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Review Article

REVIEW ON MULTI DRUG RESISTANT BACTERIA AND ITS IMPLICATION IN MEDICAL SCIENCES

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Abstract

Many procedures, use and misuse of antibiotics in man have resulted in antibiotic-resistant bacteria. Many aspects like Drug efflux pumps change in the targets in the sites which are targeted by antibiotics by mutation and many other aspects produce multi drug resistance by extruding antibiotics from bacteria. Treating these multidrug-resistant bacteria with multiple antibacterial drugs becomes very difficult. No effective treatment exists for multi drug resistant bacteria. Here below is a review on the emergence, mechanism & the reasons of the multi drug resistance bacteria.

Keywords: Drug resistance, Bacteria, antibiotic.

INTRODUCTION

Mutation and selection, together with the mechanisms of genetic exchange, enable many bacterial species to adapt quickly to the introduction of antibacterial agents into their environment. Although a single mutation in a key bacterial gene may only slightly reduce the susceptibility of the host bacteria to that antibacterial agent, it may be just enough to allow its initial survival until it acquires additional mutations or additional genetic information resulting in full-fledged resistance to the antibacterial agent.¹ However, in rare cases, a single mutation may be sufficient to confer high-level, clinically significant resistance upon an organism.¹

One of the methods used by various authors and authorities to characterize organisms as MDR is based on in vitro antimicrobial susceptibility test results, when they test 'resistant to multiple antimicrobial agents, classes or subclasses of antimicrobial agents' The definition most frequently used for Gram-positive and Gram-negative bacteria is 'resistant to three or more antimicrobial classes'. Another method used to characterize bacteria as MDR, is when they are 'resistant to one key antimicrobial agent'. These bacterial isolates may have public health importance due to resistance to only one key antimicrobial agent, but they often demonstrate cross or co-resistance to multiple classes of antimicrobials, which makes them MDR. Creating an acronym for a bacterium based on its resistance to a key antimicrobial agent (e.g. methicillin resistance in *Staphylococcus aureus*, i.e. MRSA) immediately highlights its epidemiological significance; the advantage of using this approach for surveillance purposes is that it can be easily applied.²

Multidrug-resistant organisms (MDROs) are microorganisms that are resistant to one or more therapeutic classes of antimicrobial agents. The number of MDROs will increase if

the selective pressure of antibiotic use continues and the resistant organism is able to spread from one person to another. The MDROs of greatest concern to healthcare facilities include (1) methicillin-resistant *Staphylococcus aureus* (MRSA), (2) vancomycin-resistant enterococci (VRE), (3) multidrug-resistant (MDR) gram-negative bacilli (such as *Enterobacter*, *Klebsiella*, *Acinetobacter*, and *Pseudomonas* species and *Escherichia coli*), and (4) vancomycin-resistant *Staphylococcus aureus*. For some MDR gram-negative bacilli, such as carbapenem-resistant *Enterobacter* species and extended-spectrum β -lactamase-producing *Klebsiella* species, the specific drug resistance patterns cause concern because of the challenges they present in treatment and infection prevention.³

Several factors may lead to the increase in antimicrobial resistance, such as hospital stay before ICU admission, hospitalization period before ICU admission, length of ICU stay, surgical ICU stay, the type of operation, previous antibiotic use, inappropriate use of antimicrobial drugs, and inadequate adherence to infection control practices. In particular, some patients are more vulnerable to colonization and infections including those with severe disease, those with compromised host defenses because of underlying medical conditions, patients with recent surgery, and those with indwelling medical devices. Furthermore, hospitalized patients are likely to have more risk factors and higher infection rates than non-hospitalized patients, especially those who require treatment in the ICU.⁴

It has been shown that antimicrobial drug resistance genes are present in one of the most remote areas on Earth, the Arctic. Resistant as well as multiresistant isolates of *E.coli* were detected in the normal flora of Arctic birds. This finding highlights the unique nature of bacterial adaptation and the complexity of dissemination of antimicrobial drug resistance.

