APTI women's forum

APTI Women's Forum Newsletter

Overcoming Antibiotic Resistance: Concepts and Approaches

A triannual newsletter published by Association of Pharmaceutical Teachers of India

May - Aug 2023

Editor's Note

Prof. Vandana B. Patravale

Chief Editor, APTI Women's Forum Newsletter

Dear Readers,

Writing the editorial for this volume of APTI Women's forum newsletter reminds me of 'First APTI Women leadership conclave' held at Kenilworth resort, Goa where our newsletter with thematic issue on *'Bone targeted delivery systems'* was released by the chief guest Prof. Indu Pal Kaur alongwith Dr.Mangirish and Dr. Shailesh from Goa state branch, in presence of editorial board members Prof. Suneela Dhaneshwar and Prof. Shubhini Saraf. Thanks to Dr. Rashmi Mishra and Dr. Pearl Pires Dighe for all the efforts taken to make the first conclave a huge success. Indeed, you both transformed vision of our APTI president Dr. Milind Umekar into reality. The support rendered by Dr. Raman Dang and all other members have instilled hope in women teachers. We were blessed to have great trainers Dr. Ashish Johri and Ms. Shabnam to hone our skills. The event was sponsored by Indoco Remedies, SMT, Aquatic Remedies, IPA Kolhapur branch, Maharukh Rustomjee, Mayor's lab and Tecknowledge Publications. APTI Women forum wishes to profusely thank each one of them for their generous support. This leadership conclave has generated 'sisterhood' amongst participants which is evident in many follow – up activities. Keep up this zeal dear participants!

The theme of current APTI women's newsletter is ***'Overcoming Antibiotic Resistance: Concepts and Approaches***'. According to WHO, antibiotic resistance is raising to dangerously high levels in all parts of the world as new resistance mechanisms are emerging and spreading globally. Unless action is taken, global burden of deaths from antibiotic resistance could rise to 10 million lives each year by 2050. This newsletter is a small effort in culminating articles to discuss strategies to overcome antibiotic resistance, mechanistic approaches, natural products for the same, computational approaches, antibiotic adjuvants, Kryptonite strategies, personalized medicines etc. It is very heartening to note that the newsletter features an article on 'Equine antibiotic resistance' indicating that working on 'one heath concept' (An approach to treat both human and animals) is need of the hour to effectively end this antibiotic resistance crisis. We, at the editorial board, extend our gratitude to all the authors for taking time out of their busy schedule and contributing towards this newsletter to make it a captivating read. I thank the entire editorial team for their efforts in conceptualizing this newsletter. I hope and expect that you all enjoy reading this newsletter as much as we enjoyed bringing it forth to you.

Also, I wish to thank our ICT students Sarika Jadhav, Dr. Emmanuel Chukwuebuka Umeyor, Preeya Negi, Ankita Anure, and Niyamat Chimthanawala for all the support rendered for this newsletter.

APTI Women's Forum
Editorial Board

Prof. Vandana B. Patravale
Professor of Pharmaceutics
Institute of Chemical Technology, Mumbai
(editor.aptiwomensforum@gmail.com)

Prof. Suneela Dhaneshwar
Director,
Amity Institute of Pharmacy, Amity University, Panvel,
Mumbai, Maharashtra

Prof. Shubhini Saraf
Director,
National Institute of Pharmaceutical Education and Research
(NIPER), Transit campus, Raebareli, Lucknow

Dr. Purnima Ashok
Dean R&D
East West Institutions,
Bengaluru

Prof. Kanchan Kohli
Director, Research & Publication,
Lloyd Institute of Management & Technology (Pharm.),
Greater Noida

Dr. Clara Fernandes
Associate Professor,
Bombay College of Pharmacy- Autonomous
(Affiliated to University of Mumbai)
Mumbai

Prof. Preeti K. Suresh
Professor of Pharmaceutics,
University Institute of Pharmacy
Director, Human Resource Development Centre
Pandit Ravishankar Shukla University, Raipur

Strategies to overcome antibiotic resistance: An overview

Ayushi Santosh Mokati, Lipika Uppal, Patel Mehalaka Naemoddin, Riti Chatterjee, Vaishnavi, Anil B. Jindal*

Department of Pharmacy, Birla Institute of Technology and Science, Pilani Campus, Rajasthan-333031
anil.jindal@pilani.bits-pilani.ac.in

1. Introduction

Modern medicine's remarkable advancements have saved countless lives and enhanced the quality of human existence. Antibiotics have been especially beneficial in treating infections and preventing severe complications. However, an alarming threat lurks in the future: the rise of antibiotic resistance. Antibiotic resistance is a significant and rising global danger to human, animal, and environmental health. Antibiotic resistance is the ability of microorganisms, mainly bacteria, to resist the effects of antibiotic drugs, making it more challenging to treat infections and presenting a grave threat to global health. The global resistome, made up of genes that cause antibiotic resistance, is affected by factors such as population growth, rising migration, excessive antibiotic use in medical and agricultural sectors, poor sanitation, wildlife transmission, and inadequate wastewater treatment (1). This alarming problem presents a plethora of issues, most notably affecting patient care and public health. As the efficacy of antibiotics declines, the treatment of once-easily curable infections becomes difficult, resulting in prolonged illnesses, increased medical costs, and increased mortality.

Particularly worrisome are "superbugs" or multidrug-resistant bacteria immune to all known antibiotics that can proliferate, posing treatment barriers. The UN estimates by 2050, superbugs and associated forms of antimicrobial resistance could be responsible for up to 10 million deaths, equaling the 2020 annual global mortality toll from cancer (2). The World Health Organization estimates that antimicrobial resistance (AMR) is responsible for approximately 4.9 million deaths annually, while a 2022 Lancet study revealed that 1.27 million deaths in 2019, including 860,000 in Africa, were the direct were the direct result of drug-resistant bacterial

bacterial infections (3, 4). While it is true that the risks associated with infectious diseases are a concern for all nations, it is essential to acknowledge that Low-Income Countries (LICs) and Lower-Middle-Income Countries (LMICs) shoulder a substantial portion of this burden, particularly those in Asia and Africa. As we face this imminent crisis, there is an urgent need to explore effective strategies to battle antibiotic resistance.

2. Understanding the development of antibiotic resistance: Molecular mechanisms

Antibiotic resistance is associated with various molecular and enzymatic processes through eliminating or modifying the drug. Primarily, four mechanisms are discussed here: modification of the binding target, membrane efflux protein, enzymatic inactivation, and molecular bypass (Figure 1).

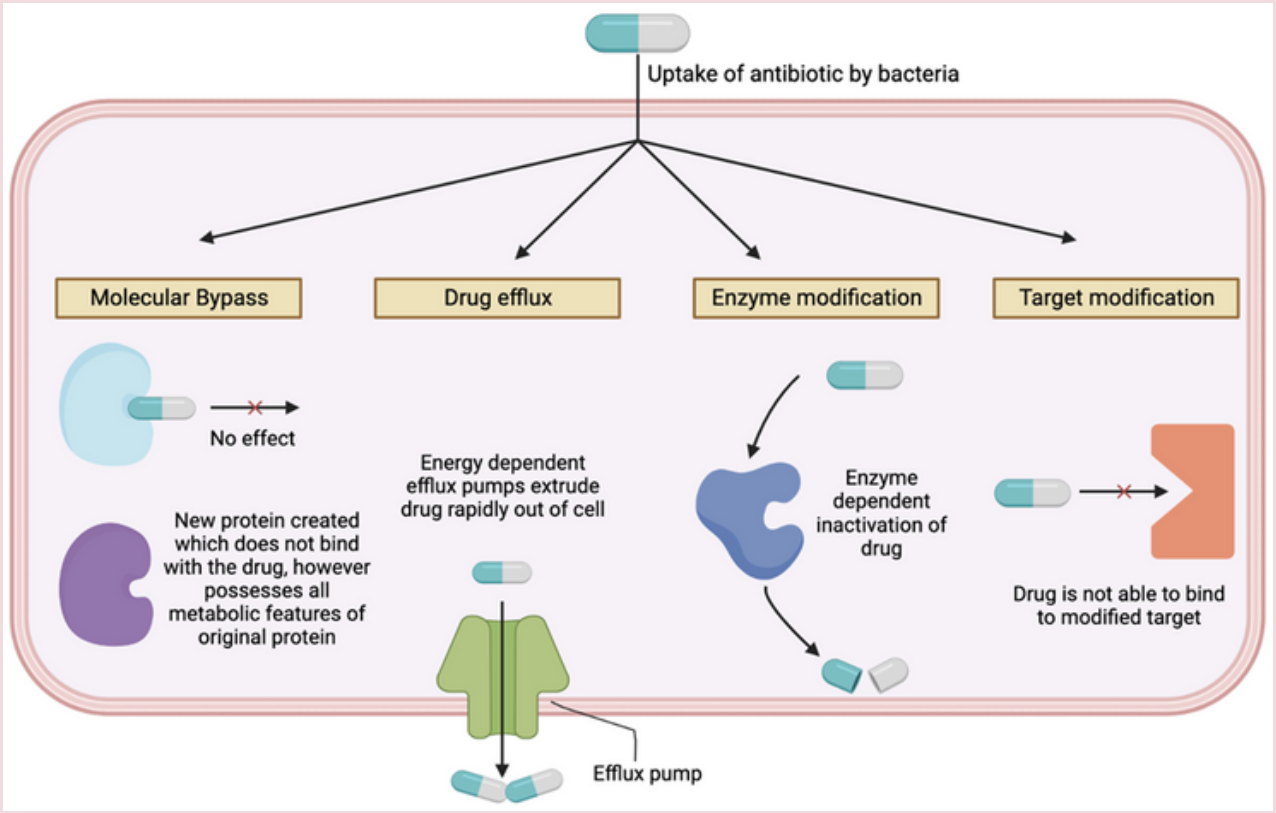


Figure 1. Molecular Mechanisms of Antibiotic Resistance.

2.1 Target modification: Modification of target binding is one of the most significant mechanism occurring due to point mutations in plasmid genes. The mutation of Serine in the GyrA gene with the bulkier group causes resistance to fluoroquinolones. The process can also involve the role of enzymes, as seen by Erm enzymes, which modify the 23sRNA ribosome, causing resistance to three classes of antibiotics: macrolides, lincosamide, and type B streptogramin in Escherichia coli (5).

2.2 Membrane efflux protein: Membrane efflux proteins are transporters involved in removing toxic substances from cells to the outside. Generally, it is the overexpression of these transporters that leads to resistance to antibiotics, as seen in Gram-negative bacteria, where the MFS (Major Facilitator Super-transporter) inhibits phenothiazines, and the RND family (Resistance Nodulation Division) inhibits quinolines. In Gram-positive bacteria, the overexpression of the MATE family inhibits Verapamil (6).

2.3 Enzyme inactivation: It can occur through the formation of a covalent enzyme intermediate followed by hydrolysis. The most important example is the Beta-lactamase enzyme, which causes the hydrolysis of beta-lactam by attacking the carbonyl carbon of B-lactam using the hydroxyl group of reactive Serine, further inhibiting the enzyme's action. Other antibiotics include fosfomycin, where the epoxide-containing ring is destroyed by a thiol-containing substrate followed by hydrolysis.

2.4 Molecular bypass: Certain bacteria can produce alternative proteins that might be ineffective against antibiotic action. The most famous example is found in Vancomycin (5). When acyl-D-Alanyl-D-Alanine is changed to acyl-D-Alanyl-D-Lactate, the amide bond is altered to an ester bond. As a result, an H donor deficiency is created, resulting in the non-productive binding of antibiotics.

3. Strategies for combating antibiotic resistance

3.1 Rational use of antibiotics

The irrational use of antibiotics has emerged as a pressing global concern, as many individuals are not adhering to the standard treatment guidelines set forth by the World Health Organization (WHO). Both the public and private sectors must be educated effectively to ensure the rational use of antibiotics. Shockingly, less than 30% of people in the private sector and only 40% in the public sector currently follow the proper guidelines for treatment, leading to the development of antibiotic resistance. A major contributing factor is the rampant use of over-the-counter (OTC) antibiotics, where individuals obtain and use these drugs without a prescription, often neglecting to complete the full course of treatment. To combat this problem, stringent measures and regulatory controls must be implemented to prevent the use of antibiotics without a prescription (7). Proper use of antibiotics should consider critical factors, such as the patient's clinical condition, the specific pathogen causing the disease, and the resistance pattern of the drug. Making decisions regarding the change of antibiotics based on microbial sensitivity and the patient's condition falls under the responsibility of registered medical practitioners. Therefore, antibiotic guidelines must be regularly updated to optimize the use of appropriate antibiotics, including considerations for the proper route of administration, treatment duration, and dosing frequency. By promoting a greater understanding of the significance of rational antibiotic use, we can safeguard the efficacy of these vital medications and combat the rise of antibiotic resistance effectively (8).

3.2 Discovery of new antibiotics

The decline in the efficacy of readily accessible antibiotics is a significant worldwide healthcare issue in the context of treating bacterial infections since these medications are susceptible to the development of resistance (9). There exists a pressing need to replace the currently available antibiotics in the market with novel drugs and pharmacological classes to sustain their efficacy for the treatment of bacterial illnesses. One potential strategy is the development of analogues of already available medications that possess the ability to combat bacterial species that have developed resistance (10).

Another potential strategy that has recently emerged involves the alteration of naturally occurring peptides, which exhibit both non-resistance to bacteria and comparable efficacy to

other categories of antibiotics. The researchers have made modifications to the naturally occurring peptide known as “thanatin” to augment its efficacy and mitigate bacterial resistance. This intervention has generated very promising outcomes. The identification of new peptides has the potential to considerably decelerate the development of antibiotic resistance.

Nevertheless, it is recommended by experts in the field of infectious diseases that the utilization of novel antibiotics be limited and reserved as a final resort for the management of the ailment. These medications are specifically designated for instances of severe resistance and should only be administered when patients have developed resistance to currently available antibiotics. As a result of this factor, the rate of discovery and advancement of novel antibiotics has diminished due to insufficient return on investments (9).

3.3 Adjuvant therapy

Antibiotic adjuvants are compounds that lack direct antimicrobial activity. Instead, they are co-administered with antibiotics to combat bacterial resistance and preserve their efficacy. Consequently, antibiotic adjuvants are classified into two groups: Class I agents that target the pathogen include - β-lactamase inhibitors, and efflux pump inhibitors (EPIs), whereas membrane permeabilizing are class II agents that act on the host.

3.3.1 β-lactamase inhibitors: β-lactam antibiotics (penicillins, cephalosporins, and carbapenems) disrupt the proteins necessary for peptidoglycan biosynthesis leading to bacterial cell lysis. However, β-lactamases, bacteria’s defense enzymes, inactivate them by hydrolyzing the β-lactam ring in their structure and developing a resistant phenotype. β-lactamases inhibitors block this enzyme's active site, boosting the potency of co-administered antibiotics. Clavulanic acid, sulbactam, tazobactam, avibactam, vaborbactam, and relebactam are approved β-lactamases inhibitors used with specific antibiotics.

3.3.2 Efflux pump inhibitors: Efflux pumps in bacterial membranes expel antibiotics, lowering intracellular concentrations and causing resistance. Adjuvants called EPIs physically hinder these pumps, increasing the intracellular antibiotic concentration and restoring sensitivity.

3.3.3 Membrane Permeabilizer: Antibiotics must penetrate the bacterial membrane to work. For Gram-negative bacteria protected by an outer membrane, antibiotic adjuvants promote membrane permeability (permeabilizer). These cationic and amphiphilic compounds destabilize the outer membrane by interacting with lipopolysaccharides or capturing cations, facilitating antibiotic passage. Substances such as polymyxin B, colistin, aminoglycosides, cationic peptide, and polyamines act as permeabilizers (11).

4. Novel drug delivery system

The emergence of novel drug delivery systems (DDS) represents a promising and innovative strategy to enhance antibiotic treatment regimens while reducing the development of antibiotic resistance. By formulating single or combinations of multiple antibiotics into diverse nano-formulations, including metal-based (e.g., gold, silver nanoparticles), lipid-based (e.g., liposomes, solid lipid nanoparticles), carbon-based (e.g., Carbon nanotubes, graphene sheets), and polymeric nanoparticles (derived from both natural and synthetic polymers), these advanced DDS offer viable alternatives to conventional antibiotics as efficient drug-delivery vehicles (12).

An essential advantage of nanoparticle-based DDS is their ability to protect antibiotics from enzymatic degradation, prolonging drug release and increasing drug half-life and bioavailability. Furthermore, these DDS can be engineered to facilitate antibiotic bacterial binding, resulting in higher antibiotic concentrations at the site of infection. Upon penetrating the microbial cell membrane, the nanomaterials induce structural alterations, disrupting the size and configuration of the bacterial membrane and altering metabolic pathways. Subsequently, when the nanoparticles enter the intracellular environment, they engage with biological pathways, impeding enzymes, deactivating proteins, instigating oxidative stress, causing electrolyte imbalances, and inducing genetic variations (13). These orchestrated interactions result in the primary eradication of microorganisms.

By optimizing drug delivery, inhibiting bacterial defense mechanisms, and fostering synergistic effects, nanoparticle-based DDS demonstrate unparalleled potential in revolutionizing the management and control of antibiotic-resistant infections (Figure 2).

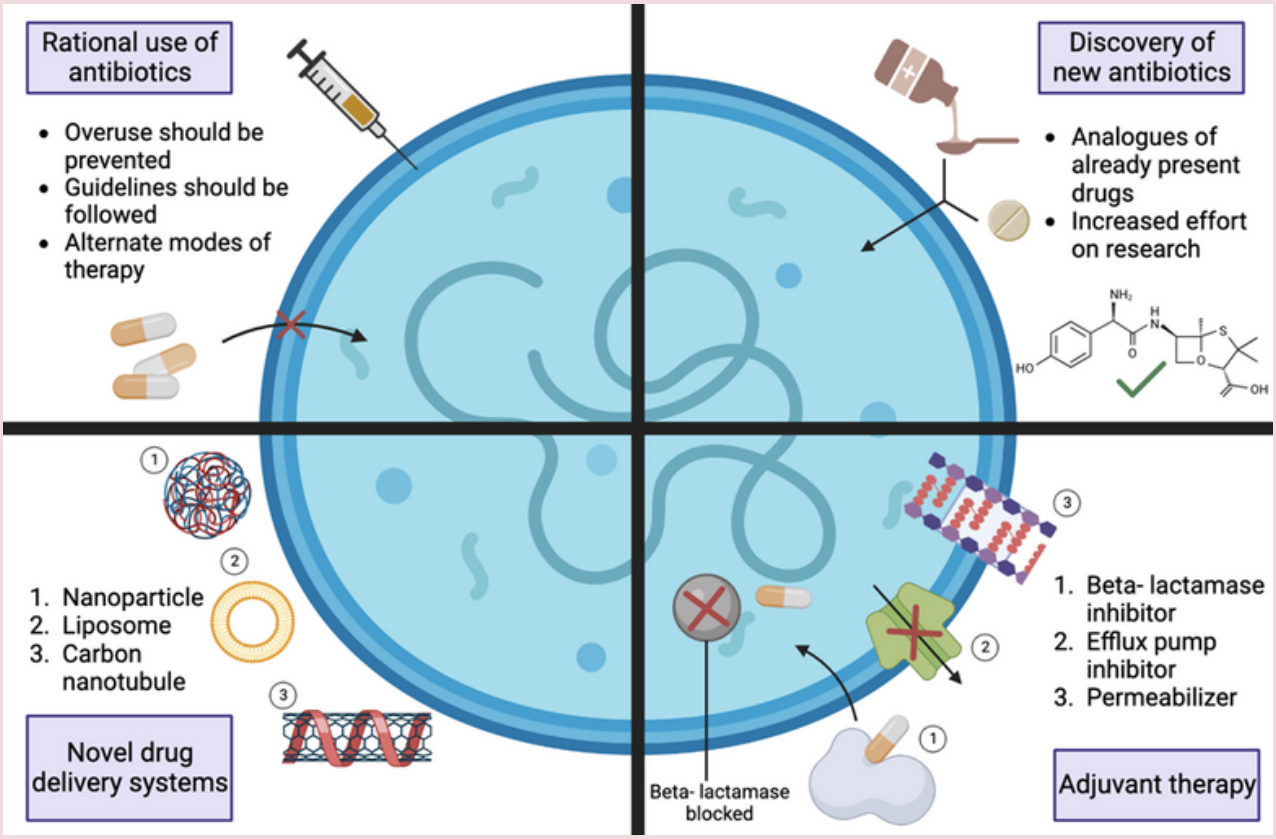


Figure 2. Strategies to combat Antimicrobial Resistance

5. Conclusion

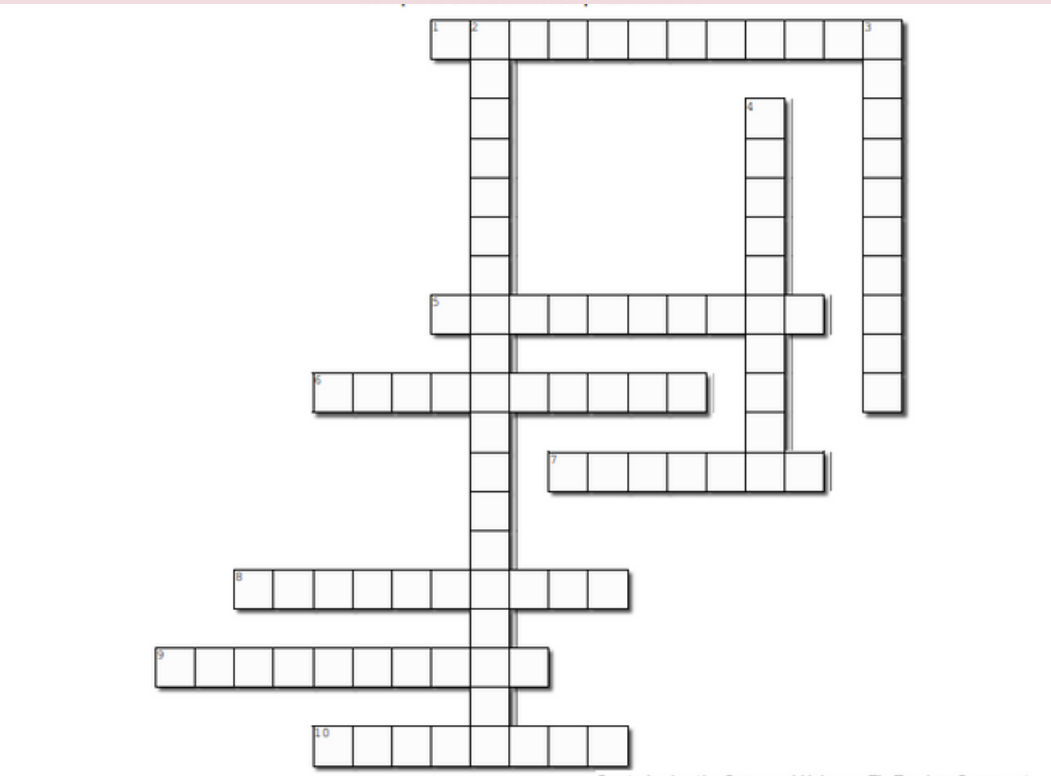
The current problem of antibiotic resistance is a serious challenge to global health and demands immediate action. An increase in multidrug-resistant pathogens and the lack of efficient existing antibiotics requires innovative solutions. Rational antibiotic use, the discovery of new antibiotics and adjuvant therapies, and novel drug delivery systems offer promising avenues to combat this crisis. Urgent collaboration between healthcare providers, policymakers, researchers, and the public is essential to preserve the effectiveness of antibiotics and treat disease. We can pave the way for a healthier and more sustainable future for future generations with concrete efforts.

References:

1. Aslam B, Wang W, Arshad M, Khurshid M, Muzammil S, et al. Antibiotic resistance: a rundown of a global crisis. Infection and Drug Resistance. 2018;11:1645-1658.
2. UNEP. Bracing for Superbugs: Strengthening environmental action in the One Health response to antimicrobial resistance. UNEP - UN Environment Programme. 2023. Retrieved July 24, 2023, from https://www.unep.org/resources/superbugs/environmental-action
3. Murray CJL, Ikuta KS, Sharara F, Swetschinski L, Aguilar GR, et al. Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis. The Lancet. 2022;399(10325):629-655.
4. O'Neill J. Tackling drug-resistant infections globally: final report and recommendations. 2016. Retrieved July 24, 2023, from https://apo.org.au/node/63983.
5. Wright GD. Molecular mechanisms of antibiotic resistance. Chem Commun. 2011;47:4055-4061.
6. Annunziato G. Strategies to Overcome Antimicrobial Resistance (AMR) Making Use of Non-Essential Target Inhibitors: A Review. Int J Mol Sci. 2019 Nov 21;20(23):5844.
7. Milani B, Scholten W. & World Health Organization. The world medicines situation 2011: access to controlled medicines (No. WHO/EMP/MIE/2011.2. 4). World Health Organization. 2011.
8. Bhagwati A. Guidelines for antibiotic usage in common situations. J Assoc Physicians India. 2010;58:49-50.
9. Ventola CL. The antibiotic resistance crisis: Part 1: Causes and threats. P T. 2015;40(4):277-83.
10. Silver LL, Bostian KA. Discovery and development of new antibiotics: the problem of antibiotic resistance. Antimicrobial agents and chemotherapy. 1993;37(3):377-383.
11. Laws M, Shaaban A, Rahman KM. Antibiotic resistance breakers: current approaches and future directions. FEMS Microbiology Reviews. 2019;43(5):490-516.
12. Lai CK, Ng RW, Leung SS, Hui M, Ip M. Overcoming the rising incidence and evolving mechanisms of antibiotic resistance by novel drug delivery approaches – An overview. Advanced Drug Delivery Reviews. 2022;181:114078.
13. Imran M, Jha SK, Hasan N, Insaf A, Shrestha J, et al. Overcoming Multidrug Resistance of Antibiotics via Nanodelivery Systems. Pharmaceutics. 2022;14(3):586.

Fun & frolic - Crossword

Solution on page 89



Equine antibiotic resistance: epidemiology, regulation and strategies there of

Purav Shah

Thoroughbred Remedies Manufacturing. TRM. Industrial Estate, Newbridge, Co Kildare, W12 ND30, Ireland.

pshah733@yahoo.in

1. Introduction

Antimicrobial resistance is the ability of the microbe to endure and multiply within the body tissues in the presence of highest tolerated antimicrobial dose. This being a global healthcare threat, it has not just remained confined to humans but has successfully stretched its presence even in animals. Of the various antimicrobials used in veterinary practices, antibiotics are quite readily prescribed by the veterinarians against infections, becoming the basis of developing bacterial resistance in animals. However, it is only recently that the antimicrobial resistance in horses has started to receive due attention, at both, public and clinical levels.

Methicillin-resistant Staphylococcus aureus (MRSA) is no longer the only bacterial species that exhibits equine antibiotic resistance; many other bacterial species, notably gram-negative bacteria like E. coli, have also exhibited similar resistance in clinical settings. The four critical targets for any antibacterial agent in the bacterial cells are – inhibition of cell wall synthesis, prohibiting genetic material (DNA/ RNA) synthesis, preventing protein synthesis or disruption of a vital metabolic pathway.

1.1 Epidemiology in equine antibiotic resistance

Researchers have made efforts in understanding the epidemiology behind single or multi-drug resistance, particularly amongst MRSA and β-lactamase producing E. coli in equine samples using both phenotypic and genotypic methodologies.

1.2 Antimicrobial resistance against MRSA

MRSA carriage has been detected on the skin and mucous layers of the nasal chambers in horses. Though all MRSA colonization may not lead to clinical infections but may possess a threat for potential infections under certain circumstances, most likely during hospitalization of horses. With a 2.3-6.4% prevalence in hospitalized horses under normal circumstances, these values can go very high under an infection outbreak (1).

At molecular levels, the epidemiology is governed by the carriage of variably sized DNA fragments of the SCCmec gene cassettes. Within this, the mecA gene encodes PBP2a, an

alternative penicillin-binding protein, which has reduced affinity for β-lactam drugs like methicillin. This mechanism induces resistance in horses (2). Further, a narrow spectrum of resistance is offered by the encoding of the blaZ β-lactamase gene, which synthesizes penicillinase that breaks down the β-lactam penicillin's, majorly encountered in staphylococci from horses (3).

1.3 Antimicrobial resistance against *E. coli*

Resistance to all antimicrobials has been encountered in E. coli for horses. Faecal carriage of E. coli has been reported quite high in hospitalized horses (60.5-81.7%) compared to non-hospitalized community (13.4-24.5%) horses. Similar estimates and faecal carriage ratios are prevalent for multidrug resistant and extended spectrum β -lactamase (ESBL) producing E. coli isolates in hospitalized horses than in the community horses (4).

Due to inherent inability for penicillin penetration through E. coli's outer layers, there is an increasing resistance to β-lactam drugs primarily by producing inactivating β -lactamase enzymes like TEM-1, TEM-2, and SHV-1, or AmpC β-lactamases, which are all encoded by different bla resistance genes (5). Recently equine faecal samples of clinically normal horses have shown ESBL producing E. coli isolates, particularly the CTX-M-15 type from the ST131 gene sequence (6). Similarly, mild ciprofloxacin resistance was observed in equine samples due to an over synthesise of AAC(6')-lb-cr, an aminoglycoside modifying acetyltransferase enzyme along with over expression of efflux pumps due to qnr gene encoding towards fluoroquinolone resistance (4).

1.4 Antimicrobial resistance against other microbes

Intrinsically resistant to cephalosporins and aminoglycosides, vanA and vanB genes have been identified exhibiting resistance to vancomycin in enterococcal isolates from horses with sample prevalence of 6.7-9.6%. Similarly, equine enterococcal resistance to macrolides and tetracycline have been identified to be caused by erm(B) and tet(L) genes (4, 7).

With only limited antibiotics available to treat Pseudomonas caused sepsis, extensive multidrug resistance has been reported in foals (4). However, epidemiologically antimicrobial resistance against Pseudomonas (particularly P. aeruginosa), a major causative agent for various equine infections is lacking.

Salmonella, another significant causative agent for various equine diseases has shown multidrug resistance in equine faecal samples. Several ESBL genes (blaCTX-M-1, blaCTX-M-15, blaSHV-12), plasmid mediated ampC genes (blaCMY-2) and integrons associated genes (DT104) have been identified in equine Salmonella isolates (8, 9). Similarly, equine sulphonamide and trimethoprim resistance is observed due to over expression of aminoglycoside adenyltrasferase enzyme and several dihydrofolate reductase enzyme analogues from dfr genes respectively (4, 10).

Acinetobacter has significantly been understood for human diseases, however very restricted information is available epidemiologically in equine cases. A. baumannii has shown various aminoglycoside resistance genes. Similarly, various Acineobacter isolates have documented carbapenemase OXA-23 genes in horses that metabolize carbapenem (4).

1.5 European Medicines Agency (EMA) Approach in assessing antimicrobial resistance in veterinary medicine

The emerging instances of antibiotic resistance in veterinary practices has posed a global healthcare threat due to limited availability of therapeutic options in combatting any infections. Repeated exposure to antimicrobial drugs has caused the microbes to undergo changes (including genetic mutations) and stop them from being killed or inactivated by the treatment.

By supporting the responsible use of antibiotics and taking effective measures in order to minimise the development of antibiotic resistance in animals, including food-producing animals like horses, EMA has been following a steadfast approach.

Under the veterinary regulatory guidelines of the EMA, antimicrobial resistance in veterinary medicines section highlights the following key topics summarised as below:

1.5.1 Committee for Veterinary Medicinal Products (CVMP) strategy on antimicrobials

As a strategic plan for 2021-2025, CVMP is focusing on the implementation of the Veterinary Medicines Regulation provision along with a guideline on the demonstration of efficacy for antimicrobial veterinary medicines. Further, it also provides guidelines on the metaphylactic use of antimicrobics in animals at the verge of catching an infection.

1.5.2 Monitoring veterinary antimicrobial consumption

As horses are considered as food-producing animals, it is vital to monitor antimicrobial consumption that may lead to resistance and potential contamination in the food chain. Within the European Union (EU) member state, from 2010 EMA has initiated the 'European Surveillance of Veterinary Antimicrobial Consumption (ESVAC)' project which collects data on the use of antimicrobials in the EU and the European Economic Area (EEA). This data can be publicly accessed through the 'European database of sales of Veterinary antimicrobial agents.

1.5.3 Analysis of antimicrobial consumption and resistance

The EMA analyses the intake of antibiotics by animals and the development of resistance thereafter in close collaboration with the European Food Safety Authority (EFSA) and the European Centre for Disease Prevention and Control (ECDC). The results get published in the 'Joint Inter-agency Antimicrobial Consumption and Resistance Analysis' (JIACRA) report.

1.5.4 Recommendations on the use of antibiotics in animals

EMA along with other relevant EU authorities, involving EFSA, issues the RONAFA opinion based on the 3R's theme – reduce, replace and rethink. This joint opinion aims to reduce the rigorous use of antimicrobial agents in animal rearing. Previous reports from EMA, EFSA, ECDC and European Commission's 'Scientific Committee on Emerging and Newly Identified Health Risk (SCENIHR)' have pressed the need for not just the wise use of antimicrobics in animals but has also strongly recommended adherence to basic animal husbandry hygiene practices. Additionally, a sturdy approach towards monitoring of antibiotic resistance, fostering novel areas of research and strategies to overcome antibiotic resistance in animals are also advised.

1.5.5 Proposed methods for preventing antibiotic resistance in horses

Across the EU and the EMA, experts/ veterinarians propose strategic approaches towards preventing/ minimising or slowing down the antibiotic resistance in horses.

- Follow the prescribed regimen thoroughly.
- Never use one antibiotic amongst different horses. Doses or frequency of dosing are tapered according to individual horse's condition.
- Just like for humans, do not stop administering the prescribed antibiotic midway if the condition is seen improving in the horse.
- Never keep outdated antibiotics for later use.
- Never skip a dose.
- Always contact a clinical expert if no improvement is seen/ conditions worsen.
-

Apart from the key advices on the use of antibiotic in horses to prevent resistance, basic good hygiene animal husbandry and biosecurity principles should be followed in preventing infections. Self-medication without a veterinarian's intervention should be avoided based on just visual inspection by the owner, which is quite common in parts with difficult access to vet clinics. Most of the times, especially in self-medication, viral infections can be misunderstood as bacterial infections and using antibiotics in viral infection may lead to severe antibiotic resistance.

2. Conclusion

Antibiotic resistance in horses is an alarming concept globally. For easy access and efficacy of antibiotic therapies, antimicrobial/ antibiotic stewardship must be embraced. Being a nexus with multifaceted approach, key emphasis must be given to the monitoring and education on the use of antibiotics in horses. Further, understanding the mechanisms causing the resistance is critical which would dictate the clinicians to effectively choose an antibiotic regimen against a particular infection. However, there is limited research work done on understanding the epidemiology of antibiotic resistance in various microbes in horses. A collaborative effort must be made between academician, industry, and regulatory bodies to foster this multifactorial research forming the basis of practicing antimicrobial/ antibiotic stewardship. Additionally, reporting and documentation of any antibiotic resistance observed in horses must be efficiently reported and must be well documented with the regulatory bodies. This would enhance evidence-based decision-making on antibiotic selection and direct the pathway towards development of more robust and novel antibiotic moieties.

References

1. Weese JS, Rousseau J, Willey BM, Archambault M, McGeer A, Low DE. Methicillin-resistant Staphylococcus aureus in horses at a Veterinary Teaching Hospital: Frequency, characterization, and association with clinical disease. J Vet Intern Med. 2006;20(1):182-186.
2. Brown DFJ, Reynolds PE. Intrinsic resistance to B-lactam antibiotics in. FEBS Lett. 1980;122(2).
3. Olsen JE, Christensen H, Aarestrup FM. Diversity and evolution of blaZ from Staphylococcus aureus and coagulase-negative staphylococci. J Antimicrob Chemother. 2006;57(3):450-460.
4. Maddox TW, Clegg PD, Williams NJ, Pinchbeck GL. Antimicrobial resistance in bacteria from horses: Epidemiology of antimicrobial resistance. Equine Vet J. 2015;47(6):756-765.
5. Hawkey PM. Molecular epidemiology of clinically significant antibiotic resistance genes. Br J Pharmacol. 2008;153(1);406-413.
6. Ewers C, Bethe A, Semmler T, Guenther S, Wieler LH. Extended-spectrum β-lactamase-producing and AmpC-producing Escherichia coli from livestock and companion animals, and their putative impact on public health: A global perspective. Clin Microbiol Infect. 2012;18(7):646-655.
7. Moura I, Radhouani H, Torres C, Poeta P, Igrejas G. Detection and genetic characterisation of vanA-containing enterococcus strains in healthy lusitano horses. Equine Vet J. 2010;42(2):181-183.
8. Niwa H, Anzai T, Izumiya H, Morita-Ishihara T, Watanabe H, et al. Antimicrobial resistance and genetic characteristics of salmonella typhimurium isolated from Horses in Hokkaido, Japan. J Vet Med Sci. 2009;71(8):1115-1119.
9. Dallap Schaer BL, Aceto H, Rankin SC. Outbreak of salmonellosis caused by Salmonella enterica serovar Newport MDR-AmpC in a large animal veterinary teaching hospital. J Vet Intern Med. 2010 Sep-Oct;24(5):1138-46.
10. Zhao S, Qaiyumi S, Friedman S, Singh R, Foley SL, et al. Characterization of Salmonella enterica Serotype Newport Isolated from Humans and Food Animals. J Clin Microbiol. 2003;41(12):5366-5371.

A novel frontier in computational approaches and personalised medicine interventions to overcome antibiotic resistance


Akey Krishna Swaroop[1], Preeya Negi[2*]

1Department of Pharmaceutical Chemistry, JSS College of Pharmacy, Ootacamund, JSS Academy of Higher Education & Research, Mysuru, India, 643001

2* Department of Pharmaceutical Sciences and Technology, Institute of Chemical Technology, Matunga, Mumbai, Maharashtra, India, 400019

negipreeya1@gmail.com

1. Introduction

The growing increase in antibiotic resistance is a health catastrophe that poses a direct challenge to global health. Programmes like Global Antimicrobial Resistance Surveillance System under WHO (World Health Organization), Global Health Security Agenda (GHSA), and Antimicrobial Resistance Action Package (GHSA Action Package Prevent-1) were created to address this problem (1). Antibiotics, sometimes called "miracle drugs," have prevented the needless deaths of many people from bacterial infections or diseases. The evolution of antibiotic-resistant bacteria, however, has been fuelled by the extensive and often improper use of antibiotics in human and animal healthcare and in agriculture (2). Some bacterial strains have developed resistance to antibiotics, making these life-saving medications useless against bacterial infections that were formerly readily treated (1). Bacteria are incredible survivors because of their innate capacity to adjust to new conditions, including exposure to antibiotics (3). Due to the high chances of survival by antibiotic resistant bacteria, genetic mutations that support antibiotic resistance have spread widely across bacterial populations. Furthermore, bacterial species may rapidly exchange resistance genes with one another through horizontal gene transfer mechanisms such as plasmid transfer. Determining how antibiotic resistance occurs is crucial for devising solutions to this significant global challenge (4). Although conventional methods have provided useful insights, they are inadequate to fully grasp the genetic and molecular complexities of antibiotic resistance. Consequently, computational methods have shown great promise as a paradigm shift in the battle against antibiotic resistance. Computational techniques provide better in-depth knowledge of the genetic basis of resistance and novel ways to design innovative therapies through the use of contemporary computers, data analysis, and sophisticated algorithms. Artificial intelligence (AI) may speed up the preclinical stage of drug development by generating numerous novel chemical

suggestions using algorithms developed with machine learning (ML) methods (5). Hence, the integration of computational methods and precision medicine has the potential to revolutionize how we tackle antibiotic resistance and safeguard the efficacy of antibiotics for future generations.

