

Fungal Types and Concentrations from Settled Dust in Normal Residences

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Analysis of settled dust collected from carpeting and furnishings is occasionally used by investigators to determine whether an environment contains unusual fungi. Little information is available concerning the types and concentrations of culturable fungi present on textile surfaces in normal residential settings not affected by unusual mold reservoirs, such as from fungal growth sites within the built environment. This study presents the results of the collection and analysis of surface dust from 26 residential environments that were pre-screened by interview, physical inspection, and air sampling to limit the surface dust collection to structures in which there was no history of water intrusion, flooding, plumbing leaks, signs of mold growth, or evidence of unusual airborne fungal spore types or concentrations. In those structures found to have no history or indications of water events or unusual fungi, surface dust was vacuumed from prescribed horizontal areas on carpet and textile-covered furnishings. These samples were then subjected to fungal culture, from which viable colonies were enumerated and identified. Based on the study results, it does not appear reasonable that the frequently quoted total fungi concentration exceeding 10^3 CFU/g is definitive evidence that a residential surface is contaminated with unusual amounts of culturable fungi. Collocated samples collected from eight side-by-side carpets sections revealed poor reproducibility. While settled dust sampling may be appropriate for determining the fungal status of a localized area, or as a gross screening tool, using settled dust results alone to establish the presence of unusual fungal types or concentrations within a structure appears to be inappropriate, and using settled dust results with other investigative methods, such as visual observations and air sampling, requires cautious interpretation.

Keywords carpet dust, fungal contamination, fungi, indoor air quality, mold, settled dust

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Collection and analysis of settled dust from surfaces in the built environment has become one of several environmental sampling and analytical techniques used during fungal evaluations by some

investigators.^(1–3) The objectives of this study were to ascertain what types and concentrations of fungi are typically present in surface dust from residential environments not known or found to be affected by mold. Measurements of total culturable fungi exceeding 10^5 colony forming units per gram (CFU/g) of dust collected from carpet or furniture surfaces have been considered by some as evidence that a building has been contaminated with mold.⁽⁴⁾ Hodgson and Scott have also suggested that the type of taxa present as dominant or major taxa, particularly *Penicillium* spp. and *Aspergillus* spp., is an indicator of fungal contamination and that nonproblem buildings “do not contain a dominant [taxon] when three or more taxa are present, unless the [taxon] is . . . *Cladosporium* or *Alternaria*.”^(4,p.273)

Horner et al.⁽⁵⁾ reported the results of fungal investigations in 50 single-family homes from the greater Atlanta, Georgia region that were selected to determine typical air and surface dust fungal results in normal homes specifically selected for their absence of significant sites of water damage and fungal infestation. They reported indoor airborne culturable fungi median values ranging from 71 to 189 CFU/m³, and dust vacuumed from surfaces in each house showed total culturable fungi up to 3.1×10^5 CFU/g (median values were approximately 2.7×10^4 to 5.2×10^5 CFU/g). Also reported was the near absence of organisms the authors attribute to water indicator fungi, namely *Ulocladium* spp., *Chaetomium globosum*, and *Stachybotrys chartarum*, from 100 dust samples. There are other studies that have reported measurements of settled dust in residences, but they do not indicate the status of the structures with respect to fungal impact^(6–12) or they were conducted solely on fungal-impacted homes.⁽¹³⁾

There is little regulatory guidance available with respect to fungi in settled dust. The basis for an Occupational Safety and Health Administration (OSHA) statement that total fungi values above 1,000,000 CFU/g may be an indicator of the presence of unusual levels of fungi dates back to a 1988 article by Brief and Bernath,⁽¹⁴⁾ who focused largely on Legionnaire’s disease.

METHODS

Study Population

A total of 26 dwellings (18 single-family homes and 8 apartment units) in the northern California area were included in this study. From the 26 dwellings, samples of indoor air, outdoor air, carpet dust, and settled dust from furniture were collected. These dwellings were selected predominantly from colleagues and volunteers who were willing to participate in the study and whose dwellings had passed specific criteria established to ensure that the residence was highly unlikely to be affected by unusual fungi. The goal of the study was to evaluate fungal types and concentrations from dust collected in nonfungal impacted structures. All homeowners or occupants were interviewed to confirm the absence of historical or current water damage, water stains, and signs of possible fungal growth sites.

In general, the single-family homes used in this study were one or two stories with wood-frame (Type V) construction. Type V construction may be of any materials allowed by the Uniform Building Code, and these structures are of 1-hour fire resistive construction throughout. The exterior envelope of the various homes consisted of exterior cement plaster (stucco) or wood siding, and the roofing systems included composition shingle, concrete tile, ceramic tile, and wood shake. Interior wall and ceiling surfaces were gypsum wallboard; floor surfaces were carpet, hardwood planks, or resilient floor covering. The approximate age of the homes ranged from 14 to 108 years (the majority of the structures were less than 30 years old). All except one house had central forced air heating and air-conditioning systems. One house had a wall-mounted heating furnace and did not have air conditioning.

The apartment units sampled were from two complexes: one constructed approximately 26 years ago and the other in the late 1990s. These complexes were two- or three-story structures with wood-frame (Type V) construction. Similar to the single-family homes, the complexes consisted of walls typically constructed of 2- by 4-inch wood members. The exterior envelope of the 26-year-old complex (sample from Apt. 1) consisted of T-111 siding and a composition shingle roofing system. For the complex constructed in the late 1990s, the exterior envelope consisted of composite wood siding with a composition shingle roofing system. All of the apartments had central forced air heating and air-conditioning systems.

Selection criteria were applied to eliminate residences that may have indoor fungal reservoirs. The selection process was performed as follows:

- At the time of inspection, occupants must have lived in their residence for at least 5 years and have had historical knowledge of the condition of their dwelling. The occupants of one house included in the study had lived in their home for only 2 years, but at the time of purchase they had the structure inspected by qualified home, pest, and mold inspectors, and no signs of unusual mold growth sites or

past water/moisture events were noted. The occupants of all structures were interviewed to determine if there had been any water intrusion or plumbing leaks, water stains, any signs of unusual mold growth, any mold-like odors, or any other fungal-related problems. If any of these issues were or had been present, the residence was eliminated from further consideration.

