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Bacteriology

Evaluation of the automated BD Phoenix CPO Detect test for detection and classification of carbapenemases in Gram negatives

Imane Saad Albichr [a,1], Ahalieyah Anantharajah [a,1], Magali Dodémont [b], Marie Hallin [b], Alexia Verroken [a], Hector Rodriguez-Villalobos [a,*]

[a] Department of Microbiology, Cliniques universitaires Saint-Luc, Université Catholique de Louvain, Brussels, Belgium
[b] Department of Microbiology, Laboratoire Hospitalier Universitaire de Bruxelles (LHUB-ULB), Hôpital Erasme-Cliniques universitaires de Bruxelles, Université Libre de Bruxelles, Brussels, Belgium

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ABSTRACT

We evaluated the performance of the automated BD Phoenix CPO Detect test (BD-CPO test) for detection and Ambler classification of carbapenemases in *Enterobacteriaceae*, *P.aeruginosa* and *A.baumannii* complex. A collection of 287 Gram-negative clinical isolates, with a reduced susceptibility to at least one carbapenem including 184 carbapenemase-producing organisms (CPO) and 103 non-CPO, was tested.
The BD-CPO test showed an overall sensitivity of 89.7% and specificity of 83.5% for carbapenemase detection. 1/7 of class A, 82.9% of class B, and 89.8% of class D carbapenemases were correctly classified. Poor detection sensitivity of 68.9% and specificity of 62.1% in *P.aeruginosa* was observed. However, combination with ceftazidime/avibactam susceptibility, provided by this panel, increased the performances for *P.aeruginosa*. The integration of an automated carbapenemase detection and classification in routine susceptibility panels would save time and help for therapeutic management. Further developments are needed to improve the accuracy of the BD-CPO test.

1. Introduction

Rapid and accurate detection methods of carbapenemase-producing organisms (CPO) are needed for collective control infection and for optimal therapeutic management. However, the CPO detection has become a critical challenge in clinical microbiology. Most carbapenemase hydrolyze carbapenems at variable levels and some of them show low-level resistance or even susceptibility to carbapenems. Moreover, decreased susceptibility to carbapenems may also be caused by reduced permeability due to porin down-regulation or over-expression of efflux pumps.

Carbapenemases are classified into 3 groups based on their active sites according to Ambler Classification: Class A carbapenemases (mostly KPC enzymes), Class B or metallo-β-lactamases (MBL), mostly VIM, NDM, and to a lesser extent IMP, and Class D (mostly OXA-48-like in *Enterobacteriaceae* and OXA-23, OXA-24/−40, and OXA-58-like in *Acinetobacter* spp).

In most routine microbiology laboratories, the first cause for CPO suspicion is elevated carbapenem minimum inhibitory concentration (MIC) values followed by phenotypic or molecular methods for confirmation of carbapenemase production. Molecular assays remain the reference standard for the identification of known carbapenemase-encoding genes but their high cost make difficult their usage in routine laboratories. During the recent years, several rapid phenotypic methods, low-cost alternative to genotypic methods, have been developed such as hydrolysis methods (e.g. Carba NP and matrix-assisted laser-desorption/ionization time-of-flight mass spectrometry [MALDI TOF MS] methods). Although these tests provide an answer within hours with excellent performances, they cannot discriminate between carbapenemase types (Dortet et al., 2018; Noel et al., 2017). The lateral flow immunochromatographic assays have proven to be a reliable alternative but are limited to the detection of the most prevalent carbapenemases (KPC, NDM, VIM and OXA-48-like) (Glupczynski et al., 2019). The characterization of the carbapenemase class is of growing interest not only for epidemiologic purposes but also for therapeutic decisions. Indeed, some new antibiotics such as ceftazidime-avibactam (CAZ/AVI) are active on class A and class D carbapenemases but have no efficacy on MBL (Liscio et al., 2015).

To reduce detection time and to deal with the lack of discrimination between the 3 different classes of carbapenemases by previous phenotypic detection methods, Becton Dickinson recently developed the BD Phoenix® CPO Detect test (Becton Dickinson, Franklin Lakes, NJ, USA) that allows (1) antimicrobial susceptibility testing (AST), (2) carbapenemase detection and carbapenemase characterization (according Ambler classification) at the same time. In this study, we evaluated the performances of the CPO Detect test on the BD Phoenix®

* Corresponding author.
E-mail address: hector.rodriguez@uclouvain.be (H. Rodriguez-Villalobos).
[1] Both authors contributed equally to this work.

