The Bioscan AN INTERNATIONAL QUARTERLY JOURNAL OF LIFE SCIENCES 21(2): S.I (2), 1140-1145,2026 www.thebioscan.com

A Comprehensive Review on Mechanism, Clinical Impact, Alternative Treatment and Challenges in drug Development of Multidrug Resistance Bacteria

Nikita Sawant, Sanika Pawar, Dipali Nikalje, Shivani Mahadik, Sakshi Shinde, Amol Jadhav*

Department of Microbiology, Yashavantrao Chavan Institute of Science, Satara, Maharashtra, India 415 001

*Corresponding author: Amol Jadhav (Email: amolsjadhav12@gmail.com)

DOI: https://doi.org/10.63001/tbs.2026.v21.i02.S.I(2).pp1140-1145

KEYWORDS

Multidrug-resistant (MDR) bacteria,
Efflux pump,
Antibiotic,
MDR Pathogen.

Received on:
28-04-2026

Accepted on:
16-05-2026

Published on:
26-05-2026

Abstract

Antibiotic resistant bacteria and resistance genes have become more prevalent in the environment due to unchecked use, despite the fact that antibiotics are widely used to prevent and treat infections in both humans and animals. Multidrug-resistant (MDR) bacteria can survive in harsh environments and are found in reservoirs created by wastewater and untreated sewage from hospitals, industries, and agricultural operations. Drinking water, soil, wastewater, groundwater, and surface water have all been found to contain resistant bacteria. *Mycobacterium tuberculosis, Enterococcus faecium, Klebsiella pneumoniae, Staphylococcus aureus, Acinetobacter baumannii*, and *Pseudomonas aeruginosa* are MDR pathogens that are significant worldwide. *Salmonella, Vibrio cholerae*, and *Shigella* are examples of MDR enteric pathogens that present additional risks to public health in developing nations. Because of the production of carbapenemase, resistance to carbapenems, which are frequently the last line of treatment for MDR Gram-negative bacteria like *K. pneumoniae* and *E. coli*, has become especially concerning. MDR bacteria use a variety of strategies, such as efflux pumps, antibiotic degradation or modification, target site mutation, decreased membrane permeability, biofilm formation, and horizontal gene transfer through conjugation, transduction, and transformation. Hospital-acquired resistance has gotten worse due to the spread of β-lactamases, ESBLs, and carbapenemases via plasmids. According to clinical research, infections brought on by MDR organisms are associated with longer hospital stays, higher treatment expenses, and higher mortality rates, especially in intensive care units and patients with compromised immune systems. Synergistic antibiotic combinations, efflux pump inhibitors, engineered endolysins (artilysins), and tactics aimed at outer membrane permeability are among the novel approaches being investigated to treat MDR infections. Control efforts are made more difficult by the spread of resistance genes via clonal outbreaks and mobile genetic elements. MDR bacteria continue to be a significant global clinical and public health concern because no single antibiotic is effective against all pathogens.

Introduction:

Antibiotics are widely used to prevent or treat microbial infections in humans and animals; however, uncontrolled and excessive antibiotic use by humans and animals leads to an increase in antibiotic resistance and the spread of resistance genes in environmental samples [1]. This method has the potential to create a reservoir of multidrug-resistant microbes in water

bodies. These organisms may have transmissible genes and can survive in harsh environmental conditions for extended periods [2]. Antibiotic-resistant species have been reported in a variety of environments, including surface waters, treated wastewater, groundwater, sediments, soils, and drinking water [3,4]. Untreated sewage from municipal services, hospitals, industries, agriculture, and veterinary operations is the primary source of contamination [4, 5].

This is especially true for developing countries. Global examples of MDR strains include *Mycobacterium tuberculosis, Enterococcus faecium, Enterobacter cloacae*, *Klebsiella Pneumoniae, Staphylococcus aureus, Acinetobacter baumanii* and *Pseudomonas aeruginosa*. In developing countries, MDR enteric disease agents like *Salmonella Enteritidis*, *Shigella flexneri*, and *Vibrio cholera* pose a threat to public health measures [6,7]. In the US and UK, 40-60% of nosocomial *S. aureus* strains are methicillin-resistant (MRSA) or multidrug-resistant (MDR). Bacteria such as *Bacillus, Staphylococcus*, and *Streptococcus* species, *P. aeruginosa* and *Candida albicans* have been reported to be resistant to commonly used antibiotics, resulting in outbreaks of several infections [8].

