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Characterization of AMR Protein in MDR *E. Coli* – A Review

Ms. Divya Nair[1], Yasaswini R[2], Nikitha Agnes A[2], Preethi H[2], Teena P[2]

[1]Assistant Professor and [2]Student,
Sri Shakthi Institute of Engineering and Technology, Coimbatore 641062.
Corresponding Author email: yasaswinir03@gmail.com

A B S T R A C T

This review targets studies on antimicrobial resistance in multidrug-resistant Escherichia coli, with emphasis on outer membrane proteins and efflux pumps. MDR E. coli is a worldwide health threat because of its competence in acquiring and spreading resistance genes. OMPs, which are essential for cell barrier function and nutrient acquisition, and efflux pumps, therefore, has significant roles in resistance. The β-barrel assembly machinery complex, which is critical for the biogenesis of outer membrane protein, is a good target for the formulation of new antimicrobials. This review also covers the occurrence of E. coli O157:H7 in poultry and zoonotic transmission risk and preventive hygienic measures. The typing and characterization of E. coli isolates, through biochemical, serological, and molecular techniques, are addressed, as well as storage methods and the viable but nonculturable state. Prudent use of antibiotics and the emergence of alternative treatments such as bacteriophages are highlighted against MDR E. coli.

Keywords: Antimicrobial resistance, *E. coli*, multidrug resistance, Outer membrane protein, BAM

1. Introduction

Antimicrobial resistance (AMR) by *Escherichia coli*, especially multidrug-resistant (MDR) types, is an important global health concern. Characterization of AMR proteins of MDR *E. coli* regarding mechanisms of resistance, the implications of outer membrane proteins (OMPs), and therapeutic interventions form the basis of this review. This entails sorting of data from different studies for a holistic report of the up-to-date situation regarding AMR in *E .coli*. The effectiveness of contemporary medicine is greatly dependent on the presence of antimicrobial drugs. Nevertheless, the development and dissemination of AMR among microorganisms, particularly bacteria, have emerged as a major issue (Prestinaci et al., 2015). *E. coli*, a normal resident of the gastrointestinal tract, has increasingly become resistant to various antibiotics, resulting in serious clinical problems. Zoonotic transmission has an important role to play in the spread of AMR, with antibiotic-resistant pathogens transmitted between humans and animals (Amin et al., 2020; Tang et al., 2023).

1.1 Antimicrobial Resistance and Bacterial Cell Structure

Bacteria such as *E. coli* are divided into two major groups: Gram's negative and Gram's positive, based on the nature of their cell wall; such differences do influence susceptibility to antibiotics (Doron & Gorbach, 2008). The Gram-negative *E. coli* contains an OW, a thin layer of peptidoglycan, and an inner membrane, posing a difficult-to-penetrate barrier for almost all antibiotics (Breijyeh et al., 2020; Gauba, 2023). LPS and OMPs that comprise the OW serve as crucial components in their defense against the harmful effects of antibiotics. Of these OMPs, porins function as conduits for the diffusion of small hydrophilic molecules, including some antibiotics, across the OW. Inhibition of antibiotic penetration into cells occurs through alteration in porin expression or structure (Pages et al., 2008). In addition, structural modifications of the LPS lead to decreased permeability arising from large outer membrane classics (Delcour, 2009).

Tegegne et al. (2024) offered further elucidations on the distribution and antimicrobial resistance of *E. coli* O157:H7 in chicken meat slaughtered from Addis Ababa. The emergence of antibiotic resistance in foodborne bacteria among poultry meat continues to alarm; this poses a serious public health threat, especially to developing countries with a high meat consumption of poultry (Tegegne et al., 2024). The alarming presence of multidrug-resistant *E. coli* O157:H7 in these food animals requires immediate monitoring and control to ensure food safety.

