

## ANTIBIOTIC SUSCEPTIBILITY PATTERN OF BACTERIAL ISOLATES FROM PATIENTS ADMITTED TO A TERTIARY CARE HOSPITAL IN LAHORE

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### ABSTRACT

*Objective:* To determine the antimicrobial sensitivity pattern of bacterial isolates from patients admitted in a tertiary care hospital of Lahore.

*Methodology:* The study was carried out in Department of Pathology, Postgraduate Medical Institute, Lahore from January 2010 to June 2010. The samples from the hospital were sent to microbiology laboratory for bacteriological examination. They were cultured onto Blood and MacConkey agar plates; organisms were identified by their colonial morphology, Gram Staining and appropriate biochemical tests using standard recommended protocol. Antimicrobial susceptibility pattern of the bacterial isolates recovered from different clinical specimens against penicillins, cephalosporins, fluoroquinolones, carbapenems, aminoglycosides and trimethoprim sulphamethoxazole was determined using modified Kirby Bauer method.

*Results:* Among the 925 different clinical samples, 379 organisms were isolated. *Escherichia coli* and *Klebsiella* species were the most prevalent isolates followed by *Pseudomonas* and *Staphylococcus* spp. High degree of resistance was observed among gram negative organisms to all groups of antibiotics. Resistance to amikacin ranged from 12 – 18% among different species of Gram negative isolates whereas the range of carbapenem resistance was 1.4 – 9.5%. The percentage of oxacillin resistance among staphylococcal isolates was 33.1%, but all were sensitive to vancomycin.

*Conclusion:* High frequency of resistance observed in the present study indicates that antibiotic resistance among nosocomial isolates is a serious problem. There is a continuous need of surveillance of sensitivity patterns of antimicrobial agents in our set up to know about the trend of this problem.

*Key Words:* Bacteria; Drug sensitivity; Hospital; Gram negative; Staphylococcus.

### INTRODUCTION

Antibiotic resistance among bacteria is becoming more and more serious problem throughout the world. It is said that evolution of bacteria towards resistance to antimicrobial drugs, including multi-drug resistance, is unavoidable because it represents a particular aspect of the general evolution of bacteria that is un-stoppable.<sup>1</sup> Antibiotic resistance emerges commonly when patients are treated with empiric antimicrobial drugs. To overcome these difficulties and to improve the outcome of serious infections in our institutions, monitoring of resistance patterns in the hospital is needed.<sup>2</sup>

A number of studies have been carried out in the west to monitor antimicrobial resistance at national level. The academic and educational value of these studies is particularly useful for microbiologists and infectious disease clinicians. The data collected from these studies are useful in improving antimicrobial use in those communities.<sup>3-6</sup> In our setting, establishment of surveillance programs to

monitor the true extent of resistance at the local, regional and national levels is urgently needed. This will help in monitoring emerging trends in resistance at the local level to support clinical decision making, infection – control interventions, and antimicrobial – resistance containment strategies.<sup>7</sup>

The present study is an attempt to know the current status of antibiotic sensitivity pattern of common bacterial isolates in a tertiary care hospital of Lahore.

### MATERIALS AND METHODS

The samples received from various departments of a tertiary care hospital during the period of January 2010 to June 2010 were included in the study. These samples were mainly urine, blood, pus, endotracheal secretions, high vaginal swabs, CSF, fluids and sputum sent to the microbiology section of Pathology Department, Postgraduate Medical Institute, Lahore.

The samples were processed for culture and

sensitivity testing in the department. Specimens inoculated onto Blood and MacConkey agar plates were incubated aerobically at 37°C overnight. The cultured plates were examined after 24 hours and organisms identified by their colonial morphology, Gram staining and appropriate biochemical tests using standard techniques.<sup>9</sup> The results were interpreted according to the guidelines of the Clinical and Laboratory Standards Institute (CLSI).<sup>10</sup>

Antibiotic susceptibility of the isolates was determined by Modified Kirby – Bauer disc diffusion method, according to CLSI recommendations.<sup>10</sup> Antibiotic discs (Oxoid) used for *Staphylococcus* spp were oxacillin (5 µg), vancomycin (30 µg), ampicillin (10 µg), amoxicillin/ clavulanic acid (20 / 10 µg), erythromycin (15 ug), cefaclor (30 µg) ciprofloxacin (5 ug), clindamycin (2µg), linezolid (30 µg), doxycycline (30 µg), trimethoprim / sulphamethoxazole (1.25 / 23.75 µg), cephhradine (30 µg), teicoplanin (30 µg) and fusidic acid (10 µg).

