Building Forensics Ltd

Mould Sampling Alternatives

Rev 1 08/19

The following are examples of different mould and water damaged building sampling techniques we use.
You don't have to be concerned as to which is your best option, because we will explain the most suitable which will fit with budget and usefulness.

These sampling techniques have benefits and shortfalls and the decision on suitability and choice will revolve around the result objectives which will vary from client to client.

We almost NEVER sample or test visible mould. It simply shouldn't be there, and it has already identified a possible health risk and should be remediated and removed. We do however specialise in identifying the invisible or hidden mould and biotoxin hazard which is probably a constant exposure source and inhalation risk.

The following lab reports are accompanied by explanation as to why the differing techniques are used.

You can see from Tables 1-3 why different forms of sampling are required

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Table 1 This shows the type of Mycotoxins which can be produced by toxic moulds and which organs they challenge

Table 1

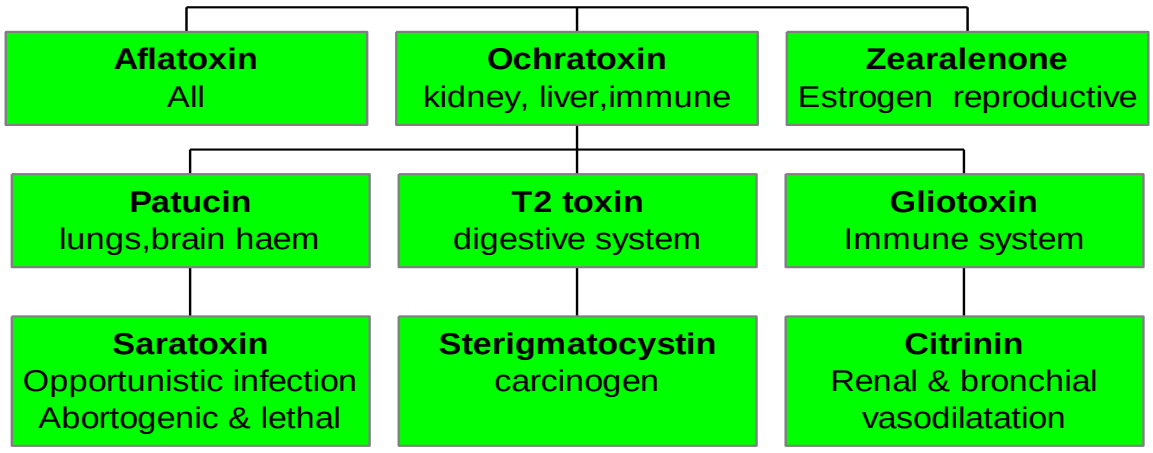


Table 2

This shows the various mycotoxins produced by individual species of mould

	Alternaria	*Chaetomium*	*Stachybotrys*	*Trichoderma*	*Asp. flavus*	*Asp ochraceous*	*Asp. versicolor*	*Asp. niger*	*Asp. fumigatus*	*Asp. nidulans*	*Pen. viridicatum*	*Pen. crustosum*	*Pen. rugulosum*
Altenuic acid	X												
Alternariol	X												
Gliotoxin	X				X			X	X				
Cochliodinol		X											
Chaetoglobosin		X											
Roridin E			X										
Satratoxins F,G,H			X	X									
Trichodermin				X									
Trichoverrins			X										
Trichoverrols			X										
Verrucarin			X										
Verruculogen			X						X				
Aflatoxin					X								
Destructin B						X							
Ochratoxin						X		X			X		
Penicillic acid						X							
Cyclopiazonic acid							X						
Sterigmatocystin					X		X			X			X
Viopurpurin											X		
Penitrem												X	

My favourite brave couple in their 80s. They couldn't enter their home after a leak from the flat above developed Bio toxin elements including toxic moulds. They had no alternative but to wear special personal protective equipment to allow them to enter their home to make important decisions.

Table 3

The harmful effects of different mycotoxins are listed below however the synergistic effects of two or more mycotoxins is currently unknown

Aflatoxin M1 (AFM1)
This is the main metabolite of aflatoxin B1, which is a mycotoxin produced by the mould species Aspergillus. Aflatoxins are some of the most carcinogenic substances in the environment. Aflatoxin can lead to liver damage, cancer, mental impairment, abdominal pain, hemorrhaging, coma, and death.

Ochratoxin A
(OTA) is a nephrotoxic, immunotoxic, and carcinogenic mycotoxin. Exposure to OTA can also come from inhalation exposure in water-damaged buildings. OTA can lead to kidney disease and adverse neurological effects.

