Clinical evaluation of IMMY MycoDDR™

Digestion/Decontamination Reagents for the Recovery of Mycobacterium

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ABSTRACT

Tuberculosis (TB) is a serious disease that affects a significant portion of the world's population and is one of the leading causes of death in developing countries. Proper digestion and decontamination of patient samples is a critical step in facilitating the diagnosis of TB. This study aimed to accelerate positive detection of mycobacterium through comparison of digestion and decontamination sample processing reagents. Of the 159 specimens, 10 tested positive for mycobacterium. In 8 of the 10 samples processed using the MycoDDR™ *(3.0% NaOH)* system, positive results were obtained in equal or less time than the paired samples processed by the Alpha-Tec *(3.0% NaOH)* system. These data show that the use of IMMY's MycoDDR™ system provides positive results approximately 1 day earlier on average than processing of samples with the NAC-PAC™ Red system (mean numbers of days to positive culture result: MycoDDR™- 8.4 days vs. NAC-PAC™ Red- 9.6 days). The remaining 2 positive samples were detected after processing with the MycoDDR™ system in an average of 26.6 days and remained negative in the paired samples using the Alpha-Tec system. Additionally, the MycoDDR™ system produced these results using less neutralization buffer to achieve the optimal pH than the NAC-PAC™ Red system.

INTRODUCTION

TB is a serious disease that affects a significant portion of the world's population and is one of the leading causes of death in developing countries. Diagnosis of TB is important not only for correct treatment of the disease but also for containment of the highly contagious infected individual. Currently, the most widely used diagnostics for pulmonary TB benefit from pre-processing of the patient respiratory samples for concentration of the sample and removal of any contaminating organisms that may interfere with the test results. Consequently, processing of the patient sample is a critical step in facilitating the diagnosis of TB. The importance of this sample processing step necessitates that the method be efficient, dependable, and cost effective.

SUMMARY

The MycoDDR™ sample processing system delivers true positive results in less or equal time than the Alpha-Tec NAC-PAC™ Red sample processing system.

The MycoDDR™ sample processing system uses approximately 50% less neutralization buffer, on average, to achieve the optimal pH for survival of mycobacterium and death of normal flora, than the NAC-PAC™ Red system.

RESULTS

1) Positive Results are Obtained in Less Time and Without False Negatives with the MycoDDR™ Sample Processing System

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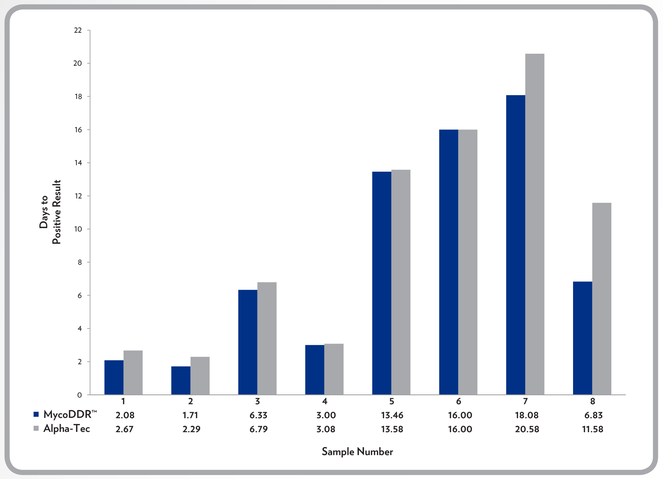

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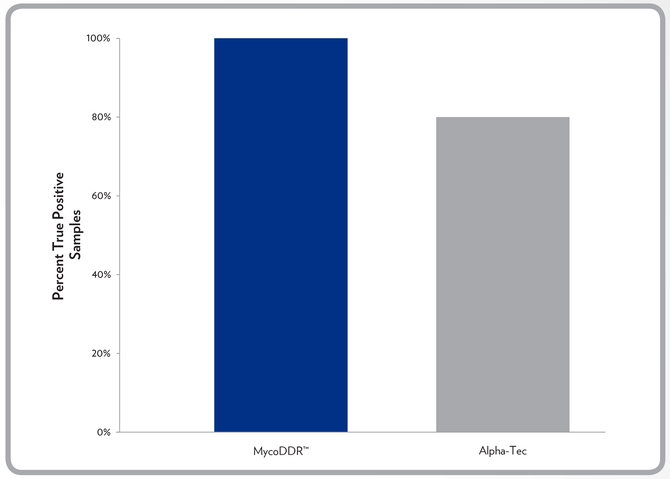

Figure 1. The MycoDDR™ sample processing system provides true positive results faster than the Alpha-Tec NAC-PAC™ Red system.
A) In 8 of the 10 positive samples processed using the MycoDDR™ *(3.0% NaOH)* system, positive results were obtained in equal or less time than the paired samples processed by the Alpha-Tec *(3.0% NaOH)* system.
B) The NAC-PAC™ Red *(3.0% NaOH)* system showed a 20% false negative rate. The remaining 2 positive samples were detected after processing with the MycoDDR™ *(3.0% NaOH)* system in an average of 26.6 days and remained negative in the paired samples using the Alpha-Tec *(3.0% NaOH)* system. The 2 discrepant specimen were confirmed positive for MTB using molecular based testing. n=159 total samples.