- Each residence was then visually inspected. Each room of the residence was examined. Particular interest was placed on the interior sides of the exterior walls, ceilings, under and around windows, near plumbing service areas (e.g., around and under sinks and hot water heaters, adjacent to showers and bathtubs, near refrigerators, dishwashers, and washing machines), and the relative number and size of interior plants. Dwellings with minor water stains (e.g., small stains under sinks or adjacent to operating sides of windows that could be opened) were evaluated on a case-by-case basis, and the decision to include or exclude homes was based on the apparent extent of the stains and the absence of obvious fungal growth. If there were visible signs of unusual mold growth, stains suggesting leaks, uncontrolled water intrusion, plumbing leaks, water stains exceeding more than several square inches, or an earthy or musty odor that could suggest areas of fungal growth, the residence was excluded from this study.
- Spore trap air samples collected from multiple indoor locations were compared with multiple outdoor air samples at each residence where there were no visual evidence of water stains or mold growth, and when occupants reported no past moisture, water stains, or signs of visible mold growth. Normal findings were interpreted as lower indoor air concentrations for each identified spore type as compared with the outdoor results for each of the indoor air samples. If any of the indoor air samples revealed fungal spore concentrations at levels higher than the outdoor concentrations for that specific residence, the residence was rejected from this study. Six homes were rejected from the study due to possible unusual spore trap results.

Sampling Procedure

To ensure the air samples collected inside each dwelling were representative of baseline indoor air conditions, occupants of each residence were asked to close their windows and doors overnight prior to sampling activities. Normal access and egress through exterior doors were permitted during this time period. Indoor air samples were collected from two to four regularly occupied rooms on each floor of the residence (typically, the living room, kitchen, master bedroom, and an additional bedroom). Multiple outdoor air samples (typically two to six) were collected in the vicinity of each residence or apartment unit on the same day indoor samples were taken.

Indoor and outdoor air samples were collected using Zefon Air-O-Cell sampling cassettes. This device consists of a microscope cover slip coated with a transparent adhesive

medium and encased in a 37 mm diameter polystyrene cassette fitted with a slotted inlet nozzle. Air was passed through the sampling cassette using a Zefon Analytical Biopump (Zefon Int., Ocala, Fla.). This portable, battery-powered vacuum pump was calibrated at a flow rate of 15.0 L/min before and after sample collection by use of a precision rotameter with calibration traceable to a primary standard calibration device (inverted 1.0 L bubble buret). Each air sample was collected for 5.0 min. At the completion of the sampling period, the sample cassettes were sealed, uniquely labeled, and shipped following standard chain-of-custody procedures to an analytical laboratory accredited by the American Industrial Hygiene Association (AIHA) for fungal spore identification and enumeration.

Settled dust samples were collected the same day and from the same rooms that indoor and outdoor fungal spore air samples were collected. However, the settled dust samples were not submitted for fungal spore identification and enumeration until indoor and outdoor air samples confirmed that no unusual fungal types or concentrations were present in the dwelling. These dust samples were always collected following the collection of the spore trap air samples to avoid disturbing the settled dust in an unusual fashion.

In each residence, settled dust samples were collected from three locations: a high-traffic carpeted area located well away from exterior thresholds (e.g., hallway entrance that leads to bedrooms), a low-traffic carpeted area (e.g., a corner of a room located against an interior wall), and the horizontal surface of either textile-covered furniture (upholstered chair or sofa) or a bedspread. Settled dust samples were collected using a 25 mm diameter, 0.8 μm pore size, mixed cellulose ester filter housed in a carbon-impregnated polyethylene cassette that was attached via a flexible tube to a Gast rotary-vane vacuum pump set to draw 25 L of air per minute. This method was selected because it is commonly used by industrial hygienists conducting fungal investigations in residences.⁽¹⁾ The area from which each sample was collected ranged from 0.09 to 0.70 square meters (1 to 7.5 square feet). The surface area sampled was based on the need to collect a minimum quantity of settled dust of approximately 100 milligrams to ensure sufficient dust was available for the culture assay procedure. The size of the area from which the sample was collected was measured and blocked with masking tape.

The cassette top was removed for sampling in an open face configuration, and the opened cassette was passed over the surface under study in two perpendicular directions. After the study area was sampled, the interior of the cassette was examined. If there was a visible dust cake within the cassette, the cassette was sealed. If there was no apparent significant dust cake, the sample area was expanded and additional sampling was performed using the original cassette. The cassettes were then capped, sealed, uniquely labeled, and shipped following standard chain-of-custody procedures to the laboratory for fungal culture and enumeration.

To evaluate the reproducibility of the surface dust sampling method, eight settled dust samples were collected from identical sized, adjacent high-traffic carpeted locations in one residence. These samples were collected from an interior hallway well away from entry thresholds to the house. The carpet in this house was medium pile nylon and was approximately 2 years old.

Laboratory Analysis

Zefon spore traps were mounted onto glass slides using a clear adhesive liquid. A mounting solution (lacto cotton blue) was applied to the slide, and a cover slip was placed on top to protect the slide during analysis. The slide was then examined under transmitted light brightfield illumination using a light microscope. The slide was scanned at various levels of magnification ranging from 200 \times to 1000 \times to assess the numbers of large and smaller spores. One hundred percent of the trace at a magnification of 200 \times was read for the larger spore types (e.g., *Alternaria*, *Chaetomium*, *Bipolaris*, *Curvularia*). The higher magnification required for counting the smaller spore types (e.g., *Penicillium/Aspergillus*, *Cladosporium*, ascospores, basidiospores) present in the trace varied depending on the amount of background debris. A percentage of the trace or the whole trace at higher magnifications of 400 \times , 600 \times , or 1000 \times was used for counting the smaller spore types depending on their distribution and amount. The spores were identified and the count for each individual type of spore was determined. The final spore densities were then calculated and reported in spores per cubic meter.