Bacteria, including *E. coli* and *K. pneumoniae*, can develop resistance to various antibiotics. Carbapenem antibiotics are now recommended as the last line of treatment for MDR strains of *K. pneumoniae* and *E. coli* infections [9,10]. However, resistance to carbapenems has complicated treatment and resulted in untreatable infections [11]. Enterobacteriaceae's resistance to carbapenems is primarily caused by the production of carbapenem-hydrolysing enzymes (carbapenemases), particularly in *K. pneumoniae* [12,13].

Drug-resistant strains first appeared in hospitals, where antibiotics were commonly used. Sulfonamide-resistant *Streptococcus pyogenes* first appeared in military hospitals in the 1930s. Penicillin-resistant. *S. aureus* emerged in London civilian hospitals shortly after the introduction of penicillin in 1940. *M. tuberculosis* developed resistance to streptomycin shortly after its discovery [14,15].

The pathogens, known as "superbugs," include *Enterococcus faecium*, *S. aureus, K.pneumoniae, A.baumannii, P.aeruginosa,* and *Enterobacter spp.* (ESKAPE). *Enterobacteriaceae*, a family of Gram-negative bacteria, produce carbapenemase and are resistant to most antibiotics.

Enterobacteriaceae, including *E. coli* and *K. pneumoniae*, can develop resistance to various antibiotics. Carbapenem antibiotics are now recommended as the last line of treatment for MDR *K. pneumoniae* and *E. coli* infections. Antimicrobials are ineffective against certain pathogens due to resistance mechanisms like drug inactivation, modification of binding sites/targets, changes in cell permeability, and mutation [9,10, 16-20].

Mechanism of Multidrug Resistance Bacteria:

Efflux pumps, unlike other resistance determinants, are typically intrinsic. Genes for these transporters are found in both susceptible and resistant bacteria, and these genes are typically part of an operon with transcriptional regulation. Mutations in regulatory proteins or promoters lead to overexpression of efflux pumps, resulting in drug resistance [23]. The bacterial efflux system can be either specific, extruding only one class, or a single class. TetA and AbaF are antibiotics that selectively exclude specific antibiotics like tetracycline and fosfomycin, respectively [24].

MDR efflux pumps are capable of releasing multiple antibiotic classes, including MexAB-OprM, NorA, and BmrA, as well as disinfectants, dyes, and detergents. Most MDR efflux pumps, such as NorA, NorB, MepA, and MdeA in *S. aureus*, are chromosomally encoded and contribute to bacteria's intrinsic resistance to antibiotics. Some pumps are encoded on plasmids (QacA/B of *S. aureus*) or transposons (MefA and MefB of *Streptococcus* spp.), allowing for transferable resistance [25,26]. Exposure to a single agent from a pump's substrate profile can cause over-expression and cross-resistance to other substrates. These could include clinically relevant antibiotics. *P. aeruginosa* mutants that over-produce MexAB are resistant to various antibiotics, including fluoroquinolones, β-lactams, chloramphenicol, and trimethoprim, as well as triclosan, a common household biocide [27]. Recently, the efflux pump inhibitors have been studied to enhance the efficacy of exported antibiotics. Inhibitors that reduce efflux pumps' impact on fluoroquinolone activity have been developed using this strategy [28].

Horizontal gene transfer (HGT) enables microbial species to acquire genetic material from outside their clonal lineages. Microbes can use HGT to sample and share a large gene pool, which may encode traits useful in their local environment [29]. When bacteria are

exposed to strong selective pressures, such as the presence of antimicrobials, horizontal acquisition of antibiotic resistance genes (ARG) allows for genome diversification and the possibility of rapid fitness increases. Indeed, HGT can be faster than spontaneous mutations in providing genes required for survival [30].

Three major HGT mechanisms in bacteria, conjugation, transduction, and natural transformation, may contribute to the spread of MDR in clinical settings. Conjugation necessitates physical contact between two cells in the same environment, followed by the formation of a bridge that allows plasmid transfer from a donor to a recipient cell [31]. β-Lactam resistance genes are often found on plasmids and spread through inter- and intraspecies conjugation in *Enterobacteriaceae, Pseudomonas*, and *Acinetobacter*. Plasmid-mediated resistance to β-lactam antibiotics highlights how HGT worsens AR challenges in hospitals. ESBLs and carbapenemases are enzymes that break down β-lactam antibiotics like penicillin, carbapenems, and cephalosporins, leading to resistance [32-35].

