ORIGINAL ARTICLE

ANTIBIOTIC RESISTANCE PATTERN IN PSEUDOMONAS AERUGINOSA SPECIES ISOLATED AT A TERTIARY CARE HOSPITAL, AHMADABAD

Rajat Rakesh M[1], Ninama Govind L[2], Mistry Kalpesh[3], Parmar Rosy[3], Patel Kanu[4], Vegad MM[5]

[1]Tutor, [2]Associate Professor, [3]Assistant Professor, Microbiology Department, GMERS, Gotri Medical College, Vadodara [4]Associate Professor, [5]Professor & Head, Microbiology Department, B. J. Medical College, Ahmadabad

Correspondence:
Dr. Rakesh M Rajat
C/o Dr. Kalpesh Mistry,
B/102,Manorath flates, B/h Mother`s School
Nr.Jalaram Nagar bus stop, Gotri, Vadodara 390021
Email: rakeshrjt@gmail.com

ABSTRACT

Introduction: Pseudomonas aeruginosa (Ps.aeruginosa) is one of the important bacterial pathogens isolated from various samples. Despite advances in medical and surgical care and introduction of wide variety of antimicrobial agents against having anti-pseudomonal activities, life threatening infection caused by Ps. aeruginosa continues to cause complications in hospital acquired infections. Several different epidemiological studies indicate that antibiotic resistance is increasing in clinical isolates.
Material and Method: This study was conducted during April 2009 to april 2010. During this period total of 630 samples were tested, in which 321 samples showed growth of bacteria. Out of 321 samples, 100 clinical isolates of Pseudomonas aeruginosa were isolated. The samples were selected on the basis of their growth on routine MacConkey medium which showed lactose Non-fermenting pale colonies which were oxidase test positive and on Nutrient agar pigmented and non-pigmented colonies with oxidase positive. Antimicrobial susceptibility of all the isolates was performed by the disc-diffusion (Modified-Kirby Baur disc diffusion method) according to CLSIs guidelines.
Result: In present study, maximum isolates of Ps. aeruginosa isolated from various samples are resistant to tobramycin (68%) followed by gentamycin (63%), piperacillin (50%), ciprofloxacin (49%) and ceftazidime (43%).
Conclusion: To prevent the spread of the resistant bacteria, it is critically important to have strict antibiotic policies while surveillance programmes for multidrug resistant organisms and infection control procedures need to be implemented.

Key words: Pseudomonas aeruginosa, Resistance, Antimicrobial agents**,** Antibiotic sensitivity

INTRODUCTION

Antibiotic when first introduced was considered as a magic bullet. A single injection of penicillin could eradicate a life threatening infection. Unfortunately with time due to malpractices of natural causes, most of the cheaper antibiotics have lost their efficacy and more and more expensive and complicated antibiotics were introduced and marketed to combat simple infection[2]. The microbial pathogens, as well as, their antibiotic sensitivity pattern, may change from time to time and place to place. Therefore knowledge of current drug resistance pattern of the common pathogenic bacteria in a particular region is useful in clinical practice.

Pseudomonas aeruginosa (Ps.aeruginosa) is one of the important bacterial pathogens isolated from various samples. Despite advances in medical and surgical care and introduction of wide variety of antimicrobial agents against having anti-pseudomonal activities, life threatening infection caused by Ps. aeruginosa continues to cause complications in hospital acquired infections. Ps.aeruginosa is increasingly recognized as an emerging opportunistic pathogen of clinical relevance that causes infections in hospitalized patient particularly in burn patients, orthopaedic related infections, respiratory diseases, immunosuppressed and catheterized patients.

Several different epidemiological studies indicate that antibiotic resistance is increasing in clinical isolates[3]. Being gram-negative bacteria, most pseudomonas spp. are naturally resistant to penicillin and majority of related beta-lactum antibiotics, but a number are sensitive to piperacillin, imipenem, tobramycin or ciprofloxacin. Nowadays more and more resistance of

Ps.aeruginosa are encountered in routine clinical practice, a serious problem, increase morbidity and mortality and also cost of treatment.

MATERIAL AND METHOD[4]

This study was conducted at the Department of Microbiology in a tertiary care hospital, Ahmedabad. It is tertiary care center, referral and teaching hospital. This study was conducted during April 2009 to April 2010.

The present study comprises 100 Pseudomonas positive samples: swab, urine, sputum, pus, pleural fluid, BAL, ascitic fluid and blood samples submitted for microbiological diagnosis to the Microbiology Department. All these samples were obtained from various wards of hospital. The clinical data was obtained from the respective units and wards of the patients.

Sample proccessing

The samples were selected on the basis of their growth on routine MacConkey medium which showed lactose Non-fermenting pale colonies which were oxidase test positive and on Nutrient agar pigmented and non-pigmented colonies with oxidase positive.