Sterigmatocystin
(STC) is a mycotoxin that is closely related to aflatoxin. STC is produced from several species of mould such as Aspergillus, Penicillium, and Bipolaris. STC is considered to be carcinogenic, particularly in the cells of the GI tract and liver. STC has been found in the dust from damp carpets.

Zearalenone
(ZEA) is mycotoxin that is produced by the mould species Fusarium, and has been shown to be hepatotoxic, haematotoxic, immunotoxic, and genotoxic.

Roridin E
(ROE) is a macrocyclic trichothecene produced by the mould species Fusarium, Myrothecium, and Stachybotrys (i.e. black mold). Trichothecenes are frequently found in buildings with water damage. Trichothecenes are considered extremely toxic and have been used as biological warfare agents. Even low levels of exposure to macrocyclic trichothecenes can cause severe neurological damage, immunosuppression, endocrine disruption, cardiovascular problems, and gastrointestinal distress

Verrucarin A
(VRA) is a macrocyclic trichothecene mycotoxin produced from Stachybotrys, Fusarium, and Myrothecium. Trichothecenes are frequently found in buildings with water damage but can also be found in contaminated grain

Testing cavity walls, usually after builders have painted over mould

In this brand-new building, mould developed on walls even before painted. The client asked to test cavities and recommend correct protocols. The cavity was completely contaminated, and all walls had to be removed back to framing

Sample type 1 QPCR--DNA airborne

In this sample type we sample airborne particles of mould and fragments to identify DNA of spores and hyphae. This can assist in the identification of mycotoxin produces

Sample ID: 5
Sample Description: Chris S
Collection Volume: 150 L
Reporting Limit: 7 Spores/Cubic Meter

Species Identification	Spores/m^3 of Air Inside	Relative Abundance (%) of Detected Species
Acremonium strictum	ND	0.00
Alternaria alternata	ND	0.00
Anigr*	ND	0.00
Aspergillus flavus/oryzae	2,301	82.98
Aspergillus fumigatus, Neosartorya fischeri	ND	0.00
Aspergillus ochraceus/ostianus	ND	0.00
Aspergillus penicillioides	ND	0.00
Aspergillus restrictus/caesillus/conicus	ND	0.00
Aspergillus sclerotiorum	ND	0.00
Aspergillus sydowii	ND	0.00
Aspergillus unguis	ND	0.00
Aspergillus ustus	ND	0.00
Aspergillus versicolor	ND	0.00
Aureobasidium pullulans	7	0.25
Chaetomium globosum	ND	0.00
Cladosporium cladosporioides svar. 1	7	0.25
Cladosporium cladosporioides svar. 2	ND	0.00
Cladosporium herbarum	ND	0.00
Cladosporium sphaerospermum	ND	0.00
Eamst*	ND	0.00
Epicoccum nigrum	ND	0.00
Muc1*	ND	0.00
Paecilomyces variotii	ND	0.00
PenGrp2*	35	1.26
Penicillium brevicompactum/stoloniferum	ND	0.00
Penicillium chrysogenum	ND	0.00
Penicillium corylophilum	ND	0.00
Penicillium purpurogenum	ND	0.00
Penicillium variabile	ND	0.00
Pspin2*	ND	0.00
Rhizopus stolonifer	ND	0.00
Scopulariopsis brevicaulis/fusca	ND	0.00
Scopulariopsis chartarum	ND	0.00
Stachybotrys chartarum	ND	0.00
Trichoderma viride/atroviride/koningii	423	15.25
Wallemia sebi	ND	0.00
Total Spores:	2,773	

Sample 2
Airborne Genus sampling

This sampling is for airborne spore counts. It helps us identify risk and hazard areas. We usually undertake 4 samples including one outside for comparison or control purposes. This is not precise but is reasonably priced tool used internationally as a base line**.**

