For many years, health implementers in high TB endemic areas with limited resources have been struggling with TB control primarily due to the lack of a rapid and reliable diagnostic test for HIV-associated TB and MDR-TB. The recent endorsement of the GeneXpert MTB/RIF, a fully-automated cartridge based nucleic acid amplification test (NAAT), by the World Health Organization in December 2010 was welcomed with enthusiasm by both implementers and donors. This novel tool reliably detects TB DNA in sputum specimens in less than two hours with 94.4 percent sensitivity for the simultaneous detection of rifampin resistance with 98.3 percent specificity (Lancet, 2011).

But many questions have since arisen as to how best integrate this technology along with existing TB diagnostic tools in countries. Experts from the Stop TB Department of the WHO, the San Raffaele Scientific Institute, the Instituto de Medicina Tropical Alexander von Humboldt, and the Foundation for Innovative New Diagnostics (FIND) discuss the feasibility of implementing GeneXpert MTB/RIF in countries with limited resources with GHDonline members from around the globe.

Key Points

- The primary consideration for integrating the GeneXpert MTB/RIF into country diagnostic and treatment plans will be knowledge of the epidemiology of TB and HIV in each setting.
- The GeneXpert MTB/RIF does not replace the need for acid-fast bacilli (AFB) microscopy, culture, and DST capacity.
- Good referral mechanisms of specimens from lower (peripheral laboratories) to intermediate level laboratories (e.g., District level) are needed for all persons at risk of MDR and HIV-associated TB. GeneXpert MTB/RIF is recommended as the initial diagnostic test for these two risk groups but can be performed as an add-on test to AFB microscopy where resources allow.
- GeneXpert MTB/RIF implementation should be gradual and not disrupt the functioning and quality assurance of existing microscopy laboratory networks.
- For Mark Perkins, the first strategy question is whether GeneXpert MTB/RIF should be used primarily to improve case detection or to screen for MDR-TB.
- A national diagnostic algorithm should be developed in partnership between the National Tuberculosis Control Programme (NTP), the National Reference Laboratory (NRL) and with Ministry of Health representatives. The algorithm must consider the local epidemiology of tuberculosis, HIV-associated TB, drug resistant TB, as well as the diagnostic and treatment capacity at all levels of the health system.
- Further testing for confirming MDR should be available especially in settings or populations with low prevalence of rifampicin resistance e.g. among new TB cases in low MDR-TB settings with no previous TB treatment history. In these instances, confirmation of rifampicin resistance should be performed using a line probe assay or conventional culture and first-line DST. Culture and second line drug susceptibility testing (DST) is still needed for all strains found resistant to rifampicin.
- The GeneXpert MTB/RIF targets sequences in the rpoB gene that are specific for M. tuberculosis. Non-tuberculous mycobacteria are not detected by the assay.
- Each GeneXpert MTB/RIF contains the following internal controls in each cartridge: 1. The sample processing control (SPC) contains non-infectious spores in the form of a dry spore cake that is included in each cartridge to verify adequate processing of MTB; Verifies that lysis of MTB has occurred if the organisms are present; Verifies the specimen processing is adequate; Detects specimen associated inhibition of the real-time PCR assay; SPC should be positive in a negative sample and SPC can be negative or positive in a positive sample; SPC passes if it meets assigned acceptance criteria.
- 2. The Probe Check Control (PCC) is a check undertaken before the start of the PCR reaction. The system measures the fluorescence signal from the probes to monitor bead rehydration, reaction-tube filling, probe integrity and dye stability. PCC passes if it meets assigned acceptance criteria.
- In South Africa, MSF has opted to run positive samples (smear +++) and a negative control on a monthly basis as part of IQC while we wait for the WHO validation panels (see “Developments below”).
- Disposal of used cartridges: The WHO currently advises to dispose of the cartridges in the same manner as sputum cups. Environmentally sound methods of burning/recycling/disposing are being explored.
- The WHO recommends that liquid culture with rapid speciation using a lateral immunoflow test for identification (e.g. Capilia test) is the gold standard method for mycobacterial culture. Biochemical testing can be used to confirm MTB when at least two different tests including inhibition by PNB, nitrate reduction, niacin accumulation and/or catalase production are performed. But, as notes a member from the Imperial College London based in Peru, “Using MGIT cultures in South Africa Chihota et al (IUTLD 2010; 14 (8): 1024-1031) demonstrated that the sensitivity and specificity of demonstration of cording for identification of MTB was identical to the use of MPB64 testing (99.0% and 97.9% respectively) confirming the findings of all groups who have published their work on MODS, including our own, who have argued that this is a reliable and safe method for differentiating M tuberculosis from MOTT/NTMs.”

