

PRACTICAL CONSIDERATIONS FOR IMPLEMENTATION OF TRUENAT



Truenat™ is a rapid, chip-based real-time PCR testing technology for detection of TB and rifampicin resistance. In January 2020 WHO issued a [Rapid Communication on Molecular assays as initial tests for the diagnosis of tuberculosis and rifampicin resistance](#), in advance of policy guidance expected in Q2 2020 that will include recommendations on use of Truenat.

Available Truenat chips for detection of TB include the Truenat MTB chip and a more sensitive Truenat MTB Plus chip. For samples found to be MTB positive, the Truenat MTB-Rif Dx chip may be used as a follow-on test for detection of rifampicin resistance, using the same DNA eluted from the sample. The manufacturer of Truenat is Molbio Diagnostics Pvt. Ltd. (Goa, India).

Conclusions from the [WHO review](#) on performance¹ of Truenat:

- The performance of Truenat MTB, MTB Plus and MTB-RIF Dx assays show comparable accuracy with Xpert® MTB/RIF and Xpert® MTB/RIF Ultra (Cepheid, USA) for TB detection and for sequential rifampicin resistance detection.
- The Truenat MTB and MTB Plus assays also show comparable accuracy to the TB-LAMP assay (Loopamp™, Eiken Chemical Company Ltd, Japan) as replacement tests for sputum smear microscopy.
- The data for Truenat MTB-Rif Dx show similar accuracy to Genotype MTBDR_{plus} (Bruker/Hain Lifescience, Germany) and Genoscholar™ NTM+MDRTB (Nipro, Japan) line probe assays.

TEST PROCEDURES

The test involves three main procedures:

1.) Sample preparation using:

- **Trueprep® AUTO MTB Sample Pre-treatment Pack**, which liquefies the sputum specimen and lyses cells

2.) Extraction and purification of DNA using:

- **Trueprep® AUTO Universal Cartridge Based Sample Prep Kit**
- **Trueprep® AUTO Universal Cartridge Based Sample Prep Device**

3.) PCR amplification and fluorescent probe-based detection using:

- **Truenat chips**
- **Truelab™ Dx Real Time Quantitative micro PCR Analyzer**

EQUIPMENT AND REAGENTS REQUIRED



3 reagent packs (Sample Pre-treatment, Sample Prep, and Truenat chip packs)



Trueprep Sample Prep Device

- Running time: 20 minutes



Truelab Micro PCR Analyzer

- Available with 1, 2 or 4 chip ports (Uno, Duo, Quattro)
- Micro-pipette included
- Running time: 35 minutes

¹ Interim analysis of data from a field evaluation study involving 744 patients (out of 1,866 enrolled) in 4 countries: overall sensitivity was 83% for Truenat MTB and 89% for MTB Plus compared to culture. Specificity was 99% for MTB and 98% for MTB Plus. Specificity was 99% for Truenat MTB and 98% for MTB Plus. Of 163 participants with Truenat MTB and MTB Plus positive results, sensitivity of Truenat MTB-Rif Dx for rifampicin resistance detection was 93% and specificity was 95%

OPERATIONAL CONSIDERATIONS

Positioning: Truenat can be positioned in laboratories as low as microscopy center level given:

- In-built batteries allow for testing without power for up to 8 hours. Note that power is still required to charge batteries as well as to possibly cool the storage room for test chips (storage at $\leq 30^{\circ}\text{C}$)
- Equipment can be used at relatively high room temperatures: $\leq 40^{\circ}\text{C}$ (for reference, GeneXpert operating temperature: $\leq 30^{\circ}\text{C}$; TB-LAMP HumaLoop: $\leq 40^{\circ}\text{C}$)
- Equipment can be used in humid settings (relative humidity: 10-80%)
- Equipment can be used in dusty settings (PCR analyzer does not require air intake)
- Need for minimal biosafety precautions, similar to microscopy, Xpert or LAMP
- A carrying case is also available, allowing for portability for active case finding purposes

Testing capacity per Truelab Micro PCR Analyzer (optimal throughput, as per manufacturer):

- **Truelab Uno** with 1 chip port: performs 10-12 tests in 8 hours
- **Truelab Duo** with 2 chip port: performs 20-24 tests in 8 hours
- **Truelab Quattro** with 4 chip port: performs 40-48 tests in 8 hours. Settings with a Quattro analyzer should procure 2 Trueprep devices to match the throughput of the analyzer

Target populations: The scope of the expected WHO policy guidance will include the use of Truenat with sputum specimens as a replacement for smear microscopy for detecting TB among people being evaluated for pulmonary TB. It is unknown whether the anticipated WHO recommendations will include particular use cases for testing children or people living with HIV or for detecting extrapulmonary TB.

Storage conditions: Truenat chips: 2 - 30°C ; Sample Pre-treatment Pack and Prep Kit: 2 - 40°C

Shelf-life: All test reagents: 2 years

Results reporting: The Truelab Analyzer has in-built connectivity allowing for transmission of results via SIM card, wifi or Bluetooth, as well as other connectivity applications. A Truelab microprinter is also an option.

Needs for training: The test procedures require multiple hands-on steps as well as micro-pipetting. Technicians should be properly trained on all procedures and proficiency maintained, and on-site SOPs and other job aids will be needed.

Multiplexing: Truenat tests for HPV and HCV are undergoing review by the WHO prequalification department; [many other tests are also available](#).

Service and maintenance: The manufacturer offers warranty packages, including possible on-site visits for comprehensive service and maintenance.

Pricing: Global pricing is not yet available. Discussions are underway with Stop TB Partnership's Global Drug Facility (GDF) for inclusion in the GDF Catalog. The manufacturer is currently offering prices based on sizes of orders. The manufacturer may also consider reagent rental agreements when test volumes are committed.

Selection of tests: While countries should consider the advantages and disadvantages of using Truenat versus other WHO-recommended rapid molecular tests for TB detection including GeneXpert or TB-LAMP, a country does not need to necessarily select only one test to meet its needs for rapid testing. Positioning Truenat or TB-LAMP in laboratories at lower levels than GeneXpert can increase patient access to rapid testing. Note that adoption of any molecular test including Truenat does not eliminate the need for sputum smear microscopy, as microscopy is still required for monitoring treatment of TB patients.

A Truenat Implementation Guide is currently under development by Stop TB, USAID and GFI and will include practical guidance for countries interested in adopting Truenat and learning from the experience of early users.