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## Spoligotyping followed by double-repetitive-element PCR as rapid alternative to IS6110 fingerprinting for epidemiological studies of tuberculosis.

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**Spoligotyping Followed by  
Double-Repetitive-Element PCR as Rapid  
Alternative to IS6110 Fingerprinting for  
Epidemiological Studies of Tuberculosis**

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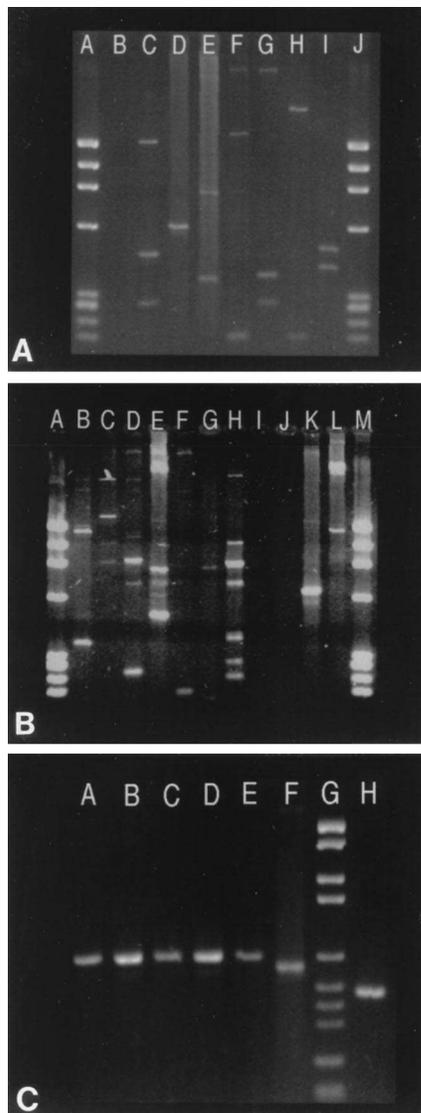


FIG. 1. Representative patterns obtained following DRE-PCR analysis of *M. tuberculosis* clinical isolates. (A and B) Results for isolates that were clustered by neither IS6110-RFLP nor spoligotyping. (A) Lanes: A, molecular weight (MW) marker; B, negative control; C to I, various unclustered isolates (GP14, GP35, GP16, GP5, GP25, GP33, and GP10); J, MW marker. (B) Lanes: A, MW marker; B to H, various unclustered isolates (GP1, PMON, GP34, GP9, A11, GP39, and GP15); I, negative control; J, an unclustered isolate, GP29, that did not generate any bands upon DRE-PCR; K, isolate B4 from cluster B; L, unclustered isolate GP37; M, MW marker. (C) DRE-PCR analysis of isolates that were clustered by IS6110-RFLP analysis followed by spoligotyping. Lanes: A to E, five isolates from four patients of cluster B; F, an unclustered isolate (GP20); G, MW marker; H, an isolate of cluster C. DNA MW markers IX (A and B) and VI (C), from Boehringer Mannheim, Meylan, France, were used.

patient isolates did not give visible bands upon DRE-PCR, which involved 4 unclustered isolates in this study). The use of DRE-PCR as a first-line test cannot be recommended, as it is not sufficiently discriminatory when used alone and both the

low (below 200 bp)- and high (above 3,500 bp)-molecular-size bands may be difficult to interpret and compare. Furthermore, interpretation of the results may be tedious when hundreds of isolates on separate gels are compared. In this sense, initial screening by spoligotyping limits the number of potentially linked isolates prior to DRE-PCR. In conclusion, the strategy described in this article is easily applicable to the handling of a very large number of samples and would be well suited to developing countries and/or countries with high prevalences of tuberculosis.

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