TO: Deputy Director General: Chief of Operations
     Chief Executive Officer: Central, Regional & District Hospitals
     Chief Operational Officers: Central, Regional & District Hospitals, General Specialist and Emergency Services
     Chief Directors: Metro & Rural District Health Services, General Specialist and Emergency Services
     Executive Director Health: City of Cape Town
     District Managers: Sub-Structures & Districts
     Facility Managers: District Health Services
     NHLS
     PCGC: Medicine and Paediatrics
     Deputy Director: Laboratory Services

ATTENTION: All healthcare workers submitting specimens for TB investigations to the NHLS laboratories.

CIRCULAR NUMBER H.157/2014

GUIDELINES ON THE USE OF XPERT MTB/RIF IN: I) CHILDREN WITH SUSPECTED PULMONARY TB & II) EXTRA PULMONARY SAMPLES IN BOTH ADULTS & CHILDREN WITHIN DISTRICT, REGIONAL AND CENTRAL HOSPITALS IN THE WESTERN CAPE

This circular replaces the Circular H13 / 2013 – Restriction on GXP test requests on specimens other than sputa and tracheal aspirates

8 Riebeeck Street, Cape Town, 8001
tel: +27 21 483 5751  fax: +27 21 483 6033
P O Box 2060, Cape Town, 8000
www.capegateway.gov.za
Background

The use of Xpert MTB / RIF on respiratory tract samples has been well documented and is endorsed by the WHO for the diagnosis of pulmonary TB in adults and is now included in the NDoH TB diagnosis algorithm. Both national and provincial policy stipulates the performing of Xpert MTB/RIF on respiratory samples (sputum, induced sputum and tracheal aspirates) from adult TB suspects. The NHLS performs these Xpert MTB / RIF tests for adult PTB suspects at all levels of care.

Recent evidence regarding the performance of Xpert in both paediatric TB as well as extra-pulmonary TB has shed more light on the potential use of Xpert testing in these settings. This document serves to clarify the availability and use of Xpert testing for EPTB and paediatric TB, and on Broncho-alveolar lavage aspirates. It does not replace current national and provincial guidelines for use of Xpert, but serves to add to them. The guidelines discussed in this document apply only to the tertiary, district and regional hospitals in the Western Cape.

1. Guideline for using Xpert MTB/RIF in children with suspected pulmonary tuberculosis (PTB)

Limitations of Xpert MTB/RIF in paediatric practice include (1) lower sensitivity than TB culture, hence this guideline recommends that both Xpert MTB/RIF and TB culture be used in combination in routine PTB diagnosis, (2) Xpert MTB/RIF is not a Mycobacterium tuberculosis specific test but identifies all mycobacteria within the Mycobacterium tuberculosis complex; therefore, if M. bovis BCG infection is a diagnostic consideration, speciation of a cultured MTB complex isolate should be requested, and (3) although Xpert MTB/RIF is able to identify rifampicin-resistant strains, it is currently unable to identify INH-mono-resistant TB strains; this is currently done by the Hain line probe assay.

General principles

1. A single Xpert MTB/RIF test will be performed as part of the workup for suspected PTB in children
2. The Xpert MTB/RIF test may be requested on any one of the following specimen types: a respiratory tract specimen (e.g. spontaneously produced sputum, induced sputum or tracheal aspirate), a gastric lavage aspirate (gastric washings) or a gastric aspirate (where no fluid is instilled in the stomach).
3. Xpert MTB/RIF, when performed, will replace smear microscopy.
4. Repeat Xpert MTB/RIF testing is not recommended for evaluating sputum conversion; this should be done by sputum culture

Specimen collection & processing

Children less than 13 years of age

Submit a single respiratory tract or gastric (lavage) aspirate specimen (minimum volume = 2ml) for screening by Xpert MTB/RIF and TB culture. This specimen will be processed in the laboratory for culture; the decontaminated deposit will be used to perform both Xpert MTB / RIF and to inoculate MGIT culture medium.

**Note: Please request Xpert MTB/RIF, culture and susceptibility testing on the request form for paediatric patients in order to facilitate the above process.**

- If the Xpert MTB/RIF result is positive for MTB complex that is rifampicin susceptible, the TB culture will be completed, and where positive, processed by Hain line probe assay to identify INH-mono-resistant isolates. Further diagnostic specimens should not be sent to the laboratory
- If the Xpert MTB/RIF result is positive for MTB complex that is rifampicin resistant, the TB culture will be completed and processed by Hain line probe assay and second-line drug susceptibility testing (DST). A second (backup) specimen should be sent for TB culture to maximize the chance of obtaining a viable isolate
that can be used for second-line DST. Ideally, the second specimen should be obtained before the child is commenced on anti-TB therapy.

- If the Xpert MTB/RIF result is either negative for MTBC or positive for MTBC but indeterminate for rifampicin, a second (backup) specimen should be submitted for TB culture to maximize the chance of confirming the diagnosis of TB. This second specimen should be obtained before the child is commenced on anti-TB therapy.

Adolescents ≥13 years of age

For these adolescents the standard recommended screening approach for adults should apply, in the Western Cape this requires that 2 samples be sent simultaneously to the laboratory. One, respiratory tract specimen will be screened by Xpert MTB/RIF only (i.e. not cultured). The second specimen is processed by MGIT culture in the following situations:

1. indeterminate rifampicin Xpert MTB/RIF result,
2. HIV-infected adolescents with a MTBC-negative Xpert MTB/RIF result (thus the HIV status must be indicated on the request form),
3. adolescents who test rifampicin resistant on Xpert MTB/RIF

If the Xpert MTB/RIF is negative, and culture is required for any other reason (such as adolescents who are being retreated for TB or who have been in contact with drug-resistant infectious TB cases), a second sample must be submitted and culture requested.