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Automated Microbiology System using phenotypically and genotypically characterized clinical isolates. We investigated the performance of the test not only on *Enterobacteriaceae* isolates including a large proportion of OXA-48 like producers that is in line with our local current epidemiology (David et al., 2019; Poirel et al., 2012) but also on non-fermentative Gram-negative isolates.

2. Material and methods

2.1. Reference strains and clinical isolates

Fourteen Gram-negative reference strains from the Belgian National Antimicrobial Comity (NAC) and 287 Gram-negative clinical isolates, including 201 *Enterobacteriaceae*, 74 *P. aeruginosa* and 12 *A. baumannii complex*, with a reduced susceptibility to at least one carbapenem (imipenem, meropenem, or ertapenem) were included (Table S1). Antimicrobial Susceptibility testing (AST) was primarily performed by BD Phoenix® NMIC-408 or UNMIC-409 panels and/or by disk diffusion method and AST results were confirmed by the BD Phoenix® NMIC-502 susceptibility testing. These strains were collected from various clinical samples (blood culture, urine, sputum, rectal swab) in 2 centers in Brussels (Cliniques universitaires Saint-Luc and Erasme Hospital) between January 2015 and March 2018 and were characterized for carbapenemase genes by an in-house multiplex PCR (Bogaerts et al., 2013). Duplicate isolates from the same patient and duplicate outbreak strains were excluded. Carbapenemase producers (n = 184) included 7 KPC producers, 85 class B producers (53 VIM, 31 NDM, and 1 IMP) and 92 class D producers (80 OXA-48-like, 8 OXA-23, 2 OXA-24/−40, and 2 OXA-58). bla_{OXA-48}-like genes were sequenced for 40 OXA-48-producing *Enterobacteriaceae* using the following primers: OXA-48F, 5′-ATGCGTGTATTAGCCTTATCG-3′ and OXA-48R, 5′-GAGCACTTCTTTTGTGATGGC-3′ (Anantharajah et al., 2019). Thirty-seven OXA-48 and 3 OXA-181 were characterized. Non-CPO isolates (n = 103) expressed resistance mechanisms other than carbapenemases, mainly including extended-spectrum-β-lactamases (ESBL), AmpC-type cephalosporinases associated or not with uncharacterized carbapenem resistance mechanisms (porin loss, efflux pumps overexpression). Combination disk testing (ESBL + AmpC Screen Kit, Rosco diagnostic, Taastrup, Denmark) was used to detect ESBL and AmpC production in *Enterobacteriaceae*. Isolates with inhibition zones suggestive of ESBL production were further characterized by multiplex PCR targeting bla_{CTX-M}, bla_{SHV}, bla_{TEM}, and bla_{OXA-1} (Bogaerts et al., 2013).

2.2. BD Phoenix® CPO Detect test

Bacteria suspension of all strains were prepared from overnight colonies grown on a Columbia plate (BD Diagnostic Systems, Sparks, MD) in a Phoenix® ID broth according to the manufacturer's recommendations. The BD Phoenix® NMIC-502 is a Phoenix antimicrobial susceptibility testing panel for Gram-negative of 24 antibiotics including ceftazidime-avibactam, ertapenem, imipenem, meropenem, fosfomycin, nitrofurantoin, temocillin, and tigecycline. The panel includes also a CPO Detect test: a growth-based method for the detection and Ambler classification of carbapenemases: Nine wells are dedicated to CPO detection/ characterization, each containing a β-lactam antibiotic, alone and in combination with various chelators and β-lactamase inhibitors. The MIC values, the carbapenemase detection and the Ambler classification were analyzed by the BD expert system s/w 6.35. The susceptibility breakpoints were interpreted according to the European Committee on Antimicrobial Susceptibility Testing (EUCAST) recommendations. Three specific alarms were indicated by the expert system: "resistant to one or more carbapenems", "carbapenemase producer" and "Class A, B or D carbapenemase producer". The isolates identified as carbapenemase producers by the system were assigned to an Ambler class or remained unclassified. When discordant results were obtained, both methods (BD Phoenix CPO test and molecular resistance gene identification) were repeated.

