International Journal of Infectious Diseases 17 (2013) e739–e743

Contents lists available at SciVerse ScienceDirect

International Journal of Infectious Diseases

journal homepage: www.elsevier.com/locate/ijid

Prevalence of carbapenemase-encoding genes including New Delhi metallo-β-lactamase in *Acinetobacter* species, Algeria

Esma Mesli [a,b], Meryem Berrazeg [a,b], Mourad Drissi [b], Souad Naïma Bekkhoucha [c], Jean-Marc Rolain [a,*]

[a] Aix-Marseille Université, Unité de Recherche en Maladies Infectieuses et Tropicales Emergentes (URMITE), UM63, CNRS 7278, IRD 198, Inserm 1095, IHU Méditerranée Infection, Faculté de Médecine et de Pharmacie, 27 Bd Jean Moulin, 13385 Marseille Cedex 05, France
[b] Laboratoire Antibiotiques, Antifongiques: Physico-chimie, Synthèse et Activité Biologique (LAPSAB), Faculté des Sciences de la Nature et de la Vie et Sciences de la Terre et de l'Univers, Université Abou Bekr Belkaid-Tlemcen, Tlemcen, Algeria
[c] Laboratoire Central, Hôpital Universitaire, Oran, Algeria

ARTICLE INFO

Article history:
Received 6 February 2013
Accepted 26 February 2013

Corresponding Editor: Eskild Petersen, Aarhus, Denmark

Keywords:
Acinetobacter spp
NDM-1-producing bacteria
Algeria

SUMMARY

Background: Nosocomial infections caused by carbapenem-resistant *Acinetobacter spp* are a global health problem. The aim of this study was to investigate the molecular epidemiology and the genetic support of carbapenem resistance in *Acinetobacter spp* clinical isolates recovered from three different hospitals in western Algeria from 2008 to 2012.

Methods: A total of 113 *Acinetobacter spp* isolates were identified by matrix-assisted laser desorption ionization-time of flight (MALDI-TOF) mass spectrometry. Antimicrobial susceptibility testing was carried out, and minimum inhibitory concentrations (MICs) were determined by the dilution method on Mueller–Hinton agar for β-lactams, aminoglycosides, fluoroquinolones, and colistin. The characterization of β-lactamases was investigated by phenotypic tests for the detection of metallo-β-lactamases and oxacillinases. Resistance genes were screened for by quantitative PCR and sequenced when positive.

Results: Among the 113 isolates, 80 (70.8%) were found to be resistant to imipenem with MICs ranging from 64 to 512 μg/ml. The $bla_{\text{OXA-23-like}}$ gene was detected in 50% (40/80) of the isolates and the $bla_{\text{OXA-24-like}}$ gene was detected in 21.2% (17/80) of the isolates. In addition, the metallo-β-lactamase $bla_{\text{NDM-1-like}}$ was detected in five isolates (6.2%).

Conclusions: This study represents the first description of autochthonous *Acinetobacter spp* producing metallo-β-lactamase $bla_{\text{NDM-1-like}}$ and oxacillinases $bla_{\text{OXA-23-like}}$ and $bla_{\text{OXA-24-like}}$ in western Algeria.

1. Introduction

Acinetobacter spp are major nosocomial pathogens. The genus currently consists of more than 40 species, including validly published species and genomic species.[1] Of these, *Acinetobacter baumannii* is the most clinically relevant *Acinetobacter* species. It has emerged as a major cause of healthcare-associated infections including pneumonia, urinary tract infection, and septicemia.[2] It has the ability to develop resistance to multiple classes of useful antimicrobial agents.[3] Closely related species, *Acinetobacter nosocomialis* (formerly named *Acinetobacter* genomic species (gen. sp.) 13TU) and *Acinetobacter pittii* (formerly named *Acinetobacter* gen. sp. 3), have also been associated with nosocomial infections and outbreaks.[4] These three clinically important species are phenotypically and genotypically difficult to differentiate, thus they are grouped together into the *Acinetobacter calcoaceticus–Acinetobacter baumannii* (ACB) complex.[1] They are so much alike that they cannot be differentiated using routine commercial systems. Genotypic methodologies can be used to differentiate them, such as the determination of specific gene sequences, including the 16sRNA, *recA*, *rpoB*, and *gyrB* genes, in combination with the technology of matrix-assisted laser desorption ionization-time of flight (MALDI-TOF) mass spectrometry (MS).[5]