2. Guideline for using Xpert MTB/RIF for extra pulmonary samples from adults and children

The evidence to date describing the performance of Xpert MTB/RIF on extra pulmonary samples (both published and unpublished), indicates that the performance of Xpert MTB/RIF in EPTB varies substantially with the specimen type. While specificity has overall been excellent across all specimen types, sensitivity varies. Poor sensitivity (<50%) has been found when performing Xpert MTB/RIF on clear pleural, ascitic and peritoneal fluids, but good sensitivity (close to 100% compared to positive culture in some studies) when using CSF, lymph node aspirates, thick (pus) pleural fluids and tissue samples.

The decision to offer Xpert MTB/RIF testing has been influenced by these factors, as well as the instrument and staff capacity in the NHLS laboratories. Testing will thus be limited to specific specimen types, as outlined below.

General Principles:

1. Xpert MTB/RIF will be performed on the following extra pulmonary specimens:
   a. CSF samples
   b. Lymph node aspirates, other tissue aspirates and pus aspirates
2. Xpert MTB/RIF will NOT be performed on clear (non-purulent) pleural, pericardial or peritoneal fluids, on urine, or on stool samples
3. Only one Xpert MTB/RIF assay will be performed per patient and specimen type. In other words, a patient may have an Xpert MTB/RIF on both a sputum and an extra-pulmonary sample (unless one is known to be positive – see point 3), but Xpert MTB/RIF will not be performed on two sputum (or two of the same extra-pulmonary samples)
4. If a patient has a positive Xpert MTB/RIF from any site, no further Xpert MTB/RIF testing will be performed on any other specimen, regardless of specimen type. For example, a patient with a positive Xpert MTB/RIF from sputum will not have an Xpert MTB/RIF performed on the CSF sample.
5. The samples listed in point 1 will be processed for both Xpert MTB/RIF and TB culture, in order not to waste the sample, or subject the patient to repeated invasive specimen collection if the Xpert MTB/RIF is negative. If specimen volumes are too small to perform both tests, Xpert MTB/RIF will be performed in preference.

6. Xpert MTB/RIF testing will replace microscopy.
   - If the Xpert MTB/RIF assay shows rifampicin resistance, a second (backup) specimen should be sent to the laboratory (if at all possible) for TB culture to maximize the chance of isolating the organism and being able to perform 2nd line susceptibility testing.

**Specimen collection:**

**CSF**

Submit in a normal sterile tube, indicating TB investigation on the form. Please send at least 5mL, but preferable 10mL, which will allow for the cell count, routine MCS and chemistry to be performed. This should leave at least 3mL for TB investigation. TB culture will only be performed if the volume of CSF left over for TB investigations is >3mL and if the Xpert MTB/RIF is negative, or shows rifampicin resistance. If the available volume of CSF for left over for TB investigation is <3mL, then ONLY Xpert MTB/RIF will be performed, and TB culture will not be performed regardless of the result of the Xpert MTB/RIF. If the lymphocyte count is normal (<5 lymphocytes), or the cell count is in keeping with acute bacterial meningitis, TB investigations will not be performed. CSF will be kept in the laboratory for one week, and the need for TB investigation can be discussed.

Remember that the greater the volume submitted, the greater the chance of detecting MTB (by Xpert and by culture). A volume of 10 mL of CSF is the optimal volume to submit and if possible and safe to perform, should be attempted.

**Lymph node, tissue aspirates and pus**

Preferably submit two samples. One for culture and one for Xpert MTB/RIF testing. Samples should be sent either in preservative-free saline, or neat (if they are large volume). For LN aspirates use special TB transport medium for culture (liceal with laboratory) or preservative-free saline, if a single sample is submitted, only Xpert MTB/RIF will be performed unless there is an adequate volume to perform culture on the Xpert specimen as well.

3. **Guideline for using Xpert MTB/RIF for samples obtained at bronchoscopy (broncho-alveolar lavage fluids) from adults and children**

Although there are limited studies that have looked at BAL fluids as a specific specimen group for Xpert MTB/RIF, the yield from this specimen group appears to be excellent (>80% sensitivity). In addition, many studies have included BAL fluids in testing algorithms, and anecdotal experience also supports the use Xpert MTB/RIF of BAL fluid. Xpert MTB/RIF testing will thus be performed on BAL samples where TB investigations have been requested, unless the patient already has a positive Xpert MTB/RIF result from another sample.

- Xpert MTB/RIF testing will replace microscopy.
- If a patient has had an MTBC-negative Xpert MTB/RIF on a sputum sample, a second Xpert MTB/RIF on an invasive respiratory tract sample (BAL) can be requested. However, if a patient has had an MTBC-negative Xpert MTB/RIF on a BAL sample, Xpert MTB/RIF will NOT be performed on a subsequent sputum sample.
- Samples will be processed for Xpert MTB/RIF and TB culture in parallel, in order not to waste the sample, or subject the patient to repeated invasive specimen collection if the Xpert MTB/RIF is negative.
If the Xpert MTB/Rif assay shows rifampicin resistance, a second specimen (either BAL or other respiratory specimen) should be sent to the laboratory (if at all possible) for TB culture to maximize the chance of isolating the organism and being able to perform 2nd line susceptibility testing.

DR BETH ENGELBRECHT
DEPUTY DIRECTOR GENERAL: CHIEF OF OPERATIONS
DATE: 2014-09-16

Contributors: Brian Elay, Andrew Whitelaw, Mark Nicol, Heather Zar, Kim Bonorchis, James Nuttall, Colleen Bamford, Simon Schaaf, Marc Mencelison, Jorjfe Tjaard, Jannie Mouton, Nicolene van der Westhuizen and various DoH & NHLS clinical, pathology and technical experts.