Confirmation of pseudomonas spp

After obtaining the pure strains, the strains were subjected to biochemical identification tests to identify Pseudomonas spp. For this purpose samples were inoculated in Triple Sugar Iron media (TSI), Citrate media, Peptone water, Urease media and kept in an incubator for 18 hrs at 37°C. Next day the results were noted on TSI, Citrate media and Urease media. Part of growth on Peptone water was subjected to Indole test with Kovac's Reagent and part for motility test by 'Hanging drop' method. A strain of Pseudomonas in the TSI medium showed alkaline slant, no reaction in butt. It showed negative reaction for indole test, negative urease test and positive citrate test. Glucose is utilised oxidatively, forming acid only

Antimicrobial disc: susceptibility test

Application of antibiotic discs to the inoculated agar plates:

Antimicrobial susceptibility of all the isolates was performed by the disc-diffusion (Modified-Kirby Baur disc diffusion method) according to CLSIs guidelines. The following antibiotics were tested by disc diffusion method, Ceftazidime, Piperacillin, Piperacillin-tazobactam, Cefipime tazobactum, Imepenam, Gentamicin , Ciprofloxacillin , Levofloxacillin, Cefoperazone, Tobramycin, Polymyxin B, Aztreonam, Netilmycin

RESULTS AND ANALYSIS

Total 630 samples were tested, out of 630 samples, 321 samples were showing growth on culture and out of 321 samples, 100 Ps.aeruginosa were isolated and tested for antibiotic sensitivity

Table 1: Sex wise distribution of cases

Sex	Total no	Percentage (%)
Male	61	61
Female	39	39
Total	100	100

Table-2 Shows sex wise distribution of samples. Ps.aeruginosa was isolated from 61(61%) males and 39(39%) females.

Table 2: Isolation of Pseudomonas aeruginosa from different clinical samples

Name of sample	No. of Sample in which Pseudomonas aeruginosa Isolated
Pus	3
Sputum	12
Stool	1
Swab	68
Urine	16
Total	100

Table 3: Antibiotic resistance of Pseudomonas aeruginosa isolated from different clinical samples

Antibiotic	Resistance (%)
Ceftazidime (AZ)	43
Piperacillin (MP)	50
Piperacillin tazobactam (PT)	04
Cefepime tazobactam (TT)	03
Cefoperazone (CM)	33
Ciprofloxacin (CG)	49
Tobramycin (TF)	68
Levofloxacin (GF)	25
Polymyxin B (AK)	45
Gentamycin (GM)	63
Aztreonam (AC)	39
Netilmycin (NT)	36
Imepenam (IM)	14

DISCUSSION

Pseudomonas aeruginosa emerged as an important pathogen and responsible for the nosocomial infections.It is one of the important causes of morbidity among hospital patients.

The pre-eminent of pseudomonas aeruginosa in hospital infections is due to its resistance to common antibiotics and antiseptics, and its ability to establish itself widely in hospitals. Being an extremely adaptable organism, it can survive and multiply even with minimum nutrients, if moisture is available.

As pseudomonas aeruginosa causes serious infections, and is one of the leading causes of hospital acquired

infections, several studies were carried out to detect antibiotic sensitivity pattern for the various drugs available. Such study helps clinicians for the better management of patients.

So the present study was conducted to determine the antibiotic sensitivity pattern of Pseudomonas aeruginosa isolated from various clinical samples. In present study the isolation rate of Pseudomonas aeruginosa was comparable with other studies.

In the present study sex wise prevalence of clinical isolates shows that infections caused by Pseudomonas aeruginosa are more common in males (61%) compared to females (39%). This is comparable with study of Javia et al.[5] , Jamshaid Ali Khan et al.[6] and Rashid et al.[7].

In present study the age wise prevalence of clinical isolates shows that most of patients 29 (29%) were aged between 31-45 years. This is comparable with study of Rashid et al.

The sex wise and age wise distribution of patients diagnosed with infections followed the natural epidemiological pattern.

In present study, the maximum clinical isolates of Ps. aeruginosa were isolated from pus/swab (71%), followed by urine (16%). These results are in line with studies of Jamshaid A K et al. 32 and other studies.[7-12]

In present study the highest percentage (48%) of Pseudomonas aeruginosa infections were observed in the surgical ward, followed by paediatric ward (23%) and medical ward (17%). Prevalence of infection was higher in surgical ward as maximum isolates were isolated from pus/swab samples.

In present study it is evident from table 7 that there is distinct difference in the sensivitity pattern of isolates of pseudomonas aeruginosa from specimen to specimen [9].

