

**Molecular analysis on resistant TB isolates in Portugal**

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Portugal has one of the highest tuberculosis (TB) incidence (20/100 000 inhabitants) in Europe. The emergence of multidrug resistant (MDR) TB and extensive drug-resistant (XDR) TB infection is the biggest threat to TB control. Most strains of MDR-TB circulating in the Lisbon area belong to a particular family of genetically related strains, the Lisboa family, detected in the 90's. The prevalence of this family of strains has increased over the years, and represented more than 85% of the MDR-TB strains in the year of 2008. XDR-TB has been recently derived from MDR-TB strains and account for about 50% of these, the majority belonging to Lisboa family. Lisboa family represents a serious problem regarding TB control in Portugal and its prevalence in recent years suggests that these strains may have selective advantages over others. In order to establish a link between the most prevalent mutations in drug-resistance associated genes and spread of Lisboa strains in the Portuguese setting, 54 resistant-TB clinical isolates in Portugal were study. The isolates were characterized by 24 loci MIRU-VNTR and analyzed for *inhA*, *katG*, *rpoB*, *rpsL*, *rrs*, *embB* and *pncA* genes, for resistance to first-line drugs. The MDR isolates (n=35) were further analyzed for mutations in *gyrA*, *tlyA*, *eis* and *rrs* for resistance to fluoroquinolones and second-line injectables. MIRU-VNTR analysis showed that Lisboa family strains and Q1 cluster were the most prevalent, with 26 (48%) and 6 (11%) isolates respectively, including the majority of the MDR-TB and XDR- TB isolates. No mutations in first-line drug resistance associated genes specifically related with MDR were found. However, mutation analysis to second-line drug resistance in 17 MDR-TB isolates shown that specific mutations are present in particular families. Therefore, XDR-TB from Lisboa family exhibits *gyrA* D94G/S91P, *tlyA* Ins755GT and *eis* G-10A mutations, and XDR-TB from Q1 presents *gyrA* D94A and *rrs* A1401G mutations. The remaining isolates are still under study, and further analyses are ongoing. We conclude that XDR-TB isolates from Lisboa family and Q1 cluster have shown a marked difference between them, regarding second-line mutations. Such analysis may be useful to define mutation profiles that distinguish Lisboa family from Q1 isolates.