Prevalence and antibiogram of *Pseudomonas aeruginosa* isolated from clinical specimens in a Teaching Hospital, Kathmandu.

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ABSTRACT

Pseudomonas aeruginosa is a leading cause of nosocomial infections. Increased resistance in this organism continues to pose a significant threat to patient care because of limited therapeutic options. Knowledge of the prevalence of *P. aeruginosa* in various infections and their antimicrobial susceptibility pattern is of utmost importance for selection of appropriate therapy. Objective of the present study was to determine the prevalence and resistance pattern of *P. aeruginosa* isolated from various clinical specimens to commonly used antibiotics in Nepal Medical College and Teaching Hospital (NMCTH), a teaching hospital in Kathmandu, Nepal. *P. aeruginosa* isolated from various clinical specimens over the period of one year (April 14, 2013 – April 13, 2014) were included. Direct microscopy with Gram stain, isolation, identification and antimicrobial susceptibility testing were performed using standard microbiological techniques. A total of 102 isolates of *P. aeruginosa* were evaluated. The prevalence rate of was 5.1% out of which 75 (73.5%) were from inpatients and 27 (26.5%) were from outpatients departments. Urine and sputum yielded highest number of isolates 37 (36.3% each) followed by pus and devices 10 (9.8%) each. Highest percentage of the organism 36 (35.3%) was isolated from the patients who were of more than 60 years of age. Nineteen (18.6%) of the organism was seen to be multi-drug resistant. The organism showed maximum resistance to piperacillin (57.1%) followed by ciprofloxacin (36.7%), ofloxacin (28.8%) and gentamycin (30.9%). Only 6.5% of the isolates were resistant to imipenem. The antibiotic susceptibility pattern of bacterial pathogens like *P. aeruginosa* in the hospital settings should be continuously monitored and the results readily made available to clinicians so as to maximize the possibility of administering an effective therapeutic agent whenever needed.

Keywords: *Pseudomonas aeruginosa,*Infection, antibiogram

INTRODUCTION

Pseudomonas aeruginosa is an invasive, gram negative opportunistic pathogen that causes a wide range of severe infections that include bacteremia, pneumonia, meningitis, urinary tract and wound infections[1].It is a leading cause of nosocomial infection and is associated with a high mortality rate. One reason for high mortality is its notable resistance to many currently available antibiotics[2]. Resistance to multiple drugs is usually the result of combination of different mechanisms in a single isolate[3]. There are variety of mechanisms involved in the resistance of *P. aeruginosa*, among them over expression of efflux pump[4], acquisition of Extended-Spectrum β-Lactamases (ESBLs) and Metallo-β-Lactamases (MBLs) [5]; target site or outer membrane modification[3]are predominant. Production of multiple β-lactamases by *P. aeruginosa* has led tremendous therapeutic consequences and posed clinical challenges[6]. ESBLs mediate resistance to extended spectrum cephalosporins such as cefotaxime, ceftriaxone and ceftazidime[7]. The carbapenems and β-lactam and β-lactamase inhibitor combination such as piperacillin plus tazobactam are the drugs active against ESBL

producing *P. aeruginosa*. However, resistance to these drugs has also been increasing worldwide[8,9,10,]. The production of MBLs, increased expression of efflux pump, reduced level of drug accumulation are the main factors involved in carbapenem resistance to *P. aeruginosa*[8,11,12]. The present study aimed to determine the prevalence of *P. aeruginosa* from clinical specimens and their antibiogram from Nepal Medical College and Teaching Hospital (NMCTH), Kathmandu, Nepal.

MATERIALS AND METHODS

This is a prospective cross-sectional study, conducted in the Microbiology unit of the Central Labortory Service, NMCTH, Kathmandu, Nepal over a period of one year from April 14, 2013 – April 13, 2014. A total of 102 consecutive isolates of*P. aeruginosa*from various clinical specimens (pus, blood, sputum, urine, high vaginal swab, catheter tip and body fluids) of patients visiting Out patients departments and Inpatients submitted for bacterial culture were included in the study. All isolates were non duplicate. Identification of the organism was done by standard microbiological techniques[13]. The antimicrobial sensitivity testing was performed on Mueller-Hinton

agar plates with commercially available discs (Hi-media, Mumbai) by Kirby Bauer disc diffusion technique. Diameter of the zone of inhibition for each antibiotic was measured and interpreted according to CLSI guidelines[14]. The antibiotic discs used were piperacillin (100µg), piperacillin/tazobactum (100/10µg), ceftazidime (30µg), ciprofloxacin (5µg), ofloxacin (5µg), tobramycin (10µg), amikacin (30µg) and imipenem (10µg). Isolates were regarded as multidrug resistant (MDR) if they were resistant to two or more than two classes of antibiotics. The data obtained were recorded in Microsoft excel sheet and analyzed in terms of percentage.

RESULTS

The total number of specimens received for culture in the clinical microbiology laboratory, NMCTH from April 14, 2013 – April 13, 2014 was 10,110 out of which culture positive was seen in 2037 (20.15%). One hundred and two (5.1%) of the isolated organisms were identified as *P. aeruginosa* out of which 66 were from male patients and 36 were from female patients. Majority of the isolates (73.5%) were recovered from patients attending inpatient departments than from outpatient departments (26.5%). Frequency of the organism isolated from various clinical specimens is shown in Table no 1.