2. Use of computational approaches in antibiotic resistance

The complex problems presented by antibiotic resistance have made computational methods significant. Researchers may analyse massive datasets, including genomic information, proteomic interactions, and microbiome data, to better understand the genetic basis of resistance mechanisms (6). Bacterial evolution to acquire resistance to antibiotics could be better understood by the combination of computational biology, bioinformatics, and artificial intelligence (7). In 2009, a team employed ML algorithms in conjunction with the quantitative structure-activity relationship (QSAR) approach to find novel antimicrobial peptides with antibacterial capabilities. With the ARGs (antibiotic resistant genes) data obtained from the COALA database (collection of all antibiotic resistance gene databases), an algorithm was developed to predict the class of antibiotics like sulphonamides, tetracyclines, beta-lactams, etc. using a DL (deep learning) derived ensemble method. This would cut down on the failure rate associated with empirical antibiotic treatment and save time compared to traditional antibiotic susceptibility testing (AST) (8).

3. Precision medicine

Precision medicine has shown promise as a personalised and targeted strategy to combating the complicated issue of antibiotic resistance. The use of genetic testing to detect genetic markers linked with antibiotic resistance is a crucial component of precision medicine in the context of antibiotic resistance (9). The mechanisms of resistance can be analysed by studying the genetic composition of patients and that of the invading bacteria to help healthcare professionals in choosing the best antibiotic therapy and steer clear of drugs that the bacteria have developed resistance to. For instance, a patient presenting with a severe bacterial infection may undergo genetic testing to determine whether the infecting bacteria harbour specific resistance genes (10). Precision medicine is a patient-centred and data-driven healthcare system that employs a wide range of techniques to provide personalised treatment plans. Some forms of precision medicine are listed in table 1 with the computational tools that can be utilised for respective study.

3.1 Genetic based precision medicine

Precision medicine based on genetic analysis seeks to determine how specific genetic compositions of patients affect their responses to antibiotics. Genes involved in drug metabolism, transport, and target interactions may be identified via genetic testing and the impact of variations in such genes on antibiotics can be studied. Antibiotic effectiveness and adverse effects might be affected by unique genetic makeup of an individual (11). Deciphering the huge amount of genetic data produced by genome sequencing relies heavily on computational methods. In order to determine which antibiotics should be used, sophisticated algorithms can detect important genetic variations and evaluate their clinical importance. Antibiotic resistance genes may be a target for novel drug discovery, and population-level investigations can reveal genetic patterns linked to antibiotic resistance (12).

Table 1. Computational tools for study of precision medicine to overcome antibiotic resistance

S. No.	Type of precision medicine technique	Computational tools		
		Use	Tool name	Link
1.	Genetic-based precision medicine	Genomic sequencing and variant calling	Burrows-Wheeler Aligner	https://bio-bwa.sourceforge.net/
			Bowtie	https://bowtie-bio.sourceforge.net/index.shtml
			GATK (Genome analysis toolkit)	https://gatk.broadinstitute.org/hc/en-us
			Samtools	http://www.htslib.org/
		Variant annotation and interpretation	ANNOVAR	https://annovar.openbioinformatics.org/en/latest/
			SnpEff	http://pcingola.github.io/SnpEff/
			Variant Effect Predictor (VEP)	https://asia.ensembl.org/info/docs/tools/vep/index.html
		Population-level genetic analysis	PLINK	https://zzz.bwh.harvard.edu/plink/
			Genome-wide Complex Trait Analysis (GCTA)	https://yanglab.westlake.edu.cn/software/gcta/
2.	Microbiome-based precision medicine	Microbiome data analysis	QIIME2	https://qiime2.org/
			MetaPhlAn 4.0	https://huttenhower.sph.harvard.edu/metaphlan/
			HUMAnN 3.0 (HMP Unified Metabolic Analysis Network)	https://huttenhower.sph.harvard.edu/humann/
		Functional analysis of microbiome data	PICRUSt 2.0 (Phylogenetic Investigation of Communities by Reconstruction of Unobserved States)	https://huttenhower.sph.harvard.edu/picrust/
			Tax4Fun	http://tax4fun.gobics.de/
		Microbial co-occurrence network analysis	CoNet	http://raeslab.org/software/conet.html
			SparCC	https://web.mit.edu/almlab/sparcc.html
			WGCNA	https://bio.tools/wgcna
3.	Pharmacogenomics	Pharmacogenetic analysis	CPIC	https://cpicpgx.org/
			PharmGKB	https://www.pharmgkb.org/
			PharmVar	https://www.pharmvar.org/
		Genetic variant-drug interaction prediction	PolyPhen-2	http://genetics.bwh.harvard.edu/pph2/
			SIFT	https://sift.bii.a-star.edu.sg/

Table 1. Computational tools for study of precision medicine to overcome antibiotic resistance (continued)

4.	Proteomics and biomarker-based precision medicine	Proteomics data analysis	MaxQuant	https://www.maxquant.org/
			Proteome Discoverer	https://www.thermofisher.com/in/en/home/industrial/mass-spectrometry/liquid-chromatography-mass-spectrometry-lc-ms/lc-ms-software/multi-omics-data-analysis/proteome-discoverer-software.html
			Skyline	https://skyline.ms/project/home/software/Skyline/begin.view
		Pathway and functional enrichment analysis	DAVID	https://david.ncifcrf.gov/
			Panther	https://pantherdb.org/
			Reactome	https://reactome.org/
		Biomarker discovery and validation	Lasso regression	https://www.xlstat.com/en/solutions/features/lasso-regression
			ROC analysis	https://www.rocplot.org/
5.	Systems biology and network pharmacology	Network analysis	Cytoscape	https://cytoscape.org/
			NetworkX	https://networkx.org/
			Gephi	https://gephi.org/
		Network-based drug target prediction Or Network-based Inference of Drug Similarity (NIDS)	NetCBP	https://netcap.io/
			NetPhos	https://services.healthtech.dtu.dk/services/NetPhos-3.1/
		Pathway and network analysis	STRING	https://string-db.org/
			Ingenuity Pathway Analysis (IPA)	https://digitalinsights.qiagen.com/products-overview/discovery-insights-portfolio/analysis-and-visualization/qiagen-ipa/
			Reactome	https://reactome.org/

3.2 Microbiome based precision medicine

The billions of bacteria found in and on the human body, collectively known as the human microbiome, have a major impact on our health and wellbeing. Antibiotic resistance may be fostered in part by bacteria in the gut that either directly harbour resistance genes or aid in the transfer of those genes to pathogenic bacteria. Large amounts of microbiome data can be analysed to find microbial fingerprints linked to antibiotic resistance through computational methods. Integration of microbiome data with patient-specific information, such as antibiotic exposure history and health status, could be implemented for the purpose of microbiome alteration and antibiotic medication optimisation. The use of antibiotics may alter the makeup

of the microbiome, and computer models can predict how this will affect the likelihood of resistance development, allowing for better informed treatment decisions (13).

3.3 Pharmacogenomics

The field of pharmacogenomics studies the impact of individual differences in response to medications like antibiotics. Pharmacogenomics aids in antibiotics selection for individual patients by analysing changes in drug-metabolizing enzymes and drug transporters. By taking these precautions, harmful medication interactions may be avoided and treatment results can be improved (14). The field of pharmacogenomics uses computational algorithms to analyse genetic data in order to predict the pharmacological reaction of an individual. The effects of numerous genetic variations on drug metabolism may be evaluated using machine learning models, allowing for fine-tuned dosing for best therapeutic results (15). Pharmacogenomics is helpful in the setting of antibiotic resistance because it allows for the identification of people at greater risk of acquiring resistance and guides the selection of medicines that are less likely to induce resistance.

3.4 Proteomics and biomarker based precision medicine

Proteomics is the study of all proteins expressed in a cell or tissue. Antibiotic treatment choices and the creation of tailored medicines may be aided by the identification of proteins or biomarkers linked with antibiotic resistance. Researchers can identify proteins that are essential to resistance mechanisms by comparing the proteomes of resistant and susceptible bacterial strains. This information can be used to create innovative and quick diagnostic techniques or to build tailored therapies to counteract resistance processes (16,17).

3.5 Systems biology and network pharmacology

Systems biology computational methods investigate regulatory networks and metabolic processes in bacteria to identify points of weakness that might be exploited in the fight against resistance. Computational techniques uncover possible intervention locations for altering resistance mechanisms by modelling the interconnection of biological systems. In network pharmacology, computational approaches are used to investigate systems-level interactions between medicines, proteins, and genes. Using this method, scientists may find possibilities for repurposing existing drugs or formulate new medication combinations with enhanced efficacy against treatment-resistant illnesses (18).

4. Conclusion and future perspectives

The meeting point of precision medicine and computational methods offers a game-changing chance to tackle the critical problem of antibiotic resistance. By tailoring medical interventions to individual patients based on their genetic makeup, microbiome composition, and other personalized factors, precision medicine offers the potential to optimize antibiotic treatment and minimize the risk of resistance development. Computational tools and advanced algorithms play pivotal roles in deciphering complex genomic, proteomic, and microbiome data, enabling researchers to predict antibiotic resistance, identify novel drug targets, and design innovative treatment strategies. These computational approaches empower healthcare providers to make informed decisions, anticipate the emergence of resistance, and implement proactive measures to combat antibiotic resistance effectively. The integration of precision medicine with computational techniques opens new frontiers in the fight against antibiotic-resistant infections. By leveraging the power of data-driven insights and personalized

approaches, we can revolutionize the way we combat antibiotic resistance and preserve the efficacy of these life-saving drugs for current and future generations. Moving forward, continued research and collaborative efforts between computational scientists, clinicians, and policymakers will be instrumental in advancing precision medicine interventions and computational tools to effectively tackle antibiotic resistance. As we embrace this innovative and patient-centric approach, we stand poised to revolutionize infectious disease management and ensure a healthier and more resilient future for all.

References

1. Aslam B, Wang W, Arshad MI, Khurshid M, Muzammil S, Rasool MH, et al. Antibiotic resistance: a rundown of a global crisis. Infect Drug Resist. 2018;11:1645–58.
2. Bhandari V, Suresh A. Next-Generation Approaches Needed to Tackle Antimicrobial Resistance for the Development of Novel Therapies Against the Deadly Pathogens. Front Pharmacol. 2022;13:838092.
3. Huemer M, Mairpady Shambat S, Brugger SD, Zinkernagel AS. Antibiotic resistance and persistence—Implications for human health and treatment perspectives. EMBO Rep. 2020;21(12):e51034.
4. Serwecińska L. Antimicrobials and Antibiotic-Resistant Bacteria: A Risk to the Environment and to Public Health. Water. 2020;12(12):3313.
5. Forte C, Voinea A, Chichirau M, Yeshmagambetova G, Albrecht LM, Erfurt C, et al. Deep Learning for Identification of Acute Illness and Facial Cues of Illness. Front Med [Internet]. 2021 [cited 2022 Oct 20];8. Available from: https://www.frontiersin.org/articles/10.3389/fmed.2021.661309
6. Imchen M, Moopantakath J, Kumavath R, Barh D, Tiwari S, Ghosh P, et al. Current Trends in Experimental and Computational Approaches to Combat Antimicrobial Resistance. Front Genet. 2020;11:563975.
7. Behling AH, Wilson BC, Ho D, Virta M, O'Sullivan JM, Vatanen T. Addressing antibiotic resistance: computational answers to a biological problem? Curr Opin Microbiol. 2023;74:102305.
8. Popa SL, Pop C, Dita MO, Brata VD, Bolchis R, Czako Z, et al. Deep Learning and Antibiotic Resistance. Antibiotics. 2022;11(11):1674.
9. Watkins RR. Antibiotic stewardship in the era of precision medicine. JAC-Antimicrob Resist. 2022;4(3):dlac066.
10. Beceiro A, Tomás M, Bou G. Antimicrobial Resistance and Virulence: a Successful or Deleterious Association in the Bacterial World? Clin Microbiol Rev. 2013;26(2):185–230.
11. Ahmed S, Zhou Z, Zhou J, Chen SQ. Pharmacogenomics of Drug Metabolizing Enzymes and Transporters: Relevance to Precision Medicine. Genomics Proteomics Bioinformatics. 2016;14(5):298–313.
12. Boolchandani M, D'Souza AW, Dantas G. Sequencing-based methods and resources to study antimicrobial resistance. Nat Rev Genet. 2019;20(6):356–70.
13. Impact of antibiotics on the human microbiome and consequences for host health - PMC [Internet]. [cited 2023 Aug 4]. Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC8756738/
14. Ventola CL. Role of Pharmacogenomic Biomarkers In Predicting and Improving Drug Response. Pharm Ther. 2013;38(9):545–60.
15. Lauschke VM, Zhou Y, Ingelman-Sundberg M. Novel genetic and epigenetic factors of importance for inter-individual differences in drug disposition, response and toxicity. Pharmacol Ther. 2019;197:122–52.
16. Amiri-Dashatan N, Koushki M, Abbaszadeh HA, Rostami-Nejad M, Rezaei-Tavirani M. Proteomics Applications in Health: Biomarker and Drug Discovery and Food Industry. Iran J Pharm Res IJPR. 2018;17(4):1523–36.
17. Khodadadi E, Zeinalzadeh E, Taghizadeh S, Mehramouz B, Kamounah FS, Khodadadi E, et al. Proteomic Applications in Antimicrobial Resistance and Clinical Microbiology Studies. Infect Drug Resist. 2020;13:1785–806.
18. Durmuş S, Çakır T, Özgür A, Guthke R. A review on computational systems biology of pathogen–host interactions. Front Microbiol. 2015;6:235.

Recommended steps to reduce antibiotic resistance and new emerging treatment approaches to treat infections




Shradha Bisht[1], Poonam Parashar[1], Pragya[1]*, Mamta F Singh[2]

1 Amity Institute of Pharmacy, Amity University Uttar Pradesh, Lucknow Campus 226028
2 School of Pharmaceutical Sciences, SBS University, Balawala, Dehradun

ingenious.pragya@gmail.com

Abstract

Antibiotic resistance is a growing public health concern, and it poses a significant threat to human health. The overuse and misuse of antibiotics have contributed to the emergence of antibiotic-resistant bacteria, making it difficult to treat bacterial infections. This has led to the need for new approaches to treating infections, and researchers are exploring various options. The recommended steps to reducing antibiotic resistance include the appropriate use of antibiotics, preventing infections, and developing alternative treatments. Additionally, people can prevent infections by practicing good hygiene, getting vaccinated, and avoiding close contact with sick individuals. Emerging treatments for bacterial infections include bacteriophages, which are viruses that target specific bacteria, and antimicrobial peptides, which are naturally occurring molecules that can kill bacteria. These treatments show promise in treating antibiotic-resistant infections, and researchers are continuing to explore their effectiveness. In conclusion, reducing antibiotic resistance requires a multifaceted approach. Researchers are making progress in identifying new treatments, but it is essential to use antibiotics responsibly to preserve their effectiveness in the long term.

Keywords: Antibiotic Resistance, Appropriate Antibiotic Use, Bacteriophages, Antimicrobial Peptides, Bacterial Infections, Overuse, Misuse.

1. Introduction

Medicines known as antibiotics are used to both prevent and treat bacterial infections. In addition to saving patients' lives, antimicrobial therapies have been vital in achieving significant improvements in surgery and medicine (1). They have effectively avoided or managed infections that can develop in patients undergoing chemotherapy, dealing with long-term conditions like diabetes, end-stage renal disease, or rheumatoid arthritis, or recovering from difficult operations like organ transplants, joint replacements, or cardiac surgery (2). Antibiotics have also helped extend expected life spans by changing the outcome of bacterial infections (3).

When microorganisms like bacteria, viruses, fungi, and parasites evolve in ways that make antimicrobials ineffective, it is termed as antimicrobial resistance, also known as drug

resistance. "Superbugs" are germs that have developed resistance to most antimicrobials. One of the major dangers to food security, and global health today is antibiotic resistance (AMR). Anyone, of any age, in any nation, can become vulnerable to AMR. The abuse of antibiotics in both humans and animals is hastening the natural occurrence of AMR. As the effectiveness of the antibiotics used to treat them declines, a rising range of illnesses, including pneumonia, TB, gonorrhea, and salmonellosis, are becoming more challenging to treat. This is a major concern because a resistant infection may kill, spread to others, and impose huge costs to individuals and the society (4).

The way antibiotics are prescribed and used in the world needs to change immediately. Without a behavioural change, AMR will continue to pose a serious hazard even if new medications are formulated. Behavioural modifications must focus on improving food cleanliness, hand washing, practicing safer sex, and being vaccinated so as to stop the spread of diseases.

2. Scope of the problem

AMR is rising to dangerously high levels in all parts of the world. New resistance mechanisms are emerging and spreading globally, threatening our ability to treat common infectious diseases. A growing list of infections – such as pneumonia, tuberculosis, blood poisoning, gonorrhoea, and foodborne diseases – are becoming harder, and sometimes impossible, to treat as antibiotics become less effective (5).

With a high prevalence of diseases that are resistant to treatment, AMR poses a serious public health threat in India. According to the Indian Council of Medical Research (ICMR), there are considerable variations by region in the prevalence of AMR. AMR is more common overall in hospitals than in the general population. According to a 2019 ICMR study, more than 70% of the bacteria causing bloodstream infections in hospitals were at least partially resistant to one of the regularly prescribed antibiotics (6). Another study published in the Lancet in 2020, reported that India has the highest burden of drug-resistant tuberculosis (TB) globally, with an estimated 27% of TB cases in India being resistant to at least one major anti-TB drug. Another study also estimated that by 2050, the annual number of deaths due to AMR in India could reach 10 million (7).

This emphasizes the urgent requirement for efficient AMR prevention methods in India; including encouraging sensible antibiotic usage, enhancing infection prevention and control procedures, and stepping up surveillance and monitoring of AMR. Without urgent action, we are heading for a post-antibiotic era, in which common infections and minor injuries can once again lead to mortality.

3. Causes of the crisis in antibiotic resistance (AMR)

3.1 Excessive antibiotic use: Sir Alexander Fleming emphasized the issue of antibiotic misuse in 1945 when he foresaw that "public demand for the drug will then bring in an era of abuses." (8). Resistance can evolve on its own through mutation. Drug-sensitive competitors are eliminated by antibiotics overuse, leaving resistant microorganisms viable to proliferate due to natural selection (9). Despite warnings regarding overuse, antibiotics are overprescribed

worldwide. In many other countries, antibiotics are unregulated and available over the counter without a prescription. This lack of regulation results in antibiotics that are easily accessible, plentiful, and cheap; promoting their overuse (10).

3.2 Inappropriate Prescribing: Incorrectly prescribed antibiotics also contribute to the proliferation of resistant bacteria. Studies have shown that treatment indication, choice of agent, or duration of antibiotic therapy is incorrect in 30 - 50% of cases. The Centers for Disease Control and Prevention (CDC) estimates that at least 2 million people are infected with antibiotic-resistant bacteria each year in the United States, resulting in 23,000 deaths (11).
Studies have shown that inappropriate prescribing of antibiotics is common in India. For example, a study published in the Journal of Antimicrobial Chemotherapy in 2017 found that 52% of outpatient antibiotic prescriptions in India were inappropriate. Another study published in PLOS ONE in 2019 found that 66% of antibiotics prescribed for respiratory tract infections in a hospital in India were inappropriate(12). Low levels of antibiotics have been shown to contribute to strain diversification in organisms such as Pseudomonas aeruginosa. Subinhibitory concentrations of piperacillin and/or tazobactam have also been shown to induce broad proteomic alterations in Bacteroides fragilis (13).

3.3 Extensive Agricultural Use: Studies have demonstrated that direct interaction with livestock can result in the spread of germs that are resistant to antibiotics, which is a direct infection with resistant bacteria from an animal's source. Staphylococcus aureus germs, which are resistant to antibiotics and can infect humans, may also be present in manure.
The most typical foodborne bacteria are Campylobacter, Salmonella, E. coli, and Listeria species that alone account for over 400,000 Americans becoming sick from antibiotic-resistant infections every year. Also, dairy products and different types of meat can harbour pathogens both resistant and susceptible to antibiotics like Enterobacteriaceae (14).

3.4 Availability of Few New Antibiotics: There have been lesser attempts to produce new antibiotics recently, which can be attributed to several things.

Regulatory obstacles: Due to the high expenditures of clinical studies and the unpredictability of consumer demand, the regulatory environment for the development of antibiotics can become difficult. The extensive use of new medicines raises the possibility of AMR, which could affect market access and regulatory approval.

Economic challenges: Due to short treatment cycles and low cost, antibiotics are less profitable than other medications that are taken for a longer duration or prescribed for chronic diseases.
Public health issues: The effects of AMR on public health is a major source of concern. However, there is a lack of globally coordinated action to address this problem. Due to this, it may be challenging for businesses to give research and development of antibiotics priority (15).

3.5 The clinical and economic burden of antibiotic resistance: AMR poses a significant clinical and economic burden, both globally and within individual countries, such as India. Because infections are harder to cure when there is AMR, there may be an increase in both - morbidity and death; especially, by bacteria that are multidrug resistant, as these diseases

Table 1. CDC Assessment of Antibacterial Resistance Threats (5).

Urgent Threats
Clostridium difficile
Carbapenem-resistant Enterobacteriaceae (CRE)
Drug-resistant Neisseria gonorrhoeae
Serious Threats
Multidrug-resistant Acinetobacter
Drug-resistant Campylobacter
Fluconazole-resistant Candida (a fungus)
Extended spectrum beta-lactamase-producing Enterobacteriaceae (ESBLs)
Vancomycin-resistant Enterococci (VRE)
Multidrug-resistant Pseudomonas aeruginosa
Drug-resistant nontyphoidal Salmonella
Drug-resistant Salmonella Typhimurium
Drug-resistant Shigella
Methicillin-resistant Staphylococcus aureus (MRSA)
Drug-resistant Streptococcus pneumoniae
Drug-resistant tuberculosis
Concerning Threats
Vancomycin-resistant Staphylococcus aureus (VRSA)
Erythromycin-resistant Group A Streptococcus
Clindamycin-resistant Group B Streptococcus

may necessitate more vigorous therapy, longer hospital stays, and possibly greater mortality rates. Patients with antibiotic-resistant infections may require longer hospital stays and more intensive care, which can increase healthcare costs and pose a significant burden on patients and their families (5, 16). The assessment of antibacterial resistance AMR shown in Table 1.

4. Recommended Steps to Reduce Antibiotic Resistance

Several actions can be taken by health care providers (HCPs) and facilities to reduce AMR, according to the CDC, other groups, and experts. What follows is an explanation of each of these actions.

4.1 Adopt Antibiotic Stewardship Programs: Antibiotic stewardship involves making a commitment to use antibiotics only when needed, choosing the proper drug, and administering the medication at the appropriate dose and duration in every case (17). Successful implementation of an antibiotic stewardship program requires an interdisciplinary team, system innovation, educational intervention, and feedback provided to health care workers (18). A review of 24 studies published from 1996 to 2010 demonstrated that antibiotic stewardship programs achieved an 11% to 38% reduction in defined daily dose per 1,000 patient-days. This result included significant reductions in total antibiotic consumption, duration, and inappropriate use (19).

4.2 Optimize Therapeutic Regimens: Antibiotics are generally prescribed according to a fixed regimen that involves a specific dose, dosage frequency, and length of treatment. Recent evidence indicates that extended regimens may be unnecessary, since many clinical trials have shown that shorter courses of therapy are often just as effective as longer ones. One study showed that patients with hospital-acquired infections (HAI), including ventilator-associated pneumonia (VAP), who had received appropriate antimicrobial therapy had good clinical responses within the first six days. Results from a multicenter, randomized controlled trial of

01 patients also indicated that clinical outcomes for patients receiving appropriate empiric therapy for microscopically proven VAP for eight days were similar to those for patients who had received treatment for 15 days (18).

4.3 Improve Diagnosis and Diagnostic Tools: Perhaps the most effective way to reduce inappropriate antibiotic use is to eliminate diagnostic uncertainty. Identifying antibiotic-resistant infections can be challenging, so selection of antibiotic treatments is often empiric. In the United States (US), a recent report showed that a microbiological diagnosis was made in only 7.6% of 17,435 patients who were hospitalized with community-acquired pneumonia (CAP) (20). Multiple antimicrobials are often administered simultaneously in the hope that one will be useful in controlling an unidentified pathogen. More commonly, general practitioners may prescribe successive courses of antibiotics until an effective treatment is found. This approach can be harmful because it subjects the patient's microbiota to intense and repeated selective pressure, which encourages the development of AMR (21).

4.4 Improve Tracking Methodologies: The CDC has recently implemented the National Healthcare Safety Network (NHSN) for use by health care facilities to electronically report infections, antibiotic use, and resistance. These data allow regions, states, and facilities to identify and track antibiotic-resistant bacteria that are responsible for many HAIs. As more hospitals submit data to the NHSN database, they will be able to track antibiotic usage and bacterial resistance, enabling areas of concern to be addressed, needed improvements to be made, and successes to be shared (22).

4.5 Prevention and control: AMR is accelerated by the misuse and overuse of antibiotics, as well as poor infection prevention and control. Prevention of infection can significantly decrease resistance by eliminating the need for antibiotics in the first place. Patients are placed at risk for antibiotic-resistant infections when pathogens are transferred from one patient to other. To accomplish this goal, compliance with infection-control guidelines established by the health care facility is essential. Diligent hand hygiene before and after all patient is critical to reduce the risk of transmitting both antibiotic-susceptible and antibiotic-resistant bacterial pathogens. Disinfection of the health care environment and patient-care equipment should also be required (8). Growing concern spurs public and private initiatives, policies, and investments. In addition to adopting the recommended steps, a clearly defined, comprehensive, national action plan needs to be established to manage the AMR crisis (23).

5. New agents for the treatment of bacterial infections

There have been a few new antibiotics that have been approved and are now available for use, as well as some in development. Cefiderocol and Fosfomycin were approved for the treatment of urinary tract infections (24,25). Lefamulin and Omadacycline were approved for the treatment of CAP caused by certain gram-positive bacteria. (26,27). Teixobactin has shown activity against a range of gram-positive bacteria, including drug-resistant strains. Clinical trials are currently ongoing (28). It is important to note that while these new antibiotics offer hope in the fight against AMR, they should be used judiciously to prevent the emergence and spread of further resistance. The prevention and control of antibiotic resistance is tabulated in Table 2

Table 2. WHO: Prevention and control of AMR

Individuals
To prevent and control the spread of antibiotic resistance, individuals can:
Only use antibiotics when prescribed by a certified health professional.
Never demand antibiotics if your health worker says you don't need them.
Always follow your health worker's advice when using antibiotics.
Never share or use leftover antibiotics.
Prevent infections by regularly washing hands, preparing food hygienically, avoiding close contact with sick people, practicing safer sex, and keeping vaccinations up to date.
Prepare food hygienically, following the WHO Five Keys to Safer Food (keep clean, separate raw and cooked, cook thoroughly, keep food at safe temperatures, use safe water and raw materials) and choose foods that have been produced without the use of antibiotics for growth promotion or disease prevention in healthy animals.
Health professionals
To prevent and control the spread of antibiotic resistance, health professionals can:
Prevent infections by ensuring that your hands, instruments, and environment are clean.
Only prescribe and dispense antibiotics when they are needed, according to current guidelines.
Report antibiotic-resistant infections to surveillance teams.
Talk to the patients about how to take antibiotics correctly, AMR and the dangers of misuse.
Talk to your patients about preventing infections (for example, vaccination, hand washing, safer sex, and covering nose and mouth when sneezing).

6. New Approaches to Treating Bacterial Infections

In addition to the development of new antibiotics, there are also several new approaches to the treatment of bacterial infections that are being explored now. Here are some examples:

6.1 Bacteriophage therapy: Bacteriophages are viruses that infect and kill bacteria. Bacteriophage therapy involves using a specific phage to target and kill bacterial pathogens. This approach has shown promise in treating multidrug-resistant bacterial infections, particularly in cases where traditional antibiotics are ineffective (29).

6.2 CRISPR-Cas systems: CRISPR-Cas systems are genetic tools that can be used to selectively target and modify DNA. They are being explored as a potential treatment for bacterial infections by targeting and disabling virulence factors or antibiotic resistance AMR genes (30, 31).

6.3 Immune-based therapies: Immune-based therapies involve harnessing the immune system to target and kill bacterial pathogens. Examples include monoclonal antibodies, which can be designed to specifically target bacterial antigens, and phagocytic cells, which can be activated to engulf and kill bacteria (32, 33).

6.4 Antimicrobial peptides: Antimicrobial peptides are naturally occurring molecules that can kill bacteria by disrupting their cell membranes. They are being explored as a potential alternative to traditional antibiotics, particularly for the treatment of multidrug-resistant bacterial infections (34).

6.5 Repurposing existing drugs: There is growing interest in repurposing existing drugs for the treatment of bacterial infections. For example, some drugs that are used to treat other

diseases, such as cancer or autoimmune disorders, have been shown to have antibacterial properties (35). It is important to note that while these new approaches offer promise in the fight against AMR, they are still in the early stages of development and further research is needed to determine their safety and efficacy.

7. Conclusion

Rapidly emerging resistant bacteria threaten the extraordinary health benefits that have been achieved with antibiotics. Despite the alarming and increasing threat posed by emerging antibiotic-resistant bacteria worldwide, the implementation of recommended steps, new policies to manage the crisis, renewed research efforts to find novel agents and approaches to treating bacterial infections could dramatically reduce these risks. HCPs, researchers, policymakers, and representatives of the pharmaceutical industries have begun to come together in these ways to fight the AMR crisis. Although success will require a considerable investment of human and financial resources, the cost of not acting would likely be much greater.

When infections can no longer be treated by first-line antibiotics, more expensive medicines must be used. A longer duration of illness and treatment, often in hospitals, increases health care costs as well as the economic burden on families and societies. AMR is putting the achievements of modern medicine at risk. Organ transplantations, chemotherapy, and surgeries such as caesarean sections become much more dangerous without effective antibiotics for the prevention and treatment of infections.

References

1. Gould IM, Bal AM. New antibiotic agents in the pipeline and how they can help overcome microbial resistance. Virulence. 2013;4(2):185-91.
2. Wright GD. Something old, something new: revisiting natural products in antibiotic drug discovery. Canadian journal of microbiology. 2014;60(3):147-54.
3. Rossolini GM, Arena F, Pecile P, Pollini S. Update on the antibiotic resistance crisis. Current opinion in pharmacology. 2014;18:56-60.
4. Prestinaci F, Pezzotti P, Pantosti A. Antimicrobial resistance: a global multifaceted phenomenon. Pathogens and global health. 2015;109(7):309-18.
5. Ventola CL. The antibiotic resistance crisis: part 1: causes and threats. Pharmacy and therapeutics. 2015;40(4):277.
6. Research ICoM. Antimicrobial Research and Surveillance Network. Available from: http://iamrsn.icmr.org.in/index.php/resources/amr-icmr-data
7. Milton R, Gillespie D, Dyer C, Taiyari K, Carvalho MJ, Thomson K, et al. Neonatal sepsis and mortality in low-income and middle-income countries from a facility-based birth cohort: an international multisite prospective observational study. The Lancet Global Health. 2022;10(5):e661-e72.
8. Spellberg B, Gilbert DN. The future of antibiotics and resistance: a tribute to a career of leadership by John Bartlett. Clinical infectious diseases. 2014;59(suppl_2):S71-S5.
9. Read AF, Woods RJ. Antibiotic resistance management. Evolution, medicine, and public health. 2014;2014(1):147.
10. Michael C, Dominey-Howes D, Labbate M. The antimicrobial resistance crisis: causes, consequences, and management. Front Public Health. 2014;2:145.
11. Prevention CfDCa. Antibiotic Resistance Threats in the United States. Available from: https://www.cdc.gov/drugresistance/pdf/threats-report/2019-ar-threats-report-508.pdf.
12. Aljayyousi GF, Abdel-Rahman ME, El-Heneidy A, Kurdi R, Faisal E. Public practices on antibiotic use: A cross-sectional study among Qatar University students and their family members. PloS one. 2019;14(11):e0225499.
13. 13. Viswanathan V. Off-label abuse of antibiotics by bacteria. Gut microbes. 2014;5(1):3-4.
14. Zaheer R, Cook SR, Barbieri R, Goji N, Cameron A, Petkau A, et al. Surveillance of Enterococcus spp. reveals distinct species and antimicrobial resistance diversity across a One-Health continuum. Scientific reports. 2020;10(1):3937.
15. Organization WH. Antibacterial agents in clinical development: an analysis of the antibacterial clinical development pipeline, including tuberculosis. World Health Organization, 2017.
16. Dadgostar P. Antimicrobial resistance: implications and costs. Infection and drug resistance. 2019:3903-10.
17. Solomon SL, Oliver KB. Antibiotic resistance threats in the United States: stepping back from the brink. American family physician. 2014;89(12):938-41.
18. Luyt C-E, Bréchot N, Trouillet J-L, Chastre J. Antibiotic stewardship in the intensive care unit. Critical care. 2014;18:1-12.
19. Bartlett JG, Gilbert DN, Spellberg B. Seven ways to preserve the miracle of antibiotics. Clinical Infectious Diseases. 2013;56(10):1445-50.
20. Nature E. The antibiotic alarm. Nature. 2013;495(7440):141.
21. Michael CA, Dominey-Howes D, Labbate M. The antimicrobial resistance crisis: causes, consequences, and management. Frontiers in public health. 2014;2:145.
22. Centers for Disease Control and Prevention OoID. Antibiotic resistance threats in the United States. Available from: http://www.cdc.gov/drugresistance/ threat-report-2013. .

23. Tavernise S, Shear M. Obama seeks to double funding to fight antibiotic resistance. The New York Times. 2015.
24. Wu JY, Srinivas P, Pogue JM. Cefiderocol: a novel agent for the management of multidrug-resistant Gram-negative organisms. Infectious diseases and therapy. 2020;9:17-40.
25. Gardiner BJ, Stewardson AJ, Abbott IJ, Peleg AY. Nitrofurantoin and fosfomycin for resistant urinary tract infections: old drugs for emerging problems. Australian prescriber. 2019;42(1):14.
26. FDA U. FDA approves new antibiotic to treat community-acquired bacterial pneumonia. Available from:https://www.fda.gov/news-events/press-announcements/fda-approves-new-antibiotic-treat-community-acquired-bacterial-pneumonia.
27. Durães F, Sousa E. Omadacycline: a newly approved antibacterial from the class of tetracyclines. Pharmaceuticals. 2019;12(2):63.
28. Kali A. Teixobactin: A novel antibiotic in treatment of gram positive bacterial infections. Journal of Clinical and Diagnostic Research: JCDR. 2015;9(3):DL01.
29. Chan BK, Abedon ST, Loc-Carrillo C. Phage cocktails and the future of phage therapy. Future microbiology. 2013;8(6):769-83.
30. Bikard D, Barrangou R. Using CRISPR-Cas systems as antimicrobials. Current opinion in Microbiology. 2017;37:155-60.
31. Citorik RJ, Mimee M, Lu TK. Sequence-specific antimicrobials using efficiently delivered RNA-guided nucleases. Nature biotechnology. 2014;32(11):1141-5.
32. de Kraker ME, Stewardson AJ, Harbarth S. Will 10 million people die a year due to antimicrobial resistance by 2050? PLoS medicine. 2016;13(11):e1002184.
33. Diago-Navarro E, Calatayud-Baselga I, Sun D, Khairallah C, Mann I, Ulacia-Hernando A, et al. Antibody-based immunotherapy to treat and prevent infection with hypervirulent Klebsiella pneumoniae. Clinical and Vaccine Immunology. 2017;24(1):e00456-16.
34. Wang S, Zeng X, Yang Q, Qiao S. Antimicrobial peptides as potential alternatives to antibiotics in food animal industry. International journal of molecular sciences. 2016;17(5):603.
35. Silver LL. Challenges of antibacterial discovery. Clinical microbiology reviews. 2011;24(1):71-109.

Fun & frolic - Wordsearch

Solution on page 96

F A F F E J G A P B U M Y E P P J T G Q
C G O K I W F C L F R Y E U X I A B F I
H S U L F O N A M I D E X J N X F X M E
D O E Z M Z E R O H P O R E D I S U C K
P T Q E G Z W P Q J B B G B R L A N D U
Y H K F H I Y I M L A Q J B I T R I T V
V I E U K Y Z M T G H J F T B E F Q B B
P W G N J P J N J G Y E K P O T V R C B
N M I R P O H T E M I R T I S R M N H D
C M G M H J B B D Y H R O M O A L N T L
Q U U A L T L W I B O B X N M C D I V G
A E L H X I V T T F H P F I E Y A H P P
T C V G C V E Y P E U U I A Y C P H Y X
N V P E G A Z O E G G I S S W L T A X W
A J A O K V O S P K B Y X X M I O A Q E
Y Y F U A X C O O J M W R C Z N M V B B
Y V E L Y T Q U C E L X B R H E Y Y P I
Z U X Z F Q A Y Y N I R I N Q X C O D H
B O V F E S T Y L H L B M P H E I M W S
U M C L S B R W G S D E E P F X N Z J A

RIBOSOME
SULFONAMIDE
SIDEROPHORE
GLYCOPEPTIDE
DAPTOMYCIN
TRIMETHOPRIM
TETRACYCLINE

Advanced and innovative therapeutic strategies for unveiling the battle of antibiotic resistance



Srishti Shetty, Chandani Muleva, Saritha Shetty*

Shobhaben Pratapbhai Patel School of Pharmacy & Technology Management, SVKM'S NMIMS (Deemed to be University), Mumbai 400056, India

shettyrsaritha@gmail.com

1. Introduction to Antibiotic Resistance

1.1 A Brief History

Antibiotics (AB), the term coined in the year 1941 has come a long way as an effective pharmacological agent in the history of integrative medicine. Antibiotic resistance is quite common which was observed for S.aureus within 5 years after introduction of first antibiotic i.e., penicillin in 1943. The truth is, the usage of AB has eventually resulted in resistance over time and are now rendered obsolete. Bacterial infection is tied to a large part of morbidity and mortality rates in medical practice and antibiotics have proved to be a boon resolving this problem. However, antibiotic resistance (ABR) has now become a massive issue with social, economic and health repercussions (1). Figure 1 depicts the mechanisms exploited by bacteria in order to tackle the action of antibiotic agents. The statistic claims that by 2050 the AMR population could reach over 10 million people thus exceedingly around 8.2 million lives which is currently accounted to cancer (2,3). This newsletter focuses on the therapeutic approaches and extensive attempts that are made to overcome for antibiotic resistance. Figure 2 depicts the underutilised tools such antibodies, stem cells, vaccines, immunomodulators and nanomedicines against antibiotic resistant.

2. Therapeutic approaches for combating antibiotic resistance

2.1 Development of new Antibiotics (Abs) with multi-effective and multidrug targets:

The ABR studies have shown that resistance to antibiotics that only target one protein emerges too quickly, shifting the concern towards development of antibiotics having multiple targets to act upon the drug candidates. The pharmacology of the drug generally speaks of its ability to interact with the multiple targets giving rise to two main categories, classified as multi effective and multi-functional. When a drug is associated with a single target but is an integral part of other process that depends on its inhibition, it is said to be multi effective. Whereas multifunctional agents have an extra antibacterial action in addition to their antibiotic target (4-6). Table 1 gives an overview of the antibiotics with their targets.