The efficacy of carbapenems against multidrug-resistant *Acinetobacter spp* has been undermined by the emergence of Ambler class B and class D carbapenemase-hydrolyzing β-lactamases.[6] The class D carbapenemase (oxacillinase) found in *A. baumannii* can be clustered into four distinct groups: OXA–23-like (OXA–23, OXA–27 and OXA–49), OXA–24-like (OXA–24/40, OXA–25, OXA–26 and OXA–72), OXA–58-like (OXA–58 and OXA–96), and OXA–51-like enzymes.[7] The last group constitutes a family of chromosomal enzymes typically present in *A. baumannii*.[8] The high-level carbapenem resistance due to the expression of genes encoding the class D carbapenemases, requires a strong

* Corresponding author. Tel.: +33 4 91 32 43 75; fax: +33 4 91 38 77 72.
E-mail address: jean-marc.rolain@univ-amu.fr (J.-M. Rolain).

1201-9712/$36.00 – see front matter
http://dx.doi.org/10.1016/j.ijid.2013.02.024

promoter such as that provided by the mobile insertion sequence *IS*Aba1.[9] This is characteristic of *A. baumannii*, and most outbreaks of carbapenem-resistant *A. baumannii* associated with bla_{OXA-23} have been identified using primers based on *IS*Aba1.[6]

The new metallo-β-lactamase (MBL), New Delhi metallo-β-lactamase 1 (NDM-1), was initially reported in *Klebsiella pneumoniae* clinical isolates from a Swedish patient who had previously been hospitalized in India.[10] Recently, cases of NDM-producing *A. baumannii* have been described in several countries worldwide, including Canada, USA, Sweden, UK, Austria, Belgium, France, Netherlands, Germany, Japan, Africa, Oman, and Australia.[11,12] At present, the worldwide caseload is probably being driven by people infected or colonized in the Indian subcontinent before traveling elsewhere. However, there is already evidence of others reservoirs of infected patients in the Balkan states, the Middle East, and Israel, suggesting that the gene may become endemic worldwide, similar to the bla_{KPC} gene, which is now endemic in Greece and Israel.[13,14]

In the present study, we evaluated the prevalence of antibiotic resistance and the genetic background of carbapenem resistance in a series of 113 *A. baumannii* strains isolated in western Algeria between October 2008 and April 2012. We report five bla_{NDM-1}-positive *A. baumannii* strains recovered from autochthonous cases in the same area.

2. Materials and methods

Bacterial isolates of *Acinetobacter spp* were recovered from three different hospitals situated in north-western Algeria (Tlemcen, Oran, and Sidi Bel Abbes). All of them were isolated from the hospital environment and patients admitted to the intensive care unit (ICU) and hematology, surgery, and neurosurgery wards, during the study period of October 2008 to April 2012. They were identified using MALDI-TOF MS, which was performed with a Bruker Daltonics Microflex (Bremen, Germany) using 96-spot polished-steel targets.

Antimicrobial susceptibility was determined by disk diffusion and agar dilution methods, in accordance with the Comité de l'Antibiogramme de la Société Française de Microbiologie (CA-SFM) 2008 guidelines. Antibiotic disks were purchased from Bio-Rad (Marnes-la-Coquette, France). The minimum inhibitory concentrations (MICs) were determined by agar dilution method in Mueller–Hinton medium (Fluka BioChemika, Spain) and E-test strips for carbapenems (imipenem, meropenem) (bioMérieux, Marcy l'Etoile, France). Isolates with MICs of imipenem >8 μg/ml and inhibition zone diameter <17 mm were investigated in this study. The double-disk synergy test (DDST) was used to detect MBL.

Strains showing non-susceptibility to carbapenems were screened for the production of acquired carbapenem-hydrolyzing class D β-lactamase: bla_{OXA-23}, bla_{OXA-24}, bla_{OXA-58}, intrinsic β-lactamase bla_{OXA-51}, and MBL bla_{NDM-1}. Quantitative real-time PCR (CFX96, C1000 Thermal Cycler, Bio-Rad) and standard PCR were carried out to detect the encoding genes. MasterMix was prepared in accordance with the manufacturer's instructions and positive controls carrying each gene were used to determine the efficacy of the real-time PCR assay. The experimental run protocol used was as follows: denaturation program (95 °C for 15 min), amplification and quantification programs repeated 35 times (95 °C for 30 s, 60 °C for 1 min). Samples were considered positive if a threshold cycle was reached during the 35 cycles or less. Standard PCR analysis was performed for $bla_{VIM-like}$, $bla_{GIM-like}$, $bla_{IMP-like}$, $bla_{KPC-like}$, $bla_{NDM-1-like}$, $bla_{CTX-M-like}$, $bla_{SHV-like}$, $bla_{TEM-like}$, $bla_{PER-like}$, and $bla_{GES-like}$. PCR screening was also performed for aminoglycoside-modifying enzyme and fluoroquinolone resistance genes (*aac(3)-Ia*, *aac(6′)-Ib*, *aadA*, *ant(2′′)-I*, *aph(3′)-VI*, *armA*, *rmtA*, *rmtF*, *arr-2*, *qnrA*, and *qnrB).* Oligonucleotide primers and probes used are listed in the **Supplementary Material (Table S1)**. Purified PCR products were sequenced using BigDye terminator chemistry on an automated ABI 3730 sequencer (PE Applied Biosystems, Foster City, CA, USA) based on Sanger's sequencing method. Data collection and analysis were performed using CodonCode Aligner 3.7.1.1 sequencing analysis software.

