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Rapid identification of *Mycobacterium tuberculosis* complex directly from broth culture vials

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Background: *Mycobacterium tuberculosis* complex (MTC) is one of most frequently cause of death among infectious disease worldwide. Although the number of tuberculosis (TB) deaths and the TB incidence rate continue to fall globally, intensive efforts to control TB dissemination are needed to achieve WHO targets for 2030. Rapid diagnosis and early treatment are main measures for detecting and controlling TB dissemination. Despite, molecular diagnosis had revolutionized TB diagnosis, they have slightly less sensitivity than culture-isolation methods and are not available in all sets. In addition, molecular approved tests are mainly restricted to a few clinical samples. Herein, we aimed to improve the detection of MTC by performing rapid immunochromatographic test directly from positive broth culture vials from diverse clinical samples. In addition, colony identification of MTC and non-MTC species were evaluated by MALDI-TOF.

Material/methods: Clinical samples from respiratory tract, urine, tissue and organic fluids submitted to *Mycobacterium* culture between Dec/2015 and Nov/2016 were included. All culture vials flagged positive on automated system (VersaTREK[®], ThermoFisher) were submitted to acid fast staining and solid medium culture using Ziehl-Neelsen and Löwenstein-Jensen, respectively. For rapid diagnosis, 5 mL from the broth culture vial were retrieved and centrifuged in an sterile recipient (3500 rpm during 5 minutes). One hundred microliters of the supernatant were introduced into the sample spot of the immunochromatographic TB Ag MPT64[®] cassette (Standard Diagnostic). Results were compared to the colony identification by mass spectrometry (Microflex[®], Bruker Daltonics). Discordant results were submitted to polymerase chain reaction restriction-enzyme analysis (PRA) of the hsp65 gene.

Results: From 527 clinical samples, 31 were positive on broth culture and acid fast staining (positivity rate of 5,9%). Immunochromatographic method yielded 21 positive results directly from broth culture vials. All of them were confirmed as MTC by MALDI-TOF. None false-negative result was observed.

Among 10 non-MTC isolates, 9 were identified by MALDI-TOF (90%) as *M. gordonae* (2), *M. intracellulare* (2), *M. chelonae* (2), *M. abscessus* (1), *M. kansasii* (1), *M. mucogenicum/ phocaicum* (1) and confirmed by PRA. Only one *M. xenopi* isolate could not be identified by MALDI-TOF.

Conclusions: The immunochromatographic TB Ag MPT64 test could rapid identified MTC isolates directly from broth vials with excellent sensitivity and specificity (100%). It is an alternative for laboratories where molecular diagnosis is not available or when results are discordant from culture-based method. Non-MTC isolates can be reliable identified by MALDI-TOF. Therefore, this study presented rapid alternative methods for clinical laboratories identified *Mycobacterium* species of clinical importance.