

Prevalence of Pseudomonas Aeruginosa in Clinical Isolates

Sumit Khatri, Sonia Bharty, Brahma Prakash, Kshitiz Chourasiya

School of Excellence and Pulmonary Medicine, Netaji Subhash Chandra Bose Medical College, Jabalpur-482003

ABSTRACT

P. aeruginosa is a leading cause of nosocomial infections ranking second among the gram-negative pathogens. Hence this study was conducted to enhance the knowledge of this particular organism. A total of 100 isolates of *P. aeruginosa* isolated from various clinical specimens like urine, pus, blood, body fluids, sputum, collected from patients, irrespective of age and sex, were identified by standard microbiological procedures. Total hundred culture positive samples were taken and found that *P. aeruginosa* was predominantly present in urine sample of male aged between 21-30 years.

KEY WORDS: *P. aeruginosa*- *Pseudomonas aeruginosa*, NNISS- National Nosocomial infection surveillance system, ICU- Intensive care unit.

INTRODUCTION:

Pseudomonas aeruginosa is the epitome of an opportunistic pathogen of human. Infection due to *P. aeruginosa* is seldom encountered in healthy adults, now the organism has become increasingly recognized as the etiological agent in a variety of serious infections in hospitalized patients with impaired immune defenses. It causes infections particularly in burns patients where the skin host defenses are destroyed, orthopaedic infections, respiratory diseases, immune suppressed and catheterized patients. It may be the cause of the chronic debilitating pulmonary infections, which is one of the major cause of death in patients with cystic fibrosis. Generally, it contributes substantially to wound related morbidity and mortality worldwide^[1]. *P. aeruginosa* is a leading cause of nosocomial infections, ranking second among the gram-negative pathogens reported to the National Nosocomial Infection Surveillance System (NNISS). Hence this study was conducted to

observe the growth of *P. aeruginosa* in various samples, according to parameters related to hospitalised patients, to enhance the knowledge about this particular organism.

MATERIALS AND METHODS:

This study was conducted in the Department of Microbiology, Pt. B.D. Sharma Post Graduate Institute of Medical Sciences, Rohtak over a period of one year.

A total of 100 isolates of *P. aeruginosa* isolated from various clinical specimens like urine, pus, blood, body fluids, sputum, etc collected from patients, irrespective of age and sex, were identified by standard microbiological procedures^[2].

Collection of specimen: Urine: clean catch midstream urine samples were collected; Pus: aspirated samples of pus or swabs were collected; Blood: blood samples were collected by aseptic venipuncture; Body fluids: body fluids were aspirated under aseptic conditions and Sputum: expectorated sputum samples were collected.

Processing and culture of organism: Microscopy and culture of all the above mentioned samples were done. Cultures were performed on blood agar and MacConkey agar. Inoculated media was examined for growth after overnight incubation at 37°C. Blood samples were cultured in glucose broth and subcultured on blood agar and MacConkey agar after incubation at 37°C for 24 hours, 48 hours, 72 hours and on 7th day. The evaluation of colony morphology on the plating media was done and the subsequent

Corresponding Author:

Dr Sumit Khatri

Senior Resident,

School of Excellence and Pulmonary Medicine, Netaji Subhash Chandra Bose Medical College, Jabalpur - 482003.

Phone No.: 7697034954

E-mail: sk36636@gmail.com



identification procedures was carried on the isolated bacteria, using standard procedures^[2,3].

Blood agar and MacConkey agar were inoculated within half an hour of collection with the specimen. Inoculation of samples on culture medium was done using an ordinary reusable inoculating loop.

IDENTIFICATION AND SCREENING OF P. AERUGINOSA:

Gram staining

The smear was prepared on clean, grease free slide, air dried and heat fixed. Crystal violet was poured on the slide, allowed to remain for one minute and rinsed with tap water. Gram's iodine was then poured on the slide, retained for one minute and then rinsed with tap water. The smear was decolorised with acetone and rinsed immediately with tap water. The slide was counter stained with carbol fuschin for 30 seconds and rinsed with tap water and air dried. The slide was finally examined under an oil immersion lens for presence of gram negative bacilli.

Detection of motility using hanging drop preparation:

A part of colony was passed into peptone water and incubated at 37°C for two hours. After two hours, hanging drop was prepared by taking a loopful of growth from peptone water. It was kept on a cover slip was inverted on a slide with a plasticine ring over it. First, the edge of the drop was focused under 10X of microscope and then it was examined under 40X. Gliding type of motility is seen in maximum number of isolates.

Biochemical Reactions:

The various biochemical reactions used were oxidase test, catalase test, motility, growth at 42°C, oxidative/fermentative medium (Glucose, Maltose, Lactose), nitrate reduction test, MR/VP, mannitol motility medium, triple sugar iron agar, indole production, urea hydrolysis, citrate utilization.

