



The Brazilian Journal of INFECTIOUS DISEASES

www.elsevier.com/locate/bjid



Letter to the Editor

False-positive results in screening for metallo- β -lactamase are observed in isolates of *Acinetobacter baumannii* due to production of oxacillinases

Dear Editor,

Carbapenemases production, either metallo- β -lactamases (MBLs), KPC or oxacillinases (OXA), is the main resistance mechanism responsible for resistance phenotype to carbapenems in *Acinetobacter*.¹ While for MBLs and KPCs there are some screening tests for their detection,^{2,3} the same is not true for oxacillinases.

During the investigation of a large outbreak, we analyzed 584 carbapenem-resistant *Acinetobacter* spp. isolates from seven hospitals. Isolates were identified using the API 20NE system (Biomérieux, Basingstoke, United Kingdom). PCR for the *bla*_{OXA-51} gene was performed as a marker of *Acinetobacter baumannii* at species level.¹ The MIC for imipenem was performed by Etest® (AB BIODISK, Solna, Sweden) and was ≥ 8 μ g/mL in all isolates.

A total of 562 (96.3%) and 553 (95%) isolates proved to be OXA-51 and OXA-23 producers respectively, by a multiplex PCR, which included primers for the *bla*_{OXA-23-like}, *bla*_{OXA-24-like}, *bla*_{OXA-51}, *bla*_{OXA-58}, *bla*_{OXA-143} genes.⁴ Among the 553 OXA-23-like *A. baumannii* producing isolates, we observed 86 (15.5%) positive isolates in the screening test for MBLs either by the disk-approximation test using a ceftazidime (CAZ) and a 2-mercaptopyruvic acid (MPA) or by the Etest MBL (Imipenem/Imipenem + EDTA – AB BIODISK, Solna, Sweden).^{2,5} The modified Hodge Test was performed in these isolates and only nine (1.5%) were positive.⁶ PCR using *bla*_{IMP-1-like}, *bla*_{VIM-2-like}, *bla*_{SPM-1}, *bla*_{NDM-1}, *bla*_{KPC-1,2} primers^{2-4,7} failed to produce any amplicon in these isolates.

Oxacillinases enzymes are formed by dimers.⁸ As divalent metals are needed to make the dimeric structure more stable, a chelator agent would affect the activity of such enzymes.⁸ Therefore, chelators, such as EDTA or MPA, may inhibit these OXA enzymes producing a result that could be interpreted as positive for the presence of MBL according to the phenotypic methods.⁸ This hypothesis can be supported by the fact that 98.5% of the isolates were Hodge Test negative. The opposite is observed when there is only oxacillinases production, since these enzymes hydrolyze carbapenems poorly; it results in a Hodge Test negative in most isolates.

The OXA-23 gene is the most prevalent oxacillinase among CRAB in our city. Although positive results have been obtained by phenotypic screening tests for MBL and the PCR was negative for genes tested. Thus, one must be careful when interpreting positive results in phenotypic screening tests for MBL in CRAB because false-positive results can occur.

Conflict of interest

The authors declare no conflict of interest.

REFERENCES

1. Peleg AY, Seifert H, Paterson DL. *Acinetobacter baumannii*: emergence of a successful pathogen. *Clin Microbiol Rev.* 2008;21:538–82.
2. Arakawa Y, Shibata N, Shibayama K, et al. Convenient test for screening metallo-beta-lactamase-producing gram-negative bacteria by using thiol compounds. *J Clin Microbiol.* 2000;38:40–3.
3. Martins AF, Kuchenbecker R, Sukiennik T, et al. Carbapenem-resistant *Acinetobacter baumannii* producing the OXA-23 enzyme: dissemination in Southern Brazil. *Infection.* 2009;35:474–6.
4. Woodford N, Ellington MJ, Coelho JM, et al. Multiplex PCR for genes encoding prevalent OXA carbapenemases in *Acinetobacter* spp. *Int J Antimicrob Agents.* 2006;27:351–3.
5. Walsh TR, Bolmström A, Qwärnström A, Gales AC. Evaluation of a new Etest for detecting metallo- β -lactamases in routine clinical testing. *J Clin Microbiol.* 2002;40:2755–9.
6. Pasteran F, Veliz O, Rapoport M, Guerriero L, Corso A. Sensitive and specific modified Hodge test for KPC and metallo-beta-lactamase detection in *Pseudomonas aeruginosa* by use a novel indicator strain, *Klebsiella pneumoniae* ATCC 700603. *J Clin Microbiol.* 2011;49:4301–3.
7. Yigit H, Queenan AM, Anderson GJ, et al. Novel Carbapenem-hydrolyzing beta-lactamase, KPC-1, from a carbapenem-resistant strain of *Klebsiella pneumoniae*. *Antimicrob Agents Chemother.* 2001;45:1151–61.

8. Danel F, Frére JM, Livermore DM. Evidence of dimerization among class D b-lactamases: kinetics of OXA-14 b-lactamase. *Biochim Biophys Acta*. 2001;1546:132-42.

Andreza F. Martins^{a,*}, Aline Borges^b, Mariana Pagano^b,
Liberia Maria Dalla-Costa^c, Afonso L. Barth^d

^a Centro Universitário IPA Metodista, Porto Alegre, RS, Brazil

^b Medical Sciences Post-Graduate Program, Universidade Federal do Rio Grande do Sul, Porto Alegre, RS, Brazil

^c Hospital de Clínicas, Universidade Federal do Paraná (HC/UFPR), Curitiba, PR, Brazil

^d Infectious Diseases Unit, Hospital de Clínicas de Porto Alegre, Porto Alegre, RS, Brazil

*Corresponding author at: Centro Universitário Metodista do IPA, Porto Alegre, 372/3 Padre Cacique Avenue, Porto Alegre, RS 90000-000, Brazil.

E-mail address: andreza.20@pop.com.br (A.F. Martins).

Received 2 January 2013

Accepted 6 January 2013

Available online 11 July 2013

1413-8670/\$ – see front matter

© 2013 Elsevier Editora Ltda. All rights reserved.

<http://dx.doi.org/10.1016/j.bjid.2013.01.010>