3. Results

3.1. Carbapenemase detection and classification

The 5 NAC strains producing a carbapenemase (1 KPC, 2 VIM, and 2 OXA-48-like producers) were correctly identified as carbapenemase producers and appropriately classified by the BD Phoenix CPO test.

Table 1
Performances of the BD-CPO test for carbapenemase detection and Ambler classification.

CPO type	Species	No of isolates tested	Carbapenemase detection				Carbapenemase Ambler classification				
			No of positive	No of negative	Sensitivity% (95% CI*)	Specificity% (95% CI*)	A	B	D	Unclassified positive	Sensitivity% (95% CI*)
All CPO		184	165	19	89.7 (84.4–93.3)	-	1	58	79	27	83.6 (77.2–88.5)
Class A	*Enterobacteriaceae*	7	7	-	100.0 (64.6–100.0)	-	1	-	-	6	14.3 (2.6–51.3)
Class B	All Class B	85	70	15	82.4 (72.9–89.0)	-	-	58	-	12	82.9 (72.4–89.9)
	Enterobacteriaceae	40	39	1	97.5 (87.1–99.6)	-	-	32	-	7	82.1 (67.3–91.0)
	P. aeruginosa	45	31	14	68.9 (54.3–80.5)	-	-	26	-	5	83.9 (67.4–92.9)
Class D	All Class D	92	88	4	95.7 (89.3–98.3)	-	-	-	79	9	89.8 (81.7–94.5)
	Enterobacteriaceae	80	79	1	98.8 (93.2–99.8)	-	-	-	71	8	89.9 (81.3–94.8)
	A. baumannii complex	12	9	3	75.0 (46.8–91.1)	-	-	-	8	1	88.9 (56.5–98.0)
negative	All non-CPO‡	103	17	86	-	83.5 (75.2–89.4)	7	-	7	3	-
	Enterobacteriaceae	74	6	68	-	91.9 (83.4–96.2)	-	-	3	3	-
	P. aeruginosa	29	11	18	-	62.1 (42.4–78.7)	7	-	4	-	-

‡ CPO = carbapenemase producing-organisms.
* 95% CI = 95% confidence interval.

The overall agreement with PCR results for carbapenemase detection (including non-CPO isolates) was 87.5% (251/287). Among the carbapenemase producing clinical isolates, the BD-CPO test showed a detection sensitivity of 89.7% (165/184), with a specificity of 83.5% (86/103) (Table 1). The panel correctly identified as carbapenemase producers 1/7, 82.4% and 95.7% of class A, B, and D CPO respectively. The BD-CPO test failed to detect 19 CPO: 2 *Enterobacteriaceae* (one OXA-48-like and one VIM producers), 14 *P. aeruginosa* (all VIM producers), and 3 *A. baumannii complex* (one OXA-23 and 2 OXA-58 producers). Of the 165 isolates identified as "carbapenemase producer" by the BD-CPO test, 138 strains were correctly classified, and 27 strains were unclassified, leading to an overall sensitivity for carbapenemase classification of 83.6% (Table 1). No CPO strain was misclassified in this study. The performances of the BD-CPO test for carbapenemase classification differed depending on the Ambler class. Indeed, the test correctly classified to Ambler level only one of the 7 KPC-producing *Klebsiella* spp. isolates. For class B and class D CPO, the BD-CPO test correctly classified 58/70 (82.9%) and 79/88 (89.8%) of the isolates, respectively, yielding a positive carbapenemase test. Of note, the 3 OXA-181 producers, included in our collection, were correctly detected and classified. In 6 *Enterobacteriaceae* isolates and 11 non carbapenemase-producing *P. aeruginosa* isolates, decrease of carbapenem susceptibility was incorrectly associated with carbapenemase production (14 class A or D enzymes and 3 unclassified) by the BD-CPO test. The 6 *Enterobacteriaceae* isolates (4 *E.coli* and 2 *K. pneumoniae*) incorrectly identified as carbapenemase producers displayed a pattern consistent with ESBL production and expressed $bla_{CTX\text{-}M1}$ or $bla_{CTX\text{-}M9}$ gene associated or not with bla_{SHV}, bla_{TEM} and $bla_{OXA\text{-}1}$.

