ORIGINAL ARTICLE

Antibiotic Susceptibility Patterns of Gram Negative Organisms Isolated from Cases of Nosocomial Infections in a Paediatric ICU

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ABSTRACT

Objective: To determine the antibiotic susceptibility patterns of Gram negative organisms isolated from cases of nosocomial infections in a paediatric ICU.
Setting: This study was carried out from November 2009 to September 2010 on 200 samples taken from patients in pediatric ICU of a tertiary care hospital, in the Department of Microbiology, Basic Medical Sciences Institute, Jinnah Postgraduate Medical Centre.
Method: This study was carried out from November 2009 to September 2010 on 200 samples taken from patients in pediatric ICU of a tertiary care hospital who were clinically suspected of having nosocomial infection and were processed for isolation of the microbes and their antibiotic susceptibility in the microbiology department Basic Medical Sciences Institute (BMSI) JPMC.
Results: Out of 200 cases having nosocomial infection, 138 samples showed bacterial growth. Among 138 isolates, 104(72.7%) were Gram -ve for which antibiotic susceptibility was determined.
Conclusion: Drug resistance to conventional antibiotics is a common problem and it grows readily
Keywords: Nosocomial, antibiotic susceptibility, PICU

INTRODUCTION

Nosocomial infection is defined as an infection which develops 48 hours after being discharged that was not incubating at the time of admission in hospital[1]. The Centre for Disease Control and Prevention (CDC) defines ICU associated infections as those that occur after 48 hours of ICU admission or within 48 hours after transfer from an ICU[2]. The incidence of resistant infection varies in different PICUs[3]. According to surveillance data of the prevention network of United States, the incidence of resistant infections in pediatric ICUs was 13.9 per 1000 patient days[4]. Nosocomial infections represent an important cause of morbidity and mortality in this population, the overall mortality being estimated at 11%[5]. Bacteremia, Pneumonia and UTI are most common infections[6]. Bacteremia is the second most common resistant infection in a PICU at 28%-43%[7].

Antimicrobial resistance is a significant and increasing problem worldwide. Resistance of Gram negative bacilli is of particular importance especially in some geographical areas. This resistance is mainly due to emergence of multidrug resistant (MDR) *Pseudomonas aeruginosa* and ESBL producing Enterobactearіaceae or stably depressed AmpC beta lactamases[8].

The drug resistant Gram negative organisms for the most part threaten only hospitalized patients whose immune systems are weak. They can survive on surfaces in the hospital and enter the body through wounds, catheters and ventilators[9]. Different regional and global ICU surveillance studies have documented resistance profile. The organisms considered most troublesome and problematic for patients in the ICU include, the Enterobacteriaceae family, Non fermenters (*Pseudomonas aeruginosa* and *Acinetobacter species*), Oxacillin resistant *Staphylococcus aureus* and Vancomycin resistant enterococci[10].

Close monitoring of the bacteria and their resistance patterns can lead to selection of an appropriate antimicrobial agent[11]. The antimicrobial surveillance programmes that provide extensive information on the pattern, development and prevalence of bacterial resistance in different geographical regions are vital in the fight against bacterial resistance[12].

MATERIALS AND METHODS

This prospective study was conducted in the Dept. of Microbiology, BMSI, JPMC in suspected cases of resistant infection. In this study 200 samples were collected from the patients admitted in Pediatric ICU of tertiary care hospital of Karachi from December 2009 to September 2010. These samples were collected from clinically suspected cases of nosocomial infections. Antimicrobial susceptibility for different species of organisms was performed by in vitro disc diffusion method according to CLSI[13].

