A New PCR Test For The Rapid Diagnosis of Tuberculosis

Paul A. Granato, Ph.D.

Tuberculosis (TB) caused by Mycobacterium tuberculosis complex (MTBC), is a global disease of pandemic proportions infecting over one third of the world's population or 2 billion people. In the United States, only 9,954 cases of TB were reported in 2012 which represents the first time that the number of yearly reported cases of TB has fallen below the 10,000 mark since tracking began over 60 years ago.

A problem associated with the complete eradication of TB in the U.S. has been the emergence of strains of TB that are multi-drug resistant, often called MOR-TB. MDR-TB is defined as any strain of Mycobacterium tuberculosis that is resistant to two of the most effective drugs used to treat TB, isoniazid and rifampin.

The emergence of MOR-TB following the AIDS outbreak in the 1990s along with other developments suggests that TB will continue to be a public health threat in the U.S. For instance, the incidence of TB increased in foreign-born individuals in the U.S. from 29% in 1993 to 63% in 2012. The countries with the highest incidence of foreign-born TB cases were Mexico, the Philippines, India, Vietnam, and China where the prevalence of multi-drug resistance is 1.8%, 5.6%, 4.3%, 3.7% and 6.4% respectively.

The problem of MOR-TB has been further complicated by the emergence of an even more drug resistant TB strain, called extensively drug resistant tuberculosis or XOR-TB. XOR-TB strains are not only resistant to isoniazid and rifampin but also to any fluoroquinolone such as ofloxacin, and at least one of three injectable second line drugs (amilracin, kanamycin, or capreomycin). With foreign-born cases making up an increasing proportion of U.S. cases and with the incidence of MOR-TB and XOR-TB increasing worldwide, particularly in the countries that are a higher source of immigration to the U.S., it seems likely that the problem of TB and drug-resistant TB in the U.S. will continue to persist, if not increase, in the foreseeable future.

Recently, a significant technologic advance has been made with the availability of nucleic acid amplification test (NAAT) for the rapid and highly reliable diagnosis of TB and MOR-TB infections. The NAAT, called the XPERT MTB/RIF Assay, detects DNA specific for MTBC and genetic mutations in the rpoB gene that is associated with rifampin resistance. The presence of the rpoB gene is a surrogate marker for MOR-TB and XOR-TB that has a sensitivity and specificity of 95% and 99% respectively. If the rpoB gene is not detected by the assay and MTBC is detected, the isolate is susceptible to the conventional drugs used for therapy.

Traditional methods for the laboratory diagnosis of TB involve the use of microscopy and culture. Microscopy is very insensitive whereas the cultural isolation and

identification of MTBC from a clinical specimen may require several weeks to several months. Because of this, in 2008, the Association of Public Health Laboratories and CDC convened a panel that recommended NAA testing as standard practice in the United States to aid in the initial diagnosis of patients with suspected TB. On the basis of the panel report and consultation with the Advisory Council for the Elimination of TB, CDC published revised NAA guidelines, including a detailed testing and interpretation algorithm for initial diagnosis. Recent studies further support NAA test use in the United States to avoid delays in diagnosis and treatment, especially for patients with suspected TB and sputum smears negative for acid-fast bacilli on microscopy. Because of rapid results, NAA testing can help avoid unnecessary respiratory isolation, treatment, and contact investigation of patients without TB and can contribute to system cost savings in patients with HIV infection, homelessness, or substance abuse, compared with smear microscopy alone.

CDC recommends that NAA testing be performed on at least one (preferably the first) respiratory specimen from each patient suspected of pulmonary TB for whom a diagnosis of TB is being considered but has not yet been established, and for whom the test result would alter case management or TB control activities. The recommendation emphasizes the need for NAA testing in the initial diagnosis and for triaging public health interventions such as contact investigations and infection control decisions. Parallel guidance for the use of NAA TB testing in patients infected with HIV has been published. NAA testing does not replace the need for culture; all patients suspected of TB should have specimens collected for mycobacterial culture.

In January of 2013, the Microbiology department of Laboratory Alliance of CNY began routinely performing the Xpert MTB/RIF assay as a new service for the detection of MTBC and MOR-TB patient sputum specimens. Ideally, the specimen of choice for testing is a first-morning expectorated sputum. The Xpert MB/RIF assay will only be performed on one patient sputum specimen even though more than one sputum specimen might be submitted to the laboratory. The NAAT can be completed with the results reported within several hours of specimen receipt and, as such, offers the most highly sensitive and rapid method for the detection of MTBC and MDR-TB in sputum specimens. Microscopy and mycobacterial culture will continue to be performed on these specimens as an adjunct to the NAAT. If additional information is needed or if one has questions, please feel free to contact me at paulgranatophd@LACNY.com .