Location:	1: Outside				2: Bedroom				3: Lounge				4: Bathroom			
Comments (see below)	None				A				None				None			
Lab ID-Version‡:	10483128-1				10483129-1				10483130-1				10483131-1			
Analysis Date:	07/17/2019				07/17/2019				07/17/2019				07/17/2019			
Sample volume (liters)	30				30				30				30			
Background debris (1-4+)††	1+				2+				3+				4+			
	raw ct.	Count/m3	DL/m3*	%	raw ct.	Count/m3	DL/m3*	%	raw ct.	Count/m3	DL/m3*	%	raw ct	Count/m3	DL/m3*	%
Hyphal fragments	3	100	33	n/a					9	300	33	n/a	15	500	33	n/a
Pollen	1	33	33	n/a	3	100	33	n/a	3	100	33	n/a	15	500	33	n/a
§ TOTAL FUNGAL SPORES	17	1,900	n/a	100	284	110,000	n/a	100	172	180,000	n/a	100	213	230,000	n/a	100
Alternaria	1	33	33	2					1	33	33	< 1	1	33	33	< 1
Ascospores	1	130	130	7	2	270	130	< 1	5	670	130	< 1	2	270	130	< 1
Basidiospores	4	530	130	29	1	130	130	< 1	2	270	130	< 1	3	400	130	< 1
Chaetomium					1	33	33	< 1	12	400	33	< 1	18	600	33	< 1
Cladosporium	8	1,100	130	57	7	930	130	1	13	1,700	130	1	12	1,600	130	1
Epicoccum													1	33	33	< 1
Other brown									1	33	33	< 1	1	33	33	< 1
Penicillium/Aspergillus types					272	110,000	390	99	135	180,000	1,300	98	172	230,000	1,300	99
Polythrincium	2	67	33	4												
Rusts	1	33	33	2					1	33	33	< 1				
Smuts, Periconia, Myxomycetes					1	33	33	< 1	2	67	33	< 1	3	100	33	< 1
Stachybotrys																
Zygomycetes																

Sample 3

Airborne sampling of genus with a mould score against other areas for comparison This is a pure calculation and report on comparison of air sampling against other areas. This is free when used with some tests

Fungi Identified	Indoor sample spores/m3 <100 1K 10K >100K	Raw count	Spores/ m3	MoldSCORE‡ 100 200 300	Score
Generally able to grow indoors*					
Alternaria		ND	< 33		100
Bipolaris/Drechslera group		ND	< 33		100
Chaetomium		ND	< 33		100
Cladosporium		22	2,900		214
Curvularia		ND	< 33		100
Nigrospora		ND	< 33		100
Penicillium/Aspergillus types†		91	30,000		300
Stachybotrys		ND	< 33		100
Torula		ND	< 33		100
Seldom found growing indoors**					
Ascospores		1	130		100
Basidiospores		1	130		100
Rusts		2	67		127
Smuts, Periconia, Myxomycetes		ND	< 33		100
Total			**33,600**	**Final Mold SCORE**	**300**

Sample 4

Sampling of settled dust fragments, spores and Hyphae to identify species and potential mycotoxin producers. Note referred to as ERMI but we analyse species and risk/hazard factors as the ERMI score has no health hazard relationship.

Group 1; Water Damage Molds

Species	SE/mg
Aspergillus flavus/oryzae	4
Aspergillus fumigatus	45
Aspergillus niger	11
Aspergillus ochraceus	ND
Aspergillus penicillioides	18
Aspergillus restrictus	10
Aspergillus sclerotiorum	ND
Aspergillus sydowii	ND
Aspergillus unguis	ND
Aspergillus versicolor	116
Aureobasidium pullulans	1,764
Chaetomium globosum	3
Cladosporium sphaerospermum	4
Eurotium (Asp.) amstelodami	597
Paecilomyces variotii	ND
Penicillium brevicompactum	203
Penicillium corylophilum	42
Penicillium crustosum	43
Penicillium purpurogenum	ND
Penicillium Spinulosum	24
Penicillium variabile	ND
Scopulariopsis brevicaulis/fusca	3
Scopulariopsis chartarum	3
Stachybotrys chartarum	ND
Trichoderma viride	32
Wallemia sebi	11

Sum of Logs 25.2

Group 2; Common Indoor Molds

Species	SE/mg
Alternaria alternata	3
Acremonium strictum	27
Aspergillus ustus	48
Cladosporium cladosporioides1	1,336
Cladosporium cladosporioides2	52
Cladosporium herbarum	1,081
Epicoccum nigrum	954
Mucor amphibiorum	44
Penicillium chrysogenum	26
Rhizopus stolonifer	ND