Dust samples were initially weighed using a microbalance, and a 0.025 g aliquot was removed for extraction and fungal culture. To each dust aliquot, 1.0 mL of 0.1% Tween surfactant was added and the sample was vortexed for 15 sec. The resulting solution was then serially diluted using the Tween solution at concentrations of 1:10, 1:100, 1:1,000, and 1:10,000 onto three formulations of fungal growth media (malt extract agar [MEA], dichloran glycol agar [DG18], and cellulose agar [CA]) in sterile, polystyrene petri plates to enhance the growth of a wide range of fungi. The resulting plates were incubated at a constant temperature of 25°C for 7 to 10 days. The plates were then examined by optical methods by an experienced analyst to identify and quantify the fungi growing on the plates. Identification was made according to morphological features of the fungi. The quantity was based on enumeration of discrete colony forming units. The developing cultures from all of the differing media and dilutions were examined and counted. The reported values are those representing the highest colony forming unit count, for each separate organism, from any of the media formulations and dilutions. The final data were normalized to represent colony forming units per gram of dust.

Statistical Analysis

Indoor air samples were compared with the corresponding outdoor air samples collected from each dwelling. These

comparisons were made using the following measures: the proportion of indoor to outdoor total spores/m³, Friedman chi-square (indoor variation), agreement ratio (indoor/outdoor), Spearman rank correlation (indoor/outdoor), and MoldSCORE (indoor/outdoor).⁽¹⁵⁾

MoldSCORE is a specialized statistical method developed by Environmental Microbiology Laboratory for examining air sampling data. It produces a score between 100 and 300, with 100 indicating a greater likelihood that the airborne indoor spores originated from the outdoors, and 300 indicating a greater likelihood that the indoor airborne spores originated from an inside source. The result displayed is based on the numeric score given and will be classified as either low, medium, or high, indicating the respective likelihood that the spores detected in the indoor air samples originated from an indoor source. Settled dust samples were collected from residences that showed “low” indoor air scores for all indoor air samples. Structures that had one or more indoor air samples with results other than “low” were not included in this study.

Surface dust sample results were grouped according to the location of the settled dust sample (i.e., high-traffic carpet, low-traffic carpet, and bedspread or furniture surface). Graphical displays and Kolmogorov-Smirnov goodness-of-fit tests were applied to verify the assumption that the log-transformed data follow a normal distribution. The geometric mean and geometric standard deviation were calculated, in addition to the arithmetic mean and standard deviation for each sample type. The samples collected from the bedspread or furniture surface were subsequently grouped according to whether they were collected from the surface of a bedspread, headrest area of a sofa, or sofa cushion. A t-test was used to determine the significance of the differences among the various sample types.

RESULTS

A total of 74 settled dust samples were collected and analyzed for culturable fungi from 18 single-family homes and 8 apartment units. These samples were collected from high-traffic carpeted areas, low-traffic carpeted areas, and textile furniture surfaces or bedspreads. A summary of the results with descriptive statistics is presented in Table I, along with the

95% confidence levels for each category. The geometric means were 124,000, 170,000, and 62,300 CFU/g for the high-traffic carpeted areas, low-traffic carpeted areas, and textile furniture surface or bedspreads, respectively.

Table II presents the total fungi for each dust sample, normalized to a relative weight (per gram), with the accompanying descriptive statistics. The range of culturable fungi for all dust samples from all structures was 5280 to 1.69×10^6 CFU/g. Table III presents a breakdown of the data in terms of the fractional distribution of total culturable fungi in varying quantitative levels, which is useful in examining the relative distributions. For example, this table indicates that approximately 54% of the surface dust samples collected from high-traffic areas harbored culturable fungi exceeding 10^5 CFU/g, and 95% of the samples were <500,000 CFU/g.

The concentration ranges of individual fungi taxa (identified to the genus or species level) found in each type of sample, with accompanying descriptive statistics, are presented in Table IV. Table V shows the relative dominance of specific organisms, along with the concentration ranges and geometric means. The results indicate that a single taxon of fungi can dominate these types of samples. “Dominant” fungal types were defined as those representing 50% or more of the organisms cultured from any one sample; “major” types were defined as those representing between 20% and 50% of the total concentration. The dominant taxa were *Cladosporium*, followed by yeasts and *Penicillium*.

The cultured fungi results from samples collected side by side from eight adjacent two-square-foot carpet sections in the same high-traffic sample area in one residence showed a considerable variation in the detected fungal types and concentrations, with organism-specific variation of ten- to fortyfold for *Alternaria* spp., *Aspergillus niger*, *Aspergillus versicolor*, *Cladosporium* spp., *Epicoccum* spp., *Penicillium* spp., *Rhizopus* spp., and yeasts (Table VI). Total culturable fungi in these samples ranged from 34,200 to 315,000 CFU/g, nearly a tenfold difference in these collocated samples. Seven of eight samples showed the same most dominant organism (*Cladosporium* spp.), although the concentration ranged from 16,000 to 160,000 CFU/g in the individual samples for this genus. One sample was dominated by *Penicillium* spp., and this genus was detected in all eight samples (the concentration range was 2800 to 150,000 CFU/g). Several organisms were detected in

TABLE I. Summary Statistics by Sample Type

Sample Type	Number of Samples	GM (CFU/g)	GSD (CFU/g)	95% Lower Confidence Interval	95% Upper Confidence Interval
High-traffic carpet	26	124,000	2.7	83,500	186,000
Low-traffic carpet	25	170,000	2.8	112,000	258,000
Bedspreads/furniture surfaces ^A	23	62,300	3.2	37,300	104,000

Notes: GM = geometric mean; GSD = geometric standard deviation; CFU/g = colony forming units per gram.

