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A Study To Know The Prevalence Of Acinetobacter Spp In Various Samples And Their Antibiotic Suspectibility Pattern In A Tertiary Care Hospital.

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Abstract: This study has been undertaken to investigate the prevalence of Acinetobacter spp infection in the clinical samples and to know their drug resistance and sensitivity pattern. To know the prevalence of Acinetobacter in different clinical wards & OPDs. It also includes the rates of Multi drug resistant Acinetobacter spp (the only isolated spp in my study is Acinetobacter baumanii). Which will help us to reduce the overuse and misuse of antimicrobial drugs in the near future.

Key Words: ICU Infections, Pericarditis, Pneumonia, Skin & wound infections.

I. INTRODUCTION

Acinetobacter species are saprophytic, ubiquitous and have emerged as an important nosocomial pathogen due to its ability for survival in the hospital environment on a wide range of dry and moist surfaces.[1] The genus *Acinetobacter* (from the Greek akinetos, i.e., non-motile) was originally suggested in 1954 by Brisou and Prevot to distinguish the organisms based on their motility in the tribe "Achromobactereae" and was composed of non-pigmented Gram-negative saprophytic bacteria comprising both oxidase-negative and oxidase-positive species. *Acinetobacter* spp are short, plump, typically 1.0–1.5 μm by 1.5–2.5 μm in size as measured during the rapid phase of their growth but often develop into coccoid in the stationary phase, usually present in pairs or long chains of variable length. *Acinetobacter* spp are non-fastidious and can be easily grown on regular laboratory media. On blood agar plates, colonies display typical shapes and size, being colorless (white or cream colored), smooth, or mucoid (when capsule is present), milky, 1–2 mm in diameter (after 18–24 h incubation at 37 °C), whereas colonies display bluish to bluish gray color on eosin methylene blue agar. Characteristic colonies on MacConkey agar are light lavender color indicating non-lactose fermenters. They are non-glucose-fermenting encapsulated cocco-bacilli rods which prevail in fluid media, particularly during the early stages of growth, Many strains are unable to reduce nitrates to nitrites and the optimum temperature at which they grow is 33–35 °C. The cell wall of *Acinetobacter* is typical of that of Gram negative bacteria, however de-staining is difficult because it keeps the crystal violet stain which can lead to erroneous detection as Gram-positive cocci. *They are* heterogeneous group of organisms that are typically free living saprophytes, found almost everywhere, commonly distributed in the environment. However, different species of the genus are generally associated with various habitats e.g. soil, water, sewage, human, foods and animal. *Acinetobacter* spp are generally considered a part of the normal flora of the skin, mucous membranes, pharynx, and human respiratory secretions.

Human infections caused by *Acinetobacter* species include pneumonia (which is most often related to endotracheal tubes or tracheostomies), endocarditis, meningitis, skin and wound infections, peritonitis in patients receiving peritoneal dialysis, UTI and bacteremia.[2,3] The main body areas populated by these microorganisms in hospitalized patients are the skin, oropharynx, and digestive tract.[4] *Acinetobacter* spp

were isolated from various locations of the healthy individual's body including the forehead, nose, ear, throat, trachea, conjunctiva, hand, vagina and perineum, inhabiting humid areas, such as axilla, the groin and toe webs.[5] *Acinetobacter* spp can endure dry conditions for long periods thus *A. baumannii* is frequently isolated from reusable medical equipments such as ventilator tubing, arterial pressure monitoring devices, humidifiers, washbasins, plastic urinals and respirometers.[6] *Acinetobacter* spp. are more frequently found on inanimate objects and hands of staff in the ICU than Staphylococcus aureus and Pseudomonas spp. It is hard to determine the significance of recovery of *Acinetobacter* spp from clinical materials, since they are frequently colonized instead of infected.[7]