To fully understand the extent of environmental and commensal reservoirs of resistance, studies of antimicrobial drug resistance in different habitats are warranted.⁵

The basic aim of this review is to know how potential these multi drug organisms are to the mankind, which includes the wisdom of antibiotics, MDROs for antibiotics, impact and possible method of prevention.

History of Antibiotics

The first antimicrobial agent in the world was salvarsan, a remedy for syphilis that was synthesized by Ehrlich in 1910. In 1935, sulphonamides were developed by Domagk and other researchers. These drugs were synthetic compounds and had limitations in terms of safety and efficacy. In 1928, Fleming discovered penicillin. He found that the growth of *Staphylococcus aureus* was inhibited in a zone surrounding a contaminated blue mold (a fungus from the *Penicillium* genus) in culture dishes, leading to the finding that a microorganism would produce substances that could inhibit the growth of other microorganisms. The antibiotic was named penicillin, and it came into clinical use in the 1940s. Penicillin, which is an outstanding agent in terms of safety and efficacy, led in the era of antimicrobial chemotherapy by saving the lives of many wounded soldiers during World War II.⁶

Ehrlich was followed by Alexander Fleming, who discovered penicillin by accident in 1928. Then, in the 1935, Gerhard Domagk discovered the sulfa drugs, thereby paving the way to the discovery of the anti-TB drug Isoniazid. Then, in 1939, René Dubos became the first scientist to discover an antibiotic after purposely looking for it in soil microbes. Dubos discovered Gramicidin, which is still used today to treat skin infections.⁷

During the subsequent two decades, new classes of antimicrobial agents were developed one after another, leading to a golden age of antimicrobial chemotherapy. In 1944, streptomycin, an aminoglycoside antibiotic, was obtained from the soil bacterium *Streptomyces griseus*. Thereafter, chloramphenicol, tetracycline, macrolide, and glycopeptide (e.g., vancomycin) were discovered from soil bacteria. The synthesized antimicrobial agent nalidixic acid, a quinolone antimicrobial drug, was obtained in 1962.⁶

Mode of action of antibiotics

Inhibition of protein synthesis & nucleic acid

Many antibiotics interfere in the different stages protein synthesis process like elongation of nascent polypeptide chains, peptidyltransferase reaction, and arrest translation. Antibiotics interfere in the activities of the enzyme like gyrase and polymerases in DNA synthesis.^{1,8,9}

Inhibition of a metabolic pathway

Many antibiotics block the metabolic pathway which includes the synthesis of the nucleotides, essential components of DNA & RNA.^{1,8,9}

Disorganizing of the cell membrane & cell wall synthesis

Antibiotic targets the glycopeptides basic components of cell wall. An antibiotic also targets the inhibition peptidoglycan biosynthesis through preferential targeting of transglycosylation. An antibiotic also attacks the channels on the membrane which are responsible for the creation isotonic on the bacterial cell, leading to cell death.^{1,8,9}

History of MDRO

The first report of an MRSA strain with reduced susceptibility to vancomycin reported as a vancomycin-intermediate *S.aureus* (VISA) appeared in Japan in 1997³⁶; this was followed by confirmed VISA cases in the United States. The first case of VRSA (vancomycin resistant *S aureus*) involved a 40-year-old woman from Michigan who was undergoing dialysis.¹

In 1959, the Japanese found *Shigella* species that were resistant to Sulfonamides, Streptomycin, Chloramphenicol, and Tetracycline. The resistance was due to plasmid, which carried different antibiotic resistance genes.⁷

