RESEARCH REPORT

HOUSE DUST:
AN EFFICIENT AND AFFORDABLE TOOL
TO ASSESS MICROBIAL CONTAMINATION
IN HOMES

**EXTERNAL
RESEARCH
PROGRAM**

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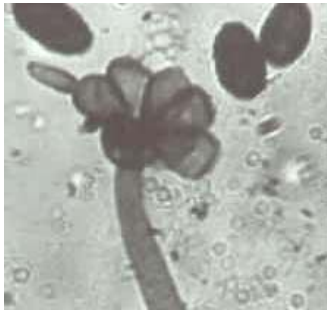
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FINAL REPORT

External research project

House dust:
an efficient and affordable tool
to assess microbial contamination in homes

February 2002

To Claude Mainville, because making the impossible come true
is just a matter of time…

ACKNOWLEDGEMENTS

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EXECUTIVE SUMMARY

In recent years, in an increasingly polluted environment, the family home, which should be a haven of peace and a shelter, has often become a source of microbial contamination, where molds, yeasts and bacteria proliferate. In such a context, the importance of a good diagnosis of the degree of microbial health in homes is therefore on the agenda. Our experience in the field has brought us to suggest the use of an inexpensive diagnostic tool: an analysis of the microbial content of dust in homes. This project was conducted to highlight the link existing between the history of water activity in a home and the microbial content of its dust. Thanks to an External Research grant from CMHC, an inspection and an analysis of the microbial content of the dust in 68 healthy homes enabled us to make a comparison with 145 unhealthy homes in our database, to confirm the validity of the method.

Homes in the Montréal area selected by an advertisement placed in two newspapers (*La Presse* and *Voir*), direct faxing, door-to-door distribution of a brochure, and word of mouth. A selection was made using a telephone questionnaire to eliminate homes that did not meet the microbial health criteria established for this project, including these main conditions: no major water damage during or since the 1998 ice storm, no health problems having appeared or worsened since moving in, at least two years of occupancy, no carpets in the basement, no poorly maintained forced air systems, with porous insulation or humidifiers.

The inspections, ensured by inspectors from the Groupe NATUR'AIR-KIWATIN of Montréal, lasted a minimum of one hour and a half and consisted of visiting the premises thoroughly both outside and inside, checking the structures with a moisture detector, conducting a complementary survey with the occupants, taking photographs and taking samples of composite dust with a vacuum over at least one square meter of surface other than the floor in inhabited rooms.

Laboratoire MICROVITAL of Montréal, in charge of performing the microbial analysis on the dust samples, proceeded at random with unidentified sample numbers. The molds and yeasts were placed on MEA culture mediums and the bacteria on PYA. The mediums were made at Concordia University. The mold counts and genus identification were done at MICROVITAL, in cooperation with Dr. Paul Widden, Ph.D., researcher at Concordia's department of biology. The molds were identified at the genus level, and at the species level in some cases.

The following results were obtained:

a)   The average mold counts in the dust from the unhealthy homes (500,000 cfus/g) are seven times higher than in healthy homes (75,000 cfus/g), and this difference is statistically significant.
b)   The average mold counts in homes are not influenced by the inspection season.
c)   Phylloplane molds (*Cladosporium* and *Alternaria*) dominate in healthy homes, while non-phylloplane molds (*Penicillium* and *Aspergillus*) dominate in unhealthy homes, and this difference is statistically significant.
d)   Bacteria are more abundant in the fall in healthy homes, and yeasts, in all homes.

**These results are conclusive: the mold content of the dust in a home
is an indicator of its degree of microbial health.**

ABSTRACT

In Greater Montreal, in 2000 and 2001, the microbial contents of dust from 68 "healthy" houses without water damage or health complaints from occupants was compared to that of 145 "sick" houses with significant water damage. Mean fungal counts were 7 times higher in the sick homes. Fungal distribution in healthy houses--with predominant *Cladosporium* and *Alternaria*-like genera--differed from that of unhealthy houses, where *Penicillium* and *Aspergillus*-like genera were predominant. These results demonstrate that the fungal contents in dust is indicative of the microbial contamination of a house.

CHAPTER 1.   MICROBIAL CONTAMINATION IN HOMES NOWADAYS

1.1   The situation

In recent years, because of our increasingly polluted environment there has been an unprecedented increase in various allergy cases, pediatric asthma, atopy and hypersensitivity in North America (CDC Report on Asthma, 2000). The family home—which is supposed to be a peace haven and a refuge—has often become a source of pollution.