Sum of Logs 17.4

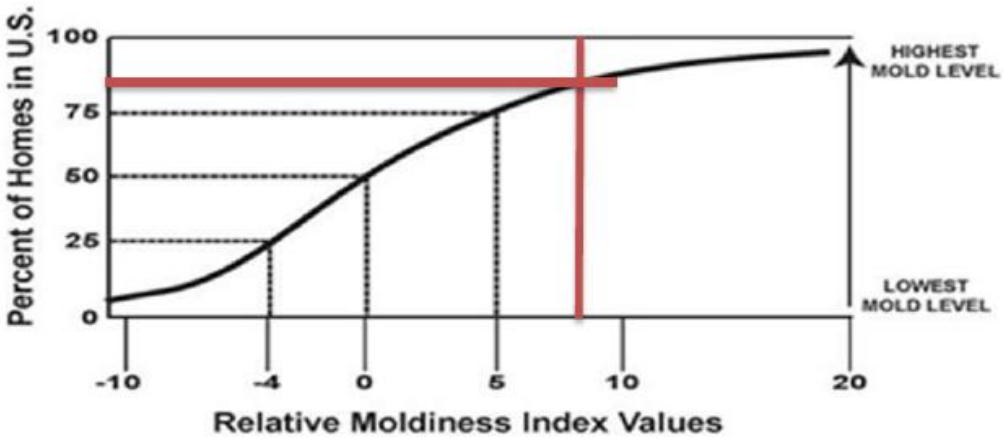
SE = Spore Equivalents
SE/mg = SE/milligrams of sample
Logs = Logarithms
ND = None Detected

Sample Size 5.0 mg

ERMI Results= (G1-G2) **7.8**

Environmental Relative Moldiness Index (ERMI)	7.8

Interpretation	Q4



Sampling 5

In this report we calculate the risk and hazard specifically for CIRS sufferers, (Mould illness) Chronic inflammatory Response. The risk levels are supported by research from over 1000 patient reports.

Species	Spore E./mg	Weighting
Aspergillus penicillioides	18	4
Aspergillus versicolor	116	6
Chaetomium globosum	3	0
Stachybotrys chartarum	ND	0
Wallemia sebi	11	0
HERTSMI-2 Score =		**10**

Colour coded interpretation	
If 10 or below	In only 17% of cases, re-occupancy of building following mould remediation has led to relapse of CIRS or WDB symptoms
If between 11 to 15	Borderline further remediation and assessment may be required
If greater than 15	Re occupation is ill advised until further remediation and re assessment are conclusive

Sampling 6

In this table we provide closely related species to the ERMI panel of species. This can be extremely useful when mould specialist doctors identify species in blood or urine tests. This is free when used with other tests

As reported	Includes
Eurotium (Asp.) amstelodami	E. chevalieri, E. herbariorum, E. rubrum and E. repens.
Penicillium spinulosum	P. glabrum, P. lividum, P. pupurescens, and P. thomii.
Trichoderma viride	T. koningii and T. atroviride.
Aspergillus restrictus	A. caesillus and A. conicus.
Mucor amphibiorum	M. circinelloides, M. hiemalis, M. indicus, M. mucedo, M. racemosus, M. ramosissimus.
Rhizopus zygosporus	R. homothalicus, R. microsporus, R. oligosporus, R. oryzae.
Penicillium crustosum	P. camembertii, P. commune, P. echinulatum, P. solitum.
Aspergillus niger	Know called Aspergillus basiliensis
Scopulariopsis brevicaulis/fusca	Has been renamed as species of Microascus[10]
Wallemia sebi	W. mellicola, W. canadensis[11]

Sample 7

In this technique we pick up visible mould for identification to genus level. We do not normally recommend sampling visible mould but sometimes the information can assist decision making. The sampling points are evaluated from the basic survey and where high-risk areas are identified