Within four years following the introduction of penicillin during the Second World War, occurrence of resistant strains was reported. According to an estimate by the The Centers for Disease Control and Prevention (USA), 13,300 patients died of antibiotic-resistant bacterial infection in the US during 1992. An incredible 150% increase in the occurrence of drug-resistant Pneumococci was noted between 1987 and 1994. A 20-fold increase in the frequency of hospital-acquired Enterococci, resistant to vancomycin, was seen between 1989 and 1993. The frequency of methicillin-resistant *Staphylococcus rose* from 2% in 1975 to 32% in 1992. By this time, resistance to virtually all the therapeutically useful antibiotics had been evidenced. Emergence of methicillin-resistant *Staphylococcus aureus*(MRSA) and vancomycin-resistant Enterococci (VRE) have raised serious concern all over the world since these two antibiotics were believed to be invincible when they were released in the market. A couple of years back, some Gramnegative bacteria (*Escherichia coli*, *Klebsiella pneumoniae*) were found to produce an enzyme, which confers resistance to virtually all the commonly used antibiotics including carbapenems, one of the last resorts in the clinical management of infections caused by multidrug-resistant organisms. The organisms were believed to be originated from India. That is why the enzyme was named New Delhi Metallo E-lactamase (NDM-1)⁹

Mechanism, Biochemical & Genetic aspects: MDRO

Antibiotic resistance can be divided into genetic & phenotypic resistance

The below mention talks about the genetic resistance:

Antibiotic inactivation by group transfer

The most diverse family of resistant enzymes is the group of transferases. These enzymes inactivate antibiotics (aminoglycosides, chloramphenicol, streptogramin, macrolides or rifampicin) by chemical substitution (adenylyl, phosphoryl or acetyl groups are added to the periphery of the antibiotic molecule). The modified antibiotics are affected in their binding to a target.⁸

Antibiotic inactivation by redox process

The oxidation or reduction of antibiotics has been infrequently exploited by pathogenic bacteria. However, there are a few of examples of this strategy. One is the oxidation of tetracycline antibiotics by the TetX enzyme. *Streptomyces virginiae*, producer of the type A streptogramin antibiotic virginiamycin M1, protects itself from its own antibiotic by reducing a critical ketone group to an alcohol at position 16.⁸

Target modification

The second major resistance mechanism is the modification of the antibiotic target site so that the antibiotic is unable to bind properly. Because of the vital cellular functions of the

target sites, organisms cannot evade antimicrobial action by dispensing with them entirely. However, it is possible for mutational changes to occur in the target that reduce susceptibility to inhibition whilst retaining cellular function.⁸

Antibiotic inactivation by hydrolysis

Many antibiotics have hydrolytically susceptible chemical bonds (e.g. esters and amides). Several enzymes are known to destroy antibiotic activity by targeting and cleaving these bonds. These enzymes can often be excreted by the bacteria, inactivating antibiotics before they reach their target within the bacteria.⁸

Efflux pumps and outer membrane (OM) permeability

The efflux pumps are the membrane proteins that export the antibiotics out of the cell and keep its intracellular concentrations at low levels. Reduced outer membrane (OM) permeability results in reduced antibiotic uptake. The reduced uptake and active efflux induce low level resistance in many clinically important bacteria.⁸

Protein synthesis interference

A wide range of antibiotics interfere with protein synthesis on different levels of protein metabolism. The resistance to antibiotics that interfere with protein synthesis (aminoglycosides, tetracyclines, macrolides, chloramphenicol, fusidic acid, upirocin, streptogramins, oxazolidinones) or transcription *via* RNA polymerase (the rifamycins) is achieved by modification of the specific target.⁸

Mutations

The first mechanism is reduced permeability or uptake of the bacteria. For example, *Neisseria gonorrhoea* porin can acquire mutations that can cause resistance to penicillin and tetracycline. Another example is *Enterobacter aerogenes* porin, which can acquire mutations that cause cephalosporin resistance.⁷

Horizontal gene transfer

A second broad category of drug resistance is due to mobile genetic elements, such as plasmids or transposons, which carry drug resistant genes. Examples as follows

- Streptomycin-resistance genes, strA- and strB, which can be carried on plasmid, and cause Streptomycin resistant.
- Sulfa drug resistance, caused by plasmids that carry the drug insensitive form of the enzyme.
- A relatively new mechanism is the plasmid-mediated qnr (quinolone resistance). The qnr gene encodes a device called pentapeptide, which is a DNA mimic. Pentapeptide binds to the DNA gyrase and thus helps prevent the quinolone drug from binding to the gyrase, thereby causing low-level resistance.