3. Results

In total, 100 human isolates and 13 hospital environment isolates were collected from Tlemcen Hospital, Oran Hospital, and Sidi Bel Abbes Hospital (51, 45, and 17, respectively). Overall, 106 strains were identified as *A. baumannii*, one strain as *A. radioresistens* (from Sidi Bel Abbes Hospital), two strains as *A. nosocomialis* (from Oran Hospital), and four strains as *A. pittii* (from Tlemcen, Oran, and Sidi Bel Abbes hospitals). All the isolates were identified to the species level with a log score >2.0; the mass spectrometry-based identification scheme yielded identical results compared against the default Bruker database. A mean spectra projection (MSP) dendrogram was generated on the basis of consensus spectra obtained from each bacterium (Figure 1).

The overall susceptibility of all the strains according to the French CA-SFM breakpoints showed that most of the isolates were characterized by resistance to β-lactams (piperacillin 92.2%, piperacillin–tazobactam 88%, ticarcillin 95.9%, ticarcillin–clavulanic acid 96.2%, ceftazidime 98.6%), to fluoroquinolones (ciprofloxacin 85%, with MICs ranging from 0.125 to 0.25 μg/ml), and to aminoglycosides (amikacin 79.1%, gentamicin 56.1%, and tobramycin 38.9%, with MICs ranging from 1 to 512 μg/ml), whilst they differed in their susceptibility to imipenem (70.8%) and showed different levels of resistance with MICs ranging from 0.5 to 512 μg/ml. However, all isolates were susceptible to colistin (MIC 0.125–0.25 μg/ml) (Table 1).

Eighty imipenem-resistant strains (with MIC ranging from 64 to 512 μg/ml), including 42 (82%) imipenem-resistant *A. baumannii* from Tlemcen Hospital, 31 (69%) imipenem-resistant *Acinetobacter spp* from Oran Hospital (30 *A. baumannii* and one *A. nosocomialis*), and seven (41%) imipenem-resistant *Acinetobacter spp* from Sidi Bel Abbes Hospital (six *A. baumannii* and one *A. radioresistens*) were screened for the presence of carbapenemase-encoding genes (Table 2). Real-time PCR results showed that 40 out of 80 imipenem-resistant isolates were positive for the bla_{OXA-23} gene (31 *A. baumannii* from Tlemcen Hospital, seven *A. baumannii* and one *A. nosocomialis* from Oran Hospital, and one *A. baumannii* from Sidi Bel Abbes Hospital) and 17 isolates harbored the bla_{OXA-24} gene (four *A. baumannii* from Tlemcen Hospital, 11 *A. baumannii* from Oran Hospital, and one *A. baumannii* and one *A. radioresistens* from Sidi Bel Abbes Hospital), of which five coexisted with the OXA–23 gene (Table 3).

In addition, among all the isolates, five from Oran were positive for the MBL NDM-1. All five isolates showed positivity by DDST. The gene was sequenced and revealed 99% identity to the sequence reported in the GenBank database under accession number **JQ739157.1**. The five NDM-1-positive isolates were from autochthonous cases in five patients admitted to the ICU and hematology wards of Oran Hospital. All five bla_{NDM-1}-positive isolates were identified as *A. baumannii*. The earliest positive isolate was collected in April 2011 from a 38-year-old man hospitalized on the hematology ward who was then transferred to the ICU of Oran Hospital for a severe cranial trauma subsequent to a stair fall. He had no relevant travel history and neither did his family. Antibiotics used were ceftazidime, amikacin, and colistin, then imipenem and colistin. The patient died in July 2011. The four other patients were all men aged up to 38 years who were admitted to the same ICU as the first patient during the period April to August 2011. Unfortunately no additional clinical records were available for these patients.