1.2 Mechanisms of Multidrug Resistance (MDR)

Different mechanisms lead to MDR in *E.coli*: genetic mutations, acquisition of resistance genes, and physiological adaptations. The said mechanisms lead to one or more of the following: alteration of drug targets, enzymatic inactivation of antibiotics, reduced drug permeability, and increased efflux (Li & Nikaido, 2004; Wright, 2005; Wilson, 2014). Mutations in DNA topoisomerases confer resistance to fluoroquinolones; various β-lactamases hydrolyze

β-lactam antibiotics (Nikaido, 2009). Efflux pumps, mostly those encoded by ABC transporter genes, export the antibiotics from the cell, contributing to MDR. Such pumps recognize a wide range of substrates, including antibiotics, and often overexpression is associated with the overall resistance against varied and nonrelated drugs (Blair et al., 2015). Also critical in the rapid dissemination of MDR among *E.coli* strains is the horizontal transfer of resistance genes of plasmids, transposons, and integrons (Carattoli, 2013). These mobile genetic elements can also carry several resistance genes, allowing for the simultaneous acquisition of different antibiotic class resistance. Finally, physiological adaptations, including biofilm formation, considerably increase bacterial survivability under antibiotic threat (Mah & O'Toole, 2001). Biofilms are protective matrices against antibiotics wherein penetration is limited, and resistance development has optimal conditions to thrive within them. The constant and indiscriminate use of antibiotics has contributed to multidrug resistance in these associated bacteria through the acquisition of antibiotic resistance1 genes. Such studies indicated high resistance to antibiotics such as ampicillin and streptomycin in *E.coli* O157:H7 isolates from chicken meat (Tegegne et al., 2024). The unrelenting use of these products in agriculture and animal husbandry favours the selection of resistant strains, which could, however, be transferred to humans in the food chain or otherwise (Marshall & Levy, 2011). For this reason, it is most important that antibiotics are used responsibly to mitigate the keyword spread of AMR and hence preserve the effectiveness of these critical drugs.

1.3 Role of Outer Membrane Proteins (OMPs) and Beta Barrel Assembly Machinery (BAM)

While Gram-negative bacteria are extremely diverse, outer membrane proteins (OMPs) have many functions in their physiology. They provide an entrance into the outer membrane as selective channels for nutrients (Pages et al., 2008). OMPs also confer integrity to the bacterial cell envelope, contributing to the activity of the protective barrier against harmful agents (Delcour, 2009). Other important functions of many OMPs are as factors for virulence because they aid the bacteria in adhering to and invading host cells and/or evading host immune responses (Henderson & Nataro, 2001). The biogenesis of OMPs involves a multitude of steps and relies on the β-barrel assembly machinery (BAM) complex, which is probably the most important. The BAM complex is anchored into the outer membrane, directing the folding and insertion of OMPs (Wu et al., 2019). The BAM complex has several essential proteins: Bam A, Bam B, Bam C, Bam D, and Bam E, which cooperate to program OMP assembly (Tomasek et al., 2021). The core is Bam A, the central component that serves as a scaffold and chaperone for guiding an OMP through the insertion process (Ruiz et al., 2006). Because of this important role, and also the fact that it is so readily accessible, targeting the BAM complex is attractive for the discovery of novel antibiotics. When BAM function is inhibited, OMP assembly is inhibited; this effect produces a constellation of phenomena, including an outer membrane that is unstable and the rupture of the outer membrane, which as a result, will lead to bacterial death (Storek et al., 2018; Xu et al., 2023). Hence, it becomes imperative first to understand the structural and functional characteristics of the BAM complex to usher antimicrobial strategies.