Inadequate ventilation and abnormal water activity (water damage episodes, excess humidity) often cause the proliferation of fungi (molds), yeasts and bacteria on visible surfaces or hidden inside structures. In scientific literature, this microbial contamination of the indoor environment is associated more often now with complaints from some occupants experiencing health problems (review article from A.L. Pasanen, 2001).

In such a context, the importance of a good diagnosis of the degree of microbial contamination in homes becomes more relevant. Unfortunately, assessment tools are few and insufficient. Air sampling, still frequently used, is an incomplete tool, not reproducible and it can lead to false negative results. On the other hand, surface samples are useful to document the nature of visible fungal contamination but insufficient to obtain a global diagnosis (ACGIH, 1999).

1.2   Project objectives

Our field experience led us to promote use of another contamination assessment tool: microbial analysis of house dust. David Miller (Ph.D.), then a mycologist from Agriculture Canada, was the first to use this method of analysis. He is convinced that dust is the "memory of a building" and gives valuable information on its microbial history (Miller, 1988). Since then, numerous other articles confirmed this assumption (Wickman et al, 1992; European community, 1993; Flannigan et al, 1994; American Industrial Hygiene Association, 1996; Pasanen and al, 1997; Veerhoeff and Burge, 1997; Dillon and al, 1999; Hodgson and Scott, 1999; Mainville and al, 1999; Miller and al, 1999).

On the other hand, it is difficult to establish a link between the microbial contents of the air occupants breathe and the microbial contents of the dust itself (Flannigan, 1997; Veerhoeff and Burge, 1997; Dillon and al, 1999). It is not our intention in this study to debate the usefulness of dust as a tool to assess the occupants exposure to micro-organisms.

This project was conducted to highlight the link between the history of abnormal water activity in a dwelling and the microbial content of the dust in that dwelling. With more than five years of field work, we have already analyzed the microbial content of fungi, yeasts and bacteria, in dust samples taken from hundreds of problem dwellings. Thanks to an External Research grant from CMHC, we were able to inspect and analyse the microbial content of dust from more than fifty healthy homes; we were also able to supplement our database and confirm the validity of the method.

CHAPTER 2.   RESEARCH METHODOLOGY

This chapter describes the work involved at every stage of the project, from the selection of healthy homes to inspection and microbial data processing.

2.1   Selecting healthy houses

2.1.1   Selection criteria

The microbial criteria did not take into account cigarette smoking, chemicals (solvents, glues…) or other factors (such as radiations, etc…).

a)   Length of occupation

At the time of inspection, the house must have been occupied for at least two years by the same occupants, unless it was built less than two years ago.

b)   Basement

Basement dwellings were not eligible for the study.

c)   Occupants' health

Exclude dwellings in which occupants had experienced health related symptoms that started or worsened since they moved in.

d)   Water activity history

The history of water activity and/or microbial contamination problems that occurred before the inspection needs to be known in detail, preferably for the last five years and at least since the 1998 ice storm. Any major water damage episode will exclude the dwelling from the study. Any minor water damage episode will be evaluated on a case by case basis, according to the way it has been remediated.

e)   Heating and humidifying

Homes with central forced-air heating are eligible if the system does not include a cold water humidifier or porous insulation and if it has been properly maintained.

f)   Carpeting

Homes with carpeting in the basement are excluded from the study, unless the carpet is new (less than one year old), covers a small area, or is installed on a dry insulated concrete floor. Wall-to-wall carpeting on the second floor of the dwelling may be acceptable as an exception, on a case by case basis, depending on its age, state and area covered. Small rugs are tolerated. Dwellings with both pets and carpeting are excluded.

g) Miscellaneous

Homes with whirlpools, saunas and indoor pools are excluded from the study, as well as those with sumps or sump pits with a wooden structure or without a cover. Dwellings storing more than twenty logs for the fireplace or using more than one cord of wood per winter are also excluded.

2.1.2   Recruiting

Applicants are selected prior to inspection through a phone survey and questionnaire to verify if they meet the above-mentioned criteria.

2.1.3   Confidentiality

When a home is selected through the phone survey, its file is forwarded by MICROVITAL to NATUR'AIR-KIWATIN where it is given a confidential number similar to the file numbers used for its other residential customers. This made it impossible for MICROVITAL to determine the origin of the corresponding sample.

2.2   Inspection protocol

All homes were inspected by professionals from Groupe NATUR'AIR-KIWATIN (NAK), a consultant firm specialized in air quality and microbial contamination in buildings. Both NAK inspectors, Robert Kelly and Alain Beaudet, have taken CMHC training sessions on building inspections (three days in 1999 and two days in 2000, respectively). They also acquired a great deal of field experience performing residential inspections for microbial contamination assessment. NAK has already inspected several hundred homes in the last five years and is the only CAA Housing accredited firm in Greater Montreal.