For gram negative isolates ceftazidime (30 µg), ceftriaxone (30 ug), amoxicillin / clavulanic acid (20 / 10 µg), cefuroxime (30 µg), ciprofloxacin (5 µg), imipenem (10 µg), trimethoprim / sulphamethoxazole (1.25/23.75 µg), aztreonam (30 µg), cefepime (30 µg) and amikacin (30 µg) were tested. Nitrofurantoin (30 µg) and norfloxacin (10 µg) were used additionally for urinary isolates. The zones of inhibition were measured and the organisms identified as sensitive or resistant based on standard criteria.<sup>10</sup> Control strains were used for checking the quality of discs and reagents. The results are expressed in percentages.

**RESULTS**

In a total of 925 different clinical samples which were processed, 379 (40.9%) organisms were isolated. These included 118 (31.1%) Gram positive and 261 (68.9%) Gram negative isolates. *Staphylococcus aureus* 88 (23.2%) was the most prevalent gram positive pathogen whereas gram negative isolates included *Escherichia coli* 121 (31.9%), *Klebsiella* 62 (16.4%) and *Pseudomonas* species 59 (15.6%) predominantly. Other species isolated were β haemolytic streptococci, *Acinetobacter* spp, *Salmonella typhi*, *Citrobacter* spp, *Enterobacter* spp and *Proteus* spp. (19%) (Table 1).

Amongst the gram negatives, *Escherichia coli* and *Klebsiella* species were the most prevalent isolates from urine (33.9% and 37.1% respectively) followed by *Pseudomonas* spp (18.6%), *Escherichia coli* (28.9%), *Klebsiella* spp (27.4%), *Staphylococcal* spp (21.6%) and *Pseudomonas* spp (13.6%) were the predominant isolates from pus samples (Table 1).

The present study showed a very high percentage of resistance among Gram negative organisms to all generations of cephalosporin antibiotics as well as β lactam/β lactamase inhibitors (Table 1). Resistance to Amikacin was 12.8% in *Klebsiella* spp, 15.7% in *E.coli* and 18.7% in *Pseudomonas* spp. Similarly Carbapenem showed 3.7% resistance among *E.coli*, 4.1% among *Pseudomonas* spp and 9.5% among *Klebsiella* spp in urinary isolates. Whereas, among Gram positive organisms 33.1% *Staph. aureus* were MRSA, but all were sensitive to vancomycin (Table 2). Sensitivity pattern of all 30 isolated strains of coagulase negative staphylococci showed that

Table 1: *Organisms isolated from various clinical samples (n = 379).*

Specimen type	<i>Staphylococcus aureus</i> (# = 88) (23.2%)	Coagulase negative <i>Staphylococci</i> (# = 30) (7.9%)	<i>Eschericia coli</i> (# = 121) (31.9%)	<i>Klebsiella</i> spp. (# =62) (16.4%)	<i>Pseudomonas</i> spp. (# = 59) (15.6%)	Others* (# =19) (5.0%)
Urine	29	13	41	23	11	4
Blood	14	9	15	3	9	7
Pus	19	–	35	17	8	3
HVS	7	–	11	3	3	1
CSF	2	2	5	4	8	–
Endotracheal secretions	3	5	9	3	7	2
Fluids	4	1	2	2	2	–
Sputum	10	–	3	7	11	2

\*β Haemolytic Streptococci, *Acinetobacter* spp, *Salmonella typhi*, *Citrobacter* spp, *Enterobacter* spp and *Proteus* spp

there was no isolate resistant to vancomycin, ciprofloxacin and linezolid. Doxycycline, fusidic acid and erythromycin were found to be effective anti-microbials with efficacy of 90.5%, 93%, and 92.1% respectively.

## DISCUSSION

The microbial pathogens, as well as their antibiotic sensitivity patterns may change from time to time and place to place. The discovery of antibiotics revolutionised the management of infectious diseases. However, the overuse and misuse of antibiotics is leading to the emergence of resistance to these life – saving drugs. Hospital antibiograms are commonly used to help guide empiric antimicrobial treatment and are an important component of detecting and monitoring trends in antimicrobial resistance.<sup>11</sup>

In the present study the most common micro-organisms isolated were *Escherichia coli* (121), *Staphylococcus aureus* (88), *Klebsiella spp* (62) and *Pseudomonas spp* (59). *Escherichia coli* was commonest gram negative organisms isolated from urine specimens. It showed a high level resistance to ampicillin (90.1%), amoxicillin / clavulanic acid (80.2%), and trimethoprim / sulphamethoxazole (81.1%). *Klebsiella* also showed marked resistance to these three antibiotics – ampicillin (88.7%), amoxicillin / clavulanic acid (78.9%), and trimethoprim / sulphamethoxazole (86.6%). This data shows that more than 85% of these isolates were resistant to these drugs. Similarly *Pseudomonas* species were also markedly resistant to ampicillin (98.2%) and amoxicillin / clavulanic acid (90.2%) as shown in Table 2. This pattern is comparable to other studies carried out at home and abroad.<sup>12-15</sup>

