

Unusual high-performance liquid chromatography profile of a strain of *Mycobacterium avium*

Mycobacterium avium complex (MAC) is the most frequently detected non-tuberculous mycobacterium, not only in AIDS patients, where it is responsible for disseminated and localized infections, but also in non-immunocompromised subjects. Among the latter, it is frequently responsible for pulmonary disease in elderly people with various predisposing conditions, and for cervical lymphadenitis in the children [1].

Several different approaches can be applied for the identification of MAC: conventional tests reveal well-defined biochemical and cultural features; highly specific commercial DNA probes (AccuProbe, USA) react with a species-specific 16S rRNA stretch; and finally, high-performance liquid chromatography (HPLC) of cell wall mycolic acids provides an easily recognizable pattern characterized by three clusters of peaks (Figure 1b).

Herein, we report the characterization of two strains isolated from sputum collected at 5-month intervals from the same immunocompetent elderly subject. Both isolates had the phenotypic features of MAC, and both reacted with AccuProbe *M. avium*, but not with AccuProbe *M. intracellulare*.

The IS1245-based restriction fragment length polymorphism analysis [2,31] of both isolates yielded different patterns (Figure 2), thus demonstrating the double origin of the infection.

Surprisingly, the HPLC profiles of the two strains were completely different (Figure 1): the one obtained from the second strain (b) is like that established for *M. avium* and for MAC in general [4], whereas the HPLC pattern of the first strain (a) is unique, never seen in our experience with over 180 MAC strains characterized with HPLC and AccuProbe, and never reported for *M. avium*.

The above data therefore add to our present knowledge of the HPLC profile of MAC; the same typical three-clustered profile is shared by strains assigned, on the basis of hybridization with specific AccuProbes, to the species *M. avium* or *M. intracellulare*, and by the members of the so-called MAC intermediates or MAI-X group [5], which react with AccuProbe MAC, but not with either AccuProbe-*M. avium* or AccuProbe-*M. intracellulare*.

A similar anomaly of HPLC profiles has been previously reported in a few isolates of mycobacteria reacting with the MAC probe but not with the species-specific ones [6]; in that case it was, however, possible to establish genetic sequences unrelated to MAC and very close to *M. simiae*. In the present case, hypervariable regions A and B of 16S rDNA [7] yielded a perfect match with the signature sequences of *M. avium* for both isolates.

This is therefore the first report of an *M. avium* strain in which the 16S rDNA species-specific markers do not correlate with the chromatographic pattern typical of this species.

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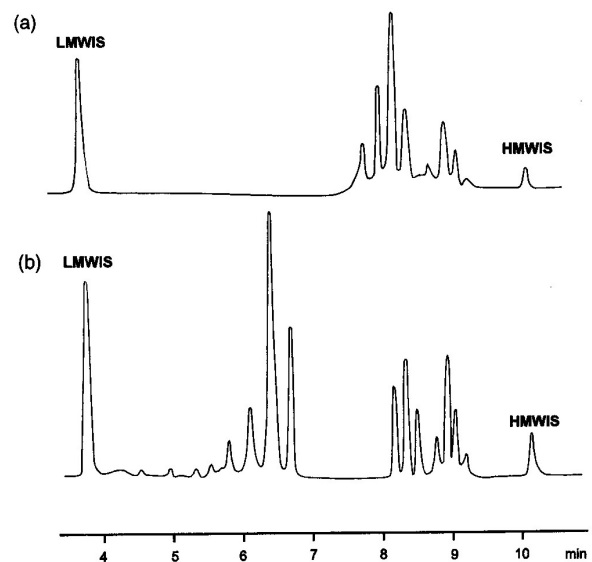


Figure 1 Mycolic acid patterns obtained by HPLC: (a) profile of the first strain (atypical); (b) profile of the second strain (MAC-typical). LMWIS and HMWIS are low and high molecular weight internal standards, respectively.

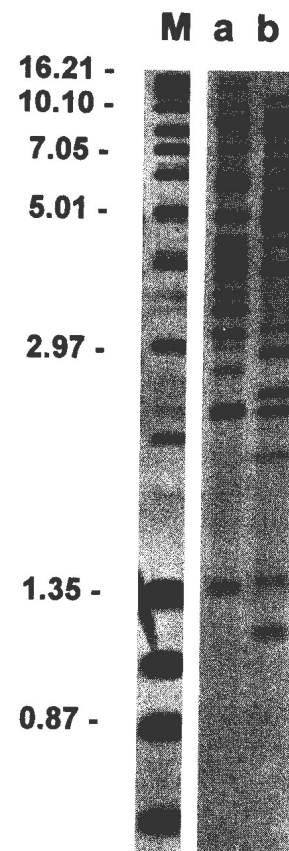


Figure 2 PvuII-generated IS1245-based RFLP fingerprints: M, molecular markers (kbp); a, second strain; b, first strain.

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