Each inspection lasts at least an hour and a half and combines the following elements:

2.2.1   Evaluation of occupants health symptoms

The inspector validates the home selection phone survey by confirming that none of the occupants have experienced new or worsened health symptoms since they moved into the inspected home.

2.2.2   Water activity history

The inspector confirms with the occupant that no major water damage episode has occurred during or since the 1998 ice storm. The inspector conducts the survey and asks questions on matters that occupants may not always think about such as: state of the attic, state of the drainage system, plumbing leakages, etc.

2.2.3 Detailed inspection of the premises and dust sampling

The inspector visits every room in the basement, attic and garage looking for traces of water activity. He takes temperature and relative humidity readings and systematically checks moisture levels in materials from the surrounding area and everywhere he considers necessary. This is done with a non-invasive moisture detector which measures the presence of water in materials through its electrical conductivity.

The inspector also checks the building exterior and verifies structural integrity (walls, roof, foundation), eavestroughs, site grade, drainage system, chimney, nature of the soil, etc.

The inspector notes his observations on the residential file and takes photographs if necessary.

Finally, the inspector takes dust samples from inhabited rooms, as described in section 2.3.

2.3 Dust sampling protocol

2.3.1 Sampling tools

For dust sampling, inspectors used a portable Hoover Portapak vacuum cleaner with disposable paper bags. Prior to use, all the movable parts of this vacuum cleaner were cleaned with a solution of 250 ml of commercial bleach in 4 litres of water with a little liquid soap. All the parts are then completely dried.

2.3.2 Sampling

Using the vacuum cleaner, the NAK inspector takes a composite sample of dry dust from the occupied rooms. To reduce outside influence, samples are not taken from the floor but higher: for example, on bookshelves, kitchen shelves, the top of a heating baseboard, door frames, etc. Depending on the level of dust accumulation, the total sampling area in the dwelling can be anywhere between 1 and 2 square meters (precisely measured). The sampling area must be vacuumed for 5 minutes.

After sampling, the vacuum cleaner bag is removed, sealed with adhesive tape and identified with a number. It is placed in a tightly sealed ziplock bag and brought to MICROVITAL where it is kept at 4 degrees C until it is placed in culture, within a maximum timeframe of 6 days.

2.4 Culturing

Once a week, MICROVITAL places in culture all the numbered samples stocked in the fridge, including those from the healthy houses inspected for this study.

2.4.1   Culture mediums

All the culture mediums used by MICROVITAL are prepared at Dr. Paul Widden's laboratory. Dr. Widden (Ph.D.) is a researcher in mycology at Concordia's Biology Department.

MEA Rose bengal medium for yeast and fungi:
Agar Sigma A-7002	15 g/l
Malt Extract Agar	20 g/l
Rose bengal	0.01 g/l

PYA medium for total bacteria:
Agar Sigma A-7002	15 g/l
Bactopeptone	5 g/l
Yeast extract	1 g/l

Difco HAJNA medium for Gram negative bacteria: catalogue # Difco 0486-17-4

2.4.2   Dust weighing and dilution

Under an extraction hood--with gloves and using scissors and tweezers that have been sterilised in 70% ethanol and flamed before each use--each dust bag is weighed, then opened. An amount of 0.100 gram of dust is then weighed and put in suspension in a volume of 10 ml. of sterile water.

The suspension is then strongly vortexed for 30 seconds to allow micro-organisms to be released from the dust in the liquid. Near the flame, and with a disposable sterile pipette, a fixed volume of the liquid is then sampled and deposited on the surface of 6 petri dishes: duplicates for total bacteria, Gram negative bacteria and fungi. A "hockey stick" (curved glass rod) previously sterilized in 70% ethanol and flamed is then used to spread the liquid evenly on the surface.

The petris are then incubated at 20 degrees C in the dark, two by two in sealed ziplock bags. Incubation lasts 48 hours for bacteria and 7 to 14 days for yeast and fungi.

2.4.3   Counting and identifying micro-organisms

a)   bacteria

Duplicates of total and Gram negative bacteria are counted under the dissecting microscope after 48 hours of incubation.

b)   yeast and fungi

Duplicates of yeast and fungi are counted under the dissecting microscope after 7 to 14 days of incubation, depending on their speed of sporulation. Note that it is impossible to identify a fungal colony that has not been sporulating.