^AFurniture surfaces sampled included both sofa cushions and headrests.

TABLE II. Settled Dust Sampling Results for Total Culturable Fungi

Structure ID	Sample Locations		
	High-Traffic Carpet Total Fungi (CFU/g)	Low-Traffic Carpet Total Fungi (CFU/g)	Bedspreads/Furniture Surfaces Total Fungi (CFU/g)
1	123,000	217,000	46,100
2	58,500	38,100 ^A	10,900 ^A
3	55,600	102,000 ^A	46,900
4	41,700	106,000 ^A	39,300
5	142,000	161,000	26,900
6	68,400	157,000 ^A	5,280
7	53,200	49,300 ^A	40,400
8	77,600	81,500	55,200
9	140,000	363,000	440,000
10	71,800	420,000	253,000
11	1.69 × 10 ⁶	529,000	75,700
12	446,000	1.29 × 10 ⁶	89,700
13	428,000	221,000	47,900
14	156,000	260,000	31,400
15	30,900 ^A	127,000	41,500
16	223,000	73,200 ^A	15,200
17	327,000	1.26 × 10 ⁶	42,900
18	267,000	55,500	183,000
19	31,300	108,000	136,000
20	203,000	—	—
21	236,000	37,400	44,800
22	45,300	40,200	258,000 ^A
23	336,000	392,000 ^A	208,000
24	33,200	306,000	490,000
25	154,000	226,000	—
26	144,000	420,000	—
Mean	215,000	228,000	114,000
Maximum	1.69 × 10 ⁶	1.29 × 10 ⁶	490,000
Minimum	30,900	37,400	5,280
Variance	1.05 × 10 ¹¹	1.1 × 10 ¹¹	1.79 × 10 ¹⁰
SD	325,000	331,000	134,000
Median	141,000	161,000	46,900
GM	124,000	170,000	62,300
GSD	2.7	2.8	3.2

Note: “—” indicates that no sample was collected.

^ASample contains >50% water-related fungi organisms (i.e., *Aspergillus*, *Chaetomium*, and *Penicillium* fungi species).

a subset of the total samples; for example, *Aspergillus versicolor* and *Aureobasidium* spp. were detected in two of the eight collocated samples. Nine types of organisms were detected in these samples, with four types found in every sample (*Alternaria* spp., *Aspergillus niger*, *Cladosporium* spp., and *Penicillium* spp.). The results of a chi-square test conducted on the measurements of different types of fungi indicate that, except for *Rhizopus* ($p = 0.74$), the measurements were not consistent ($p = 0$).

The log-transformed total fungi data for all residences, grouped according to the three sample locations (i.e., high-traffic, low-traffic, and furniture surfaces), were normally dis-

tributed. The p -values of the Kolmogorov-Smirnov goodness-of-fit tests ranged between 0.45 and 0.97, confirming that these results follow a lognormal distribution. A t-test was used to compare the geometric means of the dust concentrations in the three areas. Significant differences were found between the geometric means for the samples collected from both the high-traffic/low-traffic locations, considered as distinct groupings, and the furniture surfaces ($p = 0.029$ and $p = 0.0025$, respectively). However, no significant differences were found between the geometric means of fungal dust concentrations in the samples from high-traffic and low-traffic locations ($p = 0.44$).

TABLE III. Fractional Distribution of Total Fungi Concentrations by Concentration Range

Sample Type	<100,000	100,000 to <500,000	500,000 to <1,000,000	1,000,000 to <5,000,000	Total
High-traffic carpet	0.42 (11)	0.54 (14)	0.00 (0)	0.04 (1)	1.00
Low-traffic carpet	0.28 (7)	0.60 (15)	0.04 (1)	0.08 (2)	1.00
Bedspreads/furniture surfaces	0.70 (16)	0.30 (7)	0.00 (0)	0.00 (0)	1.00

Note: Number of samples in parentheses; concentrations listed in CFU/g.

Samples from High-Traffic Carpeted Areas

In samples collected from high-traffic carpeted areas, total fungi levels ranged from 30,900 to 1.69×10^6 CFU/g (Table II). Ten of the 26 samples (38%) had dominant fungal types (Table V) present. The most commonly detected dominant organisms were *Cladosporium* spp., followed by yeasts, *Penicillium* spp., *Aureobasidium* spp., and *Aspergillus niger*. Twenty of the 26 samples (77%) had a major fungal type present. The most commonly detected major fungal types were *Cladosporium* spp., yeasts, *Penicillium* spp., and *Aureobasidium* spp.

Two of the 26 samples from high-traffic areas were dominated by fungi often claimed to be associated with moist building conditions (i.e., *Aspergillus* spp., *Chaetomium* spp., and *Penicillium* spp.) (Table II).

Samples from Low-Traffic Carpeted Areas

In samples collected from low-traffic carpeted areas, fungi levels in surface dust ranged from 37,400 to 1.29×10^6 CFU/g (Table II). Ten of the 25 samples (40%) had dominant fungal types (Table V). The most commonly detected dominant fungal types were *Cladosporium* spp., followed by *Aspergillus niger* and *Penicillium* spp. Twenty of the 26 samples (77%) had at least one major fungal type, with the most commonly detected being *Cladosporium* spp., *Penicillium* spp., and yeasts. Seven of the 25 samples were dominated by fungi often associated with moist conditions (Table II).

Samples from Bedspreads or Furniture Surfaces

In samples collected from bedspreads or textile-covered furniture surfaces, fungi concentrations ranged from 5280 to 490,000 CFU/g (Table II). Nine of the 23 samples (39%) from these surfaces had dominant fungal types present (Table V). The most commonly detected dominant fungal taxon was *Cladosporium* spp., followed by yeasts and *Aureobasidium* spp. Fourteen of the 23 samples (61%) had at least one major fungal type. The most commonly detected major types were yeasts, followed by *Cladosporium* spp. and *Penicillium* spp. Two of the 23 samples were dominated by fungal types that are typically associated with moist conditions (Table II).

