

# Microbiology 2018: Phenotypic and genotypic characterization of carbapenem-resistant *Acinetobacter baumannii* clinical isolates from Alexandria

Elsayed Aboumagd, Alaa Abouelftouh and Aisha Torky

Alexandria University, Alexandria, Egypt

Antibiotic use in Egypt is largely under-regulated leading to the formation of resistant isolates. Carbapenems are last resort agents for treating infections of *Acinetobacter baumannii* that are resistant to other antibiotic classes. Alarming, however, carbapenem-resistant isolates are emerging. This research aimed at characterizing seventy-four carbapenem-unsusceptible A, both phenotypically and molecularly. *Baumannii* isolates from Egypt to detect the various enzymes responsible for the resistance to carbapenem.

Carbapenemase development was assessed using a variety of phenotypic methods: modified Hodge test (MHT), carbapenem inactivation (CIM), combined disk test (CDT), CarbAcineto NP test, and boronic acid testing. The polymerase chain reaction (PCR) was used to test isolates for the existence of certain genes responsible for carbapenem resistance, as well as sequences for insertion.

*Acinetobacter baumannii* has become a pathogen which threatens life. It causes nosocomial infections worldwide, including infections of the skin and soft tissue, wound and bloodstream infections, infections of the urinary tract, meningitis, and ventilator-associated pneumonia, which is the most common and fatal infection from A. *Baumannii*. These infections are especially dangerous owing to the ability of the pathogen to withstand the action of most antibacterial agents currently available, making *A. Baumannii* one of the most dangerous organisms in the ESKAPE. The number of community-acquired A, in addition. Infections of *baumannii* such as bacteremia, pneumonia, meningitis and endocarditis have been increasing progressively in the last few decades.

Resistance to Carbapenem in *A. Baumannii* strains are caused by loss or alteration of porins or, in some rare cases, by alteration of penicillin binding proteins. The key resistance mechanism, however, is the development of  $\beta$ -lactamase enzymes. Four classes of molecular  $\beta$ -lactamase (A, B, C and D) were detected at A. *Baumannii*. Just a few K. *Pneumoniae* carbapenemase (KPC) type enzymes of class A  $\beta$ -lactamases have an effect on carbapenems compared to those of class B and class D that act effectively on carbapenems. Class B  $\beta$ -lactamases are metallo- $\beta$ -lactamases (MBL) needed for their catalytic activity by zinc ions.

Detection of carbapenemases is crucial to determine the extent of the problem and to guide the application of guidelines on antimicrobial stewardship to limit further production of

carbapenem-resistant variants between A. They isolate the *baumannii*. The present study reports the prevalence of certain carbapenemases among CR-AB isolates from Alexandria, Egypt, in contrast to the various phenotypic and molecular techniques used in an Egyptian environment to detect these enzymes among CR-AB isolates.

## Discussion

*A. Baumannii* is becoming a big concern due to the awful number of nosocomial infections caused by this pathogen, mainly at ICUs around the world. Moreover, A. Due to the excessive use of antibiotics, *baumannii* has become resistant to many groups of antimicrobials, contributing to the predominance of multidrug-resistant strains, especially in hospitals. What's more, A resistance to carbapenem. *Baumannii* isolates limits the clinical treatment choices for these infections that may lead to higher morbidity and mortality rates. A few earlier studies have commented on the prevalence of Egyptian *A carbapenemases*. Clinical isolates of the *baumannii*.

This research aimed to classify 74 Egyptian A, both phenotypically and molecularly. *Baumannii* isolates the various enzymes which are responsible for carbapenem resistance. Several phenotypic methods have been used including the testing of MHT, CIM, CDT, CarbAcineto NP and boronic acid discs. Phenotypic carbapenemase detection has the advantages of low cost, simplicity of operation and lack of complicated or costly equipment; however, it suffers from poor specificities and sensitivity. As a result, PCR screening was taken as the gold standard for some genes responsible for carbapenem resistance, as well as some insertion sequences to determine the sensitivity of the various phenotypic methods.

Comparing the results of the phenotypic tests with the results of carbapenemase molecular detection, the sensitivity of MHT, CIM, CDT, CarbAcineto NP, imipenem and meropenem boronic acid was 78.4, 68.9, 79.7, 95.9, 56.8 and 70.3 per cent respectively. Four isolates carrying blaOXA-51, blaOXA-23, blaVIM in the CarbAcineto NP test including isolate no. A81 which additionally carried blaOXA-58 developed the positive outcome in less than 15 min which could be attributed to the enzyme activity in these isolates. The false negative result observed with A2 carrying blaVIM, blaOXA-51 and blaOXA-23 could be explained by low concentration of zinc in the culture medium, or by very low activity of carbapenemase in the tested isolate. These findings are consistent with the

previously reported high sensitivity of CarbAcineto NP in carbapenemase detection among *Acinetobacter* spp. Although only 21 isolates had positive allover tests, all nine isolates shown to be carrying NDM by PCR were also positively tested in MHT, CIM and CarbAcineto NP. sensitive to detect MBL 100%. On the other hand, in one isolate, CDT failed to detect NDM: A85 and boronic acid disc test with imipenem and meropenem failed to detect NDM in 2 isolates , respectively: A59 and A81 and A40 and A81. It is noteworthy that A81 was the only isolate shown to carry blaOXA-58.The insertion sequences may increase the production of  $\beta$ -lactamases when present upstream to CHDL encoding genes. The prevalence of ISAbal, ISAbal2 and ISAbal3, respectively, was 100, 2.7 and 4.1 percent in the current report. The occurrence of various sequences of insertion at A. Clinical isolates of Baumannii from Saudi Arabia were in agreement with the findings of the current study.

### Conclusions

With the exception of CarbAcineto NP which showed superior sensitivity approaching PCR findings, a combination of phenotypic tests, including MHT, CIM, CDT and boronic acid disk tests, seems to be necessary for the definitive detection of carbapenemases. NDM prevalence levels detected here are lower than previously reported from other parts of the country, suggesting the need for greater screening of different Egyptian governorates to determine the precise prevalence rate. Yet the prevalence levels of OXA-23 and VIM remain equally high.