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# Comparative analysis of bacteria with multidrug resistance (on the example of *Acinetobacter baumannii* and *Pseudomonas aeruginosa*)

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Abstract

Introduction: The study addresses the growing challenge of antibiotic resistance in *Acinetobacter baumannii* and *Pseudomonas aeruginosa*, emphasizing their resistance mechanisms and epidemiological significance both globally and in Ukraine.

Aim: The study aims to compare the main tools and the frequency of resistance of *A. baumannii* and *P. aeruginosa* in the modern population.

Material and methods: The current data on the degree of resistance to antimicrobial agents in *A. baumannii* and *P. aeruginosa* in Ukraine and worldwide were collected and analysed. Histograms were constructed using Excel to visualise comparisons between drug groups, providing a deeper understanding of the epidemiology and robustness of the mechanisms.

Results and discussion: The study identified that bacterial resistance is driven by genetic mutations, efflux pumps, and biofilm formation mechanisms, with *A. baumannii* showing high resistance to imipenem (78.2%) and *P. aeruginosa* to trimethoprim-sulfamethoxazole (100.0%). Ukrainian isolates demonstrated higher resistance levels to fluoroquinolones, aminoglycosides, and carbapenems compared to global averages, indicating a significant prevalence of resistant strains.

Conclusions: *A. baumannii* and *P. aeruginosa* showed lower resistance to ampicillin-sulbactam, with significant regional differences highlighting the need for targeted antimicrobial strategies in Ukraine.

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## 1. INTRODUCTION

In 2019, antimicrobial resistance directly caused an estimated 1.27 million deaths worldwide.[1] *Acinetobacter baumannii* and *Pseudomonas aeruginosa* are known to be extremely resistant to antibacterial drugs in hospital settings. To combat antimicrobial resistance, in 2019, the Cabinet of Ministers of Ukraine approved a national action plan that implements control over the supply of antimicrobials to the public and epidemiological surveillance of antimicrobial resistance.[2,3] However, the excessive and unregulated use of antibiotics in healthcare facilities, and subsequently the ongoing war in Ukraine, has led to the emergence of new multidrug-resistant (MDR) strains of microorganisms.[4,5]

Inadequate medical care for the wounded in the combat zone is due to the extreme conditions of the conflict.[6] The use of broad-spectrum antibiotics during transportation saves the lives of the wounded but also contributes to the complete resistance of microorganisms to further treatment. In addition, the rapid spread of resistant strains of wound infections due to forced migration poses a threat not only to Europe but also to the whole world.[7] The situation is complicated by the fact that wound infections caused by multidrug-resistant bacteria are difficult to treat with standard therapeutic regimens, which increases the risk of complications and death. A study and comparison of the mechanisms and prevalence of resistant MDR bacteria such as *A. baumannii* and *P. aeruginosa* to different antimicrobial drugs will help identify new targets for therapy and develop more effective treatment strategies.

*A. baumannii* and *P. aeruginosa* are dangerous for immunocompromised people and patients with multiorgan diseases in the intensive care unit.[8,9] Among 25 carbapenem-resistant isolates of *A. baumannii* and *P. aeruginosa*, the study of Kazemian et al.[10] showed that 84% of *A. baumannii* isolates contained the *blaOXA-23* gene, 68% – *blaOXA-58*, and 44% – *blaOXA-48*. The most abundant gene in *P. aeruginosa* was *blaVIM* (44%), while *bla IMP*, *blaNDM*, and *blaOXA-23* were present in 36%, 24%, and 12% of isolates, respectively. The combination of meropenem and tigecycline was effective against 80% of *A. baumannii* isolates and 20% of *P. aeruginosa* isolates. The combination of tigecycline and amikacin was effective against 40% of *A. baumannii* isolates. A significant synergistic effect was shown by the combination of colistin and meropenem against 84% of *A. baumannii* isolates and 68% of *P. aeruginosa* isolates.

