Chromogenic agar medium for rapid detection of extended-spectrum β-lactamases and *Klebsiella pneumoniae* carbapenemases producing bacteria from human immunodeficiency virus patients

Sir,

Bacterial infections in the human immunodeficiency virus (HIV) patients are mostly caused by the pathogenic bacteria with a high level of antibiotic resistance.[1] Extended-spectrum beta(β)-lactamases (ESBLs) are plasmid-encoded enzymes and confer resistance to penicillins, cephalosporins, and aztreonam.[2] *Klebsiella pneumoniae* carbapenemases (KPC) belong to the family of serine carbapenemases and are usually found in *Klebsiella pneumoniae* (*K. pneumoniae*) and *Escherichia coli* (*E.coli*). KPC hydrolyze β-lactam antibiotics and thereby, reducing their activity and misidentification of KPC-producing bacteria is common with standard susceptibility testing.[3] The aim of this study is to identify the ESBLs and KPC-producing gram-negative bacteria from HIV patients using HiCrome agar medium. HiCrome ESBL Agar Base (HiMedia, India) is a chromogenic medium used for the selective isolation of ESBL-producing bacteria on the basis of colour colonies. HiCrome ESBL supplement, containing antibiotics such as ceftazidime, cefotaxime, ceftriaxone, aztreonam, and flucanazole, is used to inhibit other contaminating microorganisms and non-ESBL-producing bacteria. HiCrome KPC Agar (HiMedia, India) is used in the identification of KPC-producing bacteria and the supplement contains antibiotics that inhibit the growth of yeast, gram-positive organisms, and gram-negative organisms that do not produce carbapenemases. The HIV patients were identified by standard combination rapid tests as per National acquired immune deficiency syndrome Control Organisation (NACO) guidelines (NACO, 2007).[4] The median cluster of differentiation 4 (CD4) cell count of the HIV patients was 142 cells/mm^3.

Among the 173 bacterial isolates from HIV patients, 126 were from urine, 27 from pus, 16 from sputum, 2 from blood, 1 from fine needle aspiration cytology, and 1 from vaginal swab. Using ESBL and HiCrome KPC agar media, a total of 108 ($P \leq 0.001$) and 132 ($P \leq 0.001$) bacterial isolates were found to be positive for ESBLs and KPC production, respectively. Out of 108 ESBLs-producing strains, 64 were *E. coli*, 11 *K. pneumoniae*, 18 *Klebsiella oxytoca* (*K. oxytoca*), 10 *Pseudomonas aeruginosa* (*P. aeruginosa*), and 5 *Proteus mirabilis* (*P. mirabilis*). Of the 132 KPC-producing isolates, 73 were *E. coli*, 17 *K. pneumoniae*, 15 *K. oxytoca*, 18 *P. aeruginosa*, 5 *P. mirabilis*, 3 *Proteus vulgaris*, and 1 *Acinetobacter baumannii*. A number of 59 isolates showed positive for both ESBLs and KPC production. ESBLs production was compared by the combination disc method (CDM) using cefotaxime and ceftazidime alone and in combination with clavulanic acid and KPC production by the modified Hodge test (MHT) using meropenem disc.[5] It was found that only 88 ($P \leq 0.001$) isolates showed positive for ESBLs production using CDM and 110 ($P \leq 0.001$) for KPC production using MHT. Comparison of results of the above methods in this study thus revealed that HiCrome agar is more sensitive than CDM in ESBLs production, as well as MHT in KPC production. Ongut *et al*. (2014)[6] from Turkey reported that among 237 bacterial isolates from various clinical samples from non-HIV patients, 143 showed positive for ESBLs production using Brilliance ESBL agar (Oxoid; Thermo Fisher Scientific, UK) and among 143 ESBL-positive isolates, 76 were *E.coli* and 67 *K. pneumoniae*. Samra *et al*. (2008)[7] reported a new CHROMagar KPC medium for rapid and direct detection of carbapenem-resistant *K. pneumoniae* from clinical samples. They found that among 122 swab cultures, 43 *K. pneumonaiae* isolates showed positive for KPC production. This is the first report of the detection of ESBLs and KPC-producing gram-negative bacteria from HIV patients using chromogenic agar medium in India. Identification of ESBLs and KPC production among gram-negative bacteria using HiCrome agar is cost-effective and in addition, has the advantage of rapid identification within 24 h with higher sensitivity. The limitations of this method include the occurrence of false-positive results in case of multiple β-lactamases producing bacteria and that a few other carbapenemases producing bacteria may show positive results as well.

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There are no conflicts of interest.

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