Antimicrobials and Resistance- A Review

Vikram Singh Chauhan[a], Rakesh Bhargava[b]

[a]Covance Inc., Bengaluru, India560001, India

[b]Defense Research and Development Establishment (DRDE), DRDO, Gwalior, India474002.

Abstract

The Social Discipline taught by (Covid-19) Corona consequences may also be effectively used as an awareness drive to educate the masses to restrict the intake of antibiotics as well as the medical professionals to follow the prescriptive guidelines on antibiotics. Since Anti-Microbial Resistance (AMR) entails prolonged illness and thus increases financial burden, it is prudent to be educated about the wealth of good health. In spite of efforts to develop antibiotics, new resistance mechanisms are emerging and spreading globally threatening our ability to treat common infectious diseases. The search for the treatment should be continuous by anticipating and preempting possible defense strategy of bacterial community.

KEYWORDS: Anti-Microbial Resistance, Infection, Drug-resistant organisms, Bacterial cell wall, Antibiotics

Introduction:

There has been a see-saw pattern between the bacterial fraternity and the antibiotics family to win the race. Bacteria are endowed with power to survive in the environment and are equipped with mechanism to evade the onslaught of antibiotic molecules. The ability of bacteria to neutralize the properties of antibiotics using its defense mechanism results in Antibiotic resistance.

The Arrival of antibiotics became one of the critical steps for tackling medical exigencies, the causes to elicit subsequent resistance to antibiotics can be attributed to factors like human fallacy and natural adaptation of bacteria by their inherent genetic plasticity to set up responses that results in resistance to antibiotic treatments. About half a million people developed multi-drug resistant TB globally alone in 2016[1]. It is known that antimicrobial resistance sets up gradually over time through genetic changes, the misuse and overuse of antimicrobials accelerates this process. Inappropriate use of available antibiotics and self-medication has emerged as one of the most important contributing factors for the anti-microbial-resistance (AMR)[2]. Though improper use of antimicrobials magnifies the AMR problem, there is increasing evidence that medicine quality may be another significant factor. Medicines with lower dose of the active ingredient can lead to resistance[3]. An interesting trend has been noted by Horowitz and Moehring, that resistance towards an antibiotic tends to increase as soon as the patent on that particular drug expires, as other Pharmaceutical

companies can also produce the drug resulting in fall of price and easy availability. This 'open-access' problem causes excessive use and the resistance problems ensures[4]. Meta analyses of the drug susceptibility results of various laboratories in India reveal an increasing trend of development of resistance to commonly used antimicrobials in pathogens like *Salmonella, Shigella, Vibrio cholerae, Staphylococcus aureus, Neisseria gonorrhoeae, N. meningitidis, Klebsiella, Mycobacterium tuberculosis*, HIV, plasmodium and others.[5-10] . AMR scenario exacerbates by large quantities of counterfeit and sub-standard antimicrobials permeating the pharmaceutical markets in some regions [6].

A financial factor can also be attributed towardsAnti-Microbial Resistance, where initial relieve in symptoms may lead to the discontinuation of costly(advanced) drugs and saving on money at the cost of health. Human population is also being exposed to antibiotics on regular basis from dairy products. A significant amount of antibiotics is used in animal husbandry and this sector alone accounts for almost two-thirds of the predicted growth in antibiotic usage globally. Multiple studies in India, have consistently shown that a large proportion of the milk samples contain antibiotic residues[11 -14].

A recent study from the unorganized as well as organized dairy farms reported presence of Tetracycline, Oxytetracycline, Sulfadimidine and Sulfamethoxazole above minimum recommended level (MRL) in milk samples[15]. Similarly, antimicrobial residues were reported in 23.3% dairy farm in the settings similar to the current study[16].

One of the reasons put forward for aggravating the antibiotic resistance in humans through dairy, was the continual use of antibiotics after parturition to prevent disease and sale of milk without adhering to withdrawal period. Added to this, though Methicillin Resistant *Staphylococcus aureus (*MRSA*)* infections initially originated from hospitals but are now caught in the community from other people and livestock [17]. This superbug has become resistant to beta- lactam antibiotics, a broad-spectrum group which includes Methicillin, oxacillin and the cephalosporins and is now endemic in India. Thus, increase in drug-resistant pathogens is a consequence of multiplefactors.