Transposons can also carry drug resistant genes.⁷

Impact of the MDROs

- Difficult-to-treat infections
- Untreatable infections
- Antibiotic use increases the spread of antibiotic-resistant bacteria
- Costs
- Antibiotic-resistant bacteria spread internationally¹⁰

Comparisons of mortality rate, length of stay in hospitals, and health care costs between patients with methicillin-susceptible *S aureus* (MSSA) bacteremia and those with MRSA bacteremia have shown that patients with MRSA bacteremia have a significantly higher rate of mortality, markedly longer lengths of hospital stay, and higher median hospital costs than patients with MSSA bacteremia.

Furthermore, surgical site infections caused by MRSA were shown to be associated with a 3.4- fold higher risk for death and a 2-fold increase in median hospital costs than surgical site infections caused by MSSA infections. According to Blot et al. in Belgium, 64 patients with MRSA bacteremia had a higher 30-day mortality rate than patients with MSSA bacteremia (53.2% vs. 18.4%, $p < 0.05$) and a higher in-hospital mortality rate (63.8% vs. 23.7%, $p < 0.05$).¹¹

Mode of transmission

MDROs are transmitted from one patient to another via the contaminated hands of direct care workers. As a result, healthcare facilities have evidence based recommendations for implementing precautions and hand hygiene. These guidelines are intended to interrupt transmission from direct or indirect contact with infected patients and their environment, and also to establish when precautions should be implemented and discontinued. However, these recommendations apply to known MDRO infections; a patient's MDRO status may remain unknown unless the facility performs active surveillance or until staffs completes the chart review, which may occur up to two days after admission. Active surveillance, in particular, enables facilities to improve disease control by quickly identifying MDRO-positive patients and then implementing guidelines, policies and procedures.¹²

Contact transmission is the primary mode of spread for MDRO:

Transient carriage on the hands of health care workers is a significant risk for transmission of MDROs.

Surfaces and equipment can also become reservoirs of MDRO and contribute to spread within the healthcare environment.

Droplet transmission may also be implicated in the spread of MDRO when the patient has a respiratory tract infection where the MDRO is causative organism.¹³

Future development of antibiotics

Surveillance is an important component of all long term care infection control programs. Surveillance should include maintaining a confidential line listing of residents colonized and/or infected with targeted MDROs. Monitoring culture and antibiotic susceptibility data will help determine baseline rates for MDROs in a facility, indicate the occurrence of increased transmission, and monitor the effectiveness of outbreak control measures. In some health care settings it may be appropriate to use active surveillance cultures to identify patients who are colonized with a targeted MDRO, but this would rarely be the case in a Long Term Care Facilities.^{14, 15}

Biofilm formation: Initially, the bacteria simply attaches to surfaces irreversibly, and then irreversibly. Then, early biofilms are formed, and turn into mature biofilms. They are then able to release new organisms off the structure. Biofilm bacteria are extremely resistant to antibiotics. When we compare the susceptibility of the planktonic form and biofilm, we observe that antibiotic imipenem can destroy planktonic organisms of *P. aeruginosa* effectively.⁷

Another approach to inhibiting biofilm formation is the use enzymes that can degrade the EPS of biofilm and detach established biofilm colonies. Moreover, biofilm-dispersing enzymes administered in combination treatment with antimicrobial agents will allow them to kill bacteria

embedded in EPS Kaplan *et al.* suggested that deoxyribonuclease I and glycoside hydrolase dispersin B are useful as anti-biofilm agents due to the dispersing action of EPS layers on medical devices. In addition, therapeutic treatment of combination treatments with antimicrobial peptides may result in significant synergetic-effects against MDR bacteria and the formation of biofilms.¹⁶

