



The Leeds Teaching Hospitals



NHS Trust

# Mycology Reference Centre, Leeds

## Information for Service Users 2017

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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  
Leeds General Infirmary  
LEEDS, LS1 3EX

Tel: 0113 3926787

[www.pathology.leedsth.nhs.uk/Mycology](http://www.pathology.leedsth.nhs.uk/Mycology)

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

#### Clinical and Laboratory Staff contact details:

Principal Clinical Scientist: Richard Barton, Tel 0113 3923390 or email

[Richard.Barton2@nhs.net](mailto:Richard.Barton2@nhs.net)

Clinical Scientist: Jenny Ratner, Tel 0113 392

8732 email [Jennifer.Ratner@nhs.net](mailto:Jennifer.Ratner@nhs.net)

Lead Biomedical Scientist: Rebekka Tolson Tel:

0113 392 8748 or email

[Rebekka.Tolson@nhs.net](mailto:Rebekka.Tolson@nhs.net)

#### 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 website or obtained by email from [christopher.goodall1@nhs.net](mailto:christopher.goodall1@nhs.net)

#### 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:

- i) Requests marked "Urgent"
- ii) Positive *Aspergillus* antigen result and any new or rising cryptococcal antigen titre
- iii) Antifungal resistance in clinically significant isolates

iv) Azole assay results outside standard ranges

### **Fungal Microscopy and Culture**

#### **Microscopy and Culture of Clinical Specimens**

*Use(s):* Isolation and identification of relevant fungi from skin, hair and nail specimens; oral and vaginal swabs; urine; peritoneal dialysis fluid; CSF; sputum and broncho-alveolar lavage fluid. For sample types other than skin, nail and hair, we 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.

*Limitations:* Negative fungal culture does not exclude a diagnosis of fungal infection but reduces its likelihood. Depending on the fungus isolated, positive results may indicate contamination. Clinical correlation of results is required.

**95% Turnaround Time: Microscopy: 7 days; Culture: 27 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 MALDI-TOF. 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.

*Limitations:* *In vitro* susceptibility to an antifungal agent does not guarantee clinical success with that agent, but makes clinical success more likely.

**95% Turnaround Time: 16 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.

*Limitations:* If insufficient phenotypic and/or genotypic information is available for species identification moulds may be identified to genus level only.

**95% Turnaround Time: 22 days**

#### **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 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.*

*Limitations of all antibody tests: An antibody result above the laboratory-determined cut off level is supportive, but not diagnostic, of the diagnosis under consideration. Conversely, a low antibody level does not exclude the diagnosis, but makes it less likely. Please see the mycology website for comprehensive information on interpretation of ImmunoCAP antibody results.*

#### **Aspergillus fumigatus antibodies**

*Use(s):* Diagnostic support for allergic bronchopulmonary aspergillosis, aspergilloma, paranasal sinus aspergillosis, other forms of aspergillosis in immunocompetent patients.

*Description:* Quantitation of IgG antibodies to *A. 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.

**95% 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.

**95% Turnaround Time 6 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).

**95% Turnaround Time: 7.5 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).

*Limitations:* The CFT tests are more sensitive and less specific than the immunodiffusion tests. Positive CFT in the absence of positive immunodiffusion is less likely to indicate the presence of infection than if both tests are positive.

**95% Turnaround Time: Histoplasma 14 days; Coccidioides 12 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.

**95% Turnaround Time: *Blastomyces* 6 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. *NB: For BAL samples, the referring laboratory must confirm that the sample has been tested and is negative for acid & alcohol-fast bacilli.*

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

*Limitations:* A negative *Aspergillus* antigen result does not exclude a diagnosis of aspergillosis. False positive results have been associated with some batches of beta-lactam antibiotics (e.g. piperacillin-tazobactam) and with testing babies in the neonatal period. Cross-reactive positive results may occur in patients with fungal infections other than aspergillosis (e.g. histoplasmosis). There is no universal agreement about appropriate cut off levels for BAL samples.

**95% Turnaround Time: 3 days**

*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 (CRAG)**

*Use(s):* Diagnosis of cryptococcal meningitis, systemic cryptococcosis in both

immunocompetent and immunocompromised patients.

*Description:* Determination of the presence of cryptococcal antigen and (if requested specifically) antigen titre, by lateral flow immunochromatography.

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

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

*Limitations:* The lateral flow test has been found to be as diagnostically accurate as other CRAG test types and is therefore highly sensitive and specific. However, accurate data on test sensitivity and specificity are currently limited.

**95% Turnaround Time 1.5 days**

#### **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.

**95% Turnaround Time: Itraconazole 9 days, voriconazole 8 days, posaconazole 8 days.**

*Limitations:* Adequate antifungal drug levels do not guarantee response to treatment with the antifungal agent in question, but are considered to increase the likelihood of clinical response. High levels may be associated with toxicity.

*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*

## Mycology Reference Centre, Leeds USER MANUAL

*Susceptibility; UKNEQAS Fungal Serology; UKNEQAS scheme for anti-fungal drug assay, UKNEQAS scheme for fungal biomarkers. 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 [www.pathology.leedsth.nhs.uk/mycology](http://www.pathology.leedsth.nhs.uk/mycology)**

Under "User Manual" link on the left or email [christopher.goodall1@nhs.net](mailto:christopher.goodall1@nhs.net)

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