

Mycology Reference Centre, Leeds

Information for Service Users

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The Mycology Reference Centre

The Mycology Reference Centre is situated in the Old Medical School, within the Leeds Teaching Hospitals NHS Trust (LTHT) Department of Microbiology.

Contact details:

Mycology Reference Centre
Old Medical School
Thoresby Place
LEEDS, LS1 3EX
Tel: 0113 3926787
Fax: 0113 3928782

URL:

<http://www.pathology.leedsth.nhs.uk/pathology/Departments/Microbiology/Mycology.aspx>

Hayes DX address: DX 6281504, Exchange LEEDS 90 LS.

Clinical and Laboratory Staff contact details:

Consultant in charge: Richard Hobson, Tel 0113 392 2835 or email richard.hobson@leedsth.nhs.uk

Principal Clinical Scientists:

Ruth Ashbee, Tel: 0113 392 8732 or email ruth.ashbee@leedsth.nhs.uk

Richard Barton, Tel 0113 3923390 or email richard.barton@leedsth.nhs.uk

Lead Biomedical Scientist: Chris Goodall Tel: 0113 392 8748 or email christopher.goodall@leedsth.nhs.uk

Opening hours:

The laboratory is open from 09:00-17:00 Monday to Friday. It may be possible to organise specific testing outside these times after discussion with senior staff.

Clinical/laboratory advice and report interpretation:

Advice is available from the staff members listed. Staff are also happy to advise on mycological issues that do not relate to samples submitted to the Mycology Reference Centre for processing, where possible.

Request Forms

Request forms should be filled in for every specimen sent to the laboratory. They should contain as much information as possible, as this may aid interpretation of test results. The minimum information required is: patient name, hospital number, date of birth, the place from which the specimen is being sent e.g. hospital, GP surgery, ward, specimen type and the tests requested.

Request Forms may be downloaded from the mycology website

(<http://www.pathology.leedsth.nhs.uk/pathology/Portals/0/Users/Request%20form%20100111.pdf>)

or obtained by email from

christopher.goodall@leedsth.nhs.uk

Specimen Transportation

Specimens should be sent by established transport networks. Specimens from within the Leeds Teaching Hospitals NHS Trust should be sent either via the air tube system or specimen shuttles from other sites. Specimens should be sent via the postal system or Hayes DX and be appropriately packaged. Specimens (tube or packet) must be clearly labelled with the patients name, and where appropriate a referring laboratory number.

Telephone Reporting of Results

The results of the following investigations will be phoned by the laboratory staff routinely:

- Requests marked "Urgent"

- Positive *Aspergillus* antigen result and any new or rising cryptococcal antigen titre
- Antifungal resistance in clinically significant isolates
- Azole assay results

Fungal Microscopy and Culture

Microscopy and Culture of Clinical Specimens

Use(s): Isolation and identification of all relevant fungi from the following sample types: skin, hair and nail specimens; oral and vaginal swabs; urine; peritoneal dialysis fluid; CSF; respiratory samples (e.g. sputum, broncho-alveolar lavage fluid). For most sample types (with the exception of skin, nail and hair), we would advise referring laboratories to process the primary sample and then send resultant fungal cultures for identification or further testing (See below)

Description: Microscopy for yeasts, mycelium, arthroconidia and other fungal elements; culture of any viable fungi present and identification of any clinically significant species. Antifungal susceptibility testing is undertaken where appropriate or requested (see below)

Specimens: Skin, nail and hair should be sent in Dermapak or similar card packs designed for the purpose. Preferably wet specimens are processed locally.

Results: Microscopy is reported as "No fungus seen" or positive with a description of the fungal cells seen.

Culture is reported as the identity of any significant fungi isolated, estimation of amount of fungal growth (+, ++, +++) where relevant..

**Mean Turnaround Time: Microscopy: 7 days;
Culture: 19 days (Positive cultures will take longer to report than negatives).**

Identification and Antifungal Susceptibility Testing

Yeast identification and sensitivity

Use(s): Identification of yeasts and assessment of susceptibility to antifungals

Description: Identification, usually to species level using a combination of morphological and nutritional/enzymatic tests. Molecular identification is carried out for those isolates which cannot be identified using phenotypic tests.