DISCUSSION

Surface dust sampling and fungal culture are used by some investigators to determine whether above background culturable fungal propagules are present, possibly indicating the need to conduct some form of fungal remediation. This tech-

nique is also used to assess fungal contamination of personal property or contents items within a structure. In some circumstances, fungal dust analysis is used as a surrogate measure for occupant exposure to fungi. The results from this study indicate that culturable fungi in settled dust from carpets and furnishings within the built environment, in structures where there is no indication of water damage or the presence of unusual fungi from visual observations and air sampling results, is highly variable in terms of the types and amounts of fungi, and may not necessarily be correlated with that from indoor air samples.

The presence of fungi on settled dust should not be used to evaluate occupant exposures. The results from this study are generally consistent with observations made by others.^(7,8,12) Kemp et al.⁽¹⁶⁾ and Loan et al.⁽¹⁷⁾ appear to support this perspective. Kemp et al. collected dust samples from bedroom carpets and mattresses from residences devoid of mold-related problems. Based on the rate of respiration measured for the fungi present, they concluded that bedroom carpets and mattresses in nonproblem dwellings and without moisture damage can provide a habitat with enough moisture to support fungal growth, despite the lack of an obvious moisture source. However, concentrations of total fungi present per gram were not reported. Schober⁽¹⁸⁾ collected samples of floor, bed, and furniture dust from "dry" homes (i.e., no signs of dampness problems or mold damage) and found that *Aspergillus* and *Penicillium* species were commonly encountered and that furniture can be a major source of *Penicillium* spores.

Hodgson and Scott⁽⁴⁾ stated that the taxa present, especially in dominant or major abundance, can also be an indicator of fungal contamination. They called out the presence of *Penicillium* spp. and *Aspergillus* spp. as being typically associated with problem buildings. There is no apparent wide agreement on what organisms are indicators of water damage. Mahooti-Brooks et al.⁽¹⁹⁾ defined moisture indicator fungi as a broad classification of organisms that includes many species of *Alternaria*, *Aspergillus*, *Cladosporium*, *Euotium*, *Penicillium* spp., Basidiomycetes, and several others. Horner et al.⁽⁵⁾ had a considerably smaller list of water indicator fungi, which includes *Chaetomium* spp., *Ulocladium* spp., and *Stachybotrys* spp.

In this current study, *Penicillium* spp. was a major fungi type in a significant fraction of all sample types (present in 93% of all samples) and *Aspergillus* spp. (particularly *Aspergillus niger*) was present in more than half of the samples. These two organisms were not as commonly the dominant types

TABLE IV. Summary Statistics by Taxon and Sample Type

Fungi	High-Traffic Carpet				Low-Traffic Carpet				Bedspread/Furniture Surfaces			
	Range of Concentrations Detected		Median	GM (GSD)	Range of Concentrations Detected		Median	GM (GSD)	Range of Concentrations Detected		Median	GM (GSD)
	Concentrations Detected	Concentrations Detected	GM (GSD)	Concentrations Detected	Concentrations Detected	GM (GSD)	Concentrations Detected	Concentrations Detected	GM (GSD)	Concentrations Detected	GM (GSD)	
<i>Acronium</i>	ND	ND	ND (1.0)	ND	ND	ND (1.0)	ND	ND (1.0)	ND-400	ND	ND	23 (1.9)
<i>Alternaria</i>	ND-36,000	3850	703 (19.6)	ND-45,000	3300	1030 (16)	ND-12,000	1800	582 (14.6)			
<i>Aspergillus flavus</i>	ND-400	ND	22 (1.8)	ND-32,000	ND	37 (6.3)	ND-4000	ND	29 (3.5)			
<i>Aspergillus fumigatus</i>	ND-6700	ND	34 (4.9)	ND-8000	ND	74 (10.9)	ND-4000	ND	37 (4.3)			
<i>Aspergillus glaucus</i>	ND-8000	ND	39 (5.4)	ND-8300	ND	74 (9.2)	ND-8000	ND	31 (4.3)			
<i>Aspergillus nidulans</i>	ND	ND	ND (1.0)	ND-13,000	ND	26 (3.7)	ND	ND	ND (1.0)			
<i>Aspergillus niger</i>	ND-60,000	1450	536 (16.3)	ND-72,000	4000	1920 (12.8)	ND-56,000	400	326 (15.9)			
<i>Aspergillus sydowii</i>	ND	ND	ND (1.0)	ND-17,000	ND	26 (3.9)	ND	ND	ND (1.0)			
<i>Aspergillus ustus</i>	ND	ND	ND (1.0)	ND-20,000	ND	40 (6.9)	ND	ND	ND (1.0)			
<i>Aspergillus versicolor</i>	ND-27,000	ND	32 (5.6)	ND-160,000	ND	54 (11.9)	ND-530	ND	34 (3.3)			
<i>Aspergillus</i> , other	ND-400	ND	25 (2.3)	ND	ND	ND (1.0)	ND	ND	ND (1.0)			
<i>Aureobasidium</i>	ND-200,000	7600	3760 (18.0)	ND-67,000	4400	1650 (15.9)	ND-68,000	4300	2030 (15.3)			
<i>Beauveria</i>	ND	ND	ND (1.0)	ND-6700	ND	25 (3.2)	ND	ND	ND (1.0)			
<i>Bipolaris/ Drechslera</i> group	ND	ND	ND (1.0)	ND	ND	ND (1.0)	ND-1400	ND	24 (2.4)			
<i>Botrytis</i>	ND-4000	ND	25 (2.8)	ND-3300	ND	39 (5.0)	ND-5000	ND	29 (3.6)			
<i>Chaetomium</i>	ND	ND	ND (1.0)	ND-14,000	ND	32 (5.2)	ND	ND	ND (1.0)			
<i>Cladosporium</i>	ND-350,000	25,000	17,900 (8.7)	400-1,200,000	48,000	41,400 (6.0)	ND-180,000	22,000	15,300 (8.4)			
<i>Curvularia</i>	ND-6700	ND	31 (4.5)	ND-400	ND	23 (1.8)	ND-4000	ND	25 (3.0)			
<i>Epicoccum</i>	ND-110,000	3350	644 (23.2)	ND-50,000	4,000	863 (20.5)	ND-12,000	800	375 (15.8)			
<i>Fusarium</i>	ND-4800	ND	51 (7.4)	ND-8300	ND	46 (7.4)	ND-5000	ND	43 (4.7)			
<i>Mucor</i>	ND-4000	ND	52 (5.2)	ND-11,000	ND	117 (12.8)	ND-5000	ND	58 (5.6)			
Non-sporulating fungi	ND-12,000	800	354 (8.3)	ND-8300	400	224 (9.0)	ND-1400	800	199 (6.7)			
<i>Paecilomyces</i>	ND-9500	ND	31 (4.8)	ND-25,000	ND	51 (9.3)	ND-910	ND	27 (2.7)			
<i>Penicillium</i>	200-100,000	9000	8760 (5.6)	ND-430,000	20,000	11,700 (8.4)	ND-120,000	4000	2830 (8.7)			
<i>Periconia</i>	ND	ND	ND (1.0)	ND-8300	ND	25 (3.3)	ND-4000	ND	25 (3.0)			
<i>Phoma/coelomycetes</i>	ND-8000	ND	61 (8.3)	ND-16,000	ND	74 (11.8)	ND-10,000	ND	70 (7.5)			
<i>Rhizopus</i>	ND-4000	ND	117 (7.8)	ND-11,000	400	226 (10.8)	ND-4000	ND	91 (7.7)			
<i>Stachybotrys chartarum (atra)</i>	ND-4000	ND	62 (6.2)	ND-8300	ND	25 (3.3)	ND	ND	ND (1.0)			
<i>Trichoderma</i>	ND-4000	ND	31 (3.6)	ND-8300	ND	29 (3.8)	ND-4000	ND	25 (3.0)			
<i>Ulocladium</i>	ND-9100	ND	105 (12.8)	ND-29,000	3300	730 (18.8)	ND-16,000	ND	213 (14.7)			
<i>Wallemia</i>	ND-16,000	ND	69 (10.7)	ND-12,000	ND	33 (5.6)	ND	ND	ND (1.0)			
Yeasts	ND-1,400,000	8250	5660 (16.3)	ND-140,000	8300	8360 (9.1)	ND-200,000	8800	4180 (11.1)			

