Prevalence and characterization of Carbapenem-Resistant Enterobacterales among inpatients and outpatients in Skikda, Algeria

Amina Bougouizi¹, Zohra Chekroud¹, Hamza Rahab², Ali Boumegoura², Abdelaziz Touati³

¹ Laboratoire de recherche des Interactions, Biodiversité, Ecosystèmes et Biotechnologie, faculté des Sciences de la Nature et de la Vie, Algeria
² Biotechnology Research Center - C.R.Bt Constantine, Algeria
³ Laboratoire d'Ecologie Microbienne, FSNV, Université de Bejaia, Algeria

Abstract
Introduction: The spread of Carbapenemase-producing Enterobacterales (CPEs) has become a significant concern in Algeria, with limited data available on their presence in community settings. This research investigated the resistance mechanisms of carbapenem-resistant Enterobacterales (CREs) collected from hospitals and the community in Skikda city, Algeria, between December 2020 and June 2022.

Methodology: The study collected Enterobacterales strains resistant to ertapenem from inpatient and outpatient populations. An automated system was used for identification and antibiotic susceptibility testing. β-lactamase production was evaluated through phenotypic tests and confirmed by standard PCR. Lastly, the carbapenemase genes were sequenced using the Sanger method.

Results: 17 CRE were isolated, with 9 from inpatients and 8 from outpatients. These isolates belonged to four species: Klebsiella pneumoniae (n = 8), Escherichia coli (n = 6), Enterobacter cloacae (n = 1), and Proteus mirabilis (n = 1). Of 15 CPEs, 11 were extended-spectrum β-lactamases (ESBLs) positive, 5 were plasmid-mediated cephalosporinase (AmpC) positive, and 1 harbored all three β-lactamases. All metallo-β-lactamase-producing strains carried the New Delhi metallo-beta-lactamase gene (blaNDM), including 5 NDM-1 and 7 NDM-5 variants. The presence of blaOXA-48 and blaOXA-244 was observed in one outpatient strain each. NDM was associated with Cefotaximase Munich (CTX-M) ESBL in 8 isolates, while Cephamycinase (CMY) was detected in 3 NDM-5-producing E. coli.

Conclusions: This research highlights the rising prevalence of carbapenemases NDM-1 and NDM-5 among inpatients and outpatients and supports the notion that OXA-48 is becoming increasingly widespread beyond Algerian hospitals.

Key words: Carbapenem-resistant Enterobacterales (cres); Algeria; inpatients, outpatients; Enterobacterales.


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Introduction
Enterobacterales are among the most common human pathogens, causing infections such as cystitis, pneumonia, pyelonephritis, sepsis, meningitis, peritonitis, and medical device infections. These agents, particularly Escherichia coli and Klebsiella pneumoniae are known to be the most frequent causes of both community- and hospital-acquired infections [1-2].

The global emergence of Multidrug-resistant (MDR) Enterobacterales is a growing concern and constitutes a major threat to public health. The widespread production of carbapenem-hydrolyzing β-lactamases (carbapenemases) is the leading cause of resistance to carbapenem antibiotics, leaving limited options for treating infections caused by these highly drug-resistant bacterial strains [3]. It is widely recognized that the carbapenem-resistance phenomenon can be attributed to the presence of three distinct classes of carbapenemases: Class A, including the KPC enzyme, Class B metallo-β-lactamases (MβLs), which encompass VIM, IMP, and NDM, and Class D represented mainly by the OXA-48-like enzyme [4-5].

The New Delhi MβL enzyme (NDM) is a highly problematic and rapidly spreading form of carbapenem resistance. NDM can cause resistance to a broad range of antibiotics, making it extremely difficult to treat. In recent years, cases of NDM have become endemic in the Arabian Peninsula, northern Africa, and the Balkans. Among the 25 different NDM variants, NDM-1 and NDM-5 are the two most commonly found in Enterobacterales [6].

The blaOXA-48 gene has become increasingly widespread, particularly in Mediterranean countries, since its initial detection in a K. pneumoniae strain in
Turkey [5]. The OXA-48 class of carbapenemases, known as OXA-48-like, encompasses several variants that vary in amino acids but have not become as widespread as OXA-48. Some variants, including OXA-244 and OXA-232, have weaker hydrolytic activity against carbapenems than OXA-48 [7]. North African countries are hotspots for OXA-type carbapenemases [5]. NDM and OXA-48 are frequently identified in *K. pneumoniae* and *Escherichia coli*, but can also be found in other *Enterobacterales* species [8-9].

