Four genotypes of carbapenem-resistant Acinetobacter baumannii strains lacking OXA-23 production in Korea

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Abstract

During nationwide Fantimicrobial surveillance (Korea Global Antimicrobial Resistance Surveillance System [Kor-GLASS]), the recent emergence of non-oxacillinase (OXA)-23 production by carbapenem-resistant Acinetobacter baumannii (CRAB) isolates was noted. In this study, we evaluated resistance mechanisms other than OXA-23 production to elucidate the shift in considerable CRAB clones. The presence of OXA carbapenemase genes, such as blaOXA-23, blaOXA-24, blaOXA-58, and blaOXA-51-ISAb1, was determined by PCR. Other carbapenemase genes, such as blaimp, blaOXA, blaNDM, blaKPC, blaGES, and blaOXA-48, were determined using sequencing. Strains lacking carbapenemase genes were subjected to whole genome sequencing, and resistance genes were analyzed using ResFinder. Four CRAB strains were collected through a Kor-GLASS study in 2022, in which OXA-23 production was not identified. The carbapenemase genotypes of the four CRAB strains lacking blaOXA-23 were blaOXA-51-ISAb1, blaciROX-6/ACD25, blaciROX-182, and bliciROX-48. To the best of our knowledge, this is the first study to identify CRAB producing New Delhi metallo-β-lactamase (NDM)-1 in Korea. In conclusion, domestic CRAB resistance mechanisms may undergo subtle changes. Continuous observations are required to monitor the emergence of new clones.

Keywords: Carbapenem, Resistance, Acinetobacter baumannii, NDM-1, OXA-23

Acinetobacter baumannii is an important pathogen that causes healthcare-associated infections, such as ventilator-associated pneumonia, line-associated bloodstream infections, and catheter-associated urinary tract infections [1]. Carbapenem is usually considered a treatment option for extended-spectrum β-lactamase producers. The rapid increase in carbapenem-resistant A. baumannii (CRAB) isolation has been correlated with an increased nationwide prescription rate of carbapenems [2]. The carbapenem resistance rate is very
high in strains isolated in Korea, and multidrug resistance is common, hindering the selection of therapeutic options [3]. According to Kor-GLASS (Korea Global Antimicrobial Resistance Surveillance System) data, the imipenem-resistance rate of \textit{A. baumannii} blood isolates was $> 90\%$ [3].

There are three classes of carbapenemase:Ambler class A (serine carbapenemases), class B (metallo-\(\beta\)-lactamase), and class D (oxacillinase carbapenemases) [4]. \textit{Klebsiella pneumoniae} carbapenemase (class A), New Delhi metallo-\(\beta\)-lactamase (NDM, class B), and oxacillinase-48 (class D) are common in carbapenem-resistant Enterobacteriaceae [5]. Metallo-\(\beta\)-lactamases, such as Guiana extended-spectrum \(\beta\)-lactamase, imipenemase (IMP), Verona integron-encoded metallo-\(\beta\)-lactamase (VIM), and NDM, are frequently found in carbapenem-resistant \textit{Pseudomonas aeruginosa} [6].

Carbapenemase types in carbapenem-resistant organisms other than \textit{A. baumannii} vary. However, CRAB isolates uniformly carry \textit{bla}_{OXA-23}, in Korea because of the notorious multidrug resistance clone, \textit{A. baumannii} global clone 2 with sequencing type 191, which has become predominant in clinical settings worldwide, including Korea [7]. OXA-type \(\beta\)-lactamases are the primary resistance mechanism for CRAB, and a drastic increase in \textit{A. baumannii} isolates with \textit{bla}_{OXA-23} has been observed since the mid-2000s [7]. IS\textit{Aba1}-associated \textit{bla}_{OXA-51}\textsubscript{S}, another contributor to CRAB, has decreased since the mid-2000s [7].

The recent emergence of non-OXA-23 production of CRAB isolates was noted in a Kor-GLASS study. Therefore, in the present study, resistance mechanisms other than OXA-23 production were evaluated to elucidate the shift in significant CRAB clones.

In total, 366 \textit{A. baumannii} isolates were collected according to the Kor-GLASS protocol in 2022 [8]. \textit{A. baumannii} strains were identified using Matrix-Assisted Laser Desorption/Ionization Time-of-Flight Mass Spectrometry (Bruker Biotyper; Bruker Daltonics GmbH), and positive OXA-51 polymerase chain reaction (PCR) results confirmed the species identification. PCR methods determined the existence of OXA carbapenemase genes, such as \textit{bla}_{OXA-23}, \textit{bla}_{OXA-24}, \textit{bla}_{OXA-51}, and \textit{bla}_{OXA-51}\textsubscript{S}-IS\textit{Aba1} [8]. PCR sequencing methods also determined the presence of other carbapenemase genes, such as \textit{bla}_{IMP}, \textit{bla}_{VIM}, \textit{bla}_{NDM}, \textit{bla}_{KPC}, \textit{bla}_{GES}, and \textit{bla}_{OXA-48} [8]. Strains without determination of carbapenemase genes using the aforementioned methods were subjected to whole genome sequencing, as previously described [9]. Using the NextSeq 550 instrument (Illumina), the entire genome was sequenced with 8 \(\mu\)g of input genomic DNA. Sequences were assembled using Spades (version 3.11.1) and annotated using Prokka (version 1.13.7). Resistance genes were determined using ResFinder 4.5 [10].

The resistance rate to imipenem was 85.2\% in \textit{A. baumannii} isolates in 2022, and 98.7\% in OXA-24 producers. The carbapenemase genotypes of the four CRAB strains lacking \textit{bla}_{OXA-23} were \textit{bla}_{OXA-46}/ \textit{ACD-25}, \textit{bla}_{OXA-51}/IS\textit{Aba1}, \textit{bla}_{OXA-48}, and \textit{bla}_{NDM-1} (Table 1).
Carbapenem-resistant *A. baumannii* without OXA-23 production

<table>
<thead>
<tr>
<th>Genotypes</th>
<th>No. (%)</th>
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<tbody>
<tr>
<td>OXA-23</td>
<td>308 (98.7)</td>
</tr>
<tr>
<td>Non-OXA-23</td>
<td>312 (85.2)</td>
</tr>
<tr>
<td><em>bla</em>$_{OXA-66/ACD-25}$</td>
<td>4 (1.3)</td>
</tr>
<tr>
<td><em>bla</em>$_{OXA-182/-ISABA1}$</td>
<td>1 (0.3)</td>
</tr>
<tr>
<td><em>bla</em>$_{OXA-182}$</td>
<td>1 (0.3)</td>
</tr>
<tr>
<td><em>bla</em>$_{NDM-1}$</td>
<td>1 (0.3)</td>
</tr>
</tbody>
</table>

In conclusion, the mechanism of resistance via OXA-23 in CRAB may have subtly changed with the emergence of NDM-1-producing *A. baumannii* in Korea. Continuous observation is required to monitor the emergence of new clones.

**Ethics statement**

This study was approved by the Institutional Review Board of the National Health Insurance Ilsan Hospital (No. 2024-05-016), and the requirement for informed consent was waived.

**Conflicts of interest**

Jeong Hwan Shin is an associate editor and Young Uh and Nam Hee Ryoo are editorial board members of the *Annals of Clinical Microbiology*. However, they were not involved in the review process of this article. No other potential conflict of interest relevant to this article was reported.
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References