Novel chromogenic medium for detection of extended-spectrum beta-lactamase-producing *Enterobacteriaceae*, methicillin resistant *Staphylococcus aureus* and vancomycin resistant *Enterococcus*

Sir,

Multi drug resistant (MDR) bacteria, particularly extended-spectrum beta-lactamase producing *enterobacteria* (ESBL-E), methicillin resistant *Staphylococcus aureus* (MRSA) and vancomycin resistant *Enterococcus* (VRE) are frequently the source of potentially highly epidemic hospital-acquired infections. Several phenotypic tests have been recommended for screening and confirmation of these MDR pathogens, but these are usually performed on isolated organisms following culture and antibiotic susceptibility testing.[1] Early detection of these organisms has proven useful in the treatment of patients and prevention of nosocomial outbreaks.

In this study, the novel chromogenic media chromID ESBL, MRSA and VRE (bioMérieux SA, France) were evaluated for the presumptive identification of MDR pathogens. The reliability of chromID ESBL has been shown in other studies[2,3] also but the data related to chromID MRSA and VRE is lacking.

Automated blood culture system BACTEC 9240 is being used for blood culture in our bacteriology laboratory. Depending on Gram staining result of all the positive blood culture vials, 181, 122, and 47 specimens were inoculated on chromID ESBL, MRSA and VRE media respectively along with the routine sheep blood agar and MacConkey agar. The isolates obtained were identified by standard procedures and subjected to antimicrobial susceptibility testing using Kirby Bauer disc diffusion (KBDD) method. To avoid missing any MRSA strain, we used both cefoxitin disc (30 μg) and oxacillin screen agar plate (6 μg/ml). On chromID ESBL medium, the colonies presumed to produce ESBL had the following colors [Figure 1] obtained after 24 h of incubation at 37°C (except for one *Escherichia coli* strain that took 48 h for pink color to appear): pink or burgundy for *E. coli*, blue or green for *Klebsiella* and *Enterobacter* and brown for *Proteus* sp. All nonfermenting gram negative *bacilli* and *Salmonella typhi* showed colorless colonies. The distribution of various isolates obtained is as shown in Table 1. All isolates that grew on chrom agar were reported as resistant to cefotaxime and ceftazidime by KBDD method also except one *Klebsiella* strain which was picked by chrom agar only and was reported as sensitive by KBDD method. However on repeat testing from chrom agar, it was found to be resistant to both cefotaxime and ceftazidime. This indicates that chrom agar could detect the resistant sub-population. For ESBL confirmation, out of 96 resistant *Enterobacteria*, disc potentiation method was done in 33 strains and was found to be positive in 24 strains. Out of nine negative strains double disc synergy was done in five strains (as four strains could not be revived) and was negative in all. This can be explained on the fact that none of the methods that rely on phenotypic expression of the β-lactamase can detect every ESBL-producing isolate.[4]

On chromID MRSA medium, MRSA (16/122) showed green colonies and MRCONS (49/122) grew as colorless within 24 h of incubation.

On chromID VRE medium, VRE (12/47) showed violet colonies within 24 h of incubation.

The results obtained from all the three chromID media corresponded well with the conventional methods used in routine. Neither of the chromID media missed any resistant isolate. The chromogenic and selective characters of these media enhance recovery and identification of MDR pathogens within 18-24 h and reduce unnecessary

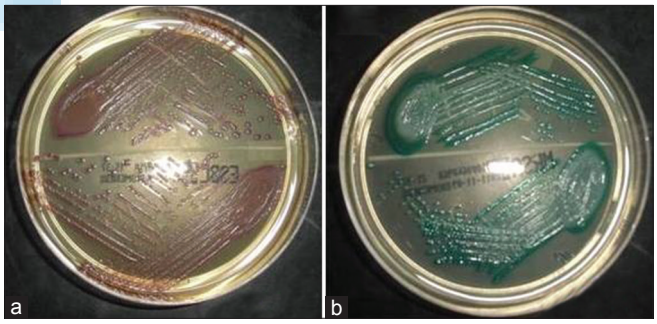

Figure 1: (a) Pink colored colonies of *Escherichia coli* on chromID extended-spectrum beta-lacatamase medium and (b) green colored colonies of methicillin resistant *Staphylococcus aureus* (MRSA) on chromID MRSA medium

Table 1: Distribution of various isolates obtained

Organism	Number
Escherichia coli	49
Klebsiella pneumoniae	45
Enterobacter sp.	10
Proteus mirabilis	1
Acinetobacter sp.	32
NFGNBs	25
Pseudomonas aeruginosa	7
Salmonella typhi	11

NFGNB - Nonfermenting gram negative *bacilli*

confirmations. The limitation of our study was that we could not perform ESBL confirmation tests on all the isolates and moreover, the genotyping characterization was not done.

To conclude these ready to use chromogenic selective media are reliable for screening and presumptive identification of ESBL-producing *Enterobacteriaceae*, MRSA and VRE directly from clinical samples.

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REFERENCES

1. Clinical and Laboratory Standards Institute. Performance Standards for Antimicrobial Susceptibility Testing; Twenteeth Informational Supplement M100-S20. Wayne: Clinical and Laboratory Standards Institute; 2010.
2. Glupczynski Y, Berhin C, Bauraing C, Bogaerts P. Evaluation of a new selective chromogenic agar medium for detection of extended-spectrum beta-lactamase-producing *Enterobacteriaceae*. J Clin Microbiol 2007;45:501-5.
3. Réglier-Poupet H, Naas T, Carrer A, Cady A, Adam JM, Fortineau N, *et al*. Performance of chromID ESBL, a chromogenic medium for detection of *Enterobacteriaceae* producing extended-spectrum beta-lactamases. J Med Microbiol 2008;57:310-5.
4. Bradford PA. Extended-spectrum beta-lactamases in the 21st century: Characterization, epidemiology, and detection of this important resistance threat. Clin Microbiol Rev 2001;14:933-51.

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