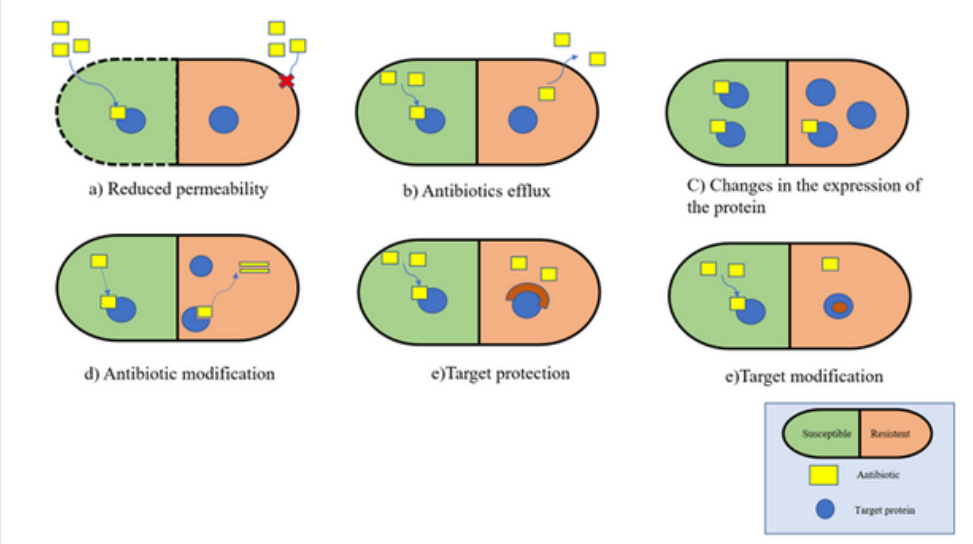


Figure 1: A graphical illustration of the mechanism of Antibiotic Resistance

2.2 Exploring nanomedicine as a therapeutic approach against AMB

Nanomedicine display characteristics feature of enhanced physicochemical property, prolongation of action and increased stability (14). For instance, liposomes were explored for its activity against intracellular strains where anti tubercular drugs were loaded within liposomes with tiny fabrications of ribonucleic acid. This system showed controlled release of drug with maximum concentration at the targeted site (15). Different liposomal types such as conventional liposomes, Fusogenic liposomes and surface modified liposomes (16-19).The most sophisticated variety of nanostructured lipid carriers are made of unstructured matrix with a blend of liquid oil matrix that reduces the rate of drug escape (20). Various lipids such as glyceryl monostearate, stearic acid, glyceryl behenate, tripalmitin are explored as a remedy to dela with antibiotic resistance (21-23). Nanosystems have notably prevented the promotion of resistance from bacteria by improving the therapeutic activity. Certain strategies that entail drug breakdown by lactamase, efflux pumps or thickening of bacterial cell walls involving nanosystems are explored widely as a therapeutic approach (24).

2.3 Immunomodulators as an alternative therapeutic approach against antibiotic resistance pathogens

Immunomodulators such as Host defence peptides (HDPs) having a broad-spectrum antimicrobial activity have been suitably used nowadays as a potential therapeutic agent to treat antibiotic resistant diseases. HDPs are component of innate immune system that comprises of three main classes such as linear α helical assembly excluding the disulfide bonds such as cathelicidins, magainins and cecropins, disulphide bridge stabilized β sheet assembly containing defensins and lastly a peptide in loop form. A self-promoted uptake mechanism explained by the pore forming ability of HDPs in the biological membrane demonstrates the cytotoxic functional abilities of such HDPs while some protection to cytotoxicity of HDPs is generally offered by the anionic phospholipids present in the mammalian membranes (25, 26).

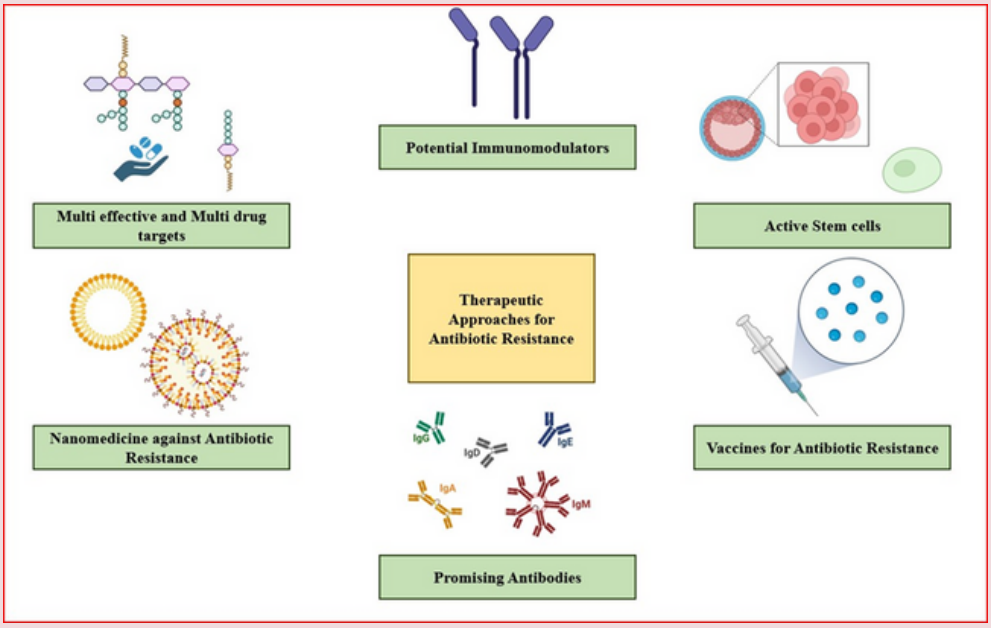


Figure 2. Therapeutic approaches as an alternative to tackle antibiotic resistance

Table 1. Summary of antibiotics with their mechanism of action to overcome ABR

S. No.	Antibiotic	Target	Mechanism of action	Ref
1.	Daptomycin	Phosphatidylglycerol, fluid lipid domains	Multi-effective- Inhibits the cell wall synthesis machinery by binding phosphatidylglycerol and then intruding into the fluid domains.	(7)
2.	Vancomycin	lipid II	Multi-effective Targets lipid II but also prevents formation of peptidoglycan and also prevents the creation of wall teichoic acid.	(8)
3.	Bacitracin	bactoprenol pyrophosphate	Multi-effective role ultimately results in inhibition of peptidoglycan and teichoic acid.	(9)
4.	Nitrofurantoin	cellular macromolecules	Multi-effective damages cellular macromolecules including DNA and membrane lipids	(10)
5.	Dapsone	dihydropteroate synthase	Multifunctional with anti-inflammatory and immunomodulatory activity	(11)
6.	Macrolides	50S rRNA	Depicts both anti-inflammatory and immunomodulatory multifunctional activity.	(12)
7.	Rifampicin	DNA-dependent RNA polymerase	anti-inflammatory and immunomodulatory	(13)

2.5 Stem cell in antibiotic resistance

Mesenchymal stem cells (MSCs) through production of antimicrobial factor appears to be a great non-antibiotic strategy for bacterial resistance. MSCs is proven to be immunomodulatory in nature and through this property it secrets immunomodulatory factors such as transforming growth factor-β, prostaglandin E2 and various antimicrobial peptides such as LL-37. Further it was seen and reported that antimicrobial peptide secreted by MSCs such as cathelicidin lL-37, hepcidin lipocalin etc indirectly led to inhibition of methicillin resistant S.aureus (30). Table 3 gives an overview of the mechanism of stem cell in antibiotic resistance.

2.6 Vaccines, particularly against antibiotic resistance bacteria (ARBs)

Prophylactic agents such as vaccines that have low probability of developing resistance and multitargeting ability proves to be the promising alternatives to the existing antibiotic treatment (33). Multiple immunogenic epitopes contained by vaccines as compared to a single targeting ability of antibiotics makes them superior in terms of developing resistance (34). The vaccine introduced against S. pneumoniae, and Neisseria meningitidis showed a complete elimination of antimicrobial resistance against these strains. Vaccines directly blocks the transmission of resistant strains of bacteria and decreases the usage of antibiotic and risk associated with acquiring and transmitting resistance (3, 35). Table 4 tabulates the advantages and disadvantages of various therapeutic approaches studied by the various scientists in combating the antibiotic resistance.

Table 3. Mechanism of stem cell in antibiotic resistance

Therapeutic approach	Mechanism	Target	Ref
Secretome from Mesenchymal stem cells (MSCs)	Inhibition of bacterial cell wall synthesis.	Antibiotic resistant Methicillin resistant *S.aureus* (MRSA)	(31, 32)
MSCs derived from human bone marrow and umbilical cord such as lipocal, hepcidin and LL-37	Act via a cationic antimicrobial peptide namely LL-37 expressed in the peroxidase-negative granules of neutrophils.	*E. coli*, *S. aureus* and *L. pneumonia* and imipenem resistant *P. aeruginosa* in human infants	(31-33)

Table 4. Advantages and disadvantages of various therapeutic approaches in antibiotic resistance

Therapeutic approach	Advantage	Disadvantage	Ref
Host Defence peptides	Dual action: Direct killing of bacteria + Immunomodulatory action Multiple complementary actions responsible for minimal resistance	Prone to degradation by the protease enzyme due to presence of L-amino acids in their structure. Less bioavailability and faster elimination due to their small size. Systemic cytotoxicity towards host cells	(36)
Monoclonal antibodies	Attractive approach in immunocompromised or elderly population which do not response to vaccines Additive action shown by antibiotic and antibodies when co-administered simultaneously Specific and selectively target a pathogen by blocking the pathogen-host, protein-receptor interaction.	Recognizes a single target Large protein structure that limits the conformational binding of MAbs Expensive, less cost effective	(28)
Stem cells	Abundant in the human body and can be easily harvested	Risk of undesired immune responses, reaction at the administration site along with fear of disease transmission among the few	(32)
Vaccines	Highly specific Efficacious in immunocompromised patient through herd immunity High longevity as it has very less or no chance of developing resistance	Vaccine takes longer time i.e., few days or weeks to show its protective action compared to antibiotics Absence of animal models that are representative of human disease to test efficacy of vaccines.	(36, 37)

4. Conclusion

Antibiotic resistance is a complex issue posing significant threat to public health and has eventually resulted the easily treatable infections to a more complex and challenging stage. Therapeutic approaches overcoming antibiotic resistance emphasizes the importance of expanding the arsenal of treatment options for such infections along with rational prescription which is a recommended standard for thorough understanding of pharmacology and therapeutics. Exploring the multi-effective roles of antibiotics, nanosystem carriers entailing drug breakdown, immunomodulators expanding the horizon of antibiotic activity and synergistically acting monoclonal antibodies approaches in future will definitely be considered as a potent option to curtail the resistant factor. Thus, we conclude that, campaign with multidisciplinary strategy and constant oversighting of approaches for the therapeutic management of ABR is a possible remedy to battle antibiotic resistance. The clinical studies undergoing on these approaches will bring a new era in the treatment of resistant diseases.

Abbreviations

Antibiotic resistance (ABR)
Antibiotics (AB)
Host defence peptides (HDPs)
Monoclonal antibodies (MAbs)
Mesenchymal stem cells (MSCs)
Methicillin resistant S.aureus (MRSA)

References

1. Dhingra S, Rahman NAA, Peile E, Rahman M, Sartelli M, et al. Microbial Resistance Movements: An Overview of Global Public Health Threats Posed by Antimicrobial Resistance, and How Best to Counter. Frontiers in Public Health. 2020;8.
2. Willyard C. The drug-resistant bacteria that pose the greatest health threats. Nature. 2017;543(7643):15.
3. Jansen KU, Anderson AS. The role of vaccines in fighting antimicrobial resistance (AMR). Human Vaccines & Immunotherapeutics. 2018;14(9):2142-2149.
4.Makhoba XH, Viegas Jr C, Mosa RA, Viegas FPD, Pooe OJ. Potential Impact of the Multi-Target Drug Approach in the Treatment of Some Complex Diseases. Drug Design, Development and Therapy. 2020;14:3235-3249.
5. Ma Y, Frutos-Beltrán E, Kang D, Pannecouque C, De Clercq E et al. Medicinal chemistry strategies for discovering antivirals effective against drug-resistant viruses. Chemical Society Reviews. 2021;50(7):4514-4540.
6. Gray DA, Wenzel M. Multitarget Approaches against Multiresistant Superbugs. ACS Infectious Diseases. 2020;6(6):1346-1365.
7. Müller A, Wenzel M, Strahl H, Grein F, Saaki TNV et al. Daptomycin inhibits cell envelope synthesis by interfering with fluid membrane microdomains. Proceedings of the National Academy of Sciences. 2016;113(45):E7077-E7086.
8. Reynolds PE. Structure, biochemistry and mechanism of action of glycopeptide antibiotics. European Journal of Clinical Microbiology and Infectious Diseases. 1989;8(11):943-950.
9.Stone KJ, Strominger JL. Mechanism of Action of Bacitracin: Complexation with Metal Ion and C55-Isoprenyl Pyrophosphate. Proceedings of the National Academy of Sciences. 1971;68(12):3223-3227.
10. Wenzel M, Dekker MP, Wang B, Burggraaf MJ, Bitter W, et al. New flat embedding method for transmission electron microscopy reveals an unknown mechanism of tetracycline. BioRxiv. 2019;820191.
11. Tan BB, Lear JT, Smoth AG. Acne fulminans and erythema nodosum during isotretinoin therapy responding to dapsone. Clinical and Experimental Dermatology. 1997;22(1):26-27.
12. López-Boado YS, Rubin BK. Macrolides as immunomodulatory medications for the therapy of chronic lung diseases. Current Opinion in Pharmacology. 2018;8(3):286-291.
13.Mendonça CO, Griffiths CEM. Clindamycin and rifampicin combination therapy for hidradenitis suppurativa. British Journal of Dermatology. 2006;154(5):977-978.
14.Patra JK, Das G, Fraceto LF, Campos EVR, Rodriguez-Torres M del P, et al. Nano based drug delivery systems: recent developments and future prospects. Journal of Nanobiotechnology. 2018;16(1):71.
15.Niu NK, Yin JJ, Yang YX, Wang ZL, Zhou ZW, et al. Novel targeting of PEGylated liposomes for codelivery of TGF-β1 siRNA and four antitubercular drugs to human macrophages for the treatment of mycobacterial infection: a quantitative proteomic study. Drug Design, Development and Therapy. 2015;9:4441-4470.
16. Castoldi A, Herr C, Niederstraßer J, Labouta HI, Melero A, Gordon S, Schneider-Daum N, Bals R, Lehr CM. Calcifediol-loaded liposomes for local treatment of pulmonary bacterial infections. European Journal of Pharmaceutics and Biopharmaceutics. 2017;118:62-67.
17. Jadhav M, Kalhapure RS, Rambharose S, Mocktar C, Singh S, et al. Novel lipids with three C18-fatty acid chains and an amino acid head group for pH-responsive and sustained antibiotic delivery. Chemistry and Physics of Lipids. 2018;212:12-25.

18.Nicolosi D, Cupri S, Genovese C, Tempera G, Mattina R, et al. Nanotechnology approaches for antibacterial drug delivery: Preparation and microbiological evaluation of fusogenic liposomes carrying fusidic acid. International Journal of Antimicrobial Agents. 2015;45(6):622-626.
19. Cavalcanti IMF, Pontes-Neto JG, Kocerginsky PO, Bezerra-Neto AM, Lima JLC, et al. Antimicrobial activity of β-lapachone encapsulated into liposomes against meticillin-resistant Staphylococcus aureus and Cryptococcus neoformans clinical strains. Journal of Global Antimicrobial Resistance. 2015;3(2):103-108.
20.Salvi VR, Pawar P. Nanostructured lipid carriers (NLC) system: A novel drug targeting carrier. Journal of Drug Delivery Science and Technology. 2019;51:255-267.
21. Fazly Bazzaz BS, Khameneh B, Zarei H, Golmohammadzadeh, S. Antibacterial efficacy of rifampin loaded solid lipid nanoparticles against Staphylococcus epidermidis biofilm. Microbial Pathogenesis. 2016;93:137-144.
22. Baig MS, Ahad A, Aslam M, Imam SS, Aqil Mohd, Ali A. Application of Box–Behnken design for preparation of levofloxacin-loaded stearic acid solid lipid nanoparticles for ocular delivery: Optimization, in vitro release, ocular tolerance, and antibacterial activity. International Journal of Biological Macromolecules. 2016;85:258-270.
23.Öztürk AA, Aygül A, Şenel B. Influence of glyceryl behenate, tripalmitin and stearic acid on the properties of clarithromycin incorporated solid lipid nanoparticles (SLNs): Formulation, characterization, antibacterial activity and cytotoxicity. Journal of Drug Delivery Science and Technology. 2019;54:101240.
24. Baptista PV, McCusker MP, Carvalho A, Ferreira DA, Mohan NM, Martins M, Fernandes AR. Nano-Strategies to Fight Multidrug Resistant Bacteria—"A Battle of the Titans." Frontiers in Microbiology. 2018:9.
25.Steinstraesser L, Kraneburg UM, Hirsch T, Kesting M, Steinau HU et al. Host Defense Peptides as Effector Molecules of the Innate Immune Response: A Sledgehammer for Drug Resistance? In International Journal of Molecular Sciences. 2009;10(9):3951-3970.
26.Nijnik A, Hancock REW. Host defence peptides: antimicrobial and immunomodulatory activity and potential applications for tackling antibiotic-resistant infections. Emerging Health Threats Journal. 2019;2(1):7078.
27.Motley MP, Fries BC. A New Take on an Old Remedy: Generating Antibodies against Multidrug-Resistant Gram-Negative Bacteria in a Postantibiotic World. MSphere. 2017;2(5):10.
28. Wagner EK, Maynard JA. Engineering therapeutic antibodies to combat infectious diseases. Current Opinion in Chemical Engineering. 2018;19:131-141.
29. Idso MN, Akhade AS, Arrieta-Ortiz ML, Lai BT, Srinivas V, et al. Antibody-recruiting protein-catalyzed capture agents to combat antibiotic-resistant bacteria. Chemical Science. 2020;11(11):3054-3067.
30. Battah, B. Mesenchymal Stem Cells: Potential Role against Bacterial Infection. Journal of Biosciences and Medicines. 2022;10(03):97-113.
31. Kumar M, Sarma DK, Shubham S, Kumawat M, Verma V, et al. Futuristic Non-antibiotic Therapies to Combat Antibiotic Resistance: A Review. Frontiers in Microbiology. 2021;12.
32. Wu X, Jin S, Ding C, Wang Y, He D, Liu Y. Mesenchymal Stem Cell-Derived Exosome Therapy of Microbial Diseases: From Bench to Bed. Frontiers in Microbiology. 2022;12.
33. Yagi H, Chen AF, Hirsch D, Rothenberg AC, Tan J, Alexander PG, et al. Antimicrobial activity of mesenchymal stem cells against Staphylococcus aureus. Stem Cell Res Ther. 2020;11(1):293.
34. Costanzo V, Roviello GN. The Potential Role of Vaccines in Preventing Antimicrobial Resistance (AMR): An Update and Future Perspectives. Vaccines. 2023;11(2).
35.Micoli F, Bagnoli F, Rappuoli R, Serruto D. The role of vaccines in combatting antimicrobial resistance. Nature Reviews Microbiology. 2021;19(5):287-302.
36.Rosini R, Nicchi S, Pizza M, Rappuoli R. Vaccines Against Antimicrobial Resistance. Frontiers in Immunology. 2020;11:1048.
37. Drayton M, Deisinger JP, Ludwig KC, Raheem N, Müller A, et al. Host Defense Peptides: Dual Antimicrobial and Immunomodulatory Action. In International Journal of Molecular Sciences. 2021;22(20).

Combating antibiotic resistance: a major challenge

Leena Sawaikar, Celina Nazareth*, Swati Keny

PES's Rajaram and Tarabai Bandekar College of Pharmacy, Farmagudi, Ponda, Goa, India - 403401.
celinanaz@yahoo.com

1.Introduction

Antibiotics are microbial metabolites or synthetic analogues inspired by them that in small doses inhibit the growth and survival of microorganisms without serious toxicity to the host (1). These drugs have revolutionized medicine and made our modern way of life possible. Antibiotics target and inhibit essential cellular processes, retarding growth and causing cell death. Antibiotics are essential for preventing and treating infectious diseases (2). From the time of discovery of the first antibiotic, the challenge of antibiotic resistance commenced. Antibiotics use different mechanisms against bacteria to prevent their pathogenesis and they can be classified as bactericidal or bacteriostatic. At the same time, bacteria are also using methods to overcome the effectivity of the antibiotics by using distinct types of mechanisms. This ability of microorganisms to survive and be viable under the influence of antimicrobial agents is called as antibiotic resistance (3). Knowing the mechanism by which the organisms develop this resistance helps to overcome the resistance and also to formulate synthetic antimicrobials to overcome the current mechanism of resistance. However, if bacteria are exposed to drugs below the dose required to kill all bacteria in a population (the minimum bactericidal concentration or MBC), they can mutate and resist antibiotic treatment via natural selection for resistance-conferring mutations. These genetic mutations can arise from the adoption of a plasmid encoding a resistance gene or by mutation to the bacterial chromosome itself.

2.Mechanism of resistance

A common mechanism used by bacteria to minimize the effects of antibiotics is to acquire or increase the expression of drug efflux pumps. Bacteria use pumps in the cell wall to expel the antibiotic. And these pumps expel drugs from the cytoplasm, limiting their ability to access their target (4).

Enzymatic inactivation: An existing bacterial enzyme is modified to interact with an antibiotic in order to make them inactive towards bacteria. It is due to the transfer of the antibiotic resistance gene carried on plasmids. The most significant examples are beta-lactamase enzymes, which hydrolyse beta-lactams (penicillins, cephalosporins).

Decreased uptake by changes in the outer membrane permeability or by presence of porins: These variations interfere with the entrance of antibiotics.
Modification of the drug target: These changes impede the binding of the antibiotic and limit its potency (5).

3.Clinical causes of antibiotic resistance (6)

The main origin of resistance to antibiotics is their misuse. An example is unnecessary prescription of antibiotics for viral infections, against which they are ineffective.
In most of the low income countries most of the antibiotics are of secondary quality and available over the counter. Wherever there is insufficient enforcement of regulatory policies on prescribing medicine, over-the-counter antibiotics are prevalent. Such availability makes it accessible for patients to do self-treatment for diseases that do not necessarily need antibiotics for treatment.

Antibiotic resistance can develop because physicians unnecessarily prescribe lengthy courses of antibiotics. Another factor contributing to overprescribing antibiotics by providers is patient's expectations from the clinicians. It is seen that clinicians consider the perceived patient request for antibiotics as one of the major barriers to adhere to standard guidelines for antibiotic prescriptions and hence to avoid the dissatisfaction of their patients prescribe antibiotics.

The usage of antibiotics in agriculture is one of the significant factors in developing antimicrobial resistance in humans. Antibiotics are added to animal feed and drinking water to cure sick animals and to prevent illness (prophylaxis) in healthy animals and in animal farming worldwide to promote the growth of livestock, particularly colistin, a critical last-line antibiotic to treat severe infections in humans (7).

Inappropriate prescribing patterns which include prescription of broad spectrum antibiotics in place of a better targeted antibiotic.

Improper use by the patient with respect to dosage or duration of the treatment which makes some of the bacteria survive and become resistant.
Development of superbugs which can adapt to the medicines that are intended to kill them and fight back against them. These multiply and cause infections despite treatment with different antibiotics. Some infections with superbugs include gonorrhoea, MRSA and tuberculosis (8).

4. Consequences of antibiotic resistance

Infections caused by resistant bacterial strains can cause more severe infections which may be fatal or difficult to treat as compared with similar infections caused by susceptible strains. Antibiotic resistant infections cause economic and health burden to the nation. When first-line and then second-line antibiotic treatment fail, more antibiotics which are more toxic and expensive have to be administered. Patients with resistant infections require significantly longer hospital stays, more doctor's visits, and may experience a higher incidence of long-term disability (9).

Finally when antibiotics don't work, it can lead to

- Illnesses lasting for a longer time with complications.
- Frequents visits to the doctor
- Usage of more potent and expensive medications
- Increased bacterial infection related mortality.

5.Bacteria resistant to antibiotics (10)

Antibiotics which were earlier used to treat a particular infection have become ineffective against a particular bacteria. The development of bacterial resistance by some strains of bacteria to most of the easily available antibiotic has created a major issue in the field of medicine. Methicillin-resistant Staphylococcus aureus (MRSA), vancomycin-resistant Enterococcus (VRE), multi-drug-resistant Mycobacterium tuberculosis (MDR-TB) and carbapenem-resistant Enterobacteriaceae (CRE) gut bacteria are some of these which causes serious disease and a major public health problem.

6. Strategies to combat antibiotic resistance (11)

6.1. Measures to be taken by the patients

Medications should be taken as per the doctor's prescription the medication as prescribed by your doctor. Treatment should not be stopped even if the patient is feeling better. If the treatment is stopped soon, the patient may fall sick again and bacteria may become resistant to the antibiotic taken.

No skipping of dose is allowed as the medicine is effective only if the blood levels of the same are maintained.

An antibiotic is meant for a particular infection at the time, hence using the leftover antibiotic is not appropriate. By taking the incorrect medication, receiving the right care on time is delayed and thus the illness gets worse.

Antibiotics prescribed for someone else should not be taken as these may not be appropriate for your illness so this can delay or worsen the treatment. If any new or unusual symptoms or side effects are observed it should be informed to the physician so that the trouble causing antibiotic can be stopped and the treatment be completed with a different antibiotic (12-13).

6.2.Measures to be taken by physicians

Prescribing a short-course antibiotic therapy: Recent randomized controlled trials show that shorter courses of antibiotic therapy are as effective as longer courses, with the added benefit of reducing the exposure of patients to antibiotics. This is because most of the signs and symptoms of bacterial infections result from the inflammatory response to the bacteria rather than the direct presence of viable bacteria (14).

Antibiotic stewardship programmes have to be conducted. These ensure that each patient is given best care for their specific condition and gets an antibiotic only when necessary. Effective sanitation, hygiene and infection prevention measures ensure that incidence of infection is reduced. Clinical guidelines to be followed. Use delayed antibiotic prescriptions (15-16).

6.3. National strategies (17)

Establishment of national committee to monitor impact of antibiotic resistance and provide intersectoral co-ordination is required. Establishing and implementing national standard treatment guidelines, having essential drug list (EDL), enhancing coverage of immunization are other essential strategies desired at national level. A national policy for containment of antimicrobial resistance (AMR) was introduced in 2011. The policy aims to understand emergence, spread and factors influencing resistance, to setup antimicrobial program, to rationalize the use of antimicrobials and to encourage the innovation of newer effective antimicrobials. Other than this, it also aims establishing antibiotic resistance surveillance system, strengthening infection prevention and control measures and educate, train and motivate all stake holders in rational use of antibiotics.

7.Conclusion

Antibiotic resistance is a serious issue with a wide range of causes. It is major cause of health concerns adding cost to oneself and to the community, directly or indirectly. The best way to combat it is prevention so that the spread of infection is reduced. The need of the hour is to develop newer antibiotics and sensible usage of presently available antibiotics. To combat the globally growing antibiotic resistance, patients, prescribers, and individuals must work together with international regulators and policy makers.

References

1. Williams DA, Foye WO, Lemke TL. Foye's principles of medicinal chemistry. 5th ed. Philadelphia, PA: Lippincott Williams and Wilkins; 2002.
2. Habboush Y, Guzman N. Antibiotic Resistance. StatPearls Treasure Island. 2022.
3. Abushaheen MA, Muzaheed, Fatani AJ, Alosaimi M, Mansy W, et al. Antimicrobial resistance, mechanisms and its clinical significance. Dis Mon. 2020;66(6):100971.
4. Richardson LA. Understanding and overcoming antibiotic resistance. PLoS Biol. 2017 Aug 23;15(8):e2003775.
5. Annunziato. Strategies to overcome antimicrobial resistance making use of non-essential target inhibitors: A review. International Journal of Molecular Sciences. 2019:20(23):5844.
6. Aslam B, Wang W, Arshad MI, Khurshid M, Muzammil S, Rasool MH. Antibiotic resistance: a Rundown of a Global Crisis. Infection and Drug Resistance. 2018;11(11):1645-58.
7.Antibiotic Resistance: What Is It, Complications & Treatment. Cleveland Clinic; 2021. Available from: https://my.clevelandclinic.org/health/articles/21655-antibiotic-resistance
8. Dadgostar P. Antimicrobial resistance: Implications and costs. Infect Drug Resist. 2019;12:3903-10.
9. Sifri Z, Chokshi A, Cennimo D, Horng H. Global contributors to antibiotic resistance. Journal of Global Infectious Disease. 2019:11(1):36.
10. Ventola CL. The antibiotic resistance crisis: part 1: causes and threats. Pharmacy and therapeutics.2015;40(4):277–83.
11.Antibiotic resistant bacteria [Internet]. Gov.au. [cited 2023 Aug 2]. Available from: https://www.betterhealth.vic.gov.au/health/conditionsandtreatments/antibiotic-resistant-bacteria
12. Wall S. Prevention of antibiotic resistance - an epidemiological scoping review to identify research categories and knowledge gaps. Glob Health Action. 2019 [cited 2023 Aug 2];12(1):1756191. Available from: http://dx.doi.org/10.1080/16549716.2020.1756191
13. Combating antibiotic resistance [Internet]. U.S. Food and Drug Administration. FDA; 2020 [cited 2023 Aug 2]. Available from: https://www.fda.gov/consumers/consumer-updates/combating-antibiotic-resistance
14. Nathan C. Resisting antimicrobial resistance. Nature Reviews Microbiology. 2020;18(5):259-60.
15. Spellberg B. The maturing antibiotic mantra: "shorter is still better." Journal of Hospital Medicine. 2018;13(5):361-2.
16. Fda.gov. [cited 2023 Aug 2]. Available from: https://www.fda.gov/consumers/consumer-updates/combating-antibiotic-resistance.
17. Nfid.org. [cited 2023 Aug 2]. Available from: https://www.nfid.org/antibiotic-resistance/antibiotic-stewardship-pledg

Approaches in the fight against Antibiotics resistance: Harnessing technology and biotechnology



Vaishnavi Chhabra[1], Reshu Virmani[1], Kamla Pathak[2]*

1School of Pharmaceutical Sciences, MVN University, Palwal, Haryana, India
2Faculty of Pharmacy, Uttar Pradesh University of Medical Sciences, Saifai, Etawah, Uttar Pradesh, India- 206130

kamlapathak5@gmail.com

1. Introduction

Since their inception, antibiotics have been regarded as one of the most important discoveries of the twentieth century, and their widespread usage has transformed healthcare. Between 1950 and 1970, known as the "golden age" of antibacterial medication discovery, empirical screening of microbial natural product fermentation provided the majority of antibacterial classes currently utilized for infection treatment. For the last 30 years, there has been a discovery gap in antibacterial drugs, with no new classes of antibacterials released to the market until 2000, when linezolid, an oxazolidinone, was approved. Inevitably, the advent of antibiotics coincided with the emergence of the phenomena of antimicrobial resistance. Based on his early results, Fleming, in 1954 predicted that indiscriminate application of this discovery would result in the selection and development of antibiotic-resistant bacterium mutants. Indeed, after only a few years of the golden age of antimicrobials, frightening signs of resistance were noticed (1).

1.1 Understanding the antibiotic resistance

The term "resistance" is a natural biological process whereby bacteria evolve and acquire genetic mutations or exchange resistance genes, enabling them to withstand the effects of antibiotics. The timeline of antibiotic resistance are depicted in Figure 1. Resistance can be divided into two groups: intrinsic or acquired resistance. Some bacterial species are innately resistant to a specific antibiotic class. Acquired resistance occurs when just select strains of a particular species are resistant to an antibiotic, rather than the entire species. This resistance can occur as a result of a spontaneous mutation in the chromosomal DNA or as an extra-chromosomal event, such as when bacteria exchange plasmids or transposons (2). There is surge in AMR, which is frequently directed towards multidrug resistance (MDR). MDR infections are difficult to treat with conventional medications. As per some studies, it is anticipated that more patients my die from MDR pathogen infections (10 million/year) than from cancer (8.2

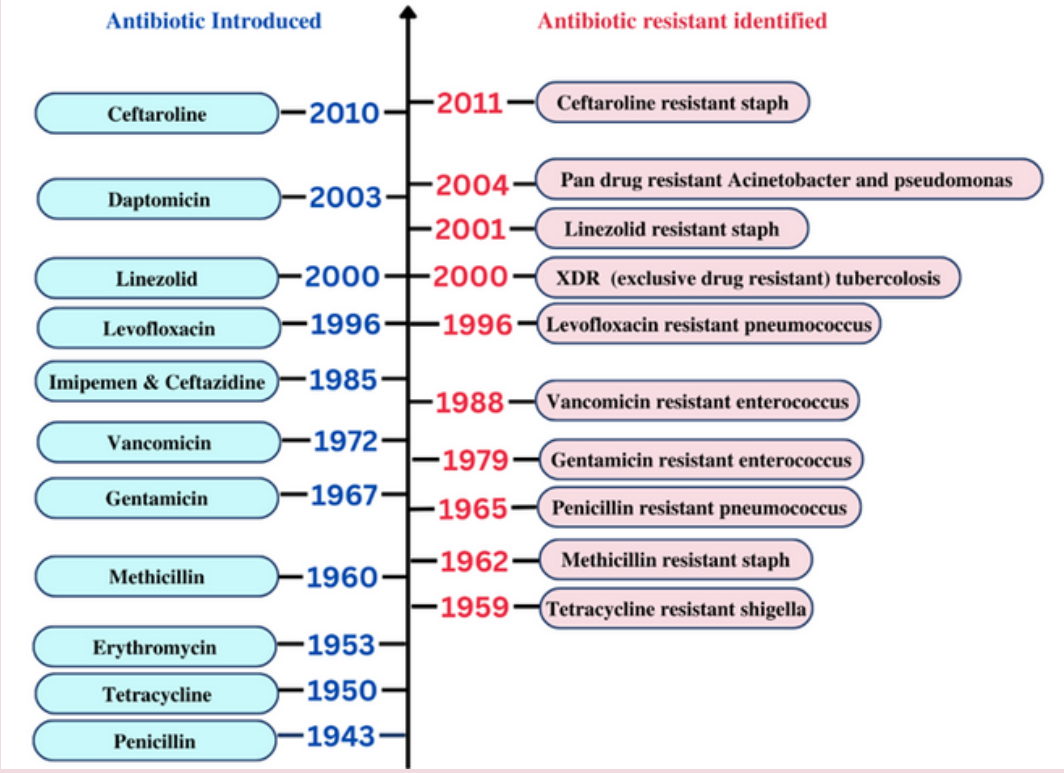


Figure 1: Antibiotic discovery and resistance timeline

million/year) by 2050 (3). Some of the most prevalent resistance mechanisms include antibiotic modification/inactivation, changes in the permeability of the external membrane, the formation of efflux pumps, and changes in the bacterial target site (4).

1.2 Causes of antibiotic resistance

The primary cause of AMR is thought to be the growing usage of antibiotics during the previous few decades. The second reason is that most patients are unable to effectively follow therapy directions. The third reason is that there are very few new medications in development within a specific class of antimicrobials to substitute those become ineffective by growing drug resistance. The use of antibiotics in agriculture to improve crop quality and yield is requisite to meet the increased demand for food and in animal husbandry to prevent infections. The use of antibiotics in agriculture contributes to the spread of resistant bacteria in the food chain and are transferred to human through direct or indirect consumption (5).

1.3 Mechanisms of antibiotic resistance

The bacteria become resistant to antibiotics through the four mechanisms (6) depicted in Figure 2.

1.4 Consequences of Antibiotic Resistance

The antibiotic resistance results in increased morbidity and mortality as resistant infections are harder to treat, leading to prolonged illnesses. Common medical procedures, such as surgeries and cancer treatments, rely heavily on effective antibiotics to prevent and treat infections. The rise of antibiotic resistance threatens the success of these procedures (6).

1.5 Approaches for overcoming antibiotic resistance

1.5.1 Discovery of new antibiotics

The most significant roadblock to novel antibiotic research is the lengthy medication production process and high cost. The current position indicates that selecting and using a new molecule identified in the laboratory would take around 15 years. As a result, instead of inventing new medications or antibiotics, researchers attempt to tweak or rediscover old ones through the various approaches, which include semi-synthetic engineering, genome mining, retro-biosynthetic algorithm and hit compounds technique (7).

1.5.2 Antibiotics adjuvants

Antibiotic adjuvants are employed not just to prevent resistance, but also to boost the efficacy of currently available medications. The majority of antibiotic adjuvants are utilised in combination therapy. Adjuvant treatments demonstrated response by (i) modifying active transport, (ii) improving drug absorption, (iii) altering drug transformation to the intestine or liver, (iv) enhancing immunological activity, and (v) decreasing excretion rate. To treat enterococcal infections, aminoglycoside and penicillin both, work significantly better than a single medicine because synergistic interactions are reached, and drug efficacy appears to be higher in this scenario rather than a single drug. As a result, germs are killed more quickly, and resistance is also inhibited (7).

1.5.3 Phytochemicals

Many infections can be efficiently treated using a combination therapy that includes botanical and nutritional approaches, such as phytochemicals, flavonoids, isoflavonoids, and many other phenolic compounds. The plant extract can also be employed as a powerful strategy to fight microbe resistance development mechanisms. Pathogens are unable to easily train resistance against phytochemical complexes derived from various plant extracts; thus, these can be employed as an alternative to antibiotics. It may include disruption of the cell membrane or increasing membrane permeability and enhancing the influx system, blockage of genome synthesis, changes in the structure of adhesion proteins and membrane-bound enzymes, and interference with cellular processes such as cytoplasm coagulation and QS inhibition. One example is guava leaf extract, which has a bactericidal effect and is also implicated in the neutralisation of pathogen-produced toxins (8).

1.5.4 Nanoparticles

In the fight against AMR, NPs perform two functions. Firstly they have bactericidal activity and the second is that they act as nanocarriers for antibiotics and AMPs. Gold nanoparticles (AuNPs) with functionalized monolayer protection have been shown to suppress clinical MDR, both against Gram-positive and G-negative bacteria. Antibiotics are conjugated or infused by cooperative or non-covalent contact with NPs in the second situation, in which they act as nanocarriers. Antibiotic efficiency is increased by this approach, resulting in high efficacy at a lower minimal inhibitory concentrationwhen compared to free antibiotic. When vancomycin and ampicillin were combined with AuNPs, they provided effective results at low MIC against Gram-positive and G-negative bacteria, respectively (9).

1.5.5 Probiotics

Integrating antibiotics and probiotics has been demonstrated to reduce the severity, length, and occurrence of antibiotic-associated diarrhoea. This encourages patients to closely follow

Figure 2: Strategic approaches used in Bone targeting

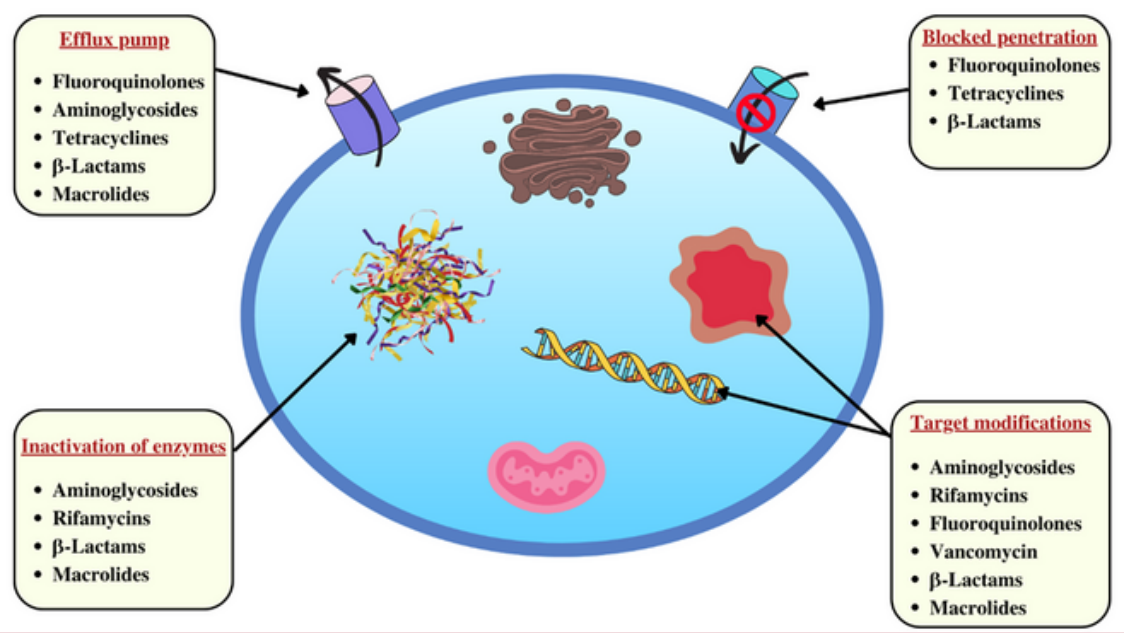


Figure 2: Mechanisms of antibiotic mechanism

their antibiotic prescriptions, slowing the spread of resistance. The extent to which probiotics directly reduce antibiotic resistance propagation is still being explored (10).