Drug resistance is also becoming a major problem in human infections involving biofilm. For example, some orthopaedic devices can have *Staphylococcus aureus* and *Staphylococcus epidermidis* infections. Once these devices are infected with the biofilm, it is extremely difficult to eliminate the biofilm completely merely by using antibiotics. Often, the orthopaedic device must be replaced.⁷

Nanoantibiotics: Nanomaterials for infection control. Nanomaterials, which either show antimicrobial activity by themselves or elevate the effectiveness and safety of antibiotics administration are called “nanoantibiotics” and their capability of controlling infections *in vitro* and *in vivo* has been explored and demonstrated. Unlike many antimicrobial agents currently being used in the clinic, antimicrobial NPs may not pose direct and acute adverse effects, although potential toxicity upon long-term exposure is questionable. Most importantly, antimicrobial NPs tackle multiple biological pathways found in broad species of microbes and many concurrent mutations would have to occur in order to develop resistance against NPs' antimicrobial activities. Preparation of antimicrobial NPs could be cost-effective, compared with antibiotics synthesis, and they are quite stable enough for long-term storage with a prolonged shelf-life. In addition, some NPs can withstand harsh conditions, such as high temperature sterilization, under which conventional antibiotics are inactivated.

Antibiotics delivery using nanomaterials offer multiple advantages:

- 1) Controllable and relatively uniform distribution in the target tissue,
- 2) Improved solubility,
- 3) Sustained and controlled release,
- 4) Improved patient-compliance,
- 5) Minimized side effects, and
- 6) Enhanced cellular internalization.¹⁷

Strategies for overcoming multidrug resistance

Targeting resistance mechanisms, Developing novel drug targets,

Mining microbial genomes,

Targeting essential genes,

Vaccines and immunomodulators,

The Use of Normal Bacterial Flora

Limiting the Spread of Drug Resistant Bacteria.^{7,18}

U turn towards the herbal medicines to fight against multi drug resistance micro organisms

Resistant bacteria strains may develop almost anywhere particularly in a pressurized environment containing previously non-resistant bacteria strains as contaminants. One of such environments can be an herbal medicinal product (HMP). HMPs have been previously implicated as a pool for such contaminations. The use of HMPs as a form of complementary medicine is becoming increasingly popular in both developing and developed countries. About 70% to 80% of the world's population particularly in the developing world has been shown to depend on the primary source of herbal

drug regimen for their health needs. It is of utmost importance to both monitor and ascertain the microbial purity of HMPs given the huge medical and economic implications of any such microbial contamination especially with multiple drug resistant strains.¹⁹

Plant based antimicrobials represent a vast untapped source. The use of plant extract for medicinal treatment has become popular when people realized that the effective life span of antibiotic is limited and over prescription and misuse of traditional antibiotics are causing microbial resistance. Traditionally used medicinal plants produce a variety of compounds of known therapeutic properties. At present, nearly 30% or more of the modern pharmacological drugs are derived directly or indirectly from plants and their extracts dominate in homeopathic or ayurvedic medicines.²⁰

In-vitro antibacterial activity of ethanolic extracts of selected commonly used herbal plants, *Ocimum gratissimum*, *Vernonia amygdalina*, *Zingiber officinale* and *Myristica fragrans* were screened against multi drug resistant bacteria including *Staphylococcus aureus*, *Proteus vulgaris*, *Bacillus subtilis*, *Salmonella typhimurium*, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* of clinical origin by agar well diffusion method. The crude extracts of the plants were fairly effective against the bacterial isolates by the values of the extracts with concentration ranging between 50 to 200mg/ml for *Ocimum gratissimum* and *Myristica fragrans* and 100 to 200mg/ml for *Vernonia amygdalina* and *Zingiber officinale*. The potency of these extracts based on their zones of inhibition (mm) and MIC values were higher in *Myristica fragrans* and *Ocimum gratissimum* which concludes that their extracts can be used against multi drug resistance bacteria capable of causing both nosocomial and community acquired infections.²¹

