

May 2010

Tuberculosis (TB) and Mycobacteriology Laboratory Update

To health care providers:

Nucleic Acid Amplification tests (NAT) for the direct detection of *M. tuberculosis* complex from specimens

The Ontario Agency for Health Protection and Promotion (OAHPP) public health laboratories (PHL) currently uses two amplification tests to detect *Mycobacterium tuberculosis*:

1. The Amplified Mycobacterium Tuberculosis Direct Test (AMTD, Gen-Probe CA) which detects *M. tuberculosis* complex rRNA
2. PCR (Seegene, Seoul, Korea) which detects *M. tuberculosis* DNA.

AMTD is performed automatically on AFB smear positive specimens from new patients.

If you would like to request AMTD on a specimen regardless of the AFB result, please contact OAHPP medical microbiologist, Dr. Jamieson at 416-235-5841, before submitting the sample to public health laboratories otherwise the entire respiratory specimen will be used for culture. Requests written on the requisition will not be accepted unless prior approval is obtained. Non-respiratory and specimens with visible blood will be tested by PCR.

Respiratory specimens

Sputum specimens should contain at least 5ml and the patient must be instructed on how to correctly produce a deep expectoration. The submission of three separately obtained specimens is recommended by the Canadian Tuberculosis Standards. If a Bronchoalveolar Lavage (BAL) is submitted, a post-bronchoscopy sputum sample should also be sent. Follow-up sputum specimens from tuberculosis patients should be submitted for AFB smear and culture at the end of first and second month's therapy respectively, and at the completion of treatment.

Cerebrospinal fluid specimens (CSF)

Specimens of CSF should be at least 2ml in volume in order to allow for concentration of the specimen. Lesser volumes may compromise the isolation of mycobacteria. Specimens that are less than 0.5ml will be rejected.

Tuberculosis (TB) and Mycobacteriology Laboratory Update (Continued)

Swabs

Swabs are inadequate specimens for the isolation of mycobacteria, as insufficient material is available for culture. Please submit aspirated material in a sterile specimen container. Dry swabs and swabs in anaerobic transport medium and dry swabs will be rejected.

Urine Samples

Urine samples should only be submitted if renal TB is suspected. AFB smears on urine will be reported as of May 15, 2010.

STAT AFB smears

STAT smears **must** be requested **by phone** to the lab. A phone number and contact person must be provided on the form for phoning the result. Specimens received in the lab by 2 p.m. will have a STAT smear phoned the same day. Specimens received after 2 p.m. will have the smear result phoned first thing the next day.

Culture reports

Cultures are reported negative if there is no growth at seven weeks. Reports of mycobacteria isolated, or of no growth, are issued within one working day of identified growth or at seven weeks negative. All new isolates of *M. tuberculosis* complex are telephoned to the submitter within 24 hours of identification. Tests to speciate within the MTB complex are performed if the strain shows resistance to pyrazinamide, or by request.

Susceptibility testing of *M. tuberculosis*

All first-identified isolates of *M. tuberculosis* from patients are tested for susceptibility to the first-line anti-tuberculosis drugs. Susceptibility testing to first line drugs is repeated at three months if cultures are still positive. The second-line panel of drugs is tested if any isolate is resistant to rifampin or to any two of the first-line drugs.

Susceptibility testing of Nontuberculous Mycobacteria (NTM)

Susceptibility testing for NTM is performed upon request. Isolates of *M. avium* complex or *M. kansasii* are screened for susceptibility to clarithromycin and rifampin respectively at the TB and Mycobacteriology Laboratory at Toronto PHL. All other isolates of NTM, and all isolates requiring additional susceptibility testing, are submitted to the National Reference Centre for Mycobacteriology (NRCM) in Winnipeg. The patient's clinical information and the reason for requesting susceptibility testing is required by the NRCM and should be faxed to the TB and Mycobacteriology Laboratory at the time of request.

Tuberculosis (TB) and Mycobacteriology Laboratory Update (Continued)

New Genotyping Request Form

For MTB isolate strain typing, please submit requests using the new TB Genotyping Request Form. This form may be obtained electronically from <http://www.oahpp.ca/resources/laboratory-materials.html>, or by telephone at 416-235-5993. Once complete, fax the form to the Mycobacteriology laboratory at 416-235-6013. Genotyping is performed using two rapid PCR methods, 24-loci MIRU-VNTR and spoligotyping.

For further information: Please contact:

- TB and Mycobacteriology Laboratory, Toronto, phone **416-235-5928**, fax **416-235-6013**, or contact your local OAHPP laboratory.
- Manager, TB and Mycobacteriology Lab. Toronto, Pam Chedore, **416-235-5993**
- Medical microbiologist, Dr Frances Jamieson, **416-235-5841**
- OAHPP Laboratory Helpline **1-800-640-7221**
- For the OAHPP Specimen Collection Guide and previous Labstracts refer to <http://www.oahpp.ca/publichealthlaboratories.php>
- For the Genotyping request form and general requisition refer to <http://www.oahpp.ca/resources/laboratory-materials.html>