Since 1966, when described as the first standardized method, the disc diffusion test of Kirby Bauer has been widely used in clinical laboratories. The method was modified later. Briefly a McFarland

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0.5 standardized suspension of bacteria is swabbed over the surface of an agar plate and paper disc containing single concentration of each antimicrobial agent is placed onto the inoculated surface within 15 minutes. After overnight incubation, the diameters of the zones produced by antimicrobial inhibition of bacterial growth were measured and the size of zone is inversely proportional to the minimum inhibitory concentration of the organism and the isolate is interpreted as susceptible, intermediate or resistant to a particular drug according to present criteria. McFarland turbidity 0.5 standard containing 99.4ml of 1% v/v solution of sulphuric acid and 0.6ml of 1% w/v solution of barium chloride is used for comparison of test suspension. The present criteria have been specified by the National Committee for Clinical Laboratory Standard[13] for disc diffusion testing recommended by World Health organization[14].

RESULT

Total samples included in our study were two hundred. Among these 200 samples, 143(71.5%) showed the growth of the organisms and no growth was observed in 57(28.5%) samples and no growth was observed in 57(28.5%) samples.

Among the total 143 isolates bacterial growth was observed in 138 cases while in 5 cases fungal growth was observed. In 138 bacterial isolates, Gram negative was 104(72.7%) and Gram positive were 39(23.3%). Table shows antimicrobial sensitivity pattern of Gram negative organisms isolated from PICU. Almost all the organisms are 100% resistant to ampicillin. Most of the organisms including *Acinetobacter baumannii* are highly sensitive to impenem and cefepime with *Pseudomonas aeruginosa* showing 66.7% sensitivity to imipenem which is least of all other organisms. Most of the organisms showed good sensitivity to Piperacillin/ tazobactum also. Most organisms are moderately or intermediately sensitive to 3rd generation cephalosporins, Ceftazidime showing good activity against *E.coli* (70.4%) and *Pseudomonas aeruginosa* (76.2%). Aminoglycosides along with ofloxacin are also moderately active but amikacin shows 81.5% activity for *E.coli*, 82.8% for *Enterobacter* and 85.7% for *Pseudomonas aeruginosa*. 1st and 2nd generation cephalosporins show least activity.

Table Susceptibility pattern of gram negative organisms isolated during the study

Antibiotics	***E.coli* (27)**		***Enterobacter* (35)**		***Klebsiella spps* (14)**	
	S	**R**	**S**	**R**	**S**	**R**
Imipenem (10μg)	92.6%	7.4%	94.3%	5.7%	85.7%	14.3%
Piperacillin/Tazobactum (100/100 μg)	77.7%	22.3%	62.8%	37.2%	64.3%	35.7%
Cefotaxime (30μg)	66.7%	33.3%	51.4%	48.6%	57.1%	42.9%
Ceftazidime (30μg)	70.4%	29.6%	54.3%	45.7%	57.1%	42.9%
Cefipime (30μg)	92.6%	7.4%	91.4%	8.6%	85.7%	14.3%
Gentamicin (10μg)	51.8%	48.2%	68.6%	31.4%	57.1%	42.9%
Tobramicin (10μg)	55.5%	44.5%	65.7%	34.3%	64.3%	35.7%
Amikacin (30μg)	81.5%	18.5%	82.8%	7.2%	71.4%	28.6%
Augmentin (30μg)	66.7%	33.3%	68.6%	31.4%	71.4%	28.6%
Cefoperazone (75μg)	70.4%	29.6%	54.3%	45.7%	57.1%	42.9%
Ofloxacin (5μg)	59.2%	40.8%	51.4%	48.5%	64.3%	35.7%
Cefoxitin (30μg)	81.5%	18.5%	60.0%	40.0%	57.1%	42.9%
Cefalexin (30μg)	55.5%	44.5%	57.1%	42.9%	57.1%	42.9%
Cefaclor (30μg)	59.3%	40.7%	62.8%	37.2%	50.0%	50.0%
Cefadrexil (30μg)	55.5%	44.5%	51.4%	48.6%	50.0%	50.0%
Ampicillin (10μg)	7.4%	92.6%	5.7%	94.3%	7.1%	92.9%
Imipenem (10μg)	66.7%	33.3%	80.0%	20.0%	100.0%	0.0%
Piperacillin/Tazobactum (100/100 μg)	85.7%	14.3%	80.0%	20.0%	100.0%	0.0%
Cefotaxime (30μg)	61.9%	38.1%	60.0%	40.0%	50.0%	50.0%
Ceftazidime (30μg)	76.2%	13.8%	60.0%	40.0%	50.0%	50.0%
Cefipime (30μg)	85.7%	14.3%	80.0%	20.0%	100.0%	0.0%
Gentamicin (10μg)	71.4%	28.6%	60.0%	40.0%	50.0%	50%
Tobramicin (10μg)	76.1%	23.9%	60.0%	40.0%	100.0%	0.0%
Amikacin (30μg)	85.7%	14.3%	60.0%	40.0%	50.0%	50.0%
Augmentin (30μg)	71.4%	28.6%	40.0%	60.0%	50.0%	50.0%
Cefoperazone (75μg)	66.7%	33.3%	60.0%	40.0%	50.0%	50.0%
Ofloxacin (5μg)	71.4%	28.6%	60.0%	40.0%	100.0%	0.0%
Cefoxitin (30μg)	66.7%	33.3%	40.0%	60.0%	50.0%	50.0%
Cefalexin (30μg)	80.9%	19.1%	40.0%	60.0%	50.0%	50.0%
Cefaclor (30μg)	52.4%	47.6%	40.0%	60.0%	0.0%	100.0%
Cefadrexil (30μg)	52.3%	47.7%	40.0%	60.0%	0.0%	100.0%
Ampicillin (10μg)	0.0%	100.0%	0.0%	100.0%	0.0%	100.0%

