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BENCHAPRON CHINRAT : EFFICIENCY OF PCR FOR DETECTION OF *MYCOBACTERIUM TUBERCULOSIS* FROM SPUTUM IMPREGNATED ON FILTER PAPER AT ROOM TEMPERATURE FOR 5 DAYS. THESIS ADVISORS : UNCHALEE TANSUPHASIRI, M. Sc. (Micro). SOMSAK RIENTHONG, M. Sc. (Med. Micro). 150 p. ISBN 974-663-167-5

With the increasing incidence of tuberculosis, more rapid detection of *M. tuberculosis* has become an important issue that may improve management of infected patients and facilitate infection control procedures. In practice, sputum specimens are sent to central laboratories for culture. However, the shipment of sputum specimens over a long distance from peripheral health centers to central laboratories may affect the culture recovery. Therefore, a rapid PCR-based method was considered in this study since it can detect both viable and non-viable organisms. The efficacy of detection by PCR and by culture from sputum impregnated on filter paper at room temperature for 5 days were investigated. The results were compared with those of staining and conventional culture before storage as the "gold standard".

Out of 231 sputum specimens examined, *M. tuberculosis* were recovered from 124 samples by culture before storage. The culture positivity rate was significantly decreased to 70% after 5 days storage. For PCR assay, the sensitivity, specificity and efficiency by first PCR combined with agarose gel electrophoresis were 70, 100 and 84.9%, respectively and with dot blot hybridization were 89.5, 96.3 and 92.6%, respectively. The same values for nested PCR were 96.0, 97.2, and 96.5%, respectively. Of these methods, the nested PCR showed excellent sensitivity and specificity with no significant difference ($p = 0.727$) from conventional culture. Thus the storage of sputum on filter paper and keeping it at room temperature for 5 days had no apparent effect on the performance of nested PCR.

It is recommend this PCR method be used in combination with culture for detection of *M. tuberculosis* from mailing sputum specimens stored on filter paper. This sputum collection and storage has the advantage that samples can be sent by post with a minimum space and without refrigeration, convenience for culture, less culture contamination, and remain viable for PCR assay. Future modifications to the PCR protocol, especially at the sample preparation step and at the detection step will make the assay more reliable, and suitable for use in routine clinical practice or in epidemiological study for analysis of large number of samples collected in the field sent by post to the well-established central laboratories for PCR analysis.