The potential modes of *A. baumannii* (which is most commonly isolated from the clinical samples) transmission into a ward are displayed through a colonized patient being the most likely mode. After its diffusion to a ward, *A. baumannii* can be transmitted from the colonized patient to the surroundings and to other susceptible patients. Transfer of *Acinetobacter* to several patients is boosted by a combination of multiple-site patient colonization, widespread environmental contamination, persistence on dry surfaces and hands for long periods, and the ability to develop or gain resistance to nearly all classes of antimicrobial agents.[8] Patients with mechanical ventilation, particularly of prolonged duration, longer hospital or ICU stay, greater degree of exposure to infected or colonized patients in the neighbouring hospital environment have an increasing risk for the acquisition of multidrug-resistant outbreak strains.[9] Strains of *A. baumannii* have acquired resistance to newly developed antimicrobial drugs; these strains are known as MDR *A. baumannii*. It became prevalent in many hospitals all over the world and has been recently recognized as a leading nosocomial pathogen.[10] MDR *Acinetobacter* spp can refer to being resistant to a minimum of three classes of antimicrobial drugs e.g. all penicillins and cephalosporins, fluoroquinolones, and aminoglycosides. Another specific definition of multidrug resistance is whenever there is resistance to more than two of the following five drug classes: anti-pseudomonal cephalosporins (ceftazidime or cefepime), anti-pseudomonal carbapenems (imipenem or meropenem), ampicillin-sulbactam, fluoroquinolones (ciprofloxacin or levofloxacin), and aminoglycosides (gentamicin, tobramycin, or amikacin).[11] However, carbapenems remain the treatment of choice for *Acinetobacter* infections.

II. Aims and Objectives

To evaluate the Sero-prevalenc of Acinetobacter spp in various clinically suspected samples.

To study the age and gender wise distribution of Acinetobacter spp.

To study the Antibiotic susceptibility pattern and their drug resistance pattern.

To know the epidemiological factors affecting transmission

III. Materials and methods

This study was carried out in Bacteriology laboratory, Department of Microbiology, PDU Govt. Medical college, Rajkot from February 2022 to May 2023.

The source of the samples for study are the clinical samples from patients admitted in different wards and outdoor patients at PDU Govt. Medical college, Rajkot.

Demographic and clinical data were obtained from wards and outdoor case files.

All samples were subjected to routine microscopy, Identification of isolates was performed by standard conventional methods based on the colony morphology, preliminaries like gram staining, catalase test, oxidase test, motility. Various biochemical tests were used to identify genus *Acinetobacter* like indole, citrate utilization test, urease test, triple sugar iron agar test, phenylalanine deaminase test. Identification of *Acinetobacter baumanni* species was made conventionally using specific tests like oxidative/fermentation glucose test, Arginine decarboxylation, and growth at 33-35°C.

Antibiotic susceptibility testing was performed by the Kirby Bauer disc diffusion method on Mueller-Hinton agar plates and interpreted according to the CLSI guidelines.

All the *Acinetobacter* isolates were tested for their antibiotic susceptibilities for various classes of antimicrobials using the following antibiotic discs: Cephalosporins (ceftazidime, ceftriaxone), Aminoglycosides, (Gentamicin, Amikacin), Fluoroquinolones (Levofloxacin, Ciprofloxacin), beta-lactam and beta-lactamase inhibitor combination drugs (Ampicillin + Sulbactam, Piperacillin + Tazobactum), carbapenems (imipenem, meropenem).

The following picture shows grams stain, colony morphology and biochemical reactions of Acinetobacter baumanii.

IV. Results

Of the total 36,131 samples, 27,989 were routine samples that were tested and 8,142 were tested for blood culture.

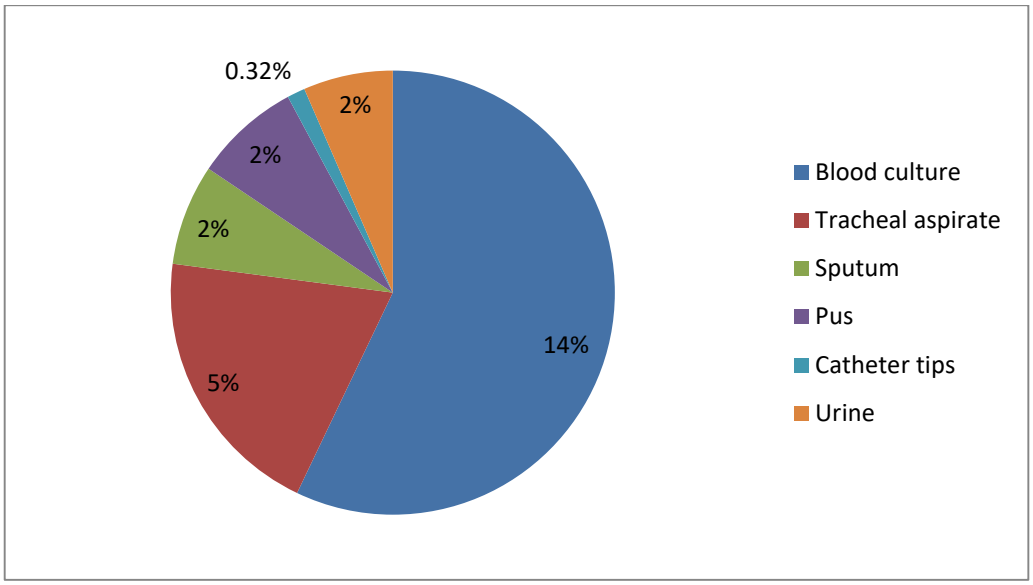
Out of 27,989 routine samples tested, 6,071 were culture positive. Out of which 3,036 were gram positive cocci, 2,011 were gram negative bacilli lactose fermenters, 535 were gram negative non-fermenters, and Acinetobacter baumanii isolated were 489.
Out of 8,142 blood culture samples tested, 1,068 were found to be positive for culture growth. Of which gram negative bacteria were 367 and rest 469 were gram positive cocci. Of total gram negative organisms 128 were non-fermenters and the isolated number of Acinetobacter baumanii from total non-fermenters was 104.

