Satish K Awasthi
Email: satishpna@gmail.com
9582087608

Paper 4204 A: Organic Chemistry
Course A: Medicinal Chemistry(Special Paper)

Drug Resistance:

Antimicrobial resistance is the ability of a microorganism to survive and multiply in the presence of an antimicrobial agent that would normally inhibit or kill this particular kind of organism. It's just one of the many adaptive traits that resilient bacterial subpopulations may possess or acquire, enabling them to out-compete and out-survive their microbial neighbors and overcome host strategies aimed against them. This phenomenon is nearly as old as the discovery of antimicrobials themselves, having been described by pioneers like Ehrlich for trypanosomes and Fleming for staphylococci. Most fearsome today is the rate at which antibiotic resistance often develops and hastily it spreads across the globe and among different species of bacteria.

Resistance to single antibiotics became prominent in organisms that encountered the first commercially produced antibiotics. The most notable example is resistance to penicillin among staphylococci, specified by an enzyme (penicillinase) that degraded the antibiotic. Over the years, continued selective pressure by different drugs has resulted in organisms bearing additional kinds of resistance mechanisms that led to multidrug resistance (MDR)— novel penicillin-binding proteins (PBPs), enzymatic mechanisms of drug modification, mutated drug targets, enhanced efflux pump expression, and altered membrane permeability. Some of the most problematic MDR organisms that are encountered currently include *Pseudomonas aeruginosa* (another microbe of soil origin), *Acinetobacter baumannii*, *Escherichia coli* and *Klebsiella pneumoniae* bearing extended-spectrum β-lactamases (ESBL), vancomycin resistant enterococci (VRE), methicillin-resistant Staphylococcus aureus (MRSA), vancomycin-resistant MRSA, and extensively drug-resistant (XDR) *Mycobacterium tuberculosis*. Some like methicillin-resistant *S. aureus* couple MDR with exceptional virulence capabilities. Others, including some strains of *P. aeruginosa*, *A. baumannii*, and *K. pneumonia*e, manage to evade every drug within the physician's arsenal.

Bacterial Resistance Strategies

To survive in the presence of an antibiotic, bacterial organisms must be able to disrupt one or more of the essential steps required for the effective action of the antimicrobial agent. The intended modes of action of antibiotics may be counter-acted by bacterial organisms via several different means. This may involve preventing antibiotic access into the bacterial cell or perhaps

removal or even degradation of the active component of the antimicrobial agent. No single mechanism of resistance is considered responsible for the observed resistance in a bacterial organism. In fact, several different mechanisms may work together to confer resistance to a single antimicrobial agent.

Four major bacterial resistance strategies:

(1) By prevention of the antimicrobial from reaching its target by reducing its ability to penetrate into the cell

(2) By expulsion of the antimicrobial agents from the cell via general or specific efflux pumps