1.5.6 Bacteriophage therapy

Bacteriophage therapy involves the use of viruses called bacteriophages to target and destroy specific bacteria. These viruses are highly specific to their host bacteria, leaving other beneficial bacteria unharmed. Bacteriophages can be adapted quickly to target antibiotic-resistant strains. Research in this area shows promise in treating infections that no longer respond to traditional antibiotics (8).

1.5.7 CRISPR-Cas9 Technology

Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) and the CRISPR-associated protein 9 (Cas9) system have revolutionized gene editing and present a potential solution to antibiotic resistance. Scientists are exploring CRISPR-Cas9 technology to directly target antibiotic resistance genes in bacteria, making them susceptible to antibiotics again. This gene-editing approach could play a crucial role in preventing the spread of resistance genes among bacterial populations (8).

1.5.8 Stewardship Programs

Antibiotic stewardship programs aim to promote the responsible use of antibiotics in healthcare settings. These programs involve strict guidelines for prescribing antibiotics, encouraging healthcare professionals to use these medications judiciously. By reducing unnecessary antibiotic prescriptions, the development and spread of antibiotic-resistant bacteria can be curbed (8).

2. Conclusion

Antibiotic resistance is a pressing global health crisis that demands immediate and collaborative action. To overcome this challenge, a multi-pronged approach is essential, involving antibiotic stewardship, the development of novel antibiotics, exploration of alternative treatments like AMPs and bacteriophage therapy, and the integration of cutting-edge technologies like CRISPR-Cas9. It is crucial for healthcare professionals, policymakers,

researchers, and the general public to work together to ensure the responsible use of antibiotics and the implementation of strategies to preserve the efficacy of these life-saving medications for future generations.

References

1. Annunziato G. Strategies to overcome antimicrobial resistance (AMR) making use of non-essential target inhibitors: A review. International journal of molecular sciences. 2019;20 (23):5844.
2. Admassie M. Current review on molecular and phenotypic mechanism of bacterial resistance to antibiotic. Sci J Clin Med. 2018;7:13.
3. Review on antimicrobial resistance. https://amr-review.org/ (accessed July 23).
4. Monserrat-Martinez A, Gambin Y, Sierecki E. Thinking outside the bug: molecular targets and strategies to overcome antibiotic resistance. International journal of molecular sciences. 2019;20(6):1255.
5. Ahmed S, Ahmed MZ, Rafique S, Almasoudi SE, Shah M, et al. Recent approaches for downplaying antibiotic resistance: Molecular mechanisms. BioMed Research International. 2023.
6. Friedman ND, Temkin E, Carmeli Y. The negative impact of antibiotic resistance. Clinical Microbiology and Infection. 2016;22(5):416-422.
7. Wright GD. Antibiotic adjuvants: rescuing antibiotics from resistance. Trends in microbiology. 2016;24(11):862-871.
8. Spellberg B, Bartlett J, Wunderink R, Gilbert DN. Novel approaches are needed to develop tomorrow's antibacterial therapies. American journal of respiratory and critical care medicine. 2015;191(2):135-140.
9. Mohanbaba S, Gurunathan S. Differential biological activities of silver nanoparticles against Gram-negative and Gram-positive bacteria: A novel approach for antimicrobial therapy. In Nanobiomaterials in Antimicrobial Therapy. 2016:193-227.
10. Ouwehand AC, Forssten S, Hibberd AA, Lyra A, Stahl B. Probiotic approach to prevent antibiotic resistance. Annals of Medicine. 2016;48(4):246-255.

Antibiotic resistance: a global phenomenon


Bharti Ahirwar* & Ruchika Gupta

Guru Ghasidas Vishwavidyalaya, Koni, Bilaspur. (CG)

ah_bharti@yahoo.com

Abstract

Antibiotic resistance is a global phenomenon that poses a significant threat to public health worldwide. The overuse and misuse of antibiotics have led to the emergence of antibiotic-resistant bacteria, making it challenging to treat infections that were once easily cured with antibiotics. Antibiotic resistance has been observed in various settings, including hospitals, communities, and agricultural practices, and it affects people of all ages and backgrounds. The consequences of antibiotic resistance are severe, resulting in prolonged illnesses, increased healthcare costs, and higher mortality rates. Therefore, urgent action is required to address this global crisis, including the development of new antibiotics, better infection prevention and control measures, and responsible use of existing antibiotics.

1. Introduction

1.1 Antibiotics

Antibiotics are substances that are produced by a microorganism (e.g. bacterium, fungli) and in dilute solutions have the ability to destroy or inhibit the growth of other microorganisms. They kill bacteria or prevent them from reproducing and spreading. Alexander Fleming discovered penicillin, the first natural antibiotic, in 1928.

1.2 How Antibiotics work?

- Antibiotics attacks the coating or we can say destroy the surrounding of bacteria.
- It slows bacteria production and protein production in bacteria.
-

1.3 How long do antibiotics take to show their action?

- Antibiotics start work after taking it or sometimes the work or action depend on the type of infection you tried to cure.
- Most of the antibiotics should be taken approximately for 2 weeks for better cure.

1.4 Antibiotics resistance

When micro-organisms are no longer controlled or eliminated by antibiotics, antibiotic resistance occurs. Human or animal do not produce resistance and only the microorganism produce resistance. They cause the infections which are harder to treat as the antibiotics used for the treatment are no longer effective.

1.5 Basic mechanisms of antibiotic resistance

1. Limiting uptake of a drug
2. Drug target alterations
3. Inactivating a drug
4. Active drug efflux
5. Alternative enzymes
6. Gram negative bacteria uses all four mechanisms and the gram positives uses all mechanism except the limiting uptake of drug and active drug efflux.

Table.1: Antibiotics, their mechanism, and mechanisms of resistance [2-14]

Category	Mode of action	Major mechanism of resistance
Beta- lactum	Inhibition of cell wall synthesis	Cleavage by beta-lactamases, ESBLs, CTX-mases, Carbapenemases altered PBPs
Aminoglycosides	Inhibition of protein synthesis	Enzymatic modification. Efflux, ribosomal mutations, 16 S rRNA methylation
Quinolones	Inhibition of DNA replication	Efflux, modification, target mutation
Glycopeptides	Inhibition of cell well synthesis	Altered cell wall, efflux
Tetracycline	Inhibition of translation	Mainly efflux
Rifamcylines	Inhibition of transcription	Altered submit of RNA polymerase
Sreptogramins	Inhibition of cell wall synthesis	Enzymatic cleavage, modification efflux
Oxazolodinones	Inhibition of formation of 70s ribosomal complex	Mutations in 23s rRNA genesfollowed by gene conversion.

1.6 Challenges to Antibiotic Resistance:

The usage of antibiotics has increased globally, new resistance are spreading in all over the world and we are not able to treat common infection. A growing number of infections, including pneumonia, tuberculosis, blood poisoning, gonorrhea, and food and water diseases, are becoming more difficult, and in some cases impossible, to treat as a result of the decrease in antibiotic effectiveness brought on by the bacterial resistance.When antibiotics bought for human and animal without prescription, the spread and emergence resistance become worse, in other words we can say that misuse and overuse of antibioticsis increasing the process of antibiotic resistance through which the condition is becoming dangerous. As more and more resistance produce effective treatment of any infection decreases. When antibiotics used to treat infection are no longer effective, then the higher dose and expensive antibiotics are prescribed which increases the duration of illness and treatment along with the cost of treatment leading to economic burden on the family and society.The occurrence of antibiotics resistance places the success of modern medicine or treatment at risk. Organ transplantation, chemotherapy and surgeries become much more dangerous without antibiotics because chances of infection are high during the chemotherapy and surgery and to prevent the infection antibiotics is required.

1.7 Prevention and control

- Use antibiotics only prescribed by certified health professional.
- Always follow the advice of your doctor regarding antibiotics.
- Never use leftover medicine.
- Follow WHO five keys (Keep clean, separate raw and cooked, cook thoroughly, keep food at safe temperatures, use safe water and raw materials.

1.7. 1 Policy makers

- Many policies are being made at national level to tackle antibiotic resistance.
- Improve surveillance of antibiotic-resistant infections.
- Strengthen polices and encourage people to follow the policy made by the authorities.
- Regulate and promote the use and proper disposal of quality medicines.

1.7.2 Health professionals

- Prevent infections by ensuring the working of instrument, and to clean environment.
- Health professional should only prescribe and dispense antibiotics when needed and should be prescribed according to the guidelines.
- Report antibiotics-resistant infections to the surveillance teams.
- Explain the patients about how to take medicine with proper dosing and proper timing and acquaint them with the knowledge about the misuse of antibiotics and how this can be dangerous to their life and health.
- Healthcare Industries- Invest more in research for the development of new antibiotics as the older one are not effective because of resistance.
- Encourage the researchers in field of antibiotics as this is very much needed.

1.7.3 Restricted antibiotics:

- Red – Highly Restricted
- Orange- Restricted
- Green-Unrestricted

1.7.4 List of restricted antibiotics are:

Amikacin, Amphoteracin B, Aztreonam, Ciprifloxacin, Ceftazidine, Chloramphenicol, Flucytosine, Linezolid, Meropenem, Nalidixin, Piperacilin + Tazobactum, Sodium Fusidate, Tobramycin, Cefuroxime, Ertapenem.

1.7.5 Current approaches and future directions:

To tackle the emergence of antibiotics resistance, alternative treatment or strategies are designed to reduce the use of antibiotics for further clinical use. Kalan and Wright 2011 explained the idea of co-administering antibiotics with other drug which enjoy the success in the past through either synergistic or additive effects reduce the antibiotic resistance. Idea of administering other drug with antibiotics also enhanced the effect of antibiotics by combatting the bacterial resistance or also allowing the lower doses of antibiotics to be used.

1.7.6 Modifying enzyme inhibitors

Bacteria employ enzymes to destroy the antibiotics and to produce resistance if the enzymes are destroyed or inhibit then antibiotics resistance should be reduced. These enzymes are categorized by both substrate and the action. Transfer a functional group to the antibiotics and the action of redox and lyase [15]

1.7.7 Beta lactamase inhibitor

- Aminoglycoside – modifying enzyme inhibitors
- Membrane permeabilisers
- Efflux pump inhibitors

1.7.8 WHO Response: [16]

Tackling antibiotic resistance is a complicated task for WHO. 1n 2015, World Health Assembly planned a global action on microorganism resistance including antibiotic resistance.

Five strategic goals make up the "Global Action Plan on Antimicrobial Resistance":

1. Educate people about the antibiotic resistance and its harmful effects.
2. Support research and surveillance related to antibiotic resistance.
3. Lower the occurring of infection
4. To make the best possible use of antibiotics.
5. To ensure sustainable investment in countering antimicrobial resistance

Heads of State supported a political statement at the United Nations General Assembly in New York in September 2016 that showed the world's commitment to adopting a comprehensive, coordinated approach to address the causes of antimicrobial resistance across many sectors, particularly human health, animal health, and agriculture. WHO is aiding Member States in developing national antimicrobial resistance action plans based on the global action plan.

Several projects have been led by WHO to address antibiotic resistance:

- *World Antimicrobial Awareness Week (WAAW):* Since 2015, WAAW is observed annually from November 18 to 24, with the goal of raising awareness of antimicrobial resistance throughout the world and motivating individuals, healthcare professionals, and policymakers to take action to prevent the emergence and spread of infections due to their drug resistance. Antimicrobials have a crucial role in the fight against infections that affect people, animals, and plants. Antibiotics, antivirals, antifungals, and antiprotozoa are some of them. The phrase "Antibiotics: Handle with Care" was originally used, but in 2020 it was changed to "Antimicrobials: Handle with Care" to reflect the growing prevalence of infections with drug resistance.
- *The Global Antimicrobial Resistance Surveillance System (GLASS):* The antimicrobial resistance data collection, analysis, and sharing system supported by the WHO encourages as a standarized approach to global decision-making and drives local, national, and regional action.
- *Global Antibiotic Research and Development Partnership (GARDP):* Through public-private partnerships, GARDP, a joint project of WHO and Drugs for Neglected Diseases Initiative (DNDi), promotes novel antibiotic research and development. By 2023, the public-private collaboration hopes to improve already available antibiotics and hasten the introduction of new antibiotic medications in order to develop and supply up to four new therapies.
- *Interagency Coordination Group on Antimicrobial Resistance (IACG):*The United Nations Secretary-General has established IACG to improve coordination and communication between international organizations and to ensure effective global action is taken against health security. The Deputy Secretary-General of the UN and the Director-General of the WHO co-chair the IACG, which is made up of high-level representatives of pertinent UN agencies, other international organizations, and individual specialists from other fields.

2. Conclusion

In conclusion, antibiotic resistance is a global phenomenon that poses a threat to public health worldwide. The overuse and misuse of antibiotics, as well as their use in agriculture, have contributed to the development of resistant bacteria. To address this issue, it is crucial to promote responsible use of antibiotics, develop new antibiotics, and implement infection prevention and control measures. Failure to take action now could lead to a future where common infections become untreatable, highlighting the urgent need for collaborative efforts from all stakeholders to combat this global crisis.

References

1. Aminov RI. The role of antibiotics and antibiotic resistance in nature. Environmental microbiology. 2009 Dec;11(12):2970-88.
2. Murray CJ, Ikuta KS, Sharara F, Swetschinski L, Aguilar GR, Gray A, Han C, Bisignano C, Rao P, Wool E, Johnson SC. Global burden of bacterial antimicrobial resistance in 2019: a systematic analysis. The Lancet. 2022 Feb 12;399(10325):629-55.
3.Bradford PA.Extended-spectrumβ-lactamases in the 21stcentury: characterization, epidemiology, and detection of this important resistance threat. Clin Microbiol Rev 2001;14:933-951.6.
4. Matagne A, Lamotte-Brasseur J, Frère JM. Catalytic properties of class A β-lactamases: efficiency and diversity. Biochem J 1998;330:581-598
5.Miguet L, Zervosen A, Gerards T, Pasha FA, Luxen A, Distèche-Nguyen M,Thomas A. Discovery of new inhibitors of resistant Streptococcus pneumoniae penicillin binding protein (PBP) 2x by structure-based virtual screening. JMed Chem 2009;52(19):5926-5936.
6.WhiteDG, Hudson C,Maurer JJ, AyersS, Zhao S, Lee MD,Bolton L, Foley T,Sherwood J. Characterization ofchloramphenicol and forfenicol resistance in Escherichia coli associated withbovine diarrhea. J Clin Microbiol2000; 38:4593-4598.
7. Marquez B.Bacterial effux systemsand ef□ux pumps inhibitors. Biochimie 2005;87:1137-1147.
8.Alcaide E,Blasco MD, Esteve C. Mechanismsof quinolone resistance in Aeromonas species isolated from humans, water and eels. Res Microbiol 2010;161(1):40-45.
9. ZakeriB, Wright GD.Chemical biology of tetracycline antibiotics.Biochem Cell Biol 2008;86:124-136.
10. Aleksandrov A, Schuldt L, Hinrichs W, Simonson T. Tetracycline-tet repressorbinding specicity: insights from experiments and simulations. Biophys J 2009;97(10):2829-2838,
11. Liaw SJ, Lee YL, Hsueh PR. Multidrug resistance in clinical isolates of Stenotrophomonas maltophilia: rolesof integrons, ef□uxpumps, phosphoglucomutase (SpgM), and melanin and bio□lmformation. Int JAntimicrob Agents 2010;35(2):126-130.
12. Liaw SJ, Lee YL, Hsueh PR. Multidrug resistance in clinical isolates of Stenotrophomonas maltophilia: rolesof integrons, ef□uxpumps, phosphoglucomutase (SpgM), and melanin and bio□lmformation. Int JAntimicrob Agents 2010;35(2):126-130.
13. Sun AH, Fan XL, Li LW, Wang LF, Ans WY, Yan J. Rapid detection ofrpoB gene mutations in rif-resistant M. tuberculosis isolates by oligonucleotide microarray. BiomedEnviron Sci 2009;22(3):253-258.
14. WangXJ, Du GJ,Zhao SQ, YanM, Gu LQ. Synthesis and antibacterial activity of a series of alpha-substituted acetylpiperazinyl oxazolidinones. Chem Biol Drug Des 2009;74(3):276-281.
15. Melander RJ, Melander C. The challenge of overcoming antibiotic resistance: an adjuvant approach?. ACS infectious diseases. 2017 Aug 11;3(8):559-63.
16.https://www.who.int/news-room/fact-sheets/detail/antibiotic-resistance#:~:text=Antibiotic%20resistance%20occurs%20when%20bacteria,caused%20by%20non%2Dresistant%20bacteria.

Multi-drug resistance to antibiotics: A way forward and challenges

Preeti Karade[1], Popat Kumbhar[2], Ashatai Patil[2], John Disouza[2]

1Department of Pharmaceutics, Appasaheb Birnale College of Pharmacy, Sangli, Maharashtra, India, 416416
2Department of Pharmaceutics, Tatyasaheb Kore College of Pharmacy, Warananagar, Tal.: Panhala, Dist.: Kolhapur Maharashtra, India, 416113

pritigkarade@gmail.com; pskumbhar@tkcpwarana.ac.in; ashabpatil.tkcp@gmail.com; jidisouza@tkcpwarana.ac.in

1.Introduction

For many decades, antibiotics have been effective in protecting the health of people, animals and plants (1). They frequently do this by impeding the production of proteins, influencing the formation of cell walls, interfering with the machinery that produces nucleic acids, disturbing metabolic pathways, or other particular activities (2).

Multidrug resistance (MDR) is a form of antibiotic resistance expressed by a species of microbe to at least one drug in different classes of antimicrobials. Antimicrobial resistance (AMR) has become a global health and development threat. The emergence and spread of drug-resistant organisms with acquired new resistance mechanisms, leading to AMR, is threatening our ability to treat common infectious diseases. Especially alarming is the rapid global spread of multi- and pan-resistant bacteria called “superbugs”, causing infections that are non-treatable with existing antimicrobials. It requires urgent multisectoral action in order to realise the Sustainable Development Goals (SDGs). WHO has declared AMR as one of the top 10 global public health threats that humanity is facing. Currently, approximately 700,000 people worldwide die due to antimicrobial resistance and are likely to be associated with 10 million deaths per year by 2050 (3). Few of the common multidrug-resistant organisms (MDROs) are listed in table 1.

The traditional oral or intravenous administration of these medications needed substantial and frequent dosing, which contributed to microbial resistance against therapeutics. Additionally, overuse and withdrawal of antibiotic therapy is also responsible for the microbial resistance. The irrational use of antibiotics in medicine and agriculture also led to increase in microbial resistance to antibiotics.

The cost of AMR to the economy is substantial. Along with death and disability, longer hospital stays due to prolonged illness, financial challenges for affected due to the need for more

Table. 1: Some common multidrug-resistant organisms, types of resistance they develop and their abbreviations

Bacteria	Antibiotic resistance	Abbreviated as
Enterococcus species	Vancomycin-resistant	VRE
Staphylococcus aureus	Methicillin-resistant	MRSA
Enterobacteriaceae *(e.g. E. coli, Klebsiella)*	Extended-spectrum β-lactamase	ESBLs
Enterobacteriaceae *(e.g. E. coli, Klebsiella)*	Carbapenem resistance	CRE *KPC
Enterobacter species, *E. coli, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa*	Multidrug-resistant Gram-negative rods	MDR GNR

* Klebsiella pneumoniae carbapenemase (KPC)

expensive medicines are the major challenges. The success of modern medicine in treating infections, is majorly dependent of availability of effective antimicrobials, especially during major surgery and cancer chemotherapy.

There are several strategies employed to overcome antimicrobial resistance; educating masses and health-care professionals on preventing AMR; and encouraging research in developing diagnostic tools to identify resistant infections and new antimicrobials are most imperative.

2. Resistance Mechanisms

Increasing abusive and overuse of antibiotics in animals (food, pets and aquatic) or humans has great role in accelerating antimicrobial resistance. Insufficient knowledge about the use of antibiotics, patients' non-compliance with prescribers' instructions, the use or sharing of leftover antibiotics from previous prescriptions, the sale of over-the-counter antibiotics, poor hospital sanitation and hygiene practices, migratory birds, and the release of antibiotics that have not been metabolized into the environment all help to foster the growth of resistant microorganisms (4,5). The spread of resistant bacteria and resistance genes might also be a result of international trade and travel. At the same time, the development of new antibiotics slowed down dramatically.

Resistance to antibiotics can be innate, acquired, cross- or multi-drug resistant. Diverse defense mechanisms employed by bacteria to evade drugs can lead to antibiotic resistance (figure 1). The variation in structure between gram-negative and gram-positive bacteria determines the mechanism of resistance. It is a complex issue with many facets that is affected by a number of factors, including;

- Alternation of membrane permeability;
- Alteration of targets that prevent antibiotics reaching and binding to the target;
- Alteration and or development of enzymes causing antibiotic inactivation;
- Drug extrusion from the cell through the efflux pump mechanism (overexpression of certain protein by bacteria);
- Alteration of metabolic pathways etc.

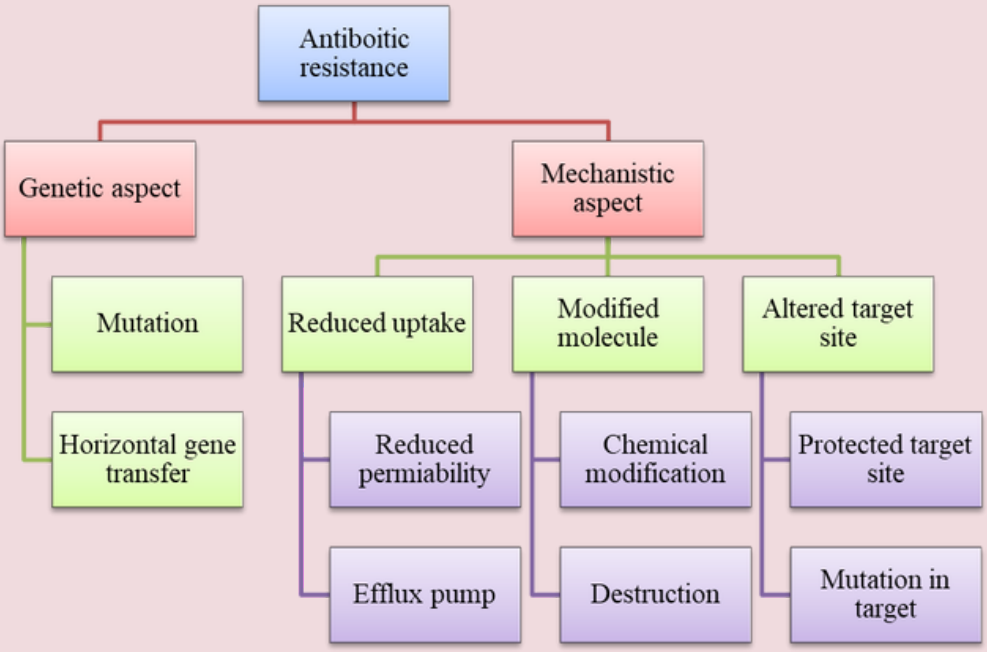


Figure 1. Mechanisms of antibiotic resistance

3.Way forward: Techniques for combating resistance

Antibiotic resistant bacteria are transferred between different sectors like people, animals and foodstuffs, and they can spread in our environment. Therefore, efforts must be made from a broad perspective to combat antibiotic resistance summarized in figure 2.

Combination therapy works in combating AMR, e.g. Combination of colistin and carbapenems for the treatment of CRE infections, ceftazidime-avibactam and aztreonam for the treatment of infections caused by ESBLs, daptomycin and linezolid for the treatment of VRE etc. However, some researchers have found that combination therapies can promote acquisition of drug

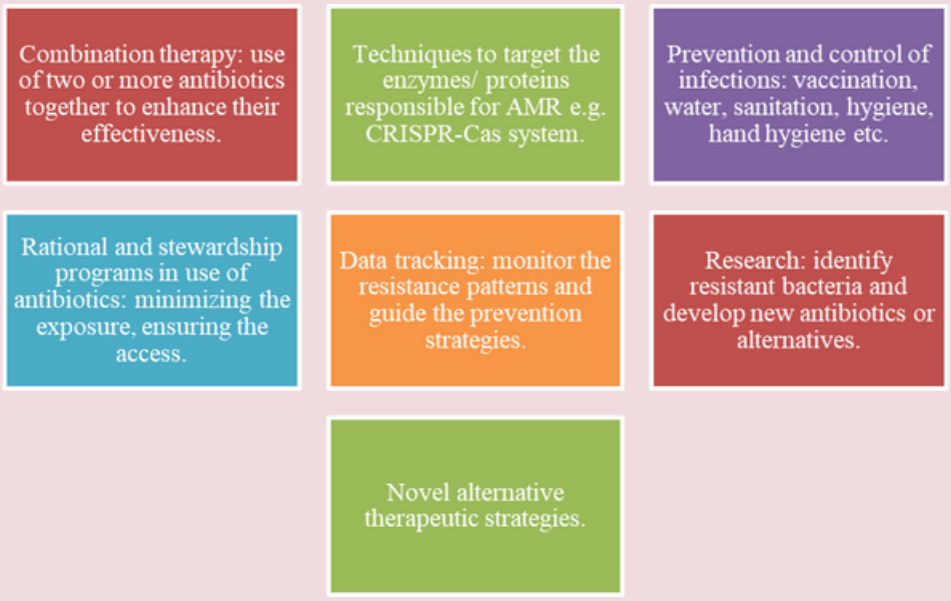


Figure 2. Strategies to combat antimicrobial resistance

resistance. CRISPR-Cas system may be repurposed to selectively target and destroy bacterial genomes, offering an attractive option for the development of the next-generation antimicrobials to combat AMR pathogens. Improvements in water sanitation and hygiene (WASH) and wastewater management in all sectors also helps reducing the spread of AMR along with various infection control measures, including hand hygiene and environmental hygiene etc. Vaccines blocks transmission of the MDR strains and prevents people developing infections which are very hard to treat.

The prevention of irrational use of antibiotics helps avoiding emergence of extended AMR, wherein the environment acts as a reservoir of antibiotic resistance genes (ARGs); completing the cycle of contamination and recontamination.
Data tracking can be successfully employed to monitor the resistance patterns and can help deciding prevention strategies combating AMR including surveillance of newly emerged AMR pathogens, prioritizing research focusing on AMR, and awareness programs among the local population etc. The development of new antibiotics needs to be prioritized by the scientific community.

New class of antibiotics known as dual-acting immune antibiotics (DAIAs) are promising in combating AMR by blocking an essential metabolic pathway in bacteria while enhancing the natural host immune response (6). Development of inhibitors of efflux pump could improve the therapeutic activity against resistant bacteria. In this regard, combination of available antibiotics with suitable adjuvants improves the effect of an antibiotic against a resistant strain of bacteria. Thus, administering an antibiotic along with an adjuvant improves the inhibitory action and decreases resistance (7). Novel alternative therapeutic strategies such as antimicrobial nanomaterials (NMs) therapy, antimicrobial photodynamic therapy, and antibiotic hybrids, have the potential to treat many infectious diseases (8). Designing antimicrobial peptides with broader spectrum of targets (9), phage therapy to kill antibiotic-resistant bacteria (10), the delivery of drugs as nanoparticles for controlled and sustained release, use of natural compounds (11) are some of other approaches that can be adopted. Furthermore, targeting the proteins or enzymes responsible for resistance, through targeted drug delivery systems can also assist to overcome the microbial resistance (12).

4.Challenges

The biggest challenge in combating AMR is quickly finding the resistance genes of bacteria responsible for AMR. For the purpose of identifying the resistance genes in bacteria resistant to antibiotics, effective molecular diagnostic approaches are required. In this regard, the government should establish research and development priorities and underline the need of developing and enhancing integrated laboratory platforms and services for antimicrobial agents. Besides, major pharmaceutical companies have driven out of antibacterial research due to the expensive clinical trials, new regulatory uncertainties over approval requirements, and a low-economic return. Development of nanoparticles laden with antibiotic is a significant challenge to treat MDR bacteria. The apt control of size and shape of NPs is highly required to achieve better penetration and interaction with microbial cells that result in significant therapeutic performance. NPs can aggregate that result in loss of stability which contribute to toxicity. An important concern in the usage of nanomaterials is toxicity.

In spite of harming humans' beneficial microbes, local and systemic toxic concerns are a serious worry allied with NPs. Various strains and disease brought from various microorganisms may have an impact on the function of nanoparticles and make treatment more difficult. The manufacture, scale-up, characterization of physiochemical properties, biocompatibility, standardization, and methodologies to compare data originating from in vivo and in vitro research are all lacking adequate guidelines.

Beside these technical challenges, quite a few measures are needed to be taken to win this war against microorganisms once again, including;

1. Educating the public about 'the risks of AMR' and 'how to prevent it?';
2. Ensuring prescribing antibiotics by healthcare professionals only when it is necessary;
3. Encouraging population to get vaccinated;
4. Ensuring 'infection prevention and control measures' including hand hygiene, safe food handling, and safe sex practices etc.;
5. Encouraging the research in development of new antibiotics and alternative treatments and new diagnostic tools to help identify resistant infections.

5.Conclusion

The responsible and prudent use of antibiotics across all health care sectors requires awareness and investment in professional education. Antibiotic resistance is international disaster that terrorizes healthcare, resulting longer hospitalization and higher rates of morbidity and mortality. Because the drivers of antimicrobial resistance lie in humans, animals, plants, food and the environment, increasing awareness among all stakeholders is most critical challenge that need to be urgently addressed. With a goal to tackle the menace of antibiotic resistance, unethical promotion of antibiotics must be controlled and everybody should have some basic knowledge. To combat microbial MDR of antibiotics, it is crucial to develop novel antibiotics. Moreover, major efforts are essential in delivering antimicrobials smartly using latest technology. Collaboration and accountability are required at all levels to protect the coming years from drug-resistant diseases. The globe needs to take immediate action to protect the future from drug-resistant diseases and the devastating effects of superbugs. To put it briefly, public awareness, national action plans, optimal use of antibiotics, and ground-breaking research in understanding molecular mechanism of resistance, developing next generation antimicrobials are the measures to be taken by 'superman' to win against 'superbugs'.

References

1. Hernando-Amado S, Coque TM, Baquero F, Martínez JL. Defining and combating antibiotic resistance from One Health and Global Health perspectives. Nature microbiology. 2019;4(9):1432-1442.
2. Levy SB, Marshall B. Antibacterial resistance worldwide: causes, challenges and responses. Nat Med. 2004;10:122-129.
3. Strathdee SA, Davies SC, Marcelin JR. Confronting antimicrobial resistance beyond the COVID-19 pandemic and the 2020 US election. Lancet. 2020; 396 (10257): 1050-1053.
4. Burow E, Käsbohrer A. Risk Factors for Antimicrobial Resistance in Escherichia coli in Pigs Receiving Oral Antimicrobial Treatment: A Systematic Review. Microb Drug Resist. 2017 Mar;23(2):194-205.
5. Iskandar K, Molinier L, Hallit F, et al. Drivers of Antibiotic Resistance Transmission in Low and Middle Income Countries from a "One Health" Perspective -A Review. Antibiotics (Basel Switzerland). 2020;9(7):372.
6. Pokrovskaya V, Baasov T. Dual-acting hybrid antibiotics: a promising strategy to combat bacterial resistance. Expert Opin Drug Discov. 2010;5(9):883-902.
7. Worthington RJ and Melander C. Combination approaches to combat multidrug-resistant bacteria. Trends Biotechnol. 2013;31(3):177-84.
8. Alqahtani FA, Almustafa HI, Alshehri RS, et al., Combating Antibiotic Resistance in Bacteria: The Development of Novel Therapeutic Strategies. J Pure Appl Microbiol. 2022;16(4):2201-2224.
9. Gordya N, Yakovlev A, Kruglikova A, Tulin D, Potolitsina E, et al. Natural antimicrobial peptide complexes in the fighting of antibiotic resistant biofilms: Calliphora vicina medicinal maggots. PLoS One. 2017;12(3):e0173559.
10. Chan BK, Sistrom M, Wertz JE., Kortright KE, et al. Phage selection restores antibiotic sensitivity in MDR Pseudomonas aeruginosa. Sci. Rep. 2016;6:26717.
11. Chandra H, Bishnoi P, Yadav A, et al. Antimicrobial resistance and the alternative resources with special emphasis on plant-based antimicrobials-a review. Plants. 2017;6:16.
12. Murugaiyan J, Kumar PA, Rao GS, et al. Progress in Alternative Strategies to Combat Antimicrobial Resistance: Focus on Antibiotics. Antibiotics. 2022;11(2):200.

Mechanistic Approaches for Combating against Antibiotic Resistance


Rajasekhar Reddy Alavala, GSN Koteswara Rao*

Shobhaben Pratapbhai Patel School of Pharmacy & Technology Management, SVKM's NMIMS, Vile Parle (W), Mumbai.
snkoteswararao.g@nmims.edu

1.Introduction

The discovery of antibiotics was one of the milestones in human history since it revolutionized the world of medicine in many ways and helped save innumerable lives. Fundamentally, antibiotics are from natural origin; a deep understanding of the chemical structure, biosynthetic routes, evolution, and mode of action of the many antibiotic compounds is however, required. Due to its complex structure with respect to the functional groups and chiral nature, it is challenging to perform the total synthesis of these compounds in the lab. During the history of antibiotics, research on modes of action has offered biochemical knowledge on these ligands and their microbial targets (1, 2).

The wider applications and the low-cost availability have offered them a non-prescription and off-label medication status in certain parts of the world. Years of continuous choice burden from human usage of antibiotic compounds, especially the underuse, overuse, and misuse, have developed the antibiotic-resistant microbial species and dispersed them in the microbial populations through the environment. This developed resistance may not be a natural process; it might be a man-made situation, which can be a finer illustration of the Darwinian concepts of selection and survival.

2.Antibiotic resistance (AR): causes and implications

The resistance of microorganisms towards the antibiotics is of two types, acquired and intrinsic. This involves the degradation or covalent transformation of key functional groups by the bacterial enzymes causing the antibiotic deactivation. The modifications include hydrolysis, glycosylation, phosphorylation, acetylation, monooxygenation, and nucleotidylation. In few cases, the modification of the bacterial target is such that the antibiotic does not bind to the target or avoids the target by utilizing an alternate pathway for producing a molecule that is crucial for the survival of bacteria. Another mechanism is the use of the efflux protein of the bacteria to actively detoxify the antibiotic from the organism. Some organisms can alter the membrane permeability, which can affect the antibiotic entry into the organism. All or some of these mechanisms are associated with acquired mutations and

genetic modifications upon prolonged exposure to the respective antibiotics class. Intrinsic resistance implies the presence of certain genes in microbial genomes that possibly will trigger a phenotypic resistance, i.e., proto- or quasi-resistance (3, 4). Different genus, species, or subspecies, etc., demonstrate different types of AR phenotypes (5, 6). Super resistant bacteria have developed a higher virulence and transmissibility in certain contexts. AR is, in reality, a pathogenicity factor (7).

3.Approaches to combat the AR

Researchers are continuously striving to combat the AR by studying several approaches but very few were successful in meeting their expectations. It is still a challenging task with the ever-increasing resistance. In this article, some important approaches have been discussed briefly.

3.1. Preventive measures

As a preventive measure, the major remedies proposed for combating AR include: strictly controlling the use of antibiotics, avoiding unnecessary prescribing of antibiotics, and building awareness among the public. However, these strategies can't control the global threat of AR that is continuously increasing due to the unrest in health conditions (2).

3.2.Antibiotic hybrids

Due to the ever-increasing resistance against antibiotics pertaining to overexpressed efflux pumps, a constant search for new strategies continues. Antibiotic hybrids are one such approach, where two or more antibiotics or an antibiotic with an adjuvant (e.g., an inhibitor of an efflux pump) are developed to target the bacteria. Antibiotic hybrids include covalently linked synthetic constructs of the two molecules. They offer the advantage of decreased risk of developing resistance, improved therapeutic effect due to additive effects of drug mechanisms thereby lowering the mortality. However, more clinical evaluations need to be conducted to confirm such advantages (8).

Li et al., has reviewed the role of supramolecular materials as a remedy to combat AR. In addition to the loading of antibiotics, the supramolecular materials also show a unique way of interacting non-covalently with biomolecules. Bioconjugated nanoparticles with penicillin G and squalene showed a different intracellular diffusion compared with penicillin G where clathrin-dependent endocytosis was observed in a neutral environment. It indicated that supramolecular-assembled bioconjugated nanoparticles have an improved activity.

Another such example for supramolecular functional materials includes beta-cyclodextrin with doxycycline, which showed an enhanced activity and a low value of minimum inhibitory concentration against S. aureus. Other examples of supramolecular materials include the application of cationic polymers, chitosan, antibacterial peptides, and metals (9).

3.3. Nano-based antibiotics

Nanotechnology has been reported to be a useful approach for combating AR. Engineered nanoparticles have the ability to disrupt the cell membranes eliminating the resistant strains. Nanotechnology also helps to predict the emergence of resistance and hence the combating measures can be implemented. By knowing the mechanisms of resistance, nanotechnology has shown to be helpful in developing nanoscale carrier systems by overcoming the existing pressures. Targeted delivery of arsenal through nanocarriers is one such promising example (10).

Nanocarriers offer improved pharmacokinetic properties for the antibiotics by restoring their efficacy against drug-resistant bacteria (11). With the ever-growing advancements in the design and development of nanotherapeutics with tailorable properties, researchers are studying their applicability to combat the AR (12). As a part of the drug-metal ion complexes, Sceptrin–Au nano-aggregates were reported to combat the drug-resistant bacterial infections. It has shown superior activity, with lower side effects, against carbapenem-resistant gram-negative bacteria (13).

3.4. Omics

Systems biology employs specific bioinformatic tools and methods that can create a comprehensive representation of the mechanisms and consequences of AR, and, sequentially, enable knowing about the events at the genomic circle by examining the increasing volume of information that gets generated in molecular biology (14). The methods include genomics, proteomics, transcriptomics, and metabolomics. Integration of the whole gene sequencing allows for the quick discovery of AR-associated genes, and higher intolerant capacity in terms of bacterial resistance contributing factors and genomic epidemiology of distinct species (15, 16).

Understanding the mechanisms of antibiotics will possibly lead to the discovery of novel ways of limiting the propagation of the resistance genes. Proteomics is extensively used to advance the understanding of the key proteome of antibiotic-resistant microbial species. Many investigators have concentrated their attempts in recent times on discovering and developing protein biomarkers that correlate with sensitivity or resistance. Such developments are made feasible with the help of enhanced proteomics methods such as matrix-assisted laser desorption ionization and time-of-flight mass spectrometry (MALDI-TOF MS) (17).

Bacterial metabolic pathways play an important role in AR and might be responsible for the acquired resistance and usually undergoing modifications in the pathway to avoid the key biomarkers which are targets for the antibiotics. For instance, elevated metabolic action is required to initiate a wide range of molecular systems, including cell wall changes, mutation steadying, transportation, energy production, and efflux pump overexpression. Metabolomics allows for a global perspective of all the compounds involved in metabolism, which are in order inherently linked to the organism’s phenotype. Metabolomic methods are critical for understanding the interactions between AR mechanisms and bacterial metabolism (18).

3.5. AI or ML based methods for new antibiotics

The traditional approaches to antibiotic research are time-consuming and expensive; hence, novel and more effective ways must be utilized to choose the most promising compounds with novel processes that the bacteria are unfamiliar with (19). Based on significant advances achieved in recent years, incorporation of artificial intelligence (AI) in antibiotic creation has initiated the discovery of newer antibiotics that can offer promising results. By swiftly recommending several new compounds based on algorithms produced by machine learning (ML) approaches such as neural networks (NN) or deep learning (DL), AI helps fasten the early drug discovery process, as the algorithms foresee antibiotic efficacy for each created molecule (20). Despite continual progress in the domain of ML, the advantage against the clinical environment is yet to be accomplished.