Infections caused by drug-resistant organisms are associated with increased mortality compared to those caused by susceptible bacteria. It is known that AMR entails prolong illness and increased financial burden. The economic costs of AMR per antibiotic consumed are high, often exceeding their purchase cost [18]. The Centers for Disease Control and Prevention estimates, that at least 23,000 people die annually as a result of infections with an antibiotic- resistant organism [19]. Moreover, according to a recent report, antibiotic resistance is estimated to cause around 300 million premature deaths by 2050 with a loss of up to $100 trillion to the global economy [20]. Consumption of Antibiotics in India is highest among the Asian countries followed by China and Pakistan. A recent pharmaceutical sales data on antibiotic usage showed an

increase of 103% between 2000 and 2015 in India. Global consumption of antibiotics in human medicine rose by nearly 40% between 2000 and 2010 [16-18,21-23].
There is considerable variation in the patterns of AMR globally, with countries often experiencing different level of problems[24-25]. However Anti-Microbial-Resistance is a problem that should concern every country. According to a recent report, if there are no new and effective antibiotics produced, there will be more than 10,000,000 deaths per year associated with antibiotic-resistant infections, with an associated cost to the global economy of ~$1 trillion by 2050[26]. The World Health Organization has named antibiotic resistance as one of the three most important public health threats of the 21st century[27].

There are a number of excellent reviews published elsewhere describing varieties of antibiotic and their actions as well as resistance mechanisms, but the current AMR scenario world over, keeps reminding us to revisit the subject. The present paper reviews certain mile stones of antibiotics development, onset of AMR and possible steps that may be useful in reducing the menace of this resistance to an extent. The social discipline taught by (Covid-19) corona consequences may effectively be used as an awareness drive to educate the masses against the AMR that kills insidiously but to the similar magnitude.

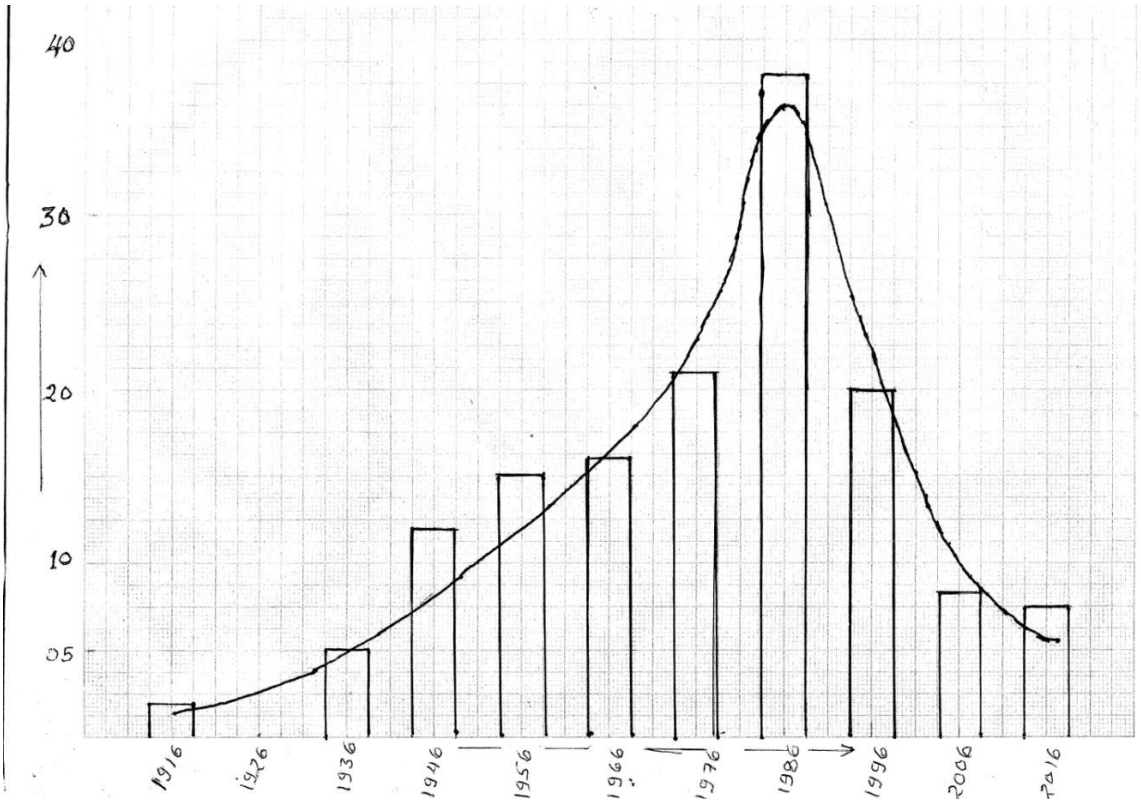
A Few mile stones:

Alexander Fleming is credited to discover the first antibiotic substance Benzylpenicillin (Penicillin G) from the mould *Penicillium notatum*in 1928, for which he shared the Nobel Prize in Physiology and Medicine in 1945 with Howard Florey and Ernst Boris Chain [28] . However, the term antibiotics was coined by Selman Waksman in 1943, the discoverer of streptomycin [29]. The first antibiotic resistance was reported in 1946 to staphylococci[30]. The first commercially available antibacterial was Prontosil, a sulfonamide developed by the German biochemist Gerhard Domagk a Nobel Prize winner in 1939[31]. Sulfonamide drugs were the first antibacterial to be used systemically, and paved the way for the antibiotic revolution in medicine. Subsequently newer drugs were introduced with mode of action to neutralize bacterial properties using various mechanisms32. The first semi-synthetic antibiotics namely Methicillin and Ampicillin were made from penicillin molecule[33]. Chloramphenicol became the first fully synthetic antibiotic, whose scaffold originated from a natural product. Fluoroquinolonesare currently the third most prescribed antibiotic to outpatients, behind macrolides and beta-lactams [34]. The antimicrobial effect is due to the formation of a DNA gyrase-quinolone-DNA complex, which hampers replication and induces cellular death in both Gram-positive and Gram-negative pathogens [34].

Figure 1, depicts a pattern of antibiotics that came into the market since 1900 till 2020[34]. They cover the antibiotics that used mechanism to kill bacteria or make them ineffective. Presently the curve is negatively skewed, as higher numbers of drugs were

marketed till 1990s. The pattern is likely to remain so in coming years as a smaller number of newer antibiotics is expected due to required extended research time which entails financial and technical consequences. They cover the antibiotics that used mechanism to kill bacteria or make them ineffective.

Figure 1



Drug development or discovery constitutes the process of designing or finding molecules that could lead to new therapies. The period of 1950s to 60s is considered as the "golden age" of antibiotic discovery due to bringing forth lifesaving drug classes, such as erythromycin, vancomycin, and metronidazole [35-37].

After mid 1930s the availability of antibiotics became fast with consistent increase in the number per decade and peaked during1980s. More than 55% of drug molecules were marketed through 70s and 80s. Thereafter, the number decreased significantly. The majority of antibiotic compounds released to market around 1980s and after, was mostly derivatives of the existing (made during 50s & 60s) classes to better target newer infections or to combat bacterial resistance. Cephalosporins, for example had 32 derivatives of β-lactam molecule resulting in five generations of antibiotics. Penicillin had 18 different derivatives to better target and control cell wall synthesis. In the current scenario almost all currently available antibiotic is a derivative of a class discovered between the early 1900s and1984[38].

Antibiotics with derivatives were found to become resistant early perhaps due to the cross-resistance that often preexisted within the microprobes. There are some molecules that were effective for very long duration like Oxazolidinones which remained useful for a period of about 46 years from 1955 to 2001. Streptogramins however had the shortest span of one year for its discovery in 1963. The use of glycopeptides in the outpatient setting has led to the search for longer-acting agents. Dalbavancin first underwent clinical trials in 2007, but it did not become available until2014[39-41].