(3) By inactivation of antimicrobial agents via modification or degradation

(4) By modification of the antimicrobial target within the bacteria

(Fig. 1) Mechanisms of Resistance Against Different Antimicrobial Classes

ANTIMICROBIAL CLASS	MECHANISM OF RESISTANCE	SPECIFIC MEANS TO ACHIEVE RESISTANCE	EXAMPLES
Beta-lactams Examples: penicillin, ampicillin, mezlocillin, peperacillin, cefazolin, cefotaxime, ceftazidime, aztreonam, imipenem	Enzymatic destruction	Destruction of beta-lactam rings by beta-lactamase enzymes. With the beta-lactam ring destroyed, the antibiotic will no longer have the ability to bind to PBP (Penicillin-binding protein), and interfere with cell wall synthesis.	Resistance of staphylococi to penicillin; Resistance of Enterobacteriaceae to penicllins, cephalosporins, and aztreonam
	Altered target	Changes in penicillin binding proteins. Mutational changes in original PBPs or acquisition of different PBPs will lead to inability of the antibiotic to bind to the PBP and inhibit cell wall synthesis	Resistance of staphylococci to methicillin and oxacillin
	Decreased uptake	Porin channel formation is decreased. Since this is where beta-lactams cross the outer membrane to reach the PBP of Gram-negative bacteria, a change in the number or character of these channels can reduce betalactam uptake..	Resistance of Enterobacter aerogenes, Klebsiella pneumoniae and Pseudomonas aeruginosa to imipenem
Glycopeptides Example: vancomycin	Altered target	Alteration in the molecular structure of cell wall precursor components decreases binding of vancomycin so that cell wall synthesis is able to continue.	Resistance of enterococci to vancomycin
Aminoglyosides Examples: gentamicin, tobramycin, amikacin, netilmicin, streptomycin, kanamycin	Enzymatic modification	Modifying enzymes alter various sites on the aminoglycoside molecule so that the ability of this drug to bind the ribosome and halt protein synthesis is greatly diminished or lost entirely.	Resistance of many Gram-positive and Gram negative bacteria to aminoglycosides
	Decreased uptake	Change in number or character of porin channels (through which aminoglycosides cross the outer membrane to reach the ribosomes of gram-negative bacteria) so that aminoglycoside uptake is diminished.	Resistance of a variety of Gram-negative bacteria to aminoglycosides
	Altered target	Modification of ribosomal proteins or of 16s rRNA. This reduces the ability of aminoglycoside to successfully bind and	Resistance of Mycobacterium spp to streptomycin

		inhibit protein synthesis	
Quinolones Examples: ciprofloxacin, levofloxacin, norfloxacin, lomefloxacin	Decreased uptake	Alterations in the outer membrane diminishes uptake of drug and/or activation of an "efflux" pump that removes quinolones before intracellular concentration is sufficient for inhibiting DNA metabolism.	Resistance of Gram negative and staphylococci (efflux mechanism only) to various quinolones
	Altered target	Changes in DNA gyrase subunits decrease the ability of quinolones to bind this enzyme and interfere with DNA processes	Gram negative and Gram positive resistance to various