An alkaloid sceptrin, isolated from *Agelas sceptrum*, has been shown to possess antimicrobial activity against *S. aureus*, *Bacillus subtilis*, *C. albicans*, *Pseudomonas aeruginosa* (*P. Aeruginosa*), *Alternaria* (fungus), and *Cladosporium cucumerinum*. Bromotyrosine alkaloids have demonstrated high antimicrobial activity against a number of Gram-positive organisms, including Mycobacteria and Staphylococci, including MRSA, VRSA and VRSH. A number of peptides have also been reported to possess antimicrobial activities. Fallaxin, a 25-mer antibacterial peptide amide, has been shown to inhibit the growth of several Gram-negative bacteria including *Enterobacter cloacae*, *E. coli*, *K. pneumoniae*, and *P. aeruginosa*. Similarly, antimicrobial activities of low molecular mass lysine dendrimers against *S. aureus*, *E. coli* and *C. albicans* have been reported earlier.²²

The synergism among herbs i.e. *Murraya koenigii* and *Coriandrum sativum* showed increased antibacterial effect against some of the MDR strains. The tested organisms had shown resistance towards different antibiotics but are sensitive towards the extract. The inhibitory effect increased on the combination of the two extracts. With the increase of multiple drug resistance bacteria the concept of synergism will prove to be helpful in the treatment of various diseases and can be used as raw material for the drugs in comparison to synthetic drugs.²³

Resistance modifying activities of plants crude extracts: the basis for isolation of potentially useful compounds: If the isolation of resistance modifying compounds from plants is to be realistic, screening for such activities in crude extracts is the first step in identifying leads for isolation of

such compounds, and some plants have provided good indications of these potentials for use in combination with antimicrobial therapy. Aqueous extracts of tea (*Camellia sinensis*) have been shown to reverse methicillin resistance in MRSA and also, to some extent, penicillin resistance in betalactamase-producing *Staphylococcus aureus*. Forty to one hundred fold dilutions of tea extracts was able to reduce the MICs of high-level resistant MRSA (256 µg/ml) to less than 0.12 µg/ml for methicillin and penicillin. Aqueous crude khat (*Catha edulis*) showed varying antibacterial activities with a range of 5-20 mg/ml-1 against periodontal bacteria when tested in isolation.²⁴

In India, spices are ethnically used as active ingredients in ayurvedic medicines and reported to possess a number of pharmacological effects to treat different human ailments. Differential antimicrobial activity of herbs against different bacteria might be due to present of different active phyto-compounds. Among those antimicrobial compounds, phenolic compounds, terpenoids, and alkaloids are very important compounds in antimicrobial or antioxidant effects. Some of the known active constituents are cuminaldehyde and monoterpene hydrocarbons like β-pinene of cumin, thymol of black cumin, sinigrin glucoside of mustard, trigonelline alkaloid of fenugreek, volatile terpenes and thymol of ajowain, monoterpene hydrocarbons like α-pinene of nutmeg and polyphenols of curry-leaf and henna.²⁵

The study demonstrated that the garlic extract has showed its effectiveness against clinical isolates of MDR M. tuberculosis. It is worthwhile to utilize garlic as natural supplement with other standard. It is corresponding that substitute medicines practices with plant extracts including garlic as a means of decreasing the burden of drug resistance and reducing the cost of management of diseases would be of public health importance.²⁶

CONCLUSION

A major factor in the emergence of antibiotic resistant organisms is overuse of antibiotics in any setting, the hospital or the community. Complicating this problem is the emergence of strains that are resistant to antibiotics commonly used against these organisms. There are numerous national efforts to reduce the use of antibiotics or promote appropriate use in the community and all healthcare delivery systems. Antimicrobial resistance took a long time to develop considering that penicillin resistance began soon after its discovery in the 1940s. So it will not be fixed overnight, solutions will not be simple, and there is no single solution. There are number of national initiatives to control antimicrobial resistance by public, private and professional groups.

