RESEARCH ARTICLE

Occurrence of MDR and ESBL as respiratory pathogens.

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Manuscript details:

Received: 24.10.2016
Accepted: 24.12.2016
Published : 06.01.2017

Editor: Dr. Arvind Chavhan

Cite this article as:
Nikita Naik (2016) Occurrence of MDR and ESBL as respiratory pathogens., *International J. of Life Sciences*, 4 (4): 610-614.

ABSTRACT

In recent times, new mechanisms of resistance have resulted in the simultaneous development of resistance to several antibiotic classes creating very dangerous multidrug-resistant (MDR) bacterial strains, known as "superbugs". The indiscriminate and inappropriate uses of antibiotics in outpatient clinics, and in hospitalized patients are the largest factor leading to antibiotic resistance. This dramatic rise in the development of drug resistance, demands new strategies for the control of these strains

Key words: Multi drug resistance, extended Beta lactamase,

INTRODUCTION

Discovery of antibiotics and their use as chemotherapeutic agents, was a belief in the medical fraternity that it would lead to the eventual eradication of infectious diseases. Though this has not proved to be true, it remains a fact, that antibiotics provide the main basis in treating majority of the microbial infections. Authorities have been strongly encouraging physicians to decrease the prescription of antibiotics to treat common upper respiratory tract infections. In addition, as viruses mainly cause these infections, antibacterial antibiotics do not significantly help in reducing the recovery time of the illness. Overuse of antibiotics has proved to be a major factor in the emergence and dissemination of multi-drug resistant strains.

The worldwide emergence of ß-lactamase producing *Escherichia coli, Klebsiella pneumoniae, Haemophilus* and many other strains have currently become a major therapeutic problem. They are resistant to the β-lactam antibiotics namely penicillin, cephalosporins and some carbapenems. These strains are increasingly being isolated from hospital and community acquired infections (Zsuzsanna *et al*., 2010).

Bacteria have a strong ability to respond to antibiotic treatment by genetic as well as physiological adaptation. In doing so they also can develop resistance towards the antibiotic used against it. This makes it important to study and develop successful antibacterial mechanisms

with reduced potential for resistance development. While the pharmaceutical industry has largely been passive in developing new antibiotics in the last decades, this has resulted in emergence of resistance to newer drugs and has become the biggest challenge to the medical profession.

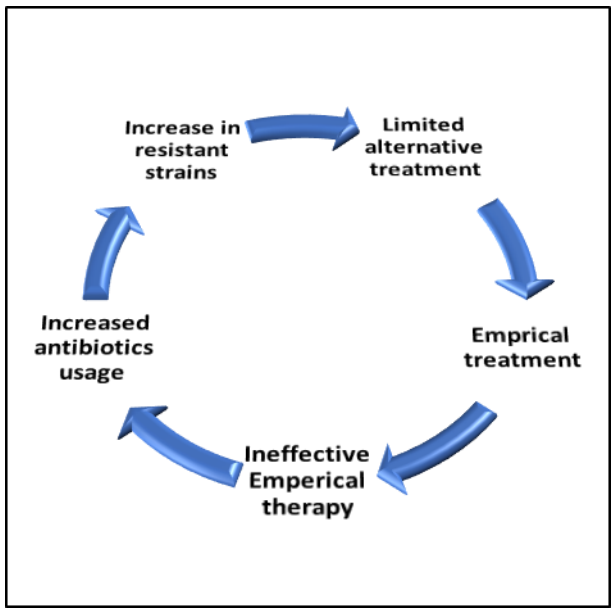

Illustration: Cycle of development of antibacterial resistance

Bacteria develop several genetic mechanisms in order to acquire resistance to antibiotics. All require either the modification of existing genetic material or the acquisition of new genetic material from another source (Jayaraman, 2009).

In 1942, first case of resistance towards Penicillin was reported and by 1960 more than 80% of both the community and hospital, acquired *Staphylococcus* isolates were resistant to penicillin. Two years, in 1961after the introduction of methicillin, *Staphylococcus aureus* developed resistance to methicillin through the acquisition of the mecA –gene (Deurenberg *et al*., 2007). The spontaneous mutation frequency of antibiotic resistance is a rare event, in the order of about of about 10^{-8}- 10^{-9}. Nevertheless, high growth rate of bacteria results in considerable resistance developed in a population. In vertical transfer, there is a direct transfer of resistance genes to all the bacterial progeny during DNA replication. The genetic determinants encoding antimicrobial resistance can be located on the bacterial chromosome or on plasmids, which may replicate independently of the chromosome.

Lateral or horizontal gene transfer (HGT), the resistance-plasmid is transferred between individual bacteria of the same species or even between different species, which spread resistance within a population. This acquisition of the genetic determinants takes place by the process of conjugation, transduction and transformation (Jayaraman, 2009).

