digestion and decontamination was done by the NaOH (Sodium Hydroxide) -NALC (N-acetyl-L-cysteine) method, followed by centrifugation for 20 minutes at 3000 revolutions/minute. 0.5 ml of each concentrate was added into the 3 media. MGIT vials were monitored by the MGIT 960 system while the Bio FM and LJ media were examined up to 42 days and 56 days respectively for mycobacterial growth. Isolates were identified by P-nitro-a-acetylaminob-hydroxypropiophenone (NAP) testing. 17 of the 20 specimens turned culture positive while three smear negative specimens remained culture negative. All isolates were identified as *M. tuberculosis* complex. Surprisingly, no discrepancy was seen in the recovery rates of the 3 media. The time to mycobacterial detection was shortest with MGIT and longest with LJ media [Table 1]. The detection time of BioFM was comparable with MGIT and outperformed the LJ media, similar to other international study findings.[4,5]

Our pilot study, thus, indicates that BioFM media may have an important role in the treatment of severely ill patients infected with VRE.

**Acknowledgments**

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**References**


**Pilot Evaluation of Commercial Liquid Culture Method for Isolation of Mycobacteria in Resource-Poor Settings**

Dear Editor,

High infrastructural costs and lack of trained manpower make setting up of automated mycobacterial culture facilities difficult. Amongst those offering culture, great disparity exists in techniques used. Though a few reference laboratories and hospitals offer automated rapid culture technology, the high cost prevents their widespread use. A vast majority of mycobacteriology laboratories continue to use the conventional Lowenstein-Jensen (LJ) media. Though the LJ media is highly specific for mycobacteria, the long time taken for growth delays diagnosis and hampers initiation of early treatment.[1,2] Evaluating cheaper and rapid culture methods is, thus, vital in tuberculosis control and management.[3] We undertook a pilot study to compare mycobacterial recovery rates and detection time of the Bio FM culture medium (M/s Bio-Rad Ltd), the mycobacteria growth indicator tube (MGIT) 960 system (M/s BD Ltd.) and LJ medium(M/s EOS labs, Mumbai, India). The Bio FM medium, a Middlebrook 7H9 medium with OADC (Oleic Acid - Albumin Fraction V, Bovine -Dextrose - Catalase (beef) - Sodium Chloride) and VCA (Vancomycin - Colistin - Amphotericin B) supplements, contains a chromogenic indicator that changes to dark blue/violet in response to mycobacterial growth. A total of 20 sputum specimens with known smear findings were included, four specimens from each category (negative, scanty, 1+, 2+ and 3+) of smear grading (Revised National Tuberculosis Control Programme). Specimen digestion and decontamination was done by the NaOH (Sodium Hydroxide) -NALC (N-acetyl-L-cysteine) method, followed by centrifugation for 20 minutes at 3000 revolutions/minute. 0.5 ml of each concentrate was added into the 3 media. MGIT vials were monitored by the MGIT 960 system while the Bio FM and LJ media were examined up to 42 days and 56 days respectively for mycobacterial growth. Isolates were identified by P-nitro-a-acetylaminob-hydroxypropophenone (NAP) testing. 17 of the 20 specimens turned culture positive while three smear negative specimens remained culture negative. All isolates were identified as *M. tuberculosis* complex. Surprisingly, no discrepancy was seen in the recovery rates of the 3 media. The time to mycobacterial detection was shortest with MGIT and longest with LJ media [Table 1]. The detection time of BioFM was comparable with MGIT and outperformed the LJ media, similar to other international study findings.[4,5]

**Table 1: Detection time of Mycobacteria by MGIT, BioFM and LJ**

<table>
<thead>
<tr>
<th>Media</th>
<th>Mean time for detection (in days)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MGIT</td>
<td>10.23</td>
</tr>
<tr>
<td>BioFM</td>
<td>10.47</td>
</tr>
<tr>
<td>LJ</td>
<td>21.82</td>
</tr>
</tbody>
</table>

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compares well with MGIT in terms of recovery rate and detection time, and detects mycobacterial growth much faster than the LJ medium. The Bio FM vial is about 30% cheaper than the MGIT vial and does not need any capital investment. Thus, it can be used in settings where cost and/or infrastructural limitations prohibit usage of fully automated systems. Also, its usage will facilitate reporting of TB cultures earlier than the conventional LJ method at a more reasonable cost to the patient, thereby improving overall patient management. Nevertheless, larger studies are recommended to evaluate the sensitivity, specificity and contamination rates of the medium, before putting to diagnostic use.

Acknowledgment

We thank M/s Biorad Ltd. for providing BioFM media for the pilot study.

References


Dear Editor,

The increasing occurrence of Enterococcus species, worldwide, since late 1980s, is of particular concern due to the emergence of Vancomycin Resistant Enterococci (VRE).[1] VRE has also been reported from some parts of India.[2,3] The appearance of VRE has limited the therapeutic options available for clinicians.

A study was undertaken in this hospital to detect vancomycin resistance in enterococcal isolates using three methods and compare the three methods. The methods included – KBDDM, Vancomycin agar screen method and MIC detection by macrobroth dilution method. A total of 200 enterococcal isolates – 65 from urine, 58 from blood, 22 from Foley’s catheter tips, 21 from wound swabs, 18 from pus and 16 from fluids (ascetic fluid 12, cerebrospinal fluid 2 and peritoneal dialysis fluid 2) were included in the study. They were identified and speciated by standard biochemical tests.[4] Susceptibility to vancomycin was performed by Kirby-Bauer Disc Diffusion Method (KBDDM)[5] on Mueller Hinton Agar by using 30µg vancomycin disc (HiMedia). Vancomycin resistance was also determined by Vancomycin agar screen method using 6µg/ml of vancomycin incorporated in Brain Heart Infusion (BHI) agar. Minimum Inhibitory Concentration (MIC) of all the isolates were done by Macrobroth dilution method, using dilutions of vancomycin ranging from 2 µg/ml to 512 µg/ml. Susceptibility to teicoplanin was also done by KBDDM, in isolates showing MIC ≥ 4 µg/ml.[5]

Out of the 200 Enterococcus species, 55% (110) were Enterococcus fecium, 31% (62) were Enterococcus fecalis and 14% (28) were other Enterococcus species.

Two isolates (1%) were resistant to vancomycin by KBDDM. By Vancomycin agar screen method, three isolates showed growth, giving an overall VRE positivity of 1.5%. The vancomycin MIC for these three isolates were 8 µg/ml for one and 128 µg/ml for two, while the remaining isolates had MIC less than or equal to 4µg/ml.

The three enterococcal isolates having MIC greater than 4 µg/ml were from two patients – one each from Foley’s catheter tip and ascitic fluid of the same patient and the third from Foley’s catheter tip of another patient. The former patient was a 11-year-old female child diagnosed with Vancomycin Resistant Enterococci in a Tertiary Care Hospital in Mumbai