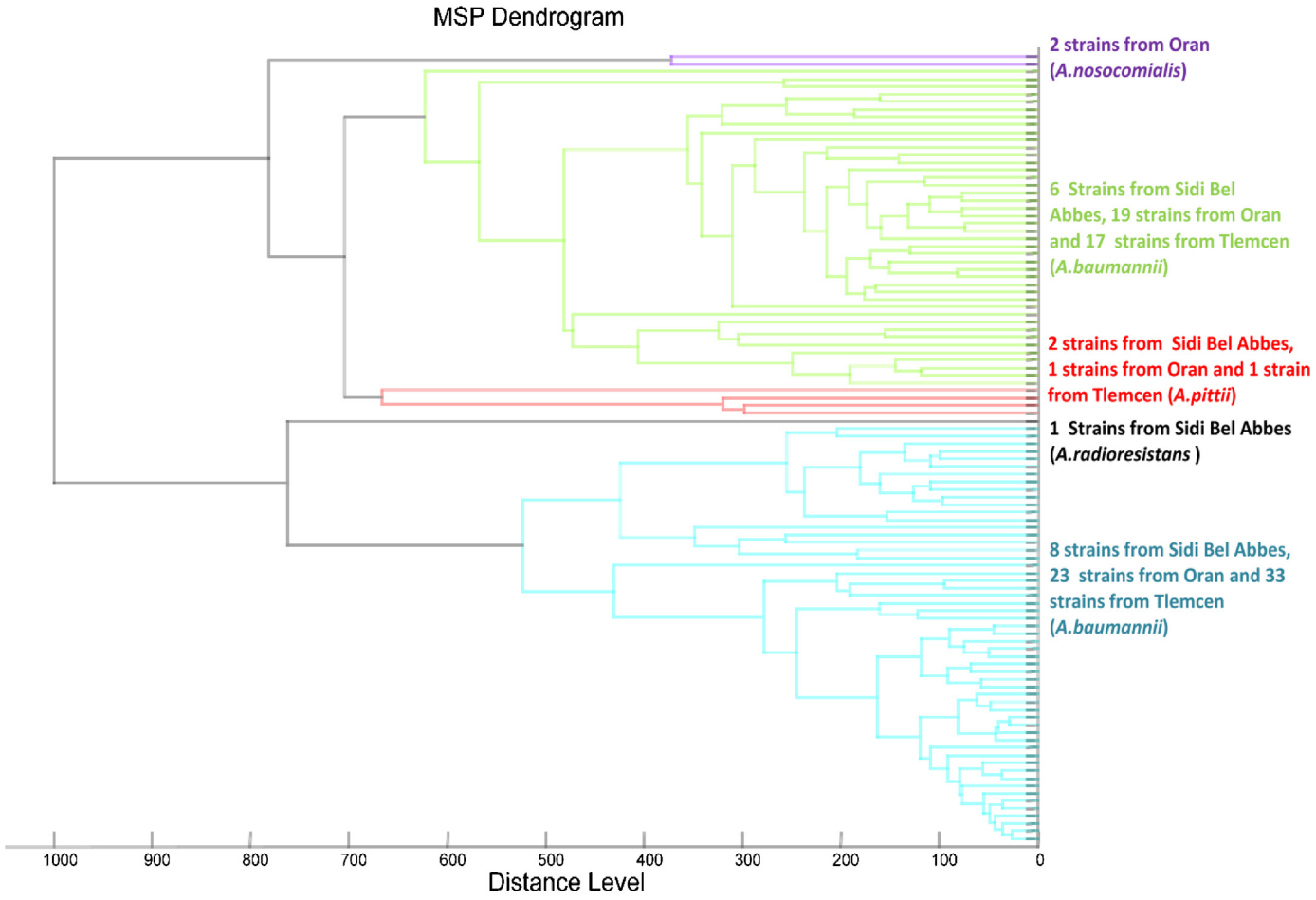

Figure 1. Mean spectra projection (MSP) dendrogram generated by BIOTYPER software (version 2; Bruker Daltonics) of *Acinetobacter spp* strains.