Mc Gann et al.[11] inoculated 3 different MDR high-risk *P. aeruginosa* strains *ST773*, *ST 357*, and *ST1047* containing carbapenemases, 16S methyltransferase and extended-spectrum β-lactamases from biomaterials of a Ukrainian wounded. The isolate *A. baumannii ST78* obtained from a patient had 18 MDR genes, including carbapenemase. A systematic review of publications on resistance of *A. baumannii* and *P. aeruginosa* resistance in urinary tract infections in 2010–2021 was conducted by Requena-Cabello et al.[12] showed that *P. aeruginosa* is susceptible to cefepime, meropenem, amikacin, tobramycin, ceftazidime, piperacillum-tazobactam and colistin. *A. baumannii* was found to be susceptible to colistin, sulbactam, minocycline, and doxycycline.

Of the 85 *A. baumannii* isolates collected from burn wounds and hospital environments, all were resistant to imipenem and had *blaOXA-51* genes. All isolates had *blaOXA-23* and *blaOXA-24/40*, indicating the prevalence of these genes as the main mechanism of resistance to carbapenems. A study by Bakhshi et al.[13] demonstrated the contamination of hospital surfaces with MDR and extensively drug-resistant (XDR) strains of *A. baumannii*, which may increase the risk of colonisation in hospital patients. Comparing 10 drug-susceptible *A. baumannii* strains and 10 MDR strains isolated from a hospital in China, Dong et al.[14] found that MDR strains differed significantly from susceptible strains in the presence of antibiotic resistance genes, plasmids, virulence factors, and the CRISPR-Cas. system. MDR genome sequences can coincide with plasmids of various bacterial genera, which is not typical for susceptible strains. MDRs contain fewer CRISPR genes but more profiles and higher levels of spacer sequences.

A study conducted by Zwittink et al.,[15] identified 58 patients from Ukraine with 75 MDR isolates, most of which originated from globally prevalent epidemic lines. Genomic analysis revealed that 60% of the isolates contained the New Delhi metallo-β-lactamase (MBL) genes. More than 50% of *A. baumannii* strains were resistant to carbapenems, fluoroquinolones, and aminoglycosides. All isolates of *A. baumannii* belonged to yet-to-be-determined sequence types. Resistance of 17%–84% to cephalosporins and carbapenems was observed among *P. aeruginosa*. Five isolates of *ST1047* and 4 isolates of *ST773* were isolated, of which 60% contained the *blaNDM-1* carbapenemase gene. The mechanisms of genetic resistance of MDR *A. baumannii* and *P. aeruginosa*, the prevalence of new strains, and the effectiveness of specific antimicrobial agents against these resistant pathogens in Ukraine and other regions remain a pressing issue.

## 2. AIM

The study consisted of a comparative analysis of the mechanisms and prevalence of MDR in the gram-negative microorganisms *A. baumannii* and *P. aeruginosa*. This study aims to examine the antibiotic resistance mechanisms of *A. baumannii* and *P. aeruginosa*, concentrating on β-lactamases, mutation-induced resistance, efflux pumps, and biofilm formation, while evaluating regional resistance disparities and identifying the most resistant strains along with their genetic determinants.

## 3. MATERIAL AND METHODS

To study antimicrobial resistance in the bacteria *A. baumannii* and *P. aeruginosa*, scientific articles and reviews in various scientific databases, such as PubMed, NCBI, Google

Scholar, and Web of Science, were analysed. The search was conducted in English and Ukrainian using the following keywords: 'mechanism of insensitivity,' 'antimicrobial drugs,''gram-negative microorganisms", 'multi-resistance", 'epidemiology,' '*Acinetobacter baumannii*,' '*Pseudomonas aeruginosa*,' 'antimicrobial resistance,' 'mechanisms of resistance,' 'multidrug-resistant bacteria,' 'Ukraine". The search included articles containing data on the mechanisms and prevalence of antimicrobial resistance in these bacteria. Studies conducted in Ukraine or using Ukrainian biomaterials, as well as articles published in the last 5 years, were prioritised. The collected scientific data were analysed to identify and compare the main mechanisms of antimicrobial resistance and epidemiological characteristics of *A. baumannii* and *P. aeruginosa*.