Target population

- GeneXpert MTB/RIF should be the initial diagnostic test for TB for persons at risk of MDR-TB or HIV-associated TB.
- Should be indentified based on country priorities for TB control and local epidemiology
- Vulnerable groups such as prisoners, women, irregular migrants, or Injecting Drug User (IDU) should be considered.

Selection of sites

- One or few implementation sites in a particular setting should be selected based on their epidemiological characteristics, capacity to implement the technology, and the ability for data collection to gather evidence for sustainability and wide-spread scale-up.
- GeneXpert MTB/RIF is expected to have greatest impact when used in settings where patients can be tested while waiting and where therapeutic decisions can directly follow. The WHO recommends placing GeneXpert MTB/RIF at intermediate level
laboratories (e.g. district level) within the TB laboratory network. Placing the technology at the lowest levels of the laboratory network may result in underutilization. Placement at intermediate level laboratories can allow for referral of samples from persons at risk from lower level laboratories and thus be accessible to and benefit a larger population. In countries where liquid cultures and LPAs are routinely performed at national or regional level, and according to national algorithm, the appropriate placement of the GeneXpert TB/MDR may be the district level.

- Sites must have continuous stable power supply. Uninterruptible power supply (UPS) devices, generators or rechargeable 12V batteries should be considered in settings with irregular power supply. Solar power can also be used as currently done in some settings in Uganda.
- Security measures to guard against computer theft should be considered.
- Temperature for storage of the cartridges should be below 30 degrees Celsius.
- The manufacturer recommended ambient operating temperature for the GeneXpert instrument is currently limited to a maximum of 30°C although GeneXpert MTB/RIF has been successfully used in laboratories where temperatures were over 40°C. When ambient temperatures are too high the device may be unable to thermocycle (rapidly heat and cool, which drives the amplification reaction) optimally and may give an error message. Similarly, the device cannot operate at temperatures that are too low.

- In Afghanistan, GeneXpert MTB/RIF will most likely be placed at the regional laboratory level. (“There will be 8 regional diagnostic labs in Afghanistan, and 8 systems are a feasible number to buy and maintain.”)
- In Brazil, GeneXpert machines are going into health centers without laboratory capacity and being run by nurses. In other countries like South Africa, a substantial amount of regional centralized testing is ongoing or planned.
- In Ethiopia, regional labs are not in the same compound with a hospital which necessitates the transport of samples. Transporting the samples increases the time gap between sample delivery and results dispatching.
- The system has not been tested at high altitude.

Supply Chain
- Given the number of tests and the space needed for delivery and storage and the cost of custom clearance, it is necessary to establish an appropriate plan for stock and supply management.
- It is expected that the cost of GeneXpert MTB/RIF will be further reduced as volumes of tests performed increases. Still, implementation will have important budgetary implications especially in the short term.
- Where to purchase the GeneXpert instrument and GeneXpert MTB/RIF assay? [FIND negotiated prices for GeneXpert® MTB/RIF. Country list, and contact information]

Developments
- WHO monitoring of Xpert MTB/RIF roll-out: Orders of GeneXperts and Xpert MTB/RIF cartridges: [Mapping project with FIND]
- Several studies are ongoing for non-respiratory samples.
- Work is being done to address temperature issues.
- Discussions are ongoing with Cepheid, the manufacturer of the assay, to make the system more accessible with additional languages and/or the use of symbols.
- The Global Laboratory Initiate Core-Group recommends the use of a validation panel of artificial sputum samples containing heat-killed MTB organisms to be shipped with each new GeneGeneXpert and following calibration of individual modules so that modules can be validated before routine test results are reported. The WHO is coordinating the initial development of validation panels. [This QA program is expected to be fully operational from July 2011.]
- [A study published by the Lancet in 2011] revealed that GeneXpert detected twice as many pediatric cases of MDR-TB than did sputum microscopy along

Key References
- Rapid implementation of the GeneXpert MTB/RIF diagnostic test. Technical and operational “how-to” Practical considerations, Stop Partnership, WHO. April 2011

Enrich the GHDonline Knowledge Base
Please consider replying to this discussion with the following information
- If roll-out of GeneXpert MTB/RIF is underway in your laboratory or health center, please share your experience.