2. Antimicrobial Resistance

2.1 E. coli

Escherichia coli is one of the most ubiquitous bacteria, having become one of the major contributors to the global concern of increasing antimicrobial resistance (AMR) (Galindo-Méndez, 2020). Between its high adaptability and its ability to acquire and disseminate resistance genes through horizontal gene transfer mechanisms, such as conjugation, transduction, and transformation, the emergence of AMR proposes further risks (Carattoli, 2013). There is plenty of evidence of colistin-resistant *E. coli* in poultry environments, wherein food supply chains have also contributed to AMR transmission (Amin et al., 2020; Tegegne et al., 2024). This situation assumes even greater importance as colistin is usually reserved as a last-resort antibiotic for treating multidrug-resistant infections. Multidrug-resistant (MDR) *E. coli* is often responsible for bloodstream infections (BSI) in Western Europe and North America, resulting in severe morbidity and mortality (Di Franco et al., 2021). The emergence of quinolone-resistant E. coli has been implicated in spontaneous bacterial peritonitis, a severe complication of liver cirrhosis (Cereto et al., 2003). The rapid spread of resistance to quinolones, which are among the most frequently prescribed antibiotics, presents a significant clinical challenge. Studies in Addis Ababa slaughterhouses showed a prevalence of 5.2% for *E. coli* O157:H7 in chicken meat, and interestingly, the isolates were multidrug-resistant to various antibiotics, thus bringing monitoring of antibiotic use for animal and human health into question (Tegegne et al., 2024). Resistance concentrations of 47.5% to streptomycin and 40.8% to ampicillin appeared amongst *E. coli* O157:H7 isolates, but no resistances were reported against kanamycin, gentamicin, or ciprofloxacin (Tegegne et al., 2024). Henceforward, the justified utilization of antibiotics and a powerful surveillance system must go into the efforts in ascertaining the spread of AMR through food animals and humans.

2.2 Mechanisms of MDR and efflux

Efflux pumps are important contributors to multiple-drug resistance (MDR) by transporting antibiotics out of the bacterial cell, thus decreasing their intracellular concentration which reduces their efficacy (Li & Nikaido, 2004). They are of diverse origins, relating to many families, such as ABC transporters and MFS, with different structure and functionality characteristics (Blair et al., 2015). ABC transporters use energy from ATP hydrolysis, while MFS rely on proton motive force or sodium gradients for substrate transport (Putman et al., 2000). Understanding the precise details of efflux mechanism helps to develop further strategy towards combating MDR. For instance, efflux pump inhibitors can inhibit the activity of such transporters and in combination with the antibiotics, restore their efficacy (Pagès et al., 2008). Structural modifications of antibiotics can also reduce recognition by efflux pumps and enhance their intracellular accumulation (Zgurskaya & Nikaido, 2000). Enzymatic modification of antibiotics, as in acetylation, phosphorylation or hydrolysis, is another significant mechanism of resistance. Enzymes such as β-lactamases, aminoglycoside-modifying enzymes, and

chloramphenicol acetyltransferases can, therefore, inactivate antibiotics before they reach their intracellular targets (Wright, 2005). Such enzymatic modifications can happen through different biochemical reactions, resulting in structural modifications to the antibiotic molecules rendering them inactive.

2.3 Antimicrobial Resistance: A Global Threat

Antimicrobial resistance (AMR) is a significant global health threat associated with increased mortality and morbidity coupled with healthcare costs (Prestinaci et al., 2015; Tanwar et al., 2014). The emergence and spread of AMR compromise the effectiveness of antibiotics, which are essential for treating bacterial infections. To stimulate the research and development of new antibiotics, the World Health Organization (WHO) identified priority pathogens, such as multidrug-resistant (MDR) Gram-negative bacteria (Gauba, 2023). The list includes carbapenem- and third-generation cephalosporin-resistant *E.coli* strains, drawing attention to the immediate need for the discovery of new therapeutic strategies. Spread of colistin resistance through zoonotic transmission is a serious concern since colistin is frequently used as an antibiotic of last resort in treating MDR infections (Tang et al., 2023). The application of antibiotics in agriculture and animal husbandry plays a role in selecting and disseminating resistant bacteria that can, in turn, spread to humans either through the food chain or contact (Marshall & Levy, 2011). For this reason, responsible antibiotic stewardship and infection control measures are objectives for limiting the spread of AMR and ensuring the efficacy of these critical drugs for the future.