QUANTITATIVE SPORE COUNT REPORT

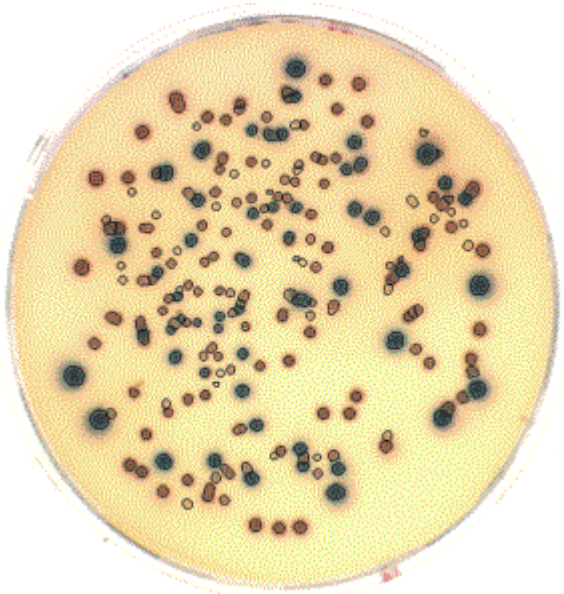
Location:	6: Bedroom				7: Carpet				8: Loft			
Comments (see below)	None				None				None			
Sample type	Tape sample				Tape sample				Tape sample			
Lab ID-Version‡:	10430649-1				10426309-1				10426310-1			
Analysis Date:	07/01/2019				07/01/2019				07/01/2019			
Background debris (1-4+)	< 1+				1+				1+			
Sample size	1 cm2				1 cm2				1 cm2			
Reporting unit	1 cm2				1 cm2				1 cm2			
	Count	Count/sample	Count/unit	%	Count	Count/sample	Count/unit	%	Count	Count/sample	Count/unit	%
Hyphal fragments	3	50	50	n/a	2	33	33	n/a	9	150	150	n/a
§ TOTAL FUNGAL SPORES	260	4,400	4,400	100	5	83	83	100	110	1,800	1,800	100
Cladosporium	261	4,400	4,400	99					16	270	270	14
Curvularia												
Epicoccum												
Fusarium												
Myrothecium												
Nigrospora												
Other colorless												
Penicillium									94	1,600	1,600	86
Penicillium/Aspergillus types	2	33	33	1	4	67	67	80				
Pithomyces												
Rusts												
Smuts, Periconia, Myxomycetes												
Stachybotrys					1	17	17	20				
Stemphylium												
Torula												
Ulocladium												
Zygomycetes												

Sampling technique 8

The World Health Organization (WHO state this form of sampling has serious limitations.
Some companies report on Colony Forming Units (CFUs) but this provides almost no useful information and can be extremely misleading.

A major issue is that CFUs are obviously only reporting on spores that grow on that specific agar within ten days. Many moulds take weeks to grow and some require different agars too. Of course, fragments of spores, hyphae and dormant spores cannot grow and according to WHO these particles are possibly 40 times more hazardous due to size and inhalation into lower respiratory system directly into blood stream.

We do NOT undertake this form of sampling or analysis as it provides no useful information to our clients. When British Standards on this type of sampling are followed the lab fees alone can be three or four times as costly as some of the techniques, we use



We Do not use these "serioulsy limited" (WHO) sampling protocols of swabs or culture based sampling (CFUs). The results cannot be used for health risk or hazard assessments

Bacteria testing

Mould is only one of the contaminates likley to lead to building related illness (CIRS etc) New research shows certain types of bacteria can have even more impact than mould.We can now include this additional testing for those with specialist doctors who will recognise trhe significance.

Gram Positive testing

Table 1

Summary of total bacteria's species

Bacteria	Types
Totals	511
Pathogen	56

Box 1

Actinomycetales	Types
Totals	154
Pathogenic	18

Box 2

Selected actinomycetales found in water damaged buildings

Mycobacteria	Types
Totals	2
Pathogenic	0

Streptomyces	Types
Totals	4
Pathogenic	0

Box 3

Non Actinomycetales	Types
Totals	357
Pathogenic	38

Box 4

The report has a score of 19 out of 40 water damage species and is a health hazard

Color-coded interpretation	
If 9 or below	Indicative of a Healthy Building
If between 10 to 15	Further investigation needed
If greater than 15	Suggestive of Building Related Illness.

Table 2

Summary of bacteria's Order

Orders Detected	Abundance BE/mg	Families	Abundance	Diversity	Pathogenic
Actinomycetales	1,802,371	31	27.4 %	23.5 %	21
Bacillales	636,429	8	9.7 %	6.1 %	11
Pseudomonadales	611,179	2	9.3 %	1.5 %	14
Sphingomonadales	555,092	2	8.4 %	1.5 %	1
Rhodobacterales	518,664	1	7.9 %	0.8 %	0
Rhizobiales	439,037	11	6.7 %	8.3 %	2
Stigonematales	268,735	1	4.1 %	0.8 %	0
Nostocales	216,855	2	3.3 %	1.5 %	0
Burkholderiales	215,014	4	3.3 %	3.0 %	3
Sphingobacteriales	170,499	3	2.6 %	2.3 %	1

Gram Negative testing

There is an elevated presence of gram-negative bacteria which is a single or synergistic health hazard

Reference Number	Date Sampled	Locations	Result EU/mg
193836-3	Jun 24, 2019	Not Given	203