World Health Organization

Rapid Communication:
Molecular assays as initial tests for the diagnosis of tuberculosis and rifampicin resistance

January 2020

Molecular assays intended as initial tests for the diagnosis of pulmonary and extrapulmonary TB and rifampicin resistance in adults and children: rapid communication. Policy update

ISBN 978-92-4-000033-9

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Suggested citation. Molecular assays intended as initial tests for the diagnosis of pulmonary and extrapulmonary TB and rifampicin resistance in adults and children: rapid communication. Policy update. Geneva: World Health Organization; 2020. Licence: CC BY-NC-SA 3.0 IGO.

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Background

Tuberculosis (TB) remains a threat to global public health and is the top infectious cause of death globally. In 2018, an estimated 10 million people developed TB and 1.5 million died from the disease.[1] At least 1 million children become ill with TB every year. In 2018, an estimated 205 000 children died of TB. About 500,000 new cases of multidrug [2] and rifampicin-resistant tuberculosis (MDR/RR-TB) are estimated to emerge annually but only one in three cases were reported by countries to have been diagnosed and treated in 2018.

Ending the global TB epidemic will only be achievable if there is intensive action by all countries that have endorsed the End TB Strategy and its ambitious targets.[3] It requires a paradigm shift from the conventional approaches to disease control used previously. Early and accurate diagnosis of all forms of TB and rapid detection of drug resistance is fundamental to this shift.

Since the approval by WHO of Xpert® MTB/RIF (Cepheid, Sunnyvale, USA, hereafter referred to as 'Xpert MTB/RIF') in 2010, significant additional evidence has been generated on its use as initial test for the diagnosis of TB and rifampicin-resistant TB. In recent months new data on the use of Xpert® MTB/RIF Ultra (Cepheid, Sunnyvale, USA, hereafter referred to as 'Xpert Ultra') and on the latest version Truenat® MTB and MTB Plus system (Molbio Diagnostics, Goa, India, hereafter referred to as 'Truenat') have also become available.

WHO commissioned a systematic review of all the available data in 2019. The results were assessed during a meeting of an independent WHO-convened Guideline Development Group (GDG) on 3-6 December 2019. Detailed recommendations will be published as part of updated WHO Consolidated Guidelines on TB diagnosis in 2020.

This Rapid Communication aims to inform national TB programmes and other stakeholders about the key implications of the latest evidence on the use of specific molecular assays as initial diagnostic tests of pulmonary and extrapulmonary TB and RR-TB, in adults and children.

The updated 2020 WHO Consolidated Guidelines will also incorporate recent WHO recommendations on other rapid tests including line probe assays, urine lipoarabinomannan lateral flow assays and molecular loop-mediated isothermal amplification assays.

Key findings

- High diagnostic accuracy and improved patient outcomes confirmed for Xpert MTB/RIF as initial test to diagnose pulmonary TB (i.e. replacing smear microscopy) and to simultaneously detect rifampicin resistance

[1] Global Tuberculosis Report 2019 https://www.who.int/tb/publications/global_report/en/

[2] Defined as combined resistance to rifampicin and isoniazid, the two most important anti-TB medicines.

[3] Implementing The End TB Strategy: the essentials. Geneva: World Health Organization; 2015 (WHO/HTM/TB/2015.31; http://www.who.int/tb/publications/2015/end_tb_essential.pdf?ua=1

Intervention: *Xpert MTB/RIF as replacement for sputum smear microscopy (bacteriological culture as reference standard)*

Data assessed: 70 studies (including 5 randomized controlled trials) involving more than 30,000 participants from 37 countries (including 14 high-burden TB countries).

Results: High diagnostic accuracy of Xpert MTB/RIF confirmed in adults with pulmonary TB: Overall sensitivity was 85% in all specimens (smear-positive and smear-negative). Overall specificity was 98% in all specimens (smear-positive and smear-negative). Sensitivity in sputum specimens from HIV co-infected participants was 81% while specificity was 98%.

Results also showed improved patient-important outcomes (cure rates, reduced mortality and reduced pre-treatment loss) when Xpert MTB/RIF replaced microscopy as the initial diagnostic test.

Intervention: *Xpert MTB/RIF for simultaneous detection of rifampicin resistance (phenotypic drug susceptibility testing as reference standard)*

Data assessed: 48 studies involving more than 8,000 participants from 33 countries (including 9 high-burden TB countries).