Mechanism ofAction:

Bacterial cell wall

Bacterial cell wall being the first line of defense has been the target of certain antibodies to breach it so as to destroy the bacteria. Antibiotics like β- lactam (Penicillins, e.g. pen V, penicillin G, procaine penicillin G, benzathine penicillin G, methicillin, oxacillin, cloxacillin, dicloxacillin, flucloxacillin, cephalosporins), had been designed to disrupt the normal synthesis of the bacterial cell wall, which is needed to remain viable. In order to counter the effect of such antibiotics, bacteria respond by increasing the synthesis of β- lactamase to remain viable and thus make the antibiotics ineffective[42]. Therefore, another antimicrobial had been designed to counter the defense mechanism of bacteria. Antibiotics with β-lactam/β-lactamase inhibitors like *imipenem, meropenem, aztreonam, ticaracillinclvulnate* and *piperaciin-tazobactam* were discovered. *cefotaxime, ceftriaxone, ceftazidime,* and *cefepime* are other β-lactam inhibitors[43-45]. Resistance to a particular antimicrobial or its derivatives is acquired through multiple biochemical pathways. As bacterial cells utilize a range of mechanisms to survive the effect of antibiotics and turn resistant[46]. In certain cases, different mechanisms of resistance have been observed in gram-negative and gram-positive bacterial species. The predominant mechanism of resistance to β-lactams in gram-negative bacteria is the production of β-lactamases, whereas resistance to β-lactams in gram-positive organisms is mostly achieved by modifications of their target site, the penicillin-binding proteins (PBPs)[47-48].

Not all bacteria from the population get killed by the action of potent antibiotics. Every time a bacterium multiplies and grows, it may develop a mutation in its DNA. The replication error from the survived bacteria create mutant population that become resistant to the subsequent antibiotics dosage. The resistance acquired by mutational changes is diverse and complex and is achieved by adopting strategies like decreasing the affinity for the imposed drug, the drug uptake reduction, activation of efflux mechanisms to eject the harmful molecule, or inducing the overall changes in metabolic pathways [49].

Bacterial membrane

Besides the cell wall, the bacterial membrane provides a unique and attractive target for

antimicrobial agents. It is known that the bacterial membrane is arranged differently from a mammalian membrane where anionic lipids are exposed on the surface of bacterial membranes, while in eukaryotic membranes, anionic lipids are sequestered to the monolayer facing the interior of the cell or organelle. The cytoplasmic membrane, which covers the cytoplasm, serves as a selective barrier and controls the internal composition of the cell. The bacterial membrane barrier is responsible for establishing concentration and electrical gradients between the cell and its environment. These gradients can be disrupted by damage to the cell membrane by the antibiotics. One such gradient is the transmembrane proton gradient. Several drugs or drug combinations have been shown to be potent antimicrobial agents as a consequence of inhibiting drug efflux as well as being effective against resistant bacterial strains. Polymyxin B and colistin are antibiotics that disrupt the cell membrane. Antimicrobials, which destroy bacteria by targeting the cell wall or cell membrane of the bacteria, are termed bactericidal. Whenever these functional roles of the cytoplasmic membrane get disturbed, molecules and ions will outflow, resulting in cell destruction or death. Many antimicrobial agents are designed cationic so that they have greater selectivity for bacterial membranes [50-52].

Certain antimicrobials have been found to inhibit the synthesis of lipopolysaccharide , a component of the Gram-negative bacteria by binding to the outer membrane and block passage of solutes between the periplasm and the cell exterior, resulting in bacterial toxicity. Recently, fosfomycin has been used in the treatment of multidrug-resistant Gram-negative bacteria[53-59].

Antibiotics affecting protein synthesis: Like any other living organism, bacteria also possess DNA having proteins and enzymes codes for survival and development. Quinolones were developed to target gyrase to stop the process of protein synthesis and hence to act as bacteriostatic. These antibiotics have multiple roles in that they wade through the cell wall and cell membrane and reach the nucleus to stop the process of protein synthesis. Though the antibiotics properties of Quinolones were discovered in 1962 and patent granted in 1978, it came to the market in USA in 1986. The other antibiotics of this class include Ofloxacin (Floxin), Norfloxacin(Noroxin), Ciprofloxicin (Cipro) and Moxifloxacin (Avelox). These have broad spectrum of activities and are effective for both Gram +ve and gram –ve bacteria. Tigecycline is a protein synthesis inhibitor. It belongs to the glycylcycline class and has a broad spectrum of activity. It is a derivative of tetracycline, and was introduced in 2005. It was the first broad- spectrum agent to be licensed since moxifloxacin in 2000. It binds reversibly to the 30S bacterial ribosomal subunit, and blocks the binding of amino-acyl-tRNA to the acceptor site on the mRNA complex. It restricts the inclusion of amino acids to the developing peptide chain, thereby inhibiting further protein synthesis. Likewise, Erythromycin, Clarithromycin and Azithromycin work as inhibitors of protein synthesis and comes under are macrolides. Macrolides are protein synthesis inhibitors, which bind to the 50S ribosomal subunits, impeding peptidyl transfer. Tetracycline and glycylcycline (tigecycline) bind to the 30S ribosomal subunit. Protein translation gets disturbed by these inhibitors. Streptogramins interfere in protein translation through prevention of initiation, elongation, and translocation