4.Future directions

With the existing knowledge and new technologies that are coming into force, researchers can focus on hybrid mode of combining several outcomes in order to combat the AR. Computer-aided drug design and development can also be taken into count for getting better remedies within reasonable screening time. In addition, nanotechnology needs to be explored for finding more alternatives for overcoming the drug resistance. DNA-based targeted therapy could also be a possible answer to overcome this drug resistance and has a scope for future exploration.

5.Conclusion

AR which is considered as a global risk, is one of the major challenges that lies in front of the healthcare professionals. If unanswered, it will certainly turn out to be a severe disaster as the existing antibiotics cannot address these resistant infections. It is becoming very hard to understand the smart behaviour of bacteria that is changing its pattern of developing drug resistance from time to time. Awareness among the public is one of the important preventive measures and that should help the researchers identify reasonable solutions for undesired drug resistance.

References

1. Aminov RI. The role of antibiotics and antibiotic resistance in nature. Environ Microbiol. 2009;11(12):2970-88.
2. Davies J, Davies D. Origins and evolution of antibiotic resistance. Microbiol. Mol. Biol. Rev. 2010; 74(3): 417-33.
3. Nicolaou KC, Montagnon T. Molecules that changed the world. Weinheim: Wiley-VCH. 2008.
4. Von Döhren H. Antibiotics: Actions, origins, resistance, by C. Walsh. 2003. Washington, DC: ASM Press. Protein Sci. 2004;13(11):3059-60.
5. Alekshun MN, Levy SB. Molecular mechanisms of antibacterial multidrug resistance. Cell. 2007;128(6):1037-50.
6. Kumar S., Varela M.F. Molecular mechanisms of bacterial resistance to antimicrobial agents. Formatex Research Center; Badajoz, Spain: 2013. pp. 522–534.
7. Kakoullis L, Papachristodoulou E, Chra P, Panos G. Mechanisms of antibiotic resistance in important gram-positive and gram-negative pathogens and novel antibiotic solutions. Antibiotics. 2021;10(4):415.
8. Gupta V, Datta P. Next-generation strategy for treating drug resistant bacteria: Antibiotic hybrids. Indian J. Med. Res. 2019; 149(2): 97-106.
9. Li X, Bai H, Yang Y, Yoon J, Wang S, Zhang X. Supramolecular Antibacterial Materials for Combatting Antibiotic Resistance. Adv. Mater. 2019; 31(5): e1805092.
10. Muteeb G. Nanotechnology-A Light of Hope for Combating Antibiotic Resistance. Microorganisms. 2023; 11(6): 1489.
11. Le H, Karakasyan C, Jouenne T, Le Cerf D, Dé E. Application of Polymeric Nanocarriers for Enhancing the Bioavailability of Antibiotics at the Target Site and Overcoming Antimicrobial Resistance. Appl. Sci. 2021; 11(22): 10695.
12. Munir MU, Ahmad MM. Nanomaterials Aiming to Tackle Antibiotic-Resistant Bacteria. Pharmaceutics. 2022; 14(3): 582.
13. Jong Min An, Sangrim Kang, Chang Woo Koh, Sungnam Park, Myung Sook Oh, Dokyoung Kim. Sceptrin–Au nano-aggregates (SANA) for overcoming drug-resistant Gram-negative bacteria. Nanoscale Horiz. 2022;7:873-882.
14. Francine P. Systems Biology: New Insight into Antibiotic Resistance. Microorganisms. 2022;10(12):2362.
15. Gupta SK, Padmanabhan BR, Diene SM, Lopez-Rojas R, Kempf M, Landraud L, Rolain JM. ARG-ANNOT, a new bioinformatic tool to discover antibiotic resistance genes in bacterial genomes. Antimicrob. Agents Chemother. 2014;58(1):212-20.
16. Zankari E, Allesøe R, Joensen KG, Cavaco LM, Lund O, Aarestrup FM. PointFinder: a novel web tool for WGS-based detection of antimicrobial resistance associated with chromosomal point mutations in bacterial pathogens. J. Antimicrob. Chemother. 2017;72(10):2764-8.
17. Pluskal T, Castillo S, Villar-Briones A, Orešič M. MZmine 2: modular framework for processing, visualizing, and analyzing mass spectrometry-based molecular profile data. BMC bioinformatics. 2010;11(1):1-1.
18. Shen X, Zhu ZJ. MetFlow: an interactive and integrated workflow for metabolomics data cleaning and differential metabolite discovery. Bioinformatics. 2019;35(16):2870-2.
19. Popa SL, Pop C, Dita MO, Brata VD, Bolchis R, Czako Z, Saadani MM, Ismaiel A, Dumitrascu DI, Grad S, David L. Deep learning and antibiotic resistance. Antibiotics. 2022;11(11):1674.
20. Martorell-Marugán J, Tabik S, Benhammou Y, del Val C, Zwir I, Herrera F, Carmona-Sáez P. Deep learning in omics data analysis and precision medicine. Exon Publications. 2019:37-53.

Revolutionizing the Fight Against Antibiotic Resistance: Innovative Concepts and Approaches



Anjali Sharma[1]*, Vishnu Mittal[1], Devkant Sharma[2]

1Guru Gobind Singh College of Pharmacy, Yamunanagar (135001) Haryana
2CH. Devi Lal College of Pharmacy, Jagadhari (135003) Haryana

sharma.sharmaa.anjali@gmail.com

Abstract

A serious concern to world health is antibiotic resistance. The fast increase in germs that are resistant to antibiotics and the slow discovery of new medications in recent years have made it clear that this problem needs immediate attention. In order for antibiotics to be effective in treating illnesses, bacteria must be able to tolerate their effects. This is known as antibiotic resistance. Multiple methods, such as genetic changes, horizontal gene transfer, and the development of biofilms, might lead to this resistance. Several approaches have been proposed to overcome antibiotic resistance. Additionally, enhancing surveillance and diagnostic techniques can aid in the early detection of resistant infections and guide appropriate treatment decisions. Overcoming antibiotic resistance requires a multifaceted approach that combines the discovery of new antibiotics, optimization of existing treatments, exploration of alternative therapies, and addressing the underlying factors driving resistance. The current review aims to discuss the concept along with various approaches to overcome antibiotic resistance which will be beneficial for pharmaceutical scientists working in the field of biotechnology and related diseases.

Keywords: Antibiotic Resistance, Bacteria, Infection, Microbes, Biofilm, Biotechnology.

1. Background

An increasing problem throughout the world is antibiotic resistance, which is when bacteria learn to resist the effects of medicines, making them useless for treating bacterial diseases. Antibiotic resistance poses significant challenges to healthcare systems and can lead to higher medical costs, prolonged hospital stays, increased mortality rates, and limited treatment options for common infectious diseases. When antibiotics are used, bacteria are exposed to these drugs, and some bacteria may possess inherent or acquired mechanisms to resist their effects (1). Bacteria develop resistance through various mechanisms, including the acquisition of plasmids encoding resistance genes or mutations in their own genetic material. Exposure to suboptimal antibiotic concentrations or incomplete treatment courses can promote the survival of resistant bacteria, allowing them to multiply and spread. The consequences of antibiotic resistance are significant. As antibiotics lose their efficacy, common illnesses including pneumonia, TB, blood poisoning, gonorrhea, and foodborne diseases are getting harder to cure and occasionally becoming incurable. Since then, bacteria have created a number of defenses against the effects of antibiotics, making them useless (2).

2. What is Antibiotic Resistance?

Antibiotic resistance is the ability of bacteria or other microorganisms to resist the effects of medications that were formerly effective in treating diseases brought on by these organisms. In other words, bacteria become resistant to drugs meant to eradicate them or halt their growth (3).

3.Causes of Antibiotic Resistance

Antibiotic resistance has a number of root causes, including:

3.1. Antibiotic misuse and overuse: One of the primary causes of antibiotic resistance is the inappropriate use of antibiotics, such as taking them for viral infections like colds or flu, which are not affected by antibiotics (4).

3.2. Agricultural use: In order to encourage growth and avoid illness, antibiotics are frequently used in cattle production. Antibiotic abuse and overuse in animals lead to the growth of germs resistant to antibiotics, which can subsequently be passed to people through tainted food items (5).

3.3. Inadequate infection control: Inadequate hygiene practices, improper sanitation, and lack of infection control procedures in healthcare facilities may cause patients to develop antibiotic-resistant microorganisms. This is particularly problematic in hospitals and long-term care facilities (6).

3.4. Insufficient new antibiotics: There are fewer treatment options available to combat resistant bacteria, making it harder to effectively treat infections (7).

3.5. Biological evolution and genetic mutations: Bacteria have the ability to evolve through natural selection, bacteria that possess genetic mutations allowing them to survive exposure to antibiotics will survive and multiply, passing on their resistant traits to subsequent generations (8).

4. Mechanisms of antibiotic resistance development

The four primary causes of antibiotic resistance are as follows:

- Limiting a drug's intake: Bacteria can develop mechanisms to prevent antibiotics from entering the cell.
- Modifying a drug target: Bacteria can change the structure of their target proteins so that antibiotics can no longer bind to them.
- Inactivating a drug: Bacteria can produce enzymes that break down antibiotics, making them ineffective.
- Active drug efflux: Bacteria can pump antibiotics out of the cell before they can have an effect (9).

5. Approaches to Overcoming Antibiotic Resistance

5.1 Developing novel antibiotics

Developing novel antibiotics is indeed one of the key approaches to overcoming antibiotic resistance. The new antibiotics can help address the growing challenge of resistance by providing effective treatment options against resistant bacteria (10).

5.2 Exploration of new microbial sources: Exploration of new microbial sources is a vital aspect of developing novel antibiotics. By investigating untapped microbial diversity, scientists can

discover new microorganisms that produce bioactive compounds with potential antimicrobial properties. The process of exploring new microbial sources typically involves:

1. Sampling
2. Isolation
3. Screening
4. Identification
5. Extract preparation
6. Compound isolation and purification
7. Testing, characterization, optimization and development (11).

5.3 Utilizing advanced technologies (e.g., genomics, metagenomics): Utilizing advanced technologies such as genomics and metagenomics in developing novel antibiotics is a powerful approach to combat antibiotic resistance (12).

5.4 Enhancing antimicrobial stewardship

Enhancing antimicrobial stewardship is a crucial approach to combating antibiotic resistance. Antimicrobial stewardship refers to the coordinated efforts and strategies aimed to control the development of bacterial resistance. Here are several ways in the context of antibiotic resistance:

1. Education and awareness
2. Clinical guidelines and best practices
3. Surveillance and monitoring
4. Multidisciplinary antimicrobial stewardship teams
5. Antibiotic review and prior authorization
6. Technology and decision support systems
7. Collaboration and communication
8. Research and development (13, 14)

5.5 Implementing infection control and prevention measures

Implementing infection control and prevention (ICP) measures is crucial in the approach to antibiotic resistance. By preventing infections from occurring in the first place, we can reduce the need for antibiotics and subsequently minimize the development of antibiotic-resistant bacteria. Here are some key steps in implementing IPC measures:

1. Personal hygiene
2. Personal protective equipment (PPE)
3. Cleaning and disinfection of environmental
4. Cough etiquette and respiratory cleanliness
5. Isolation precautions
6. Antimicrobial stewardship
7. Vaccination programs
8. Surveillance and outbreak investigation
9. Education and training
10. Public awareness (15, 16)

5.6 Alternative therapies and treatment option

Alternative therapies and treatment options can play a supportive role in addressing antibiotic resistance. While it is essential that note the primary and most effective form of treatment for bacterial infections, alternative approaches can complement conventional treatments and

help reduce the risk of antibiotic resistance. Here are some alternative therapies and treatment options that have been explored (17).

- Probiotics: Probiotics are beneficial bacteria that can be consumed to restore the natural balance of microbes in the body.
- Phage therapy: Bacteriophages, often known as phages, are viruses that can target and eradicate particular bacteria. Phage therapy involves using specific phages to infect and eliminate pathogenic bacteria.
- Essential oils: Some essential oils possess antimicrobial properties and have been studied for their potential as alternatives to antibiotics. For example, tea tree oil, oregano oil, and garlic extract (18, 19).
- Herbal medicine: Traditional herbal remedies have been used for centuries to treat various ailments, including infections. Certain herbs, such as berberine-containing plants (e.g., goldenseal, Oregon grape), have demonstrated antimicrobial properties.
- Immunomodulators: Enhancing the body's immune response can help combat infections. Immunomodulators, such as certain vitamins, minerals, and herbal supplements, aim to boost the immune system's ability to fight off pathogens (19, 20).

5.7 Advancements in diagnostic techniques

Advancements in diagnostic techniques have played a significant role in addressing antibiotic resistance by improving the identification and characterization of resistant bacteria. These advancements help guide appropriate antibiotic use, prevent unnecessary prescriptions, and facilitate timely intervention strategies (21). Here are some key advancements in diagnostic techniques related to antibiotic resistance:

- Rapid Diagnostic Tests: Traditional methods of identifying bacteria and determining their susceptibility to antibiotics can be time-consuming, taking several days or more. Rapid diagnostic tests polymerase chain reaction (PCR) and nucleic acid amplification tests (NAATs), provide results within hours.
- Whole Genome Sequencing (WGS): WGS is a powerful technique that establishes an organism's whole genome's DNA sequence (22).
- Mass Spectrometry: Using clinical samples, matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) provides a quick and precise way to identify different bacterial species (23).
- Point-of-Care Testing (POCT): POCT devices are compact, user-friendly diagnostic tools designed at the patient's bedside, producing quick results.
- Metagenomic Sequencing: Metagenomic sequencing involves analyzing the genetic material extracted from a complex mixture of microorganisms present in a sample.
- Digital Health Technologies: Digital health technologies, such as mobile applications and electronic health records, can support the collection, analysis, and sharing of diagnostic data (24, 25).

6. Challenges and Future Directions

Overcoming antibiotic resistance poses several challenges, but there are also promising future directions that can help address this global health threat. Some of the key challenges and future directions include:

6.1 Development of New Antibiotics: Developing new antibiotics is complex, time-consuming, and costly. Pharmaceutical companies have been reluctant to invest in antibiotic research due to financial challenges and the potential for a low return on investment (1).

6.2 Antibiotic Stewardship: Achieving widespread implementation of antibiotic stewardship programs in healthcare settings remains a challenge. Improving prescribing practices, reducing inappropriate use, and optimizing antibiotic use requires changes in behavior among healthcare professionals, patients, and caregivers. Overcoming barriers to implementing stewardship programs, such as limited resources and resistance to change, will be crucial to preserving the effectiveness of existing antibiotics (26).

6.3 One Health Approach: Antibiotic resistance is an interconnected problem across human, animal, and environmental sectors. Adopting a one-health strategy that interplay between the environment and the health of humans and animals is essential (27).

6.4 Surveillance and Monitoring: Strengthening surveillance systems for antibiotic resistance is critical for tracking trends, detecting emerging threats, and informing public health interventions. There is a need for standardized surveillance methodologies, data-sharing mechanisms, and improved laboratory capacity globally to enhance our understanding of the magnitude and spread of antibiotic resistance (28).

6.5 Alternative Approaches: Exploring alternative approaches to combat bacterial infections is gaining attention. This includes developing new treatment modalities like phage therapy, immunotherapies, and novel antimicrobial agents (29).

6.6 Global Collaboration: Addressing antibiotic resistance requires global collaboration and commitment from multiple stakeholders, including governments, healthcare organizations, pharmaceutical companies, researchers, and international agencies. Strengthening international partnerships, sharing best practices, and aligning efforts will be crucial to tackle this global health threat effectively (30).

6.7 Public Awareness and Education: Raising public awareness about antibiotic resistance is essential to drive behavior change. Educating the public about appropriate antibiotic use, the consequences of misuse, and the importance of infection prevention measures can help reduce unnecessary antibiotic consumption and promote responsible use (31).

7. Conclusion

In conclusion, overcoming antibiotic resistance is a complex and urgent global challenge that requires a multi-faceted approach. Antibiotic resistance is the capacity to create mechanisms that render them ineffective, endangering human health and cutting-edge medical technology. To address this issue, various approaches have been developed. These include promoting responsible antibiotic use through education and awareness campaigns, implementing antibiotic stewardship programs in healthcare settings, strengthening surveillance systems to monitor resistance patterns, and fostering international collaboration and partnerships. Additionally, innovation and research are crucial in developing new antibiotics and alternative treatment modalities. This involves incentivizing pharmaceutical companies to invest in antibiotic research, supporting scientific advancements, and exploring novel therapies such as phage therapy, immunotherapies, and nanotechnology-based approaches. It is also important to adopt a one-health strategy in the spread of antibiotic resistance.

References

1. Aslam B, Wang W, Arshad MI, Khurshid M, Muzammil S, Rasool MH et al. Antibiotic resistance: a rundown of a global crisis. Infect Drug Resist.2018;11:1645-58.
2. Reygaert WC. An overview of the antimicrobial resistance mechanisms of bacteria. AIMS Microbiol. 2018;4(3):482-501.
3. Uddin TM, Chakraborty AJ, Khusro A, Zidan BRM, Mitra S, Emran TBet al. Antibiotic resistance in microbes: history, mechanisms, therapeutic strategies and future prospects. J Infect Public Health. 2021Dec1;14(12):1750-66.
4. Llor C, Bjerrum L. Antimicrobial resistance: risk associated with antibiotic overuse and initiatives to reduce the problem. Ther Adv Drug Saf.2014;5(6):229-41.
5. Martin MJ, Thottathil SE, Newman TB. Antibiotics overuse in animal agriculture: A call to action for health care providers. Am J Public Health.2015;105(12):2409-10.
6. Collins AS. Preventing healthcare–associatedinfections. E-book: Hughes, RG (toim.).2008. Patient safety and quality: an evidence-based handbook for nurses (chapter 41). Rockville, (MD): Agency for Healthcare Research and Quality.
7. Terreni M, Taccani M, Pregnolato M. New antibiotics for multidrug-resistant bacterial strains: latest research developments and future perspectives. Molecules.2021;26(9):2671.
8. Davies J, Davies D. Origins and evolution of antibiotic resistance. Microbiol Mol Biol Rev. 2010;74(3):417-33.
9. Peterson E, Kaur P. Antibiotic resistance mechanisms in bacteria: relationships between resistance determinants of antibiotic producers, environmental bacteria, and clinical pathogens. Front Microbiol. 2018 Nov 30;9:2928.
10. Coates AR, Hu Y. Novel approaches to developing new antibiotics for bacterial infections. Br J Pharmacol. 2007;152(8):1147-54.
11. Monciardini P, Iorio M, Maffioli S, Sosio M, Donadio S. Discovering new bioactive molecules from microbial sources. Microb Biotechnol. 2014;7(3):209-20.
12. Sukhum KV, Diorio-Toth L, Dantas G. Genomic and metagenomic approaches for predictive surveillance of emerging pathogens and antibiotic resistance. Clin Pharmacol Ther. 2019;106(3):512-24.
13. Doron S, Davidson LE. Antimicrobial stewardship. Mayo Clin Proc. 2011;86(11):1113-23.
14. Infectious Diseases Society of America (IDSA). Combating antimicrobial resistance: policy recommendations to save lives. ClinInfect Dis. 2011 May 1;52(suppl_5):S397-428.
15. Ackers L, Ackers-Johnson G, Welsh J, Kibombo D, Opio S. Infection prevention control (IPC) and antimicrobial resistance (AMR). Anti-Microb Resist Glob Perspect.2020:53-80.
16. Harun MGD, Anwar MMU, Sumon SA, Hassan MZ, Mohona TM, Rahman A et al. Rationale and guidance for strengthening infection prevention and control measures and antimicrobial stewardship programs in Bangladesh: a study protocol. BMC Health Serv Res. 2022;22(1):1239.
17. Baars EW, Zoen EB, Breitkreuz T, Martin D, Matthes H, von Schoen-Angerer Tet al. The contribution of complementary and alternative medicine to reduce antibiotic use: A narrative review of health concepts, prevention, and treatment strategies. Evid Based Complement Alternat Med.2019;2019:5365608.
18. Abedon ST, Kuhl SJ, Blasdel BG, Kutter EM. Phage treatment of human infections. Bacteriophage.2011;1(2):66-85.
19. Divya Ganeshan S, Hosseinidoust Z. Phage therapy with a focus on the human microbiota. Antibiotics (Basel, Switzerland). 2019;8(3):131.
20. Düzgüneş N, Sessevmez M, Yildirim M. Bacteriophage therapy of bacterial infections: the rediscovered frontier. Pharmaceuticals (Basel). 2021;14(1):34.
21. Vasala A, Hytönen VP, Laitinen OH. Modern tools for rapid diagnostics of antimicrobial resistance. Front Cell Infect Microbiol. 2020;10:308.
22. Ljungström L, Enroth H, Claesson BE, Ovemyr I, Karlsson J, Fröberg Bet al. Clinical evaluation of commercial nucleic acid amplification tests in patients with suspected sepsis. BMC Infect Dis. 2015 Dec;15:199.
23. Peri AM, Stewart A, Hume A, Irwin A, Harris PNA. New microbiological techniques for the diagnosis of bacterial infections and sepsis in ICU including point of care. Curr Infect Dis Rep. 2021 Aug;23(8):12.
24. Bursle E, Robson J. Non-culture methods for detecting infection. AustPrescr. 2016 Oct;39(5):171-5.
25. Wu TF, Chen YC, Wang WC, Fang YC, Fukuoka S, Pride DTet al. A rapid and low-cost pathogen detection platform by using a molecular agglutination assay. ACS Cent Sci. 2018 Nov 5;4(11):1485-94.
26. Amin AN, Dellinger EP, Harnett G, Kraft BD, LaPlante KL, LoVecchio F et al. It's about the patients: practicalantibiotic stewardship in outpatient settings in the United States. Front Med (Lausanne). 2022; 9:901980.
27. Velazquez-Meza ME, Galarde-López M, Carrillo-Quiróz B, Alpuche-Aranda CM. Antimicrobial resistance: one Health approach. VetWorld. 2022;15(3):743-9.
28. Johnson AP. Surveillance of antibiotic resistance. PhilosTrans R Soc Lond B Biol Sci. 2015;370(1670):20140080.
29. Chan BK, Abedon ST, Loc-Carrillo C. Phage cocktails and the future of phage therapy. Future Microbiol. 2013 Jun;8(6):769-83.
30. Veepanattu P, Singh S, Mendelson M, Nampoothiri V, Edathadatil F, Surendran S et al. Building resilient and responsive research collaborations to tackle antimicrobial resistance-Lessons learnt from India, South Africa, and UK. Int J Infect Dis. 2020;100:278-82.
31. Mathew P, Sivaraman S, Chandy S. Communication strategies for improving public awareness on appropriate antibiotic use: bridging a vital gap for action on antibiotic resistance. J Fam Med Prim Care.2019;8(6):1867-71.

Emerging frontier approaches for the treatment of antimicrobial resistance


Saba Khan, Jaya Agnihotri*
Department of Pharmaceutics, MES H.K College of Pharmacy, Jogeshwari (W), Mumbai, Maharashtra-400083, India
jaya.agnihotri@hkcp.edu.in

1.Introduction

Antimicrobial resistance (AMR) represents a far-reaching and substantial danger to public health. The emergence of AMR leads to complications in treating infections, contributing to a rise in mortality rates. The broad occurrence of AMR, along with the excessive utilization of antimicrobial drugs, creates a substantial challenge for the restoration of human and animal populations. Urgent attention is required to devise innovative treatments and therapeutics that can effectively address the emergence and spread of resistant strains. Research indicates that AMR gene development and dissemination are primarily linked to overreliance on health drugs, improper use in veterinary medicine, agricultural practices, and vaccine hesitancy. Thankfully, promising approaches have surfaced to combat resistance effectively. These methods encompass antivirulent therapy, passive immunization, antimicrobial peptides, vaccines, phage therapy, as well as botanical and liposomal nanoparticles. Each of these innovative techniques aims to alleviate the strain on antibacterial drugs, representing cutting-edge approaches in the field. This review article focuses on the importance of employing these advanced therapeutics to combat AMR. To effectively tackle AMR, a comprehensive strategy is essential. This involves harnessing current cutting-edge therapeutics, advancing antimicrobial susceptibility testing and diagnostic techniques, and ensuring prompt clinical responses to contain AMR's spread. Furthermore, there is a need for research to explore new pharmacodynamic properties of antimicrobials and develop methods to maintain host homeostasis after AMR-caused infections. Addressing AMR calls for a multifaceted approach, involving the appropriate use of advanced antimicrobial drugs in conjunction with diverse cutting-edge therapeutics. In summary, AMR represents a pressing global health challenge. However, by employing current state-of-the-art therapeutics, advancing diagnostic and treatment methods, and continuously conducting research, we can aspire to reduce its impact and safeguard the health of both human and animal populations.

2. New antibiotic discovery

2.1. Genome-scale screening technique

Genome mining uncovers new antibiotics through biosynthetic gene clusters (BGC). Tools like anti-SMASH and PRIS identify BGCs. Strategies include ribosome engineering, CRISPR-Cas9, and small molecule elicitor use. Examples include lactocillin, halophile, taromycin A, and piperidine A/B (1-4).

2.2. Semisynthetic engineering

An innovative strategy is employed to enhance existing glycopeptide and lipopeptide antibiotics, resulting in the formation of improved drugs with broader activity (PK) (5). This semisynthetic engineering process utilizes both enzymatic and chemical methods to synthesize novel medications (6). Notably, three vancomycin derivatives, namely telavancin, dalbavancin, and oritavancin, have been successfully produced using this approach (7). Examples of successful applications of this method encompass azithromycin and clarithromycin (erythromycin derivatives), minocycline, doxycycline (tetracycline derivatives), rifampicin, and tigecycline (rifamycin derivatives) (6).

2.3. Innovative retro-biosynthetic algorithm technique

An innovative retro-biosynthetic algorithm proves to be a valuable tool for new antibiotics (8). The compounds Griselimycins and telomycins show promising interactions with specific proteins (9). Computational strategies, such as the hit compound technique, play a key role in the identification of antimicrobial compounds. The open-access facility CO-ADD significantly contributes to the discovery of antimicrobial drugs (10). Notably, initiatives like CARB-X and IMIENABLE actively support the ongoing fight against AMR (11). Prioritizing the challenges posed by AMR and seeking effective broad-spectrum formulations remain crucial goals. Researchers are actively exploring alternative methods to combat AMR through innovative discoveries.

2.4. Intestinal microbiota

The intestinal microbiota serves as a battleground for tackling multidrug-resistant bacteria effectively. Utilizing the intestinal microbiota presents a promising approach in this endeavour. Two potential strategies are preserving the microbiota and employing faecal microbial therapy (FMT). FMT involves the transplantation of beneficial probiotics to combat pathogens (12).

2.5. Antibiotic adjuvants

Preserving antibiotics is vital amid the search for new ones. Adjuvants can block resistance and boost current drugs, improving outcomes. Combining aminoglycoside and penicillin shows synergy against enterococcal infections. Adjuvants enhance drug efficacy when used together, providing a promising approach to fight resistance. Research explores new adjuvants like polyamino-isoprenyl derivatives, farnesyl spermine, and pegylated azelaic acid (13-15).

3. Novel therapy approaches for the treatment of antimicrobial resistance

3.1. Monoclonal antibodies

Monoclonal antibodies (mAb) are considered potential agents to kill bacteria. Humanized monoclonal antibodies (mAbs) are created to reduce immunogenicity. These specialized mAbs function by neutralizing pathogens through complement-mediated responses, eliminating exotoxins, inducing the production of antivirulent antibodies, or directly targeting and killing bacteria. Clinical trials have demonstrated the effectiveness of mAbs, either used alone or in combination with antibiotics, in treating various diseases (16-18).

3.2. Quorum-sensing inhibitors

Antivirulent therapy uses QS inhibitors to deplete bacterial toxicities without hindering pathogen growth (19). QS inhibitors disrupt cell-to-cell communication, reducing adaptive immunity and pathogenicity (20, 21). Quorum quenching is carried out through sequestration, competition, and signal destruction.

3.3. Vaccination

Vaccines play a significant role in addressing AMR. They reduce infection rates caused by difficult-to-treat bacteria and lower the need for antibiotics, decreasing selective pressure for resistance development (22). Specific vaccines have effectively eliminated certain pathogens and reduced antibiotic usage, preventing resistance evolution (23). Pneumococcal and Hib conjugate vaccines have shown success in reducing MDR infections (24, 25). Respiratory virus vaccines combat resistant influenza, curbing antibiotic use for flu treatment (27-28). Vaccination-based strategies offer the potential in combating AMR.

3.4. Antimicrobial peptides

Antimicrobial peptides (AMPs) are small, cationic molecules produced by fungi, bacteria, plants, and vertebrates, including humans (29). They display broad-spectrum activities against various microorganisms, making them promising therapeutic agents (30). AMPs differ in structure, consisting of ribosomal proteins or nonribosomal compounds (31). Their mechanism of action involves membrane disruption, enzyme inhibition, cell division disruption, macromolecular synthesis inhibition, or autolysis induction.

3.5. Algae-mediated treatment

Domestic and agricultural wastewaters play a significant role in the emergence of antibiotic-resistant bacteria and genes due to the incredible use of antibiotics in human and animal health (32). Antibiotic-resistant bacteria and genes are commonly found in wastewater treatment plants (WWTPs) due to the presence of domestic wastewater. Conventional treatment processes like preliminary, primary, and secondary treatments are used to remove traditional pollutants, but specific methods like UV254, ozonation, and chlorination have been investigated to degrade antibiotic-resistant genes (33-36). Microalgae, particularly green algae, have shown potential in efficiently removing contaminants from wastewater and are important for small- to medium-scale municipal wastewater treatment (37). Although green algae have been used for antibiotic treatment, their impact on antibiotic-resistant bacteria and nontarget organisms needs further exploration (38). Eco-friendly biotechnology must be efficient in waste removal while having a low environmental impact. Selective pressure on antibiotic-resistant bacteria by green algae and potential negative effects on nontarget organisms must be examined. The impact of the target antibiotics and effluent after algal treatment was also assessed using rotifers (38, 39). Studies have reported successful algae-mediated treatments for the removal of antibiotic-resistant genes. Reductions in plasmid transformation and induced ciprofloxacin resistance have been observed with freshwater algae treatment (40, 41). Comparisons between algal-based systems and conventional wastewater treatment have shown a greater reduction in antibiotic-resistant bacteria in the former (42). However, further research is needed to fully understand the potential and environmental implications of using green algae in wastewater treatment.

3.6. Bacteriophage therapy

Bacteriophages act as a biocontrol to combat AMR, and their interest has been renewed due to antibiotic resistance (43). Phage therapy utilizes specific lytic bacteriophages as another option to antibiotics, targeting pathogen receptors and causing cell lysis (44). Live phages effectively treat infectious strains and show promise in managing respiratory and systemic diseases (45). Phage therapy offers advantages over antibiotics, including targeted killing of antibiotic-resistant and MDR organisms with minimal side effects on normal flora (46). However, efforts are required for bacteriophage isolation and genetic modifications. Genetically engineered

phages were created to combat E. coli by multiplying and killing the bacteria without causing cell lysis. This approach minimizes inflammatory effects, unlike antibiotics and live lytic phages, which release more endotoxin (47). However, phage therapy cannot fully replace antibiotics due to intrinsic limitations. It is not effective against deeply intracellular pathogens and cannot be administered intravenously due to host immune responses. Therefore, phage therapy is more suitable for easily accessible infections such as pneumonia and wounds. Additionally, there are challenges related to controlling and storing phages, and a proper regulatory framework is needed (48). To address specificity issues, phage cocktails are being developed, combining multiple phages to target various strains in a single infection (49). This allows for enhanced efficacy as one phage can compensate for the inhibition of another. Researchers are also exploring the use of phage endolysins, such as N-RephasinSAL200, as another option to living phages for targeting bacteria (50). Phage therapy has shown promise in combating MDR urinary tract infections by not only lysing bacteria but also inhibiting biofilm formation through polysaccharide depolymerase induction (51).

3.7. Antimicrobial resistance and COVID-19 pandemic

The COVID-19 pandemic has disrupted antibiotic stewardship efforts and led to increased antibiotic usage worldwide. The use of azithromycin and hydrochloroquinone has surged, and hospital admissions have risen, increasing the risk of hospital infections. A study in India showed a 40% increase in antimicrobial-resistant bacterial pathogens due to excessive antibiotic use. This can lead to the transmission of multi-drug-resistant strains. Biocidal agents used outside hospitals may also promote drug-resistant strains. Healthcare workers need to be vigilant, and medical devices should be appropriately maintained to prevent healthcare-related infections (52).

3.8. Nanoparticles

Nanotechnology shows promise in combating antimicrobial resistance (AMR) by using nanoparticles (NPs) with bactericidal activity and acting as drug carriers for antibiotics and AMPs. Metallic NPs damage bacterial membranes and proteins, while functionalized NPs enhance drug efficiency. CAL02 liposomal NPs are studied for severe pneumococcal pneumonia. Nanocarriers deliver inhibitors to target cancer cell resistance. Hydrogels with antibiotics and NPs are explored for AMR control, needing further study (53-56).

3.9. Phytochemicals

Combination therapy with botanical and nutritional approaches, using phytochemicals and plant extracts, is effective against infections and can combat resistance development in microbes. Plant secondary metabolites disrupt microbes through various strategies, making them valuable for enhancing immune response and combating antibiotic resistance (57-59).

4. Conclusions

The long-term outlook for antimicrobial resistance (AMR) is unpredictable. Reflecting on the early days of antibiotic discovery, scientists are both impressed by the positive impact of drug development and concerned about the rise of resistance. A French microbiologist once predicted that certain bacteria would always remain sensitive to penicillin, but now we are facing difficulties in treating common infections due to AMR.

References

1. Liu G, Chater KF, Chandra G, Niu G, Tan H. Molecular regulation of antibiotic biosynthesis in streptomyces. Microbiology and Molecular Biology Reviews. 2013;77(1):112-43.
2. Mao D, Okada BK, Wu Y, Xu F, Seyedsayamdost MR. Recent advances in activating silent biosynthetic gene clusters in bacteria. Current Opinion in Microbiology. 2018;45:156-63.
3. Niu G, Li W. Next-Generation Drug Discovery to Combat Antimicrobial Resistance. Trends in Biochemical Sciences. 2019;44(11):961-72.
4. Ren H, Wang B, Zhao H. Breaking the silence: new strategies for discovering novel natural products. Current Opinion in Biotechnology. 2017;48:21-7.
5. Drug Discovery [Internet]. Hoboken, NJ, USA: John Wiley & Sons, Inc.; 2013 [cited 2023 Jul 26]. Available from: http://dx.doi.org/10.1002/9781118354483
6. Wright PM, Seiple IB, Myers AG. The Evolving Role of Chemical Synthesis in Antibacterial Drug Discovery. Angewandte Chemie International Edition. 2014;53(34):8840-69.
7. Bouza E, Burillo A. Oritavancin: a novel lipoglycopeptide active against Gram-positive pathogens including multiresistant strains. International Journal of Antimicrobial Agents. 2010;36(5):401-7.
8. Johnston CW, Skinnider MA, Dejong CA, Rees PN, Chen GM, Walker CG, et al. Assembly and clustering of natural antibiotics guides target identification. Nature Chemical Biology. 2016;12(4):233-9.
9. Butler MS, Blaskovich MA, Owen JG, Cooper MA. Old dogs and new tricks in antimicrobial discovery. Current Opinion in Microbiology. 2016;33:25-34.
10. Cooper MA. A community-based approach to new antibiotic discovery. Nature Reviews Drug Discovery. 2015;14(9):587-8.
11. Blaskovich MAT, Butler MS, Cooper MA. Polishing the tarnished silver bullet: the quest for new antibiotics. Essays in Biochemistry. 2017;61(1):103-14.
12. Treatment of Recurrent Clostridium difficile Infection With Fecal Transplantation. Gastroenterology Nursing. 2014;37(2):164-5.
13. Borselli D, Brunel JM, Gorgé O, Bolla JM. Polyamino-Isoprenyl Derivatives as Antibiotic Adjuvants and Motility Inhibitors for Bordetella bronchiseptica Porcine Pulmonary Infection Treatment. Frontiers in Microbiology. 2019;10.
14. Troudi A, Fethi M, Selim El Asli M, Bolla JM, Klibi N, Brunel JM. Efficiency of a Tetracycline-Adjuvant Combination Against Multidrug Resistant Pseudomonas aeruginosa Tunisian Clinical Isolates. Antibiotics. 2020;9(12):919.
15. Wang Z, Xiang H, Dong P, Zhang T, Lu C, Jin T, et al. Pegylated azelaic acid: Synthesis, tyrosinase inhibitory activity, antibacterial activity and cytotoxic studies. Journal of Molecular Structure. 2021;1224:129234.
16. Oleksiewicz MB, Nagy G, Nagy E. Anti-bacterial monoclonal antibodies: Back to the future? Archives of Biochemistry and Biophysics. 2012;526(2):124–31.
17. McConnell MJ. Where are we with monoclonal antibodies for multidrug-resistant infections? Drug Discovery Today. 2019;24(5):1132–8.
18. Morrison C. Antibacterial antibodies gain traction. Nature Reviews Drug Discovery. 2015;14(11):737-8.
19. Köhler T, Perron GG, Buckling A, van Delden C. Quorum Sensing Inhibition Selects for Virulence and Cooperation in Pseudomonas aeruginosa. PLoS Pathogens. 2010;6(5):e1000883.
20. Høyland-Kroghsbo NM, Paczkowski J, Mukherjee S, Broniewski J, Westra E, Bondy-Denomy J, et al. Quorum sensing controls the Pseudomonas aeruginosa CRISPR-Cas adaptive immune system. Proceedings of the National Academy of Sciences. 2016;114(1):131-5.
21. Tonkin M, Khan S, Wani MY, Ahmad A. Quorum Sensing - A Stratagem for Conquering Multi-Drug Resistant Pathogens. Current Pharmaceutical Design. 2021;27(25):2835-47.
22. Lu N, Hu Y, Zhu L, Yang X, Yin Y, Lei F, et al. DNA microarray analysis reveals that antibiotic resistance-gene diversity in human gut microbiota is age related. Scientific Reports. 2014;4(1).
23. Chang HH, Cohen T, Grad YH, Hanage WP, O'Brien TF, Lipsitch M. Origin and Proliferation of Multiple-Drug Resistance in Bacterial Pathogens. Microbiology and Molecular Biology Reviews. 2015;79(1):101-16.
24. Ladhani SN, Slack MPE, Andrews NJ, Waight PA, Borrow R, Miller E. Invasive Pneumococcal Disease after Routine Pneumococcal Conjugate Vaccination in Children, England and Wales. Emerging Infectious Diseases. 2013;19(1):61-8.
25. Laxminarayan R, Matsoso P, Pant S, Brower C, Røttingen JA, Klugman K, et al. Access to effective antimicrobials: a worldwide challenge. The Lancet. 2016;387(10014):168-75.
26. Fleming-Dutra KE, Hersh AL, Shapiro DJ, et al. Prevalence of inappropriate antibiotic prescriptions among us ambulatory care visits, 2010-2011. JAMA. 2016;315:1864-1873.
27. Deresinski S, Schirmer P. Faculty Opinions recommendation of Inappropriate antibiotic prescribing in managed care subjects with influenza. Faculty Opinions – Post-Publication Peer Review of the Biomedical Literature. 2011.
28. Chen CS. Combinational Synergy of Antibiotics and Antimicrobial Peptides. Journal of Translational Proteomics Research. 2017;3:6-11.
29. Pushpanathan M, Gunasekaran P, Rajendhran J. Antimicrobial Peptides: Versatile Biological Properties. International Journal of Peptides. 2013;2013:1-15.
30. Zalewska M, Błażejewska A, Czapko A, Popowska M. Antibiotics and antibiotic resistance genes in animal manure–consequences of its application in agriculture. Frontiers in Microbiology. 2021;12:640.
31. Crowther GS, Baines SD, Todhunter SL, Freeman J, Chilton CH, Wilcox MH. Evaluation of NVB302 versus vancomycin activity in an in vitro human gut model of Clostridium difficile infection. Journal of Antimicrobial Chemotherapy. 2012;68(1):168-76.
32. Chang PH, Juhrend B, Olson TM, Marrs CF, Wigginton KR. Degradation of Extracellular Antibiotic Resistance Genes with UV254 Treatment. Environmental Science & Technology. 2017;51(11):6185-92.
33. Czekalski N, Imminger S, Salhi E, Veljkovic M, Kleffel K, Drissner D, et al. Inactivation of Antibiotic Resistant Bacteria and Resistance Genes by Ozone: From Laboratory Experiments to Full-Scale Wastewater Treatment. Environmental Science & Technology. 2016;50(21):11862-71.
34. Destiani R, Templeton MR. Chlorination and ultraviolet disinfection of antibiotic-resistant bacteria and antibiotic resistance genes in drinking water. AIMS Environmental Science. 2019;6(3):222-41.
35. He H, Zhou P, Shimabuku KK, Fang X, Li S, et al. Degradation and Deactivation of Bacterial Antibiotic Resistance Genes during Exposure to Free Chlorine, Monochloramine Chlorine Dioxide, Ozone, Ultraviolet Light, and Hydroxyl Radical. Environ. Sci. Technol. 2019;53(4):2013-2026.
36. Mohsenpour SF, Hennige S, Willoughby N, Adeloye A, Gutierrez T. Integrating micro-algae into wastewater treatment: A review. Science of The Total Environment. 2021;752:142168.

