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Original Research Article

Prevalence and Antibiotic Susceptibility Profile of Acinetobacter Species Isolated from Urinary Tract Infection at Tertiary Care Hospital, Gujarat

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Abstract:

Introduction: Recently Acinetobacter has emerged as an important nosocomial pathogen and various studies demonstrated it as a primary pathogen involved in outbreaks of hospital acquired infections. This ubiquitous organism has ability to survive in the hospital environment due to its capacity to colonize human and environmental surfaces of the hospital. Indiscriminate use of antimicrobials as a part of empiric or definitive treatment leads to emergence of multidrug resistance which is constant threat to healthcare setting. This signifies the need of continuous surveillance of antimicrobial susceptibility profile of Acinetobacter in various geographical areas to decide appropriate empiric antibiotics which strengthens the antimicrobial stewardship programme. Even now adays trend of increasing prevalence of Acinetobacter is observed in community acquired infection.

Aims & Objectives: This study was aimed to determine prevalence of Acinetobacter species& to know antimicrobial susceptibility profile of Acinetobacter species from patients having Urinary Tract Infection. This study was carried out to avoid the injudicious use of antibiotics and outcome of this study would be useful to establish policy on effective empiric antimicrobial treatment in this geographical area.

Materials and Methods: In this observational retrospective study, data was collected at the Department of Microbiology B.J Medical college, Ahmedabad from January-2017 to August-2018 using Laboratory Information System (LIS)and considering only urine samples collected at Microbiology laboratory, from various areas of hospital including wards, Intensive care unit and Outpatient department. Identification of organism was done by conventional culture and antimicrobial susceptibility using manual biochemical test performed form growth obtained on culture medias& Antimicrobial drug sensitivity by employing Kirby-Bauer disk diffusion techniques on Muller Hinton Agar according to recent CLSI guidelines. Colistin screening agar was used to screen colistin. Final data analysis was done using Microsoft Excel data sheet.

Results: Out of 19, 247 urinary samples received at Microbiology laboratory, only 317 Acinetobacter species were isolated from urine samples. The highest number of isolates 111from Surgery department and least from Gynaecology ward 17. Colistin susceptibility was 97.74% followed by Imipenem and Meropenem susceptibility profile, 65.62% and 64.98% respectively. Least sensitive were Ampicillin- Sulbactam, Cefepime and Ceftazidime with percentage of 32.18%, 37.54% and 36.28% respectively.

Conclusion: Non-fermenter Gram negative bacilli emerge in recent years as an important health care associated pathogen and to prevent the spread of the bacteria having resistant profile, it is critically important to implement antibiotic policies, surveillance programmes for multidrug resistant organisms and infection control practices very judiciously.

Keyword: Acinetobacter, Antimicrobial Susceptibility Pattern, Urinary Tract Infection, Antimicrobial resistance

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Introduction

Acinetobacter species, Gram negative nonfermentative bacteria, has emerged as an important nosocomial pathogen involved in outbreaks of various hospital acquired infections. It has become a global threat especially in developing countries like India because this ubiquitous organism drug carrying numerous resistance mechanisms either constitutive or acquired. Moreover, most of the outbreaks were caused by multi-drug resistant (MDR) strains of this organism. Various studies have established the fact that this organism has been recovered from hospital environmental surfaces, from colonized or infected patients or from staff (Hand carriage). [1]Studies conducted in various countries regarding Acinetobacter and its pathogenic role have shown that the most frequent infections caused by this organism were urinary tract infection and respiratory tract infection. As per the global literature search Acinetobacter baumannii is the second most common non fermenting gram-negative organism after Pseudomonas species.[2,3]The treatment of infections caused by Acinetobacter species is challenging due to high prevalence of multidrug resistance and also there are limited therapeutic interventions available for such multi drug resistant Acinetobacter that results in poor patient outcome, longer hospital stay and higher healthcare cost.

Still community acquired infection caused by Acinetobacterare susceptible to most broad-spectrum antibiotics like Imipenem, Meropenem, Sulbactum and combinations of antimicrobials. But now a days emerging resistance of Carbapenems against this versatile genus also documented even in community acquired Acinetobacter infection.[2,4,5].Combination therapy with sulbactam are indicated in severe infections produced by multi drug resistant Acinetobacter species[6]. This tendency of aggressive transformation of bacterial antibiotic susceptibility profile varies with time and geographical locations. In India, we require such studies reflecting this emerging critical priority pathogen and its increasing importance in causing nosocomial infections.[7]In the present study, attempt was made to know the prevalence of Acinetobacter species in urine samples and to determine their antimicrobial susceptibility profile. We need to track this changing susceptibility profile and also species predominance in various geographical area to tackle the future events judiciously.