Notes: ND = not detected; concentrations listed in CFU/g; GM = geometric mean; GSD = geometric standard deviation; CFU/g = colony forming units per gram.

TABLE V. Dominance Among Fungi in Samples Collected

Fungi	High-Traffic			Low-Traffic			Bedspread/Furniture Surfaces		
	Present (%)	Major	Dominant	Present (%)	Major	Dominant	Present (%)	Major	Dominant
<i>Cladosporium</i>	25 (96)	11	4	25 (100)	11	7	22 (96)	8	7
<i>Penicillium</i> spp.	26 (100)	6	1	24 (96)	10	1	21 (91)	5	0
Yeasts	22 (85)	8	2	23 (92)	7	0	20 (87)	9	1
<i>Aureobasidium</i>	21 (81)	3	1	19 (76)	3	0	18 (78)	2	1
<i>Aspergillus niger</i>	16 (62)	2	1	20 (80)	1	2	13 (57)	2	0
Nonsporulating fungi	18 (69)	0	0	15 (60)	0	0	14 (61)	0	0
<i>Alternaria</i>	17 (65)	0	0	18 (72)	0	0	15 (65)	0	0
<i>Epicoccum</i>	15 (58)	1	1	16 (64)	0	0	13 (57)	0	0
<i>Ulocladium</i>	8 (31)	0	0	16 (64)	0	0	11 (48)	0	0
<i>Rhizopus</i>	12 (46)	0	0	14 (56)	0	0	9 (39)	0	0
<i>Mucor</i>	7 (27)	0	0	8 (32)	0	0	7 (30)	0	0
<i>Stachybotrys chartarum</i> (<i>atra</i>)	8 (31)	0	0	0	0	0	0	0	0
<i>Phoma/coelomycetes</i>	6 (23)	0	0	5 (20)	0	0	7 (30)	0	0
<i>Wallemia</i>	6 (23)	1	0	1 (4)	0	0	0	0	0
<i>Fusarium</i>	5 (19)	0	0	3 (12)	0	0	5 (22)	0	0
<i>Aspergillus fumigatus</i>	3 (12)	0	0	6 (24)	0	0	4 (17)	0	0
<i>Aspergillus glaucus</i>	4 (15)	0	0	7 (28)	0	0	2 (9)	0	0
<i>Trichoderma</i>	3 (12)	0	0	1 (4)	0	0	1 (4)	0	0
<i>Aspergillus flavus</i>	1 (4)	0	0	3 (12)	0	0	2 (9)	0	0
<i>Aspergillus</i> , other	2 (8)	0	0	0 (0)	0	0	0	0	0
<i>Curvularia</i>	2 (8)	0	0	1 (4)	0	0	1 (4)	0	0
<i>Paecilomyces</i>	2 (8)	0	0	4 (16)	0	0	2 (9)	0	0
<i>Aspergillus versicolor</i>	2 (8)	0	0	4 (16)	0	0	4 (17)	0	0
<i>Acremonium</i>	0	0	0	0	0	0	1 (4)	0	0
<i>Aspergillus nidulans</i>	0	0	0	1 (4)	0	0	0	0	0
<i>Aspergillus sydowii</i>	0	0	0	1 (4)	0	0	0	0	0
<i>Aspergillus ustus</i>	0	0	0	3 (12)	0	0	0	0	0
<i>Beauveria</i>	0	0	0	1 (4)	0	0	0	0	0
<i>Bipolaris</i> / <i>Drechslera</i> group	0	0	0	0	0	0	1 (4)	0	0
<i>Botrytis</i>	1 (4)	0	0	4 (16)	0	0	2 (9)	0	0
<i>Chaetomium</i>	0	0	0	2 (8)	0	0	0	0	0
<i>Periconia</i>	0	0	0	0	0	0	1 (4)	0	0

Note: "Dominant" is defined as those fungi in a cultured sample constituting greater than 50% of the total fungal concentration. "Major" is defined as those fungi in a cultured sample constituting 20–50% of the total fungal concentration.

