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**USE OF NOVEL SAMPLE PROCESSING METHODOLOGY FOR IMPROVED
DETECTION OF Mycobacterium tuberculosis FROM CLINICAL SAMPLES AND
ITS APPLICATION TO DETECTION OF DRUG RESISTANCE BY MOLECULAR
METHODS**

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OBJECTIVES

1. Comparison of sample processing by the Petroff's method and the universal sample processing solution in detection of AFB by smear microscopy.
2. Comparison of culture positivity of samples processed by Petroff's method and the universal sample processing solution .
3. Detection of drug resistance among M. tb isolates by culture.
4. Detection of M. tb DNA from processed samples by PCR including drug resistance to both first line and second line drugs.

METHODOLOGY

Study design: Crosssectional. Study period: Oct 2010 to Oct 2012. Study subjects: Patients who were clinically suspected to be suffering from tuberculosis attending DOTS centre and patients admitted to hospital for treatment. Samples collected: Sputum, pleural fluid, peritoneal tissue, aspirates, urine, biopsies. All samples were subjected to both the processing methods and ZN smear microscopy, culture on LJ Medium and PCR were done. Samples from which Rifampicine resistant M.tb was cultured were subjected to PCR amplification of rpoB gene and grey gene and subjected to sequencing to detect the mutation possible.

RESULTS

In the study it was found that there was no significant difference between Modified Petroff's Method and Universal sampling Processing Method for processing of samples for detection of M.tb by culture and smear microscopy. However it was found that samples processed by the USP method gave better results with PCR with both the targets as compared to the Modified Petroff's Method.

CONCLUSION

It is concluded that Universal sample processing method can be used for processing of samples for recovery of inhibitor free DNA where molecular detection of M.tb and drug resistance is desired. This method is not recommended for use in routine where facilities for molecular studies do not exist.