The mechanisms of resistance were classified and described based on typical categories, such as the classification of β-lactamases, basic concepts of resistance in *A. baumannii* and *P. aeruginosa*, reduction of cell membrane permeability, resistance caused by mutations, expression of efflux pumps, ability to form biofilms, resistance to certain antibiotics, resistance level of individual clones, and gene transfer. Data was collected on the level of resistance to antimicrobial agents of *A. baumannii* and *P. aeruginosa* both in Ukraine and in other countries.

The data was systematised and entered Microsoft Excel spreadsheet software for further analysis. To compare the severity of resistance to different groups or individual antimicrobials, an average value was obtained from the collected data. The average function in Microsoft Excel was used to calculate the arithmetic mean of a given range of numbers. Function (1):

$$=\text{AVERAGE(range of resistance cells)}, \quad (1)$$

where: AVERAGE – Excel function for calculating the average value in percentage, the range of cells is a range (for instance, B2:B4) that contains the data for which to calculate the average value.

Based on the data obtained, histograms were created in Microsoft Excel, which demonstrates the comparison of the level of resistance of *A. baumannii* and *P. aeruginosa* to antibacterial agents in the world and Ukraine. After that, conclusions were made about the degree of resistance of these bacteria. With the information gathered, Microsoft Excel was used to make histograms that show how resistant *A. baumannii* and *P. aeruginosa* are to antibacterial agents around the world and in Ukraine. The analysis included the identification of the most common mechanisms, the frequency of their occurrence in different studies, and an assessment of their clinical significance. The proposed approach was used to systematise the available data on the mechanisms of resistance in *A. baumannii* and *P. aeruginosa*, which contributed to a deeper understanding of the problem of multidrug resistance and the identification of ways to overcome it.

## 4. RESULTS AND DISCUSSION

### 4.1. Resistance of *A. baumannii* and *P. aeruginosa* in different regions

The prevalence and degree of resistance of *A. baumannii* and *P. aeruginosa* to antibiotics can vary from country to country, influenced by environmental factors, genetic diversity from migration, infection control policies, and other circumstances. *P. aeruginosa* isolates from Greece, Spain and Italy show high antibiotic resistance, especially in Greece, where the prevalence of MDR, XDR and PDR isolates is the highest (88.9%), with colistin being the most effective antibiotic, and resistance to new drug combinations was associated with the presence of *blaVIM-2*.[16] Motbainor et al.[8] determined that nosocomial MDR *A. baumannii* and *P. aeruginosa* were in 8.4% of patients among 238 patients. 3.8% of patients had *A. baumannii*, and 4.6% had *P. aeruginosa*. All isolates were 100% MDR. The extent of resistance of *A. baumannii* and *P. aeruginosa* to certain antimicrobial agents, including data from Europe, the USA, Africa, and West and East Asia, is presented in Table 1.

The bacterium *P. aeruginosa* was highly resistant to trimethoprim-sulfamethoxazole (100%), ceftriaxone (88.9%), amoxicillin-clavulanic acid (84.2%), and gentamicin (69.1%). The results indicate considerable diversity in the resistance profiles of *A. baumannii* and *P. aeruginosa* across various antimicrobial drugs and geographical areas. *A. baumannii* has significant resistance to cefotaxime and ampicillin, hence limiting treatment alternatives due to the restricted effectiveness of combination therapy. Correspondingly, concerning resistance rates in *P. aeruginosa* to antibiotics such as trimethoprim-sulfamethoxazole and ceftriaxone, significant therapeutic constraints are underscored. These trends highlight the necessity for region-specific antibiotic stewardship, ongoing surveillance, and the advancement of alternative medicines, shaped by local prescription habits and genetic determinants. A multidisciplinary strategy that combines microbiological research, clinical protocols, and public health measures is crucial to address the worldwide issue of antibiotic resistance.