Genus identification of fungi is done by observing general characteristics of colonies under the dissecting microscope: color, shape, size. Genus and species identification of a fungal colony are confirmed under the phase contrast microscope. In some cases, particularly for certain *Aspergilli* and *Penicillia*, species identification requires subculturing the colony on specific culture medium. When in doubt, colonies are identified in the Biology Department of Concordia University, with the collaboration of Dr. Paul Widden (Ph.D.), mycologist.

Since yeasts grow very fast in a culture medium, it is often impossible to count individual colonies. Quantification of the amount of yeasts present on a culture plate is done by evaluating the percentage of surface they occupy on the plate. If necessary, the same method is used for quantification of certain fast growing fungi that are often difficult to count, *Trichoderma* and *Mucor*, for example. See section 3.4 for comments on the reliability of this method.

Non-countable yeasts and fungi, evaluated using the occupied surface on the dish:
Traces (less than 10% of surface)
1+ (10 to 25%)
2+ (25 to 50%)
3+ (50 to 75%): large amount
4+ (75 to 100%): very large amount

2.4.4   Forwarding results

Every week, Microvital sends NAK all the lab reports, identified by the sample numbers, including those of the healthy houses in this study.

2.5   Data analysis

NAK sends back to MICROVITAL the completed files for every healthy home, including the work report, phone questionnaire, inspection report and lab report. All the data are entered by MICROVITAL into an EXCEL data base with more than 50 information parameters on home characteristics, inspection observations and microbial results from dust sample analysis.

Statistical analysis was made possible using the Excel data base. With the statistical software Jump'in 4.3, ANOVA or Wilcoxon/Kruskal-Wallis (for non normal distributions) tests were performed to compare results.

CHAPTER 3.   RESULTS

3.1   General characteristics of inspected homes

3.1.1   Number of inspected homes

In the scope of this project: 52 healthy homes were inspected.
From the NAKdata bank: 16 healthy homes already met the selection criteria for this project. These homes were inspected by NAK for a customer either prior to selling or buying, for a customer to select a new apartment to rent, for putative problems with chemicals, etc.

Total number of healthy homes:

68 = 52 inspected in this study + 16 already inspected by NAK

3.1.2   Contamination level of inspected homes

CONTAMINATION LEVEL	NUMBER OF HOMES
Healthy	68 (25%)
Unhealthy	145 (75%)
Very unhealthy	82 out of 145 (31%)
Total	213

Table 12.   Contamination level of the selected data bank homes

The purpose of this study was to compare microbial contents of dust samples taken from healthy homes inspected in this study and from unhealthy homes inspected by NAK since 1997.

a) Healthy homes

Water activity (duration): none or less than 24 hours
Water damage history: none or light

Possible examples of light water damage history:
-       A one-time plumbing leakage underneath the kitchen sink with repairs done less than 24 hours after.
-       A window left open for a few hours during a rain storm, resulting in some water on the wooden floor but dried rapidly.

These criteria were used to select the 16 healthy houses from the NAK data bank that were added to the 52 healthy homes inspected in this study.

b) Unhealthy homes

Water activity (duration): 24 hours and more, or one week and more, or chronic;
Water damage history: moderate to extensive

Possible examples of moderate water damage history:
- The clothes washer overflowed on the linoleum floor covering in the basement and water was improperly drained by the floor drain and the walls were not touched.
- Water infiltrated through a crack in the wall underneath a window between the bricks of the exterior wall on a 2 square meter surface and the situation was only remediated a year later.
- Every spring, there is condensation in the attic and the mineral insulation wool is blackened. The wood structure shows traces of water damage.

Possible examples of extensive water damage history:
- The flat roof started to leak during the 1998 ice storm and there was one inch of standing water in the living room and kitchen for several days before the occupants returned to their home.
- During a fire, firefighters watered the structures extensively and the inside of the house stayed drenched with water during the entire winter.
- Sewage backups occur every spring in the basement and the wet walls were not opened; the owner washed the carpet with bleach and uses a deodorant to mask the odor of rot.

c) Very unhealthy homes (a sub-group inside the unhealthy homes group)

Water activity: one week and more, or chronic
Water damage history: extensive

Selection bias:

Section 2.1 describes the selection criteria that were determined to choose healthy homes, so as to avoid useless inspections. These criteria have eliminated from the start many risk factors, such as: no basement apartments, no basement carpets, no houses with both pets and carpets. Because of that selection, the results in sections 3.1.3 to 3.1.8 are only informative and have no statistical value.