This study shows that the clinical isolates of Pseudomonas aeruginosa are becoming resistant to commonly used antibiotics and gaining more and more resistance to newer antibiotics. The antimicrobial agents are losing their efficacy because of the spread of resistant organisms due to indiscriminate use of antibiotics, lack of awareness, patient non compliance and unhygienic condition.

It is the need of the time that antibiotic policies should be formulated and implemented to resist and overcome this emerging problem. Every effort should be made to prevent spread of resistant organisms.

SUMMARY AND CONCLUSION

This study was conducted during April 2009 to april 2010. During this period total of 630 samples were tested, in which 321 samples showed growth of bacteria. Out of 321 samples, 100 clinical isolates of Pseudomonas aeruginosa were isolated.

Out of 100 clinical isolates of Ps. aeruginosa, maximum isolates (71%) are isolated from pus/swab followed by 16% from urine and 12% from sputum, 3% from other samples.

Out of 100, 61% are males and 39% are females. Most of patients were aged between 31-45 years. Most of samples were collected from surgical wards, followed by paediatric ward, medical ward, orthopaedic and gynaecology and obstetrics ward and ICU. Maximum resistant isolates of Pseudomonas aeruginosa were isolated from pus/swab samples.

In present study, maximum isolates of Ps. aeruginosa isolated from various samples are resistant to tobramycin (68%) followed by gentamycin (63%), piperacillin (50%), ciprofloxacin (49%) and ceftazidime (43%).

It is evident from the study that now a days Ps. aeruginosa is becoming less sensitive to cephalosporins, aminoglycosides and B-lactamase inhibitors.

To prevent the spread of the resistant bacteria, it is critically important to have strict antibiotic policies while surveillance programmes for multidrug resistant organisms and infection control procedures need to be implemented. In the meantime, it is desirable that the antibiotic susceptibility pattern of bacterial pathogens like Ps. aeruginosa in specialized clinical units to be continuously monitored and the results readily made available to clinicians so as to minimize the resistance.

The solution can be planned by continuous efforts of microbiologist,clinician, pharmacist and community to promote greater understanding of this problem. Frequent hand washing to prevent spread of organism should be encouraged. Better surgical and medical care should be provided to patients during hospital stay.

REFERENCES

1. Irvani A. Efficacy of lomefloxacin as compared to norfloxacin in the treatment of uncomplicated urinary tract infections in adults. Am J Med 1992;92(suppl.4A), 75-81.
2. Betty A. F. Daniel F. S. and Alice S. W. Bailey & Scott's Diagnostic Microbiology, Twelfth edition, 2007, Pseudomonas, Burkholderia and similar organisms, 340-350, mosby Elsevier, St. Louis, Missouri.
3. Koneman, Koneman's color Atlas and textbook of diagnostic Microbiology, Sixth Edition, 2006, The Nonfermantative Gram-Negative Bacilli, 303-391, Lippincott Williams & Wilkins.
4. Javiya VA, ghatak SB, Patel KR, Patel JA. Antibiotic susceptibility patterns of Pseudomonas aeruginosa at a tertiary care hospital in Gujarat, India. Indian J Pharmacol 2008;40:230-4.
5. Jamshaid A K, Zafar I, Saeed U R, K. Farzana, Abbas K. Prevalence and resistance patterns of Pseudomonas aeruginosa against various antibiotics. Pak. J. Pharm. Sci. Vol21, No. 3, July 2008:311-315.
6. Rashid A, chowdhury A, Sufi HZ R, Shahin A B, Naima M. infections by Pseudomonas and antibiotic resistance pattern of the isolates from Dhaka Medical college Hospital. Bangladesh J Med Microbiol 2007;01(02):48-51

7. Shenoy S, baliga S, Saldanha DR, Prashanth HV. Antibiotic sensitivity patterns of Pseudomonas aeruginosa strains isolated from various clinical specimens. Indian J Med Sci. 2002 Sep;56(9)427-30.

8. 34. Arshi syed, Manzoor thakur, Syed Shafiq, Mr. assad Ullah sheikh, In vitro sensitivity patterns of Pseudomonas aeruginosa strains isolated from patients at skims-role of antimicrobials in the emergence of multiple resistant strains. JK-Practitioner 2007;

9. Murase M, Miyamoto H, Handa T, Sahaki S and Takenchi . Activity of antipseudomonal agent against clinical isolates of Psedomnas aeruginosa. Jpn. J. Antibiot,1995., 48(10):1581-1589.

10. Stark RP, Maki Dg (1984) Bacteriuria in the catheterized patient. New Engl. Med. 311:560-564.

11. Henwood CJ, Livermore DM, James D, Warner M and the Pseudomonas study group (2001). Antimicrobial susceptibility of Pseudomonas aeruginosa:results of a UK survey and evaluation of the British society for Antimicrobial chemotherapy disc susceptibility test. J. Antimicrobial chemother. 47:789-799.