## Molecular analysis on resistant TB isolates in Portugal

C. Silva[a], J. Perdigão[a], L. Jordão[b] and I. Portugal[a]

[a]*Centro de Patogénese Molecular, Av. Prof. Gama Pinto, 1649-003 Lisbon, Portugal;* [b]*Inst. Nac. de Saúde, DDI, Av. Padre Cruz, 1649-016 Lisbon, Portugal*
*carlamsilva@ff.ul.pt*

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.