3.1.3   Housing types

HOME TYPES	HEALTHY	UNHEALTHY	VERY UNHEALTHY
Bungalow	21 (31%)	31 (21%)	17 (21%)
Cottage	16 (23.5%)	47 (32%)	28 (34%)
Semi-detached	8 (12%)	20 (14%)	7 (8.5%)
Town house	3 (4.5%0	5 (3.5%)	5 (6%)
Main floor apartment	4 (6%)	8 (5.5%)	6 (7%)
Second floor apartment	16 (23.5%)	22 (15%)	12 (15%)
Basement apartment	0 (0%)	9 (6%)	4 (5%)
Total	68	145	79

Table 1.   Housing types

3.1.4 Home age

AGE OF DWELLING	HEALTHY	UNHEALTHY	VERY UNHEALTHY
Less than 10 years	13 (19%)	13 (9%)	9 (11%)
10 to 50 years	29 (43%)	86 (59%)	45 (55%)
50 years and more	24 (35%)	32 (22%)	18 (22%)
Unknown	2 (3%)	14 (10%)	10 (12%)
Total	68	145	82

Table 2.   <u>Home age</u>

3.1.5 Heating types

HOME HEATING	HEALTHY	UNHEALTHY	VERY UNHEALTHY
Electrical	48 (70.5%)	87 (61%)	50 (61%)
Forced air	7 (10%)	19 (13%)	10 (12%)
Hot water	4 (6%)	10 (7%)	4 (5%)
Mixed	9 (13%)	25 (17%)	14 (17%)
Total	68	143	82

Table 3.   <u>Heating types</u>

3.1.6 Foundations and crawl spaces

FOUNDATIONS	HEALTHY	UNHEALTHY	VERY UNHEALTHY
No foundation	0 (0%)	17 (12%)	8 (10%)
Concrete crawl space	6 (9%)	0 (0%)	0 (0%)
Concrete slab under floor	3 (4%)	0 (0%)	0 (0%)
Concrete	54 (79%)	110 (76%)	60 (73%)
Concrete blocks	2 (3%)	9 (6%)	8 (10%)
Stone	1 (1.5%)	5 (3.5%)	2 (2.5%)
Stone and concrete	1 (1.5%)	3 (2%)	3 (3.5%)
Total	68	145	82

Table 4.   <u>Foundation types</u>

EARTH CRAWL SPACES	HEALTHY	UNHEALTHY	VERY UNHEALTHY
None	55 (81%)	123 (85%)	71 (86%)
Partial	3 (4.5%)	8 (5.5%)	4 (5%)
Complete	8 (12%)	13 (9%)	6 (7%)
Total	68	145	82

Table 5.   <u>Earth crawl spaces</u>

3.1.7   Carpeting

CARPETING	HEALTHY	UNHEALTHY	VERY UNHEALTHY
No carpet	35 (51%)	9 (6%)	26 (32%)
Carpet (minority of surfaces)	18 (26.5%)	28 (19%)	14 (17%)
Basement carpet (minority of surfaces)	11 (16%)	56 (39%)	14 (17%)
Carpet (majority of surfaces)	4 (6%)	41 (28%)	10 (12%)
Basement carpets (majority of surfaces)	1 (1.5%)	11 (7.5%)	10 (12%)
Carpets all over	0 (0%)	2 (1%)	0 (0%)
Total	68	145	82

Table 6.   Presence of carpets in homes

3.1.8   Pets

PRESENCE OF PETS IN HOMES	HEALTHY	UNHEALTHY	VERY UNHEALTHY
None	40 (59%)	104 (72%)	63 (77%)
One	23 (34%)	26 (18%)	10 (12%)
Two and more	5 (7.5%)	15 (10%)	9 (11%)
Total	68	145	82

Table 7.   Presence of pets in homes

3.1.9   Inspection season

INSPECTION SEASON	HEALTHY	UNHEALTHY	VERY UNHEALTHY
Winter	11 (16%)	28 (19%)	9 (11%)
Spring	15 (22%)	22 (15%)	11 (13%)
Summer	6 (9%)	45 (31%)	31 (38%)
Fall	36 (53%)	50 (34%)	31 (38%)
Total	68	145	82

Table 8.   Season of inspection

Table 8 shows that more than half of the healthy homes were inspected during the fall period compared to one third of the unhealthy homes. The influence of season on the microbial contents of house dust is taken into account in Table 14. It shows, using double criteria ANOVA, that the contamination level in a home will determine the fungal contents of its dust, whatever the season.