### 4.2. The level of resistance of *A. baumannii* and *P. aeruginosa* in Ukraine

The available data on the resistance of *A. baumannii* and *P. aeruginosa* resistance in Ukraine over the last 5 years are shown in Table 2.

The study revealed that colistin, eravacycline, and omadacycline were sensitive to both bacteria. In some studies, *A. baumannii* and *P. aeruginosa* were susceptible to cefiderocol.[11] It is noticeable that both bacteria in question show a high level of resistance to many classes of antibiotics. According to international definitions, these pathogens can be classified as MDRs, as they demonstrate resistance to antibiotics from different classes, such as aminoglycosides, carbapenems, and fluoroquinolones. Some strains of both types can also be considered XDRs, as they are resistant to most but one or two classes of antibiotics. *P. aeruginosa* does not show

**Table 1. Resistance to groups, combinations and individual antimicrobial agents of *A. baumannii* and *P. aeruginosa* in different regions.**

| Antibiotic | *A. baumannii* (%) | *P. aeruginosa* (%) |
|---|---|---|
| Meropenem | 80, 50, 36.4, 22 (47.4)* | 33, 3, 19.3 (18.4) |
| Ciprofloxacin | 70, 44.5, 55.5, 54 (56) | 66.7, 95.3, 38.7, 45.5 (61.55) |
| Imipenem | 91, 79.6, 64.1 (78.2) | 100, 3, 22.4 (41.8) |
| Ceftazidime | 100, 34 (67) | 33.1, 63.6 (48.35) |
| Gentamicin | 90, 37, 4.4 (43.8) | 93.4, 44.8 (69.1) |
| Doxycycline | 4.7 | N/A |
| Ampicillin sulbactam | 7, 5.7 (6.35) | 20 |
| Trimethoprim-sulfamethoxazole | 70 | 100 |
| Aztreonam | N/A | 77.7, 21.5 (49.6) |
| Amoxicillin-clavulanic acid | 8 | 84.2 |
| Tetracycline | 7, 77.8 (42.4) | 8.5, 90.6 (49.55) |
| Ceftriaxone | 82 | 88.9 |
| Cefotaxime | 98 | 88.9, 100 (94.45) |
| Tobramycin | 55, 21.1, 4 (26.7) | 33.3, 100, 47 (60.1) |
| Amikacin | 90, 33.3, 1 (41.4) | 44, 11.7, 27 (27.57) |
| Cefepime | N/A | 53.7, 26.9, 4.8 (28.47) |
| Ampicillin | 92 | N/A |
| Levomycetin | N/A | 8 |
| Cefiderocol | N/A | 1.4 |
| Ceftazidime-avibactam | N/A | 24, 5.5 (Europe), 3 (USA) (10.83) |
| Ceftolozan-tazobactam | N/A | 22.6, 38–32, 6.7 (26.43) |
| Imipenem-relebactam | N/A | 2.7 |
| Imipenem-cilastatin-relebactam | N/A | 5.7 |
| Piperacillin-tazobactam | N/A | 39.8, 38 (38.9) |

Comments: * – average values. Compiled by the authors based on available sources.[8,16–29]

**Table 2. Resistance to groups, combinations, and individual antimicrobial agents of *A. baumannii* and *P. aeruginosa* from isolated biomaterials from Ukraine.**