stages and free tRNA depletion [60-62].Daptomycin by virtue of its structural characteristics causes cell membrane depolarization and potassium ion efflux leading to the inhibition DNA, RNA and protein synthesis resulting in bacterial cell death. Daptomycin, which targets both membrane function and peptidoglycan synthesis, is especially effective in treating staphylococcal infections. Though, Daptomycin was first evaluated in the late 1980s but, trials were halted owing to reported adverse musculoskeletal effects, but the agent was resuscitated and launched in the USA in 2003 [61-65].

Antibiotic resistance to such antibiotics is developed by the acquisition of tiny fragments of DNA known as plasmids or transposons either from another bacterium by conjugation or through virus invasion, and inject DNA fragments by a process known as transduction. Another genetic mechanism of antibiotic resistance is the acquisition and accumulation of resistance genes from neighboring bacteria through a process known as transformation[62].

Newer Antibiotics?

Detailsoffollowingtennewantibioticshavebeenpresentedin IDweek2017symposium088in SanDiegoCA[66-67]. They are *Meropenem and Vaborbactam* (Vabomere Medicines Company) is a fixed dose combination of Meropenem and Vaborbactam is effective for complicated urinary tract infection or acute pyelonephritis. It is effective against both Gram+ ve as well as Gram –ve bacteria. *Lefamulin* (Nabria Therapeutics AG) antibiotic is for community acquired bacterial pneumonia and also showed activity for common STD. *Fosfomycin* (Zavante Therapeutics) developed for complicated urinary tract infections, hospital acquired bacterial pneumonia, ventilator associated bacterial pneumonia, skin infections and complicated abdominal infections. It is effective against both Gram+ ve and Gram –ve bacteria. *Cefiderocol*is found to be effective against Gram negative pathogens including that are highly resistant to Colistin and Carbapenem as well as resistant strains of pseudomonas aeruginosa, Acinetobacter baumannii and enterobacteriaceae.However, no appreciable activity was reported against Gram positive organisms. *Plazomicin*(Achaohen) is effective in the treatment of blood stream infections due to Carbapenem resistant enterobacteriaceae. *Omadacycline, an* oral and intravenous antibiotic for the treatment of community acquiredbacterialinfections.*Iclaprim*antibioticshasbeendevelopedforskinandhospitalacquiredinfections.

*Relebactam,*a beta lactamase inhibitor developed for hospital acquired pneumonia or ventilator associated bacterial infections. *Delafloxacin* (Baxdela- Melintatherapeutics) aims to treat acute bacterial skin and skin structure infections and for community acquired bacterial pneumonia. It is in the phase 3 trial.

Concluding remarks:

Rather than depending solely on medicine, it better to take precautions to avoid getting trapped by bacteria by following good hygiene practices, avoiding antibiotics for viral infection and not skipping prescribed antibiotic regime. The survival wisdom endowed to bacteria by Mother Nature seems to outpace human imagination in designing new antibiotics. If one traces the key events of antibiotic resistance and arrival of new antibiotics from Penicillin (1943) to Ceftaroline (2010), it appears that whatever the mode of action of these drugs on bacteria is, will adjust accordingly and survive. New resistance mechanisms are emerging and spreading globally, threatening our ability to treat common infectious diseases. However, efforts to develop antibiotics should be continuous, anticipating and preempting possible defense strategy of bacterial community. The other aspect to reduce the antibiotic resistance is to discipline ourselves towards medication by respecting the prescription. Intense and continual education drive to be enforced for letting people knows the menace of drug resistance. Though World Antibiotic Awareness Week is held every November since 2015 with the theme "Antibiotics: Handle with care", it has not reached the level of fervor itdeserves.

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