3.1.10 Surfaces with visible mold

VISIBLE MOLD	HEALTHY	UNHEALTHY	VERY UNHEALTHY
None	52 (76%)	11 (7.5%)	5 (6%)
Less than 1 m^2	16 (24%) *	42 (29%)	26 (32%)
1 to 3 m^2	0 (0%)	63 (43.5%)	20 (24%)
3 to 10 m^2	0 (0%)	1 (0.5%)	20 (24%)
More than 10 m^2	0 (0%)	27 (19%)	10(12%)
Total	68	145	82

Table 9.   Surfaces with visible mold in homes
* a few cm ^{2}

Even if it is obvious that unhealthy homes show many more surfaces with visible mold than healthy homes, most of these surfaces were visible without invasive inspections. In several cases, it is only the "tip of the iceberg", because most fungal growth occurs on the site of abnormal water activity, often in hidden inside structures. Realistically, opening of walls, ceilings or floors is not always allowed during inspection. Dust sampling is especially useful in these situations.

3.1.11 Surfaces with traces of water damage

TRACES OF WATER DAMAGE	HEALTHY	UNHEALTHY	VERY UNHEALTHY
None	47 (69%)	9 (6%)	3 (3.5%)
Less than 1 m^2	17 (25%)	28 (19%)	10 (12%)
1 to 3 m^2	4 (6%)	56 (38%)	24 (29%)
3 to 10 m^2	0 (0%)	41 (28%)	35 (24%)
More than 10 m^2	0 (0%)	11 (7.5%)	10 (12%)
Total	68	145	82

Table 11.   Surfaces with traces of water damage in homes

3.2   Fungal analysis of dust

3.2.1   Fungal counts in dust and contamination level in homes

HOME CONTAMINATION LEVEL	SAMPLE NUMBERS	MEAN FUNGAL COUNTS IN DUST cfus/gram of dust	STANDARD ERROR
Healthy	71	74 366 *	143 396
Unhealthy	184	447 837 *	89 138
Very unhealthy	95	548 179 *	124 053

cfus: colony forming units (viable fungal spores)

* Significant difference as shown by Wilcoxon/Kruskal-Wallis testing ($p < 0.0001$)

Table 13.   Fungal counts in dust and contamination in homes

These results show that the microbial content of house dust is a good indicator of contamination level and represents the "microbiological memory" of houses. Dust from unhealthy homes can contain up to seven times more mold than that of their healthy counterparts.

These results confirm the Ontario Wallaceburg study (Miller and al, 1999): fungal counts from dust sampled in 20 out of 400 homes, with the most extensive water damage episodes, were 10 times higher than fungal counts from the 20 homes without excessive water activity. We must however make it clear that results in Table 13 include a much higher number of samples, which allowed us to validate them statistically. Other publications also link contamination levels in buildings and fungal contents in carpet dust (Hodgson and Scott, 1999) and in dust deposited on smooth surfaces (Mainville and al, 1999).

3.2.2   Fungal counts in dust in relation to season

Wilcoxon/Kruskal-Wallis rank testing showed no statistically significant difference between fungal counts in relation to the season of sampling, neither for healthy nor for unhealthy and very unhealthy homes.

3.2.3   Fungal counts in dust: combined effects of season and contamination levels

INFLUENCE FACTOR	TWO CRITERIA ANOVA p value
Contamination level only	0.0108 *
Season only	>0.05 **
Contamination level and season	>0.05 **

*   Significant influence of contamination level on the fungal counts in home dust
**   No influence of season, or season and home contamination level combined

Table 14.   Combined influence of season and level of contamination on fungal counts
                in home dust

**These results confirm section 3.2.2: they show that the season of inspection does not
have any influence on fungal counts in house dust and that only the contamination
level matters.**

3.2.4   Overall fungal distribution in the dust from inspected homes

Cladosporium and *Penicillium* are by far the most frequent fungal genera in the dust from
inspected homes for this study and from the NAK data bank. But around thirty other
fungal genera can mix with *Cladosporium* and *Penicillium* in the dust, the most frequent
being: *Alternaria*, *Aspergillus*, *Chaetomium*, *Epicoccum*, *Mucor*, *Paecilomyces*,
Rhizopus, *Trichoderma* and *Ulocladium*.

Occasionally the following fungal genera could also be found:
Botrytis, *Dreschlera*, *Neurospora*, *Nigrospora*, *Phoma*.

The following fungal genera were also found, but rarely, in the house dust we sampled:
Acremonium, *Curvularia*, *Eidamella*, *Eurotium*, *Fusarium*, *Geotrichum*, *Gliocladium*,
Leptographium, *Monodictys*, *Rhinocladiella*, *Stachybotrys* and *Verticillium*.