BD-CPO test yielded a poor carbapenemase detection sensitivity of 68.9% (31/45) in *P. aeruginosa*. The test detected 30/44 and correctly classified 25/30 of VIM isolates. The IMP-producing *P. aeruginosa* isolate was accurately detected and classified. Among the 29 non-carbapenemase-producing *P. aeruginosa*, the test showed a specificity of 62.1% with falsely positive results for 11 isolates associated to imipenem and meropenem MICs >8 mg/L. These isolates were misclassified as class A (n = 7) and class D (n = 4) CPO.

3.2. Ceftazidime/avibactam susceptibility

This new BD panel provides for the first time an automated evaluation of ceftazidime/avibactam (CAZ/AVI) susceptibility which could help the clinician to deliver effective therapy against Gram-negative isolates resistant to carbapenems. As expected, all MBL-producing organisms were resistant to CAZ/AVI. In addition, 15 OXA-48 producers, 2 KPC-producers and 11 non-CPO were resistant to this β-lactamase inhibitor combination. Interestingly, based on these results, the accuracy of carbapenemase detection in *P. aeruginosa* could be improved by taking into consideration the CAZ/AVI resistance. Indeed, all the 45 carbapenemase-producing *P. aeruginosa* isolates and only 5/28 of non-CPO *P. aeruginosa* isolates were resistant to CAZ/AVI. Therefore, the sensitivity and the specificity of CAZ/AVI resistance to predict the production of class B carbapenemase in *P. aeruginosa* were respectively 100% and 82.1%, respectively.

4. Discussion

Rapid and accurate detection of CPO is essential for both the optimization of antimicrobial therapy and the prompt implementation of infection control measures to prevent outbreaks in clinical settings (Bonomo et al., 2018). The selection of a carbapenemase detection test depends on several factors, including type of organisms (i.e., *Enterobacteriaceae* and/or glucose-nonfermenting Gram negatives), labor intensity, cost, accuracy and turnaround time of the test (Tamma and Simner, 2018).

BD-CPO test combines antimicrobial susceptibility testing and an inhibitor-based method for the detection and Ambler classification of carbapenemases in *Enterobacteriaceae*, *P. aeruginosa* and *A. baumannii* complex. Moreover, this panel provides also wells testing the susceptibility to ceftazidime-avibactam, active against isolates producing class A (KPC) and some class D β-lactamases (e.g. OXA-48). (Falcone and Paterson, 2016) However, the emergence of resistance to CAZ/AVI leading to therapeutic failure has been recently reported (Humphries et al., 2015; Shields et al., 2017). In this study, 17 isolates were resistant to CAZ/AVI among class A and class D CPO.

BD-CPO test presents the advantage, over the traditional inhibitor-based methods, of an easier computer-assisted algorithm-based detection (EUCAST, n.d.). The test is not only less expensive than molecular methods but also easy to perform with low hands-on-time he test is affordable and easy to perform with low hands-on time (~1–2 min) and the results are available within 18 h from cultured bacterial growth (Tamma and Simner, 2018).

Accuracy of CPO detection and classification was heterogeneous depending on the type of species and carbapenemase. For *Enterobacteriaceae* the BD-CPO test displayed excellent performances for NDM, VIM, and OXA-48-like carbapenemase detection. OXA-48-like enzymes, known to possess a weak carbapenemase activity, are important to detect taking into account the large dissemination of OXA-48-like producers in several European, Middle East and North African countries (Albiger et al., 2015; David et al., 2019; Poirel et al., 2012).