Transduction occurs when viral particles transmit bacterial genes. Bacterial DNA may be accidentally packaged in a bacteriophage capsid following bacteriophage infection. A capsid containing bacterial DNA can fully bind to a recipient cell and inject foreign DNA. Transduction occurs when bacterial DNA is transferred and recombined into the recipient cell's genome [36]. Transduction in Gram-negative bacteria has been observed to transfer multiple ARGs (Antibiotic Resistance Genes), including ESBL genes, from *Pseudomonas* strains [37].

Natural transformation happens when naturally competent bacteria take up extracellular DNA and recombine an imported gene(s) into the host genome. *Acinetobacter, Haemophilus, Neisseria, Pseudomonas, Staphylococcus,* and *Streptococcus* are among the clinically relevant antibiotic-resistant pathogens that can take up DNA and transform naturally [38,39].

Antimicrobial resistance in bacteria is caused by the alteration of target sites. Most bacterial groups of clinical origin have several mechanisms of resistance, including *A. baumannii* [40]. Target site changes are generally caused by mutational processes in the bacterial genome; these mutations happen spontaneously in the genes coding for the target sites and frequently occur due to selective pressure phenomena in the presence of the antimicrobial [41,42].

Mutations in the RNA polymerase and DNA gyrase genes are the most common changes, making the bacterium resistant to rifampicin and quinolones, respectively [43].

Resistance mechanisms mediated by a change in the target site can be divided into four major groups. The first mechanism of antimicrobial inactivation is mediated by modifying enzymes, which are responsible for irreversibly destroying the antimicrobial target site [40]. The second group refers to the modification of the target site structure that decreases its affinity for the antimicrobial, preventing binding at the cell surface; the typical mechanisms are Lipopolysacchride modification, Protein binding peptide2 protein modification, and peptidoglycan modification [44-46].

The third category includes the phenomenon in which bacteria prevent antimicrobials from accumulating inside the cell; this is due to the mutation or complete loss of the outer membrane channels, the CarO type in *A. baumannii*, as well as the efflux systems of RND, MFS, ABC, and others. The fourth and final group discusses the persistence process and biofilm formation. In the first, the bacteria become tolerant of the presence of the antimicrobial when phenotypic changes, such as a quiescent metabolism, occur in small subpopulations [40].

Bacteria can prevent antibacterial molecules from accumulating on their targets by either reducing absorption, increasing discharge, or using both mechanisms simultaneously. Antibiotics typically penetrate bacteria's outer membrane to reach their targets. Gram-negative bacteria's outer membrane consists of lipid bilayers and porins [47]. Hydrophobic antibiotics (quinolones and macrolides) penetrate the lipid bilayer, while hydrophilic antibiotics (beta-lactams) pass through porins. [47,48]

Porins are selective molecules for the transfer of molecules from the outside to the inside of the cells, limiting the entry of certain antimicrobials when a mutation occurs in the porin protein. Bacteria can reduce permeability in their membranes to prevent antimicrobials from entering the cells when the chemical nature of the bacterial membranes intervenes, such as the nature of lipopolysaccharides. This mechanism reduces the concentration of antibiotics within the bacterial cytoplasm, and the minimum inhibitory concentration of antibiotics required for it to take effect is not reached [49-52].

Natural antibiotics like aminoglycosides (kanamycin, tobramycin, and amikacin) are often resistant due to

enzymatic phosphorylation by aminoglycoside phosphoryl transferase (APH), Aminoglycoside acetyltransferase (AAC), aminoglycoside adenyltransferase or nucleotidyl transferase, and β-lactams (penicillins, cephalosporins, and carbapenems like imipenem) are inactivated by β-lactamases in the periplasm. Modifications reduce the net positive charge of aminoglycosides, rendering them inactive as polycationic antibiotics [53,54]. Resistance to aminoglycosides has emerged in bacteria, including Gram-negatives, Gram-positives, and *Mycobacterium*, due to the spread of aminoglycoside-modifying enzymes (AMEs) via mobile genetic elements [55]. Certain microorganisms naturally produce beta-lactam antibiotics. Even before humans, microorganisms produced beta-lactamases to survive against antibiotics [56].