Susceptibility testing by CLSI M44A disc diffusion (fluconazole) or microbroth dilution (fluconazole, itraconazole, voriconazole, posaconazole, amphotericin B, flucytosine, caspofungin, anidulafungin and micafungin). Specific antifungal(s) tested depend on the identity and source of the isolate and the clinical details supplied. Microbroth dilution testing is undertaken where indicated by isolate identity, disc diffusion results, or where requested specifically. The identity of the yeast isolate is always confirmed or carried out on isolates sent for sensitivity testing.

Specimens: Culture of yeast, ideally on a Sabouraud's agar slope in a bijou or universal.

Results: Susceptible; Intermediate/Susceptible-dose dependent; Resistant/Non-susceptible (where breakpoints have been established). If microbroth dilution testing is carried out, a Minimum Inhibitory Concentration (MIC) can be reported on request.

Mean Turnaround Time: 8 days

Mould Identification

Use(s): Identification of moulds

Description: Identification, usually to species level on the basis of macroscopic and microscopic morphology. Molecular identification is carried out for those isolates which cannot be identified using phenotypic tests.

Specimens: Culture of mould, ideally on a Sabouraud's agar slope in a bijou or universal.

Results: Identity of the mould, usually to species level.

Mean Turnaround Time: 12 days

Note: Susceptibility testing of mould isolates can be carried out on request.

Identification of environmental fungi

Identification of yeasts and moulds from environmental sources can be carried out after discussing your requirement with the laboratory. Please contact Dr Richard Barton or Dr Ruth Ashbee to discuss.

Note: Culture of environmental specimens is carried out by prior arrangement and on medical or environmental health referral only. Costs are dependent on the extent and complexity of the investigations.

Antibody Testing

Note: For antibody tests, please send serum or clotted blood in a plain tube; EDTA blood is not suitable.

Aspergillus Antibodies

Use(s): Diagnosis of allergic bronchopulmonary aspergillosis, aspergilloma, paranasal sinus aspergillosis, other forms of aspergillosis in immunocompetent patients.

Description: Quantitation of IgG antibodies to *Aspergillus fumigatus* in serum using a commercial automated Fluorescent Immuno Enzyme Assay (ImmunoCAP).

Specimens: Serum 100 µl minimum or 1 mL clotted blood.

Results: Results are returned as mg Antibody per litre (mgA/L) and range from <2.0 to >200. Please see the mycology website (<http://www.pathology.leedsth.nhs.uk/pathology/ClinicalInfo/ClinicalServices/MycologyServices/DiscussionofImmunoCAPtesting/tabid/226/Default.aspx>) for comprehensive information on interpretation of Aspergillus ImmunoCAP results.

Mean Turnaround Time: 3 days

Farmers Lung Antibodies

Use(s): Detection of antibodies to *Saccharopolyspora rectivirgula* (previously known as *Micropolyspora faenii*) to diagnose Farmers' Lung.

Description: Determination of the presence of antibodies to *Saccharopolyspora rectivirgula*. (Detection of antibodies to *Thermactinomyces vulgaris* and *T. thalophilus* is no longer possible as antigens are not available).

Specimens: Serum 500 µl minimum or 2 mL clotted blood.

Results: Negative, Positive (weak, strong).

Mean Turnaround Time: 6 days.

Avian Antibodies

Use(s): Detection of antibodies to pigeon serum for diagnosis of Bird Fanciers' Lung.

Description: Determination of the presence and levels of IgG antibodies to pigeon serum. The method used is a commercial automated Fluorescent Immuno Enzyme Assay (ImmunoCAP).

Specimens: Serum 500 µl minimum or 2 mL clotted blood.

Results: Results are returned as mg Antibody per litre (mgA/L) and range from <2.0 to >200.

Please see the mycology website (<http://www.pathology.leedsth.nhs.uk/pathology/ClinicalInfo/ClinicalServices/MycologyServices/DiscussionofImmunoCAPtesting/tabid/226/Default.aspx>) for comprehensive information on interpretation of Avian ImmunoCAP results.