In Algeria, the emergence and spread of clinically significant CPE strains have been documented, with NDM-1, NDM-5, and OXA-48 enzymes being the most frequently identified in hospital and community settings [4,10–12]. To gain a comprehensive understanding of the presence and distribution of CPE strains in both hospital and community settings, particularly in smaller cities, this study was conducted in Skikda, Algeria. The aim was to characterize the *Enterobacterales* isolates that showed decreased susceptibility to carbapenems obtained from hospitalised and outpatients.

**Methodology**

**Bacterial isolates and species identification**

In this study, ertapenem-resistant isolates of *Enterobacterales* were collected from inpatient samples (urine and pus) obtained from the main hospital in Skikda and outpatient samples obtained from private laboratories. These specimens were taken from a variety of pathological sources. Isolates were obtained by aseptic plating of specimens on three different culture media: Nutrient agar, Hektoen, and CHROMagar, followed by incubation for 24 hours at 37°C. The susceptibility of the isolates to Ertapenem was determined using the established disc diffusion method on Mueller Hinton Agar.

Identification of the isolates was confirmed with automated systems, the API20E biochemical gallery (BioMérieux in Marcy-l'Étoile, France) and the Vitek® 2 Compact 15 automated system (BioMérieux).

**In vitro antibiotic susceptibility testing**

To determine the minimum inhibitory concentrations (MICs) of Ertapenem-resistant *Enterobacterales* isolates, the Vitek® 2 Compact 15 automated system was utilized. The AST 365 card, which comprises a panel of antibiotics including Ampicillin, Amoxicillin + clavulanic acid, Piperacillin + tazobactam, Cefazolin, Cefoxitin, Cefotaxime, Ceftazidime, Imipenem, Ertapenem, Amikacin, Gentamycin, Ciprofloxacin, Chloramphenicol, Nitrofurantoin, Trimethoprim-Sulfamethoxazole, and Fosfomycin for *K. pneumoniae*, was used in the analysis. The obtained MIC values were interpreted based on the guidelines set by the Clinical Laboratory Standard Institute (CLSI) (CLSI 2020, Version of M02 M07 M11, 30th ed) [13].

The wild-type control strain *E. coli* ATCC 25922 was utilized, and the Broth Microdilution method was used to determine colistin's minimum inhibitory concentration (MIC) [13].

**Phenotypic Characterization of Carbapenemase, ESBL, and AmpC production**

The presence of carbapenemase was determined phenotypically using the Modified Carba NP test described previously by Bakour et al [14].

The inhibitory effect of ethylene-diamine-tetraacetic acid (EDTA) on MβL activity was studied using the method of Yong et al. [15] with a slight modification.

### Table 1. Primers used in the PCR reaction.

<table>
<thead>
<tr>
<th>Target gene</th>
<th>Primer sequence 5'-3'</th>
<th>Amplicon Size (Pb)</th>
<th>Annealing Temperature (°C)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>blaNDM</em></td>
<td>F-CATTTTCGCCGTTTATG</td>
<td>1022</td>
<td>52</td>
<td>[42]</td>
</tr>
<tr>
<td></td>
<td>R-CTGGTACCAGGATATG</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>blaOXA</em></td>
<td>F-TGGTACCAGGATATG</td>
<td>744</td>
<td>54</td>
<td>[43]</td>
</tr>
<tr>
<td></td>
<td>R-GAGCCTTTTGGTATG</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>blaKPC</em></td>
<td>F-ATGGTACGATGATCCGCTT</td>
<td>893</td>
<td>55</td>
<td>[42]</td>
</tr>
<tr>
<td></td>
<td>R-ATGAAAGTGGTGGAGAC</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>blaVIM</em></td>
<td>F-ATGGTACGATGATCCGCTT</td>
<td>382</td>
<td>54</td>
<td>[44]</td>
</tr>
<tr>
<td></td>
<td>R-ATGAAAGTGGTGGAGAC</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>blaIMP</em></td>
<td>F-CATTTTCGCCGTTTATG</td>
<td>448</td>
<td>53</td>
<td>[43]</td>
</tr>
<tr>
<td></td>
<td>R-ATATTTTGGCGCTTATTGG</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>blaTEM</em></td>
<td>F-ATGGTACGATGATCCGCTT</td>
<td>840</td>
<td>50</td>
<td>[17]</td>
</tr>
<tr>
<td></td>
<td>R-ATGAAAGTGGTGGAGAC</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>blaSHV</em></td>
<td>F-ATGGTACGATGATCCGCTT</td>
<td>1051</td>
<td>53</td>
<td>[45]</td>
</tr>
<tr>
<td></td>
<td>R-ATGCCCTTTTTCGAGCT</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>blaCTX-M</em></td>
<td>F-CATTTTCGCCGTTTATG</td>
<td>500</td>
<td>50</td>
<td>[17]</td>
</tr>
<tr>
<td></td>
<td>R-CTGGTCCGCTTTTCAAGAATGCG</td>
<td>1200</td>
<td>50</td>
<td>[17]</td>
</tr>
</tbody>
</table>