37. Yu Y, Zhou Y, Wang Z, Torres OL, Guo R, Chen J. Investigation of the removal mechanism of antibiotic ceftazidime by green algae and subsequent microbic impact assessment. Scientific Reports. 2017;7(1).
38. Du Y, Zhang S, Guo R, Chen J. Understanding the algal contribution in combined UV-algae treatment to remove antibiotic cefradine. RSC Advances. 2015;5(74):59953-9.
39. Grimes KL, Dunphy LJ, Kolling GL, Papin JA, Colosi LM. Algae-mediated treatment offers apparent removal of a model antibiotic resistance gene. Algal Research. 2021;60:102540.
40. Grimes KL, Dunphy LJ, Loudermilk EM, Melara AJ, Kolling GL, Papin JA, Colosi LM. Evaluating the efficacy of an algae-based treatment to mitigate elicitation of antibiotic resistance. Chemosphere. 2019;237:124421.
41. Cheng X, Delanka-Pedige HMK, Munasinghe-Arachchige SP, Abeysiriwardana-Arachchige ISA, Smith GB, Nirmalakhandan N, et al. Removal of antibiotic resistance genes in an algal-based wastewater treatment system employing Galdieria sulphuraria: A comparative study. Science of The Total Environment. 2020;711:134435.
42. Gravitz L. Turning a new phage. Nature Medicine. 2012;18(9):1318-20.
43. Viertel TM, Ritter K, Horz HP. Viruses versus bacteria--novel approaches to phage therapy as a tool against multidrug-resistant pathogens. Journal of Antimicrobial Chemotherapy. 2014;69(9):2326-36.
44. Kaur G, Rajesh S, Princy SA. Plausible Drug Targets in the Streptococcus mutans Quorum Sensing Pathways to Combat Dental Biofilms and Associated Risks. Indian Journal of Microbiology. 2015;55(4):349-56.
45. Chatterjee M, Anju CP, Biswas L, Anil Kumar V, Gopi Mohan C, Biswas R. Antibiotic resistance in Pseudomonas aeruginosa and alternative therapeutic options. International Journal of Medical Microbiology. 2016;306(1):48-58.
46. Matsuda T, Freeman TA, Hilbert DW, Duff M, Fuortes M, Stapleton PP, et al. Lysis-deficient bacteriophage therapy decreases endotoxin and inflammatory mediator release and improves survival in a murine peritonitis model. Surgery. 2005;137(6):639-46.
47. Verbeken G, De Vos D, Vaneechoutte M, Merabishvili M, Zizi M, Pirnay JP. European regulatory conundrum of phage therapy. Future Microbiology. 2007;2(5):485-91.
48. Hurley MN, Cámara M, Smyth AR. Novel approaches to the treatment of Pseudomonas aeruginosa infections in cystic fibrosis. European Respiratory Journal. 2012;40(4):1014-23.
49. Jun SY, Jung GM, Yoon SJ, Choi YJ, Koh WS, Moon KS, et al. Preclinical Safety Evaluation of Intravenously Administered SAL200 Containing the Recombinant Phage Endolysin SAL-1 as a Pharmaceutical Ingredient. Antimicrobial Agents and Chemotherapy. 2014;58(4):2084-8.
50. Chegini Z, Khoshbayan A, Vesal S, Moradabadi A, Hashemi A, Shariati A. Bacteriophage therapy for inhibition of multi drug-resistant uropathogenic bacteria: a narrative review. Annals of Clinical Microbiology and Antimicrobials. 2021;20(1).
51. Ahmed S, Ahmed MZ, Rafique S, Almasoudi SE, Shah M, Jalil NA, Ojha SC. Recent approaches for downplaying antibiotic resistance: Molecular mechanisms. BioMed Research International. 2023.
52. Choudhury S, Medina-Lara A, Smith R. Antimicrobial resistance and the COVID-19 pandemic. Bulletin of the World Health Organization. 2022;100(05):295-295A.
53. Redl FX, Black CT, Papaefthymiou GC, Sandstrom RL, Yin M, Zeng H, et al. Magnetic, Electronic, and Structural Characterization of Nonstoichiometric Iron Oxides at the Nanoscale. Journal of the American Chemical Society. 2004;126(44):14583-99.
54. Piras AM, Maisetta G, Sandreschi S, Gazzarri M, Bartoli C, Grassi L, et al. Chitosan nanoparticles loaded with the antimicrobial peptide temporin B exert a long-term antibacterial activity in vitro against clinical isolates of Staphylococcus epidermidis. Frontiers in Microbiology. 2015;06.
55. Li X, Robinson SM, Gupta A, Saha K, Jiang Z, Moyano DF, et al. Functional Gold Nanoparticles as Potent Antimicrobial Agents against Multi-Drug-Resistant Bacteria. ACS Nano. 2014;8(10):10682-6.
56. Ahangari A, Salouti M, Heidari Z, Kazemizadeh AR, Safari AA. Development of gentamicin-gold nanospheres for antimicrobial drug delivery toStaphylococcalinfected foci. Drug Delivery. 2013;20(1):34-9.
57. Choudhari S, Priya VV, Gayathri R. Herbal Remedies for Swine Flu. Research Journal of Pharmacy and Technology. 2016;9(10):1789.
58. Potroz M, Cho NJ. Natural Products for the Treatment of Trachoma and Chlamydia trachomatis. Molecules. 2015;20(3):4180-203.
59. Birdi T, Gupta P, Daswani P. Approaches in fostering quality parameters for medicinal botanicals in the Indian context. Indian Journal of Pharmacology. 2014;46(4):363.

Methicillin-resistant *Staphylococcus aureus* (MRSA) efflux pump inhibitors from natural products

Pallavi Ahirrao
Chandigarh College of Pharmacy, CGC, Landran, Mohali, Punjab – 140307
pallavi.ccp@cgc.edu.in

Abstract
Among different nosocomial infections, *Staphylococcus aureus*, a Gram-positive bacterium, is a highly adaptive human pathogen. Over the years it had acquired resistance to multiple classes of antibiotics including methicillin. The multidrug resistance towards multiple antibiotics and poor pipeline of safe and effective drugs has rendered bacterial infections a life-threatening problem. Multidrug efflux pumps play an essential role in antibiotic resistance by extrusion of drugs via different mechanisms. Natural products especially derived from plants have emerged as an important source of effective efflux pump inhibitors. In this article different classes of plant- and microbe-derived natural products have been described as efflux pump inhibitors of MRSA that act synergistically in combination with antibiotics to modulate efflux pump-mediated extrusion of antibiotics and thereby help in combating the multidrug resistance.

1. Introduction

Staphylococcus aureus is a major cause of nosocomial- and community-acquired infections. the emergence of methicillin-resistant *Staphylococcus aureus* (MRSA) has occurred over the years which contributes to the development of new strains resistant against multiple classes of antibiotics (1). Around 90–95% of S. aureus strains worldwide are found to be resistant to penicillin, and methicillin-resistant strains account for 70–80% of its total count in most Asian countries (2). At least 50, 000 deaths are recorded due to S. aureus infection in Europe every year, and it is predicted that infections due to drug resistance would be the reason for the deaths of nearly ten million people worldwide by 2050 (3). MRSA is a leading cause of endocarditis, bacteremia, soft tissue skin infections, and hospital acquired infections. Since 1990, there has been a rapid spread of MRSA infections in the human community which poses a great challenge for their treatment. It is already known that staphylococci are able to act against each new antimicrobial agent by adopting one or more resistance mechanisms. Several mechanisms associated with the resistance to antibiotics include target protein mutation, antibiotic inactivation by enzymes, or antibiotic accumulation inhibition due to overexpression of the efflux system in bacterial cells. Among these, drug efflux is the most widespread reason for antimicrobial resistance (4). Gram-positive bacteria such as S. aureuslacks an outer membrane. Efflux pumps are helpful in limiting the accumulation of toxic compounds within the cell. So far, various efflux pumps have been discovered in microorganisms. These EPs aremainly classified in five different superfamilies, such as (1) adenosine triphosphate-binding cassette transporters (ABC), (2) multidrug and toxic compound extrusion (MATE), (3) major facilitator superfamily (MFS), (4) small multidrugresistance (SMR), and (5) resistance nodulation division (RND) family (Figure 1) (5-7).

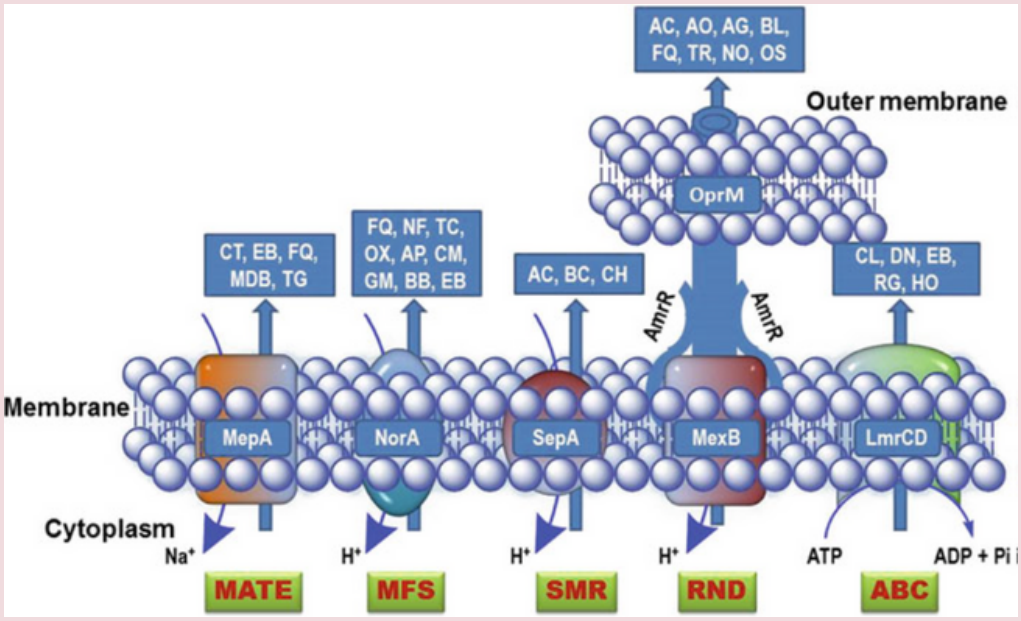


Figure 1. EPs and the substrates that are effluxed out of the bacterial cell (Jachak et al. 2012). ABC ATP-binding cassettes, RND resistance nodulation division, SMR small multidrug resistance, MFS major facilitator superfamily, MATE multidrug and toxic compound extrusion, AC acriflavine, AG aminoglycoside, AO acridine orange, AP ampicillin, BB berberine, BL β-lactam, BC benzalkonium chloride, CH chlorhexidine, CL cholate, CM chloramphenicol, CT ceftazidime, DN daunomycin, EB ethidium bromide, FQ fluoroquinolones, GM gentamicin, HO Hoechst 33342, MDB monovalent and divalent biocides, NF norfloxacin, NO novobiocin, OS organic solvents, OX oxacillin, RG rhodamine, TC tetracyclines, TG tigecycline, TR triclosan (adopted from: Pallavi Ahirrao et.al. In: Kumar, V., Shriram, V., Paul, A., Thakur, M. (eds) Antimicrobial Resistance. Springer, Singapore. (2022), https://doi.org/10.1007/978-981-16-3120-7_19)

The above efflux pump families have been classified on the basis of amino acid sequence similarity, single or multiple components, specificity, energy source, and the number of transmembrane spanning regions. The ABC, MATE, SMR, and MFS superfamilies have been widely distributed in Gram (+)ve and Gram (-)ve bacteria whereas the RND family is distributed in Gram (-)ve bacteria only.

Among all MDR microorganisms, MRSA strain is of main concern, liable for both hospital- and community-acquired infections (8). Among MRSA, NorA overexpressed strains are the most common ones (9). MsrA efflux pump of the ABC transporter family causes resistance to streptogramins and macrolides. MsrA is a transmembrane protein made up of 488 amino acids with two ATP-binding motifs.

Efflux pumps are an important and major antibacterial drug target. Hence, it is necessary to identify and develop potent efflux pump inhibitors (10-11). Efflux pumps can be inhibited by the following strategies: a) interference in genetic regulation by deregulating the EP expression, b) antibiotic redesigning that are considered as substrates earlier, c) suppressing the functional EPs assembly, d) avoiding the substrate binding by blocking the active site, and e) disintegrating the energy mechanism liable for reinvigorating the Eps (12). To tackle antibiotic resistance, drug resistance reversal agents especially efflux pump modulators/inhibitors would be promising leads. These compounds may possess either

antimicrobial activity of their own or possess the ability to enhance the activity of ineffective antibiotics by inhibiting/modulating efflux pumps. Thus, the susceptibility or sensitivity of resistant strains to antibacterial agents can be reinstated with the aid of EPIs (13-14). The actions of EPI mechanisms are not precisely known yet. But, it has been suggested that the inhibitor binds directly to the pump and thus blocks it competitively or noncompetitively with the substrates. By inhibiting ATP binding or by disrupting the proton gradient, EPIs can cause depletion of energy. A complex of EPI with an antibiotic enhances the entry of an antibiotic into the bacterial cell and further inhibits the efflux of an antibiotic due to the larger complex size (15-16). To combat antibiotic resistance, presently the researchers are working on the development of synergistic antibiotic combinations to reduce the dose of antibiotics many times. In this regard, drug resistance reversal agents especially efflux pump inhibitors (EPIs)/modulators would be the promising agents.

2. Natural product inhibitors of efflux pumps

Historically NPs have been a major source of biologically active molecules exhibiting numerous scaffolds and displaying various activities against both noninfectious and infectious diseases. The NPs are formed biogenetically by the processes catalyzed using enzymes that are highly regio-, enantio-, and diastereospecific. A few efflux pumps selectively efflux out a particular class of antibiotics, while other EPs extrude a diverse class of antibiotics; these are termed as MDR. EPIs may be useful in restoring the clinical efficacy of some earlier antibiotics, by enhancing the potency of antibiotics or by decreasing their resistance development. Numerous natural products are known to act as EPIs on different EPs located on the bacterial cell membrane (17).

2.1. *S. aureus* NorA multidrug efflux pump inhibitors

In MF superfamily, the most studied example is NorA multidrug transporter that contributes to the resistance of S. aureus. Berberine and fluoroquinolones like norfloxacin and ciprofloxacin are effluxed out of the cell by NorA EP (18).

2.1.1 Polyphenols

2.1.1.1 - Arylbenzofuran

SpinosanA from Dalea spinosa, at 48 µM, Reduced Berberine MIC by Eightfold against Wild-Type S. aureus, whereas, (+)-Medicarpin at 56 µM decreased Berberine MIC by Fourfold (19).

2.1.1.2- N-Caffeoylphenylkylamides

N-Trans-Feruloyl-4´-O-Methyldopamine isolated from Mirabilis jalapa exhibited moderate EPI activity against NorA-Overexpressed S. aureus 1199B strain. It showed an eightfold reduction in the Minimum Inhibitory Concentrationof Norfloxacin at 100 µg/mL (20).

2.1.1.3 Caffeoylquinic Acids

40,5'-O-caffeoylquinic acid isolated from chloroform extract of Artemisia absinthium potentiated activity of Berberine by eight-fold, EtBr by 4-fold,and Fluoroquinolones, Ciprofloxacin, and Norfloxacin by 4–Eight-Fold in NorA overexpressed S. aureus Strain (21). Curcumin was reported to exhibit significant inhibition of NorA EP in S. aureus. There is an Eightfold Reduction of Ciprofloxacin MIC at 25 µM by curcumin. The molecular modeling study of curcumin with the human Pgp and NorA efflux protein revealed favorable binding interactions (22).

2.1.1.4 Chalcones

A chalcone compound characterized from Dalea versicolor showed a four-fold increase in berberine activity against MDR S. aureus at 10 µg/mL (23). 3, 4'-dihydroxy-3,4,5-trimethoxy-chalcone isolated from the flowers of Arrabidaea brachypoda showed a significant decrease in MIC of norfloxacin byfourfold (24).

2.1.1.5 Coumarins

A 20-fold reduction of norfloxacin MIC in S. aureus-resistant strains (MRSA16565, 9543, 5, and 7) was observed due to coumarins, viz., 4-{[(E)-5-(3,3-dimethyl-2-oxiranyl)-3-methyl-2-pentenyl]oxy}-7H-furo(3,2-g)chromen-7-one and 7-{[(E)-5-(3,3-dimethyl-2-oxiranyl)-3-methyl-2-pentenyl]oxy}-2H-2-chromenone isolated from grapefruit oil at 35.7 µg/mL and 30 µg/mL concentrations, respectively (25). The ciprofloxacin MIC reduced from 10-80 µg/mL to 2.5-5 µg/mL and of EtBr from 4-16 µg/mL to0.5–2 µg/mL by galbanic acid (A10), a sesquiterpene coumarin isolated from theroots of Ferulaszowitsiana, against several S. aureus-resistant clinical isolatestrains, at 300 µg/mL (26).

2.1.1.6 Flavones and flavonols

Chrysosplenol-D and chrysoplenetin, methoxy flavonols reported from the herbaceous plant Artemisia annua, showed inhibition of S. aureus growth at a sub inhibitory concentration (30 µg/mL) with MIC of 25 µg/mL and6.25 µg/mL, respectively (27).

2.1.1.7 Flavonolignans

50-Methoxyhydnocarpin-D (5´-MHC-D), a flavonolignan reported from Berberis aetnensis leaves, exhibited EPI activity by reducing norfloxacin MIC to 0.25 µg/mL at 10 µg/mL for wild-type S. aureus (27).

2.1.1.8 Isoflavones

Isoflavones, viz., genistein, orobol, and biochanin A reported from Lupinus argenteus, decreased norfloxacin MIC by 2–4-folds againstS. aureus mutant strain, at10 µg/mL. Spinosan A, an isoflavone compound characterized from Dalea spinosa, at 48 µM concentration showed an eightfold reduction of berberine MIC (89 µM) in wild-type S. aureus (28).

2.1.1.9 Tannins

Catechin compounds, viz., epicatechin gallate and epigallocatechin gallate, increased the norfloxacin MIC by fourfold in wild-type (SA 1199) and NorA-overexpressed S. aureus (SA1199B) strain, at 20 µg/mL (29).

2.1.2 Terpenoids

Ferruginol characterized from Chamaecyparis lawsoniana exhibited NorA pump inhibitory activity in S. aureus-resistant strain. Ferruginol at a subinhibitory concentration (2 µg/mL) showed a twofold potentiation of norfloxacin againstSA1199B strain (30).

2.1.3 Oligosaccharides

Five murucoidins (XII–XVI) were isolated and characterized from Ipomoea murucoides. Murucoidin XIV at 5 µg/mL exhibited a fourfold increase in norfloxacin activity against S. aureus strains (31).

2.1.4 Alkaloids

Piperine, a piperidine alkaloid characterized from Piper nigrum fruits, displayed no growth of S. aureus mutant at 1 µg /mL concentration of ciprofloxacin when it was co administered at 50 µg /mL (32).

2.2. Miscellaneous S. aureus and MRSA efflux pump inhibitors of natural product origin

The essential oil extracted from Origanum vulgare L. as well as its constituents, viz.,carvacrol and thymol, showed EPI activity. The essential oil showed a four fold reduction of tetracycline MIC (64 µg/mL to 16 µg/mL) whereas carvacrol and thymol exhibited a twofold reduction of tetracycline MIC (64 µg/mL to 32 µg/mL) against S. aureus IS-58 strain over expressing TetK efflux pump (33). Essential oil and its major constituent, α-pinene(C03), extracted from Croton grewioides leaves showed EPI activity in SA-1199B (NorA-overexpressed strain) and IS-58 (TetK-overexpressed strain) by 64-fold and four-fold modulation in MIC of tetracycline and norfloxacin respectively (34). Nigella sativa essential oil and its constituents carvacrol, thymoquinone, and p-cymene were studied for their antibacterial effect and modulation of antibiotic resistance in methicillin-sensitive ATCC25923 and methicillin-resistant MRSA 272123 clinical isolate of S. aureus. All these constituents and essential oil displayed MIC values in mM range, indicating a weak antibacterial effect.

A microbial natural product, 2-(2-aminophenyl) indole isolated from Streptomyces sp. IMTB 2501 was found as the most potent NorA inhibitor, decreasing the MIC of ciprofloxacin, moxifloxacin, norfloxacin, and chloramphenicol in SA-1199B (NorA-overexpressed strain) by 64-, 16-, 4-, and 4-folds (FICI_0.5), respectively (35).

3. Conclusion

Antibiotic resistance is emerging at an alarming rate. Thus, there is an urgent unmet need to develop alternative therapies that either reduce the bacterial resistance to antibiotics or potentiate the activity of existing antibiotics. Efflux pumps are one of the major targets that confer resistance to clinically used antibiotics. Presently no efflux pump inhibitor combination with existing antibiotics is clinically approved to tackle antimicrobial resistance. Over the decades natural products have demonstrated multiple biological activities in biomedical research and served as an important source of lead molecules in drug discovery and development.

In this article, natural compounds with potential efflux pump inhibition activity are described. This article describes the natural products that showed activity against NorA, efflux pump responsible for extrusion of drugs in S. aureus and MRSA. However, no plant/fungus/marine-derived antibiotic is used clinically yet. Since isolation and identification of plant-derived drugs are tedious and time consuming, in silico methodology and high-throughput screening can be utilized. A majority of bioactives discussed above have shown promising results as potent EPIs, determined using in vitro studies. The in vivo animal model studies and human clinical trials would be required to determine antibacterial action, efficacy, and toxicity studies to optimize a high therapeutic efficacy dosage of EPIs at an acceptable toxicity level.

References

1. Alibayov B, Baba-Moussa L, Sina H, Zdeňková K, Demnerová K. Staphylococcus aureus mobile genetic elements. Mol Biol Rep. 2014;41:5005-18.
2. Lindsay JA, Holden M. T. Staphylococcus aureus: superbug, super genome. Trends Microbiol. 2004;12:378–385.
3. de Kraker ME, Stewardson AJ, Harbarth S. Will 10 million people die a year due to antimicrobial resistance by 2050? PLoS Med. 2016;13:e1002184.
4. Alekshun MN, Levy SB. Molecular Mechanisms of Antibacterial Multidrug Resistance. Cell. 2007;128:1037-1050.
5. Bohn C, Bouloc P. The Escherichia coli cmlA gene encodes the multidrug efflux pump Cmr/MdfA and is responsible for isopropyl-β-D-thiogalactopyranoside exclusion and spectinomycin sensitivity. J Bacteriol. 1998;15;180(22):6072-5.
6. Jachak SM, et al. In: Antimicrobial drug discovery: emerging strategies. CAB International, Wallingford, UK. 2012;62-76.
7. Zechini B, Versace I. Inhibitors of multidrug resistant efflux systems in bacteria. Recent Pat Antiinfect. Drug Discov. 2009;4:37-50.
8. Craft KM, Nguyen JM, Berg LJ, Townsend SD. Methicillin-resistant Staphylococcus aureus (MRSA): antibiotic-resistance and the biofilm phenotype. Med Chem Comm. 2019;10:1231-1241.
9. Costa SS, Viveiros M, Amaral L, Couto I. Multidrug efflux pumps in. Staphylococcus aureus: an update. Open Microbiol J. 2013;7:59.
10. Edelsberg J, Weycker D, Barron R, Li X, Wu H, et al. Prevalence of antibiotic resistance in US hospitals. Diagn Microbiol Infect Dis. 2014;78:255-262.
11. Mahmood YH, Jamshidi S., Sutton J.M., Rahman K.M. Current advances in developing inhibitors of bacterial multidrug efflux pumps. Curr Med Chem. 2016;23:1062-1081.
12. Sharma A, Gupta VK, Pathania R. Efflux pump inhibitors for bacterial pathogens: From bench to bedside. Indian J Med Res. 2019;149:129.
13. Costa SS, Viveiros M, Amaral L, Couto I. Multidrug efflux pumps in. Staphylococcus aureus: an update. Open Microbiol J. 2013;7:59.
14. Pagès JM, Amaral L. Mechanisms of drug efflux and strategies to combat them: challenging the efflux pump of Gram-negative bacteria. Biochim Biophys Acta. 2009;1794:826-833.
15. Opperman TJ, Nguyen ST. Recent advances toward a molecular mechanism of efflux pump inhibition. Front Microbiol. 2015;6:421.
16. Zloh M, Kaatz LW, Gibbons S. Inhibitors of multidrug resistance (MDR) have affinity for MDR substrates. Bioorg Med Chem Lett. 2004;14:881–885.
17. Ahmed MA, Borsch CM, Neyfakh AA, Schuldiner S. Mutants of the Bacillus subtilis multidrug transporter Bmr with altered sensitivity to the antihypertensive alkaloid reserpine. J Biol Chem. 1993;268(15):11086-9.
18. Kumar S, Mukherjee MM, Varela MF. Modulation of bacterial multidrug resistance efflux pumps of the major facilitator superfamily. Int J Bacteriol. 2013;204141.
19. Belofsky G, Carreno R, Lewis K, Ball A, Casadei G, Tegos GP. Metabolites of the “smoke tree”, Dalea spinosa, potentiate antibiotic activity against multidrug-resistant Staphylococcus aureus. J Nat Prod. 2006;69(2):261-4.
20. Michalet S, Cartier G, David B, Mariotte AM, Dijoux-Franca MG, et al. N-caffeoylphenalkylamide derivatives as bacterial efflux pump inhibitors. Bioorg Med Chem Lett. 2007;17(6):1755-8.
21. Fiamegos YC, Kastritis PL, Exarchou V, Han H, Bonvin AM, et al. Antimicrobial and efflux pump inhibitory activity of caffeoylquinic acids from Artemisia absinthium against gram-positive pathogenic bacteria. PLoS One. 2011;6(4):e18127.
22. Joshi P, Singh S, Wani A, Sharma S, Jain SK, et al. Osthol and curcumin as inhibitors of human Pgp and multidrug efflux pumps of Staphylococcus aureus: reversing the resistance against frontline antibacterial drugs. Med Chem Comm. 2014;5(10):1540-7.
23. Holler JG, Slotved HC, Mølgaard P, Olsen CE, Christensen SB. Chalcone inhibitors of the NorA efflux pump in Staphylococcus aureus whole cells and enriched everted membrane vesicles. Bioorg Med Chem. 2012;20(14):4514-21.
24. Rezende-Júnior LM, Andrade LM, Leal AL, Mesquita AB, Santos AL, et al. Chalcones isolated from Arrabidaea brachypoda flowers as inhibitors of NorA and MepA multidrug efflux pumps of Staphylococcus aureus. Antibiotics. 2020;9(6):351.
25. Abulrob AN, Suller MT, Gumbleton M, Simons C, Russell AD. Identification and biological evaluation of grapefruit oil components as potential novel efflux pump modulators in methicillin-resistant Staphylococcus aureus bacterial strains. Phytochemistry. 2004;65(22):3021-7.
26. Bazzaz BS, Memariani Z, Khashiarmanesh Z, Iranshahi M, Naderinasab M. Effect of galbanic acid, a sesquiterpene coumarin from Ferula szowitsiana, as an inhibitor of efflux mechanism in resistant clinical isolates of Staphylococcus aureus. Braz J Microbiol. 2010;41:574-80.
27. Stermitz FR, Tawara-Matsuda J, Lorenz P, Mueller P, Zenewicz L, Lewis K. 5‘-Methoxyhydnocarpin-D and Pheophorbide a: Berberis species components that potentiate Berberine growth inhibition of resistant staphylococcus a ureus. J Nat Prod. 2000;63(8):1146-9.
28. Morel C, Stermitz FR, Tegos G, Lewis K. Isoflavones as potentiators of antibacterial activity. J Agric Food Chem. 2003;51(19):5677-9.
29. Gibbons S, Udo EE. The effect of reserpine, a modulator of multidrug efflux pumps, on the in vitro activity of tetracycline against clinical isolates of methicillin resistant Staphylococcus aureus (MRSA) possessing the tet (K) determinant. Phytother Res. 2000;14:139-40.
30. Smith EC, Williamson EM, Wareham N, Kaatz GW, Gibbons S. Antibacterials and modulators of bacterial resistance from the immature cones of Chamaecyparis lawsoniana. Phytochemistry. 2007b;68(2):210-7.
31. Chérigo L, Pereda-Miranda R, Gibbons S. Bacterial resistance modifying tetrasaccharide agents from Ipomoea murucoides. Phytochemistry. 2009;70(2):222-7.
32. Khan IA, Mirza ZM, Kumar A, Verma V, Qazi GN. Piperine, a phytochemical potentiator of ciprofloxacin against Staphylococcus aureus. Antimicrob Agents Chemother. 2006;50(2):810-2.
33. Cirino IC, Menezes-Silva SM, Silva HT, de Souza EL, Siqueira-Júnior JP. The essential oil from Origanum vulgare L. and its individual constituents carvacrol and thymol enhance the effect of tetracycline against Staphylococcus aureus. Chemotherapy. 2014;60(5-6):290-3.
34. de Medeiros VM, do Nascimento YM, Souto AL, Madeiro SA, de Oliveira Costa VC, et al. Chemical composition and modulation of bacterial drug resistance of the essential oil from leaves of Croton grewioides. MicrobPathog. 2017;111:468-71.
35. Tambat R, Jangra M, Mahey N, Chandel N, Kaur M,et al. Microb – Derived Indole Metabolite Demonstates Potent Multidrug Efflux Pump Inhibition in Staphylococcus aureus. Frontiers Microbiol. 2019;10:2153.

Antibiotic adjuvants: A promising approach to overcome antibiotic resistance

Bindu Yadav
Bombay college of Pharmacy, Mumbai, India
bindu.yadav@bcp.edu.in

Abstract
The problem of antibiotic resistance is on the rise, with multidrug-resistant strains emerging even to the last resort antibiotics. In such a scenario, it is prudent to delve into the varying mechanisms of resistance to existing antibiotics and target them to improve antibiotic efficacy. Non-antibiotic compounds called antibiotic adjuvants which target bacterial resistance can be used in combination with obsolete drugs for an improved therapeutic regime. The field of "antibiotic adjuvants" has gained significant traction in recent years where mechanisms other than β-lactamase inhibition have been explored. The major focus of this review is how to target these resistance mechanisms by the use of antibiotic adjuvants. Different types of direct acting and indirect resistance breakers are discussed including enzyme inhibitors, efflux pump inhibitors, inhibitors of teichoic acid synthesis, and other cellular processes. Antibiotic–adjuvant combinatorial therapy indeed has immense potential to be used as an upcoming orthogonal strategy to conventional antibiotic discovery.

1. Introduction

Microbial infections are one of the most serious risks to public health globally, putting significant cost strain on global healthcare. Antimicrobial resistance (AMR) contributed to the current issue (1). According to a recent analysis, Antimicrobial resistance (AMR) kills roughly 0.7 million people per year, which is anticipated to rise to 10 million by 2050 (2). This would impose a significant burden on global spending on healthcare. Even after launching the Global Antimicrobial Resistance and Use Surveillance System (GLASS) in 2015, lower respiratory tract infections and diarrheal diseases were among the top ten worldwide causes of mortality in 2019, accounting for 4 million fatalities (3).

Rather than investing in random semi-synthetic adaptations of existing medications, it is better to recycle the present antibiotic arsenal by targeting particular causes of resistance and conquering them. Physical compound combinations provide structural and mechanistic understanding that may be used to develop sensible semi-synthetic changes. As a result, antibiotic development would be more simplified. Aside from combinations of two active compounds, another interesting class of combinations is those composed of an antibiotic and an non-antibiotic molecule that only targets the mechanism of resistance to the said antibiotic, indirect resistance elements, or nonessential resistance pathways. This combination would eliminate the redundant usage of an antimicrobial by making it beneficial in the treatment of multidrug-resistant types of bacteria (4, 5).

Combinations of β-lactam/β-lactamase inhibitors, for example, are clinically approved and

more are under clinical studies. Given the effectiveness of these combinations, it is worthwhile to investigate new mechanisms of resistance that may be addressed by similar compounds in order to restore the efficacy of existing antibiotics. These substances are known as "antibiotic adjuvants" because they have little or no antibiotic action (6).

2. Antibiotic adjuvants

Antibiotic adjuvants are compounds that enhance the action of current antibiotics by reducing or directly inhibiting resistance mechanisms. The concept of antibiotic adjuvants stems from clinically effective combinations of two or more antibiotics. These have been empirically employed to produce synergy, broaden the range of activity, and overcome resistance. In contrast to traditional antibiotic combinations, antibiotic adjuvants have no or little antibacterial action on their own. Antibiotic adjuvants can be roughly characterized as direct, indirect, or host-modulating resistance breakers based on their target profile. To potentiate antibiotics, antibiotic adjuvants target many active and passive resistance mechanisms in bacteria. All these different types of resistance breakers are discussed in the subsequent sections (6).

2.1 Direct resistance breakers

These are Class I adjuvants that function with antibiotics on resistance-causing bacterial targets. These adjuvants directly suppress antibiotic resistance mechanisms such as inhibiting enzymes, efflux pumps, or new targets that compensate for the original targets. The only clinically approved adjuvants in use now are β-lactamases lactamase inhibitors, which inactivate β-lactamases (7).

2.2 Indirect resistance breakers

Antibiotic resistance in bacteria can be caused by an inherent genetic connection, physiological variables, or the presence of generic treatment evasion mechanisms. These comprise a variety of interdependent aspects that are critical for bacterial resistance components. These characteristics, in addition to the actual aspects of resistance, can be possible targets for creating adjuvants. Through this technique, antibiotic action can be resurrected by discovering non-evident synergy in the enormous nonessential gene space. Membrane-targeting chemicals are another type of adjuvant that can have many effects such as inhibiting efflux machinery and permeabilizing the bacterial membrane for antibiotic penetration (7, 8).

2.3 Adjuvants targeting host processes

The potency of antibiotics within infected organisms can be improved by using a number of host defence mechanisms. Antibiotics, for example, have been demonstrated to function well together and can even boost antibiotic activity in difficult-to-treat biofilms. These peptides have a wide variety of strengths in terms of potent antibacterial activity or Class Ib adjuvant capabilities, as well as Class II adjuvant properties. These immunomodulatory peptides influence the host immune system in a number of ways, including lowering inflammation to prevent an infection from eliciting an overly aggressive immunological response that leads to sepsis and stimulating host-cell-based antimicrobial actions such as enhanced phagocytosis (9).

3. Future perspective

In order to combat complex types of infection, such as bacterial biofilms, metabolically

suppressed bacterial subpopulations like stationary phases or persister bacteria, and intracellular infections, antibiotic-adjuvant combinations need also be researched. This would make it possible to use it in actual therapeutic settings. This would additionally require for a detailed examination of the precise mechanism underlying such combination activity and the impact adjuvants have on the pathogenicity and quorum sensing of bacteria. Additional effects such as immunomodulation, induction/inhibition of autophagy, quorum sensing and pathogenicity, influence on intracellular pathogens, etc. would be an interesting area to investigate, especially for tiny molecule membrane targeting adjuvants and other indirect resistance breakers. Further research and development in this field could lead to the development of novel and effective treatments for infectious diseases, benefiting patients in the future.

4. Conclusions

Recent study has shown that adjuvant treatment can be used to combat multiple mechanisms of antibiotic resistance in bacteria. Direct resistance breakers, indirect resistance breakers, and host-modulating agents are among them. Direct resistance breakers, particularly β-lactamase inhibitors, have been the most effective class of adjuvants to date, with several being authorized for clinical use. Concurrently, continued attempts to fill gaps in approved adjuvants are generating results, with new candidates appearing in clinical trials.

References

1. McCloskey B, Dar O, Zumla A, Heymann DL. Emerging infectious diseases and pandemic potential: status quo and reducing risk of global spread. The Lancet infectious diseases. 2014;14(10):1001-10.
2. Top Ten causes of death. 2020. https://www.who.int/news-room/fact-sheets/detail/the-top-10-causes-of-death (accessed July 26, 2023).
3. Global AntimicrobialResistance and Use Surveillance System (GLASS) Report 2022. 2022. https://www.who.int/publications/i/item/9789240062702 (accessed July 26, 2023).
4. Tyers M, Wright GD. Drug combinations: a strategy to extend the life of antibiotics in the 21st century. Nature Reviews Microbiology. 2019;17(3):141-55.
5. Blair JM, Webber MA, Baylay AJ, Ogbolu DO, Piddock LJ. Molecular mechanisms of antibiotic resistance. Nature reviews microbiology. 2015;13(1):42-51.
6. Wright GD. Antibiotic adjuvants: rescuing antibiotics from resistance. Trends in microbiology. 2016;24(11):862-71.
7. Ashwath P, Sannejal AD. The Action of Efflux Pump Genes in Conferring Drug Resistance to Klebsiella Species and Their Inhibition. Journal of Health and Allied Sciences NU. 2021;12(01):24-31.
8. Ahmad A, Hussain S, Mehmood R, Rana A, Mustafa G. Antibiotic Resistance Breakers and Nano-Antibiotics in Mediating Antimicrobial Resistance [Internet]. Antibiotic Resistance - New Insights [Working Title]. IntechOpen; 2023. Available from: http://dx.doi.org/10.5772/intechopen.111761
9. Dhanda G, Acharya Y, Haldar J. Antibiotic Adjuvants: A Versatile Approach to Combat Antibiotic Resistance. ACS omega. 2023;8(12):10757-83.

Rational approaches in combating antibiotic resistance


Kiranmai Mandava[1]* & Anusha Are[2]

1 Department of Pharmaceutical Chemistry, St. Pauls College of Pharmacy, Turkayamjal, Ranga Reddy, Telangana, India-501510.
2 Department of Pharmacy Practice, St. Pauls College of Pharmacy, Turkayamjal, Ranga Reddy, Telangana, India-501510.

drkiranmaim@stpaulscollege.ac.in

Abstract

Antimicrobial resistance, a natural process in which microorganisms evolve in such a way that they resist the action of medications, has unfortunately increased in correlation with the advent of the antibiotic era. Bacterial pathogens have been a significant contributor to disease and mortality throughout human history. To combat this, finding adjuvants that boost the activity of existing AB may lead to an increase in the antibiotic spectrum, broaden its effectiveness against resistant bacteria, and reduce the dose necessary for antibiotics. The current level of investment in their development, particularly in the fields of natural-product-derived and synthetic small molecules, is in stark contrast to the ever-increasing demand for novel antimicrobials to treat life-threatening infections brought on by the global spread of multidrug-resistant bacterial pathogens.