Mean Turnaround Time 3 days

Histoplasma and Coccidioides Antibodies

Use(s): Diagnosis of histoplasmosis and coccidioidomycosis.

Description: Histoplasma: Determination of the presence of antibodies to *Histoplasma capsulatum* by immunodiffusion (mycelial antigen) and complement fixation test (CFT; mycelial and yeast antigens).

Coccidioides: Determination of the presence of antibodies to *Coccidioides* species by immunodiffusion and CFT.

Specimens: Serum 500 µl (*Histoplasma*) or 1ml (*Coccidioides*) minimum or 2 mL clotted blood.

Results: Histoplasma: Immunodiffusion: Negative, Positive (M or M+H band); CFT: Negative, Positive (Titre) to mycelial and/or yeast antigens. *Coccidioides:* Immunodiffusion: Negative, Positive; CFT: Negative, Positive (Titre).

The CFT is only carried out once a week.

Mean Turnaround Time: Histoplasma 9 days; Coccidioides 7 days

Note: Inclusion of travel history is useful for confirming potential exposure to these fungi.

Blastomyces and Paracoccidioides Antibodies

Use(s): Diagnosis of blastomycosis or paracoccidioidomycosis.

Description: Determination of the presence of antibodies to *Blastomyces dermatitidis* or *Paracoccidioides brasiliensis* by immunodiffusion.

Specimens: Serum 500 µl minimum or 2 mL clotted blood.

Results: Negative, Positive.

Mean Turnaround Time: *Blastomyces* 5 days; *Paracoccidioides* 6 days

Note: Inclusion of travel history is useful for confirming potential exposure to these fungi.

Antigen Testing

Note: For the following tests, please send serum or clotted blood in a plain tube; EDTA blood is not suitable.

Aspergillus Antigen (Galactomannan)

Use(s): Diagnosis of invasive aspergillosis usually in immunocompromised patients.

Description: Determination of the presence of *Aspergillus* galactomannan in serum or BAL by ELISA.

Specimens: Serum or BAL 700 µl minimum or 5 mL clotted blood.

Results: Negative; Positive with the index value (indicating the relative concentration of galactomannan)

Mean Turnaround Time: 1 day

Note: Positive results are confirmed before reporting by re-testing the specimen submitted. To improve specificity positives should be confirmed by submission of a second specimen.

Cryptococcal antigen

Use(s): Diagnosis of cryptococcal meningitis, systemic cryptococcosis in both immunocompetent and immunocompromised patients.

Description: Determination of the presence of cryptococcal antigen and the titre in the specimen, by lateral flow device.

Specimen: Serum or CSF, 300 µl minimum or 3 mL clotted blood.

Results: Negative, Positive (No titration required), Positive (Titration to follow), Positive (Titre).

Mean Turnaround Time <1 day

Antifungal Drug Assays

Antifungal agents are assayed by liquid chromatography-tandem mass spectroscopy (LC-MS-MS). The results are highly specific and are not influenced by the use of antifungal combination therapy.

Itraconazole, Posaconazole, Voriconazole

Use(s): Confirmation of adequate levels and alerting to toxic levels in patients receiving antifungal azoles for treatment or prophylaxis of fungal disease.

Specimens: Serum 200 µl minimum or 2 mL clotted blood

Results: Drug concentration in mg/L, with advice on target levels.

Mean Turnaround Time: 4 days for all drugs

Note: The assay is currently carried out twice weekly on Tuesday and Thursday, although specimens need to be received the preceding day for processing.

External Quality Assurance

The Mycology Reference Centre participates in the following EQA schemes: UKNEQAS Fungal Identification; UKNEQAS Antifungal Susceptibility; UKNEQAS Fungal Serology; SKML scheme for anti-fungal drug assay. We also operate an informal EQA scheme (serum exchange programme) with the Mayo Clinic for endemic mycoses, as no formal EQA scheme is available.

Further information

To obtain further copies of this document, go to <http://www.pathology.leedsth.nhs.uk/pathology/Departments/Microbiology/Mycology.aspx> under "User Manual" link on the left or email christopher.goodall@leedsth.nhs.uk

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