No $bla_{VIM-like}$, $bla_{GIM-like}$, $bla_{IMP-like}$, $bla_{CTX-M-like}$, $bla_{SHV-like}$, $bla_{TEM-like}$, $bla_{PER-like}$ $bla_{GES-like}$, $bla_{KPC-like}$, or bla_{OXA-58} were detected in the collected strains. Resistance to aminoglycosides (gentamicin, tobramycin, and amikacin) observed in almost all the isolates was due to the expression of *aac(3)-Ia* (77 isolates), *aadA* (57 isolates), *ant(2″)-I* (60 isolates), *aph(3′)* (70 isolates), and *aac(6′)-Ib* (one isolate) genes. No isolates were positive for *rmtA*, *rmtF*, *armA*, *arr-2*, or the *qnr* genes.

4. Discussion

Acinetobacter spp has recently emerged as one of the most important opportunistic nosocomial pathogens. Although *A. baumannii* is the most important species in clinical settings, the other *Acinetobacter spp*, such as *A. pittii* and *A. nosocomialis*, are also frequently isolated in hospitals and have been involved in a number of outbreaks in ICUs.[3] The analysis of the dendrogram generated by BIOTYPER software showed that the protein signatures formed five separate clusters related to each one of the species, excluding *A. baumannii* strains that form two separates clusters. This is consistent with the findings of Espinal et al., who showed that MALDI-TOF MS is able to identify and class Acinetobacter strains in separate clusters.[5]

In our study, we investigated the high prevalence of carbapenemase-encoding genes (OXA-type carbapenemase and bla_{NDM-1}) in *Acinetobacter spp*. OXA-type carbapenemase-producing

Table 1
Resistance rates for *Acinetobacter spp* isolates in this study

Antimicrobial agent	Resistance rate (%)			
	Tlemcen ($n=51$)	Oran ($n=45$)	Sidi Bel Abbes ($n=17$)	Total ($n=113$)
Piperacillin	94.1	95.5	87.0	92.2
Piperacillin–tazobactam	84.3	91.0	88.8	88.0
Ticarcillin	98.0	97.7	92.0	95.9
Ticarcillin–clavulanic acid	100.0	97.7	91.0	96.2
Ceftazidime	98.0	100.0	98.0	98.6
Imipenem	78.0	71.0	35.0	61.3
Meropenem	84.0	77.7	38.0	66.5
Gentamicin	50.9	26.6	91.0	56.1
Tobramycin	23.0	57.7	36.0	38.9
Amikacin	82.3	71.0	84.0	79.1
Ciprofloxacin	88.2	91.0	76.0	85.0
Colistin	0.0	0.0	0.0	0.0

Table 2
Isolates of *Acinetobacter spp* in relation to the presence of carbapenemase enzymes

Carbapenemases					Hospital location	Species	No. of isolates	Samples
OXA–51	OXA–23	OXA–24	OXA–58	NDM				
+	+	+	–	–	Tlemcen	*A. baumannii*	4	Tracheal aspirate, rectal swab, urine, environment
					Oran	*A. baumannii*	1	Tracheal aspirate
					Sidi Bel Abbes	-	0	-
+	+	–	–	–	Tlemcen	*A. baumannii*	27	Tracheal aspirate, rectal swab, urine, wound environment
					Oran	*A. baumannii* ($n=6$) *A. nosocomialis* ($n=1$)	7	Wound, tracheal aspirate
					Sidi Bel Abbes	*A. baumannii*	1	Wound, environment
+	–	+	–	–	Tlemcen	-	0	-
					Oran	*A. baumannii*	10	Urine, tracheal aspirate
					Sidi Bel Abbes	*A. baumannii* ($n=1$) *A. radioresistens* ($n=1$)	2	Tracheal aspirate
+	–	–	–	–	Tlemcen	*A. baumannii* ($n=13$) *A. pittii* ($n=1$)	14	Tracheal aspirate, environment, urine
					Oran	*A. baumannii* ($n=8$) *A. pittii* ($n=1$)	9	Tracheal aspirate, urine
					Sidi Bel Abbes	*A. baumannii* ($n=5$) *A. pittii* ($n=1$)	6	Tracheal aspirate
+	–	–	–	+	Tlemcen	-	0	-
					Oran	*A. baumannii*	5	Urine
					Sidi Bel Abbes	-	0	-
–	–	–	–	–	Tlemcen	-	0	-
					Oran	*A. nosocomialis*	1	Urine
					Sidi Bel Abbes	*A. pittii*	1	Wound

