Gene Xpert in diagnosis of female genital tuberculosis: an upcoming modality

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Abstract
Female genital tuberculosis (FGTB) poses a great challenge for treating clinicians in terms of diagnosis and treatment in a woman of reproductive age. It causes significant morbidity or short and long term sequelae, especially infertility for the affected women.The disease often remains silent or may present with non-specific symptomatology. As a result, the prevalence of genital tuberculosis is largely underestimated. Incidence of patient's with FGTB in infertility varies from 3% to 16% in India**.** A high degree of suspicion aided by intensive investigations is important in the diagnosis of the disease. Any method that is used to diagnose GTB should be highly sensitive to diagnose the disease reliably in its early stage, so that treatment may improve the prospects of cure before the tubes are damaged beyond recovery. In recent years, Gene Xpert MTB/RIF technique has evolved as a useful and rapid technique for the diagnosis of pulmonary and extra-pulmonary tuberculosis (EPTB).

Keywords: FGTB, Infertility, Gene Xpert MTB/RIF, EPTB.

Introduction
According to Global TB Report 2016, there were an estimated 10.4 million new tuberculosis (TB) cases worldwide (3.5 million in women), with 2 milliondeaths.(1) India has one of the highest incidences of TB in the world.It is estimated that 5-10% of infertile women over the world has genital tuberculosis although this varies from less than 1% in the United States to nearly 18% in India.(2) It is therefore, suggested that every patient consulting for infertility in developing countries should be investigated for female genital tract tuberculosis (FGTB).

Unlike pulmonary tuberculosis, the clinical diagnosis of genital tuberculosis is difficult because in majority of cases, the disease is either asymptomatic or has varied clinical presentation. Routine laboratory investigations like microscopy and culture are of little value in the diagnosis. An absolute diagnosis cannot be made from characteristic features in hystero-salpingography (HSG) or laproscopy. Due to paucibacillary nature of genital tuberculosis, diagnosis by mycobacterial culture has limitations and low detection rate. Polymerase chain reaction (PCR) has highest sensitivity as compared to other methods for diagnosis of tuberculosis but due to false positive results, specificity is low.(3) Only histo-pathological evidence in premenstrual endometrial tissue biopsy can provide diagnosis with certainty.

Alarming increases in multi-drug resistance tuberculosis (MDR-TB) incidence, the global emergence of extensively drug resistant tuberculosis (XDR-TB) leading to rapid mortality in MDR-TB and XDR-TB patients with HIV co-infection have highlighted urgent need for rapid screening method which is 'accurate', 'feasible', 'affordable' for use in resource limited setting.(4) The empirical use of anti-tubercular therapy (ATT) in infertile women without any diagnostic evidence has leads to resistance. Thus, a diagnostic test which is less time consuming with high sensitivity and specificity is required.

Nucleic acid amplification techniques for tuberculosis
During the 1990s, several nucleic acid amplification (NAA) techniques evolved that dramatically altered the way in which we can detect and identify *M. tuberculosis.* Unlike the signal amplification technique, which relies on large numbers of organisms for detection and, thus, is useful only for cultured isolates, NAA techniques allow for detection of *M. tuberculosis* from samples containing relatively few *M. tuberculosis* bacilli. For this reason, NAA techniques can be utilized to identify *M. tuberculosis* directly from clinical specimens, avoiding the most time-consuming aspect of *M. Tuberculosis* identification, the time required to culture the isolate.(4)

Nucleic acid amplification technique (NAAT) methods detect Mycobacterial DNA or RNA directly from the specimen's. Several molecular methods have been developed for direct detection, identification, and susceptibility testing of Mycobacteria. These methods can reduce the diagnostic time from weeks to days. Examples of these include the Enhanced Mycobacterium tuberculosis Direct Test (E-MTD; Gen-Probe, San Diego, CA) and the Amplicor Mycobacterium Test (Amplicor; Roche Diagnostic Systems, Inc, Branchburg, NJ).(4)

WHO Recommendations
Gene Xpert MTB/RIF is a single test recently endorsed by WHO in 2004. The development of the XPERT MTB/RIF assay for the Gene Xpert platform was completed in 2009. WHO recommended use of Xpert assay by 'December 2010'. It is an important

breakthrough in fight against tuberculosis and has been successfully used in pulmonary TB diagnosis, due to the affordability, rapidity and acurancy.[5]

WHO has issued recommendations about using of Gene Xpert MTB/RIF to diagnose extra pulmonary TB and to detect Rifampicin resistance.

Using Gene Xpert MTB/RIF to diagnose extra pulmonary TB in infertile women

Gene Xpert MDR/RIF is a cartridge-based test on NAAT [nucleic Acid Amplification Technique]. It represent major advance in diagnosis of tuberculosis with use of amplification system, nucleic acid sequences unique to MTB can be detected directly in clinical specimen offering better accuracy than microscopy and greater speed than culture.[6]

Gene Xpert test produces rapid, accurate and reliable TB results. They are specific to DNA of MTB, also can detect multidrug resistant MTB, which is an important aspect in TB patient management.[5] In 2015, WHO recommended the use Gene Xpert on both pulmonary and extra-pulmonary specimens due to its high sensitivity and specificity instead of conventional microscopy smear.[7] The MTB/RIF test can effectively be used in low-resource settings to simplify patient's access to early and accurate diagnosis, thereby potentially decreasing morbidity associated with diagnostic delay, drop-out and mistreatment.[4]

Predictive value of Gene Xpert MTB/RIF

The sensitivity of Xpert MTB/RIF in detecting TB is quite high (88%).[5] Gene Xpert if positive on endometrial biopsy is a reliable test for FGTB and treatment can be started on its basis.[8]

The negative predictive value (NPV) is greater than 98% both in settings with a low prevalence of TB and in those with a high prevalence of TB; that is, a negative result accurately excludes TB in most situations.

When Xpert MTB/RIF does not detect *M, tuberculosis*, the disease can be ruled out in most cases unless there is still a strong suspicion of TB.[5]

Fig. 1: A Gene-Xpert Machine

Fig. 2: Gene Xpert instruments with 1, 2, 4 and 16 modules

Public health impact of Gene Xpert MTB/RIF

The widespread introduction of new diagnostic testing platforms will allow TB to be diagnosed early and accurately. Less advances forms of TB will be diagnosed; treatment delays will be reduced; disease transmission will decrease; case-fatality rates will decrease; adverse sequelae will be prevented; and patient outcomes will improve.

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