| Antibiotic | *A. baumannii* (%) | *P. aeruginosa* (%) |
|---|---|---|
| Fluoroquinolones (ciprofloxacin/levofloxacin) | 95 | 81.4 |
| Aminoglycosides (gentamicin/tobramycin) | 83, 67.3 (75.1) | 82.1, 58.3 (70.2) |
| Carbapenems (imipenem/meropenem) | 73, 52, 67.9; 73 (66.4)* | 71.3, 46-87.5, 63, 67.9, 55.6, 58.3, 57, 78 (64.9) |
| Cefiderocol | 24/sensitive | 24; 38 (31)/sensitive |
| Ceftazidime-tazobactam | N/A | 48 |
| Ciprofloxacin | 98.1 | 79.2 |
| Imipenem-relebactam | N/A | 84 |
| Meropenem vaborbaktam | N/A | 80 |
| Piperacillin-tazobactam | N/A | 75.5 |
| Ceftazidime-avibactam | N/A | 80 |
| Ceftazidime | N/A | 81.4 |
| Colistin | Sensitive | Sensitive |
| Eravacycline | Sensitive | Sensitive |
| Omadacycline | Sensitive | Sensitive |

Note: * – average values. Compiled by the authors based on available sources.[5,11,30–33]

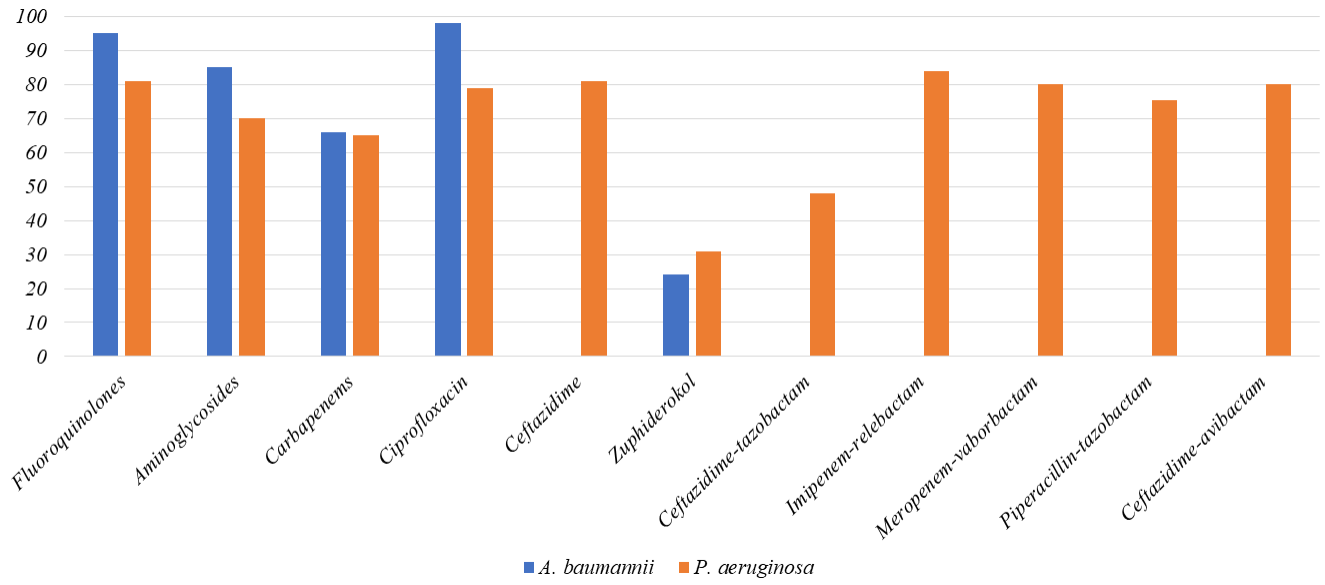


**Figure 1. Histogram comparing the mean values of resistance of *A. baumannii* and *P. aeruginosa* to antimicrobial agents in Ukraine.**