Table 3
Distribution of carbapenem-encoding genes in the three hospitals

No. of imipenem-resistant	OXA–23 (%)	OXA–24 (%)	OXA–58 (%)	NDM-1 (%)
Tlemcen ($n=42$)	31 (74.0%)	4 (9.5%)	0 (0%)	0 (0%)
Oran ($n=31$)	8 (25.8%)	11 (35.4%)	0 (0%)	5 (16.0%)
Sidi Bel Abbes ($n=7$)	1 (14.0%)	2 (28.0%)	0 (0%)	0 (0%)
Total ($n=80$)	40	17	0	5

A. baumannii are increasingly reported from Europe, South America, Asia Oceania, and Africa.[11,15,16] There is a worldwide variation in the rate of carbapenem resistance of *A. baumannii* from one geographical area to another.[12] In Algeria, the dissemination of OXA–23 carbapenemases among *A. baumannii* isolates has also been reported since 2010.[15–17] In our series of isolates, the main molecular support explaining the resistance to carbapenems is the presence of bla_{OXA-23} carbapenemase-encoding genes, along with the coexistence of bla_{OXA-24}. Consequently, the isolates demonstrated high rates of co-resistance to all other classes of antimicrobial agents tested. A limited number of antimicrobial agents maintain reliable levels of activity against OXA–23-producing *A. baumannii*.[18] Neither of the non-*baumannii Acinetobacter* showed the coexistence of bla_{OXA-23} with bla_{OXA-24}, in contrast to *A. baumannii* isolates, of which five harbored both genes at the same time.

OXA–23 (formerly ARI-1) was originally reported in an *A. baumannii* detected in Scotland in 1985.[19] In a report by Opazo et al., the bla_{OXA-23} gene is reported to have originated in the chromosome of *A. radioresistens*, which might be the natural reservoir of these enzymes[20] that are currently emerging as the sources of carbapenem resistance in *A. baumannii* worldwide.[21] Although OXA–58 has previously been detected in Tlemcen and Annaba hospitals,[16,22] none of the isolates in this series were positive for this gene. The $bla_{OXA-51-like}$ gene, originally intrinsic to *A. baumannii*, was detected in all the isolates except one *A. pittii* and one *A. nosocomialis*. These $bla_{OXA-51-like}$ genes, all preceded by *IS*Aba1, may confer a high level of carbapenem resistance. They were probably located on plasmids that might have emerged between different clones of non-*baumannii Acinetobacter* species and also between *A. baumannii* clones. The plasmid-borne *IS*Aba1–$bla_{OXA-51-like}$ in non-*baumannii Acinetobacter* species not only contributes to a high level of carbapenem resistance, but also affects the accuracy of using $bla_{OXA-51-like}$ detection as a tool for differentiating *A. baumannii* from other *Acinetobacter* species.[23]

In the present study, we found five strains producing the bla_{NDM-1} gene in autochthonous cases in the ICU of Oran Hospital between April and August 2011. No bacterial isolates harboring the bla_{NDM-1} gene were detected in Algeria from the beginning of the study (2008) until this period. The global distribution of the bla_{NDM-1} gene has been extensively described.[24] It has been found in diverse isolates since it was first discovered in *K. pneumoniae* in 2008.[10] The potential presence of this gene in non-*baumannii Acinetobacter* should receive proper attention. All five of the bla_{NDM-1}-positive isolates were identified as *A. baumannii*, suggesting that this species, which has a robust survival capability, can easily acquire foreign resistance genes.[25] NDM-1-producing *A. baumannii* has already been described in two Algerian patients. The patients were hospitalized in Oran ICU and transferred to French[26] and Belgian[27] hospitals. Patient histories were confirmed as lacking any foreign travel, suggesting that NDM-producing *A. baumannii* isolates may have already spread in North Africa.[26,27] Reports describing NDM-type carbapenemase-producers isolated from patients previously hospitalized in high-prevalence countries is increasing.[28] However, the geographic origin and the time of the first appearance of this gene are unknown.[12] A recent study has suggested that the putative original source of the bla_{NDM-1} gene could be from the chromosome of plant pathogens, such as

Pseudoxanthomonas and related bacteria widespread in the environment.[29] The spread of strains carrying the bla_{NDM-1} gene will enhance the likelihood of variants emerging. Interestingly, we have evidence that NDM-encoding genes may be widespread in *A. baumannii*, and further molecular surveys will be necessary to evaluate their distribution in that species. Many studies have constituted reports on carbapenem-resistant *A. baumannii* whose carbapenem resistance is mediated mainly by OXA-type carbapenemases. Despite being less commonly identified in *A. baumannii* than oxacillinase, NDM-1 is currently spreading worldwide and could be reported with a high frequency as a mediator of carbapenem resistance. It is thus critical to survey the presence of this gene in multidrug-resistant (MDR) *A. baumannii* isolates worldwide.

