Mould Analysis

Introduction

Moulds are a type of fungus distinct from yeasts and mushrooms. Unlike single-celled yeasts or the large fruiting bodies of mushrooms, moulds form thread-like structures and grow in speckled patches, typically in moist environments. They play a vital ecological role by decomposing dead organic material but can also cause health problems and structural damage indoors.

Moulds reproduce by releasing tiny spores that spread through air, water, or animals. These spores can grow into new mould colonies when they land on suitable surfaces with moisture and nutrients. Common indoor moulds include *Alternaria*, *Aspergillus*, *Cladosporium*, and *Penicillium*.

Mould is found nearly everywhere and is visible in various colours depending on its species and age. It thrives in damp indoor spaces, damaging buildings, spoiling food, and causing health issues like allergies, asthma, and infections. Visual identification is unreliable, and professional testing is needed to determine mould types and assess water damage.

Analytical Methods

Spore Trap Analysis

Spore trap mould testing is a method used to detect and identify airborne mould spores in indoor and outdoor environments. It involves collecting airborne particles—including mould spores—on a sticky surface in a spore trap cassette as air is drawn through it by a pump.

The method of analysis is performed in accordance with the following standard - ASTM D7391 – 20 Standard Test Method for Categorization and Quantification of Airborne Fungal Structures in an Inertial Impaction Sample by Optical Microscopy.

The method is for slit impaction air samples where a known volume of air (at the rates described by the manufacturer) has been drawn through the filter. The slide impacted sample is mounted onto a clean microscope slide with lacto-cotton blue stain, followed by a scan at X100 – X200 magnification to give the analyst an overview of sample deposition and the diversity of the spores present on the slide. The slide is then analysed at X200 – X600 magnification by identifying and counting spores in 25%-100% of the sample deposition area. Spores occurring in chains are counted individually. Raw counts at each magnification are converted to fungal structures/m3 of air based on the sample air volume supplied. Spores lacking distinguishing characteristics are reported as "Unidentified/Miscellaneous spores". Where the analyst is able to identify the group to which the spores belong but not the mould they belong to, the spores may be recorded as "Basidiospores or Ascospores". Spores of *Aspergillus* spp. and *Penicillium* spp. and others i.e. *Acremonium* spp., *Paecilomyces* spp., *Scopulariopsis* spp. are difficult to distinguish and are reported as *Aspergillus/Penicillium* -Like and differing morphology is noted in the comments if encountered.

Results are presented in both fungal structures per sample (fs/sample) and fungal structures per cubic metre (fs/$m^{3)}$. The range of results depend on the amount of particulate matter, the size of the spores and the percentage of the sample counted on the spore trap sample trace as well as the volume of air through the filter. Fungal structures are identified regardless of if they would be viable or not as a culture growth. This method only addresses the analysis of the samples and not the process of sampling or the interpretation of results.

Where 100 of the same spore is counted a stopping rule is in place as per ASTM D7391[1] as when a total of 300 fungal structures per sample is found. These will be reported as estimated counts and indicted with a #.

A scale of 0 to 5+ is used to rate abundance of non-fungal material (debris) see fungal and particle loading categories below. Large amounts of debris may obscure small spores. Counts from samples with 2-4 non-fungal loading may be treated as negatively biased and with 5+ may be treated as overloaded.

1 D7391 − 20 Standard Test Method for Categorization and Quantification of Airborne Fungal Structures in an Inertial Impaction Sample by Optical Microscopy

For more information, freephone 0508 HILL LAB 44 555 22 mail@hill-labs.co.nz hill-labs.co.nz

Direct Examination

Direct examination of mould samples using tape lifts is a simple and effective method to visually inspect and identify mould growth on surfaces. It involves pressing a piece of clear adhesive tape against a suspect surface to collect mould spores, hyphae, or fragments, which are then analysed under a microscope.

The method of analysis is performed in accordance with the following standard - ASTM D7658 – 17 Standard Test Method for Direct Microscopy of Fungal Structures from Tape.

The method is for the direct examination of tape samples. Results are presented in Fungal Loading Categories which include both fungal growth structures, spore types and other fungal particulates. Slides are prepared directly from tape samples for microscopic examination at 100x – 1000x magnification as necessary. Identification is based on current fungal taxonomic keys and reference material. Moulds that do not show enough distinguishing characteristics or structures used for identification are reported as “Unidentified/Miscellaneous moulds.” Where possible, moulds are identified to genus or species noting fungal hyphae, fruiting bodies, or clumps and chains of spores for each fungal type detected. Fungal structures are identified regardless of if they would be viable or not as a culture growth. This method only addresses the analysis of the samples and not the process of sampling or the interpretation of results.

Fungal particles and non-fungal particles loading categories are represented as observed in the microscope field of view and are outlined below.

A scale of 0 to 5+ is used to rate abundance of non-fungal material (debris) see fungal and particle loading categories below. Large amounts of debris may obscure small spores. Counts from samples with 2-4 non-fungal loading may be treated as negatively biased and with 5+ may be treated as overloaded.