2) The MycoDDR ™ Sampling Processing System Uses Less Neutralization Buffer to Achieve Optimal pH

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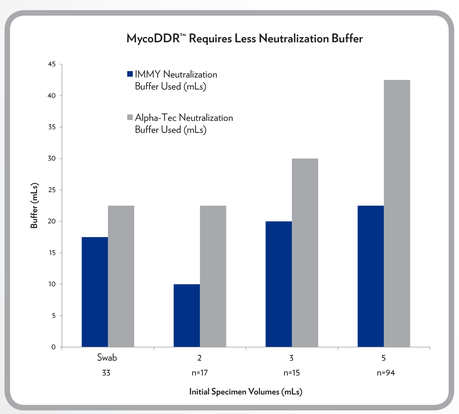

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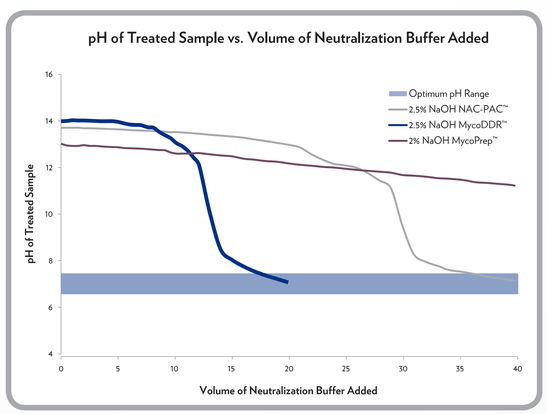

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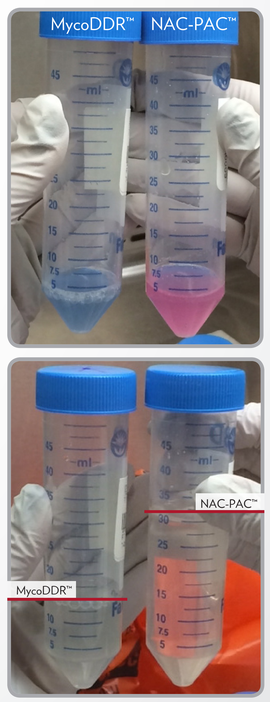

Figure 2. The MycoDDR™ sample processing system uses less neutralization buffer than the Alpha-Tec NAC-PAC™ Red system.
A) The decontamination and concentration processes were followed in accordance with the product procedure for either the MycoDDR™ *(3.0% NaOH)* system or the Alpha-Tec NAC-PAC™ Red *(3.0% NaOH)* system. Less neutralization buffer was required for neutralization of the samples using the MycoDDR™ system, regardless of the starting sample volume or type.
B) The initial pH of 5 mL simulated samples was recorded and then each sample was processed separately using the NAC-PAC™ Red System (grey), the MycoDDR™ system (blue), or the MycoPrep™ system (purple). The final pH and the final volume of each of the samples was evaluated at the end of each respective protocol. The MycoDDR™ system was able to achieve optimal pH using less neutralization buffer than the other systems. *Chart reproduced from results found in Crider 2014, MycoDDR™ White Paper. NaOH concentrations in this chart (2% and 2.5%) do not reflect the NaOH concentration (3%) used in the clinical evaluation study results presented in this poster.*
C) Representative picture demonstrating the difference in neutralization buffer volumes between the MycoDDR™ system and the NAC-PAC™ system.

MATERIALS & METHODS

Specimen Processing
Specimens were received, aliquoted into two 50mL falcon tubes (~3-5ml each), labeled and prepared for processing by either the MycoDDR™ *(3.0% NaOH)* sample processing system or the Alpha-Tec NAC-PAC™ Red *(3.0% NaOH)* system. The decontamination and concentration processes were followed in accordance with the product procedure for each corresponding system. At the end of decontamination and concentration procedures, each specimen was inoculated into solid LJ media and liquid BD MGIT™ media. All cultures were incubated at 35°C and held for a total of 6 weeks. In addition all specimens were transferred to microscope slides for flurochrome and acid fast microscopic examination.

Specimen Evaluation
The evaluation was performed simultaneously for all specimens; cultures and stains were set-up in duplicate, one aliquot was processed using the MycoDDR™ *(3.0% NaOH)* system and one aliquot using the NAC-PAC™ Red *(3.0% NaOH)* system. Solid media cultures were examined weekly for the presence of bacterial growth. Liquid BD MGIT™ cultures are continually monitored and alert the microbiologist if bacterial growth is detected. If either culture phase is detected to have growth the microbiologist confirms the bacterial growth by preparing microscope slides of the specimens to determine if the organisms are acid fast or routine bacterial contamination. If the liquid phase is detected, an additional solid LJ slant is inoculated and incubated to enhance further recovery of the organism. All positive cultures are confirmed using molecular based assays.

OBSERVATIONS & CONCLUSIONS

1. When acid fast bacilli smear and fluorochrome staining was performed on the paired processed samples, the MycoDDR ™ reagent set delivered greater clarity and quality than the Alpha-Tec NAC-PAC™ system.

2. The ability to visualize neutralization in bloody samples was facilitated with the blue pH indicator provided in the MycoDDR™ reagent set.

3. The use of approximately 50% less neutralization buffer, on average, with the use of the MycoDDR™ reagent set ultimately results in a substantial cost savings.

In conclusion, this study demonstrates that the MycoDDR™ *(3.0% NaOH)* system is superior to the Alpha-Tec NAC-PAC™ Red *(3.0% NaOH)* system for the digestion and decontamination of patient samples for quicker positive detection of mycobacterium at a substantial cost savings.

ACKNOWLEDGEMENTS

We would like to thank IMMY for providing the MycoDDR™ kits for this study at no cost.