Although polymyxins such as colistin (polymyxin E) have not typically been included in regimens to treat Acinetobacter infections because of their neurotoxicity and nephrotoxicity, they are now considered as one of the last resorts against MDR Acinetobacter infections. Owing to the increasing use of colistin against Gram-negative pathogens and the high recombination rate of *Acinetobacter spp*, it is of concern that colistin resistance in *Acinetobacter spp* isolates may increase rapidly.[30]

In conclusion, the spread of NDM-1-positive *A. baumannii* isolates in the hospital setting reemphasizes the need for strict adherence to surveillance programs in order to prevent the colonization, the infection, and the dissemination of this gene in Algeria.

Acknowledgements

We thank Linda Hadjadj for technical assistance.

Funding: This work was partly funded by CNRS and IHU Méditerranée Infection in Marseille, France, and LAPSAB Tlemcen Laboratory, Algeria.

Conflict of interest: None to declare.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.ijid.2013.02.024.

References

1. Dijkshoorn L, Nemec A, Seifert H. An increasing threat in hospitals: multidrug-resistant *Acinetobacter baumannii*. *Nat Rev Microbiol* 2007;**5**:939–51.
2. Kempf M, Rolain JM. Emergence of resistance to carbapenems in *Acinetobacter baumannii* in Europe: clinical impact and therapeutic options. *Int J Antimicrob Agents* 2012;**39**:105–14.
3. Park S, Kim HS, Lee KM, Yoo JS, Yoo JI, Lee YS, et al. Molecular and epidemiological characterization of carbapenem-resistant *Acinetobacter baumannii* in non-tertiary Korean hospitals. *Yonsei Med J* 2013;**54**:177–82.
4. Nemec A, Krizova L, Maixnerova M, van der Reijden TJ, Deschaght P, Passet V, et al. Genotypic and phenotypic characterization of the *Acinetobacter calcoaceticus–Acinetobacter baumannii* complex with the proposal of *Acinetobacter pittii* sp. nov. (formerly *Acinetobacter* genomic species 3) and *Acinetobacter nosocomialis* sp. nov. (formerly *Acinetobacter* genomic species 13TU). *Res Microbiol* 2011;**162**:393–404.
5. Espinal P, Seifert H, Dijkshoorn L, Vila J, Roca I. Rapid and accurate identification of genomic species from the *Acinetobacter baumannii* (Ab) group by MALDI-TOF MS. *Clin Microbiol Infect* 2012;**18**:1097–103.
6. Livermore DM. The impact of carbapenemases on antimicrobial development and therapy. *Curr Opin Investig Drugs* 2002;**3**:218–24.
7. Poirel L, Nordmann P. Carbapenem resistance in *Acinetobacter baumannii*: mechanisms and epidemiology. *Clin Microbiol Infect* 2006;**12**:826–36.
8. Heritier C, Poirel L, Fournier PE, Claverie JM, Raoult D, Nordmann P. Characterization of the naturally occurring oxacillinase of *Acinetobacter baumannii*. *Antimicrob Agents Chemother* 2005;**49**:4174–9.
9. Turton JF, Ward ME, Woodford N, Kaufmann ME, Pike R, Livermore DM, Pitt TL. The role of *IS*Aba1 in expression of OXA carbapenemase genes in *Acinetobacter baumannii*. *FEMS Microbiol Lett* 2006;**258**:72–7.
10. Yong D, Toleman MA, Giske CG, Cho HS, Sundman K, Lee K, Walsh TR. Characterization of a new metallo-beta-lactamase gene, bla(NDM-1), and a novel erythromycin esterase gene carried on a unique genetic structure in *Klebsiella pneumoniae* sequence type 14 from India. *Antimicrob Agents Chemother* 2009;**53**:5046–54.
11. Rolain JM, Parola P, Cornaglia G. New Delhi metallo-beta-lactamase (NDM-1): towards a new pandemia? *Clin Microbiol Infect* 2010;**16**:1699–701.
12. Wilson ME, Chen LH. NDM-1 and the role of travel in its dissemination. *Curr Infect Dis Rep* 2012;**14**:213–26.