1.Introduction

In every region of the world, antibiotic resistance (ABR) is increasing to dangerously high levels. Our ability to cure widespread infectious diseases is being threatened by the emergence and global dissemination of new mechanisms of resistance (1). As antibiotics (AB) lose their effectiveness, a rising number of infections, including gonorrhea, blood poisoning, pneumonia, and tuberculosis, have now become difficult to cure and occasionally termed non-curable. Individuals can take a few steps to stop the spread of ABR: infections can be prevented by washing your hands often, cooking your food in a clean manner, avoiding close contact with sick patients, engaging in safer sex, and maintaining a current vaccination schedule (2).

2. Origin, spread, and mechanism of resistance

The emergence of particular mechanisms of resistance hampered the therapeutic use of the first potent antimicrobials, the sulfonamides, since their debut in 1937 (3). The same mechanisms that led to sulfonamide resistance in the late 1930s exist even today; around 70 years later. A bacterial penicillinase was discovered by two members of the penicillin discovery team in 1940, several years before penicillin was made available (4).

AB have been used indiscriminately when necessary. Some instances are for example, when the wrong ABs are taken, when the dosage and duration is inappropriate, when patients skip doses because they feel better quickly after stopping them, and when viral illnesses are treated with AB (3). ABR is on the rise as a result of these misuses (5). Details of the chronological evolution of ABR is mentioned in figure 1.

AB are frequently provided to farm animals to promote growth and prevent sickness in non-medical settings. These medications are exposed to numerous animals and consequently, germs, for a longer time and at lower concentrations (6). Resistance evolves as a result of this. Humans acquire these resistant germs by consuming the animals or coming in close contact with them. Hospitals, poultry farms, and other locations, all have antibiotic-resistant germs that are disseminated via the environment (7).

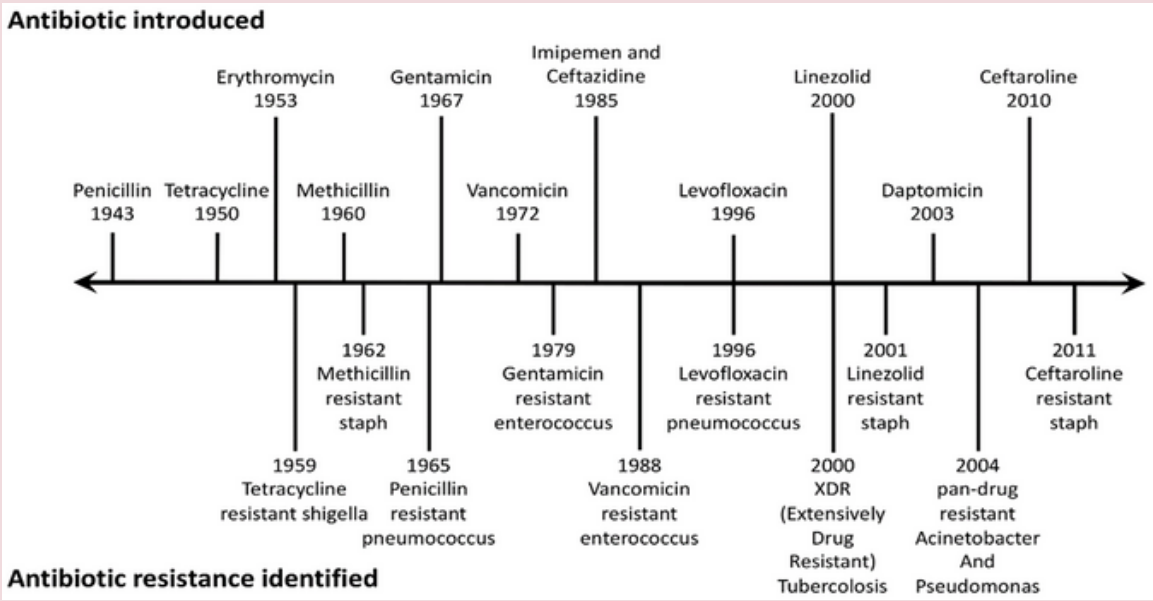


Figure 1. ABR pipeline. On the top, antibiotic introduced and, on the bottom, ABR identified.

Notably, numerous biochemical routes can typically be used to establish resistance to a single antimicrobial class, and a single bacterial cell may be able to utilize a variety of resistance mechanisms to fend off an antibiotic's effects (figure 2) (8). For instance, mutations in the genes encoding the target site of fluroquinolones (FQs; DNA gyrase and topoisomerase IV), overexpression of efflux pumps that extrude the drug from the cell, and protection of the FQ target site by a protein known as Quasi-Newton-Raphson are three different biochemical routes through which resistance to FQs can develop. All three of these biochemical pathways may coexist in the same bacteria at the same time, producing an additive effect and frequently increasing the levels of resistance (9).

3.Techniques/novel methods to combat resistance

A limited availability of a set of ABs in our arsenal has forced us to search for better ways to modify dosing and use a combination of drugs to increase efficacy and decrease resistance. Various techniques that have been employed to combat resistance are sequential regimen, synergistic ABs, optimal dosing regimen, promoting synergy between the immune system and the Abs, and sensitizing the resistant bacteria with antisense technologies (10).

Apart from the above-mentioned techniques, photodynamic therapy in endodontics (11), alternative strategies to develop inhibitors of resistance, development of new ABs (Computer aided drug design/ combinatorial techniques) (12), developing effective vaccines (13), exploring bacteriophage therapy, nano AB (futuristic), single cell pathogen diagnostics (14), and natural compounds quorum sensing are the novel methods used to treat drug resistant pathogens (15).

4. Overcoming ABs challenges in biofilm-associated infections

There is a dire need to adopt alternative treatments for biofilm-associated infections other than ABs. For the biofilm environment, many researchers have proposed a conjugative, mixture of phages, and polymeric nanoparticle-based delivery systems (16). Nanoparticles and nanoantibiotics are the future nanotechnologies that can combat biofilm mediated infections. Nanoparticles of metals like Ag, Au, Zn, Mg, Ca, Cu, CuO_2, Fe_2O_3, TiO_2, Bi, Gd- Fe_2O_3 in combination with ABs have paved a way to address the challenges laid down by multidrug resistance (17).

Natural products/ plant extracts/ secondary metabolites from natural origin like marine alkaloids, and their analogs, polyphenols, and other natural sources including, the plant-based approach – plant-based quorum sensing (QS), can be explored to break the pathways of resistance (18).

5.Modern alternative therapies

5.1. Anti-virulent therapy/ quorum sensing inhibitors (QSI)

Bacteria are the most common pathogen that proliferate within quorum sensing circuits-mediated biofilms. QSI or quorum quenchers can quench the bioactive compounds which can disrupt the biofilms. Acylated homoserine lactones, patulin (isolated from fungus Pe. coprobium), halogenated furonones, cyclic disulphur compounds extracted from garlic, crown vetch, soybean, carrot, water lily, tomato, pea seedling (Pisum sativum), habanerol (Chillies) have been found to be promising QSIs under investigation (19-20).

5.2. Passive immunization/ monoclonal antibodies

Passive immunization through nasal, oral, and topical administration of egg yolk-derived IgY antibodies from immunized chicken have shown to be effective in treating infections caused by resistant pathogens as listed by WHO (21). Anti-glucosaminidase (GMD) monoclonal antibodies can alter the growth habit of S. aureus (Methicillin Resistant Staphylococcus aureus), and suggest that GMD may be a target for direct growth inhibition and focusing on having an effect on the immune system This study shall be a ray of hope to treat osteomyelitis by crossing the barriers of resistance (22). However, using a cocktail of monoclonal antibodies can be the most promising approach to treat resistance.

5.3. Antimicrobial peptides

The combined application of antimicrobial peptides with ABs or other antimicrobials is another proven but yet to be investigated strategy for the development of antimicrobial peptide-based therapeutics against ABR (23).

5.4. Chemoprophylaxis

Vaccines contain a variety of benefits that make them significant among the available approaches to treat ABR. Few aspects of vaccines like vaccines being less likely to develop

resistance; vaccines and antimicrobial agents working in tandem for postponing the tolerance to a medication; and long -term effects produced with a few doses of vaccines alone, have placed vaccines at the forefront (24).

5.5. Phage therapy

A strategy that harnesses the potency of bacteriophages to drive strong selection on their bacterial host is the phage therapy. It mostly relies on target bacteria resisting the phage. This therapy not only kills bacterial cells but also “steers” survivors towards resistance (25).

5.6. Phytochemicals

Delivery of antimicrobial phytoactives using potential nanocarriers like liposomes, nanoemulsions, Solid lipid nanoparticles (SLNs) and nanostructured lipid carriers (NLCs) , and micelles can be strategies developed to face the ABR. Apigenin, capsaicin, carvacrol, thymol, p-cymene, c-terpinene, citral, curcumin, ellagic acid, menthol, oregano, lemon grass, hydroquinone, eugenol, α-tocopherol, theaflavin, quercetin, and thymoquinone are the various natural products used as antimicrobial phytoactives. Few essential oils are also employed for this purpose (26-27).

5.7. Nanoparticles (NPs)

NPs/ nano scale materials/ nanocomposites can be tailored and combined with a variety of antimicrobial agents to overcome antibiotic resistance. It is essential to determine the mechanisms by which these NPs or their complexes inhibit or kill bacteria. Moreover, synergistic effects of NPs with ABs can be a promising regimen to combat bacterial resistance (28).

6. Conclusion

In conclusion, this article helps in providing an overview and the futuristic antimicrobial therapies to combat this resistance.

References

1. Julian DDD. Origins and Evolution of antibiotic resistance. Microbiology and molecular biology reviews. 2010;74(3):417-433.
2. Giannamaria A. Strategies to overcome antimicrobial resistance (AMR) making use of non-essential target inhibitors: A review. International journal of molecular sciences. 2019;20(23):5844.
3. Lauren AR. Understanding and overcoming antibiotic resistance. PLOS Biology. 2017;2:1-5.
4. Jose MM, Cesar AA. Mechanisms of antibiotic resistance. Microbiology and Spectrum. 2016; 4(2):1-37.
5. Roberta JM and Christian M. The Challenge of Overcoming antibiotic resistance: an adjuvant approach? ACS Infectious Diseases. 2017;3:559-563.
6. Tomihiko Y, Katsumi S, Toshiro S, et al. Correlation of over expression of efflux pump genes with antibiotic resistance in Escherichia coli strains clinically isolated from urinary tract infection patients. Journal of clinical microbiology. 2011;49(1):189-194.
7. Christina MJE, Vandenbroucke G, Jan AJWK. Antimicrobial resistance tracing the origins of antibiotic resistance. Nature Medicine. 2022;28:638-640.
8. Rustam IA. A brief history of the antibiotic era: lessons learned and challenges for the future. 2010;1:134.
9. Gianamaria A. Strategies to overcome antimicrobial resistance (amr) making use of non-essential target inhibitors: a review. International journal of molecular sciences. 2019;20 (23):5844.
10. MacLean RC, Millan AS. The evolution of antibiotic resistance. Science. 2019;365(6458):1082-1083.
11. Haitham AE, Isabel PA, Joseph AD. Photodynamic therapy in endodonticS: a helpful tool to combat antibiotic resistance, a literature review. Antibiotics. 2021;10(9):1106.
12. Hughes D. Exploiting genomics, genetics and chemistry to combat antibiotic resistance. Nature reviews genetics. 2003;4:432-441.
13. Naveed M, Waseem M, Aziz T et al., Identification of bacterial strains and development of an mRNA-based vaccine to combat antibiotic resistance in Staphylococcus aureus via in vitro and in-silico approaches. Biomedicines. 2023;11(4):1039.
14. Kuangwen HL, Wong PK, Mach KE, et al. Single cell pathogen diagnostics for combating antibiotic resistance. Nature Reviews Methods Primers. 2023;3(1):6.
15. Javairia K, Sumbal MT, Iram G, et al. Challenges of antibiotic resistance biofilms and potential combating strategies: a review. Biotechnology. 2021;11(4):169.
16. Gholizadeh P, Kose SDS, Ganbarov K, et al. How CRI SPR-cas system could be used combat anti-microbial resistance. Infectious Drug Resistance. 2020;13:1111-21.
17. Saima MSH, Fakhar-e-Alam M. Nano antibiotics: Future naotechnologies to combat antibiotic resistance. Frontiers in Bioscience, Elite. 2018;10:352-374.
18. 1.Lan Lu, Wei Hu, Zeru Tian, et al. Developing natural products as potential anti-biofilm agents. Chinese Medicine. 2019;14:11.
19. 2.Paluch E, Rewak-Soroczynska J, Jedrusik I. Prevention of biofilm formation by quorum quenching. Applied Microbiology Biotechnology. 2020;104:1871-81.
20. 3.Thomas BR, Michael G. Quorum sensing inhibitors: a bargain of effects. Microbiology. 2006;152:895-904.
21. 4.Sherif AE, Aymn TA, Christopher O, et al. IgY antibodies: The promising potential to overcome antibiotic resistance. Comparative Immunology. 2023;14:1-14.
22. 5.John JV, Dan Li, John D, et al. Anti-glocosaminidase monoclonal antibodies as a passive immunization for methicillin- resistant Staphylococcus aureus orthopaedic infections. Bonekey Osteovision 2011;8:187-194.
23. 6.Maria SZ, Dmitriy SO, Olga YG, et al. Application of antimicrobial peptides of the innate immune systems in combination with commercial antibiotics-A novel way to combat antibiotic resistance. Frontier in cellular infections microbiology. 2019;9:1-23.
24. 7.Saad A. The role of vaccines in combating antimicrobial resistance (AMR) bacteria. Saudi Journal of Biological Science. 2021;28:7505-7510.
25. 8.Games G, Sam PB, Oliver K, et al. Steering phages to combat bacterial pathogens. Trends in Microbiology. 2020;28(2):85-94.
26. 9.Homesh Y, Anand M, Madhulika P, et al. Exploring the potential of phytochemicals and nanomaterial: A boon to antimicrobial treatment. Medicine in Drug Discovery. 2023;17:100151.
27. 10.Thanguiyan S, Issac AS, Vasantha P, et al. Tackling multiple-drug-resistance bacteria with conventional and complex phytochemicals. Frontiers in Cellular Infection Microbiology. 2022;12:883889.
28. 11.NanYL, Wen-Chien K, Po-Ren H. Nanoparticles in the treatment of infections caused by multi-drug-resistant organisms. Frontiers in Pharmacology. 2019;10:1153.

“Kryptonite” strategies to weaken the “Super-Bugs” a menace to the world

Archana Upadhya

Associate Professor of Pharmaceutics, Maharashtra Educational Society’s Humera Khan College of Pharmacy, Oshiwara, Jogeshwari (W), Mumbai-102, Maharashtra, India

arch271@gmail.com

1.Introduction

With the discovery and clinical use of antibiotics, starting with penicillin in the 1940’s, humans have been protected from many bacterial diseases. However, the reckless and rampant use of antibiotics has led to the emergence of antimicrobial resistance (AMR) which has caused treatment failures, increased morbidity, mortality, and skyrocketed treatment costs. The World Health Organization (WHO) has declared AMR as one of the top global health threats in 2019. The Indian National Health Policy of 2017 acknowledged the threat posed by antimicrobial resistance (AMR) by recognizing the need for rapid standardization of guidelines regarding the use of antibiotics, to limit the use of antibiotics as growth promoters in animal livestock and to recommend pharmacovigilance of prescription audits with respect to antibiotic usage in the community and in healthcare facilities.

Hitherto susceptible bacteria can develop resistance to antibiotics- “acquired resistance”, by casual mutation or by procuring external genetic material through horizontal gene exchange from other bacteria via plasmids or transposons (1). There are 5 mechanisms of antibiotic resistance (1); (i) inactivation of the antibiotic by enzymes (ii) antibiotic extrusion from the bacterial cell by efflux pumps (iii) decreased uptake of the antibiotic by changes in bacterial membrane permeability (iv) modification of the antibiotic targeting moiety, and (v) development of alternative metabolic pathways.

Besides bringing about awareness to decelerate the progression of AMR by optimizing the use of antibiotics and maintaining hygienic environments, scientists in India and from around the globe are working on strategies to combat the “super bugs” that have emerged which are non-susceptible or resistant to available generations of antibiotics (Figure 1). The approaches include 1) Development of newer antibiotics 2) Development of superior antibiotic delivery systems 3) Inhibition of biofilm formation 4) Co-administration of antibiotic adjuvants 5) Bacteriophage therapy 6) Maintenance of a microbiome.

2.Strategies to combat the development of antimicrobial resistance (AMR)

2.1.Strategy 1: Development of newer antibiotics

In 2016, the WHO member states requested the organization to create a priority list of antibiotic resistant bacteria to direct research and development of new and effective drugs. The list is tabulated in the article by Tacconelli and co-authors (2). The WHO centre in India,

collaborated with the Department of Biotechnology to develop a list of drug resistant microbial strains of national relevance to propel research in Indian research centres and pharmaceutical companies to come up with newer drugs to combat the problem of AMR in the country (Table 1). Mycobacteria (including *Mycobacterium tuberculosis*) drug resistant pathogens are not included in the list as these strains are well recognized as global and national threat for which new treatments are required urgently.

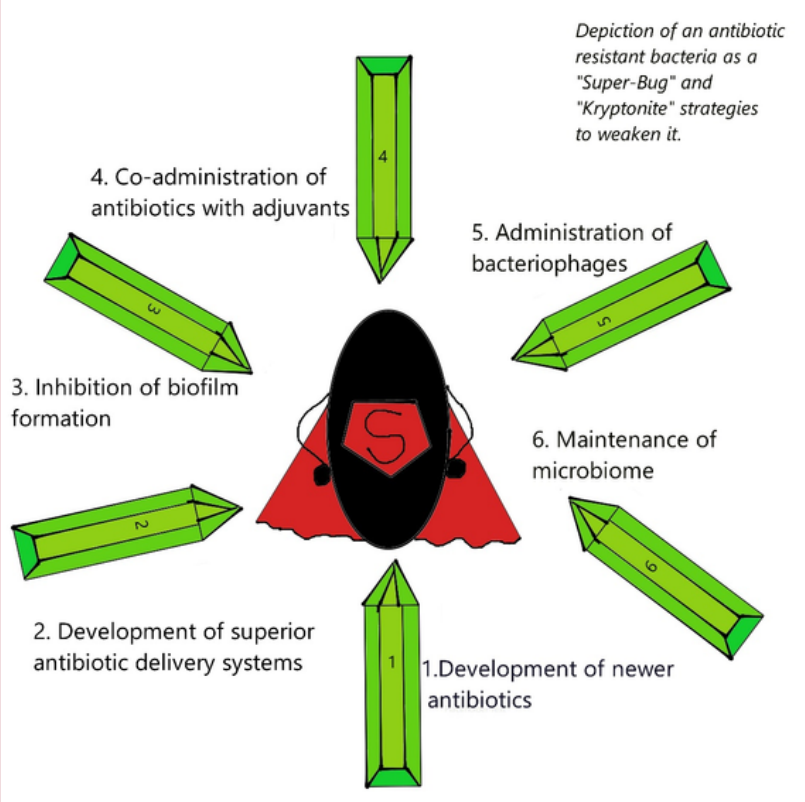


Figure 1: Strategies to combat the development of antimicrobial resistance (AMR)

To tackle the avalanching problem of AMR, new antibiotics or their combinations thereof for treatment of microbial infections have been explored and approved for human use by US FDA (United States Food and Drug Administration) since 2016 (Table 2). Many of these belong to existing classes of antibiotics based on their structural chemistry and mechanism of action. With computer aided design, rational drug design approaches and the advent of artificial intelligence to aid the drug development process, discovery of new antibiotics will be accelerated. We should be able to match the evolution of disease-causing microbial cells with the revolution of newer medicines and mechanisms to stump their growth and pathogenicity.

2.2. Strategy 2: Development of superior antibiotic delivery systems

Nanoparticles (Nps) are of small size (1~100 nm) with high surface area-to-volume ratio which enables them to interact with bacteria and cross both bacterial and mammalian cell membranes. “Nanobiotics” which is used to describe nano-systems loaded with antibiotics may be considered superior antibiotic delivery systems (5). Nanobiotics permit higher drug concentrations at infection sites and boost antibiotic association with the bacteria (6). Nano biotics may evade the antimicrobial resistance mechanisms by facilitating the passage through the microbial membrane by altering the shape, size of membrane, interfere with bacterial biological pathways, interfere with bacterial enzymes, deactivate proteins, cause oxidative

stress within bacterial cell, cause an electrolyte imbalance and interfere with the development of genetic variants (6). Nanobiotics may be created by adsorbing, covalently coupling, encapsulating or entrapping antibiotics in nanocarriers (liposomes, dendrimers, cyclodextrins, polymeric, metallic nanoparticles) to improve their pharmacokinetic, pharmacodynamic properties (7).

Antibiotics conjugated with metallic nanoparticles with inherent anti-bacterial activity like silver and gold nanoparticles show improved potency, for example, penicillin conjugated to silver nanoparticles. When penicillin G and erythromycin were combined with silver nanoparticles, an increased antibiotic activity against E. coli was observed in-vitro with respect to the diameter of inhibition zones (12mm (with antibiotic-silver Nps), as against 8mm with only the antibiotic) (7).

Antibiotic potency of several antibiotics (macrolides, rifamycins, quinolones, beta-lactams, cephalosporins and tetracyclines) has shown a marked improvement when incorporated into nano-systems of beta-cyclodextrins and their derivatives (8). Minimum inhibitory concentration (MIC) of ampicillin, amoxicillin was decreased by 4-fold and cefadroxil MIC by 16-fold against Staphylococcus spp., Klebsiella spp., Escherichia.coli, Pseudomonas aeruginosa, Enterobacter spp., Citrobacter spp., when the antibiotics were formulated with beta-cyclodextrin carriers (9). The beta-cyclodextrin carriers enhanced the stability and solubility of the antibiotics and probably improved antibiotic permeation through bacterial membranes (8). Liposomes are attractive nano delivery systems and can incorporate both hydrophobic and hydrophilic drugs. Moreover, they are mainly composed of relatively biocompatible and biodegradable systems and do not generally pose problems of toxicity or activation of the immune system (5). The US FDA has approved some liposomal antibiotic formulations and some of these are undergoing clinical trials. Arikayce®, Arikace™ and ALIS are amikacin liposomal formulations that have been approved by the FDA against Mycobacterium avium complex (MAC) lung disease, Pseudomonas aeruginosa infection (cystic fibrosis patients) and non-tuberculosis mycobacterial lung infections respectively (5). Lipoquin (NCT00889967) and Pulmaquin (NCT02104245) are ciprofloxacin liposomal formulations currently under clinical trials.

2.3. Strategy 3: Inhibition of biofilm formation

Biofilm formation is one of the main reasons of bacterial resistance. A biofilm is an ecosystem of heterogenous sessile bacteria wrapped in an extracellular polymeric substance (EPS) which supports its existence and flourishing. The sessile bacteria can survive and reproduce in harsh environments with the EPS forming a physicochemical barrier guarding the bacterial colonization against anti-microbial exposure and preventing diffusion of antibiotic to therapeutic concentrations within the film, thus spurring resistance to the antibiotic. Bacteria in biofilms are 1000 times less sensitive to antibiotics than bacteria in a planktonic state (10). Formation of antibiotic tolerant biofilms within the host threaten the efficacy of therapy and cause recurrent chronic infections. New chemotherapeutic agents that can prevent or inhibit biofilm formation, maturation and break up mature biofilms are required, so that the conventional drugs can gain access to bacteria and destroy them. The most common biofilm-forming bacteria include Staphylococcus epidermidis, Pseudomonas aeruginosa,

Staphylococcus aureus, Klebsiella pneumoniae, Enterococcus faecalis, Streptococcus viridans, Escherichia coli, and Proteus mirabilis (11). For inhibiting biofilm formation, the bacterial surface adhesions must be not permitted, the quorum-sensing systems should be disrupted, second nucleotide messenger signalling should be interfered with, biofilm maturation must be inhibited, and mature biofilms should be dispersed (11, 12). A thorough knowledge understanding of biofilm formation and maturation is now leading the way for discovery of molecules that could disrupt the process (12). Preliminary research has led to discovery of compounds such as N-acylhomoserine lactone analogues, patriniae, quercetin, parthenolide, phloretin, hordenine, cinnamaldehyde, ginkgolic acids (GAs) and plant extracts (Hymoenocallis littoralis, Rhodomyrtus tomentosa, Piper betle, Bergenia crassifolia, Zingiber officinale) that have the potential to inhibit biofilm formation (13). Antibiofilm agents are EPS degrading enzymes, anti-microbial peptides, anti-quorum sensing molecules, compounds targeting cellular components and secondary metabolites (14).

Table 1: Indian priority list of antibiotic resistant bacteria (3) (Indian Priority Pathogen List, 2021)

Priority	Bacteria	Antibiotic
Critical	Enterobacteriaceae (*Klebsiella pneumoniae* and *Escherichia coli*)	Carbapenem – R Tigecycline – R Colistin – R
	Non-fermenting bacteria (*Acinetobacter baumannii* and *Pseudomonas aeruginosa*)	Carbapenem – R Colistin – R
High	*Staphylococcus aureus*	MRSA, hVISA Daptomycin – NS Linezolid – R
	Enterococcus species	Vancomycin – R Linezolid – R Daptomycin – NS
	Salmonella species (Typhoidal and Non-typhoidal)	Azithromycin – NS Third generation cephalosporins – NS Carbapenem – NS
Medium	*Streptococcus pneumoniae*	Cephalosporin – R Fluoroquinolones – R Linezolid – R
	Staphylococcus, coagulase-negative	Vancomycin – R Linezolid – R
	Shigella species	Third generation cephalosporins – R Azithromycin – R
	Haemophilus influenzae	Third generation cephalosporin – NS Carbapenem – NS
	Neisseria meningitidis	Fluoroquinolones – NS Third generation cephalosporins – NS
R - Resistant, NS - Nonsusceptible, MRSA - methicillin resistant *Staph. aureus*, hVISA - heterogenous vancomycin intermediate *Staph. Aureus*		

Table 2: New Antibiotics introduced in the market since 2016 (4).

Class of Antibiotics	Basic Chemical Structure	New Antibiotics or their combinations (FDA approved year) since 2016
DNA topoisomerase IV inhibitors (Antibacterial)	Quinolones	Nemonoxacin (2016), Ozenoxacin (2017), Delafloxacin meglumine (2017), Levonadifloxacin arginine salt (2019), Alalevonadifloxacin mesylate (2019), Lascufloxacin hydrochloride (2019).
Protein synthesis inhibitors acting on ribosomal subunits (Antibacterial)	Tetracyclines	Sarecycline hydrochloride (2019), Omadacycline (2018), Eravacylcine (2018)
	Aminoglycosides	Plazomicin (2018)
	Oxazolidinones	Contezolid (2021)
	Pleuromutilins	Lefamulin (2019)
Interference with bacterial cell wall synthesis	Cephalosporins	Cifiderocol (2019).
	Carbapenems	Meropenem/vaborbactam (2017), Imipenem/cilastatin/relebactam (2019)
Interference with fungal cell membrane synthesis	Triterpenoids	Ibrexafungerp (2021)
Anti-tuberculosis Drugs-Cell synthesis Inhibitor	Nitroimidazole	Pretomanid (2019)

2.4. Strategy 4: Co-administration of adjuvants

Antibiotic adjuvants are molecules which possess weak or absent antimicrobial activity. These molecules can enhance the activity of antibiotics and minimize or block resistance development in the bacteria. These can also block intrinsic resistance expanding the activity of the combination (antibiotic plus adjuvant) to a wider range of micro-organisms. Antibiotic adjuvants fall under three categories: beta -lactamase inhibitors, efflux pump inhibitors and outer membrane permeabilizers. Beta-lactamase inhibitors include clavulanic acid, sulfobactam, tazobactam, diazabicyclooctane (DBO) among others. Efflux pump inhibitors comprise of phenotiazine, arylpiperazine, quinoline, thioridazine derivatives. Polimyxin B, colistin, aminoglycosides, polycationic/cationic antimicrobial peptides, glycine basic peptide (GBP), caragenins, menadione are examples of membrane permeabilizer antibiotic adjuvants (1).

2.5. Strategy 5: Bacteriophage therapy

Phage therapy advocates the use of bacteriophage to treat bacterial infection (15). Antibiotics are chemical compounds while bacteriophages are complex biological entities that are hosted in bacteria. In view of an upsurge in antimicrobial resistant infections, bacteriophages are being looked upon as suitable alternatives to treat antibiotic-unresponsive disease. Phages are categorized as lytic (causing destruction of bacterial cells) or lysogenic (inserting their genetic material into the bacterial genome) depending on their development cycle. Lytic phages are preferred in a therapeutic context against bacterial infections (16).

Phages are specific in their invasion of bacteria (strain specific), most phages will infect only those bacteria which carry the complementary receptor, while a few phage members can infect a plethora of bacteria (17). The exclusivity of phages for certain strains of bacteria before they

are used therapeutically. Phage libraries are screened for the suitable virus against the given bacterial strain in-vitro before therapy is initiated.

Phage therapy may be used alone or in combination with probiotics, antibiotics or synbiotics. Phage-antibiotic synergy (PAS) refers to an interaction between the two components which potentiates the activity of both and is more efficacious. The phenomenon of PAS may be attributed to; cell elongation or filamentation by antibiotics, increased plaque size mediated by antibiotics, decreased development of phage/antibiotic resistant mutant, increased antibiotic susceptibility, lowered MIC of antibiotics, depolymerization of bacterial polysaccharides to increase antibiotic diffusion and penetration (18).

There are two situations in which phage therapy can be employed, first is compassionate use when the bacterial infection is life-threatening and cannot be controlled by approved medication and methods. The second situation is in a clinical trial setting to determine the safety, efficacy, dosage, and other clinical parameters of the treatment. While the use of bacteriophages in cases of compassionate use have caused relief and the patient has been cured of the infection (19), clinical trials conducted have not yet been able to prove significant advantages of the phage therapy. Currently few clinical trials have been conducted for phage therapy; PhagoBurn (NCT02116010)-used a cocktail of phages against Escherichia coli and Pseudomonas aeruginosa and the drug silver sulfadiazine and oral phage therapy against diarrhoea (a T4 cocktail phage employed) (NCT00937274). Other clinical trials on therapeutic bacteriophages against Staphylococcus aureus, conducted or underway are NCT00663091, NCT04787250, NCT0432345, NCT02664740 (20).

2.6. Strategy 6: Maintenance of microbiome

Microbiome or microbiota refers to the microbial communities of the gastrointestinal (GI) tract, “gut microbiota” and of the mouth “oral microbiota”. The GI tract contains a vast majority of bacteria, archae, viruses, phages, yeast and fungi (21). Gut microbiota is essential for the proper development of the intestinal tract and maturation of the immune and nervous system (22). The normal gut microbiota does not permit the colonization and proliferation of pathogenic bacteria, but if there is an irrational use of antibiotics, gut microbiota loses its diversity and the patient is susceptible to infections of GIT, diarrhoea and colitis (23). The microbiota exerts anti-microbial effects on pathogenic bacteria by competing for nutritional/metabolic resources, producing a wide range of inhibitory compounds (anti-microbial peptides and bile acids), and cause lysis of specific invading bacteria via phages and viruses that are part of the gut microbiome. Further, the microbiota maintains a mucosal barrier, primes the innate immune defence system and, produces cytokines that perturb the colonization of the invader (24).

Modulation of gut microbiota can be achieved through diet, probiotics (naturally occurring live micro-organisms eg. Yakult, SYMPROVE™), prebiotics (substrates that are used by host micro-organisms), synbiotics (a combination of prebiotic and probiotic). Gut microbiota may also be replaced or replenished by a procedure called faecal microbiota transplantation (FMT).

In this procedure, stool from screened healthy donors is administered to recipients for improving their health. FMT is recommended for patients with recurrent Clostridium difficile infections. FMT re-establishes the health microbiota and hinders the proliferation of Clostridium difficile. There are several clinical trials that are investigating FMT for eradication of GIT of other resistant organisms (NCT05632315, NCT03802461, NCT04188743, NCT04181112, NCT04746222, NCT04759001, NCT04431934, NCT04760665, NCT04146337, EUCTR2019- 001618-41, NCT03063437, NCT03061097, EUCTR2019-004402-10-FR) (24).

3. Conclusion

Although there are several counter-offensive measures to combat AMR organisms, and an ever-growing need for antibiotics, health care practitioners and patients should be disciplined when prescribing or administering the medicine respectively. If bacteria are exposed to antibiotics below the minimum bactericidal concentration, they have the propensity to mutate and turn resistant. Resistance could be acquired from plasmids from other neighbouring micro-organisms or by mutation within the bacterial chromosome itself. Medical practitioners should not prescribe the newest generation of antibiotics, when the infection can be curtailed by an older antibiotic and the patients should complete the entire course of the antibiotic and not stop taking the treatment as the symptoms of infection subside.

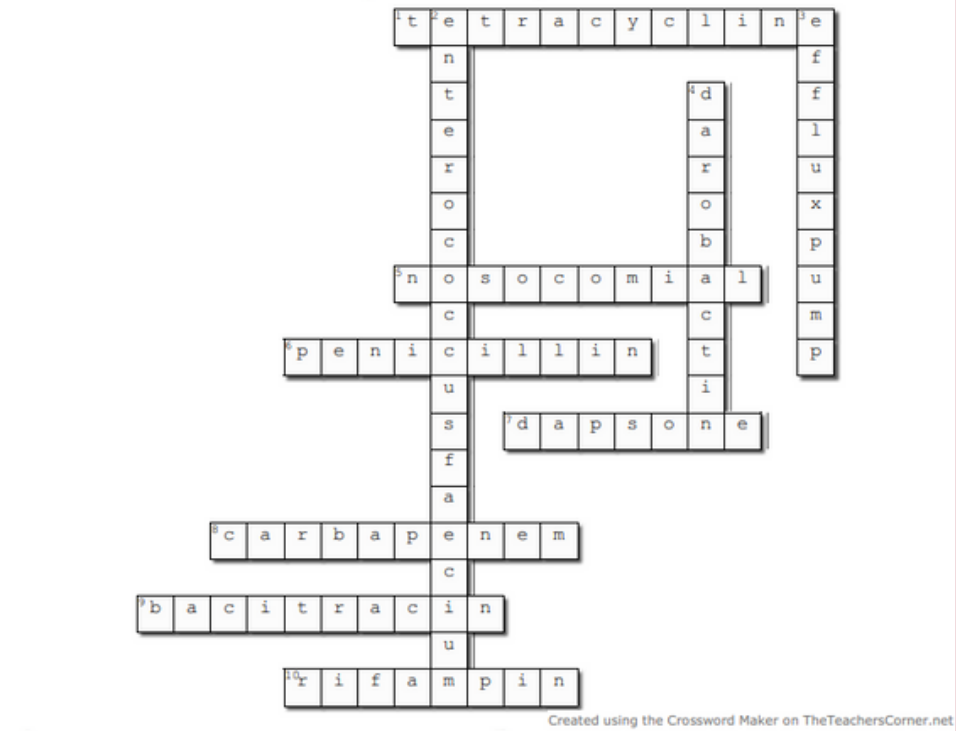
References

1. Annunziato G. Strategies to Overcome Antimicrobial Resistance (AMR) Making Use of Non-Essential Target Inhibitors: A Review. Int J Mol Sci. 2019;20(23).
2. Tacconelli E, Carrara E, Savoldi A, Harbarth S, Mendelson M, Monnet DL, et al. Discovery, research, and development of new antibiotics: the WHO priority list of antibiotic-resistant bacteria and tuberculosis. Lancet Infect Dis. 2018;18(3):318-27.
3. Indian Priority Pathogen List 2021 [Available from: https://dbtindia.gov.in/sites/default/files/IPPL_final.pdf.
4. Shi Z, Zhang J, Tian L, Xin L, Liang C, Ren X, et al. A Comprehensive Overview of the Antibiotics Approved in the Last Two Decades: Retrospects and Prospects. Molecules. 2023;28(4).
5. Ferreira M, Ogren M, Dias JNR, Silva M, Gil S, Tavares L, et al. Liposomes as Antibiotic Delivery Systems: A Promising Nanotechnological Strategy against Antimicrobial Resistance. Molecules. 2021;26(7).
6. Imran M, Jha SK, Hasan N, Insaf A, Shrestha J, Shrestha J, et al. Overcoming Multidrug Resistance of Antibiotics via Nanodelivery Systems. Pharmaceutics. 2022;14(3).
7. Vassallo A, Silletti MF, Faraone I, Milella L. Nanoparticulate Antibiotic Systems as Antibacterial Agents and Antibiotic Delivery Platforms to Fight Infections. Journal of Nanomaterials. 2020;2020:1-31.
8. Yang X, Ye W, Qi Y, Ying Y, Xia Z. Overcoming Multidrug Resistance in Bacteria Through Antibiotics Delivery in Surface-Engineered Nano-Cargos: Recent Developments for Future Nano-Antibiotics. Front Bioeng Biotechnol. 2021;9:696514.
9. Athanassiou G, Michaleas S, Lada-Chitiroglou E, Tsitsa T, Antoniadou-Vyza E. Antimicrobial activity of beta-lactam antibiotics against clinical pathogens after molecular inclusion in several cyclodextrins. A novel approach to bacterial resistance. J Pharm Pharmacol. 2003;55(3):291-300.
10. Bi Y, Xia G, Shi C, Wan J, Liu L, Chen Y, et al. Therapeutic strategies against bacterial biofilms. Fundamental Research. 2021;1(2):193-212.
11. Ghosh A, Jayaraman N, Chatterji D. Small-Molecule Inhibition of Bacterial Biofilm. ACS Omega. 2020;5(7):3108-15.
12. Ma R, Hu X, Zhang X, Wang W, Sun J, Su Z, et al. Strategies to prevent, curb and eliminate biofilm formation based on the characteristics of various periods in one biofilm life cycle. Front Cell Infect Microbiol. 2022;12:1003033.
13. Shahid A, Rasool M, Akhter N, Aslam B, Hassan A, Sana S, et al. Innovative Strategies for the Control of Biofilm Formation in Clinical Settings. Bacterial Biofilms2020.
14. Shrestha L, Fan HM, Tao HR, Huang JD. Recent Strategies to Combat Biofilms Using Antimicrobial Agents and Therapeutic Approaches. Pathogens. 2022;11(3).
15. D'Herelle F. Bacteriophage as a treatment in acute medical and surgical infections. Bulletin of the New York Academy of Medicine. 1931;II(5):329-48.
16. Brives C, Pourraz J. Phage therapy as a potential solution in the fight against AMR: obstacles and possible futures. Palgrave Communications. 2020;6(1).
17. Lin DM, Koskella B, Lin HC. Phage therapy: An alternative to antibiotics in the age of multi-drug resistance. World J Gastrointest Pharmacol Ther. 2017;8(3):162-73.
18. Lusiak-Szelachowska M, Miedzybrodzki R, Drulis-Kawa Z, Cater K, Knezevic P, Winogradow C, et al. Bacteriophages and antibiotic interactions in clinical practice: what we have learned so far. J Biomed Sci. 2022;29(1):23.
19. Hatfull GF, Dedrick RM, Schooley RT. Phage Therapy for Antibiotic-Resistant Bacterial Infections. Annu Rev Med. 2022;73:197-211.

20. Plumet L, Ahmad-Mansour N, Dunyach-Remy C, Kissa K, Sotto A, Lavigne JP, et al. Bacteriophage Therapy for Staphylococcus Aureus Infections: A Review of Animal Models, Treatments, and Clinical Trials. Front Cell Infect Microbiol. 2022;12:907314.
21. Cani PD. Human gut microbiome: hopes, threats and promises. Gut. 2018;67(9):1716-25.
22. Pilmis B, Le Monnier A, Zahar JR. Gut Microbiota, Antibiotic Therapy and Antimicrobial Resistance: A Narrative Review. Microorganisms. 2020;8(2).
23. Buffie CG, Jarchum I, Equinda M, Lipuma L, Gobourne A, Viale A, et al. Profound alterations of intestinal microbiota following a single dose of clindamycin results in sustained susceptibility to Clostridium difficile-induced colitis. Infect Immun. 2012;80(1):62-73.
24. Merrick B, Sergaki C, Edwards L, Moyes DL, Kertanegara M, Prossomariti D, et al. Modulation of the Gut Microbiota to Control Antimicrobial Resistance (AMR)-A Narrative Review with a Focus on Faecal Microbiota Transplantation (FMT). Infect Dis Rep. 2023;15(3):238-54.