Particle Loading and Fungal Loading Categories

Fungal Loading and Particle loading categories						
	Category 0 - None	Category 1 - Trace	Category 2 - Light	Category 3 - Moderate	Category 4 - Heavy	Category 5 - Overloaded
Fungal and Particulate matter – percentage observed	0%	0-5%	5-25%	25-75%	75-90%	Greater than 90%

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List Analytes

Spores [2]

Fungal Type/ Group	Notes
Alternaria spp[3]	Common allergen
Ascospores	Most pose a low risk to indoor air quality, although they can still grow indoors on damp materials. However may be allergenic.
Aspergillus/ *Penicillium*-like	Common indoor moulds; may produce mycotoxins
Basidiospores	Mushroom spores may produce toxins and can act as allergens.
Bipolaris Group	Plant pathogens, Rarely observed indoors, and where found is often associated with indoor house-plant disease and amplification. May produce allergens
Botrytis spp	Plant pathogens, common allergen
Chaetomium spp	Cellulose degrader; indicator of water damage. May produce allergens and mycotoxins
Cladosporium spp	Most common indoor/outdoor mould; allergenic
Curvularia spp	Plant pathogen; possible allergen
Epicoccum spp	May produce allergens and mycotoxins
Fusarium spp	Often found after severe flooding events indoors, where saturation of building products or items has occurred. May produce allergens and mycotoxins.
Ganoderma spp	Mushroom spores m
Memnoniella	Similar to *Stachybotrys*; possible toxin producer
Nigrospora spp	Plant pathogen, May produce allergens
Oidium spp	Plant pathogen producing mildews
Other Colourless Spores	Various unidentified non-pigmented spores
Other Pigmented Spores	Misc. pigmented spores, not classified above
Pithomyces	Grows on cellulose including damp paper and ceiling tiles. May be found in carpeting and mattress dust. May produce allergens
Rusts	Plant pathogens, May produce allergens
Smut-like	Plant pathogens, May produce allergens
Spegazzinia spp	Common in humid areas, outdoor air samples and plant debris as a cellulose decomposer
Stachybotrys	"Black mould", common in leaky, damp or flooded buildings. May produce allergens and mycotoxins
Torula	May produce allergens
Miscellaneous	Any unclassified or less common fungal spores

[2]**Spores** are reproductive units produced by moulds and fungi that are specially adapted for dispersal. They can spread through the air, or via insects, animals, or humans. Once dispersed, spores can remain dormant on a surface for years, waiting for favourable conditions to trigger growth.

[3]**Spp.** An abbreviation for 'species', used where identification to species level for genera observed is not typically required

Growth Structures

Fungal Type/ Group	Notes
Aspergillus and associated Hulle Cells	Hulle cells are thick-walled cells found in some *Aspergillus* species
Alternaria	Common allergen
Ascomycetes	Largest phylum of fungi; produce spores in sac-like structures * see Ascospores
Basidiomycetes	Includes mushrooms and rusts; produce spores on basidia * see Basidiospores
Bipolaris group	Plant pathogens, Rarely observed indoors, and where found is often associated with indoor house-plant disease and amplification. May produce allergens
Cladosporium	Common in air; appears as darkly pigmented structures
Chaetomium and associated asexual state *Botryotrichum*	Cellulose degrader; indicator of water damage. May produce allergens and mycotoxins
Curvularia	Plant pathogen; possible allergen
Memnoniella	Similar to *Stachybotrys*; possible toxin producer
Myxotrichum	Cellulose degrader especially paper, drywall and carpets. Growth common in lower temperatures
Mucor	Fast-growing, Common indoors in high humidity and water damage. May produce allergens and infections
Myxomycetes	Slime moulds, found indoors in moisture rich areas. May produce allergens
Penicillium	Common indoor mould especially in damp environments and water damaged buildings, May produce allergens and toxins
Petriella	Common in wet wood, indoors it is commonly found around sinks where there is persistent wet wood.
Stachybotrys	“Black mould", common in leaky, damp or flooded buildings. May produce allergens and mycotoxins
Scopulariopsis	Some species are human and animal pathogens. Cellulose decomposer in damp indoor environments. Known to break down a wide range of indoor substrates. Common mycosis producer and common allergenic.
Ulocladium	Similar to *Alternaria*; found in water-damaged buildings
Zygomycetes	Common cellulose decomposer including gypsum board, paper, paint, tapestries, jute, other straw materials.
Other	Any unclassified or less common fungal growth structures

Reporting Limits

Spore Trap

Reporting of Results

All results shall be reported as whole numbers. They are to be reported in raw count Fs/S (fungal structures per sample) and in concentration Fs/m³ (fungal structures per m^3)

Reporting Limits & Method Detection Limits

For each spore category, report results are not lower than the minimum reporting limit for that category. For spore categories in which no spores were counted, report results will show (-).

$$\text{minimum reporting limit (fungal structures/samples)} = 100^* \frac{\text{1 Fungal structure counted during traverse}}{\text{percentage scanned}}$$

$$\text{method detection limit (fungal structures/m}^3\text{)} = \frac{\text{minimum reporting limit (fungal structures/samples)}}{\text{volume (m}^3\text{)}}$$

Direct Examination

Reporting of Results

All results shall be reported as Fungal Loading Categories as well as spore and particles in a detected/not detected format.

Please refer to the **Particle Loading and Fungal Loading Categories** table above.

Sampling and Remediation Information

Sampling	
Spore trap	Zefron Air-O-Cell® Sampling Cassette Operating & Instruction Manual
Tape Samples	Tape Sampling for Mould – International Association of Certified Home Inspectors
Remediation	
WorkSafe New Zealand	Preventing and cleaning up mould
United States Environmental Protection Agency	Mould Testing, Sampling and Clean up
MBIE Building performance	Mould in damp buildings

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