoor air, enabling an assessment of the possible health consequences of mycotoxins for residents of water-damaged buildings. In future studies, the ubiquitousness of mycotoxins in indoor environments

TABLE 3. Proportion of mycotoxin-containing samples to
mycotoxin-free samples among samples contaminated
with the different fungal genera[a]

Genus or sp.	No. of mycotoxin-containing samples/no. of mycotoxin-free samples for material type				
	Cellulose	Gypsum	Mineral wood	Plaster, sand, or soil	Synthetic material
Stachybotrys	2/9	1/3	0/3	0/2	1/2
Aspergillus	2/7	6/19	1/5	0/9	3/8
Penicillium	7/30	3/8	1/6	1/7	2/5
Fusarium	1/7	0/1	0/0	0/2	1/2
Acremonium	3/18	1/4	0/1	0/4	1/6
Chaetomium	1/9	1/1	0/0	0/1	0/0
Trichoderma	1/7	0/0	0/5	0/1	0/1
Phoma[b]	0/0	0/1	0/0	0/2	0/1
Verticillium[b]	0/2	0/5	0/0	0/2	0/1
Paecilomyces variotii[b]	0/5	0/0	0/1	0/2	0/0
Total ratio of myco-toxin-containing samples to myco-toxin-free samples	19/22	9/8	1/5	1/9	3/4

[a] Only mycotoxins characteristic of each fungal genus were taken into account when distinguishing mycotoxin-containing samples from mycotoxin-free samples.
[b] No toxins characteristic of these genera or this species were included in the analysis protocol.

can be evaluated when more mycotoxins are added to the analysis protocol and when more moldy materials are sampled. There are techniques available to analyze most fungi present in environmental samples. Identifying the fungi responsible for producing mycotoxins in building materials will require using such techniques in combination with the enrichment of pure fungal isolates on building materials and extraction of mycotoxins from these isolates. The present study underlines the need for such research. In the largest screen from indoor environments with respect to the number of mycotoxins and samples analyzed, we found mycotoxins in more than 40% of samples.

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