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**DETECTION OF *IS6110* (INSERTION SEQUENCE) IN SERUM OF
TUBERCULOSIS PATIENTS REPORTED TO KANDY CHEST CLINIC**

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Tuberculosis (TB) remains a global health challenge, necessitating the development of more sensitive and non-invasive diagnostic tools. This preliminary cross-sectional study explores the potential of detecting the *IS6110* insertion sequence, a unique genomic marker of *Mycobacterium tuberculosis*, in serum as a novel diagnostic approach. The study protocol was approved by the Ethics Review Committee, General Hospital, Kandy. Blood samples were collected from Active TB (n = 20), Latent TB (n = 22), and contact tracing (n = 28) patients attending to the Kandy chest clinic. RNA was extracted from the serum samples using the guanidium thiocyanate-phenol-chloroform extraction method. Then extracted RNA was quantified and subjected to complementary DNA (cDNA) synthesis using GoScript™ Reverse Transcription System according to the manufacturer's guidelines. A conventional Polymerase Chain Reaction (PCR) was carried out using 6 ng of template cDNA with *IS6110* insertion sequence-specific primer pairs Pt8: 5'-GTGCGGATGGTCGCAGAGAT-3', Pt9: 5'-CTCGATGCCCTCACGGTTCA-3'. The H37Rv bacterial DNA was used as a positive control for this study. The amplified PCR products were run on 2% agarose gel along with 100 bp ladder and the negative control. The positive results obtained from the conventional PCR amplifications were confirmed with real-time PCR by using the primer pairs specific for *IS6110* insertion gene, INS1, F: 5'-CGTGAGGGCATCGAGGTGGC-3' and INS2, R: 5'-GCGTAGGCGTCGGTGACAAA-3'. The results revealed that the *IS6110* gene was detected in 45% of serum samples (9 out of 20) collected from active TB patients. Latent TB and contract tracing patients did not possess the *IS6110* gene in their serum samples. In conclusion, the serum samples in this study did not demonstrate the potential to detect the MTB-specific mRNA transcript in individuals with latent TB (LTB) and contact tracing (CT). However, it may suggest detecting the bacterial transcript in active TB patients. Nevertheless, further improvements are necessary for it to be considered a reliable biomarker for active TB.

Keywords: Active tuberculosis, *IS6110* insertion sequence, latent tuberculosis, polymerase chain reaction, and serum

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