Fig. 1: Sensitivity pattern of E.coli

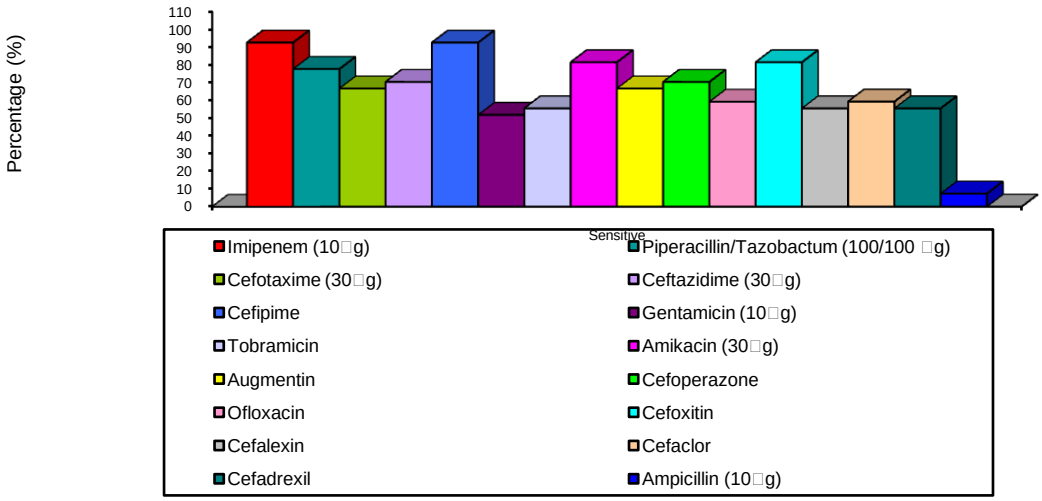


Figure 2: Sensitivity pattern of *enterobacter*

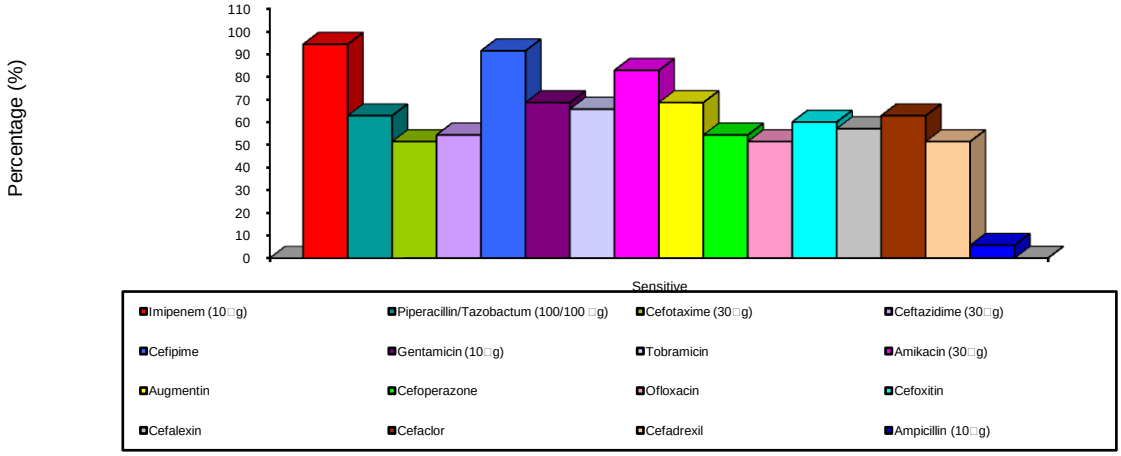