The modern medicine turned its way towards the herbs. The herbs have shown the significant activity in inhibiting the MDROs. It is experimentally proved by many herbs in controlling the infection of the MDROs.

REFERENCES

1. Fred C. Tenover. Mechanisms of Antimicrobial Resistance in Bacteria The American Journal of Medicine 2006; 119 (6A): S3-S10
2. A.-P. Magiorakos, A. Srinivasan, R. B. Carey, Y. Carmeli, M. E. Falagas, C. G. Giske, S. Harbarth, J. F. Hindler, G. Kahlmeter, B. Olsson-Liljequist, D. L. Paterson, L. B. Rice, J. Stelling, M. J. Struelens, A. Vatopoulos, J. T. Weber and D. L. Monnet. Extensively drug-resistant and pandrug-resistant bacteria: an international expert proposal for interim standard definitions for acquired resistance 2011; 28(3):268-281.
3. Adam L. Cohen, David Calfee, Scott K. Fridkin, Susan S. Huang, John A. Jernigan, Ebbing Lautenbach, Shannon Oriola, Keith M. Ramsey, Cassandra D. Salgado, Robert A. Weinstein Recommendations for Metrics for Multidrug-Resistant Organisms in Healthcare Settings: SHEA/HICPAC Position Paper, infection control and hospital epidemiology.2008; 29(10):901-913.
4. Shu-Hui Tseng a, Chun-Ming Lee b, Tzou-Yien Lin c, Shan-Chwen Chang d, Feng-Yee Chang, Emergence and spread of multi-drug resistant organisms: Think globally and act locally, Journal of Microbiology, Immunology and Infection 2011;44: 157-165. <http://dx.doi.org/10.1016/j.jmii.2011.03.001> PMID:21524608
5. Maria Sjölund, Jorge Hernandez, Stina Bengtsson, Gunilla Cederbrant, Jarone Pinhassi, Gunnar Kahlmeter, and Björn Olsen. Centers for Disease Control and Prevention, Atlanta, Georgia, USA. Emerging Infectious Diseases 2008; 14(1):70-72. <http://dx.doi.org/10.3201/eid1401.070704> PMID:18258081
6. Tomoo SAGA, Keizo YAMAGUCHI. History of Antimicrobial Agents and Resistant Bacteria. JMAJ, March/April 2009 — Vol. 52, No. 2
7. Ying ZHANG. Mechanisms of Antibiotic Resistance in the Microbial World, Baltimore, USA. 2007
8. Senka D'idi, Jagoda [u {kovi} and Bla'enka Kos. Antibiotic Resistance Mechanisms in Bacteria: Biochemical and Genetic Aspects, Food Technol. Biotechnol 2008; 46 (1) :11-21
9. Saswati Sengupta and Madhab K Chattopadhyay, Antibiotic Resistance of Bacteria: A Global Challenge, Resonance, February 2012; 177-191.
10. U.S. Congress, Office of Technology Assessment. (1995) Impacts of Antibiotic-Resistant Bacteria, OTA-H-629 (Washington, DC: U.S. Government Printing Office, September 1995.
11. Shu-Hui Tseng a, Chun-Ming Lee b, Tzou-Yien Lin c, Shan-Chwen Chang d, Feng-Yee Chang, Emergence and spread of multi-drug resistant organisms: Think globally and act locally, Journal of Microbiology, Immunology and Infection 2011;44: 157-165. <http://dx.doi.org/10.1016/j.jmii.2011.03.001> PMID:21524608
12. Linda Oliver, Liz Martino, Debbie Maaz, Michelle Cahill, Paula Foster, Prevention and Control of Multi-Drug Resistant Organisms Using Standardized Cross-Setting Communication, Medicine & Health/Rhode Island, September 2010; 93(9): 279-282.
13. Canterbury DHB. Volume 10 – Infection Prevention and Control Guidelines for the Control of Multidrug Resistant Organisms. 2012; 6: (1-19)