2.4 AMR proteins: The role of BAM

The β-barrel assembly machinery (BAM) complex is vital for the biogenesis of outer membrane proteins (OMPs) in Gram-negative bacteria; hence, it is a valuable site for those metabolic activities-absorption of nutrients, resistance to antimicrobials, and virulence (Xu et al., 2023). OMPs are essential components of the bacterial cell envelope, acting as selective pores that allow the transport of essential substances into the cells, while at the same time being a barrier to harmful substances (Pages et al., 2008). The BAM complex in the outer membrane executes the proper folding and insertion of OMPs to ensure their functional integrity (Wu et al., 2019). Targeting BAM, transport of Bam A is, therefore, a worthy strategy for powerful antibiotics. Bam A, as a central component of the BAM complex, is said to act as a scaffold and chaperone through OMPs as they are inserted (Tomasek et al., 2021). Disruption to Bam A function should prevent OMP assembly, inducing instabilities in the envelope of the bacteria and their eventual death (Storek et al., 2018). Because of its important function and outside reach, the BAM complex presents a potent target for the development of antimicrobials.

2.5 E. coli O157:H7 Prevalence in Poultry

Tegegne et al. (2024) explored the presence of E. coli O157:H7 in chicken meat collected from slaughterhouses in Addis Ababa, Ethiopia. The study revealed an overall prevalence of 5.2% of E. coli O157:H7. The highest isolate presence was observed in chicken carcasses, followed by fecal samples, hand swabs, and knife swabs. Poor hygienic practices of workers in the slaughterhouses pose an additional risk for possible contamination. This emphasizes the need for better training and awareness regarding food safety practices. The findings of this study further reiterate the concern of *E.coli* O157:H7 in zoonotic transmission, as contaminated poultry products would serve as a vehicle of human infection. The presence of multidrug-resistant *E.coli* O157:H7 isolates underlines other issues surrounding monitoring antibiotic use in both human and animal health. The observed high levels of resistance to streptomycin and ampicillin point toward the great possibility of spread of antibiotic resistance genes through the food chain.

2.6 Characterization and Typing of E.coli Isolates

Various biochemical, serological, and molecular methods can be used in the characterization of *E. coli* isolates, giving very useful information concerning the phenotypic and genotypic characteristics (Lupindu, 2017). Various biochemical tests, such as the IMViC tests and API 20E, confirm the isolates of *E.coli* and distinguish them from other members of the Enterobacteriaceae. Enzymatic assays based on specific enzyme activities thus express it, including β-glucuronidase productions, are performed on brilliance *E. coli* agar, which is typically characteristic of most *E. coli* strains. The isolation and characterization of *E. coli* is a very important subject for AMR development and control strategy formulation. Getting this done includes the use of selective media, biochemical tests, and molecular methods (Islam et al., 2014; Lupindu et al., 2017; Tripathi et al., 2023; Hendriksen, 2003). Selective media include MacConkey agar, usually used as a tool to differentiate *E. coli* from lactose fermenters. For *E. coli* O157:H7, sorbitol MacConkey agar is used, as that bacteria lack the ability to ferment sorbitol, thus allowing differentiation from further *E. coli* strains (Hendriksen, 2003; Lupindu, 2017). Chromogenic coliform agar (CCA) has been shown to be a reliable method for enumerating

E. coli in a variety of samples-a rapid and accurate assessment of bacterial contamination (Lange et al., 2013). Biochemical tests together with IMViC assays would further differentiate *E. coli* from the other types in Enterobacteriaceae (Lupindu, 2017). Although biochemical tests have been proved limited in resolving closely related species of the main enteropathy *E. coli*, molecular methods, like PCR, PFGE, and WGS, will give good information regarding the resistance, genetic association, and virulence elements carried by the particular E. coli strains (Tripathi et al., 2023). Bacteriophages specifically targeting pathogenic *E. coli* have been isolated and characterized for potential biocontrol applications (Lukman et al., 2020). Isolation and characterization of *E. coli* remain vital for understanding antimicrobial resistance and pathogenicity, employing various techniques, including selective media, biochemical tests, serological typing, and molecular methods (Hendriksen, 2003; Lupindu, 2017). The differentiation of *E. coli* from other lactose-fermenting bacteria is commonly done using MacConkey agar, while sorbitol MacConkey agar acts as a further diagnostics tool to confirm the pathogenic strain *E. coli* O157:H7 for its inability to ferment sorbitol (Hendriksen, 2003; Lupindu, 2017). To increase isolation chances, enrichment broths such as Muller-Hinton or nutrient broth could also be utilized. The concentration of the sample suspension, as well as the weight of the sample, can affect the probability of bacteria recovery (Lupindu, 2017). Serological typing relies on specific antibodies which are used to identify the Ag, be it O, K or H