R: Reverse; F: Forward; bp: base pair.
modification - the imipenem disc was substituted with an ertapenem disc. The boronic acid test was used to identify Ambler class A carbapenemase KPC, as described by Tsakris et al [16].

The production of ESBL was evaluated using the Double Disc Synergy Test (DDST) [17]. This test was further conducted in a cloxacinill-supplemented medium [18]. The DDST involved placing the following antibiotic discs in a 2 cm inter-disk distance: cefotaxime, cefepime, aztreonam, ceftazidime, and cefoxitin, along with a disc of amoxicillin + clavulanic acid.

Molecular characterization of β-lactamase production

The extraction of DNA was carried out using the boiling lysis method, previously described by Feria et al [19].

The sequence of the most prevalent genes encoding carbapenemase-hydrolyzing enzymes, such as \( \text{bla}_{\text{OXA-48}}, \text{bla}_{\text{NDM}}, \text{bla}_{\text{KPC}}, \text{bla}_{\text{VIM}}, \text{and} \text{bla}_{\text{IMP}}, \) as well as the common ESBL gene \( \text{bla}_{\text{CTX-M}} \), and the major plasmid-mediated AmpC gene \( \text{bla}_{\text{CMY}} \), were detected using standard simplex PCR technology and specific primers. The primer sequences for each targeted gene are listed in Table 1. In the case of strains showing resistance to colistin, the plasmid-mediated mcr-1 gene was also targeted using the primer sequences and protocol described by Rebello et al [20].

The amplification process was carried out using the following conditions: After a preliminary denaturation step at 95 °C for 15 minutes, 30 cycles of denaturation at 94 °C for 30 seconds, annealing for 30 seconds at a temperature determined by the primer sequence (as listed in Table 1), and elongation at 72 °C for 2 minutes were performed. The procedure was concluded with a final elongation step at 72 °C for 10 minutes. The amplified products were analyzed on a 1.5% agarose gel, stained with SYBR Safe DNA gel stain (Invitrogen, Spain), and visualized using a UV transilluminator (GEL Doc XR + Gel Documentation System from BIORAD, USA /Thermo Fisher Scientific).

The sequences of positive PCR products were obtained through purification and sequencing with a 3500 XL Genetic Analyzer (Applied Biosystems, Thermo Fisher Scientific in California, USA). The sequences were then compared to the Antibiotic Resistance Gene-ANNOTATION database (ARG-ANNOT) using the BLAST program available on the National Center for Biotechnology Information's website (www.ncbi.nlm.nih.gov).

Results

Bacterial isolates identification

17 CRE-positive isolates were identified using the disk diffusion method, with 11 isolates collected from urine and 6 from pus. Most isolated strains came from elderly patients (87.24%) aged 51 to 93. The patient information and strain data can be found in Table 2.

The identified species were \( K. \) pneumoniae (\( n = 9 \)), \( E. \) coli (\( n = 6 \)), Enterobacter cloacae (\( n = 1 \)), and Proteus mirabilis (\( n = 1 \)). \( K. \) pneumoniae was the dominant pathogen among outpatients, while \( E. \) coli was the most frequently encountered species among hospitalized patients (\( n = 5 \)).

In vitro antibiotic susceptibility

The results from the Vitek-2 automated system showed that 11 out of the 17 CRE isolates were multidrug-resistant, meaning that 54% of the CPE were resistant to at least three different classes of antibiotic.