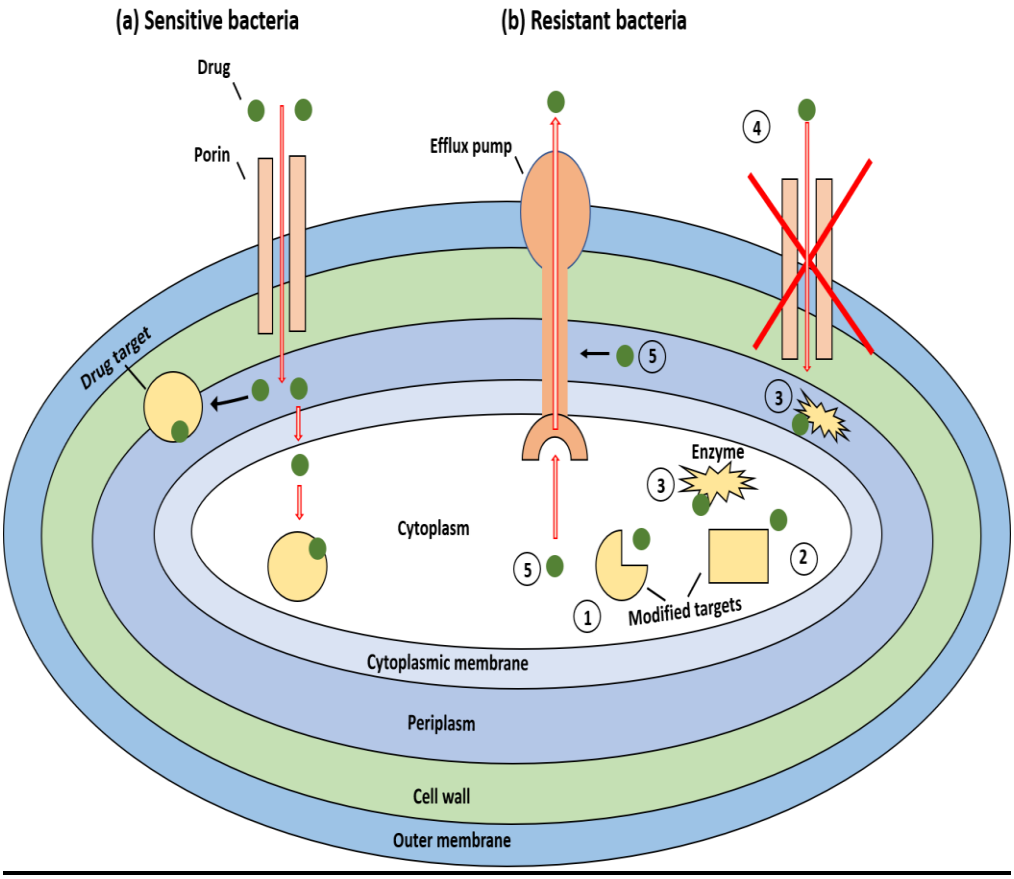
Molecular mechanisms of resistance

The abilities of bacterial organisms to utilize the various strategies to resist antimicrobial compounds are all genetically encoded.

Intrinsic Resistance

Intrinsic resistance is the innate ability of a bacterial species to resist activity of a particular antimicrobial agent through its inherent structural or functional characteristics, which allow tolerance of a particular drug or antimicrobial class. This can also be called "insensitivity" since it occurs in organisms that have never been susceptible to that particular drug. Such natural insensitivity can be due to:

- lack of affinity of the drug for the bacterial target
- inaccessibility of the drug into the bacterial cell
- extrusion of the drug by chromosomally encoded active exporters
- innate production of enzymes that inactivate the drug



Acquired Resistance

Acquired resistance is said to occur when a particular microorganism obtains the ability to resist the activity of a particular antimicrobial agent to which it was previously susceptible. This can result from the mutation of genes involved in normal physiological processes and cellular structures, from the acquisition of foreign resistance genes or from a combination of these two mechanisms.

Unlike intrinsic resistance, traits associated with acquired resistance are found only in some strains or subpopulations of each particular bacterial species. Acquired resistance results from successful gene change and/or exchange that may involve: mutation or horizontal gene transfer via transformation, transduction or conjugation.

Mutation

A mutation is a spontaneous change in the DNA sequence within the gene that may lead to a change in the trait which it codes for. Any change in a single base pair may lead to a corresponding change in one or more of the amino acids for which its codes, which can then change the enzyme or cell structure that consequently changes the affinity or effective activity of the targeted antimicrobials.

In prokaryotic genomes, mutations frequently occur due to base changes caused by exogenous agents, DNA polymerase errors, deletions, insertions and duplications. For prokaryotes, there is a constant rate of spontaneous mutation of about 0.0033 mutations per DNA replication that is relatively uniform for a diverse spectrum of organisms. The mutation rate for individual genes varies significantly among and within genes (Gillespie, 2002).

Horizontal Gene Transfer

Horizontal gene transfer, or the process of swapping genetic material between neighboring "contemporary" bacteria, is another means by which resistance can be acquired. Many of the antibiotic resistance genes are carried on plasmids, transposons or integrons that can act as vectors that transfer these genes to other members of the same bacterial species, as well as to bacteria in another genus or species. Horizontal gene transfer may occur via three main mechanisms: transformation, transduction or conjugation.

Transformation involves uptake of short fragments of naked DNA by naturally transformable bacteria. Transduction involves transfer of DNA from one bacterium into another via bacteriophages. Conjugation involves transfer of DNA via sexual pilus and requires cell –to-cell contact. DNA fragments that contain resistance genes from resistant donors can then make previously susceptible bacteria express resistance as coded by these newly acquired resistance genes.

Test Methods in Detecting Antimicrobial Resistance

There are several antimicrobial susceptibility testing methods available today, and each one has their respective advantages and disadvantages. They all have one and the same goal, which is to provide a reliable prediction of whether an infection caused by a bacterial isolate will respond therapeutically to a particular antibiotic treatment. Some examples of antibiotic sensitivity testing methods are:

- Dilution method (broth and agar dilution method)
- Disk-diffusion method
- E-test
- Automated methods

- Mechanism-specific tests such as beta-lactamase detection test and chromogenic cephalosporin test
- Genotypic methods such as PCR and DNA hybridization methods

Selection of the appropriate method will depend on the intended degree of accuracy, convenience, urgency, availability of resources, availability of technical expertise and cost. Among these available tests, the two most commonly used methods in veterinary laboratories are the agar disk-diffusion method and the broth microdilution method.