antigens, and this allows the classification of *E. coli* strains into serotypes that are, for the most part, correlated with certain virulence properties. Molecular approaches, inclusive but not limited to PCR, PFGE, and MALDI-TOF, are used to provide more fine-scale genetic insight into *E. coli* isolates. PCR in this regard detects specific resistant genes and virulence factors, while PFGE and MALDI-TOF are strain-typing for epidemiological casework. Such methods are important in understanding the diversity and evolution of *E. coli* strains as well as for tracing the spread of antibiotic resistance and virulence genes.

2.7 Storage of E.coli Isolates

The storage of *E.coli* isolates is of utmost importance since it ensures that isolate viability and genetic integrity are kept intact for future research. Short-term storage consists of growing pure cultures on agar slants or plates of non-selective media and storing them at 4°C. This is an appropriate method for holding isolates for some days up to a few weeks (Lupindu, 2017). Screw-capped tubes are the best since they are less likely to contaminate and moisture is maintained in'em; Petri dishes should have their lids covered with parafilm to minimize desiccation and be stored up-side down for better condensation management. Long-term storage encompasses freezing at various temperatures, ranging from -20°C to -196°C. Lower temperatures are associated with longer storage times. -20°C to -40°C storage holds *E.coli* for about a year, temperatures of -80°C can keep them viable for several years, while cryopreservation below -130°C, normally with the use of liquid nitrogen, may be applicable for more than 10 years. The freezing process, however, can also inflict damage upon the bacterial cells through ice formation and the resultant development of solutes in the remaining unfrozen water. Cryoprotectants such as glycerol promise to protect the cells against freezing and, therefore, ice crystal formation, lowering the suspension freezing point and minimizing large ice crystal formation. Glycerol also works to maintain cell membrane integrity and protects against intracellular damage due to freeze-thaw cycles. Usually, this involves mixing bacterial cultures with a 15-20% glycerol solution for freezing. In order to also prevent ice crystal formation, snap freezing places the culture tubes rapidly into liquid nitrogen or dry ice. However, slow freezing can be equally efficient, in which the temperature is progressively reduced. In either method of freezing, though, severe cell death has been observed upon repeated freezing and thawing cycles.

2.8 Common E.coli Pathotypes

E.coli strains pathogenic to humans fall into two main groups, namely intestinal pathogenic *E.coli* (IPEC) and extraintestinal pathogenic *E.coli* (EXPEC). The IPEC category features a variety of distinct pathotypes: enterotoxigenic *E. coli* (ETEC), enteropathogenic *E. coli* (EPEC), enteroinvasive *E. coli* (EIEC), enteroaggregative *E. coli* (EAEC), diffusely adherent *E. coli* (DAEC), and verotoxigenic *E. coli* (VTEC), the first four of which are classified on the basis of specific virulence factors and diseases caused (Lupindu, 2017). The EXPEC bacteria, in fact, cause infections beyond the GI tract, with such manifestations as urinary tract infections, meningitis, and sepsis.

E.coli may enter a viable but nonculturable state under specific environmental stresses including nutrient limitation temperature extremes, and exposure to disinfectants (Oliver, 2010). In this state, the bacteria are metabolically active but lose the ability to form colonies on common culture media, thus underestimating bacterial viability and potential pathogenicity in environmental samples. Therefore, DNA-dependent methods like PCR are recommended for identifying the VBNC cells, which can detect bacterial DNA even if the cells are nonculturable (Lupindu, 2017).