13. Livermore DM, Walsh TR, Toleman M, Woodford N. Balkan NDM-1: escape or transplant? *Lancet Infect Dis* 2011;**11**:164.
14. Nordmann P, Poirel L, Walsh TR, Livermore DM. The emerging NDM carbapenemases. *Trends Microbiol* 2011;**19**:588–95.
15. Bakour S, Kempf M, Touati A, Ait Ameur A, Haouchine D, Sahli F, Rolain JM. Carbapenemase-producing *Acinetobacter baumannii* in two university hospitals in Algeria. *J Med Microbiol* 2012;**61**:1341–3.
16. Touati M, Diene SM, Racherache A, Dekhil M, Djahoudi A, Rolain JM. Emergence of bla(OXA–23) and bla(OXA–58) carbapenemase-encoding genes in multidrug-resistant *Acinetobacter baumannii* isolates from University Hospital of Annaba, Algeria. *Int J Antimicrob Agents* 2012;**40**:89–91.
17. Kempf M, Bakour S, Flaudrops C, Berrazeg M, Brunel JM, Drissi M, et al. Rapid detection of carbapenem resistance in *Acinetobacter baumannii* using matrix-assisted laser desorption ionization-time of flight mass spectrometry. *PLoS One* 2012;**7**:e31676.
18. Dalla-Costa LM, Coelho JM, Souza HA, Castro ME, Stier CJ, Bragagnolo KL, et al. Outbreak of carbapenem-resistant *Acinetobacter baumannii* producing the OXA–23 enzyme in Curitiba, Brazil. *J Clin Microbiol* 2003;**41**:3403–6.
19. Paton R, Miles RS, Hood J, Amyes SG, Miles RS, Amyes SG. ARI 1: beta-lactamase-mediated imipenem resistance in Acinetobacter baumannii. *Int J Antimicrob Agents* 1993;**2**:81–7.
20. Opazo A, Dominguez M, Bello H, Amyes SG, Gonzalez-Rocha G. OXA-type carbapenemases in *Acinetobacter baumannii* in South America. *J Infect Dev Ctries* 2012;**6**:311–6.
21. Poirel L, Figueiredo S, Cattoir V, Carattoli A, Nordmann P. *Acinetobacter radioresistens* as a silent source of carbapenem resistance for *Acinetobacter spp*. *Antimicrob Agents Chemother* 2008;**52**:1252–6.
22. Drissi M, Poirel L, Mugnier PD, Baba AZ, Nordmann P. Carbapenemase-producing *Acinetobacter baumannii*, Algeria. *Eur J Clin Microbiol Infect Dis* 2010;**29**:1457–8.
23. Lee YT, Kuo SC, Chiang MC, Yang SP, Chen CP, Chen TL, Fung CP. Emergence of carbapenem-resistant non-*baumannii* species of *Acinetobacter* harboring a blaOXA–51-like gene that is intrinsic to *A. baumannii*. *Antimicrob Agents Chemother* 2012;**56**:1124–7.
24. Kumarasamy KK, Toleman MA, Walsh TR, Bagaria J, Butt F, Balakrishnan R, et al. Emergence of a new antibiotic resistance mechanism in India, Pakistan, and the UK: a molecular, biological, and epidemiological study. *Lancet Infect Dis* 2010;**10**:597–602.
25. Peleg AY, Seifert H, Paterson DL. *Acinetobacter baumannii*: emergence of a successful pathogen. *Clin Microbiol Rev* 2008;**21**:538–82.
26. Boulanger A, Naas T, Fortineau N, Figueiredo S, Nordmann P. NDM-1-producing *Acinetobacter baumannii* from Algeria. *Antimicrob Agents Chemother* 2012;**56**:2214–5.
27. Bogaerts P, Rezende de CR, Roisin S, Deplano A, Huang TD, Hallin M, et al. Emergence of NDM-1-producing *Acinetobacter baumannii* in Belgium. *J Antimicrob Chemother* 2012;**67**:1552–3.
28. Hrabak J, Stolbova M, Studentova V, Fridrichova M, Chudackova E, Zemlickova H. NDM-1-producing Acinetobacter baumannii isolated from a patient repatriated to the Czech Republic from Egypt, July 2011. *Euro Surveill* 2012;17. pii: 20085.
29. Sekizuka T, Matsui M, Yamane K, Takeuchi F, Ohnishi M, Hishinuma A, et al. Complete sequencing of the bla(NDM-1)-positive IncA/C plasmid from *Escherichia coli* ST38 isolate suggests a possible origin from plant pathogens. *PLoS One* 2011;**6**:e25334.
30. Park YK, Jung SI, Park KH, Cheong HS, Peck KR, Song JH, Ko KS. Independent emergence of colistin-resistant *Acinetobacter spp*. isolates from Korea. *Diagn Microbiol Infect Dis* 2009;**64**:43–51.