One major finding of our study was the inability of the BD-CPO test to classify KPC producers (1/7) in contrast to previous studies (Ong et al., 2018; Thomson et al., 2017). Indeed, Thomson et al. classified 91 of the 107 class A CPO detected by the test (85%) and Ong et al. correctly classified 6 of the 7 KPC CPO (85.7%) detected. Recently Simon et al. also reported a low sensitivity of Ambler class A classification by the BD-CPO test. (Simon et al., 2019) KPC is the most clinically significant enzymes among the carbapenemase-producing *Klebsiella* spp. in Europe, especially in Italy, Greece and Portugal (David et al., 2019). Although KPC-2 is very frequently encountered worldwide, at present more than 20 KPC variants have been identified and their epidemiology varies geographically with endemic or sporadic spread (Lee et al., 2016; Nordmann and Poirel, 2014). In this sense, the differences between the performance observed in current study and the other studies could be due to the presence of different KPC variant or different *K. pneumoniae* clones harboring *kpc* gene (e.g. ST258, ST11) depending on the geographical area. Furthers studies are needed to investigate this hypothesis. Although the susceptibility to ceftazidime-avibactam is provided by the BD-CPO AST panel, the inability of the BD-CPO test to classify KPC producers could impact the therapeutic decisions for infections caused by KPC producing organisms and prevent, for example, the use of new combinations β-lactam-β-lactamase inhibitors such as meropenem-vaborbactam and imipenem-relebactam. Our overall analytical performances for carbapenemase detection in *Enterobacteriaceae* were higher than those reported by Ong et al. (sensitivity 89.4% and specificity 66.7%). However, the carbapenemase type diversity of the 2 collections was different, reflecting the local current carbapenemase epidemiology. They demonstrated that the BD-CPO test failed to detect IMI-1, a prevalent class A carbapenemase in Singapore, not present in our isolates collection.

For *A. baumannii* complex the performances of the BD-CPO test are promising for the detection and classification of carbapenemases. However, a larger number of isolates is required to confirm these observations. The BD-CPO test results stand out in comparison with other phenotypic approaches reporting low sensitivities for the detection of carbapenemase-producing *Acinetobacter* spp., known for being more difficult to detect, probably due to the low carbapenem hydrolytic activity of the class D carbapenemases found in these organisms (Simner et al., 2017).

For *P. aeruginosa* isolates the BD-CPO test showed poor sensitivity and specificity in our study. In Belgium, the resistance to carbapenems in this species is predominantly mediated by non-carbapenemase

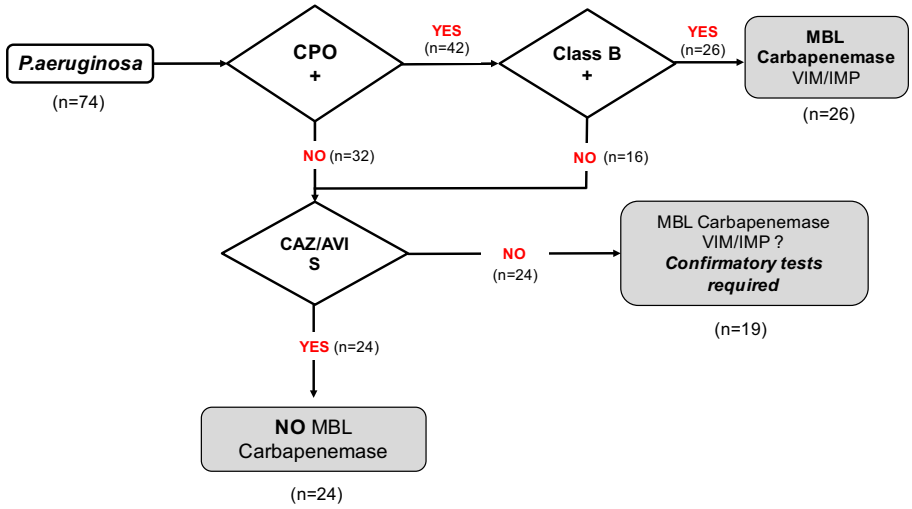

Fig. 1. Algorithm proposed for the detection of Class B carbapenemases for *Pseudomonas aeruginosa* with BD-CPO test. If *P. aeruginosa* is classified as class B carbapenemase producer, the test can be validated. If it is not detected as a CPO or not classified as class B carbapenemase producer, the ceftazidime/avibactam (CAZ/AVI) sensitivity should be verified. If the strain is susceptible to CAZ/AVI, a metallo-beta-lactamase (MBL) can be excluded. If the isolate is resistant to CAZ/AVI, a MBL cannot be ruled out and a confirmation is needed.

