

TITLE: NON-TUBERCULOUS MYCOBACTERIA IDENTIFICATION BY THE SYSTEM OF REAL-TIME PCR MULTIPLEX

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ABSTRACT:

The diagnosis of non-tuberculous Mycobacteria (NTM) is currently a challenge to laboratories. With the description of new species of Mycobacteria in recent times and the increase in the number of infections, has been increasingly difficult to accurately identify these species. A main point in identifying in Mycobacteria is differentiating between *m. tuberculosis* and the MNT; However has been increasingly important for the differentiation of species of MNT. Due to the phenotypic and genotypic similarities is required the application of methodologies that best characterized the MNT to determine precisely the species causing infection. The aim of this work was to evaluate a system of multiplex real-time PCR (qPCR multiplex) in the identification and differentiation of non-tuberculous Mycobacteria of clinical importance. 79 clinical isolates were used for culture and the correlation between multiplex qPCR and sequencing, considered gold standard was 91%. In the first step of the qPCR, differentiation of *m. tuberculosis* and MNT sensitivity and specificity were 100 and 90%, respectively. In steps 2 and 3 of the multiplex qPCR had the purpose of identifying the species of MNT characterized in step 1. . Therefore, in step 2, it was identified the species of *M. kansasii*, *M. abscessus* and *M. avium*, with sensitivities of 94%, 80% and 50% respectively. And in step 3 was identified the species *m. fortuitum* sensitivity of 58%. At the end of the stages of the multiplex qPCR could not identify 10 samples, being characterized by sequencing as a species of *M. wolinsky*, three *M. celeriflavum*, four *M. fortuitum* and two; *M. avium*. It can be concluded that the development and use of multiplex qPCR system for differentiation of TB and NTM, and identification of the species of NTM using demonstrated to be an effective and safe technique.

Keywords: Identification. Mycobacteria., multiplex, Real time PCR

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