NARA

Nationales Referenzlaboratorium zur Früherkennung neuer Antibiotikaresistenzen und Resistenzmechanismen

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Screening for Multidrug-Resistant Bacteria (Enterobacteriaceae Producing Extended-Spectrum β-Lactamases or Carbapenemases and Vancomycin-Resistant Enterococci): Using Selective Enrichment Broths Increases Sensitivity

Dr. D. Blanc, Dr. L. Poirel, Prof. P. Nordmann

The use of chromogenic media containing defined concentrations of antibiotics improves the identification and isolation of multidrug-resistant bacteria (MDR, Enterobacteriaceae producing extended-spectrum β-lactamases [ESBL] or carbapenemases [CPE] and vancomycin-resistant enterococci [VRE]) from fecal samples.

In a previous study (using spiked stool samples), we have determined the optimal conditions for selective broth enrichment to significantly increase the detection rate of multidrug-resistant bacteria (Sadek M, Poirel L, Nordmann P. Diag Microbiol Infect Dis 96, 2020, 114919). In a second and recent study involving over 1000 patient rectal swabs, we demonstrated that a selective pre-enrichment broth culture prior to inoculation on chromogenic agar media significantly increased the detection of carbapenemase producers by nearly 60%, and that of ESBL and VRE producers by 20% (manuscript in preparation).

Based on these results, NARA recommends the use of selective enrichment broths for screening ESBL, CPE, and VRE, taking into account detection sensitivity, analysis time, and cost. This gain in sensitivity is particularly crucial during repeated screenings in epidemic settings to identify the highest number of colonized patients.

Criteria to Consider When Choosing a Screening Method for MDR Bacteria:

	Sensitivity	Turn-around time (TAT)	Cost
Rapid test (PCR)	High	3-4 h	High
Primary culture	Medium	24 h	Medium
Culture with enrichment broth	High	48 h	Medium

Preparation of Enrichment broths:

Screening for ESBL:

- Place 10 discs of 5 µg cefotaxime in 10 ml of 0.9% NaCl in a glass tube (concentration: 5 µg/ml). Shake vigourously, wait for 30 minutes, then shake again.
- Add 100 µl of the cefotaxime stock solution (0.5 µg) to each tube containing 5 ml of TSB (final concentration: 0.1 µg/ml).

Screening for CPE:

- Place 5 discs of 10 µg ertapenem in 10 ml of 0.9% NaCl in a glass tube (concentration: 5 µg/ml). Shake vigourously, wait for 30 minutes, then shake again.
- Add 100 µl of the ertapenem stock solution (0.5 µg) to each tube containing 5 ml of TSB (final concentration: 0.1 µg/ml).

Screening for VRE:

- Place one disc of 5 µg vancomycin in a tube containing 5 ml of TSB (final concentration: 1 µg/ml).

Alternatively, each antibiotic provided as powder form can be weighed to achieve the indicated final concentrations.

All solutions must be prepared freshly. Since those antibiotics are unstable in solution, those latter should not be stored for future use. After inoculation, incubate the broths for 18–24 hours at 37°C (best on a shaker) before subculturing on appropriate selective chromogenic agar plates for detection of either VRE, ESBL or CPE.

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