<table>
<thead>
<tr>
<th>Source</th>
<th>Ward</th>
<th>Gender</th>
<th>Age</th>
<th>Specimen</th>
<th>ID</th>
<th>MCNP</th>
<th>EDTA</th>
<th>Ab</th>
<th>ESBL</th>
<th>AmpC</th>
<th>Carbanepenase genes</th>
<th>Additional beta-lactamase genes</th>
</tr>
</thead>
<tbody>
<tr>
<td>H</td>
<td>Nephrology</td>
<td>F</td>
<td>75</td>
<td>Urine</td>
<td>( E. ) coli</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>H</td>
<td>Nephrology</td>
<td>F</td>
<td>82</td>
<td>Urine</td>
<td>( E. ) coli</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>H</td>
<td>Surgery</td>
<td>M</td>
<td>57</td>
<td>Diabetic pus</td>
<td>( K. ) pneumoniae</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>H</td>
<td>Surgery</td>
<td>F</td>
<td>71</td>
<td>Pus</td>
<td>( K. ) pneumoniae</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>H</td>
<td>Nephrology</td>
<td>F</td>
<td>55</td>
<td>Pus</td>
<td>( E. ) cloacae</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>None</td>
<td></td>
</tr>
<tr>
<td>H</td>
<td>ICU</td>
<td>M</td>
<td>60</td>
<td>Urine</td>
<td>( E. ) coli</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>H</td>
<td>Neonatology</td>
<td>M</td>
<td>Nb</td>
<td>Pus</td>
<td>( E. ) coli</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>H</td>
<td>Neonatology</td>
<td>F</td>
<td>Nb</td>
<td>Urine</td>
<td>( E. ) coli</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>H</td>
<td>Nephrology</td>
<td>M</td>
<td>72</td>
<td>Urine</td>
<td>( K. ) pneumoniae</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>Private</td>
<td>F</td>
<td>64</td>
<td>Pus</td>
<td>( K. ) pneumoniae</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>Private</td>
<td>F</td>
<td>88</td>
<td>Urine</td>
<td>( K. ) pneumoniae</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
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</tr>
<tr>
<td>C</td>
<td>Private</td>
<td>F</td>
<td>80</td>
<td>Urine</td>
<td>( K. ) pneumoniae</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>Private</td>
<td>F</td>
<td>93</td>
<td>Urine</td>
<td>( K. ) pneumoniae</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>Private</td>
<td>M</td>
<td>82</td>
<td>Urine</td>
<td>( K. ) pneumoniae</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>None</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>Private</td>
<td>M</td>
<td>72</td>
<td>Pus</td>
<td>( P. ) mirabilis</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>Private</td>
<td>F</td>
<td>51</td>
<td>Urine</td>
<td>( K. ) pneumoniae</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
<tr>
<td>C</td>
<td>Private</td>
<td>F</td>
<td>7</td>
<td>Urine</td>
<td>( E. ) coli</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>( \text{bla}<em>{\text{NDM}} ), ( \text{bla}</em>{\text{CMY}} )</td>
<td></td>
</tr>
</tbody>
</table>

H: Hospital; C: Community; M: Male; F: Female; Nb: Newborn, MDR: Multi-Drug-Resistance; MCNP: Modified Carba Np; Ab: Acid boronic; EDTA: Ethylene diamine tetra-acetic acid; ESBL: Extended-spectrum beta-lactamase.
including fluoroquinolones, sulfonamides, and one or two aminoglycosides in addition to β-lactams.

Most community- and hospital-acquired CPE isolates were resistant to all tested β-lactams and β-lactams combined with β-lactamase inhibitors. However, one strain was susceptible to ceftazidime with a MIC value of 4μg/mL, and five isolates had intermediate resistance to imipenem with a MIC value of 2μg/mL. Additionally, 82.35%, 64.70%, 47.05%, and 47.06% of the isolates were resistant to ciprofloxacin, trimethoprim-sulfamethoxazole, gentamicin, and nitrofurantoin, respectively. On the other hand, only 23.53% of the strains showed resistance to amikacin with a MIC value of ≥ 64 μg/mL. Aminoglycosides and chloramphenicol were found to be the most effective antibiotics against both hospital and community-acquired CRE isolates, as shown in Figure 1 and Table 3.

Table 4 lists the MIC50 and MIC90 values of the antibiotics against the CRE isolates. The MIC values of ertapenem were ≥ 8 μg/mL for 12 CRE strains (70.59%), 4 μg/mL for two isolates (11.76%), and 2 μg/mL for 4 isolates (23.53%). For imipenem, the MIC values were ≥ 16μg/mL for 11 strains (64.7%), 8 μg/mL for one isolate (5.88%), and 2 μg/mL for five isolates (11.76%).