Material and Method

This was an observational retrospective study. Data was collected at the Department of Microbiology B.J Medical College, Ahmedabad from January-2017 to August2018 with the use of Laboratory Information System (LIS). It is tertiary care center, referral & teaching hospital. Brief history of patients was obtained about frequency of micturition, retention of urine, burning micturition, fever, and chills. The samples were collected in pre-sterile, dry, widemouthed and leak proof universal plastic containers. From the infant, urine samples we recollected by attaching plastic bags after careful cleaning of the genital area. In patients within dwelling catheters, the catheter well was disinfected near its junction with drainage tube and urine was collected by aspiration into sterile syringe. Samples were immediately transported to the laboratory where they were processed promptly. All these samples were obtained from various areas of hospital including

wards, intensive care units and outpatient department. The clinical correlation was done through verbal communication with clinicians of respective area. Unique Laboratory identification number was given to each sample, after receiving the samples in Microbiology laboratory using LIS (Laboratory information system). A measured amount of urine was inoculated on blood agar, MacConkey agar and nutrient agar. The urine was mixed thoroughly before plating. The plates were inoculated using a calibrated loop designed to deliver known volume 0.01ml of urine. These loops (4mm in size) were made up from platinum. The calibrated loop that delivers the fixed volume of urine (0.01ml) recommended to detect lower number of organisms in urine specimen.

Table1: Colony Count Interpretation				
Loop Diameter	Volume	No. Of Colony	Colony Count	
4 mm	0.01 ml	<10	<10000CFU/m1	
		10-100	<10000-100000CFU/ml	
		>100	>100000CFU/ml	

Table1: Colony Count Interpretation

Interpretation criteria of urine culture (ICMR sop BACTERIOLOGY) <10⁻³ CFU/ml - Insignificant bacteriuria, UTI is unlikely $10^{3} - 10^{5}$ CFU/ml - probably significant bacteriuria, UTI probable >10⁵ CFU/ml - significant bacteriuria, UTI is certain Inoculated urine sample were incubated at 37°C for 18-48 hours and growth recorded. Urinary culture infection interpretation for and contamination-based on number of CFU (Colony forming Unit) and clinical non-fermenting correlation. Lactose colonies were followed. Morphology and motility of the organisms were determined by Gram staining and hanging drop method respectively and oxidase test was done.

All the Gram-negative bacilli or coccobacilli grew on MacConkey agar or blood agar and oxidase negative colonies were inoculated on Triple sugar iron agar medium (TSI).[8] Organisms grew on Triple Sugar Iron and produced an alkaline reaction were provisionally considered to be non-fermenter gram negative bacilli, and were inoculated into Hugh and Leifson" smedium for glucose, lactose, sucrose, and maltose to find out whether a particular organism was oxidizer or non-oxidizer.9 Identification of organism was done by manual biochemical test & Antimicrobial drug sensitivity by Kirby-Bauer disk diffusion susceptibility test on Muller Hinton Agar the next day. Colistin sensitivity was done by agar dilution method.[10]

Results

Total numbers of 59,123 samples were received from January 2017 to august 2018. Out of which, 19, 247 (32.55%) urinary samples were collected. Out of 19247 urine samples, 6519 (33.87%) were positive in culture and in rest no growth isolated. Growth of Acinetobacter isolates was in 317 samples, among them 72 patients were catheterized and 245 patients were non catheterized. Out of total isolates (n=6519), 317 (04.86%) were Acinetobacter causing urinary tract infection. Out of this, 272 (85.8%) Acinetobacter baumannii, 17 (5.36%) Acinetobacter lwoffii and28 (8.83%) Acinetobacter species isolated. Most common gram-negative bacteria causing UTI in this study was Escherichia coli (n=3330, 51.08%) followed by Klebsiella pneumoniae (n=1589, 24.37%) whereas Acinetobacter species was 4.86% in isolates.

Age Group	Male	Percentage	Female	Percentage	Total	Percentage
0-10	30	14.63%	26	23.21%	56	17.66%
11-20	37	18.04%	18	16.07%	55	17.35%
21-30	35	17.07%	23	20.53%	58	18.30%
31-40	30	14.63%	17	15.17%	47	14.82%
41-50	25	12.19%	06	5.35%	31	09.78%
51-60	21	10.28%	10	8.92%	31	09.78%
61-70	17	8.29%	08	7.14%	25	07.89%
71-80	09	4.39%	03	2.67%	12	03.78%
81-90	01	0.48%	01	0.89%	02	00.63%
Total	205	100%	112	100%	317	100%

 Table 2: Gender Distribution According to Age Group

Acinetobacter infection was more common in male as compared to female in our study.