Our study showed a high ceftazidime resistance in *Klebsiella spp* and *Escherichia coli* (50.2% and 45.7% respectively); ceftriaxone resistance amongst these two isolates was also found to be high (47.6% and 53.3%). *Pseudomonas spp* showed a very high resistance to ceftriaxone (77.7%) as compared to *Escherichia coli* and *Klebsiella spp* (Table 2). However, resistance to ceftazidime was lower (32.4%) than that among *Escherichia coli* and *Klebsiella* species. Similar findings regarding drug resistance patterns of *Klebsiella*, *Escherichia coli*, *Pseudomonas* and non-fermenting gram negative bacteria have been observed by other researchers.<sup>13,16,17</sup>

*Klebsiella* also showed high resistance to

Table 2: Percentage of antibiotic resistance among *Escherichia coli*, *Klebsiella* and *Pseudomonas* isolates.

Antibiotics	Resistance (%) among Gram negative bacilli isolates (n = 242)		
	<i>E.coli</i> (no = 121)	<i>Klebsiella</i> (no = 62)	<i>Pseudomonas</i> (no = 59)
Ampicillin	90.1	88.7	98.2
Amoxicillin / Clavulanic acid	80.2	78.9	90.2
Cefepime	33.1	33.4	36.5
Trimethoprim / sulphamethoxazole	81.1	86.6	88.5
Amikacin	15.7	12.8	18.7
Imipenem	3.7	9.5	4.1
Ciprofloxacin	30.5	64.2	30.3
Cefuroxime	71.2	73.8	92.4
Ceftazidime	50.2	45.7	32.4
Ceftriaxone	53.3	47.6	77.7
Aztreonam	26.1	30.9	24.1

Table 3: Percentage of antibiotic resistance among *Staphylococcus aureus* isolates from different clinical samples.

Antibiotics	Resistance (%) among <i>Staphylococcus aureus</i> (n = 88)	Resistance (%) among Coagulase negative <i>Staphylococci</i> (n = 30)
Ampicillin	99.6	32.7
Oxacillin	33.1	12.4
Amoxicillin / Clavulanic acid	33.1	7.8
Cephadrine	98.6	23.3
Cefaclor	68.7	26.7
Vancomycin	0	0
Teicoplanin	0	0
Erythromycin	72.7	7.9
Clindamycin	20.3	11.9
Doxycycline	66.9	9.5
Ciprofloxacin	19.7	0
Linezolid	1.2	0
Trimethoprim / sulphamethoxazole	80.7	65.7
Fusidic acid	18.4	7.0

ciprofloxacin; 64.2% as compared to that of *Escherichia coli* (30.5%) and *Pseudomonas spp* (30.3%). Antibiotics which retained their usefulness and showed less resistance for these three gram negative isolates in our study, were amikacin (12.8%, 15.7%, 18.8%) and carbapenem (3.7%, 4.1%, 9.5%) against *Klebsiella*, *Escherichia coli* and *Pseudomonas* species respectively. According to a study conducted by Azra et al in 2007 at Abbottabad,<sup>18</sup> amikacin and ceftaxime were the antibiotics still effective against gram – negative isolates. In another study conducted at Lahore in the year 2009 by Saghir et al,<sup>19</sup> imipenem was the most effective drug against gram negative bacterial strains.

In 2007 Anguzu and Olila<sup>20</sup> from Uganda reported that most of the gram – negative bacteria isolated in their study were resistant to ampicillin, chloramphenicol and amoxicillin, results which are comparable to our study. Raghunath<sup>21</sup> reported from India in 2008 that coliforms have changed their susceptibility patterns extensively. According to them,  $\beta$ -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  $\beta$ -lactam drugs amongst *Escherichia coli* and *Klebsiella* is alarming with resistance ranging from 35.5% to 43.82%.<sup>3,22</sup>

Methicillin resistant *Staphylococcus aureus* (MRSA) is a global phenomenon with a prevalence rate ranging from 2% in the Netherlands and Switzerland, to 70% in Japan and Hong Kong<sup>24,25</sup>. In the present study, 33.1% of *Staphylococcus aureus* isolates were MRSA. However all were sensitive to vancomycin. A comparable prevalence rate of 31% to 38.56% have been reported from different studies conducted in Pakistan and India.<sup>6,18,26</sup>

The results of the present study highlighted the alarming resistance to almost all the drugs – with the exception of a few – included in this study. Antibiotic resistance is becoming a big problem for the public health which threatens the lives of hospitalized individuals as well as those with chronic conditions. This also adds considerably to health care cost. Therefore, it is an important issue which has to be addressed by the policy makers in order to formulate strict antibiotic prescription policy in our country. Moreover, this study indicates that majority of the gram negative isolates were more sensitive to imipenem and amikacin as compared to the other antibiotics tested and therefore these may be considered the drugs of choice for the treatment of nosocomial infections at our tertiary care hospitals.

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