DETECTION OF RESISTANT *MYCOBACTERIUM TUBERCULOSIS* STRAINS TO ISONIAZID AND RIFAMPIN, RIFAMIPICIN USING DNA SEQUENCING

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Introduction
The emergence of drug resistant strains of *Mycobacterium tuberculosis* is an increasing problem in developed and developing countries. Early detection is essential for the efficient treatment and control of drug resistant tuberculosis.

Objective
To determine the pattern of drug resistance of *M. tuberculosis* from tuberculosis patients attending the Central Chest Clinic, Kandy and to develop a simple and rapid assay based on PCR and DNA sequencing, targeting the mutations in *rpoB*, *inhA*and, *katG* genes.

Methodology
Acid Fast Bacteria (AFB) positive sputum specimens from patients with tuberculosis were cultured on Lowenstein-Jensen medium. Antibiotic susceptibility tests for isoniazid and rifampin were carried out on LJ/Middle brook 7H10 medium, using the proportion method. DNA extraction from the culture isolates (using CTAB), and directly from the sputum specimen (Boom's method) was carried out. PCR and direct automated sequencing of the PCR products of *inhA*, *katG*, and *rpoB* genes from *M. tuberculosis* culture isolates were carried out.

Results
Of the patients 83.2% were within the economically productive age group of 21 to 60 years. One of the 71 *M. tuberculosis*/*M. tuberculosis* complex isolates showed isoniazid resistance and 23.3% were resistant to rifampin. 1.4% was multi-drug resistant. All the susceptible strains tested for DNA sequence possessed wild-type sequences whereas all the resistant strains tested had novel mutations.

Conclusions
High rate of rifampin resistance was evident among the tuberculosis patients in Kandy. This emphasizes the need for a rapid and reliable method of diagnosing drug resistance among the tuberculosis patients in this country.