Clinical Impact:

Clinical research has consistently shown that patients with multidrug resistance organism infection have a higher risk of dying compared to those without the infection. The improper use of antibiotics during the empirical stage may be connected to this [57]. Because multidrug resistance organism appears to be more common in the ICU than other wards, patients there are more likely to contract it, which increases their risk of infection and longer hospital stays. Patients with a history of antibiotic exposure, organ transplant recipients, immunocompromised individuals, and central venous catheters [58,59].

The infections caused by ESBL-producing Enterobacteriaceae are associated with increased hospital Length of stay and costs. Bloodstream infections brought on by ESBL-producing Enterobacteriaceae are linked to higher death rates. Increased hospital Length of stay and mortality are linked to MDR *P. aeruginosa* (resistance to four or more antipseudomonal agents). Bloodstream infections and nosocomial ICU infections brought on by carbapenem-resistant isolates of *Acinetobacter spp*. are linked to higher mortality rates. Although they are linked to higher Length of stay and hospital expenses, other infection types have not been conclusively linked to higher mortality rates. MDR gram-negative bacteria have a significant and extremely concerning clinical and economic impact. A coordinated response against these pathogens might be made easier by an international agreement on the definitions of such bacteria [60].

A variety of bacteria are present in the surgical theatres' materials and

equipment. *Salmonella* species (42.8%) is the most prevalent bacterium, followed by *Escherichia coli* (28.6%), *Enterobacter sp*. (14.3%), and *Klebsiella sp*. (14.3%). Nevertheless, this outcome demonstrated the multidrug resistance among three distinct bacterial isolates, demonstrating the widespread use of plasmids containing genes that code for multidrug resistance in this field of study. Therefore, in order to prevent the accumulation of these bacteria, especially multidrug-resistant bacteria, in theatres that could result in infections that are challenging to control or curtail, the government and healthcare providers must raise awareness about resistant bacteria [17].

MDR bacteria have a moderate clinical impact on older patients with community-acquired Urinary tract infections. While there is no increase in mortality, cases brought on by MDR bacteria have longer hospital stays and higher IEAT. Nearly half of the community-acquired urinary tract infection cases in our series are caused by MDR bacteria, with prior antimicrobial therapy and living in a nursing home serving as independent risk factors [19].

Alternative treatment to control MDR infection:

To find a new substitute for colistin or carbapenem has been carried out. Despite obvious flaws, engineered endolysins (artilysins) are especially intriguing. Because of its distinct mode of action, a lytic enzyme that breaks down bacterial peptidoglycan is a promising new class of antimicrobial agents. Inhibition of peptidoglycan synthesis is a promising target for antimicrobial agents, much like β-lactam antibiotics, which are among the most effective antibiotics. The likelihood of a resistance mechanism developing is minimal since lytic enzymes directly break down peptidoglycans but not proteins. Furthermore, efflux pumps do not inhibit enzymes with comparatively high molecular weights. The enhanced artilysin may be a useful treatment option for *A. baumannii* infections that are resistant to carbapenems or colistin, if the short stability of artilysin in serum and its high production cost compared to small molecules can be addressed [40].

While last-line strategies combine colistin with rifampin or polymyxin B with tigecycline, MDR isolates are typically successfully treated with synergistic therapeutic combinations with either beta-lactamase inhibitor sulbactam or colistin [45].

Gram-negative bacteria's sensitivity to a wide range of antibiotics is influenced by the function of the general diffusion porin channels found in the outer membrane. It is obvious that any weakening of the LPS bilayer by targeting LPS-synthesising enzymes will sensitize bacteria to hydrophobic and some hydrophilic antibiotics, opening the door to combinatorial drug therapy [46].

Numerous protein channels, known as porins, are found in the outer membrane and are involved in the influx of different substances, including several classes of antibiotics. Together with efflux systems, bacterial adaptation to decrease influx through porins is a growing global issue that aids in the development and spread of antibiotic resistance. Recent advances in molecular approaches are enhancing our understanding of the physicochemical parameters that govern bacterial responses to antibiotic stress, including altered membrane permeability [50].

Combinations of curcumin and antibiotics may offer a different strategy for treating infections caused by extensively drug-resistant (XDR) and multidrug-resistant (MDR) bacteria [19].

Challenges in drug development:

In developing nations like ours, bacterial antibiotic resistance has grown to be a serious problem. This is due to a number of factors, such as widespread antibiotic use, easy access to antibiotics off the shelf, sometimes without a doctor's prescription, non-compliance—an incomplete course of antibiotic treatment, poor hygiene, which speeds up microbial growth and the spread of pathogens, poor water quality, and poor handling of human waste. As a result of all these factors, there is currently no antibiotic that works against every type of bacteria [3].

