

Carbapenemase-producing Enterobacteriaceae (CPE) Detection Directly from Surveillance Rectal Swab by Immunochromatographic Test

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Background.

The prevalence of CPE is increasing worldwide. Controlling their spread is important in the hospital settings and it should relies on the using of rapid diagnostic techniques.

Objectives:

Herein we evaluated an immunochromatographic device, RESIST-3 O.O.K.®, which rapidly detected OXA-48-; OXA-163- and KPC-producing Enterobacteriaceae directly from rectal swabs using enrichment broth at different incubation periods.

Material and Methods:

Samples: All rectal swabs for CPE detection were included in this study between Aug and Nov/2017.

Culture based method (CBM): The samples were inserted and homogenized into a 3 mL BHI tube and subcultured onto MacConkey (MAC) agar plate with carbapenem discs. Ten microliters were delivered onto a Chromagar KPC® plate for bacterial colony count (BCC). All plates were incubated at 35±2° C overnight. The BHI tube was incubated with a MER disc (10 µg). After a 4-hour incubation period, all the BHI tubes were re-submitted to subculture onto MAC and Chromagar KPC.

Immunochromatographic procedure: In parallel, the bacterial pellet recovered from 400 µL of BHI suspension was submitted to RESIST-3 O.O.K. at time zero (T0). The BHI tube was incubated with a MER disk (10 µg) and the RESIST-3 O.O.K. test was repeated at T1h; T2h; T4h; T6h and T8h.

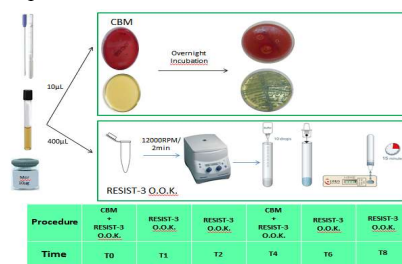


Figure 1. CBM and immunochromatographic procedures used in this study.

Susceptibility testing: Carbapenem susceptibility testing was performed with all bacterial colonies recovered from MAC.

Detection of carbapenemase encoding-genes:

The K-Set results were compared to CBM and the discordant results were submitted to detection of *bla*_{KPC}, *bla*_{NDM} and *bla*_{OXA-48}. Statistical analysis were performed according to the incubation period and BCC.

Results:

From 503 samples, 148 (29%) were positive for CPE by CBM. The bacterial species isolated were *K. pneumoniae* (97%), *Enterobacter* spp (1%), *Serratia* spp (1%) and *E.coli* (1%). RESIST-3 O.O.K. sensitivity (SE) and specificity (SP) were 91% and 100%, respectively. SE was proportional to the incubation period (T0, 25%, T1, 37%, T2, 52%, T4, 69%, T6, 79% and T8, 91%). In 33 samples the results were discrepant. Among them, *bla*_{KPC} (n = 10) and *bla*_{NDM} (n = 1) were detected in 11 samples. None of the carbapenemase-encoding genes included in this study were detected in the remaining 22 samples.

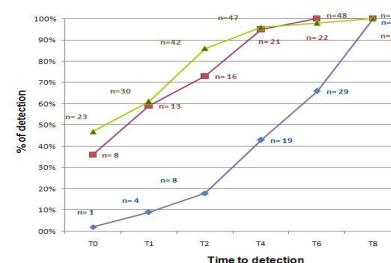


Figure 2. Detection of CPE by RESIST-3 O.O.K. according to the incubation time and bacterial inoculum in the rectal swab.

Table 1. False-negative results observed on RESIST-3 O.O.K. T test

N°	CPE - ID	BCC T0	BCC T4	OOK-KSeT result	Molecular detection
48	<i>K. pneumoniae</i>	$\leq 10^3$	$\geq 10^5$	NEG	<i>bla</i> _{KPC}
139	<i>K. pneumoniae</i>	No grow	$\leq 10^3$	NEG	<i>bla</i> _{KPC}
155	<i>K. pneumoniae</i>	10^4	10^4	NEG	<i>bla</i> _{KPC}
212	<i>K. pneumoniae</i>	$\leq 10^3$	$\leq 10^3$	NEG	<i>bla</i> _{KPC}
214	<i>K. pneumoniae</i>	No grow	$\leq 10^3$	NEG	<i>bla</i> _{KPC}
269	<i>K. pneumoniae</i>	$\geq 10^5$	$\geq 10^5$	NEG	<i>bla</i> _{KPC}
400	<i>K. pneumoniae</i>	$\leq 10^3$	$\leq 10^3$	NEG	<i>bla</i> _{KPC}
448	<i>K. pneumoniae</i>	No grow	$\leq 10^3$	NEG	<i>bla</i> _{KPC}
464	<i>E. asburiae</i>	$\leq 10^3$	10^4	NEG	<i>bla</i> _{KPC}
509	<i>K. pneumoniae</i>	$\leq 10^3$	$\leq 10^3$	NEG	<i>bla</i> _{KPC}
49	<i>K. pneumoniae</i>	$\leq 10^3$	$\leq 10^3$	NEG	<i>bla</i> _{NDM}

Conclusion:

The K-Set OOK proved to be a sensitive test for detecting KPC-producing isolates directly from rectal swabs at the same day. The test is designed to detect only OXA-48-like, OXA-163-like and KPC-like carbapenemases. False-negative results may occur due to the presence of other carbapenemases or low bacterial inoculum. Therefore, it is recommended to be performed in parallel to the culture.

References:

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CDC guideline. Laboratory Protocol for Detection of Carbapenem-Resistant or Carbapenemase-Producing, *Klebsiella* spp. and *E. coli* from Rectal Swabs. https://www.cdc.gov/hai/pdfs/labsettings/klebsiella_or_ecoli.pdf