Results: High overall sensitivity (96%) and specificity (98%) when compared to phenotypic drug susceptibility testing.

- High diagnostic accuracy of Xpert Ultra as initial test to diagnose pulmonary TB (i.e. replacing smear microscopy) and to simultaneously detect rifampicin resistance

Intervention: *Xpert Ultra as replacement for sputum smear microscopy (bacteriological culture as reference standard)*

Data assessed: 6 studies involving more than 2,000 participants from 14 countries (including 8 high-burden countries). More data are needed on relevance of 'trace calls' and on patient outcomes.

Results: High diagnostic accuracy of Xpert Ultra in adults with pulmonary TB: Overall sensitivity (including 'trace' calls as positive) was 90% in all specimens (smear-positive and smear-negative). When 'trace' calls were excluded the overall sensitivity decreased to 86%. Overall specificity (including 'trace' calls) was 96% in all specimens (smear-positive and smear-negative). Excluding 'trace' calls resulted in a slight increase in specificity to 98%. Sensitivity in sputum specimens from HIV co-infected participants was 88% while specificity was retained at 95%.

Intervention: *Xpert Ultra for simultaneous detection of rifampicin resistance (phenotypic drug susceptibility testing as reference standard)*

Data assessed: 5 studies involving close to 1,000 participants from 12 countries (including 7 high-burden TB countries).

Results: High overall sensitivity (94%) and specificity (99%) when compared to phenotypic drug susceptibility testing.

- ## Use of Xpert MTB/RIF and Xpert Ultra to diagnose TB and detect rifampicin resistance in children from sputum, stool, nasopharyngeal and gastric specimens

Given the difficulties in obtaining sputum specimens from children and the technical limitations of conventional bacteriological methods to aid diagnosis, various non-pulmonary specimens and composite reference standards are often used in evaluating the performance of new diagnostic technologies in paediatric TB.

Intervention: *Xpert MTB/RIF or Xpert Ultra as initial diagnostic test for TB in children (bacteriological culture as reference standard)*

Data assessed for Xpert MTB/RIF: 43 studies involving more than 6,000 participants from 21 countries (including 14 high-burden countries. Studies evaluated sputum specimens, gastric specimens, nasopharyngeal specimen; and stool specimens).

Results: Sensitivity varied by specimen type (46% for nasopharyngeal specimens; 61% for stool; 65% for sputum; and 73% for gastric specimens). Specificity for all specimens varied from 98% to 100%.

Data assessed for Xpert Ultra: Three studies involving close to 700 participants in three high-burden TB countries. These studies all evaluated sputum specimens while one study also evaluated nasopharyngeal specimens.

Results: Sensitivity varied by specimen type (46% for nasopharyngeal specimens; 73% for sputum). Specificity for all specimens varied from 97% to 98%.

Intervention: *Xpert MTB/RIF to detect rifampicin resistance in children (phenotypic drug susceptibility as reference standard)*

Data assessed: Six studies involving more than 200 participants from four high-burden TB countries.

Results: High overall sensitivity (90%) and specificity (98%) when compared to phenotypic drug susceptibility testing.

- ## Use of Xpert MTB/RIF and Xpert Ultra to diagnose TB and detect rifampicin resistance in adults with extra-pulmonary TB

Given the difficulties in obtaining extra-pulmonary specimens and the technical limitations of conventional bacteriological methods to aid diagnosis, various non-pulmonary specimens and composite reference standards are often used in evaluating the performance of new diagnostic technologies in extra-pulmonary TB.

Intervention: *Xpert MTB/RIF as initial diagnostic test in adults with extra-pulmonary TB (bacteriological culture as reference standard)*

Data assessed: 59 studies from 26 countries (including seven high-burden countries). MTB/RIF in adults with extra-pulmonary TB. Studies evaluated cerebrospinal fluid specimens, lymph node aspirate, lymph node biopsy, pleural fluid, urine, synovial fluid, peritoneal fluid, pericardial fluid, and blood.

Results: Sensitivity of Xpert MTB/RIF varied by specimen type (from 50% for pleural fluid up to 97% for synovial fluid). The specificity of Xpert MTB/RIF also varied by specimen type (from 79% for lymph node biopsy up to 99% for pleural fluid).