In epidemics of phenotypically identical organisms, the genes responsible for antibiotic resistance are easily transferred between cocirculating strains [32]**.** The spread of resistance to carbapenems, an antibiotic class of last resort, may result from plasmid transfer between *Enterobacteriaceae* species, making it nearly impossible to treat common infections linked to healthcare [33]. NDM enzymes are mostly found in *Enterobacteriaceae*, but they are also found in *Vibrionaceae* and non-fermenters. The blaNDM-1 gene is mostly spread through clonal outbreaks and promiscuous plasmids. Since NDM-1 bacteria are usually resistant to almost all antibiotics, accurate detection and monitoring are essential [12].

The control of antibiotic transportation through bacterial membranes, modification of the antibiotic target site, and enzymatic changes leading to antibiotic neutralization are the primary mechanisms of antimicrobial resistance [45]. mutations in DNA gyrase and RNA polymerase that cause resistance to quinolones and rifamycins. Acquisition of resistance may entail genetic exchange (conjugation, transduction, or transformation) of resistance genes from other organisms. Acquisition of the different van genes in enterococci that encode resistance to glycopeptides and the mecA genes in *Staphylococcus aureus* that encode methicillin resistance [41].

Although no efflux pump inhibitors have reached clinical practice, this area of drug development holds significant promise for reintroducing previously underutilised antimicrobial drugs [21]. Gram-negative bacteria have many efflux pumps. These pumps can transport different types of molecules, including antibiotics, out of the bacterial cell. This process reduces the amount of antibiotics inside the cell, helping bacteria survive even in high antibiotic levels. When some efflux pumps are overproduced, they can lead to significant antibiotic resistance in Gram-negative pathogens [22].

Colistin, a cationic peptide, disrupts Gram-negative bacteria's outer membrane. Colistin resistance is primarily caused by post-translational modification or loss of the lipopolysaccharide molecules inserted into the outer leaflet of the outer membrane. Lipopolysaccharide modification prevents polymyxin from binding to the bacterial surface, which may affect bacterial virulence. Antimicrobial pressure drives the evolution of antimicrobial resistance, and resistance is frequently associated with lower bacterial fitness. Therefore, alterations in lipopolysaccharide may induce changes in the fitness of *A. baumannii* [43].

Multidrug-resistant bacteria use various molecular and cellular mechanisms to resist antimicrobials. Antimicrobial resistance mechanisms include reduced drug entry into pathogen cells, enzymatic metabolism to inactive products, biofilm formation, altered drug targets, and antimicrobial target protection [49].

References:

1. Iversen, A., Kühn, I., Franklin, A., & Möllby, R. (2002). High prevalence of vancomycin-resistant enterococci in Swedish sewage. *Applied and Environmental Microbiology, 68*(6), 2838–2842.

https://doi.org/10.1128/AEM.68.6.2838-2842.2002

2. Larsson, D. G. J. (2014). Antibiotics in the environment. *Upsala Journal of Medical Sciences, 119*(2), 108–112. https://doi.org/10.3109/03009734.2014.896438
3. Nain, V. K., et al. (2015). Antibiotic resistance pattern in bacterial isolates obtained from different water samples of Delhi region. *Journal of Undergraduate Research and Innovation, 1*(3), 219–227.
4. Williams, M. R., Stedtfeld, R. D., Guo, X., & Hashsham, S. A. (2016). Antimicrobial resistance in the environment. *Water Environment Research, 88*(10), 1951–1967.
5. Food and Agriculture Organization. (2016). *Drivers, dynamics and epidemiology of antimicrobial resistance in animal production*. FAO.
6. Welch, J. T., Fricke, F. W., Patrick, F., McDermott, F. P., White, G. D., Rosso, L. M., ... & Ravel, J. (2007). Multiple antimicrobial resistance in plague: An emerging public health risk. *PLoS ONE, 2*(3), e309.
7. Gales, A. C., Jones, R. N., Turnidge, J., Rennie, R., & Ramphal, R. (2001). Characterization of *Pseudomonas aeruginosa* isolates: Occurrence rates, antimicrobial susceptibility patterns and molecular typing in the global SENTRY antimicrobial surveillance program, 1997–1999. *Clinical Infectious Diseases, 32*(2), 146–155.
8. Chitnis, V. S., Chitnis, D. M., Patil, S. L., & Kant, R. (2000). Hospital effluent: A source of multiple drug resistant bacteria. *Current Science, 79*, 989–991.
9. David, S., Reuter, S., Harris, S. R., Glasner, C., Feltwell, T., Argimon, S., ... & Grundmann, H. (2019). Epidemic of carbapenem-resistant *Klebsiella pneumoniae* in Europe is driven by nosocomial spread. *Nature Microbiology, 4*(11), 1919–1929.
10. Sharahi, J. Y., Maleki, D. T., Rad, Z. R., Rad, Z. R., Goudarzi, M., Shariati, A., et al. (2019). In vitro antibacterial activity of curcumin–meropenem combination against extensively drug-resistant bacteria isolated from burn wound infections.