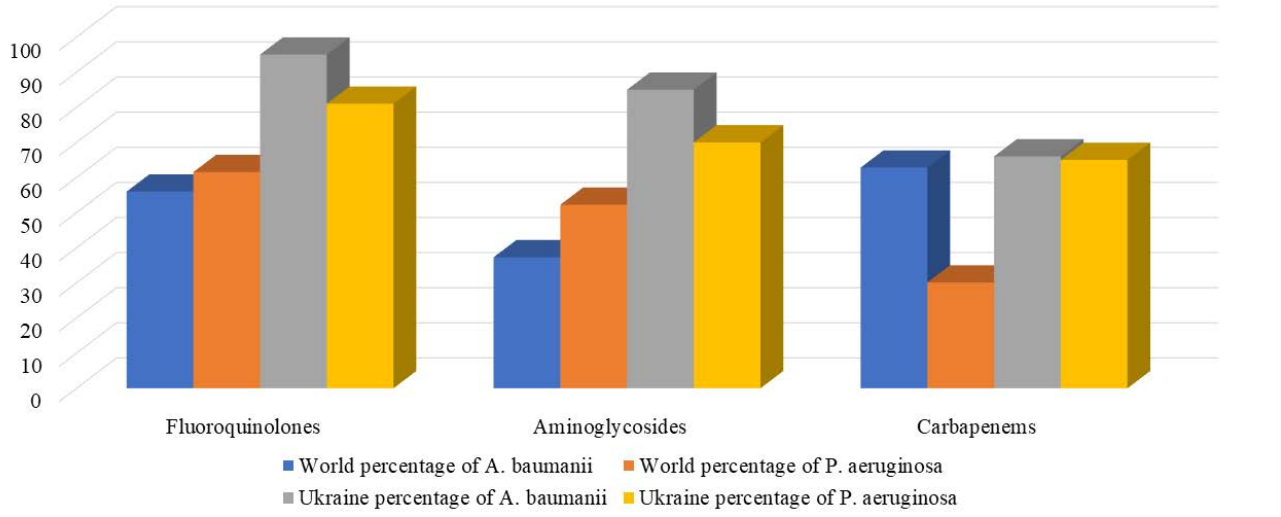


**Figure 2. Histogram comparing the mean values of resistance of *A. baumannii*, and *P. aeruginosa* to fluoroquinolones, aminoglycosides and carbapenems in Ukraine and worldwide.**

PDR as it remains susceptible to some antibiotics, such as colistin, eravacycline, and omadacycline. The comparative analysis of antimicrobial resistance in *A. baumannii* and *P. aeruginosa* demonstrates notable discrepancies in resistance patterns among several antibiotic classes, as seen in Figure 1.

As can be seen from Figure 1, data on the resistance of *A. baumannii* resistance in Ukraine is lacking. The bacterium was found to be highly resistant to ciprofloxacin (98%) and fluoroquinolones (95%), moderately resistant to aminoglycosides (85%), and less resistant to carbapenems (66%) and cefiderocol (24%). *Pseudomonas aeruginosa* showed marked resistance to a wide range of antibiotics, including ciprofloxacin (79%), fluoroquinolones (81%), aminoglycosides (70%), and carbapenems (65%). Other studies have confirmed that carbapenems are ineffective against *A. baumannii*.[34] Combination therapy can still be effective, but with different success rates, as shown in Figure 1. By studying hospital-acquired strains of *P. aeruginosa* and *A. baumannii* from 2017 to 2022 in Wroclaw, Mączyńska et al.[35] determined the highest levels of resistance to cephalosporins, carbapenems, aminoglycosides, and quinolones in 2018. For *A. baumannii*, resistance to these antibiotics has shown a significant increase in recent years.

Following Salmanov et al.[30] and the World Health Organization,[36] among non-enzymatic Gram-negative bacteria in Ukraine, in particular *A. baumannii*, carbapenem resistance was 71.3%–77.0%. At the same time, Denysko et al.[37] determined the MDR of *A. baumannii* isolates since the beginning of the full-scale war in Ukraine at 75.0%. The frequency of detection of MDR isolates was higher in wound samples (71.8%) compared to urine (9.4%), blood (7.8%), and sputum (3.2%) samples, as detected by Mirzaei et al.[38]

*Acinetobacter baumannii* and *P. aeruginosa* showed significantly higher resistance rates in Ukraine compared to the global average, especially for fluoroquinolones and aminoglycosides (Figure 2). These data indicate a potentially higher prevalence of resistant strains in Ukraine. The results of the comparison highlight an alarming trend of higher antibiotic resistance in Ukraine for both *A. baumannii* and *P. aeruginosa* in all three classes of antibiotics, potentially related to martial law in the country and wound infection. The study highlights the need for increased caution and effective infection prevention measures to control the spread of MDR in the context of increased migration and medical evacuation of patients from Ukraine.