RESULTS:

A total of 100 isolates of P. aeruginosa isolated from various clinical specimens like urine, pus, blood, body fluids, sputum, etc collected from patients, irrespective of age and gender, were included in the present study. P. aeruginosa isolates were identified on the basis of gram staining, motility and biochemical

reactions. Out of 48218 clinical samples received in the laboratory during the study period, 12854 (26.66%) showed bacterial growth, while rest 35364 samples (73.34%) were either culture sterile, or showed the growth of bacterial contaminants or fungal isolates. The overall isolation rate of P. aeruginosa was 12.21%.

Gender wise distribution of patients with P. aeruginosa infection among different age groups. The male to female ratio among patients with P. aeruginosa was 1.27:1. Majority of the patients from which P. aeruginosa was isolated belonged to age group 21-30 years (40%), followed by age group 31-40 years (17.0%) and by age group 41-50 years (16.0%) (Table 1).

The distribution of P. aeruginosa isolates among a total of 100 clinical isolates. The maximum number of P. aeruginosa isolates were from urine samples (49%), followed by pus samples (20%), blood samples (19%), sputum (11%) and body fluids (1%) (Table 2).

DISCUSSION:

The purpose of this study was to enhance the knowledge about P. aeruginosa according to various patient related parameters.

Nosocomial infections caused by P. aeruginosa are frequently life threatening and often challenging to treat. In the current study, the rate of isolation of P. aeruginosa isolates from culture positive samples was 12.21% which was lower than studies by other authors who have reported an isolation rate of 19% to 31.71% from all culture positive samples^[4,5,6]. This discordance may be due to implementation of better infection control measures in our hospital like barrier precautions, frequent hand washing by hospital staff, removal of catheters at frequent intervals, regular environmental sampling from ICUs, operation theatres and wards. However, Sherertz et al have reported an isolation rate of 12.5% which was similar to current study^[7]. Gales et al and Khan et al have reported an isolation rate of 9.46% and 6.67% respectively from culture positive samples which was low as compared to this study^[1,8]. This may be due to different prevalence rates of P. aeruginosa isolates in different geographical areas. In addition, prevalence rate may also vary from hospital to hospital^[8].

The present study showed maximum rate of isolation of P. aeruginosa isolates from urine samples (49.0%), followed by pus samples (20.0%), blood samples (19.0%), sputum (11.0%) and body

TABLE 1: Age and gender wise distribution of patients from which 100 isolates of *P. aeruginosa* were taken.

Age groups (years)	Male		Female		Total	
	n	%	n	%	n	%
0-10	2	3.57	5	11.36	7	7.0
11-20	9	16.07	5	11.36	14	14.0
21-30	17	30.36	23	52.27	40	40.0
31-40	11	19.64	6	13.64	17	17.0
41-50	12	21.43	4	9.09	16	16.0
51-60	3	5.36	1	2.27	4	4.0
>60	2	3.57	0	0.0	2	2.0
Total	56	56.0	44	44.0	100	100.0

TABLE 2: Distribution of *P. aeruginosa* isolates among various clinical samples.

Name of sample	Number of <i>P. aeruginosa</i> isolates(n)	Percentage (%) of <i>P. aeruginosa</i> isolates
Urine	49	49.0
Pus	20	20.0
Blood	19	19.0
Sputum	11	11.0
Body fluids	1	1.0
Total	100	100.0

fluids(1.0%). The results of current study were in accordance with Pitout et al who have also reported maximum rate of isolation of *P. aeruginosa* isolates from urine samples (43%), followed by pus samples (21%) and respiratory tract samples(20%) and blood samples(7%)^[9]. However ,Khan et al reported maximum rate of isolation of *P. aeruginosa* isolates from pus samples(57.64%) followed by urine (24.2%) samples. The difference in rates of isolation may be due to difference in type of samples received in different laboratories^[1].

The male to female ratio among the patients with *P. aeruginosa* infections in the present study was 1.27:1, which was in accordance with the study done by Sherertz et al who also reported the male to female ratio in patients with *P. aeruginosa* infection to be 1.3:1.^[7] Khan et al reported the male to female ratio among patients with *P. aeruginosa* infection to be 1.6:1. Higher incidence of infection among males in the present study was in accordance with these studies.^[1]

In the present study the majority of patients from which *P. aeruginosa* was isolated belonged to age group 21-30 years (40.0%), followed by age group 31-40 years (17.0%) and age group 41-50 years (16.0%).

Another study by Ruhil et al revealed the occurrence of *P. aeruginosa* infection in patients

aged 16-40 years^[10]. However, Mahmoud et al reported more *P. aeruginosa* infections in patients with age group more than 45 years and Sherertz et al reported majority of *P. aeruginosa* infections in patients in age group 50-80 years^[6,7].

CONCLUSION:

This study concluded that *P. aeruginosa* was grown predominantly in urine samples especially in young adults hospitalised patients.

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