11. Tavakolian, S., Goudarzi, H., & Faghihloo, E. (2020). LPS induces microRNAs-9, -192, and -205 overexpression in colorectal cell lines SW480, HCT116. *Middle East Journal of Cancer, 11*(1), 72–79.
12. Nordmann, P., Poirel, L., Walsh, T. R., & Livermore, D. M. (2011). The emerging NDM carbapenemases. *Trends in Microbiology, 19*(12), 588–595.
13. Nasiri, M. J., Mirsaeidi, M., Mousavi, S. M. J., Arshadi, M., Fardsanei, F., Deihim, B., et al. (2020). Prevalence and mechanisms of carbapenem resistance in *Klebsiella pneumoniae* and *Escherichia coli*: A systematic review and meta-analysis. *Microbial Drug Resistance, 26*(12), 1491–1502.
14. Abbasi, A. M., Mosaffa, N. D. M., Taheri, S., & Jaafari, M. K. (2007). Effect of antibacterial skin secretion of *Rana ridibanda* frog on methicillin-resistant *Staphylococcus aureus*. *Yakhteh Medical Journal, 9*, 146–155.
15. Rodrigues, O. D., Cezário, C. R., & Filho, G. P. P. (2009). Ventilator-associated pneumonia caused by multidrug-resistant *Pseudomonas aeruginosa* vs. other microorganisms: Risk factors and outcomes. *International Journal of Medicine and Medical Sciences, 1*(10), 432–437.
16. Blair, J. M., Webber, M. A., Baylay, A. J., et al. (2015). Molecular mechanisms of antibiotic resistance. *Nature Reviews Microbiology, 13*(1), 42–51. https://doi.org/10.1038/nrmicro3380
17. Oyedum, U., Olatunji, O., & Abu, H. (2023). Isolation and characterisation of multidrug-resistant bacteria from surgical equipments in general hospital. *African Journal of Health Safety and Environment, 4*(1), 59–64. https://doi.org/10.52417/ajhse.v4i1.393
18. Monegro, A. F., & Regunath, H. (2018). Hospital-acquired infections. *StatPearls*. StatPearls Publishing.
19. Madrazo, M., Esparcia, A., López-Cruz, I., Alberola, J., Piles, L., Viana, A., et al. (2021). Clinical impact of multidrug-resistant bacteria in older hospitalized patients with community-acquired urinary tract infection. *BMC*