Two K. pneumoniae isolates were resistant to colistin, MIC value of 4μg/mL, and isolates were also resistant to ertapenem and imipenem, MIC values of ≥ 8μg/mL and ≥ 16μg/mL, respectively.

**Phenotypic Characterization of Carbapenemase, ESBL, and AmpC production**

The modified Carba-NP test confirmed carbapenemase production in 15 isolates. The EDTA assay, which suggests the presence of MBL, was positive for 13 of the 15 isolates. However, all strains tested negative for the boronic acid test. The DDST, with and without cloxacillin, showed that 10 CPE strains produced ESBL and 5 AmpC. In two strains, both ESBL and AmpC mechanisms were detected.

**Molecular characterization of β-lactamase production**

Among the 17 CREs, six β-lactamase encoding genes were identified, as listed in Table 2. The most frequently detected carbapenemase gene was \( \text{bla}_{\text{NDM-5}} \)
(7 strains), followed by blaNDM-1 (5 strains). Other genes detected included blaOXA-48 (1 strain) and blaOXA-244 (1 strain). The blaNDM-5 was present in five E. coli and one K. pneumoniae, while blaNDM-1 was exclusively found in K. pneumoniae. The blaOXA-48 and blaOXA-244 genes were detected in one E. coli and one K. pneumonia strain, respectively. The targeted carbapenemase-encoding genes blakPC, blavIM, and blaimp were absent in all CRE strains.

Six of the 8 hospital-associated strains carried blaNDM-5 and two carried blaNDM-1. In contrast, among the 7 community strains, blakPC was the most frequently detected (3 strains), followed by blaNDM-5 (1 strain).

All 12 phenotypically confirmed ESBL-producing isolates carried the blacTX-M gene. The four non-ESBL E. coli isolates were AmpC producers, and three carried the blacMY gene. In two colistin-resistant K. pneumoniae mcr-1 gene was not detected.

Discussion

This study aimed to analyze the prevalence of CRE strains among hospitalized patients and outpatients in Skikda, Algeria. The findings revealed the presence of CRE strains in both hospital and community settings. Extended hospital stays, exposure to antibiotics, invasive medical devices, and severe secondary infections are major risk factors for acquiring carbapenem-resistant strains in hospitals. On the other hand, self-medication and excessive use of antibiotics are considered the main factors contributing to the spread of these strains in the community, posing a significant challenge to public health [21].

The results of this study on tested samples suggested that NDM could be the most prevalent carbapenemase mechanism in both hospital and community groups, which contradicts previous studies in Algeria that have identified OXA-48 as the most commonly isolated class D β-lactamase [4,5]. The CPE strains in this study were resistant to third-generation cephalosporins (3GC) and carried either the blacTX-M or blacMY gene, which was confirmed through the phenotypic detection of ESBL or AmpC production. The lower occurrence of OXA-48 in this study can be attributed to its less resistance to 3GC, which is only present when associated with another β-lactamase such as ESBL or AmpC.

Table 3. Antibiotic Minimum Inhibitory Concentrations (MICs) and Resistance Patterns of Tested Carbapenem-Resistant Enterobacteria (CREs).

<table>
<thead>
<tr>
<th>AMP</th>
<th>AUG</th>
<th>TAZ</th>
<th>CZ</th>
<th>FOX</th>
<th>CTX</th>
<th>CAZ</th>
<th>ERT</th>
<th>IMP</th>
<th>AK</th>
<th>GEN</th>
<th>CIP</th>
<th>FOS</th>
<th>NIT</th>
<th>CHL</th>
<th>SXT</th>
<th>COL</th>
<th>Resistance profile</th>
</tr>
</thead>
<tbody>
<tr>
<td>≥ 32</td>
<td>≥ 32</td>
<td>≥ 128</td>
<td>≥ 64</td>
<td>≥ 64</td>
<td>≥ 64</td>
<td>≥ 8</td>
<td>2</td>
<td>≤ 2</td>
<td>≤ 1</td>
<td>≥ 4</td>
<td>≤ 16</td>
<td>≤ 16</td>
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NR: Naturally resistant; AMP: Ampicillin; AUG: Amoxicillin + clavulanic acid; TAZ: Piperacillin + Tazobactam; CZ: Cefazolin; FOX: Cefoxitin; CTX: Cefotaxime; CAZ: Ceftazidime; ERT: Ertapenem; IMP: Imipenem; AK: Amikacin; GEN: Gentamicin; CIP: Ciprofloxacin; FOS: Fosfomycin; NIT: Nitrofurantoin; CHL: Chloramphenicol; SXT: Trimethoprim-Sulfamethoxazole. COL: Colistin.

