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Status of antibiotic sensitivity pattern of clinically isolated bacteria collected from Rajshahi City, Bangladesh

Moni Krishno Mohanta*, Ananda Kumar Saha, Md. Fazlul Haque, Sabiha Akter Mahua and Md. Ariful Hasan

Genetics and Molecular Biology Laboratory, Department of Zoology, University of Rajshahi, Bangladesh

Abstract: Five clinically isolated pathogenic bacteria *viz., Escherichia coli*, *Shigella* sp., *Salmonella* sp., *Staphylococcus* sp. and *Klebsiella* sp., were collected from different pathological laboratory of renowned clinic of Rajshahi City, and were tested using disc diffusion method against seven commonly used antibiotics *viz*., ampicillin (10µg), azithromycine (15µg), cephradine (30µg), ciprofloxacin (5µg), doxycycline (30µg), gentamycin (10µg), and nitrofurantoin (30µg). Antimicrobial sensitivity test results revealed that *E. coli* was highly sensitive to azithromycine but exhibited very low sensitivity towards ciprofloxacin and nitrofurantoin. *Shigella* and *Salmonella* spp. showed higher sensitivity patterns with cephradine. *Staphylococcus* sp. however, was sensitive to gentamycin and nitrofurantoin, but the bacterium demonstrated resistance in the following order cephradine> ampicillin > doxycycline >azithromycine> ciprofloxacin. *Klebsiella* sp. was found to be highly sensitive to ciprofloxacin and gentamycin but did not show resistance towards any of the antibiotics tested.The present results suggest that *E. coli*, *Shigella* and *Salmonella* infections in the study area are relatively easy to handle using the available antibiotics from the local market.

Keywords: Pathogenic bacteria, antibiotics, disc diffusion methods

Introduction

Antibiotic resistance among bacteria is becoming more and more serious problem throughout the world. For example, multi-drug resistant nosocomial infectious diseases are among the leading causes of death and morbidity amongst hospitalized patients, accounting a major burden on patients and public health system of any country (Shehabi & Baadran, 1996) especially in developing world.

It is said that evolution of bacteria towards resistance to antimicrobial drugs is unavoidable because it represents a particular aspect of the general evolution of bacteria that is un-stoppable (Courvalin, 2005). According to El-Azizi (2005), antibiotic resistance emerges commonly when patients are treated with empiric antimicrobial drugs.

According to Montefiore *et al*.(1989), bacteria are the most common etiology in hospital patients. However, there is a phenomenal increase in antibiotic resistant bacterial pathogens which is one of the major problems facing medicine and science today. This calls for regular review of the antimicrobial sensitivity pattern among bacteria of clinically significance in our environment (Montefiore & Okubadejo, 1970). Eke & Rotini (1987) observed that there are many reasons for this alarming phenomenon and one of them is widespread and indiscriminate use of antibiotics which have been implicated in the development of serious problems of resistance to the older and less expensive antimicrobial agents, these include penicillin, ampicillin, cotrimoxazole, tetracycline and chloramphenicol. Studies in National Institute of Cardiovascular Disease (NICVD) hospital of Dhaka have also shown that the strains of *Pseudomonas* species are 100% resistant to many of the commonly available antibiotics (*viz.* penicillin, amoxicillin, vancomycin) and this, in many cases, has led to the use of newer and more expensive agents (Shahidullah *et al.,* 2012). Infection with antibiotic resistant bacteria will make the therapeutic options for treatment rather difficult or virtually impossible (Smyder *et al.,* 2000).The knowledge of prevailing susceptibility patterns is therefore vital to the selection and use of antimicrobial agents and to the development of appropriate prescribing policies (El-Astal, 2004). The objective of the present study is an attempt to know the current status of antibiotic sensitivity

*Corresponding author: mkmohanta_zool@yahoo.com

pattern of common bacterial isolates in Rajshahi City. This is to guide to antibiotic choice as well as in formulation of policy for the rational and effective use of antimicrobial agents.

Materials and Methods

Collection of samples: The clinically isolated samples of pathogenic bacteria *viz*., *E. coli, Shigella sp., Salmonella, Staphylococcus* and *Klebsiella* species were collected from different pathological laboratory of renowned clinics of Rajshahi City, Bangladesh. The collected samples were taken immediately to the Laboratory of Genetics and Molecular Biology, Department of Zoology, University of Rajshahi for further study.