Acinetobacter baumanii isolated from blood samples were 104 (14%), followed by respiratory samples (endotracheal secretion, tracheal secretion) 298 (5%), pus 121 (2%), sputum 110 (2%) and urine 79 (2%) and various catheter tips 20 (0.32%). Maximum *Acinetobacter* species isolated were from Intensive Care Units (ICUs) (39%) followed by surgery ward (21%), medicine ward (15%), orthopedics ward (12%), pediatric ward (11%), gynecology ward (3%) and out patients department (3%). Significant difference was noted in infections in ICU caused by other non-fermenters (9.2%) and *Acinetobacter* species (8.3%). In the present study, isolated species were only *A. baumannii*.
There was a higher incidence of infection among males (59%) than the females (41%).
Acinetobacter infection was more common in patients in age group >51 years followed by <10 years.
The disc diffusion susceptibility testing shows the percentages of resistance and sensitivity among all isolates. Maximum resistance was recorded for ceftriaxone (60%), ceftazidime (75%), cefepime (80%), cefotaxime (63%), amikacin (40%), ciprofloxacin (45%), Cefoparazone + Sulbactam (59%) and ceftazidime + clavulanic acid (33%). The increased resistance of antibiotics for ICUs isolates in comparison to wards isolates was seen. Of 593 total positive isolates of Acinetobacter baumanii, 201 isolates were multidrug resistant strains (isolates resistant to at least one agent in three or more antimicrobial categories-penicillins, cephalosporins, aminoglycosides, fluoroquinolones, and carbapenems).

Table No:1 Comparing the routine samples with blood culture for Acinetobacter baumanii.

Sr No:	Parameters	Samples tested
1.	Routine Samples Tested	27,989
2.	Routine Samples Total Culture Positive	6,071
3.	Routine Samples Positive for Only Acinetobacter baumanii	489
4.	Blood Culture Tested	8,142
5.	Blood Culture Total Culture Positive	1,068
6.	Blood Culture Only Positive for Acinetobacter baumanii	104

Pie chart No 1: Pie Chart showing the isolated rates of Acinetobacter baumanii in various clinical samples.



Pie chart No 2: Pie Chart depicting the isolated rates of Acinetobacter baumanii from different wards and OPD.