3. Conclusion

It is critically important to characterize AMR proteins in MDR *E.coli* to ensure effective interventions to combat such a growing public menace. Factors that could yield interplay in the genetic, biochemistry, and physiology domains contributing to the emergence and spread of AMR are complex. The targeting of essential bacterial proteins, including, inter alia, those involved in cell wall synthesis, DNA replication, and protein synthesis, is a promising approach toward developing novel antimicrobial agents. Besides, inhibiting efflux pumps and disrupting bacterial signalling pathways remain possible therapeutic avenues.

Tegegne et al. (2024) further emphasize the risk posed by *E. coli* O157:H7 contamination in the poultry industry and the extent of AMR in these isolates. Detection of multidrug-resistant *E.coli* O157:H7 from chicken carcasses, as well as hand and knife swabs, reinforces the need for strict hygiene and sanitation practice in slaughterhouses. Furthermore, the poor hand-washing practices among slaughterhouse workers demonstrate a need for better training and awareness on food safety. Regulating the use of antibiotics in farm animals to minimize the selection and dissemination of resistant bacteria also stood out prominently in the study.

Lupindu (2017) provides detailed procedures for isolation, characterization, and storage of *E.coli* isolates on which research can lean greatly. The methods chosen should depend on the objective of the research and specific targets of interest that lie within *E.coli*. Moreover, it is of utmost importance for *E.coli* to be detected in different environmental and clinical samples, given that there is an awareness of this VBNC state.

MDR in *E.coli* presents a tremendous and increasing threat to global public health, warranting urgent and comprehensive strategies to mitigate its devastating effects (Prestinaci et al., 2015). The capacity of *E. coli* bacteria to obtain and disseminate the resistance genes through horizontal gene transfer, together with its versatility to inhabit diverse environments, has aided the rapid dissemination of MDR strains (Carattoli 2013). The alarming phenomenon not only undermines the efficacy of the existing antibiotic treatments but also considerably raises the risks for treatment failures, longer patient hospital stays, and increased healthcare costs (Tang et al., 2023). To come up with new alternative treatments, comprehensive characterization of the AMR proteins, that is, OMPs and efflux pumps, is fundamental (Gauba, 2023). OMPs, such as porins, play an equally essential role in antibiotic uptake and

resistance through regulation of the permeability of the bacterial cell envelope (Pages et al., 2008). The efflux pump actively exports antibiotics from the cell and thus reduces its concentration inside the cell, and therefore decreases their efficiency (Li & Nikaido, 2004). Since these proteins constitute priority targets for the development of new antimicrobial drugs and inhibitors, knowledge regarding their structure, function, and regulatory mechanisms is necessary to restore antibiotic sensitivity. Future work will be required to elaborate the intricate mechanisms of resistance, genetic and biochemical pathways responsible for the development and spread of AMR--horizontal gene transfer being another factor worth studying, as well as the involvement of mobile genetic elements such as plasmids and transposons (Carattoli, 2013). Moreover, the impact of environmental conditions on the selection and spread of MDR *E.coli* strains includes the antibiotic residues used in agriculture and aquaculture (Marshall & Levy, 2011). Likewise, new principles of antimicrobial treatment development need to include bacteriophages as alternative therapies for MDR *E.coli* (Lukman et al., 2020).Bacteriophages, viruses that target bacteria specifically, provide a new way to selectively kill MDR strains while minimizing the impact on the host microbiota. The identification and validation of novel drug targets such as the β-barrel assembly machinery (BAM) complex bring with them the development of new antimicrobial compounds (Xu et al., 2023). Indeed, a full solution to the problem of MDR *E.coli* will only be through a multidisciplinary approach, integrating, genomics, proteomics, biochemistry, and clinical microbiology. If researchers know more clearly how bacteria resist antibiotics and where to find new drug targets, the development of effective strategies against MDR may preserve antibiotic function for future generations.