MDR or **Multiple drug resistance** is a condition enabling the microorganism to resist the inhibition by antimicrobials of a wide variety of structure and function targeted at eradicating the organism. These bacteria show to accumulate multiple genes, each coding for resistance to a single drug, within a single cell. This accumulation occurs typically on resistance (R) plasmids. There is an increased expression of genes that code for multidrug efflux pumps, extruding a wide range of drugs. Such strains also show a decrease in outer membrane permeability through mutations in Porin genes.

These mechanisms are augmented in some Gram-negative species. Many times, persistence of pathogenic microorganisms occurs in an antibiotic treated patient. These organisms get into a physiologically resistant state, leading to a PERSISTER population of microbes that resist the action of almost all antibiotics.

In July 2010, a team in New Delhi reported a cluster of three cases of *Acinetobacter baumannii* bearing $bla_{\text{NDM-1}}$ that were found in the intensive care unit of a hospital in Chennai, India. The bacteria were fully resistant to all the aminoglycoside β-lactam and quinolone antibiotics ***Karthikeyan K, 2010.***

ESBL-EXTENDED SPECRTUM BETA LACTAMASES: The extensive use of third generation cephalosporins as a first line of treatment in hospital set up has led to the development of resistance towards cephalosporins. Until 1980 resistance to β-lactam antibiotics were known to be limited to organisms with inducible chromosomal β-lactamase genes (in this form the resistance in not transportable).

Consequently, it came as an unwelcome surprise when species of *Klebsiella* with plasmid mediated resistance was isolated in Germany in1983 (Vuye, 2012). β - lactamase are a group of enzymes act upon β - lactam

antibiotics (antibiotics have a four carbon atoms ring, called as β –lactam) and render them ineffective.

NDM-1 (New Delhi Metallo-beta-lactamase-1) was first detected in a *Klebsiella pneumoniae* isolate from a Swedish patient (of Indian origin in 2008). It was later detected in bacteria in India, Pakistan and the United States, is one of the newly developed resistant strain in this group (Kumarasamy *et al*., 2010).

The effectiveness of the cephalosporins relies on their ability to reach the penicillin binding protein (PBP) and bind to these PBP. Many times, to rule out infections caused by these β -lactamase producers, the antibiotic is co-administered with a β -lactamases inhibitor such as Clavunalic acid. The Clavulanic acid molecule is designed to overpower all β –lactamase by irreversibly binding to the enzyme molecule. They serve as an antagonist, so that the antibiotic is no longer affected by β -lactamase.

MATERIAL AND METHOD

Antibiotic Sensitivity testing by the Kirby Bauer s method, after 18 to 24 hours of incubation, each plate was examined. The diameters of the zones of complete inhibition were measured, including the diameter of the disc. Zones were measured to the nearest whole millimetre. When g blood-supplemented medium was used for testing *Streptococci*, the zone of growth inhibition was measured, not the zone of inhibition of haemolysis. The results were interpreted as per the sensitivity chart and the percentage of resistant strains were calculated.

ESBL identification double disc synergy assay was performed as a standard disc diffusion assay on Mueller Hinton Agar (MHA). Phenotypic confirmatory test using Cephalosporin/Clavulanate combination discs was done.

RESULT AND DISCUSSION

Out of 300 samples considered, Isolates from 186 (46.3%) were considered as probable or confirmed pathogens and were used as test organisms. In 215 samples (53.8%), no growth was observed.

Distribution of Gram character among pathogenic isolates showed 65 (34.9%) isolates as Gram positive and 121 (65.1%) isolates to be Gram negative.

Table 1: Antimicrobial sensitivity testing towards antibiotics

GRAM POSITIVE ISOLATES		**GRAM NEGATIVE ISOLATES**	
Antibiotic	% Resistant Isolates	Antibiotic	% Resistant Isolates
Penicillin	36.2%	Ampicillin	67.8%
Ampicillin	59.6%	Amocy Clav	64.5%
Amoyclav	17.0%	Amikacin	40.5%
Cefotaxime	44.7%	Azithromycin	68.6%
Erythromycin	44.7%	Cefuroxime	77.7%
Vancomycin	14.9%	Ceftriaxone	67.8%
Oxacillin	34.0%	Ceftazidime	66.9%
Co-trimaxole	14.9%	Cefta+Clavanalic acid	49.6%
		Ciprofloxacin	67.8%
		Gentamycin	29.8%
		Imipenem	13.2%
		Meropenem	17.4%
		Piper Tazobactam	24.0%
		Netilmicin	10.7%

ESBL Identification and confirmation showed **58** of the Gram-negative isolates to as β-lactamase enzyme producer.