(defined as >50% of the cultured organisms) as compared with *Cladosporium* spp., but did dominate in approximately 7% of the carpet dust samples. If the presence of moisture indicator fungi as defined by Mahooti-Brooks⁽¹⁹⁾ is used in examining the settled dust samples, all of the houses/apartments sampled during this current study would be misclassified as affected by unusual indoor fungal reservoirs. If the data are examined in the context of the water indicator fungi suggested by Horner et al.,⁽⁵⁾ approximately 50% of the structures in the current study would be misclassified. While there may be some appropriate classification for using the presence of these water indicator fungi in settled dust samples, based on

this current data set, the mere presence of these organisms is not appropriate. Applying a quantitative value to the detected presence of these high-moisture-dependent organisms in dust may be necessary to determine if unusual fungi are present from an indoor moisture related reservoir.

The fungi listed in Table IV represent all types that were detected in at least one or more dust samples: *Penicillium* spp., *Cladosporium* spp., *Aureobasidium* spp., and yeasts were present in more than 80% of all samples. Nonsporulating fungi, *Alternaria* spp., *Aspergillus niger*, *Epicoccum* spp., and *Rhizopus* spp. were present in approximately 40% to 80% of all samples and sample types. *Cladosporium* spp. was detected as

TABLE VI. Results of Collocated High-Traffic Area Samples Collected from One Residence

Sample Number	<i>Alternaria</i>	<i>Aspergillus niger</i>	<i>Aspergillus versicolor</i>	<i>Aureobasidium</i>	<i>Cladosporium</i>	<i>Epicoccum</i>	<i>Penicillium</i>	<i>Rhizopus</i>	Yeasts	Total Fungi
S1	8000	20,000	12,000	ND	88,000	16,000	4000	ND	60,000	233,000
S2	4300	8700	1300	ND	120,000	ND	150,000	430	ND	315,000
S3	400	2400	ND	ND	16,000	2000	4400	400	6800	34,200
S4	800	24,000	ND	ND	16,000	20,000	4400	400	4800	72,500
S5	4000	1600	ND	ND	100,000	16,000	2800	400	9600	144,000
S6	420	2500	ND	1700	18,000	2500	6300	420	8800	42,500
S7	430	1700	ND	1300	20,000	ND	3900	430	3900	34,300
S8	4000	1200	ND	ND	160,000	24,000	6800	ND	9600	220,000
GM	1500	4100	75	59	45,000	2000	6900	200	4300	98,000
GSD	3.5	3.3	12.4	7.4	2.8	19.8	3.6	4.1	10.2	2.5

Note: ND = not detected; concentrations listed in CFU/g.

a dominant fungi in all sample types (i.e., high-traffic carpeted area, low-traffic carpeted area, and furniture surface or bed-spread). Of the 74 dust samples collected, *Cladosporium* spp. was the dominant fungal type in 41%. *Penicillium* spp., *Cladosporium* spp., *Aureobasidium* spp., yeasts, and *Aspergillus niger* were detected as major fungi in samples from all surface types.

Horner et al.⁽⁵⁾ also found that *Cladosporium* spp., yeasts, and *Penicillium* spp. were the most commonly detected culturable fungi from dust collected in the 50 houses studied in the Atlanta area. Horner et al. reported the highest median values as 4.8×10^4 and 5.2×10^4 CFU/g when using DG18 medium to culture fungi from dust samples collected in the winter and the summer. The results from this current study were higher, with the median value of 1.2×10^5 CFU/g for the entire data set. The highest concentration observed (1.7×10^6 CFU/g) was significantly higher than the highest concentrations observed in the Atlanta houses.

There were several differences that could account for this observation. The climatic differences are notable (Atlanta vs. Northern California). The sample collection devices were also different (Horner et al. collected samples using vacuum dust collection bags, whereas our samples were collected using open-face, 25 mm diameter, 0.8 μ m pore size, mixed cellulose ester filters housed in cassettes). Horner et al. cultured dust that was finely sieved and sprinkled directly on two different types of culture media (MEA and DG18), while our samples were mixed and vortexed with a liquid surfactant and plated at differing dilutions on three different growth media (MEA, DG18, and cellulose agar).

A likely substantial difference is in how the resulting fungal colonies were counted. Horner et al. presents individual counts for the two different media, whereas the data received for the current study present the highest counts of each identified organism, from each growth media (the results for the individual growth medium are not presented individually). This practice is common to many commercial environmental fungal laboratories and could explain, in part, the apparent higher values found in this current study as compared with what was observed by Horner et al.

Hodgson and Scott⁽⁴⁾ postulated that total fungal concentrations greater than 10^5 CFU/g will most likely be associated with buildings contaminated with fungi. The data from their control (i.e., nonproblem) buildings are limited and may not be representative. All samples reported in the Hodgson study were collected from office buildings, which likely are different from residential environments partly because of the way in which outside air enters the structure (e.g., filtered vs. unfiltered outdoor air). Commercial office buildings are likely to be cleaned at different frequencies and by different methods than residential settings, and the sources of fungi-containing dusts from occupant activities are likely to be different from residential settings (e.g., different occupant activities, such as no or few children and pets in commercial settings).