mechanisms (loss of OprD porin expression and/or upregulation MexAB-OprM efflux pumps) and less frequently by VIM-2 production (Heinichs et al., 2015). The specificity of the BD-CPO test might be impaired by the unspecific effects of inhibitors used in the test or by chelation that disrupts the permeability of the outer bacterial membrane. In addition, low carbapenem hydrolytic activity, unexpressed or minimally expressed carbapenemase genes and slow bacterial growth could explain the detection failure by the BD-CPO test yet not investigated in this study. Interestingly, include CAZ/AVI susceptibility in the algorithm of Class B carbapenemase detection for *P. aeruginosa* considerably increases sensitivity (100%) and specificity (82.1%). We observed that all *P. aeruginosa* isolates producing class B carbapenemase were resistant to CAZ/AVI, while only 5/28 were non-CPO strains. Based on these findings, isolates reported as resistant to CAZ/AVI without any carbapenemase detection by the BD-CPO test must be verified by another method for the presence of carbapenemase activity (Fig. 1).

Co-production of KPC or OXA-48-like and a MBL by *K. pneumoniae* has been recently described in various regions of the world (Balm et al., 2013). In this study, we did not evaluate the accuracy of the test for the simultaneously detection of different carbapenemases. However, according to Thomson et al. and Ong et al., the test failed to detect dual carbapenemase carriage (Ong et al., 2018; Thomson et al., 2017). For these types of isolates, molecular assays or immunochromatographic tests will remain more performant.

Overall, given the low specificity of this test (83.5%), laboratories should perform a confirmatory test for isolates identified as carbapenemase producer by the BD-CPO test to avoid unnecessary control measures and isolation precautions. Recently, the study of Simon et al. (2019) revealed that the use of hydrolysis methods such as β-CARBA assay as a confirmatory test could increase the specificity of the BD-CPO test from 55.3% to 100%. Nevertheless, all the colorimetric assays are based on a subjective visual observation of color change, which can result sometimes in erroneous interpretations.

We acknowledge that this study suffers from several limitations. First, we could not exclude carbapenemase expression among our non-CPO isolates. Indeed, these isolates might harbor less common carbapenemase genes not targeted by our in-house multiplex PCR. Second, the isolate collection tested was a reflection of our local CPO epidemiology and was limited in the diversity of carbapenemase. Therefore, additional testing of isolates with GES, IMI, NMC, SPM, GIM, and DIM carbapenemase is warranted to challenge the performances of the BD-CPO test.

5. Conclusion

The BD-CPO test is an automated phenotypic inhibitor-based test, included in the routine susceptibility testing, allowing detection and classification of carbapenemase producing organisms in one step. This test appears to be a highly sensitive screening test to exclude carbapenemases in *Enterobacteriaceae,* as reported by previous studies (Ong et al., 2018; Simon et al., 2019; Thomson et al., 2017). Of note, its ability to classify carbapenemases can help to optimize therapeutic choices. However, modifications to this test may be necessary to accurately classify KPC-producing *Klebsiella* spp. The poor accuracy for MBL-producing *P. aeruginosa* detection could be overcome by the use of an algorithm including susceptibility to CAZ/AVI which can help to establish the need of confirmatory testing.

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Declaration of competing interest

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References

Albiger B, Glasner C, Struelens MJ, et al. Carbapenemase-producing Enterobacteriaceae in Europe: assessment by national experts from 38 countries, May 2015. Euro Surveill 2015;20.

Anantharajah A, Tossens B, Olive N, et al. Performance evaluation of the MBT STAR((R))-Carba IVD assay for the detection of carbapenemases with MALDI-TOF MS. Front Microbiol 2019;10:1413.

Balm MN, La MV, Krishnan P, et al. Emergence of *Klebsiella pneumoniae* co-producing NDM-type and OXA-181 carbapenemases. Clin Microbiol Infect 2013;19:E421–3.

Bogaerts P, Rezende de Castro R, de Mendonca R, et al. Validation of carbapenemase and extended-spectrum beta-lactamase multiplex endpoint PCR assays according to ISO 15189. J Antimicrob Chemother 2013;68:1576–82.

Bonomo RA, Burd EM, Conly J, et al. Carbapenemase-producing organisms: a global scourge. Clin Infect Dis 2018;66:1290–7.

