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High-resolution melting curve analysis for rapid detection of streptomycin and ethambutol resistance in Mycobacterium tuberculosis

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Background: Development of molecular techniques for rapid detection of drug resistant tuberculosis allows for the prompt initiation of appropriate anti-TB treatment. We aimed to assess high-resolution melting (HRM) analysis for the detection of *rpsL*, *rrs* and *embB* mutations to identify streptomycin and ethambutol resistance in *Mycobacterium tuberculosis.*

Material/methods: A total of 76 clinical isolates of *M.tuberculosis* including 25 SM-R, 21 EB-R and 30 drug susceptible (determined by the proportion method of drug susceptibility testing (DST)) were analyzed by HRM analysis, and the results were confirmed using DNA sequencing.

Results: The sensitivity and specificity of the HRMA compared to phenotypic DST were 88% and 100.0% respectively for the detection of streptomycin resistance(SM-R), and 90.4% and 96.6% respectively for ethambutol resistance (EB-R). Three SM-R and two EB-R isolates had no mutations in the studied regions of *rpsL*, *rrs* and *embB* genes determined by DNA sequencing and therefore were not identified as resistant by HRM assay. Interestingly, one phenotypic EM-S isolate was found by sequencing to have a mutation at codon 423 (Met →Ilue) of *embB* gene and was clustered as resistant by HRM as well.

Conclusions: The sensitivity and specificity of HRM curve assay was in consistent with DNA sequencing which is the gold standard method for genotypic DST. This assay can be utilized as a

screening method for detection of drug-resistant tuberculosis offering the advantages of high throughput, single step, cost effectiveness, and rapid work flow method.