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In the present study, NDM-1 and NDM-5 were identified as the only carbapenemases in the hospital setting, with NDM-5 being the most predominant. This finding aligns with previous studies in Algeria that have reported the circulation of both NDM-1 and NDM-5 in Algerian hospitals [22–25]. The detection of NDM-1 and NDM-5 in outpatients was also reported, with the detection of NDM-5 being the first in Algeria [10]. The presence of NDM-5 has been previously documented in non-hospitalized patients, healthy individuals, and animals in other countries, including Latin America [26] China [27], and Madagascar [28]. The presence of NDM-5 in the community could be due to various factors, including transmission from hospital patients or animals or even the environment, which could serve as a reservoir. NDM-5 has also been detected in Algeria in long-distance migratory birds [29], dogs [30], and raw milk [31].

The present study detected OXA-48 in one outpatient, which aligns with previous reports of its emergence in community settings in Batna and Annaba, Algeria [11-12]. Additionally, we detected the first clinically significant isolate carrying the blaOXA-244 gene in Algeria, specifically in a strain of K. pneumoniae. The OXA-244 carbapenemase, a variant of OXA-48 with a single substitution (Arg214Gly), is known to have decreased carbapenemase activity compared to OXA-48.[32] This variant of OXA has been widely reported in several countries, including Russia, Germany, France, the UK, Egypt, the Netherlands, Colombia, Turkey, and Lebanon, primarily in E. coli, but also in K. pneumoniae and Enterobacter aerogenes [32-33]. In Algeria, OXA-244-producing E. coli was detected in the Soummum River in Bejaia [34]. It is important to note that OXA-244-producing strains are known to have weak carbapenemase activity, making them difficult to detect and potentially contributing to their silent spread [32].

In the present study, NDM-producing strains were multi-drug resistant, unlike the OXA-48-like strains. This aligns with previous research which shows that NDM production is consistently associated with a multidrug-resistant phenotype [35]. On the other hand, OXA-type carbapenemases have limited activity against carbapenems and can only induce significant resistance when they are combined with an ESBL [36-37].

In our study collection, carbapenemase production was not detected in two isolates. Both isolates produced AmpC β-lactamase and one isolate was an ESBL producer. Isolates with AmpC production usually have ertapenem resistance due to hyperproduction of the enzyme [38].

This aligns with previous studies [10,39], suggesting that carbapenem resistance in these two strains is likely a result of a combination of ESBL or AmpC production and non-enzymatic resistance mechanisms [40-41].

Conclusions

The occurrence of CPE strains in both hospital and community settings is a cause for concern, as these bacteria are multi-drug resistant and challenging to treat. This study highlights the presence of NDM and OXA-48-like carbapenemases in Algeria, particularly in Skikda, where the NDM-1 and NDM-5 were found to be the most prevalent. The detection of the first human case carrying the blaOXA-244 gene and the first NDM-5-producing K. pneumoniae in the community underscores the need for immediate action to control the spread of these infections.

The spread of MDR bacteria in the community is a serious threat to public health, as it can increase the risk of healthcare-associated infections and impede effective treatment. The presence of MDR bacteria in the community also increases the risk of their transmission from person to person, exacerbating the problem.

In conclusion, the occurrence of CPE strains in hospital and community settings is worrisome and requires immediate attention and action. Effective monitoring, improved infection control measures, and implementation of infection prevention strategies are crucial for preventing the spread of these bacteria and ensuring the safety of public health.

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Authors’ contributions

AB collected the data, analyzed them, and wrote the article. ZC analyzed the microbiological data. HR and AB analyzed the molecular data AT analyzed the data and corrected the article.
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**Corresponding author**

Prof. A. Touati

Laboratoire d'Ecologie Microbienne, FSNV, Université de Bejaia, 06000 Bejaia, Algeria

Email: abdelaziz.touati@univ-bejaia.dz

**Conflict of interests:** No conflict of interests is declared.