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CHROMagar mSuperCarba screening followed by Rapidec Carba NP test for detection of carbapenemase producers in Enterobacteriaceae

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OBJECTIVE: Study of sensitivity and specificity of the novel chromogenic medium CHROMagar mSuperCARBA to detect carbapenemases including OXA-48-type producers compared to SUPERCARBA medium.

INTRODUCTION

The number of carbapenemase-producing Enterobacteriacea (CPE) is increasing worldwide. Mortality rates as high as 69% due to infections caused by these bacteria have been described. The main groups of carbapenemases that have been identified in Enterobacteriaceae are Ambler class A (KPC-type), which are able to hydrolize all β-lactams except cephamycins, the zinc-dependent Ambler class B carbapenemases (NDM, VIM and IMP), which are metallo-β-lactamases (MBL) incapable of hydrolizing aztreonam, and the Ambler class D (OXA-48-like) carbapenemase, which hydrolyzes carbapenems and weakly (or not at all) broad-spectrum cephalosporins.

Chromogenic and nonchromogenic screening methods for detecting CPE bacteria have been developed. Chromogenic media include substrate molecules for specific enzymes that result in a change of colour after substrate degradation. Among these, CHROMagar KPC is effecive in detecting VIM and KPC carbapenemases, but poorly detects OXA-48 producers; whereas ChromID ESBL was shown to detect not only ESBL producers but also class A and B carbapenemases. Brilliance CRE was reported to more efficiently detect KPC- and MBL-producing Enterobacteriaceae, but not other β-lactamases.

The chromogenic medium chromID CARBA contains carbapenems for the detection of CPE, however, this medium does not efficiently identify OXA-48 producers, and for this reason the chromID OXA-48 medium was developed. ChromID CARBA SMART is a selective chromogenic media bi-plate that selects for OXA-48 on one side and other carbapenemases, notably KPC and NDM on the other. Recently, SUPERCARBA medium containing ertapenem, cloxacillin and zinc sulfate was developed and compared to ChromID CARBA and ChromID OXA-48 in one study, and also to Brilliance CRE and CHROMagar KPC in a separate study. SUPERCARBA medium is able to detect KPC, MBL and OXA-48 producers with high sensitivity, however, this medium does not includes chromogenic molecues for indentification of enterobacterial species.

We are testing a novel chromogenic screening medium called CHROMagar mSuperCARBA that has been designed for the detection and isolation of carbapenemase-producing Enterobacteria-ceae, including strains with low-level resistance to carbapenems, by employing chromogenic molecules for that detect enterobacterial species. This medium is inhibitory for many microorganisms, mostly Gram positive and other non-CPE Gram negative bacteria.

Limits of detection of CHROMagar mSuperCARBA and SUPER CARBA media and Rapidec Carba NP test for OXA-48-type-producing enterobacterial isolates.

Strains	β-Lactamase content	MIC (mg/L)			Lowest detection limit (CFU/mL)[b]		
		IPM[a]	ETP	MEM	CHROMagar mSuperCARBA	SUPER CARBA	RAPIDEC CARBA NP
Carbapenemase OXA-48-type							
Enterobacter spp TUR9	OXA-48[c]	0.38	3	0.38	10^1	10^1	+
E. cloacae TUR10	OXA-48	0.38	4	0.38	10^1	10^1	+
K. pneumoniae T12	OXA-48	1.5	4	0.75	10^1	10^1	+
K. pneumoniae OM14	OXA-48 + TEM1	0.5	1	0.38	10^1	10^1	+
K. pneumoniae ROU	OXA-48 + CTX-M-15	0.5	1.5	0.25	10^1	10^1	+
K. pneumoniae DUW	OXA-48 + CTX-M-15 + SHV-28 + TEM-1	32	32	32	10^1	10^1	+
K. pneumoniae SIC	OXA-48 + CTX-M-15 + SHV28	0.25	1	0.25	10^1	10^1	+
K. pneumoniae AMS	OXA-48 + CTX-M-15 + TEM-1 + OXA-1	0.5	2	0.38	10^1	10^1	+
K. pneumoniae ELK	OXA-48 + CTX-M-15 + TEM-1 + SHV-11	0.5	3	0.38	10^1	10^1	+
K. pneumoniae VSG	OXA-48 + CTX-M-15 + OXA-1 + TEM-1	0.75	3	0.75	10^1	10^1	+
K. pneumoniae OM11	OXA-48 + CTX-M-14 + TEM-1	0.5	0.75	0.25	10^1	10^1	+
E. coli BOU	OXA-48 + CTX-M-15	0.5	0.75	0.12	10^1	10^1	+
E. coli BON	OXA-48 + CTX-M-24 + TEM-1	0.38	0.5	0.19	10^1	10^2	+
E. cloacae TUR	OXA-48 + SHV-5	0.5	0.5	0.5	10^1	10^1	+
K. pneumoniae Af53	OXA-181	3	>32	4	10^1	10^1	+
K. pneumoniae DEL	OXA-181 + SHV11 + CTX-M-15 + TEM-1	>32	>32	>32	10^1	10^1	+
E. coli LIEU	OXA-181 + CTX-M-15	1	1	0.25	10^2	10^2	+
K. pneumoniae 479	OXA-204 + CMY-4	8	16	8	10^1	10^1	+
E. coli DUP	OXA-204 + CMY-4 + CTX-M-15 + OXA-1	2	1	0.25	10^1	10^2	+
E. coli BAR	OXA-204 + CMY-4 + CTX-M-15	2	2	0.5	10^2	10^2	+
K. pneumoniae DEL	OXA-232 + SHV-1 + TEM-1 + CTX-M-15 + OXA-1	8	>32	32	10^2	10^2	-
K. pneumoniae RAN	OXA-232 + SHV-1 + TEM-1 + CTX-M-15 + OXA-1 + NDM-1	>32	>32	>32	10^1	10^1	+

[a] IPM = Imipenem; ETP = ertapenem; MEM = meropenem.
[b] Boldened β-lactamase names correspond to carbapenemase.
Underlined colony-forming unit counts are considered as negative results (cut-off values set at ≥1 x 10^3 CFU/plate).

METHODS: A total of 117 clinical strains of enterobacteria were used. This collection included 13 strains with reduced susceptibility to carbapenems (ESBL, overexpressed AmpC and/or porin deficiency), 18 isolates susceptible to carbapenems, 36 OXA-48-type producers, 17 KPC producers, 12 NDM producers, 13 VIM producers, and 8 IMP producers. The novel chromogenic screening medium is called CHROMagar mSuperCARBA (CHROMagar company, France) which has been designed for the detection and isolation of carbapenemase-producing Enterobacteriaceae, including those isolates with low-level of resistance to carbapenems. This medium contains chromogenic molecules that permit the identification of enterobacterial species. We compared our results with those using the SUPERCARBA medium, which is able to select KPC, MBL and OXA-48-type producers, but is not chromogenic.

RESULTS: CHROMagar mSuperCARBA is as sensitive and as specific as SUPERCARBA medium (100% and 100%, respectively) for detecting KPC, MBL and OXA-48-type producers and is compatible with posterior testing using RAPIDEC NP.

CONCLUSIONS: Our results suggest that a good workflow would be to perform initial screening using the novel chromogenic CHROMagar mSuperCARBA medium to select carbapenem-resistant isolates followed by the use of the commercial RAPIDEC NP test for detecting carbapenemase activity.