Figure 3: Sensitivity pattern of *klebsiella species*

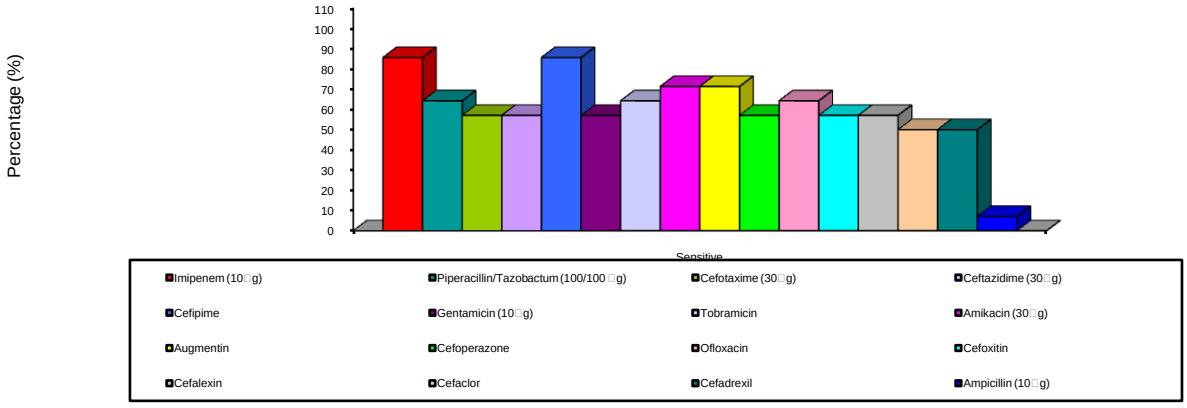


Figure 4: Sensitivity pattern of *pseudomonas aeruginosa*