David S, Reuter S, Harris SR, et al. Epidemic of carbapenem-resistant Klebsiella pneumoniae in Europe is driven by nosocomial spread. Nat Microbiol 2019;4(11):1919–29.

Dortet L, Tande D, de Briel D, et al. MALDI-TOF for the rapid detection of carbapenemase-producing Enterobacteriaceae: comparison of the commercialized MBT STAR(R)-Carba IVD Kit with two in-house MALDI-TOF techniques and the RAPIDEC (R) CARBA NP. J Antimicrob Chemother 2018;73:2352–9.

EUCAST. Guidelines for detection of resistance mechanisms and specific resistances of clinical and/or epidemiological importance. http://www.eucast.org/fileadmin/src/media/PDFs/EUCAST_files/Resistance_mechanisms/EUCAST_detection_of_resistance_mechanisms_170711.pdf.

Falcone M, Paterson D. Spotlight on ceftazidime/avibactam: a new option for MDR Gram-negative infections. J Antimicrob Chemother 2016;71:2713–22.

Glupczynski Y, Evrard S, Huang TD, Bogaerts P. Evaluation of the RESIST-4 K-SeT assay, a multiplex immunochromatographic assay for the rapid detection of OXA-48-like, KPC, VIM and NDM carbapenemases. J Antimicrob Chemother 2019;74(5):1284–7.

Heinichs A, Huang TD, Berhin C, et al. Evaluation of several phenotypic methods for the detection of carbapenemase-producing *Pseudomonas aeruginosa*. Eur J Clin Microbiol Infect Dis 2015;34:1467–74.

Humphries RM, Yang S, Hemarajata P, et al. First report of ceftazidime-avibactam resistance in a KPC-3-expressing *Klebsiella pneumoniae* isolate. Antimicrob Agents Chemother 2015;59:6605–7.

Lee CR, Lee JH, Park KS, et al. Global dissemination of carbapenemase-producing *Klebsiella pneumoniae*: epidemiology, genetic context, treatment options, and detection methods. Front Microbiol 2016;7:895.

Liscio JL, Mahoney MV, Hirsch EB. Ceftolozane/tazobactam and ceftazidime/avibactam: two novel beta-lactam/beta-lactamase inhibitor combination agents for the treatment of resistant Gram-negative bacterial infections. Int J Antimicrob Agents 2015;46:266–71.

Noel A, Huang TD, Berhin C, et al. Comparative evaluation of four phenotypic tests for detection of carbapenemase-producing Gram-negative bacteria. J Clin Microbiol 2017;55:510–8.

Nordmann P, Poirel L. The difficult-to-control spread of carbapenemase producers among Enterobacteriaceae worldwide. Clin Microbiol Infect 2014;20:821–30.

Ong CH, Ratnayake L, Ang MLT, et al. Diagnostic accuracy of BD Phoenix CPO Detect for carbapenemase production in 190 Enterobacteriaceae isolates. J Clin Microbiol 2018;56.

Poirel L, Potron A, Nordmann P. OXA-48-like carbapenemases: the phantom menace. J Antimicrob Chemother 2012;67:1597–606.

Shields RK, Chen L, Cheng S, et al. Emergence of ceftazidime-avibactam resistance due to plasmid-borne blaKPC-3 mutations during treatment of carbapenem-resistant *Klebsiella pneumoniae* infections. Antimicrob Agents Chemother 2017;61.

Simner PJ, Opene BNA, Chambers KK, et al. Carbapenemase detection among carbapenem-resistant glucose-nonfermenting Gram-negative bacilli. J Clin Microbiol 2017;55:2858–64.

Simon M, Gatermann S, Pfeifer Y, et al. Evaluation of the automated BD Phoenix CPO Detect panel in combination with the beta-CARBA assay for detection and classification of carbapenemase-producing Enterobacterales. J Microbiol Methods 2019;156:29–33.

Tamma PD, Simner PJ. Phenotypic detection of carbapenemase-producing organisms from clinical isolates. J Clin Microbiol 2018;56.

Thomson G, Turner D, Brasso W, et al. High-stringency evaluation of the automated BD Phoenix CPO Detect and Rapidec Carba NP tests for detection and classification of carbapenemases. J Clin Microbiol 2017;55:3437–43.