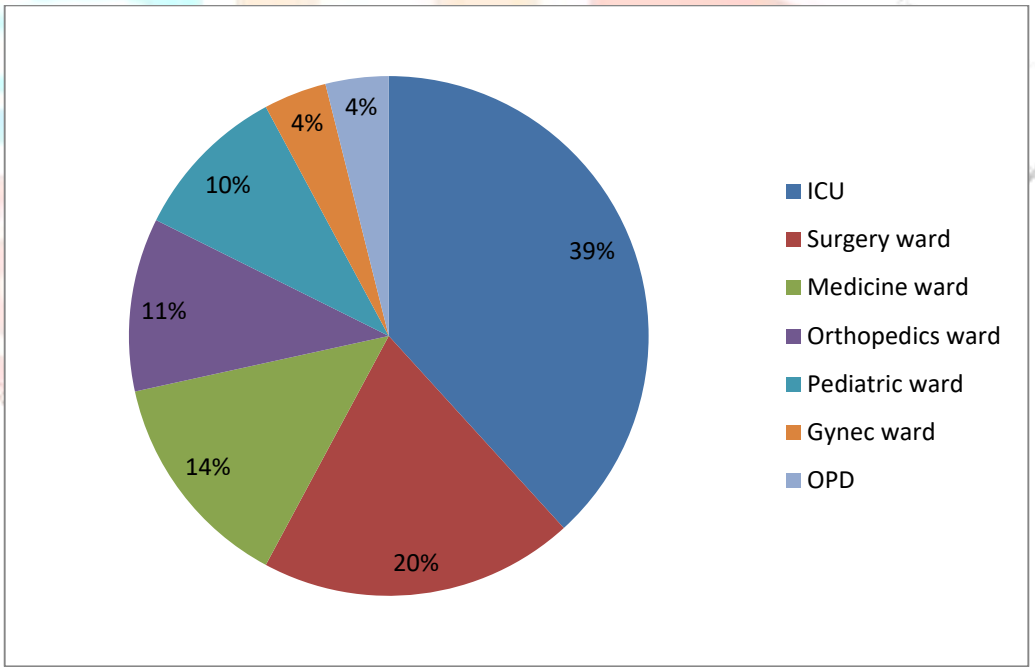


Table No:2: Table comparing the Acinetobacter baumanii isolated rate according to the gender wise distribution.

Sr No:	Gender	Acinetobacter isolated rate in %
1.	Males	59%
2.	Females	41%

Chart No:3: Following chart depicts the isolated rates of Acinetobacter in different age groups.

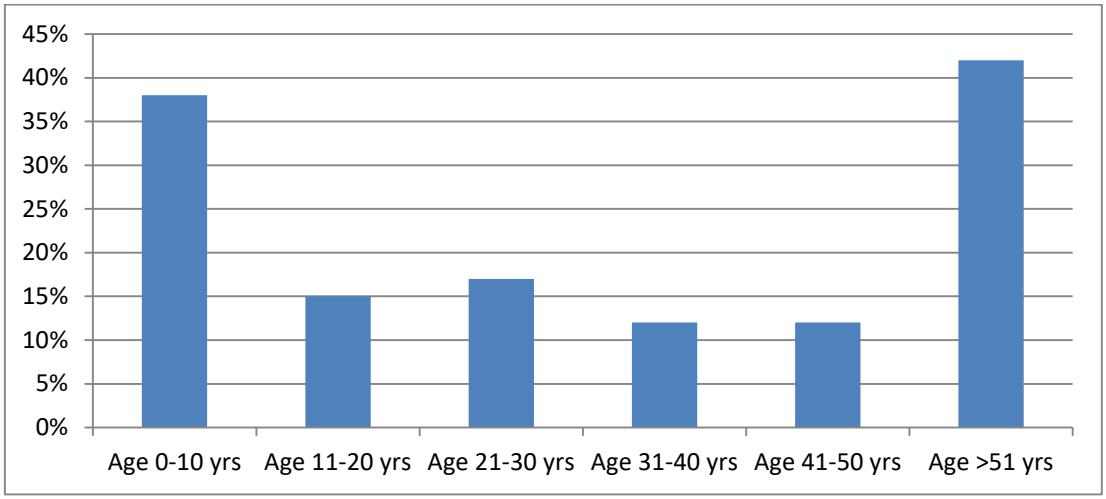


Table No:3: Table showing the antibiotic sensitivity and resistance pattern.

Sr No:	Drugs	Sensitivity in %	Resistance in %
1.	Gentamycin	58%	42%
2.	Amikacin	60%	40%
3.	Piparacillin + Tazobactam	66%	34%
4.	Ampicillin + Sulbactam	41%	59%
5.	Cefuroxime	22%	78%
6.	Cefepime	20%	80%
7.	Cefotaxime	37%	63%
8.	Ceftriaxone	40%	60%
9.	Ceftazidime + Clavulanic acid	33%	67%
10.	Cotrimoxazol	87%	13%
11.	Ceftazidime	25%	75%
12.	Cefoparazone	33%	67%
13.	Cefoparazone + Sulbactam	41%	59%
14.	Cefotaxime + Clavulanic acid	20%	80%
15	Meropenem	100%	0%
16	Imipenem	88%	12%
17.	Levofloxacin	80%	20%
18.	Ciprofloxacin	55%	45%

V. Discussion

Acinetobacter is a nosocomial pathogen. Its ability to infect healthy hosts and its propensity to develop antimicrobial drug resistance is a cause for concern among infectious disease speciality.