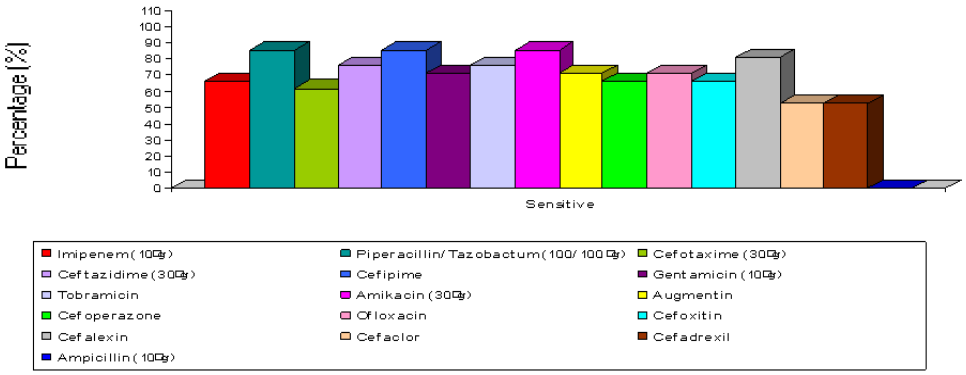


Figure 5: Sensitivity pattern of *proteus species*

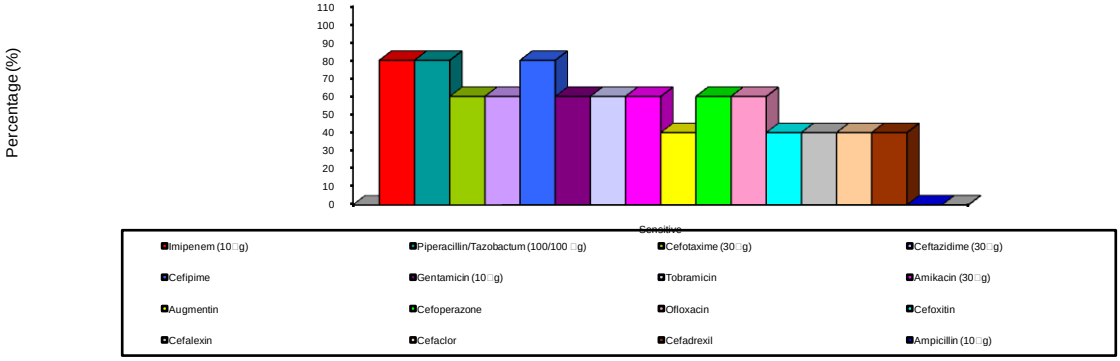
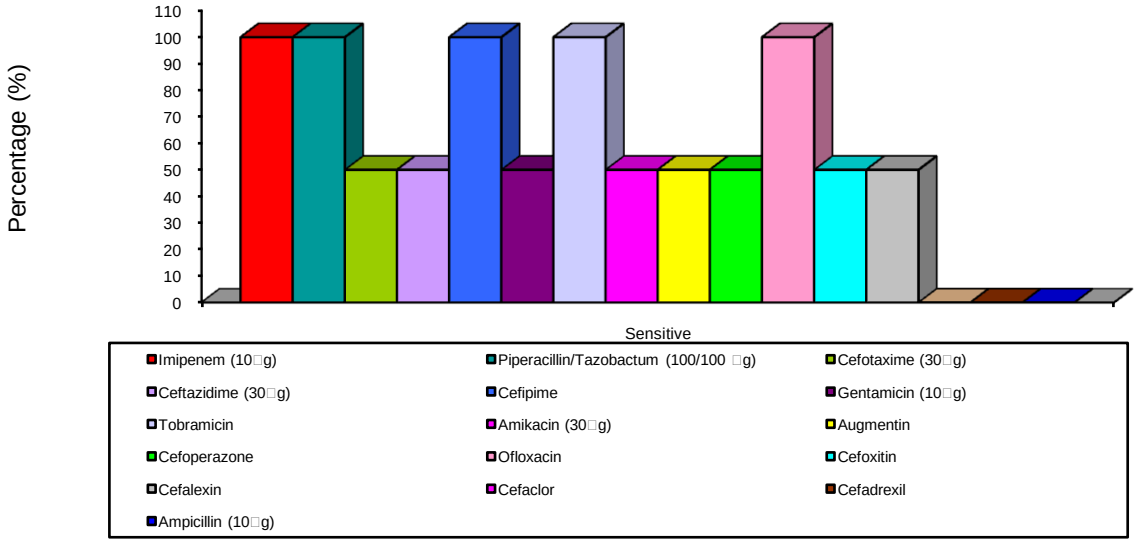