Antibiotic sensitivity tests: The collected bacterial strains were grown overnight in nutrient broths that were placed in the shaker at 37°C temperature and 120 rpm for the antibiotic sensitivity test. A serial dilution technique was made for the test respective. Seven commercially available antibacterial drug discs were used*viz.* Cephradine (30µg), Ampicillin (10µg), Doxycycline (30µg), Azithromycin (15µg), Ciprofloxacin(30µg), Nitrofurantoin (30µg), and Gentamicin (30µg). One ml of test samples were poured separately in nutrient agar plate by using micropipette and then antibiotic discs were placed centrally on the respective plates and incubated overnight at 37°C. After overnight incubation the zone of inhibition on the plate was observed and measured with mm scale.

Results and Discussion

The antibiotic sensitivity tests against five pathogenic bacteria were done by the disc diffusion method using nutrient agar medium. After overnight incubation at 37°C the diameter of the inhibition zone, if any was measured. Antibiotic sensitivity tests are presented in Figure 1. From the figure it is evident that *Staphylococcus* sp. was resistant to cephradine, ampicillin, doxycycline, azithromycin and ciprofloxacin respectively, but sensitive to gentamycin and nitrofurantoin. Very low sensitivity of *E. coli* towards ciprofloxacin and nitrofurantoin was observed. Higher sensitivity pattern was observed in cephradine for *Shigella* and *Salmonella*, and in azithromycin for *E. coli* and *Klebsiella.* Thus, the finding of this study indicates that sensitivity pattern possibly susceptible enough to consider for the easy management of infection of *E. coli*, *Shigella*, *Klebsiella* and *Salmonella* in the area under study by using commonly available antibiotics while management of infection of *Staphylococcus* will be comparatively more challenging.

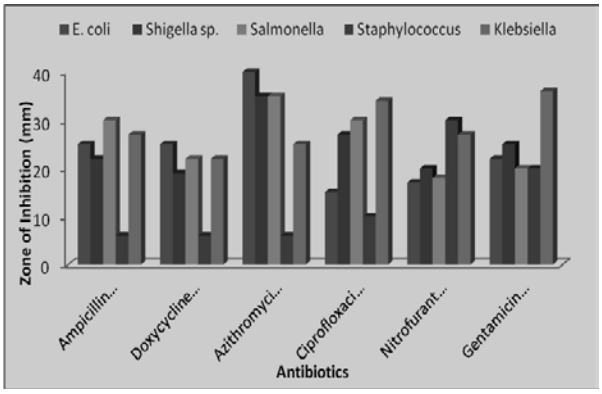

Fig.1. Comparison of different antibiotics against clinically isolated pathogenic bacteria.

(5-10mm) = Resistant to antibiotics (R), (10-15mm) = intermediate resistance (I), (15-20mm) = Sensitive to antibiotics (S).

Antibiotics are used to find out their effectiveness against pathogenic bacteria. The microbial pathogens, as well as their antibiotic sensitivity patterns may change from time to time and place to place (Mahmoud *et al*., 2012). The discovery of antibiotics revolutionized the management of infectious diseases. However, the over-use and misuse of antibiotics is leading to the emergence of resistance to these lifesaving drugs. Hospital antibiograms are commonly used to help guiding empiric antimicrobial treatment and are an important component of detecting and monitoring trends in antimicrobial resistance.

The present results show that different types of microorganisms vary in their response to different types of antibiotics. The *Staphylococcus* showed resistance to cephradine, ampicillin, doxycycline, azithromycine and ciprofloxacin respectively whereas susceptibility to gentamycin and nitrofurantoin. Cephradine for *Shigella*, *Salmonella*, and azithromycine for *E. coli* and *Klebsiella* have shown sensitivity pattern possibly susceptible enough to be drugs of choice for the management of infection caused by *E. coli*, *Shigella* and *Salmonella* in the area under study.

According to Hasan *et al.* (2007) at Abbottabad, amikacin and cefotaxime were the antibiotics still effective against gram negative bacteria. In another

study conducted by Saghir *et al.* (2009) at Lahore, imipenem (primaxin) was the most effective drug against gram negative bacterial strains.