DISCUSION

The situation of antibiotic resistance in developing countries like India is of particular concern because the use of antibiotics without medical guidance is largely facilitated. Self-medication is also noted in the major cities in India because of the high cost of medical consultations and dissatisfaction with medical practitioners. Authorities have been strongly encouraging physicians to reduce prescribing of antibiotics.

The resistance pattern of Gram-positive cocci to various antibiotics reported as, Cotrimoxazole 66.3%, Chloramphenicol 9.0%, Oxacillin 15.9% and Erythromycin 2.8%. The present study noted resistance towards Oxacillin in 11.9% and Erythromycin in 5% cases. There by presenting an increase in resistance towards Erythromycin.

Gagneja *et al*., (2011) reported a dramatic rise amongst Gram negative bacteria. Resistance towards Cephalosporin namely ceftriaxone **(**67.8%) Cefuroxime (77.7%) ceftazidime (66.9%) Cefta-Clavanalic acid (49.6%) were reported.

Treating these strains with antimicrobial chemotherapy poses a larger risk of therapeutic failure and is major cause of concern in controlling these infections. Ceftazidime aided with Clavulanic acid is known to restore the antimicrobial activity of β-lactam antibiotics against these lactamase-secreting bacteria. The present study shows 49.6% of bacterial isolates have emerged resistant to such combinations. Our study confirmed 58 isolates as ESBL by Double disc tests.

Rodriguez *et al.,* (2007) Studies on patients in the critical care, report the use of *Combination antibiotic therapy* to improve survival with community-acquired pneumonia**.** needs further consideration. Multidrug resistance (to a combination of drugs) was seen to be predominant in Gram negative species. In the present study, 36.55% isolates were resistant to the combination of Ampicillin-Azithromycin, and 15. 59% to the combination of Azithromycin-Gentamycin-Ciprofloxacin**.** This is of concern as these infection by these strains may spread among people in hospitals and long-term care facilities.

Antibiotic usage increases the out-of-pocket costs and does not significantly reduce recovery time of illness Rather this promotes the development of resistant strains within a population. ***Zsuzsanna et al., 2010.***

Beta-Lactamase threat in respiratory tract infections

ESBL positive strains of *Klebsiella pneumoniae, Escherichia coli, Acinetobacter baumannii* and *Pseudomonas aeruginosa* are increasingly found in hospitals. They are associated with respiratory tract infections. These strains become resistant to available, antibiotics and they can pass this resistance gene to other clinical strains. Therefore, quick detection of these strains in clinical laboratories is very important. These bacteria spread rapidly and have become a serious threat to human health worldwide.

Antibiotic resistance results in a great therapeutic and economic burden in the treatment of infectious diseases and threatens the success of antimicrobial chemotherapy.

Conflicts of interest: The authors stated that no conflicts of interest.

REFERENCES

Gagneja D, Goel N, Aggarwal R, Chaudhary U (2011) Changing trend of antimicrobial resistance among gram-negative bacilli isolated from lower respiratory tract of ICU patients: A 5-year study. *Indian Journal of Critical Care Medicine*, vol-5(3): pp 164–167.

Jayaraman (2009) Review Article-Antibiotic resistance: an overview of mechanisms and a paradigm shift. Current *Science*, vol-96(11).

Karthikeyan K, Thirunarayan MA, Krishnan (2010) Coexistence of bla-OXA-23 with blaNDM-1 and arm A in clinical isolates of *Acinetobacter baumannii* from India. *Journal of Antimicrobial Chemotherapy*, vol-65(10): pp 2253–2254.

Kumaraswamy KK, Toleman MA, Walsh TR and Woodford N (2010) Emergence of a new antibiotic resistance mechanism in India, Pakistan, and the UK: a molecular, biological, and epidemiological study. *Lancet Infectious Diseases*, vol- 10(9): pp597–602.

Rodríguez Ale Jandro, Mendia, Angel, Sirvent (2007) Combination antibiotic therapy improves survival in patients with community-acquired pneumonia and shock. *Critical Care Medicine*, vol-35(6): pp 1493-1498.

Vuye A, Verschraegen G, Claeys G (2012) Molecular description of plasmid-mediated AmpC β-lactamases among nosocomial isolates of *Escherichia coli* & *Klebsiella pneumoniae* from different hospitals in India. *Indian Journal of Medical Research*, vol-135: pp 114-119

Zsuzsanna Schelz, Judit Hohmann, Joseph Molnar, (2010) Recent advances in research of antimicrobial effects of essential oils and plant derived compounds on bacteria. Ethnomedicine: A Source of Complementary Therapeutics, Publisher-Research Signpost, ISBN-978-81-308-0390-6, pp179-201