

P2400 Combined use of commercial and sequencing analysis methods for the identification of non-tuberculous mycobacteria in a tertiary hospital of Athens: eleven years of experience

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Background: Non-tuberculous Mycobacteria (NTM) are environmental bacteria that cause opportunistic infections in humans. The clinical relevance and the optimal treatment regimens of NTM differs strongly by species. We describe our experience regarding the molecular identification of NTM species recovered from clinical specimens in Attikon University Hospital Athens, by using commercial and more advantaged molecular identification methods as sequence analysis.

Materials/methods: Specimens submitted for mycobacterial culture between 12/2006-10/2017 and the recovered NTM were identified by the commercially available DNA strip test Genotype Mycobacterium Common Mycobacteria (CM) and Additional Species (AS) (Hain-Lifescience), two reverse hybridization-based assays. Sequencing analysis of 16S *rDNA* (1500bp) and *hsp65* (440bp) genes was performed when necessary.

Results: In total, 294 NTM strains were recovered; 285 belonged to 31 known *Mycobacterium* species with most frequent the *M. avium* (n= 53), *M. lentiflavum* (n= 45) and *M. goodii* (n=35), while 9 strains were not belonged to any known species. The 84% (247) of strains (17 species) were correctly identified at the species level by the commercial assays. Nineteen (6.5%) strains identified by the commercial assays only to the genus level and belonged to rare NTM species *M.bohemicum*, *M. neoaurum*, *M. arupense*, *M. elephantis*, *M. monacense*, *M. canariense*, *M. agri*, and *M.celeriflavum*. Twenty-two strains erroneously identified by the commercial assays as *M. intracellulare*. Sequencing analysis showed that belonged to the recently described species *M.yongonense*, *M. marseillense*, *M. timonense*, *M. chimaera* and *M. mantonii*, while 7 strains had unique sequences and belonged to *M. avium* complex. Also a *M.kumamotoense* strain was incorrectly identified as *M. celatum*. Finally, two strains which were identified by the commercial assays only to the genus level had unique sequences. Phylogenetic analysis demonstrates that the 9 strains with unique sequences didn't belong to a previously reported species and represented probably novel *Mycobacterium* species which will be documented with further genomic studies.

Conclusions: Our findings suggest that the combined use of molecular commercial identification tests with sequencing analysis improve the ability to correctly identify not only the common NTM but also the rare, the recently described as well as to detect probably the novel mycobacterial species.