Anguzu & Olila (2007) from Uganda reported that most of the gram negative bacteria isolated from septic postoperative wounds were resistance to ampicillin, chloramphenicol and amoxicillin, the results are comparable to the present study. Raghunath (2008) reported that coliforms have changed their susceptibility patterns extensively in India. According to the authors, B-lactam resistance is widespread among coliform bacteria due to vertical as well as horizontally acquired resistance factors. Researchers from Lahore have reported that resistance to B-lactam drugs amongst *E. coli* and *Klebsiella* is alarming with resistance ranging from 35.5% to 43.82% (Anwar *et al.,* 2007; Hafeez *et al.,* 2009). Methicillin resistant *Staphylococcus aureus* is a global phenomenon with a prevalence rate ranging from 2% in the Netherlands and Switzerland, to 70% in Japan and Hong-Kong (Diekema *et al.,* 2001; Saikia *et al.,* 2009). Okesola & Oni (2009) reported that *Pseudomonas aeruginosa* (53.3%) and *E. coli* (50%) showed high resistance to cefuroxime. A high resistance rate to streptomycin (86.2%) and chloramphenical (75.9%) were also recorded in *Staphylococcus aureus* by Okesola & Oni (2009). Pattanayak *et al.* (2013) observed on antibiotic sensitivity pattern of major six bacterial isolates *viz. E. coli, Klebsiella* sp., *Proteus* sp., *Pseudomonas* sp., *Staphylococcus* and *Enterococcus* species. The authors also noticed that the *E.coli* was most commonly sensitivity to polymixin B (100%), gatifloxacin (56.7%) and ceftriaxone (51.6%), *Pseudomonas aeruginosa* was sensitive to gatifloxacin (80%) and netilmicin (50%), *Staphylococcus aureus* was sensitive to vancomycin (100%) and linezolid (100%), *Klebsiella pneumonia* was sensitive to sparfloxacin (100%), levofloxacin (100%), piperacillin-tazobactam (100%), *E. fecalis* to amoxicillin (100%). The mentioned bacteria showed a very high rate of resistance to the cephalosporins namely cefuroxime, ceftazidime, cefixime and cefpodoxime respectively. There are also high rates of antimicrobial resistance reported among these bacterial pathogens, which are in keeping with the results of studies conducted by Goosens (2000) in Belgium, El-Astal (2004) in Palestine and Bakare*et al.* (1999) in Nigeria. Some strains of *Klebseilla, Pseudomonas and Proteus* species were resistant to ciprofloxacin, quinolone, with MICs values 8µg/ml (Okesola& Oni, 2009). Ogunsola *et al.* (1997) documented a 100% sensitivity of *Pseudomonas aeruginosa* to ciprofloxacin at Loges, Nigeria. Javeed *et al.* (2011) reported that *E.coli* showed a high level resistance to ampicillin (90.1%), amoxicillin/clavulanicacid (81.2%), and trimethoprim /sulfamethoxazole (81.1%). *Klebsiella*sp. also showed resistance to these three antibiotics *viz.* ampicillin (88.7%), amoxicillin/ clavulanic acid (78.9%), and trimethoprim /sulfamethoxazole (86.6%).

Nerurkar *et al.* (2012) reported that the isolates of most of species of *E.coli, Enterobacter, Klebsiella, Citrobacter, Staphylococcous, Pseudomonas Proteus and Streptococcus* sp. exhibited high rate of resistance to ampicillin, cotrimoxazole, cefozolin, norfloxacin and nitrofurantion. Resistance to antibiotics develops due to its frequent misuse. The pattern of resistance has also been reported by Gupta *et al.* (2009) and Tankhiwale *et al.* (2004). Such pattern has been reported from many other countries of the world (Uwaezuoke & Ogbulie, 2006).

The variations in antimicrobial susceptibility in Bangladesh depends mostly upon the easy availability of antimicrobial drugs over the counter. Ampicillin, cephradine, doxycycline, azithromycin, ciprofloxacin, gentamycin etc. are very commonly used over the counter drugs for infection by bacterial pathogen in our country. Frequent use and overdose of these drugs without any pathological screening resulted in evolution of resistance among a number of bacteria against these drugs.

Conclusion

In this study *Staphylococcus* species exhibited high level of resistance power against the test antibiotics. The study has further confirmed the intrinsic resistance of *Staphylococcus* sp. to examine antibiotics, a situation which favors their continued existence in hospital environment. This high resistance rate may be connected with widespread, indiscriminate and inappropriate use of antibiotics which is rampant in this environment. This calls for the education of both medical and paramedical staff on the rational use of antibiotics. Furthermore, the community at large must also be enlightened through regular health education programs on the dangers inherent in self-medication.

It is pertinent to say here that continued surveillance of changes in resistance patterns of this pathogen to antimicrobial agents is of utmost importance if effective management of infectious diseases is to be ensured.

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