Evaluation of diagnostic accuracy and agreement between four phenotypic carbapenemase detection methods using clinical enterobacteriaceae isolates at a diagnostic laboratory of Pakistan

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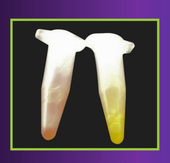
Background

- Antimicrobial resistance is a grave concern, specially in the developing world where injudicious antibiotic use is leading to rapid emergence of resistance
- Carbapenems are being used increasingly due to resistance to extended spectrum beta lactams in gram negative organisms
- Plasmid borne resistance to carbapenems is easily transmissible and a giant threat to patient management and infection control
- It is extremely important for hospitals and their laboratories to employ rapid and cost effective methods for the detection and control of spread of carbapenem resistant enterobacteriaceae (CRE)
- The Clinical Laboratory and Standards Institute (CLSI) recommends the Modified Hodge Test, the Carba NP test, the modified Inactivation Method (mCIM) and molecular methods as screening tests for the detection of CRE

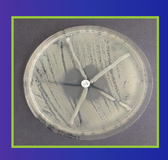
Purpose

This study aimed to compare the diagnostic performance of the Carba NP test, the modified Hodge test, the EDTA disk synergy test and the modified carbapenem inactivation method

Materials and Methods



Carba NP



Modified Hodge Test

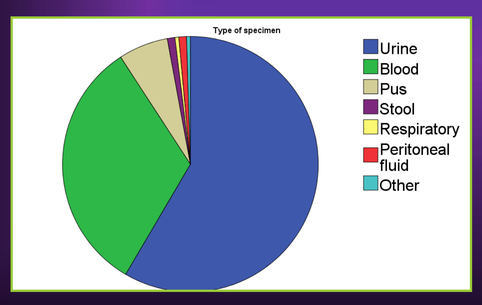
EDTA Disk Synergy Test

Modified Carbapenem Inactivation Method

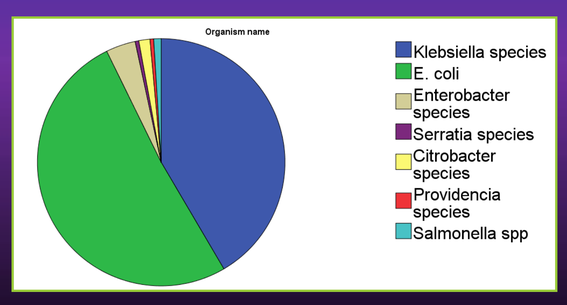
- Setting: Section of Microbiology, department of pathology, Aga Khan University Hospital, Karachi, 2014-2016
- Non probability consecutive sampling for carbapenem resistant enterobacteriaceae from clinical samples + 114 isolates from a previous study
- Identification: conventional biochemical tests
- Susceptibility testing: disk diffusion and/or minimum inhibitory concentrations when required as per CLSI recommendations
- Carba NP: Bacterial colonies + 6 mg/ml imipenem + phenol red (pH based indicator) → 2 hours : yellow color = positive
- Modified Hodge Test: A lawn of *E. coli* ATCC 25922 + 10 µg imipenem disk + test organisms streaked from the center to periphery → overnight incubation: a clover leaf shaped indentation = positive
- EDTA DST: A lawn of *E. coli* ATCC 25922 + 10 µg imipenem disk + blank filter disk with 10 µl of 0.5 mM EDTA solution → overnight incubation: synergistic zone of inhibition = positive
- mCIM: Bacterial colonies + 2 ml of Trypticase Soy Broth + 10 µg imipenem disk → 4 hours incubation -> disk on a lawn of *E. coli* ATCC 25922 → overnight incubation: zone size ≤15 mm = positive
- *bla*NDM-1 PCR was performed on 114 saved isolates
- Statistical analysis: Sensitivity, specificity, *Kappa* scores

Results

Total enterobacteriaceae: 207



Source of specimen



Categorization of isolates

Results

Diagnostic accuracy of the phenotypic tests using PCR as gold standard

Test name	Sensitivity % (CI)	Specificity % (CI)	Diagnostic accuracy %
Carba NP	94.34 (88.2-97.38)	25 (7.15-59.07)	89.4
EDTA DST	79.25 (70.57-85.888)	25 (7.15-59.07)	75.4
Modified Hodge Test	75.47 (66.49-82.68)	37.50 (13.68-69.43)	72.4
mCIM	98.11 (93.38-99.48)	00 (0.00-32.44)	91.22

Diagnostic accuracy of the phenotypic tests using meropenem MIC as gold standard

Test name	Sensitivity % (CI)	Specificity % (CI)	Diagnostic accuracy %
Carba NP	93.7 (87.9 – 97.2)	97.5 (91.2 – 99.7)	95.1 (91.3-97.6)
Modified Hodge Test	75.5 (67.1-82.7)	92.5 (84.3-97.2)	82.1 (76.2-87.09)
EDTA DST	77.9 (69.7-84.8)	98.7 (93.2-99.9)	85.9 (80.5-90.4)
mCIM	97.6 (93.2-99.5)	82.5 (72.3-90.1)	91.8 (87.1-95.1)

Agreement between the tests

Methods	Cohen's *Kappa*
Carba NP and EDTA DST	0.721
Carba NP and MHT	0.682
Carba NP and mCIM	0.677
mCIM and EDTA DST	0.529
mCIM and MHT	0.471

Discussion

- The Carba NP test is a rapid, cost effective and reliable method that can be safely used for the screening of CREs in case of outbreak or infection control in the hospital setting
- The mCIM is a very sensitive and specific test, but its main limitation is its turnaround time
- The MHT and EDTA DST are easy to perform but have long turnaround times and lower sensitivity and specificity than the Carba NP and mCIM

Conclusion

- Due to increased rates of carbapenem resistance, there is a need to employ mechanisms in hospitals that can identify such organisms as early as possible, both from a clinical and epidemiological standpoint
- Laboratories must, therefore, identify and employ methods for screening of CRE according to their feasability

References

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