Intervention: *Xpert Ultra as initial diagnostic test in adults with extra-pulmonary TB (bacteriological culture as reference standard)*

Data assessed: Six studies involving from three countries (including two high-burden TB countries). Studies evaluated cerebrospinal fluid, pleural fluid, urine, synovial fluid and urine.

Results: Sensitivity of Xpert Ultra varied by specimen type (from 71% for pleural fluid up to 100% for lymph node biopsy). The specificity of Xpert Ultra also varied by specimen type (from 71% for pleural fluid up to 100% for urine).

Intervention: *Xpert MTB/RIF or Xpert Ultra for simultaneous detection of rifampicin resistance (phenotypic drug susceptibility testing as reference standard)*

Data showed overall high performance of both assay for detection of rifampicin resistance: sensitivity 96-97% and specificity 99%.

- High diagnostic accuracy of Truenat as initial test to diagnose TB (i.e. replacing sputum smear microscopy) and to sequentially detect rifampicin resistance

Intervention: *Truenat MTB and MTB Plus as replacement for sputum smear microscopy (bacteriological culture as reference standard)*

Data assessed: Interim data from a FIND-coordinated multi-central, prospective field evaluation study in four countries (India, Ethiopia, Peru, Papua-New Guinea) involving 744 participants (out of 1,866 enrolled) with final results on bacteriological culture. The review was based on interim evidence, more data are needed including on patient outcomes.

Results: Overall sensitivity of the Truenat MTB assay was 83% and that of the MTBPlus assay 89%. Specificity was 99% for the MTB and 98% for the MTBPlus assay. The interim analysis also included an assessment of the operational characteristics (provided by the manufacturer) and ease of use of the assays (provided by FIND from the study sites).

Intervention: *Truenat MTB-Rif Dx for sequential detection of rifampicin resistance (phenotypic drug susceptibility testing as reference standard)*

Data assessed: Interim data from a FIND-coordinated multi-central, prospective field evaluation study in four countries (India, Ethiopia, Peru, Papua-New Guinea) involving 163 participants (out of 744 with MTB and MTB Plus positive results) and with final phenotypic drug susceptibility testing.

Results: Sensitivity of the Truenat MTB-Rif Dx assay was 93% and specificity was 95%.

Overall conclusions

The latest evidence reviewed supports the continued use of Xpert MTB/RIF and Xpert Ultra as initial diagnostic tests for pulmonary TB in patients of all ages. It also supports the use of Xpert MTB/RIF and Xpert Ultra in the diagnostic work-up of all patients with extra-pulmonary TB and in children with TB (specifically in gastric specimens, nasopharyngeal specimens and stool specimens). Both assays also show high accuracy in the simultaneous detection of rifampicin resistance.

The performance of Truenat MTB, MTB Plus and MTB-RIF Dx assays show comparable accuracy with Xpert MTB/RIF and Xpert Ultra for TB detection (Truenat MTB and Truenat MTB Plus) and for sequential rifampicin resistance detection (Truenat MTB-Rif Dx).

The Truenat MTB and MTB Plus assays also show comparable accuracy to the TB-LAMP® assay (Eiken Chemical Company Ltd (Tokyo, Japan) as replacement tests for sputum smear microscopy.

The data for Truenat MTB-Rif Dx show similar accuracy to WHO-approved commercial line probe assays (GenoType MTBDRplus ®VER 1 and 2 (Hain Lifescience, Germany); Nipro NTM+MDRTB detection kit 2® (Nipro, Japan).

Next steps

The updated policy guidelines on molecular assays intended as initial tests for the diagnosis of TB and rifampicin resistance will be released by April 2020, as part of anticipated consolidated WHO Guidelines on TB diagnostics during 2010. Updated implementation guidance will also be developed in 2020.

National TB Programmes and other stakeholders are encouraged to conduct high-quality implementation/operational research to collect more evidence on the accuracy, effectiveness, feasibility, acceptability, cost and impact of WHO-recommended diagnostic tools for TB.

WHO will convene a Global Consultation in 2020 to inform Member States, technical partners, donors and civil society on the key changes in the updated guidelines. The meeting will aim to support countries to update their national guidelines, inform program budgets and enable monitoring systems to facilitate rapid transition to more effective diagnosis and treatment for TB and drug-resistant TB patients.

Acknowledgements

We gratefully acknowledge the work of the GDG members, systematic reviewers, consultants with additional technical expertise, data contributors and - most importantly – the TB patients who participated in the studies.

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