

New whole blood assay may help overcome roadblocks to TB eradication

January 6 2015

One of the roadblocks to the eradication of tuberculosis (TB) is the difficulty in identifying patients with latent TB infections (LTBI). Neither the tuberculin skin test (TST) nor interferon-gamma release assays (IGRAs) are capable of distinguishing active from latent infection or predicting the chance of reactivation. A new multiple-target, real-time reverse transcription-PCR (real-time RT-PCR) TaqMan assay targeting eight human immune markers can differentiate active pulmonary TB from LTBI, according to a study in the *Journal of Molecular Diagnostics*.

"The World Health Organization reports that one third of the world's population is latently infected with *Mycobacterium tuberculosis* (MTB). It has been estimated that in 5% to 10% of LTBI individuals, the infection progresses to an active disease state, and the conversion rate is greater in immunosuppressed individuals such as those with HIV," explains Hyeyoung Lee, PhD, of the Department of Biomedical Laboratory Science, College of Health Sciences, Yonsei University (Republic of Korea). "Therefore, rapid diagnostic tests and effective treatment of LTBI are important to reduce and control the TB burden."

In previous work, the researchers quantified interferon- γ (IFN- γ) mRNA expression levels as an indicator of IFN- γ levels in an IGRA test. However, the results of IFN- γ RT-PCR showed poor specificity and sensitivity, and the test could not be used to distinguish between active and latent TB.

With these results in mind, the investigators developed a multiple-target

RT-PCR TaqMan assay that could target eight human immune markers: Th1-type factors (IFN- γ , TNF- α , and IL-2R), Th2-type cytokines (IL-4 and IL-10), and IFN- γ -induced chemokines [CXCL9 (MIG), CXCL10 (IP-10), and CXCL11 (I-TAC)]. MTB-specific, antigen-dependent mRNA expression levels were measured in blood samples from 28 patients with active pulmonary TB, 22 with LTBI, and 29 non-TB controls.

When five of the human immune markers were evaluated individually, three were found to be suitable for detecting active pulmonary TB: TNF- α , IL-2R, and CXCL10, with sensitivities ranging from 96.43% to 100%. Two individual markers, IL-2R and CXCL10, were able to detect LTBI, with sensitivities of 86.36% and 81.82%, respectively.

To optimize sensitivity, Dr. Lee and her colleagues used the assay to simultaneously detect multiple targets. They found that the combination of TNF- α , IL-2R, CSCL9, and CSCL10 could differentiate active pulmonary TB from healthy controls (P

Citation: New whole blood assay may help overcome roadblocks to TB eradication (2015, January 6) retrieved 25 April 2024 from <https://medicalxpress.com/news/2015-01-blood-assay-roadblocks-tb-eradication.html>

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