Fun & frolic - Crossword

Solution from page 9



Across

1. First drug against which resistance was identified (**tetracycline**)
5. Hospital-Acquired Infections (**nosocomial**)
6. Drug cleaved by beta lactamase (**penicillin**)
7. Antimetabolite (**dapsone**)
8. .....Resistant K. pneumoniae is the pathogen on the CDC threat list (**carbapenem**)
9. Antibiotic against cell wall synthesis (**bacitracin**)
10. Drug affected by mutation in RNA polymerase (**rifampin**)

Down

2. E of ESKAPE (**enterococcusfaecium**)
3. Responsible for antimicrobial resistance (**effluxpump**)
4. Recent bacterial secondary metabolite drug against gram negative superbugs (**darobactin**)

Emerging avenues of flavonoids in defying antibiotic resistance


Dhruv Sanjay Gupta & Divya Suares*

Shobhaben Pratapbhai Patel School of Pharmacy & Technology Management, SVKM's NMIMS, V.L. Mehta Road, Vile Parle (W), Mumbai - 400056, India

divyasuares@gmail.com; divya.suares@nmims.edu

1.Introduction

Antibiotic resistance is a critical issue in the clinical use of antibiotics for the management of bacterial infections, as well as more serious conditions such as cancer (1). The growing development and spread of multidrug resistant infections has accelerated the demand for novel antibiotics, directing research focus towards natural compounds, such as flavonoids (2).
Flavonoids are bioactive with tremendous health potentials and are widely present in a variety of food materials, such as fruits and vegetables. Some of the most studied benefits include antioxidant and anti-inflammatory activities, and their impact on biological functioning has been validated through a variety of preclinical and clinical studies (3). The topic at hand deals with the possibilities of harnessing flavonoids in the management of antimicrobial resistance, and the authors have aimed to provide a structured understanding of the phenomenon and prospective intervention strategies.

At the outset, the authors have discussed the various types of antibiotic resistance with an insight into the mechanisms that may be responsible for their occurrence. Additionally, the prime role played by efflux pump and transport proteins during the onset of resistance have also been touched upon (4). Moreover, the potential adjuvant treatments for bacterial infections using actives that exhibit inhibitory activity towards these pathways, i.e., the therapeutic role of flavonoids will be delved (5). Nevertheless, the claims discussed have been suitably backed by recent preclinical and clinical studies, to further support the use of flavonoids in reducing dependence on synthetic compounds and minimizing the growing burden on healthcare systems, and in turn, improving the quality of life of patients.
Finally, in order to present the most recent developments on this front, the authors offer a discussion on nanotechnological interventions, aimed at improving targeted delivery and offering other allied benefits, such as improved bioavailability and lowering of dose (6). The authors have concluded the article with a short discussion on the key challenges and future perspectives that will prompt continued research efforts and further ones understanding on the plethora of benefits offered by flavonoids.

2.Brief overview of types of antibiotic resistance and resistance mechanisms

Antibiotic resistance exhibited by bacteria may be broadly classified into three sub-types, i.e. intrinsic, acquired, and adaptive (7). Intrinsic resistance primarily entails the resistance that is seen to develop owing to the inherent properties, such as glycopeptide resistance that is widely seen in case of gram-negative pathogens (8). Acquired resistance, on the other hand, develops due to sensitisation over time. This phenomenon is seen to arise due to bacterial gene mutations, or horizontal gene transfer (9). The major mechanisms involved in bacterial gene transfer include chromosomal incorporation due to transformation, genetic transduction and conjugation, mediated by physical contact (10).

Adaptive resistance occurs due to exposure to environmental factors, such as ionic or nutrient concentrations, surrounding pH and the presence of stressors. This type of resistance is a transient, largely reversible process, mediated by epigenetic modulations (11). Some mechanisms that have been attributed to adaptive resistance include DNA methylation, modified expression of porin channels and efflux pumps (12).

In order to obtain a holistic understanding of antimicrobial resistance, other key mechanisms that are explored include, (a) the inactivation or modification of chemical structure of antibiotics, for e.g., acetylation of gentamicin by Pseudomonas aeruginosa (13). (b) The alteration of target site of the antibiotic, which is widely observed in case of beta-lactam antibiotics, such as vancomycin. (c) Staphylococcus aureus resistance due to transpeptidase mutations, which is a cause for clinical concern (14). Such mutations prevent binding of the antibiotic moiety to the target site, permitting retention of the integrity of cellular structure. Efflux pumps also play a crucial role in the mechanism of antimicrobial resistance. They mediate the elimination of antibiotics from bacterial cells, for e.g., AcrAD efflux pump is responsible for aminoglycoside resistance in Escherichia coli (15).

3.Therapeutic significance of flavonoids

An extensive literature analysis undertaken by Biharee et al. sheds light on various mechanisms of flavonoids that may explain their benefits in the management of antimicrobial resistance. Some of these include the prevention of development of cell envelops, regulating the activity of efflux pumps, preventing biofilm formation, triggering membrane rupture, as well as by slowing down bacterial motility (16).

Addressing resistance to antibiotics in cancer management is another emerging avenue. Flavonoids offer a cost-effective, non-toxic alternative, and have been observed to elicit positive health outcomes through a variety of pathways. With a special focus on neoplasms, flavonoids, chiefly flavonolignans, have been seen to inhibit Adenosine triphosphate Binding Cassette (ABC)-transporters. Flavonoids are also responsible for modulating the action of other transporters involved in the pathogenesis, including glucose transporters, small multidrug resistance (SMR) and toxic compound extrusion (MATE) transporters (17).
In addition to this, biotransformation reactions including hydroxylation and conjugation have been observed to improve the biological activity of these agents. Flavonoids such as quercetin and naringenin have been observed to inhibit cell wall synthesis, control DNA replication and

enable the targeting of multiple sites in bacteria. This, in turn, improves the sensitivity of cells to these compounds, minimising the possibility of resistance (18). Isoflavones (such as equol, daidzein), flavonones (naringenin, hesperitin) and flavonols (quercetin, galangin) include someof the most widely studied classes of compounds in combating opportunistic fungal resistance, majorly those belonging to the Candida species (19). The usage of these bioactives, as monotherapy or in combination with widely used antibiotics, may be explored as an effective strategy in the management of antimicrobial resistance. Their usage has been linked with a reduction in the ability of microorganisms to adapt to treatment regimens, opening up the doors for exciting research opportunities (20). Figure 1 depicts the skeleton structure of flavonoids, alongside the representative structure from certain classes discussed, and their members.

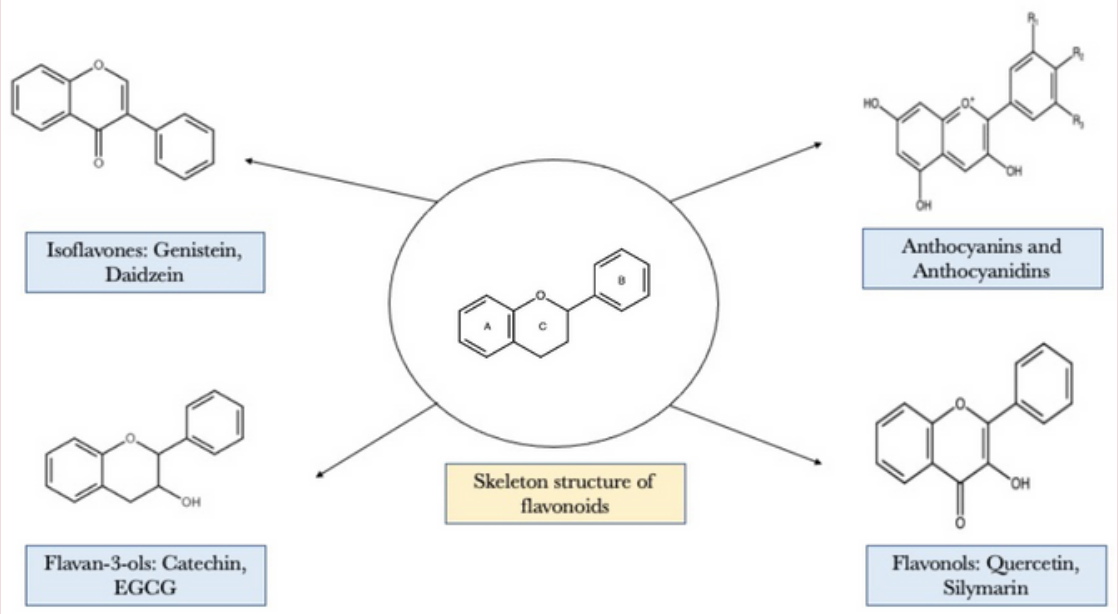


Figure 1. Skeleton structure of flavonoids, alongside the representative structure from certain class and their members

4. Supporting evidence from preclinical studies

This section aims to offer an overview of recent preclinical studies that elucidate the role of flavonoids as antimicrobials. Some of the key mechanisms explored in preclinical trials include the inhibition of cell envelop formation, arrested synthesis of key biomolecules such as nucleic acids, and lowered bacterial motility.

A study by Zhang et al. highlighted the potentials of flavonoids such as quercetin and apigenin in inhibiting cell envelop synthesis, by inhibiting the dehydrase enzyme that catalyses the process (21). With respect to bacterial motility, flavonoids have been observed to arrest swarming movement, thereby reducing pathogen adhesion to target cells. Naringenins and catechins have been widely explored in this domain, and have been linked with a suppression of biofilm formation, resulting from lowering of quorum sensing among bacterial populations (22). Bacterial endotoxins and exotoxins are of particular interest with respect to disease onset and progression, and have been explored as a potential target. A study by Silva et al. reported the usage of fisetin, genistein and luteolin, among other compounds, against the haemolysins produced by S. aureus (23).

A study by Cuccioloni et al. aimed to understand the benefits of epigallocatechin gallate (EGCG) in the inhibition of bacterial folic acid that revealed key enzyme inhibition across different species, including Streptomonas, Mycobacterium and Escherichia sp. (24). These studies highlight the potentials for further exploration, and clinical validation of preliminary outcomes. Figure 2 offers a diagrammatic representation of the major mechanisms of flavonoids in the management of antibiotic resistance, based on insights from preclinical studies.

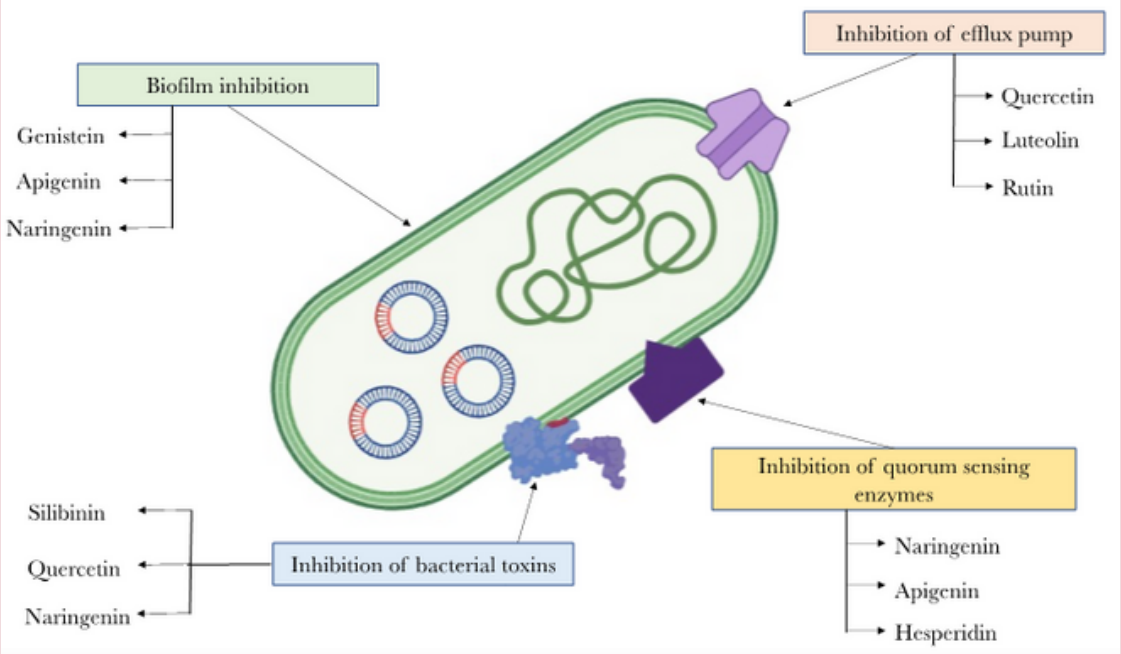


Figure 2. Mechanisms of flavonoids in the management of antibiotic resistance, gleaned from preclinical studies (16) (Created using Biorender)

5. Clinical picture

While results from preclinical studies are a crucial stepping-stone in evaluating the benefits of flavonoids in disease management, it is pertinent to include relevant data from clinical trials. Table 1 offers an insight into the outcomes and inferences from recently conducted clinical trials, to further support the usage of phytoconstituents such as flavonoids in the management of the condition at hand. While there have been a number of studies over the last decade, there is a need for a greater number of focused trials, exploring combinations of different agents to establish their pharmacodynamic profiles, safety as well as possibilities for exploring newer targets.

6.Nanotechnological interventions

While natural actives are widely distributed across various dietary food materials and provide a wide range of health benefits, their usage is marred by a number of limitations, including degradation owing to poor stability, and low solubility. Nanotechnological strategies aim to pave the way for improved delivery, as well as lowered side effects. This section offers an exploration of recent advances in the domain, focused on the delivery of nanocarrier loaded with flavonoids for their antimicrobial benefits.

A study by Das et al. explored the application of flavonoid-loaded gold nanoparticles for controlling the growth of gram-negative bacteria. The antioxidant benefits of quercetin were harnessed to hamper microbial growth, and a synergistic effect with gold assisted cell

Table 1. Insight into recently conducted clinical trials to establish the clinical efficacy of flavonoids in antimicrobial resistance

Dose and duration	Volunteers	Design	Outcome	Conclusion	Ref
Cranberry proanthocyanidins (Doses 1-18.5 mg, twice a day), for 24 weeks	145 healthy women with history of Urinary tract infections (UTIs)	Randomized, controlled clinical trial	Decrease in the incidence of infections, no major adverse effects reported.	Proanthyocyanidin administration was associated with protective effects, and reduced rate of recurrence. No significant benefits observed with higher dosing.	(25)
Naringenin (5mg/ kg) v/s Azithromycin, over a 5 day period	180 children with bronchial pneumonia	Randomized, controlled clinical trial	Decreased latency period, lowered inflammation, satisfactory safety profile (compared to azithromycin) and overall health improvement.	Naringenin may be clinically employed in pneumonia management, owing to its anti-inflammatory and antimicrobial benefits.	(26)
Cranberry juice (rich in proanthocyanidins), at concentrations 17 and 35%, for a period of 8 weeks	522 *Helicobacter pylori* positive adults	Double-blind, randomized, placebo-controlled trial	Satisfactory patient compliance, no adverse effects observed, lowered positivity rates over an 8-week period.	Intake of foods rich in flavonoids, such as cranberries, may be beneficial in suppressing microbial growth and proliferation.	(27)
Silymarin (140mg/ day), in combination with oral doxycycline (100mg/ day), over 4 months	60 patients with acne vulgaris	Randomised, controlled clinical trial	Therapeutic synergism, lowered occurrence of lesions on combining a flavonoid moiety with synthetic therapy.	While work remains to be done in evaluating the benefits of silymarin monotherapy, it may be used as adjunct therapy with conventional treatment regimens.	(28)
Plantago lanceolata herbal tea (prepared from its flowers and leaves), infusion	44 healthy adolescents	Randomised, controlled clinical trial	Inhibition of growth of cariogenic bacteria (*Streptococci*, *Lactobacilli*),	Phytochemicals such as flavonoids, coumarins and tannins may be used in checking the growth of cariogenic	(29)

membrane disruption (30). Li et al. evaluated the usage of nanoparticles and nanofibers loaded with actives from Chinese traditional medicine, in order to elicit an antibacterial effect. It was observed that the combination of flavonoid glycosides with nanoparticles enhanced bacteriostatic activity, offering interesting opportunities for drug delivery and combating antimicrobial resistance (31).

Nanotechnological interventions have also been employed to improve the pharmacokinetic profiles of natural actives, thereby improving their bioavailability (32). By the process of valorization, products that would have otherwise been treated as waste are now finding biomedical applications. Pomegranate peel, known to be rich in anthocyanins among other polyphenols, offers a host of antimicrobial benefits. Various strategies such as nanoemulsions and lipid nanoparticles have been utilised to deliver pomegranate peel extract to target sites, offering advantages such as easy production, low cost and promising biocompatibility (33). As interest in this domain continues to grow, other biological compounds such as peptides have also been explored for their benefits as natural nano-antibiotics (34).

7.Conclusion

Antimicrobial resistance is a growing issue in the medical space, affecting millions across the globe. This has sparked the need for exploring alternative strategies for its management. Extensive research and development in the domain has permitted the discovery of potential targets and mechanisms, and nanotechnology offers interesting opportunities for targeted delivery, minimising the occurrence of side effects. Notwithstanding the massive number of benefits offered and prospects for future development, a greater number of focused studies are necessary, to further support and scale-up the usage of these agents.

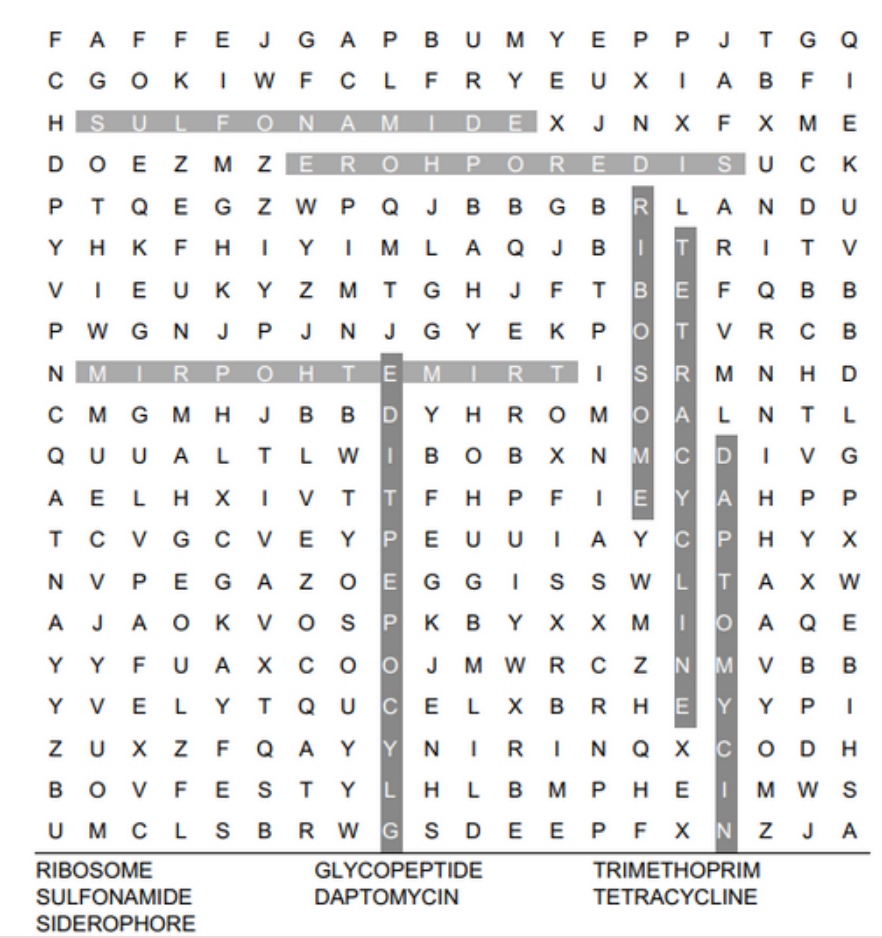
References

1. Huemer M, Mairpady Shambat S, Brugger SD, Zinkernagel AS. Antibiotic resistance and persistence—Implications for human health and treatment perspectives. EMBO Reports. 2020 Dec 3;21(12):e51034.
2. Mączka W, Duda-Madej A, Grabarczyk M, Wińska K. Natural Compounds in the Battle against Microorganisms—Linalool. Molecules. 2022 Oct 15;27(20):6928.
3. Pei R, Liu X, Bolling B. Flavonoids and gut health. Current Opinion in Biotechnology. 2020 Feb;61:153–9.
4. Kumawat M, Nabi B, Daswani M, Viquar I, Pal N, Sharma P, et al. Role of bacterial efflux pump proteins in antibiotic resistance across microbial species. Microbial Pathogenesis. 2023 Aug;181:106182.
5. Waditzer M, Bucar F. Flavonoids as Inhibitors of Bacterial Efflux Pumps. Molecules. 2021 Nov 16;26(22):6904.
6. Ghosh S, Nandi S, Basu T. Nano-Antibacterials Using Medicinal Plant Components: An Overview. Front Microbiol. 2022 Feb 22;12:768739.
7. Lee JH. Perspectives towards antibiotic resistance: from molecules to population. J Microbiol. 2019 Mar;57(3):181–4.
8. Pang Z, Raudonis R, Glick BR, Lin TJ, Cheng Z. Antibiotic resistance in Pseudomonas aeruginosa: mechanisms and alternative therapeutic strategies. Biotechnology Advances. 2019 Jan;37(1):177–92.
9. Partridge SR, Kwong SM, Firth N, Jensen SO. Mobile Genetic Elements Associated with Antimicrobial Resistance. Clin Microbiol Rev. 2018 Oct;31(4):e00088-17.
10. Ward RA, Fawell S, Floc'h N, Flemington V, McKerrecher D, Smith PD. Challenges and Opportunities in Cancer Drug Resistance. Chem Rev. 2021 Mar 24;121(6):3297–351.
11. Ghosh D, Veeraraghavan B, Elangovan R, Vivekanandan P. Antibiotic Resistance and Epigenetics: More to It than Meets the Eye. Antimicrob Agents Chemother. 2020 Jan 27;64(2):e02225-19.
12. Motta SS, Cluzel P, Aldana M. Adaptive Resistance in Bacteria Requires Epigenetic Inheritance, Genetic Noise, and Cost of Efflux Pumps. Ryu CM, editor. PLoS ONE. 2015 Mar 17;10(3):e0118464.
13. Poole K. Aminoglycoside Resistance in Pseudomonas aeruginosa. Antimicrob Agents Chemother. 2005 Feb;49(2):479–87.
14. Marsot A, Boulamery A, Bruguerolle B, Simon N. Vancomycin: A Review of Population Pharmacokinetic Analyses. Clinical Pharmacokinetics. 2012 Jan;51(1):1–13.
15. Nikaido H. Multidrug efflux pumps of gram-negative bacteria. J Bacteriol. 1996 Oct;178(20):5853–9.
16. Biharee A, Sharma A, Kumar A, Jaitak V. Antimicrobial flavonoids as a potential substitute for overcoming antimicrobial resistance. Fitoterapia. 2020 Oct;146:104720.
17. Chambers CS, Viktorová J, Řehořová K, Biedermann D, Turková L, Macek T, et al. Defying Multidrug Resistance! Modulation of Related Transporters by Flavonoids and Flavonolignans. J Agric Food Chem. 2020 Feb 19;68(7):1763–79.
18. Lobiuc A, Pavăl NE, Mangalagiu II, Gheorghiţă R, Teliban GC, Amăriucăi-Mantu D, et al. Future Antimicrobials: Natural and Functionalized Phenolics. Molecules. 2023 Jan 22;28(3):1114.

19. Srivastava D, Yadav A, Naqvi S, Awasthi H, Fatima Z. Efficacy of Flavonoids in Combating Fluconazole Resistant Oral Candidiasis. CPD. 2022 Jun;28(21):1703–13.
20. Al Aboody MS, Mickymaray S. Anti-Fungal Efficacy and Mechanisms of Flavonoids. Antibiotics. 2020 Jan 26;9(2):45.
21. Zhang L, Kong Y, Wu D, Zhang H, Wu J, Chen J, et al. Three flavonoids targeting the β-hydroxyacyl-acyl carrier protein dehydratase from Helicobacter pylori : Crystal structure characterization with enzymatic inhibition assay. Protein Sci. 2008 Nov;17(11):1971–8.
22. Pejin B, Ciric A, Markovic J, Glamoclija J, Nikolic M, Stanimirovic B, et al. Quercetin Potently Reduces Biofilm Formation of the Strain Pseudomonas aeruginosa PAO1 in vitro. CPB. 2015 Jun 6;16(8):733–7.
23. Silva LN, Zimmer KR, Macedo AJ, Trentin DS. Plant Natural Products Targeting Bacterial Virulence Factors. Chem Rev. 2016 Aug 24;116(16):9162–236.
24. Cuccioloni M, Mozzicafreddo M, Spina M, Tran CN, Falconi M, Eleuteri AM, et al. Epigallocatechin-3-gallate potently inhibits the in vitro activity of hydroxy-3-methyl-glutaryl-CoA reductase. Journal of Lipid Research. 2011 May;52(5):897–907.
25. Babar A, Moore L, Leblanc V, Dudonné S, Desjardins Y, Lemieux S, et al. High dose versus low dose standardized cranberry proanthocyanidin extract for the prevention of recurrent urinary tract infection in healthy women: a double-blind randomized controlled trial. BMC Urol. 2021 Dec;21(1):44.
26. Yao W, Zhang X, Xu F, Cao C, Liu T, Xue Y. The therapeutic effects of naringenin on bronchial pneumonia in children. Pharmacol Res Perspect. 2021 Aug;9(4):e00825.
27. Li Z, Ma J, Guo Y, Liu W, Li M, Zhang L, et al. Suppression of Helicobacter pylori infection by daily cranberry intake: A double-blind, randomized, placebo-controlled trial. Journal of Gastroenterology and Hepatology. 2021 Apr;36(4):927–35.
28. Shie Morteza M, Hayati Z, Namazi N, Abdollahimajd F. Efficacy and safety of oral silymarin in comparison with oral doxycycline and their combination therapy in the treatment of acne vulgaris. Dermatologic Therapy [Internet]. 2019 Nov [cited 2023 Jul 27];32(6). Available from: https://onlinelibrary.wiley.com/doi/10.1111/dth.13095
29. Ferrazzano GF, Cantile T, Roberto L, Ingenito A, Catania MR, Roscetto E, et al. Determination of the In Vitro and In Vivo Antimicrobial Activity on Salivary Streptococci and Lactobacilli and Chemical Characterisation of the Phenolic Content of a Plantago lanceolata Infusion. BioMed Research International. 2015;2015:1–8.
30. Das S, Pramanik T, Jethwa M, Roy P. Flavonoid-Decorated Nano-gold for Antimicrobial Therapy Against Gram-negative Bacteria Escherichia coli. Appl Biochem Biotechnol. 2021 Jun;193(6):1727–43.
31. Li T, Wang P, Guo W, Huang X, Tian X, Wu G, et al. Natural Berberine-Based Chinese Herb Medicine Assembled Nanostructures with Modified Antibacterial Application. ACS Nano. 2019 Jun 25;13(6):6770–81.
32. Huang T, Liu Y, Zhang C. Pharmacokinetics and Bioavailability Enhancement of Baicalin: A Review. Eur J Drug Metab Pharmacokinet. 2019 Apr;44(2):159–68.
33. Andishmand H, Azadmard-damirchi S, Hamishekar H, Torbati M, Kharazmi MS, Savage GP, et al. Nano-delivery systems for encapsulation of phenolic compounds from pomegranate peel. Advances in Colloid and Interface Science. 2023 Jan;311:102833.
34. Rahman L, Asif S, Ullah A, Khan WS, Rehman A. Biofunctionalized Nano-antimicrobials - Progress, Prospects and Challenges. CTMC. 2022 May;22(13):1046–67.

Fun & frolic - Wordsearch

Solution from page 27



APTI Forum News

Dr Varsha Pokharkar, professor and Ms Preeti Tamane, assistant professor, Poona College of Pharmacy, Bharati Vidyapeeth (Deemed to be University), Pune, were granted an Indian patent (patent no. 435008) for their platform technology which is a dual drug loaded formulation for oral cancer. The formulation is a nanoemulsion based mucoadhesive gel for site specific delivery of 5-fluorouracil (the first line chemotherapy drug used in oral cancer) and Berberine, a phytoconstituent.

Dr. Asha Thomas, professor and head, department of pharmaceutical chemistry was granted patents for novel indole compounds (patent no. 427211, date of grant: 28/03/2023), and for extraction, preparation, and processing of chicken skin oil by recycling poultry waste and its formulation comprises of biodegradable base (patent no. 437775, date of grant: 07/07/2023).

Dr. Arati Malpani, associate professor, department of pharmacology, HKES's Matoshree Taradevi Rampure Institute of Pharmaceutical Sciences, Kalaburagi has got one patent published for in vivo Ballon method for measuring uterine contractions in Wistar rats (patent application no. 2023410299997A, date of publication: 26/05/2023). Also, she has been appointed as the editorial board member of International Journal of Chinese Medicine.

Women Leadership Conclave

APTI and APTI's Women's Forum organized a first of its kind and exclusive **"Women Leadership Conclave"** from 27 th to 29 th April 2023 at the Kenilworth Resort, Utorda-Goa. The residential conclave was attended by 50 women academic leaders from all over India. Aimed at capitalizing the leadership skills of our women pharmacy teachers, understanding the challenges that women face in leadership and to help women leaders rejuvenate and foster their leadership qualities, the residential conclave was a vision of our luminary APTI leaders, Dr. Milind Umekar, President APTI India and Dr. Vandana Patravale, National Convenor, APTI Women's Forum.

The delegates checked into the beautiful venue at the Kenilworth Resort, Utorda-Goa, on 27 April 2023. The program began on 28 April with the divine inauguration and the blessings of almighty. The icing on the cake for the program was the presence of the distinguished Chief Guest, Dr. Indu Pal Kur, Professor and Chairperson and Ex Dean, University Institute of Pharmaceutical Sciences, Panjab University, who emphasised on being perseverant, having grit and yet striking a balance in the lives of women. This was followed by a report by Dr. Vandana Patravale, National Convenor, APTI Women's forum wherein she showcased all the various activities carried out by APTI Women's forum zone wise. The release of the APTI Women's forum Newsletter on the theme "Bone Targeted Delivery" was released at the hands of the Chief guest Dr. Indu Pal Kur, Dr. Vandana Patravale, Dr. Mangirish Deshpande, APTI, Goa State President, Dr. Shailendra Gurav APTI, Goa State Vice President, the members of the editorial board of the Newsletter Dr. Shubhini Saraf, Dr. Suneela Dhaneshwar and the program coordinators Dr. Rashmi Trivedi and Dr. Pearl Dighe. Dr. Milind Umekar, respected National President of APTI shared his thoughts through a video message to promote more women pharmacy teachers at the upfront in APTI's leadership roles. In her powerful video message, Ms. Maharukh Rustomjee, Founder & Managing Partner, Amaterasu Life Sciences LLP, Mumbai expressed the desire that women should occupy leadership roles in various sectors owing to their remarkable qualities of compassion and perseverance. The inaugural program was coordinated by Dr. Rashmi Trivedi.

The two power packed exhilarating days were filled with sessions from renowned resource persons. Dr. Ashish Johri who holds a PhD in the area of Authentic Leadership Development and has a total of 21 years of experience is a versatile behavioural trainer, capacity development and talent development professional for various types of corporate and academic requirements.

His activity based sessions on the areas of Emotional Resilience and Academic Leadership, Cultivating Coalitions, Managing Negative Beliefs, subconscious bias and stereotypes and developing grit were thought provoking and full of life and energy. The second resource person, Ms. Shabnam Gaitonde is a renowned HR professional with over 25 years of experience across industries and currently serving at Gland Pharma as VP HR. She is known for being an executive coach and her strength lies in building teams and developing leaders. Her intriguing sessions revolved around challenges faced by women in leadership positions, how to be a confident leader and managing what is most important.

The APTI Women's forum hosted a Gala dinner at the Aquatic by the poolside with entertainment program comprising of a one man and a Goan folk dance troupe, which was enjoyed by all. The hospitality of Kenilworth Resort and the opportunity to network with co-academicians, researchers and mentors was savoured by all the delegates.

The residential retreat ended with felicitation of the organisers, sharing of feedback by thedelegates, the distribution of certificates, mementos and personalised gifts to every delegate.

Dr. Pearl Dighe proposed the vote of thanks. The event was sponsored by Indoco remedies Ltd., Sahajanand Medical Technologies, Mumbai, Amaterasu Life Sciences LLP, Aquatic Remedies Ltd. Mumbai, Mayons Pharmaceuticals, Nagpur, IPA Kolhapur Branch, Mumbai.

The entire event was convened by Dr. Vandana Patravale, National Convenor, APTI Women's Forum and coordinated by Dr. Rashmi Trivedi, Member APTI Women's Forum and Dr. Pearl Dighe, APTI Goa State - Vice President.

Pharma News Round-Up

2 May, 2023: In accordance to the guidelines established by the Global Initiative for Asthma (GINA), AstraZeneca India has announced plans to launch five Severe Asthma Centres of Excellences (CoEs) across New Delhi, Ahmedabad, Goa and Cochin. At these CoEs, focus will be on specialty and clinical assessment, diagnosis, patient counselling, biologics initiation, and data management related to severe asthma. Under AstraZeneca's model of Severe Asthma CoEs, disease management is centred on four crucial pillars. The first pillar involves utilising identification of patient profile for severe asthma. The second pillar involves addressing knowledge gaps in patient awareness by providing crucial information throught interactive education forum. The third pillar focuses on curating specialised curriculum for assisting respiratory specialists in tailoring therapy to meet the patient needs. Finally, fourth pillar focuses on establishing an academy for asthma counsellors to revolutionise patient care and asthma control.

8 May, 2023: Common Services Centers (CSCs), under the Ministry of Electronics & IT, and Mumbai-based startup Medikabazaar has entered into Memorandum of Understanding (MoU). This partnership will strengthen rural healthcare systems as well as Tier 3 & 4 cities via simpler and faster access to medical supplies and equipment. Additionally, under this agreement, training will be imparted to VLEs (Village Level Entrepreneurs), who run the CSCs, for developing and establishing online B2B medical supply business.

12 May, 2023: Euvichol-Plus, the world's first andonly oral cholera vaccine (OCV), in a low-density polyethylene (LDPE) unidose pack has been jointly launched by Mumbai-based startup TechInvention Lifecare and Eubiologics Co. in South Korea in India. TechInvention recently received approval by the regulatory body Central Drugs Standard Control Organisation (CDSCO) for the launch of Euvichol-Plus following the completion of a phase III clinical study. The potential advantages of the LDPE unidose pack is ease to use and minimal issues of breakage, storage,transportation,distribution, and waste management.

13 May, 2023: Ministry of Ayush has sponsored first-of-its-kind initiative, AyurTech, the Centre of Excellence (CoE), part of the Centre of Excellence in Artificial Intelligence (AI) based Precision Healthcare at the school of Artificial Intelligence and Data Science (AIDE) at IIT Jodhpur. At this centre, an AI-driven 'Evidence-based Ayurveda', integrative framework for population and individual risk stratification and early actionable precision health interventions will be established by combining Electronics, Digital health and AI and multi-omics technologies for realising solutions in a transdisciplinary framework.

16 May, 2023: Andhra Pradesh government has released Andhra Pradesh Action Plan for Containment of Antimicrobial Resistance (APAPCAR) a proactive decision taken to combat the pertinent challenge of antimicrobial resistance (AMR). The initiative to facilitate implementation of this plan was taken under the able stewardship of M T Krishna Babu, IAS, Special Chief Secretary Health, Medical and Family welfare, Andhra Pradesh Govt, and Commissioner J Nivas, IAS.

22 May, 2023: SKAN Research Trust, Bangalore and UK-based Quadram Institute Bioscience have entered into a Comprehensive Partnership Agreement to collaborate on research programmes focused on gut health to develop microbiome based products . Under this agreement, both organisations will undertake microbiome based research projects to develop Indian specific therapies that will alleviate ageing and neurological diseases. Further to this, the agreement will enable technology transfer between the organisations to augment cutting-edge Indian research capabilities as well as impart training to young Indian research scientists with Quadram Institute Bioscience.

20 Jun, 2023: Under Corporate Social Responsibility (CSR), Lupin Human Welfare and Research Foundation (LHWRF), has signed a Memorandum of Understanding (MoU) with the Government of Maharashtra to implement 5 years program to create awareness about the risks, significance of early screening, diagnosis and of Cardiovascular Diseases (CVD) and Chronic Obstructive Pulmonary Disease (COPD) in the Palghar district of Maharashtra.

21 Jun, 2023: Max Healthcare Institute, and the Indian Institute of Technology Bombay (IIT-B), have entered into a Memorandum of Understanding (MoU) to create a partnership intended to undertake joint research projects focusing on healthcare technologies, medical devices, artificial intelligence, data analytics, and other cutting-edge areas. In addition to this, the collaboration will entail development of joint training programmes and workshops to enhance the skills of healthcare professionals, researchers, and students.

29 Jun, 2023: Procter & Gamble (P&G) India will invest Rs 2000 crore to set up a new state-of-the-art personal healthcare manufacturing facility in Sanand, Gujarat to manufacture products of superior quality in digestive wellness. It will use state-of-the-art technologyviz. the latest vision systems for quality checks, robotic equipment for material movement and operator cockpits to be fully automated based on the modern concept of 'Industry 4.0'.

Source:

1. https://www.biospectrumindia.com
2. www.expresspharma.in

PREPARATION OF THE ARTICLE FOR SUBMISSION TO APTI NEWSLETTER

General points

- Word limit for the article is 1500 – 2000 words
- The font to be used in the article shall be Times New Roman (or Times) with a font size of 12
- Any abbreviations used in the article must be explained in its full form at the first instance of their use
- The title must be concise and self explanatory
- Name of all authors must be mentioned along with their detailed current affiliation with an asterisk mark (*) for the corresponding author
- Email address of the corresponding author should be mentioned for further communication
- A high resolution photograph of the corresponding author should be embedded below the author affiliation, before start of the article main text
- Kindly use a suitable naming convention for the submitted file of your article for better understanding at our end
- The article should be submitted in word format only
- Please do not insert any header or footer to the article apart from page number
- Submit articles to: editor.aptiwomensforum@gmail.com

Figures and Tables

- The figures/illustrations and tables should be embedded next to the relevant text in the article and not at the end and number them according to their sequence of appearance in the text
- The caption of the figures and tables should be placed along with the figures and tables respectively
- Kindly ensure that all figures submitted are of high resolution with a minimum size of 300dpi and in .jpeg or .tiff format

References

- The reference style to be followed is Vancouver style for citation in the text
- Use of a referencing software is encouraged

PUBLISHED BY: Secretary APTI, on behalf of APTI

Created by : Chandramouli R

ASSOCIATION OF PHARMACEUTICAL TEACHERS OF INDIA (APTI)

APTI SECRETARIAT: Krupanidhi College of Pharmacy, # 12/1 Chikka Bellandur, Carmelram Post, Varthur Hobli, Bangalore – 560035, Karnataka, India, Phone :+91 90088 88415

LOTUS LOGO STORY

As a lotus is able to emerge from muddy waters un-spoilt and pure it is considered to represent a wise and spiritually enlightened quality in a person; it is representative of a woman who carries out her tasks with little concern for any reward and with a full liberation from attachment. Lotus-woman in the modern sense of women's qualities: she is superbly intelligent, highly educated, and totally committed to individualism. She is politically astute and works incessantly for a better and more humane society. She is exquisite in her taste for music, art and culture, abounds in social graces and performs brilliantly in communication.