Figure 6: Sensitivity pattern of *a.baumannii*



DISCUSSION

A knowledge of the antibiotic susceptibility of the organisms isolated in the ICU helps to formulate an antibiotic policy for the ICU. This also avoids un-necessary use of broad-spectrum empirical antibiotics and prevents emergence of drug resistant bacterial strains. Also, reviewing antibiotic susceptibility pattern of common organisms isolated from the ICU and formulating appropriate antibiotic protocols for ICU and hospital help in managing nosocomial infections. This would prevent emergence of drug resistant strains[15]. This study is an attempt to know the antibiotic sensitivity pattern of the common isolates causing nosocomial infections in paediatric ICU.

Overall sensitivity of Gram negative organisms was highest for imipenem, sensitivity ranges from 80% for *Proteus spp*. to 92.6% and 94.3% for *E.coli* and *Enterobacter* species respectively. The sensitivity, of different Gram negative organisms isolated, was also high for piperacillin tazobactam with 85.7% for P. aeroginosa, while Klebsiella and *Enterobacter* being more resistant (i.e., 35.7% and 37.2% resistance respectively) as compared to others.

The sensitivity of Gram negative organisms was also high for cefepime (4th generation cephalosporin) with range of 85.7% for *Klebsiella* and *Pseudomonas aeruginosa* to 91.4% for *Enterobacter* and 92.6% for *E. coli*. Most of the organisms showed moderate or intermediate type of sensitivity to cefotoxime, ceftazidime, and cefoperazone.

Like 3rd generation cephalosporins, organisms were moderately or intermediately sensitive to aminoglycosides also, with amikacin showing higher activity against *E.coli* (81.5%), *Enterobacter* (82.8%), Klebsiella (71.4%) and *P. aeruginosa* (85.7%).

In present study quinolones (e.g., ofloxacin) also showed moderate sensitivity ranging from lowest of 51.3% for *Enterobacter*, to 71.4% for *P.aeruginosa*. Cefoxitin showed highest activity against *E.coli* and *P.aeruginosa* i.e., 81.5% and 66.7% respectively. All the Gram negative organisms were least sensitive to 1st and 2nd generation cephalosporins.

In a study by Ahmad et al, *P. aeruginosa* were sensitive to amikacin (89.79%), tobramycin (75.81%), norfloxacin (68.48%), piperacillin (68.2%), ceftazidime (58.8%) and few isolates were resistant to gentamicin, cerbenicillin tobramycin, ceftazidime and augmentin[16]

In present study, sensitivity of *P. aeruginosa* was nearly similar to above study with negligible difference i.e. amikacin (85.7%), tobramycin (76.1%), ofloxacin (71.4%), piperacillin-tazobactum (85.7%) and gentamicin and augmentin (71.4% each). The ceftazidime sensitivity was comparatively more i.e., 76.2%.

The sensitivity of *P. aeruginosa* to imipenem was 76% in study by Naeem et al[17] while it is 66.7% in our study. All isolates were resistant to augmentin, ceftazidime, chloramphenicol, gentamicin, tobramycin and cotrimoxazole in study by Naeem et al[17]. Antibiotic resistant Gram negative organisms are a significant risk to severely ill children and in many instances are imported into the unit or rapidly acquired from environment reservoirs as observed by Toltiz et al[18].

