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Emergence of Ertapenem Susceptible (ETP S) KPC-Producing *Enterobacteriaceae* (KPC-PE) with Conflicting Susceptibility Results by Reference and Routine Methods

O. Veliz, F. Pasteran, L. Guerriero, S. Gomez, D. Faccone, A. Corso;
INEI ANLIS, Bs. As., ARGENTINA.

Background: ETP non-susceptibility has been recommended as the most sensitivity marker (100%) for KPC-PE screening. Ten clinical isolates suspected of KPC production due to a positive Hodge test and synergy with boronic acid but ETP S were submitted to the Natl. Reference Lab. **Aim**: to characterize ETP S KPC-PE. **Methods**: Disk diffusion (DD), agar dilution (AD), Etest (ET) and broth microdilution (BMD) against carbapenems were performed in triplicates and with the same standardized bacterial inocula (CLSI). AD, ET and DD were also tested at higher inoculums. ETP true MICs were defined by population analysis profiling (PAP). PCR and sequencing were used to detect the KPC variant and the surrounding genetic structure, and PFGE to establish the genetic relatedness. **Results**: Strains were (n): *K. pneumoniae*-Kpn (6), *C.freundii*-Cfr (2), *K. oxytoca*-Kox (1) and *E. coli*-Eco (1), isolated from 9 hospitals (4 provinces) during 2011-13. *bla*KPC-2 and the surrounding genetic Variant 1a were obtained in all cases. All Kpn and Cfr strains displayed different PFGE patterns. ETP results are shown in Table. % of S to imipenem/meropenem: AD 80/100; DD 50/70; ET 100/100; BMD 10/30. **Conclusions**: By PAP, we confirmed ETP S KPC-PE as determined by AD, ET, and DD. BMD showed an alarming disagreement compared to other methods and were not due to resistant sub-populations or inoculum effect. Remarkably, ETP S KPC-PE belonged to different clones and none of the Kpn isolates were associated to ST258. The clinical impact of the overestimation of MICs by BMD should be further explored. Clinical labs should rethink ETP-based screening, as KPCs may be hidden in the wild population. Only then, the real impact of the dissemination of ETP S KPC-PE will be known

Id	Specie	Carbapenemase and other relevant *blas*	ERTAPENEM zone of inhibition or MIC value by:				
			DD	AD	ET	BMD	PAP
13233	KOX	KPC-2	19^{I}	0.015^{S}	1^{I}	8^{R}	0.03^{S}
13403	KPN	KPC-2	26^{S}	0.015^{S}	0.12^{S}	0.03^{S}	0.25^{S}
13521	ECO	KPC-2	19^{I}	0.12^{S}	0.5^{S}	2^{R}	0.25^{S}
13965	CFR	KPC-2 + PER-2	24^{S}	0.25^{S}	0.25^{S}	2^{R}	0.25^{S}
13984	KPN	KPC-2	23^{S}	0.5^{S}	0.38^{S}	4^{R}	0.25^{S}
15075	KPN	KPC-2 + CTX-M-2	22^{S}	0.25^{S}	0.38^{S}	4^{R}	0.5^{S}
15113	KPN	KPC-2	23^{S}	0.5^{S}	1^{I}	4^{R}	0.5^{S}
15222	KPN	KPC-2 + CTX-M-2	24^{S}	0.25^{S}	0.25^{S}	4^{R}	0.25^{S}
15254	CFR	KPC-2	23^{S}	0.25^{S}	0.38^{S}	2^{R}	0.25^{S}
15455	KPN	KPC-2	27^{S}	0.25^{S}	0.19^{S}	4^{R}	0.5^{S}
25922	ECO	wild type (ATCC)	34	0.12	0.12	0.12	0.06

15455	KPN	KPC-2	27^{S}	0.25^{S}	0.19^{S}	4^{R}	0.5^{S}
25922	ECO	wild type (ATCC)	34	0.12	0.12	0.12	0.06
27853	PAE	wild type (ATCC)	16	4	4	4	4

DD in mm; AD, ET, BMD, PAP in mg/L. S: susceptible; I: intermediate; R: resistant by CLSI. PAE: Pseudomonas aeruginosa.