



High Frequency of OXA-253-Producing *Acinetobacter baumannii* in Different Hospitals in Recife, Brazil

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ABSTRACT Here, we report the isolation of 31 *Acinetobacter baumannii* strains producing OXA-253 in a single large Brazilian city. These strains belonged to five different sequence types (STs), including 4 STs not previously associated with *bla*_{OXA-253}. In all strains, the *bla*_{OXA-253} gene was located in a plasmid within a genetic environment similar to what was found previously in Brazil and Italy. The reported data emphasize the successful transmission of the *bla*_{OXA-253} gene through a large area and the tendency for this resistance determinant to remain in the *A. baumannii* population.

KEYWORDS *Acinetobacter*, MLST, antibiotic resistance, genome analysis, oxacillinase

Acinetobacter baumannii is an opportunistic Gram-negative pathogen responsible for a large number of outbreaks of hospital-acquired infections. This bacterium has been linked with serious infections affecting mainly debilitated patients in intensive care units (1), and it can become resistant to a wide range of antibiotics, leading to serious impediments to treatment. Carbapenems, such as imipenem and meropenem, have been often used as drugs of last resort, but outbreaks of carbapenem-resistant *A. baumannii* have been reported in several countries, including Brazil (2).

Resistance to carbapenems in *A. baumannii* may be due to various mechanisms, including the loss of outer membrane protein (OMP), overexpression of efflux pumps, or through alterations in the penicillin-binding protein (3). An increase in reported carbapenem resistance by *A. baumannii* isolates, however, has been mostly attributed to yet another mechanism, the production of carbapenemases, such as those belonging to the class D OXA type (4).

Here, we report OXA-253-producing *A. baumannii* strains isolated from different hospitals localized within the city of Recife, state of Pernambuco, in northeastern Brazil. This variant of the OXA-143-like carbapenemase belongs to the Ambler class D of β -lactamases (5), which has previously been the subject of studies discussing its incidence in Brazil. So far, however, the *bla*_{OXA-143} gene has only been reported for samples from the states of São Paulo and Rio de Janeiro, over 1,800 km to the south of Recife (2, 6–8).

Identification of the OXA-253-producing strains occurred after sequencing of the genomes of 45 *A. baumannii* strains displaying an extensively drug-resistant (XDR) phenotype and isolated from different individuals hospitalized in five public hospitals in Recife between 2010 and 2014. These strains were considered resistant to the two carbapenems tested (imipenem and meropenem) according to the Brazilian Committee

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on Antimicrobial Susceptibility Testing (BrCAST) guidelines, using the broth microdilution method. A detailed report of the whole-genomic data analysis, in the context of resistance and virulence characteristics, will be provided in future publications. Analysis of the sequencing data led to identification in 31 strains of the recently described class D β -lactamase *bla*_{OXA-253} (9). According to a multilocus sequence typing (MLST) analysis based on the sequenced genes, these strains were assigned to five different STs (ST 1, ST 15, ST 25, ST 79, and ST 113). So far, the only report of *bla*_{OXA-253} in Brazil (9) is from a single *A. baumannii* strain from the city of Belo Horizonte. This strain belongs to ST 113, the same ST associated with 12 of the *bla*_{OXA-253}-positive strains from Recife. Considering that the two cities are geographically far apart (~2,000 km), the evidence indicates a large distribution of both the ST and the resistance gene in Brazil. Nevertheless, since 19 strains producing OXA-253 in Recife are distributed between the four other STs (ST 1, ST 15, ST 25, and ST 79), and to our knowledge, this is the first report of multiple STs related to strains carrying the *bla*_{OXA-253} gene and distinct from ST 113, the data highlight the potential for this gene to spread further.

To confirm the plasmid localization of the *bla*_{OXA-253} gene, and in order to minimize assembly errors due to lower sequence coverage in some strains, two different approaches were carried out in order to assemble plasmid-derived DNA. For the first approach, the whole set of sequenced reads was used for a *de novo* assembly of the *bla*_{OXA-253} gene and neighboring sequences. For the second approach, the reads used for the *de novo* assembly were only those which were first seen to map to plasmid-derived DNA sequences deposited at the European Nucleotide Archive. The two approaches yielded similar results, although the second approach sometimes resulted in shorter contigs. Within the different genomes, the resulting contigs harboring the *bla*_{OXA-253} gene also varied in size, depending on the strain and the sequence coverage. Nevertheless, multiple strains (from various STs) yielded large contigs, which represented up to 91% identity and 97% coverage of the previously described plasmid pABVA01, found in two OXA-24-producing *A. baumannii* strains in Italy (10). Manual inspection of smaller contigs and reads matching to the plasmid pABVA01 also led to the identification, for most strains, of overlapping contigs covering nearly all of the plasmid and compatible with circular DNA elements 9 to 10 kb in length. The genetic environment surrounding the *bla*_{OXA-253} gene was also investigated and, in general, the genetic organization seen was similar to what was found in *A. baumannii* isolates producing OXA-253 and OXA-24 in Brazil and Italy, respectively (9, 10). In almost all isolates, the *bla*_{OXA-253} gene was found to be flanked by conserved elements, including genes coding for hypothetical proteins, a membrane protein, and a TonB-dependent receptor gene.