Klebsiella pneumoniae was 3rd most common organism (13%) causing pediatric nosocomial infections (Naeem et al[17]. In our study *Klebsiella pneumoniae* is 4th most common organism causing nosocomial infections, large number of it (42.9%) is resistant to 3rd generation cephalosporins which agrees with Becerra et al[19] who also observed big resistance i.e., (85% resistance) and partially coincides with a study by Gorgan et al[20], who in his study isolated two cases of *Klebsiella pneumoniae* from a PICU resistant to 3rd generation cephalosporins. Higher incidence of strains of *Klebsiella pneumoniae* resistant to 3rd generation cephalosporins have been observed by Kolar et al.[21]. Resistance to amikacin and cefepime is also observed in the study by Naeem et al. [17] which is in contrast to our study in which amikacin and cefipime show 71.4% and 85.7% sensitivity respectively against *Klebsiella pneumoniae.* Usually prescribed above antimicrobials failed to clear this organism and most effective antimicrobials against such organisms were meropenem (100%), imipenem (100%) (Naeem et al.[17], present study demonstrated 85.7% sensitivity of imipenem against *Klebsiella pneumonia*e.

The *E.coli* is one of the most abundantly existing oganisms in the pediatric NIs (Naeem et al[17]. *E. coli* being the 2nd most common organism causing nosocomial infections in present study is in accordance with the above study.

Our study showed high sensitivity of *E.coli* to imipienem (92.6%) and cefepime and good sensitivity to amikacin (81.5%) and cefoxitin (81.5%) which partially coincides with a study by Patzer et al[22]. showing sensitivity of *E.coli* as; imipenem 100%, cefepime 100%, while aminoglycosides (gentamicin 58.8% and tobramycin 55.1%), ceftazidime 76.5%, cefotaxime 69%. Present study also corroborates with the above study showing sensitivity of *E.coli* as: gentamicin 51.5%, tobramycin 55.5%, ceftazidime 70.4% and cefotaxime 66.5%. In the study of Patzer et al[22], ciprofloxacin showed 98.9% activity against *E.coli*. Ofloxacin in present study showed 59.2% activity which is in accordance to study by Bayram and Balci[23] showing 60.5% activity against *E.coli*. It is

recommended that except in life threatening infections and cystic fibrosis, quinolones should not be used in pediatric patients even if good in vitro sensitivity is observed.

Data are limited on the prevalence, patterns of resistance and risk factors associated with resistant organisms including *E.coli* in children as observed by Naeem et al[17].In recent years *A. baumanii* species have emerged as important pathogens of ICUs, most of them being rersistant to several antibiotics especially third generation cephalosporins and aztreonam, ticracillin and gentamicin (i.e. 94.6%, 93.2% and 85.1% respectively) as observed by Bayram and Balci[23],. Multiple drug resistant (MDR) bacteria continue to be of concern world wide[4].

The susceptibility of *A. baumanii* in a study by Patzer et al[22] was imipenem (98.5%), piperacillin/ tazobactum (70.1%), cefotaxime (52.5%), ceftazidime (73.3%), cefepime (72.3%), gentamicin (81.7%), tobramycin (92.9%) and ciprofloxacin (84.7%). Whereas the susceptibility of *A.baumanii* in the present study was 100% for imipenem and cefepime which coincides partially with the above study, high resistance (100%) for cephalexin, cephadrexil and ampicillin, 50% sensitivity for piperacillin/tazobactum and third generation cephalosporins, aminoglycosides and ofloxacin. However this may not reflect true picture as it represents sensitivity of only two *A. baumanii* isolates.

The challenges for the future are to limit the emergence of antibiotic resistant organisms especially in critically ill children with optimal and cost effective care, along with minimizing infection in the PICU.

CONCLUSION

Resistance to antibiotics poses a serious and growing problem, because such resistant bacteria are becoming more difficult to treat. The empirical and the indiscriminate use of antibiotics should be avoided in order to curtail the emergence and the spread of drug resistance among nosocomial pathogens.

Reduction of nosocomial infections and antimicrobial resistance is both a challenge and goal of all ICU's around the world. Strict infection control measures like universal precautions and stringent adherence to hand washing practices, formulation of antibiotic policy, surveillance activities, might be required for the same. The antimicrobials like imipenem, meropenem, amikacin, vancomycin (especially in MRSA or BRSA), and some of the 3rd generation cephalosporins were found most effective and hence can be useful provided that these are used in appropriate dosage and regime

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