Thr270Ile in *embC* (Rv3793) Is Not a Marker for Ethambutol Resistance in the *Mycobacterium tuberculosis* Complex

We thank Goude et al. for their extensive study of the role of *embC* (Rv3793) in ethambutol (EMB) resistance in *Mycobacterium tuberculosis* (9). Their finding that the Thr270Ile change on its own plays no role in EMB resistance is of particular importance (9). Clinical studies had found this mutation in numerous EMB-resistant but also some EMB-sensitive isolates (13, 15, 16, 19–21). However, given that this mutation usually coincided with additional mutations in *embCAB*, its relevance for EMB resistance remained unclear (15).

We note that this particular *embC* single nucleotide polymorphism (SNP) had been detected in a number of further instances but not discussed in the context of EMB resistance. First, it is present in *Mycobacterium bovis* AF2122/97 and the two closely related vaccine strains *M. bovis* BCG strain Pasteur 1173P2 and BCG strain Tokyo 172, both of which are sensitive to EMB (1, 8, 17, 18, 22). Second, it was found in all *Mycobacterium africanum* West African 1 strains (S444 04 and 11821 03) and West African 2 strains (4141 04 and GM 0981) sequenced as part of the *M. tuberculosis* Phylogeographic Diversity Sequencing Project (3, 7, 10). Third, it occurs in all *M. tuberculosis* Indo-Oceanic lineage 1 strains (K21, K93, T17, T83, and 95 0545), for which valid sequence information is available (7). Fourth, the orthologs of the closest, fully sequenced relatives of the *M. tuberculosis* complex (MTBC) (*M. leprae* TN, *M. avium* 104, *M. avium* k10, *M. ulcerans* Agy99, and *M. marinum*; deposited in the TB Database under gene family cluster ID 360180271) also display an Ile at this position (7). In contrast, all evolutionary “modern” *M. tuberculosis* strains (lineages 2 to 4) have the Thr (2, 4, 5, 7, 11, 12, 14, 23).

This demonstrates that Ile is the ancestral amino acid at position 270 of *embC* in MTBC rather than a marker for EMB resistance, which agrees with the findings by Goude et al. (9). Moreover, the geographic variation in the frequency of Thr270Ile in the aforementioned clinical studies is in line with this conclusion. With 47 and 32% of strains, this mutation was most common in two studies from India (20, 21). Conversely, lineage 1 strains are known not to be frequent in either Germany or Uzbekistan, as confirmed by the fact that Thr270Ile was found only in 3% of strains (6, 10, 15).

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