Acinetobacter isolated from normal skin and mucous membrane are reported to cause serious and sometimes fatal infections.[12] Bacteremia due to *Acinetobacter* occur most frequently in critically ill patients particularly admitted in ICUs as these patients usually require prolonged hospital stay, need repeated invasive procedures and frequently receive treatment with broad spectrum antimicrobials.[13]

Delay in receiving adequate empirical antimicrobial therapy has an adverse effect on clinical outcomes in hospital-acquired infections caused by A. baumannii.[14] Acinetobacter-associated nosocomial infections in critically ill patients are on the rise.[15,16]

Table No:4: Table comparing the rates of isolated Acinetobacter in my current study and studies conducted by different authers.

Authers	% of Isolation in blood culture and Respiratory samples (tracheal aspirate & sputum)	% of isolation in ICU	Age wise distribution	Gender wise distribution	% of Drug resistance
Pradnya Mahapure et al (year 2024)	Acinetobacter isolated rates from tracheal aspirates (5%) & sputum (2%) and predominately isolated from blood culture is (14%).	Maximum isolation from ICU (39%)	Most common age groups involved are adults >55 yrs and <10yrs.	Acinetobacter was predominately isolated from Males.	Maximum resistance shown by Cefuroxime (78%) Cefepime (80%) Ceftazidime (75%) Cefotaxime (63%) Ceftazidime + Clavulanic acid (80%)
Neetu Gupta et al (year 2015)	Acinetobacter was predominately isolated from Blood culture (36%)	Maximum isolation from ICU (38%)	Most common age groups affected were >50 yrs and <10 yrs.	Acinetobacter was mainly isolated from Males, in neonates.	Maximum resistance shown by Piperacillin (55%) Ceftriaxone (46%) Ceftazidime (46%) Cefepime (44%)
Sudhaharan Sukarya et al (year 2014)	Acinetobacter was predominately isolated from Blood culture (40%)	Maximum isolation from ICU	Most common age groups affected were >55 yrs.	Acinetobacter was mainly isolated from Males than Females	Maximum resistance shown by Ceftriaxone (80%) Ceftazidime (82%)

					Gentamicin (73%) Levofloxacin (64%) Carbapenems (70%)

Nevertheless, it is essential to periodically perform such prevalence and sensitivity assays as it will help clinicians in better management of Acinetobacter infections.

VI. Conclusion

This study is undertaken in Bacteriology Laboratory, Department of Microbiology of P.D.U Govt Medical College & Hospital, Rajkot over a period of one year from February 2022 to May 2023.
Of the total 36,131 samples, out of which 27,989 were routine samples and 8,142 were tested for blood culture.
Out of 27,989 routine samples tested, 6,071 were culture positive. Out of which 3,036 were gram positive cocci, 2,011 were gram negative bacilli lactose fermenters, 535 were gram negative non-fermenters, and Acinetobacter baumanii isolated were 489.
Out of 8,142 blood culture samples tested 1,068 were found to be positive for culture growth. Of which gram negative bacteria were 367 and rest 469 were gram positive cocci. Of total gram negative organisms 128 were non-fermenters and the isolated number of Acinetobacter baumanii of total non-fermenters was 104 from the blood culture samples.
Acinetobacter baumanii isolated from blood samples are 104 (14%), followed by respiratory samples (endotracheal secretion, tracheal secretion) 298 (5%), pus 121 (2%), sputum 110 (2%) and urine 79 (2%) and various catheter tips 20 (0.32%). Maximum *Acinetobacter baumanii* isolated were from Intensive Care Units (ICUs) (39%) followed by surgery ward (21%), medicine ward (15%), orthopedics ward (12%), pediatric ward (11%), gynecology ward (3%) and out patients department (3%).

A slight difference was noted in infections in ICU caused by other non-fermenters (9.2%) and *Acinetobacter* species (8.3%). In the present study, isolated species were (*A. baumannii*). Maximum isolation rate of Acinetobacter baumanii was seen in ICUs (39%).

High level of antibiotic resistance was observed in my study, for ceftriaxone (60%), ceftazidime (75%), cefepime (80%), cefotaxime (63%), amikacin (40%), ciprofloxacin (45%), Cefoparazone + Sulbactam (59%) and ceftazidime + clavulanic acid (33%).
However Carbapenems (Meropenem, Imipenem) remains the drug of choice.
The occurrence of *Acinetobacter* species among non-fermenters is high in hospital settings. Rationale use of antibiotics is important and necessary to prevent microbial resistance catastrophe.
Therefore simple phenotypic methods can be used to recognize these enzymes. Resistant antibiotic after sensitivity report should be discontinued and in place a sensitive drug should be given.
A continued awareness of the need to maintain good housekeeping and control of the environment, including equipment decontamination, strict attention to hand washing should undertake to control the spread of *Acinetobacter* in hospitals.

VII. References

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