14. Massachusetts Department of Public Health Division of Epidemiology and Immunization, Multi Drug-Resistant Organisms Infection Control Guidelines for Long Term Care Facilities, 2009; 1-9
15. Chand Watal, Neeraj Goel, JK Oberoi, Reena Raveendran, S Datta, KJ Prasad, Surveillance of Multidrug Resistant Organisms in a Tertiary Care Hospital in Delhi, India Supplement to Japi December 2010; 58:32-36.
16. Seong-Cheol Park, Yoonkyung Park and Kyung-Soo Hahm, The Role of Antimicrobial Peptides in Preventing Multidrug-Resistant Bacterial Infections and Biofilm Formation, Int. J. Mol. Sci. 2011; 12: 5971-5992 <http://dx.doi.org/10.3390/ijms12095971> PMID:22016639
17. Ae Jung Huh & Young Jik Kwon a.c, "Nanoantibiotics": A new paradigm for treating infectious diseases using nanomaterials in the antibiotics resistant era Journal of Controlled Release 2011; 156: 128-145 <http://dx.doi.org/10.1016/j.jconrel.2011.07.002> PMID:21763369
18. Keith Poole. Overcoming multidrug resistance in Gram-negative bacteria, Current Opinion in Investigational Drugs 2003; 4 (2):128-139
19. Oleghe Peace Omoikhudu, Odimegwu Damian Chukwu, Udofia Ema and Esimone Charles Okechukwu, Multi-Drug-Resistant Bacteria Isolates Recovered from Herbal Medicinal, Preparations in a Southern Nigerian Setting, Rural Trop Public Health 2011; 10 : 70 - 75.
20. Md. Al Nayem Chowdhury, M. Ashrafuzzaman, Md. Hazrat Ali, Lutfun Nahar Liza, Kazi Mohammad Ali Zinnah, Antimicrobial Activity of Some Medicinal Plants against Multi Drug Resistant Human Pathogens, Advances in Bioscience and Bioengineering 2013; 1:(1)1-24
21. T.A. Ibrahim, B.O. Opawale and J.M.A. Oyinloye, Antibacterial Activity of Herbal Extracts against Multi Drug Resistant Strains of Bacteria from Clinical Origin, Life sciences Leaflets 2011; 15:490 – 498.
22. Rosina Khan, Mohammad Zakir, Sadul H. Afaq, Abdul Latif, Asad U. Khan, Activity of solvent extracts of Prosopis spicigera, Zingiber officinale and Trachyspermum ammi against multidrug resistant bacterial and fungal strains, J Infect Dev Ctries 2010; 4(5):292-300. <http://dx.doi.org/10.3855/jidc.621> PMID:20539061
23. Avantika Pandey, O. P. Verma, Natasha Nageswaran and Rachna Gupta Synergistic Antimicrobial Activity of Edible Herbs Against Multi Drug Resistant Bacteria 2012; 7(1) : 103-105.
24. T. Sibanda, and A. I. Okoh. The challenges of overcoming antibiotic resistance: Plant extracts as potential sources of antimicrobial and

- resistance modifying agents African Journal of Biotechnology Vol. 6 (25), pp. 2886-2896, 28 December, 2007
25. Das, S., Anjeza, C. and Mandal, S., Synergistic or additive antimicrobial activities of Indian spice and herbal extracts against pathogenic, probiotic and food-spoiler micro-organisms, International Food Research Journal 2012;19 (3): 1185-1191
26. Abdul Hannan , Muhammad Ikram Ullah , Muhammad Usman, Shahid Hussain , Muhammad Absar and Khursheed Javed, Anti-Mycobacterial Activity of GARLIC (*Allium sativum*) against Multi-Drug Resistant and Non-Multi-Drug Resistant Mycobacterium Tuberculosis, Pak. J. Pharm. Sci. 2011;24(1): 81-85.