Of the five control buildings studied by Hodgson and Scott, three had been problem buildings previously and had been

subsequently remediated. It is unclear what this means for surface dust, but it seems likely that some type of treatment, specialized cleaning, or remediation of surfaces would have been conducted. In addition, the study reports that "seven control building culture plates and two problem building culture plates did not contain fungal growth."⁽⁴⁾ This finding is unusual, given the ubiquitous nature of fungal spores and the expectation that horizontal surfaces in normal buildings should contain some detectable levels of culturable fungi (e.g., in this current study, there were no dust samples in which fungi was not detected). However, some control samples were from new carpet and, in two cases, the samples were collected after the application of biocidal treatment to the carpets. It is likely that the samples from control buildings in the Hodgson and Scott study are not representative of the typical surface dust found in residences or in normal office buildings or other common building environments.

Based on the results from this current study, it does not appear reasonable that a total fungi concentration of $>10^5$ CFU/g is definitive evidence that an indoor surface is contaminated with unusual amounts of culturable fungi. Had this criterion been applied to the data from this current study, it would suggest that 54% of the samples were indicating "mold contaminated" and, when examined on a per structure basis, this criterion could suggest that almost 90% of the structures were mold contaminated, whereas other evidence indicates that the structures are not affected by unusual fungal reservoirs. If a numerical value of 10^6 CFU/g of total culturable fungi were used as a contamination criterion, the data from this study would indicate that an erroneous conclusion would be drawn for 4% of the samples. Horner et al.⁽⁵⁾ did not attempt to determine what might be a quantitative value suggesting unusual fungal contamination. A significant fraction of the samples collected from the 50 normal (nonfungal or water impacted) structures examined by Horner et al. were above a value of 10^5 CFU/g (approximately 10% to 20% of their samples exceeded this value).

The ten- to fortyfold variation in culturable fungi concentrations from the eight collocated dust samples collected in a high-traffic location is unexpected. This difference is likely attributable to the variability in the sampling and analytical methods and the variation inherent to the deposition and collection of culturable fungi on textile surfaces in this type of environment. This high degree of variability demonstrates that vacuum dust samples collected from immediately adjacent sections of carpet are not reliably reproducible. Because of the wide variation in results, if carpet dust samples are to be used as an investigative method, multiple samples should be collected and the type of variation demonstrated in this limited study must be considered in the interpretation of the results. If these data were to be used to reach important and significant conclusions (such as determining whether the space is contaminated and in need of some type of remedial activity to restore it to normal conditions), the apparent high degree of variability indicates the need to collect many dust samples from any single structure or area within a large structure. Collecting

a small number of surface dust samples could easily lead to erroneous conclusions. However, if a small number of samples reveal very high concentrations of a clearly unusual type of fungi (e.g., $>10^6$ CFU/g of *Stachybotrys* spp. or *Fusarium* spp.) as the dominant or sole detected organism, it may not be necessary to collect a large number of samples to provide convincing evidence that unusual types and concentrations of fungi are present.

Limitations

One possible study limitation is the size of some of the sample aliquots (i.e., <0.01 g), which could result in widely varying results and bias high the reported concentration values. However, within this study, most sample aliquots were of uniform size (i.e., 0.025 g), and only a limited number of dust samples from low-traffic areas were less than 0.01 g. In the experience of these investigators, it is not uncommon to see settled dust samples collected for which the mass is as small as 0.01 g. The results from the analysis of such small samples, which could bias high determinations of per gram colony counts, should be viewed carefully and should be avoided if possible.

Fungal growth media used in the culture process can also affect the results of a culture assay because the medium selected may exclude, suppress, or exaggerate the presence of some types of fungi.⁽⁸⁾ In this current study, three popular and commonly used growth media that would support a wide range of fungi were used in an attempt to maximize fungal growth and support a broad spectrum of fungal types.

It is notable that no widely accepted standardized equipment or method for collection and analysis of settled dust for culturable fungi is recognized. The methods employed varied among the studies reviewed in this article. Thus, while others, such as Spurgeon,⁽¹⁰⁾ have reported results from carpet dust samples collected from areas known to be subject to water intrusion, comparing such data with those from other studies remains challenging if not impossible.⁽²⁰⁾

CONCLUSIONS

These data will continue to be supplemented with additional data with the intent of establishing a database of typical background fungi levels in residential settings unaffected by unusual mold sources. The study results suggest that total fungi levels from carpet dust or furniture surfaces exceeding 10^5 CFU/g is not definitive evidence that a residential surface is contaminated with unusual amounts of culturable fungi. While settled dust sampling may be appropriate for determining the mold status of a localized area, such as a mattress, or as a gross screening tool, the use of settled dust results alone to establish the mold status of a structure appears to be inappropriate, unless highly unusual findings are found, such as a set of samples dominated by elevated levels (i.e., $>10^6$ CFU/g) of an unusual fungal type typically associated with water intrusion, such as certain species of fungi like *Aspergillus*, *Chaetomium*, or *Stachybotrys*. Fungi are present in normal residential structures

from a variety of pathways and reservoirs, such as the migration of outdoor air, dust and debris from foot traffic, common indoor reservoirs such as potted plants, normal decaying foodstuff occasionally present in kitchens, and from other less recognizable sources. As fungal propagules age, some lose viability at a faster rate than others; thus, what becomes culturable using common environmental sample testing methods may not be a very good indicator of the past history of the presence of fungi in any particular environment.

Collection and analysis of surface dust for fungal identification alone is likely a poor measure of determining if unusual types and concentrations of fungi are present. Traditional investigative procedures, such as visual inspections, measurement of surface moisture levels (when possible), and collecting representative surface samples from stained locations, is a more reliable and straightforward way of assessing the built environment for unusual fungal contamination. Because highly variable results were observed in collocated samples, if collection and analysis of surface dust is being used, multiple samples should be collected from a single structure. The results from a small number of samples may not be a reliable measure of the type or concentration of fungi in settled dust. A standardized dust collection and analytical method should be considered if one expects to compare results from different studies.

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