The disparities in antibiotic resistance trends between Ukraine and other locations arise from a complex interaction of healthcare practices, antimicrobial stewardship regulations, socioeconomic situations, and geopolitical considerations. The protracted conflict in Ukraine has resulted in a rise in serious wound infections and extended hospitalisations, facilitating the proliferation of MDR microorganisms. The extensive application of broad-spectrum antibiotics in the absence of appropriate diagnostics exacerbates resistance.[39,40] Unlike regions with stringent antibiotic laws, Ukraine has a history of non-prescription antibiotic sales and empirical prescribing methods, leading to improper usage and heightened resistance. Self-medication is prevalent, exerting selection pressure on bacterial populations.[41,42] The genetic variability of *A. baumannii* and *P. aeruginosa* isolates in Ukraine may explain the observed differences in resistance. High-risk clones possessing carbapenemase genes are more frequent, indicating enhanced horizontal gene transfer throughout Ukrainian healthcare environments.

Infection control strategies in hospitals substantially affect resistance patterns. More strict regulations in other nations effectively contain MDR infections. However, resource constraints in Ukrainian facilities impede efficient infection management, hence promoting the dissemination of resistant microorganisms. Environmental factors also play a role. Antibiotics present in wastewater and agricultural runoff might facilitate the development of resistance. In contrast to Western Europe, where restrictions limit agricultural antibiotic usage, Ukraine's ongoing antibiotic residues may promote the spread of MDR strains. International migration and medical evacuations can bring resistant strains into new healthcare facilities, complicating global resistance patterns as Ukrainian patients acquire MDR diseases abroad and potentially reintroduce them upon their return.

## 5. CONCLUSIONS

(1) *Acinetobacter baumannii* and *P. aeruginosa* demonstrate significant adaptability and antibiotic resistance via several mechanisms, including β-lactamase synthesis, efflux pumps, target site alterations, and biofilm development, hence complicating treatment approaches.

(2) Resistance development is driven by genetic changes and horizontal gene transfer, involving critical resistance genes (*blaVIM*, *blaIMP*, *OXA-23*, *NDM-1*) and efflux pump systems (*AdeABC*, *MexAB-OprM*) that diminish antibiotic effectiveness. Regional disparities in resistance indicate that environmental, genetic, and healthcare-related variables affect the worldwide dissemination of MDR isolates. *Acinetobacter baumannii* had the greatest resistance to cefotaxime (98%) and ampicillin (92%), whereas *P. aeruginosa* demonstrated the highest resistance to trimethoprim-sulfamethoxazole (100%) and ceftriaxone (88.9%). Ukrainian isolates showed significant resistance to fluoroquinolones and aminoglycosides while maintaining susceptibility to colistin, eravacycline, and omadacycline.

(3) The extensive resistance of *A. baumannii* and *P. aeruginosa* in Europe, especially to cefotaxime and ciprofloxacin, requires combination treatment. In Ukraine, resistance rates are significantly elevated, particularly against fluoroquinolones and aminoglycosides, largely attributable to wartime circumstances and the high incidence of wound infections.

(4) Variability in data sources and methodologies may affect resistance evaluations, confounding direct worldwide comparisons. An extensive, interdisciplinary approach that combines microbiological, genetic, clinical, and epidemiological research is crucial for comprehending resistance processes and formulating effective control strategies.

## Conflict of interest

The authors confirm that they have no conflicts of interest to disclose.

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