Note that the culture medium used influences the variety of fungal genera found. The
MEA medium used for culturing has characteristics that allow growth of a good number
of fungal species with moderate to high water requirements.

3.2.5   Non phylloplane vs phylloplane ratios in dust related to home contamination
levels

HOME CONTAMINATION LEVELS	SAMPLES	PHYLLOPLANE (Clado + Alt) **mean % (standard error)**	NON PHYLLOPLANE (Pen +Asp) **mean % (standard error)**	RATIO NON PHYLLOPLANE/ PHYLLOPLANE
Healthy	67	45% * (3.7)	31.6% ** (4.05)	0.70
Unhealthy	189	30% * (2.2)	45.5% ** (2.41)	1.51

*   Significant difference in the % of the most frequent phylloplane fungi
(*Cladosporium* and *Alternaria*) in the dust of homes in relation to their
contamination levels: ANOVA (p=0.00040)

**   Significant difference in the % of the most frequent non phylloplane fungi
(*Penicillium* and *Aspergillus*) in the dust of homes in relation to their
contamination levels: ANOVA (p=0.012)

Table 15.   Non phylloplane vs phylloplane ratios in dust related to home
contamination levels

***Cladosporium* and *Alternaria* are the phylloplane, and *Penicillium* and *Aspergillus*
the non phylloplane genera found most frequently overall in the dust of the
inspected homes, whatever their contamination levels. On the other hand, Table 15
shows that phylloplanes predominate more often in healthy homes, while non
phylloplanes predominate in unhealthy homes.**

These results suggest the possibility of assigning to residential dust samples a
characteristic ratio defined as non phylloplane divided by phylloplane fungal percentage.
A ratio of one or less indicates a healthy home and the more the ratio rises, the more
unhealthy the house.

3.3 Dust contents in bacteria

3.3.1 Bacterial counts in dust and home contamination levels

HOME CONTAMINATION LEVEL	SAMPLE NUMBERS	MEAN BACTERIAL COUNTS cfus/gram of dust	STANDARD ERROR
Healthy	68	678 088 *	522 444
Unhealthy	171	1 414 664 *	329 455
Very unhealthy	95	1 504 579 *	442 010

* No significant difference in bacterial counts from healthy and unhealthy homes, and from unhealthy and very unhealthy homes using the Wilcoxon/Kruskal-Wallis test ($p > 0.05$)

Table 17.   Bacterial counts in dust and home contamination levels

Mean counts in bacteria are more than twice as high in unhealthy homes compared to healthy homes, but the standard deviation is too high to confer statistical significance to these data.

Many factors can explain these findings:

- presence of pets
- sewage backups
- bird nests, bats, rodents
- sump pumps with standing water
- cold water humidifiers without proper maintenance
- season (see Table 18)

In the data bank, there are not enough homes with each of these separate characteristics to allow us to perform a statistical analysis of their dust bacterial counts. NAK's inspection findings indicate however that all these factors do have an influence on counts of bacteria in house dust.

3.3.2 Counts of bacteria in dust in relation to season

In unhealthy and very unhealthy homes, no statistically significant difference was found between dust counts and the season of sampling, according to the Wilcoxon/Kruskal-Wallis rank test. However, in healthy homes, there is a connection between the sampling season and the bacterial counts in dust, as shown in Table 18.

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SEASON	SAMPLE NUMBERS	MEAN BACTERIAL COUNTS cfus/gram of dust	STANDARD ERROR
Winter	12	43 542 *	60 042
Spring	15	244 833	474 111
Summer	7	578 571	694 028
Fall	32	1 120 703 *	324 602

*   Significant difference in mean bacterial counts from healthy homes between winter and fall as shown with the Wilcoxon/Kruskal-Wallis test (p=0.0216)

Tableau 18.   Dust bacterial counts in healthy homes and season

3.4   Amounts of yeasts in dust and season

SEASON :	FALL September, October, November	SPRING March, April, May
Number of samples with a large or very large amount of yeasts	30.3% *	18.8 % *

*   Significant difference between the fall and spring number of home dust samples with a large or very large amount of yeasts : X^2 (p=0.010)

Table 19.   Influence of season on the yeast levels in home dust

Regardless of the home contamination level, yeasts tend to be more numerous during the fall season. However, these results are preliminary because of the limitations of the evaluation method used to measure yeasts in the samples. In fact, because of the very fast growth rate of yeasts in culture, it became impossible for us to count individual colonies, so yeast numbers were evaluated from the surface they occupied on the culture dish. This is a very approximate and rather inaccurate method (see section 2.4.3 of Chapter 2).