Table 1: Frequency of P. aeruginosa from various clinical specimens

Clinical Specimen	Number	Percentage
Sputum	37	36.27
Urine	37	36.27
Pus	10	9.8
Devices	10	9.8
Blood	6	5.89
Stone	2	1.97

Maximum number of *P. aeruginosa* was recovered from patients who were more than 60 years of age (35.3%) followed by those who were more than 40 upto 60 years of age (29.4%) as shown in Table no 2.

Table 2: Distribution of P. aeruginosa in various age groups

Age groups	Number	Percentage
Upto 1 yr	2	1.9
>1-20 yrs	12	11.8
>20-40 yrs	22	21.6
>40-60 yrs	30	29.4
>60 yrs	36	35.3

A total of 19 (18.6%) of the organism was found to be multidrug resistant (MDR). Resistance pattern of the organism to various antibiotics is shown in Table no 3.

Table 3: Resistance pattern of P. aeruginosa to various antibiotics

Antibiotic	Percentage
Piperacillin	57.14
Ciprofloxacin	36.67
Ofloxacin	28.79
Piperacillin Tazobactum	19.72
Tobramycin	15.70
Ceftazidime	13.85
Amikacin	13.11
Imipenem	6.58

DISCUSSION

The present study revealed the prevalence rate of 5.1% which is almost comparable to the prevalence rate shown in studies done in Afghanistan (6.67%) and India (9.28%).[15,16] A similar study done in another teaching hospital of Kathmandu in 2012 showed 17.05% of the isolates as *P. aeruginosa*.[17] Higher prevalence rate (32-40%) was documented by other studies[18,19]. Duration of stay is directly proportional as infection was much higher (73.5%) in inpatients than in outpatients.

The most common source of the isolate in our study were urine and sputum (36.27% each). Only 9% of the isolates were recovered from pus in our study. Viren *et al* also reported maximum percentage of *P aeruginosa* from urine, sputum and pus (26.7% each).[20] Other studies reported maximum isolation from pus followed by urine and sputum[17,21]. Distribution of specimens may vary with each hospital as each hospital facility has different environment. Genderwise, male patients (66) constituted a larger group in our study. Other studies from India have also shown similar findings.[16,22] When factors such as age of the patients were considered, we found the occurrence of the isolates to be higher in the age group of patients who were more than 60 years of age (35.3%). Similar observation was made by Somporn *et al* (29.5%), Srinivas *et al* (37%) and Chander *et al* (43.92%). *P. aeruginosa* infection was more common in the old age group patients.[2,16,17]This could be explained as due to decreased immunity, prolonged hospitalization and other associated co-morbidities in these age groups.

The emergence of resistance in *P. aeruginosa* is an increasing clinical problem which not only limits future therapeutic choices but is also associated with increased rates of mortality, morbidity and higher costs. The present study showed 18.6% of the isolate as MDR which is comparable to the study done by Mishra*et al* who reported 20.5% of the *P. aeruginosa* to be MDR.[23] In the present study, resistance to Ureidopenicillin like piperacillin was seen in 57.14% of the isolates which closely matches the findings of

Angadi *et al* (54.4%).[21]Various studies in India have reported higher rate of resistance to piperacillin (66.67%, 75%, 85%) [16,24,25]. Similarly, our study showed lower rate of resistance to piperacillin/tazobactum (19.7%) whereas Chaudhary *et al* showed only 3.5% resistance but Srinivas *et al* showed 36.99% of resistance to piperacillin/tazobactum.[26,16]

Among the quinolones and aminoglycosides, our study showed 28.9% resistance to ofloxacine, 36.67% to ciprofloxacin, 13.11% to amikacin and 15.79% to tobramycin. Similar studies from India had reported higher resistance to quinolones and aminoglycosides ranging from 40%-70% to quinolones,57-90% to amikacinand 60.19% to tobramycin.[21,27,16,18] Ceftazidime is a reserved drug for *P. aeruginosa* infections because of its unique antipseudomonal activity. Only 13.85% of the isolates exhibited resistance against ceftazidime in our study which is very low in comparison to the rate of resistance (69.64-89%) shown by various other studies.[16,18,27]

Imipenem is a carbapenem antibiotic which is highly active against *P. aeruginosa*. In recent years, there has been an alarming increase in the *P. aeruginosa* resistant to imipenem. In the SENTRY Antimicrobial Surveillance program (SASP), 10-30% of *P. aeruginosa* strains from various countries have been found to be resistant to imipenem[28]. Fortunately, our study showed only 6.58% of resistance to imipenem which was slightly lower to the resistance rate revealed by study done in a different university of Kathmandu (9.1% and 15.4%) [29,23]. Higher rate of resistance was seen in study done by Taneja *et al* (36.4%) and Chaudhary *et al* (66.5%).[30,26]

The prevalence and sensitivity of *P. aeruginosa* often varies between communities, hospitals in the same community and among different patient populations in the same hospital. It is therefore, important to institute a system of surveillance in a hospital so that clinicians have access to recent data on prevalence and antimicrobial resistance which will help them in making clinical judgement in therapy.Increase in antibacterial resistance in *P aeruginosa* is a cause of concern. So, continuous monitoring of bacterial resistance trends should be done and therapy should be based on antibacterial susceptibility testing.

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