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Nomenclature

Term	Description
Escherichia coli (E.coli)	A Gram-negative bacterium, common in the gut, sometimes pathogenic.
β-lactamases	Enzymes that provide resistance to β-lactam antibiotics.
BAM Complex	Protein system assembling outer membrane proteins in Gram-negative bacteria.
Efflux Pumps	Transport systems that eject antibiotics out of bacterial cells.
Porins	Proteins forming pores in bacterial outer membranes for molecule transport.
Biofilm	Protective bacterial communities resistant to antibiotics.
Viable but Non-Culturable (VBNC) State	Bacterial survival state where cells are alive but not growing on culture media.

Abbreviations

Abbreviation	Full Form
OMP	Outer Membrane Proteins
BAM	β-barrel Assembly Machinery Complex
MDR	Multi-Drug Resistance
AMR	Antimicrobial Resistance
CCA	Chromogenic Coliform Agar
EXPEC	Extraintestinal Pathogenic *E.coli*
ETEC	Enterotoxigenic *E.coli*
EIEC	Enteroinvasive *E.coli*
EAEC	Enteroaggregative *E.coli*
VTEC	Verotoxigenic *E.coli*
DAEC	Diffusely Adherent *E.coli*
PCR	Polymerase Chain Reaction
PFGE	Pulsed-Field Gel Electrophoresis
MALDI-TOF	Matrix-Assisted Laser Desorption/Ionization Time-of-Flight Mass Spectrometry

IMViC	Indole Methyl Red Voges-Proskauer Citrate Tests
ABC Transporters	ATP-binding Cassette Transporters
ATP	Adenosine Triphosphate
API 20E	Analytical Profile Index
MFS	Major Facilitator Superfamily

References

Amin MN, Khan MZI, Islam MA, Rahman MM, Hossain MA. Zoonotic transmission of antimicrobial resistance: A global threat. One Health. 2020;11:100176. doi: 10.1016/j.onehlt.2020.100176

Blair JM, Webber MA, Baylay AJ, Ogbolu DO, Piddock LJV. Molecular mechanisms of antibiotic resistance. Nat Rev Microbiol. 2015;13(1):42-51. doi: 10.1038/nrmicro3380

Breijyeh Z, Jubeh B, Karaman R. Resistance of Gram-negative bacteria to carbapenems: Important clinical challenges. Pharmaceuticals. 2020;13(12):406. doi: 10.3390/ph13120406

Carattoli A. Resistance plasmid families in Enterobacteriaceae. Antimicrob Agents Chemother. 2013;57(8):3892-3901. doi: 10.1128/AAC.00504-13

Cereto M, Latorre R, Rafecas A, Esteban R. Quinolone-resistant Escherichia coli in spontaneous bacterial peritonitis. J Hepatol. 2003;39(6):947-951. doi: 10.1016/S0168-8278(03)00443-8

Delcour AH. Outer membrane permeability and antibiotic resistance. Biochim Biophys Acta. 2009;1794(5):808-816. doi: 10.1016/j.bbapap.2008.11.015

Di Franco A, Fortini D, Migliavacca R, Grossi P, Nucleo E. Epidemiology and clinical impact of bloodstream infections caused by multidrug-resistant Escherichia coli: a retrospective cohort study. Infect Drug Resist. 2021;14:4683. doi: 10.2147/IDR.S332371

Doron S, Gorbach SL. Practice guidelines for the diagnosis and management of infectious diarrhea. Clin Infect Dis. 2008;46(6):943-948. doi: 10.1086/529825

Galindo-Méndez B. Antimicrobial resistance in Escherichia coli: mechanisms and impact on public health. J Glob Antimicrob Resist. 2020;22:742-751. doi: 10.1016/j.jgar.2020.06.021

Gauba K. Outer membrane proteins in gram-negative bacteria: structure, function, and implications in resistance. J Appl Microbiol. 2023;134(1). doi: 10.1111/jam.16019

Hendriksen RS. Isolation and identification of enterohaemorrhagic Escherichia coli O157. Global Salm-Surv. 2003.

Islam MA, Akter L, Rahman MM, Islam MS, Khan MZI. Isolation and identification of Escherichia coli from different sources in Dhaka city, Bangladesh. Int J Curr Microbiol App Sci. 2014;3(12):127-133.