Overall, the data presented confirm the plasmid localization of the *bla*_{OXA-253} gene for most, if not all, strains studied here, suggesting a constant and efficient dispersion of the *bla*_{OXA-253} gene between different *A. baumannii* isolates. This report then draws attention to the highly successful transmission of OXA-253-producing *A. baumannii* in Brazil and the tendency of the *bla*_{OXA-253} gene to remain in the bacterial population over the years, reinforcing the potential for this gene to expand further.

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REFERENCES

- Howard A, O'Donoghue M, Feeney A, Sleator RD. 2012. *Acinetobacter baumannii*: an emerging opportunistic pathogen. *Virulence* 3:243–250. <https://doi.org/10.4161/viru.19700>.
- Mostachio AK, Levin AS, Rizek C, Rossi F, Zerbini J, Costa SF. 2012. High prevalence of OXA-143 and alteration of outer membrane proteins in carbapenem-resistant *Acinetobacter* spp. isolates in Brazil. *Int J Antimicrob Agents* 39:396–401. <https://doi.org/10.1016/j.ijantimicag.2012.01.021>.
- Kim YJ, Kim SI, Kim YR, Hong KW, Wie SH, Park YJ, Jeong H, Kang MW. 2011. Carbapenem-resistant *Acinetobacter baumannii*: diversity of resis-

- tant mechanisms and risk factors for infection. *Epidemiol Infect* 140: 137–145.
4. Kulah C, Mooij MJ, Comert F, Aktas E, Celebi G, Ozlu N, Rijnsburger MC, Savelkoul PH. 2010. Characterisation of carbapenem-resistant *Acinetobacter baumannii* outbreak strains producing OXA-58 in Turkey. *Int J Antimicrob Agents* 36:114–118. <https://doi.org/10.1016/j.ijantimicag.2010.03.017>.
 5. Higgins PG, Poirer L, Lehmann M, Nordmann P, Seifert H. 2009. OXA-143, a novel carbapenem-hydrolyzing class D β -lactamase in *Acinetobacter baumannii*. *Antimicrob Agents Chemother* 53:5035–5038. <https://doi.org/10.1128/AAC.00856-09>.
 6. Antonio CS, Neves PR, Medeiros M, Mamizuka EM, Elmor de Araújo MR, Lincopan N. 2011. High prevalence of carbapenem-resistant *Acinetobacter baumannii* carrying the *bla*_{OXA-143} gene in Brazilian hospitals. *Antimicrob Agents Chemother* 55:1322–1323. <https://doi.org/10.1128/AAC.01102-10>.
 7. Werneck JS, Picão RC, Girardello R, Cayô R, Marguti V, Dalla-Costa L, Gales AC. 2011. Low prevalence of *bla*_{OXA-143} in private hospitals in Brazil. *Antimicrob Agents Chemother* 55:4494–4495. <https://doi.org/10.1128/AAC.00295-11>.
 8. Gionco B, Pelayo JS, Venancio EJ, Cayô R, Gales AC, Carrara-Marroni FE. 2012. Detection of OXA-231, a new variant of *bla*_{OXA-143} in *Acinetobacter baumannii* from Brazil: a case report. *J Antimicrob Chemother* 67: 2531–2539. <https://doi.org/10.1093/jac/dks223>.
 9. Girlich D, Damaceno QS, Oliveira AC, Nordmann P. 2014. OXA-253, a variant of the carbapenem-hydrolyzing class D β -lactamase OXA-143 in *Acinetobacter baumannii*. *Antimicrob Agents Chemother* 58:2976–2978. <https://doi.org/10.1128/AAC.02640-13>.
 10. D'Andrea MM, Giani T, D'Arezzo S, Capone A, Petrosillo N, Visca P, Luzzaro F, Rossolini GM. 2009. Characterization of pABVA01, a plasmid encoding the OXA-24 carbapenemase from Italian isolates of *Acinetobacter baumannii*. *Antimicrob Agents Chemother* 53:3528–3533. <https://doi.org/10.1128/AAC.00178-09>.