Infectious Diseases, 21, 1232. https://doi.org/10.1186/s12879-021-06939-2

20. Centers for Disease Control and Prevention. (2018). *Carbapenemase-producing carbapenem-resistant Enterobacteriaceae (CP-CRE) case definition*. CDC.
21. Piddock, L. J. (2006). Clinically relevant chromosomally encoded multidrug resistance efflux pumps in bacteria. *Clinical Microbiology Reviews, 19*, 382–402.
22. Blair, J. M., Richmond, G. E., & Piddock, L. J. (2014). Multidrug efflux pumps in Gram-negative bacteria and their role in antibiotic resistance. *Future Microbiology, 9*, 1165–1177.
23. Webber, M. A., & Piddock, L. J. (2003). The importance of efflux pumps in bacterial antibiotic resistance. *Journal of Antimicrobial Chemotherapy, 51*, 9–11.
24. Sharma, A., Sharma, R., Bhattacharyya, T., Bhando, T., & Pathania, R. (2017). Fosfomycin resistance in *Acinetobacter baumannii* is mediated by efflux through the MFS transporter AbaF. *Journal of Antimicrobial Chemotherapy, 72*, 68–74.
25. Costa, S. S., Ntokou, E., Martins, A., Viveiros, M., Pournaras, S., Couto, I., et al. (2010). Identification of the plasmid-encoded QacA efflux pump gene in methicillin-resistant *Staphylococcus aureus*. *International Journal of Antimicrobial Agents, 36*, 557–561.
26. Santagati, M., Iannelli, F., Cascone, C., Campanile, F., Oggioni, M. R., Stefani, S., et al. (2003). TN1207.3 carries the macrolide efflux gene mef(A) in *Streptococcus pyogenes*. *Microbial Drug Resistance, 9*, 243–247.
27. Chuanchuen, R., Beinlich, K., Hoang, T. T., Becher, A., Karkhoff-Schweizer, R. R., & Schweizer, H. P. (2001). Cross-resistance between triclosan and antibiotics in *Pseudomonas aeruginosa* is mediated by multidrug efflux pumps. *Antimicrobial Agents and Chemotherapy, 45*, 428–432.
28. Lomovskaya, O., Warren, M. S., Lee, A., Galazzo, J., Fronko, R., Lee, M., et al. (2001). Identification and characterization of inhibitors of multidrug

resistance efflux pumps in *Pseudomonas aeruginosa*. *Antimicrobial Agents and Chemotherapy, 45*, 105–116.

29. Sørensen, S. J., Bailey, M., Hansen, L. H., Kroer, N., & Wuertz, S. (2005). Studying plasmid horizontal transfer in situ: A critical review. *Nature Reviews Microbiology, 3*, 700–710.
30. Charpentier, X., Polard, P., & Claverys, J.-P. (2012). Induction of competence for genetic transformation by antibiotics. *Current Opinion in Microbiology, 15*, 570–576.
31. Paterson, D. L., & Bonomo, R. A. (2005). Extended-spectrum β-lactamases: A clinical update. *Clinical Microbiology Reviews, 18*, 657–686.
32. Valenzuela, J. K., Thomas, L., Partridge, S. R., van der Reijden, T., Dijkshoorn, L., & Iredell, J. (2007). Horizontal gene transfer in a polyclonal outbreak of carbapenem-resistant *Acinetobacter baumannii*. *Journal of Clinical Microbiology, 45*, 453–460.
33. Conlan, S., Thomas, P. J., Deming, C., Park, M., Lau, A. F., Dekker, J. P., et al. (2014). Single-molecule sequencing to track plasmid diversity in carbapenemase-producing Enterobacteriaceae. *Science Translational Medicine, 6*(254), 254ra126.
34. Phan, H. T. T., Stoesser, N., Maciuca, I. E., Toma, F., Szekely, E., Flonta, M., et al. (2018). WGS to characterize an NDM-1 *Serratia marcescens* outbreak in Romania. *Journal of Antimicrobial Chemotherapy, 73*, 672–679.
35. Stanczak-Mrozek, K. I., Manne, A., Knight, G. M., Gould, K., Witney, A. A., & Lindsay, J. A. (2015). Within-host diversity of MRSA antimicrobial resistances. *Journal of Antimicrobial Chemotherapy, 70*, 2191–2198.
36. Blahová, J., Králiková, K., Krčméry, V., & Ježek, P. (2000). Transduction of imipenem resistance by bacteriophage AP-151 in *Pseudomonas aeruginosa*. *Journal of Chemotherapy, 12*, 482–486.
37. Johnston, C., Martin, B., Fichant, G., Polard, P., & Claverys, J. P. (2014). Bacterial transformation: Distribution, shared mechanisms and divergent control. *Nature Reviews Microbiology, 12*, 181–196.