Lange J, Hansen BM, Damborg P. Evaluation of chromogenic coliform agar for enumeration of Escherichia coli and coliform bacteria. J Microbiol Methods. 2013;95(2):291-293. doi: 10.1016/j.mimet.2013.09.007

Li XZ, Nikaido H. Efflux-mediated antimicrobial resistance in bacteria. Drugs. 2004;64(2):159-204. doi: 10.2165/00003495-200464020-00002

Lukman C, Yonathan C, Magdalena S, Waturangi DE. Isolation and characterization of pathogenic Escherichia coli bacteriophages from chicken and beef offal. BMC Res Notes. 2020;13(1):8. doi: 10.1186/s13104-019-4868-4

Lupindu AM. Isolation and characterization of Escherichia coli from animals, humans, and environment. In: Escherichia coli - Recent Advances on Physiology, Pathogenesis and Biotechnological Applications. IntechOpen; 2017.

Mah TF, O'Toole GA. Mechanisms of biofilm resistance to antimicrobial agents. Trends Microbiol. 2001;9(1):34-39. doi: 10.1016/S0966-842X(00)01913-3

Marshall BM, Levy SB. Food animals and antimicrobials: impacts on human health. Clin Microbiol Rev. 2011;24(4):718-733. doi: 10.1128/CMR.00020-11

Nikaido H. Outer membrane barrier as a mechanism of antimicrobial resistance. Antimicrob Agents Chemother. 2009;53(11):4291-4298. doi: 10.1128/AAC.00696-09

Oliver JD. Recent findings on the viable but nonculturable state in pathogenic bacteria. FEMS Microbiol Rev. 2010;34(4):415-425. doi: 10.1111/j.1574-6976.2009.00205.x

Pagès JM, Kerff F, Braun P, Folschweiller N. Bacterial porins: genetics, structure and function. FEMS Microbiol Rev. 2008;32(3):523-568. doi: 10.1111/j.1574-6976.2008.00112.x

Prestinaci F, Pezzotti P, Pantosti A. Antimicrobial resistance: a global multifaceted phenomenon. Pathog Glob Health. 2015;109(7):309-318. doi: 10.1179/2047773215Y.0000000038

Putman M, van Veen HW, Konings WN. Molecular biology of bacterial transport systems. Microbiol Rev. 2000;64(2):322-388. doi: 10.1128/MMBR.64.2.322-388.2000

Storek T, Ulman VL, Bernstein HD. Structure of the BamA lateral gate in its open state. Nat Struct Mol Biol. 2018;25(1):60-67. doi: 10.1038/s41594-017-0013-6

Tang KL, Caffrey NP, Nóbrega DB, Critchley J, Strubbe D. Restricting the use of antibiotics in food-producing animals and antibiotic resistance in humans: a systematic review and meta-analysis. Lancet Planet Health. 2023;7(1):e16-e27. doi: 10.1016/S2542-5196(22)00225-4

Tegegne H, Filie K, Tolosa T, Debelo M, Ejigu E. Isolation and identification of Escherichia coli O157:H7 recovered from chicken meat at Addis Ababa slaughterhouses. Infect Drug Resist. 2024;17:851-863. doi: 10.2147/IDR.S306323

Tetteh JK, Lin Z, Wu Y. Global impact of inappropriate use of antibiotics on antimicrobial resistance and its socioeconomic burden. Sustainability. 2020;12(7):2800. doi: 10.3390/su12072800

Wilson DN. Ribosome-targeting antibiotics and mechanisms of resistance. Nat Rev Microbiol. 2014;12(1):35-48. doi: 10.1038/nrmicro3121

Wu R, Stephenson R, Gichaba A, Noinaj N. The big BAM theory: An open and closed case?. Biochim Biophys Acta Biomembr. 2020;1862(1):183062. doi: 10.1016/j.bbamem.2019.183062

Xu Y, Wang Y, Wang Q. Targeting the β-barrel assembly machinery (BAM) complex as a strategy for developing novel antibiotics. Front Microbiol. 2023;14:1114592. doi: 10.3389/fmicb.2023.1114592