CHAPTER 4.   CONCLUSIONS AND OUTLOOK

This comparative study between the microbial contents of dust from healthy and unhealthy homes confirms the reliability of house dust sampling as a complementary diagnosis tool for the assessment of microbial contamination indoors.

The results are as follows:

a)   Mean fungal counts in the dust from unhealthy homes were 7 times higher than counts in healthy homes; this difference is statistically significant.
b)   Mean fungal counts in homes are not influenced by the season of inspection.
c)   Phylloplane fungi dominate in healthy homes while non phylloplane fungi dominate in unhealthy homes; this difference is statistically significant.
d)   Mean bacterial counts are twice as high in unhealthy homes than in healthy homes, but deviation from the mean is too great for the difference to be statistically significant.
e)   During the fall season, bacteria are more numerous in healthy homes and yeasts are more numerous in all the homes.

4.1   How to use house dust sampling

All the experts in the field of indoor microbial contamination know that it is IMPOSSIBLE to rely only on lab analysis for a thorough and reliable diagnosis.

In fact, four elements must be considered to obtain a complete microbial contamination diagnosis:
-       evaluation of the occupants' health complaints: symptoms enhanced when at home and decreased when outside;
-       abnormal water activity history in the house;
-       detailed inspection of the premises, including ventilation: looking for water in structures using a moisture detector, looking for visible mold and measuring the moldy surfaces, taking photos supporting each case, etc.; and
-       laboratory sampling if necessary.

Laboratory sampling must be adapted to the situation, as the following examples show:
-       in the presence of visible mold on a surface: surface sampling, air sampling nearby to measure potential exposure of occupants, in particular for certain court cases; and
-       in the case of water damage without extensive visible mold: dust sampling to evaluate the overall contamination level of a floor, a room, a ventilation system, a carpet, etc.

No sampling method is perfect.

Air samples measure the microbial contents of air only at the precise time of sampling, with frequent risks of false negative results.

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Surface samples on adhesive tape do not always allow identification of the mold nor do they tell us if the mold is growing.

Finally, in approximately 10% of cases, we observed that dust microbial contents do not correspond with the inspection data on the field. *Stachybotrys chartarum conidia*, for example, cannot survive very long away from a very humid environment such as wet materials inside a wall. When spores from such a humid growth environment migrate into the dryer dust found in inhabited rooms, the vast majority are already dead and will not grow in culture. This situation is associated to risks of false negative results, with counts as low as 2,500 cfus per gram in houses where visible moldy surfaces sometimes measured more than a square meter. There is also a risk of false positives, with very high counts in the absence of a contamination source. This type of situation can result from residual contaminated dust that was not properly removed during remediation work, when the contamination source was eliminated.

Therefore, sound judgment is to be used on a case by case basis to choose the most appropriate sampling method(s) and avoid unnecessary sampling.

4.2   Why use house dust sampling

According to scientific literature, approximately 15% of the population is at risk of experiencing health problems in the presence of microbial contaminants indoors. Their immune system is either not working well, weakened or immature. These include:
-        the elderly and the very young;
-        people with immune system disorders, AIDS patients for example;
-        people taking immunosuppressive medication (cortisone, cyclosporin, chemotherapy);
-        patients with chronic illnesses;
-        asthma patients, atopic persons;
-        environmentally hypersensitive persons, etc.

Where there is indoor microbial contamination, these people are often helpless and unwilling to pay the costs for a complete microbial contamination inspection. Generally, their health problems prompt them to ask for help, especially if their symptoms are new or enhanced since they moved into their house. Unfortunately, water damage and fungal growth problems in homes are often hidden for several reasons:
-        damage has been covered up by the previous owner or before the new tenants moved in (new gypsum, fresh paint); and
-        water damage inside structures occurs and the occupants are not aware of it: fungal growth is hidden beneath floors, inside ceilings or walls, etc.

Microbial analysis of house dust, where samples are taken by the occupants themselves, is an affordable way to obtain indicative information in these situations. Depending on the lab results, the occupants would be better informed to make the decision whether to proceed to have an inspection and do remedial work.

On the other hand, inadequate remedial work in contaminated homes can make the problem worse. Unfortunately, most building contractors do not know the proper procedures for microbial decontamination of homes. Without good microbial diagnosis and sampling, many contractors will not even know the nature of the problem when doing remedial work and, sooner or later, it will have to be done again especially when dealing with hidden mold.

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