38. Traglia, G. M., Chua, K., Centrón, D., Tolmasky, M. E., & Ramírez, M. S. (2014). Whole-genome sequence analysis of naturally competent *Acinetobacter baumannii* clinical isolate A118. *Genome Biology and Evolution, 6*, 2235–2239.
39. De Oliveira, D. M. P., Forde, B. M., Kidd, T. J., Harris, P. N. A., Schembri, M. A., Beatson, S. A., et al. (2020). Antimicrobial resistance in ESKAPE pathogens. *Clinical Microbiology Reviews, 33*(3), e00181-19.
40. Lee, C. R., Lee, J. H., Park, M., Park, K. S., Bae, I. K., Kim, Y. B., Cha, C. J., Jeong, B. C., & Lee, S. H. (2017). Biology of *Acinetobacter baumannii*: Pathogenesis, antibiotic resistance mechanisms, and treatment options. *Frontiers in Cellular and Infection Microbiology, 7*, 55.
41. Lambert, P. A. (2005). Bacterial resistance to antibiotics: Modified target sites. *Advanced Drug Delivery Reviews, 57*, 1471–1485.
42. Ruiz, J. (2003). Mechanisms of resistance to quinolones. *Journal of Antimicrobial Chemotherapy, 51*, 1109–1117.
43. Da Silva, G. J., & Domingues, S. (2017). Interplay between colistin resistance, virulence, and fitness in *Acinetobacter baumannii*. *Antibiotics, 6*(28).
44. Tipton, K. A., & Rather, P. N. (2019). Extraction and visualization of capsular polysaccharide from *Acinetobacter baumannii*. *Methods in Molecular Biology, 1946*, 227–231.
45. Kyriakidis, I., Vasileiou, E., Pana, Z. D., & Tragiannidis, A. (2021). *Acinetobacter baumannii* antibiotic resistance mechanisms. *Pathogens, 10*(373).
46. Delcour, A. H. (2009). Outer membrane permeability and antibiotic resistance. *Biochimica et Biophysica Acta, 1794*, 808–816.
47. Chapman, J. S., & Georgopapdakou, N. H. (1988). Routes of quinolone permeation in *Escherichia coli*. *Antimicrobial Agents and Chemotherapy, 32*, 438–442.
48. Lee, C. R., Lee, J. H., Park, M., Park, K. S., Bae, I. K., Kim, Y. B., Cha, C. J., Jeong, B. C., & Lee, S. H. (2017). Biology of *Acinetobacter baumannii*: Pathogenesis, antibiotic resistance mechanisms, and treatment

options. *Frontiers in Cellular and Infection Microbiology, 7*, 55.

49. Varela, M. F., Stephen, J., Lekshmi, M., Ojha, M., Wenzel, N., Sanford, L. M., et al. (2021). Bacterial resistance to antimicrobial agents. *Antibiotics, 10*(593).
50. Pagès, J. M., James, C. E., & Winterhalter, M. (2008). The porin and permeating antibiotic barrier in Gram-negative bacteria. *Nature Reviews Microbiology, 6*, 893–903.
51. Kobylka, J., Kuth, M. S., Müller, R. T., Geertsma, E. R., & Pos, K. M. (2020). AcrB: A drug efflux machine. *Annals of the New York Academy of Sciences, 1459*, 38–68.
52. Davies, J., & Wright, G. D. (1997). Bacterial resistance to aminoglycoside antibiotics. *Trends in Microbiology, 5*, 234–240.
53. Wright, G. D. (1999). Aminoglycoside-modifying enzymes. *Current Opinion in Microbiology, 2*, 499–503.
54. Bush, K., & Miller, G. H. (1998). Bacterial enzymatic resistance: β-lactamases and aminoglycoside-modifying enzymes. *Current Opinion in Microbiology, 1*, 509–515.
55. Bhullar, K., Waglechner, N., Pawlowski, A., et al. (2012). Antibiotic resistance in an isolated cave microbiome. *PLoS ONE, 7*, e34953.
56. Davies, C., White, S. W., & Nicholas, R. A. (2001). Crystal structure of a deacylation-defective mutant of penicillin-binding protein 5. *Journal of Biological Chemistry, 276*, 616–623.
57. Busani, S., Serafini, G., Mantovani, E., Venturelli, C., Giannella, M., Viale, P., et al. (2017). Mortality in septic shock by multidrug-resistant bacteria. *Journal of Intensive Care Medicine, 34*, 48–54.
58. McConville, T. H., Sullivan, S. B., Gomez-Simmonds, A., Whittier, S., & Uhlemann, A. C. (2017). Carbapenem-resistant Enterobacteriaceae colonization and mortality. *PLoS ONE, 12*, e0186195.
59. Ling, M. L., Tee, Y. M., Tan, S. G., Amin, I. M., How, K. B., Tan, K. Y., et al. (2015). Risk factors for acquisition of carbapenem-resistant Enterobacteriaceae. *Antimicrobial Resistance and Infection Control, 4*(26).
60. Giske, C., Monnet, D., Cars, O., & Carmeli, Y. (2008). Clinical and

economic impact of MDR gram-negative bacilli. *Antimicrobial Agents and Chemotherapy, 52*(3), 813–821.