Most common age group affected were 0-10, 11-20 and 21-30 (17.66%,17.35%,18.30% respectively). Least common age group affected was 81-90(n=2, 0.63%) followed by 71-80(n= 12, 3.78%).No significant difference observed among both the gender. Hospital area wise distribution of isolates are shown in Table 3. The highest number of isolates 111 (35.02%) were from patient admitted in surgery department. Other locations of isolation were in Medicine wards 96(30.28%), OPD 93(29.34%), Paediatric wards (16.09%); 51 Orthopaedics 42 (13.25%), ICU 40(12.62%) and least from Gynaecology ward 17(05.36%).

Ward	Male	Female	Number Of Patients	Percentage
Medicine	57	39	96	30.28%
Surgery	86	25	111	35.02%
Orthopedic	38	04	42	13.25%
Paediatric	24	27	51	16.09%
Gynecology	00	17	17	05.36%
Opd	52	41	93	29.34%
Icu	30	10	40	12.62%

 Table 3: Area Wise Distribution of Acinetobacter Species

No	Antibiotics	Sensitive	Resistant
1	Amikacine	147 (46.37%)	170 (53.63%)
2	Ampiciline Sulbactum	102 (32.18%)	215 (67.82%)
3	Cefepime	119 (37.54%)	198 (62.46%)
4	Ceftazidime	115 (36.28%)	202 (63.72%)
5	Cotrimoxazole	167 (52.68%)	150 (47.32%)
6	Levofloxacin	151 (47.63%)	166 (52.37%)
7	Ciprofloxacin	155 (48.90%)	162 (51.10%)
8	Gentamicin	153 (48.26%)	164 (51.74%)
9	Imipenam	208 (65.62%)	109 (34.38%)
10	Meropenam	206 (64.98%)	111 (35.02%)
11	Pipericiline Tazobactum	152 (47.95%)	165 (52.05%)
12	Colistin	310 (97.80%)	007 (02.20%)

Table 4: Antimicrobial Susceptibility Profile of Acnetobacter Species Isolated In This Study

Out of 317 isolates, average sensitivity of colistin was sensitive 97.74% followed by imipenem and meropenem with 65.62% and 64.98% respectively. Least sensitive antimicrobials were Ampicillin-Sulbactam, Cefepime and Ceftazidime with percentage of 32.18%, 37.54% and 36.28%

respectively. Average susceptibility of other antimicrobials is enlisted in table 4. Colistin sensitivity was done by agar dilution method and other antimicrobials sensitivity by disk diffusion method. [Table4/figure1]



Figure 1: Sensitivity Pattern of Acnetobacter Spp Isolated In This Study

Discussion

During routine clinical microbiology work in most laboratories, non-fermentative Gram-negative bacilli (NFGNB) other than Pseudomonas aeruginosa are not taken seriously as a pathogen. They are not pursued for identification and dismissed as contaminants.11This study was conducted as a part of dissertation to make post graduate student aware about importance of isolation, susceptibility profile and its pathogenic role in hospital and community acquired infection. We took up this study when we regularly encountered isolates of non-fermentative Gram-negative bacilli (NFGNB) from various clinical samples.

This study has generated authentic results and data for future use for preparation of antibiotic susceptibility policy in our geographical area. These isolates were identified as Acinetobacter species as per standard criteria.[12]

Study	Prevalence
Present Study	04.86%
Preeti B. Mindolli Et Al[13]	04.66%
Malini, Et Al[14]	06.97%
M. Idomir Et Al.[9]	06.50%
Hasan Ejaz Et Al[15]	01.00%
Dr.Shobha Kl Et Al.[16]	19.16%



Figure 2: Prevalence of Acinetobacter Spp In Uti In Various Studies

In present study the prevalence of Acinetobacter species in urinary tract infection was 04.86 % which is comparable to study done by Preeti B Mindolli et al, where prevalence was 04.66%. Whereas the lesser prevalence rate reported by HASAN EJAZ et al (1%). The study done by Malini et al and M. IDOMIR et al reported nearly same prevalence rate (6.97% and 6.5% respectively) and Dr. Shobha KL et al reported 19.16% of prevalence which was much higher than our study.

Ward Present Study		Lt Col Kk Lahiri Et Al.[17]	M.Idomir Et	
		N= 152	Al.[9]n=108	
Medicine	30.28%	04.60%	13.88%	
Surgery	35.02%	16.40%	11.11%	
Orthopaedics	13.25%	03.90%	19.44%	
Paediatrics	16.09%	-	-	
Gynaecology	05.36%	-	-	
Opd	29.34%	17.10%	-	
Icu	12.62%	07.20%	18.51%	

 Table 6: Comparision of Different Studies of Ward-Wise Distribution Of Acinetobacter Isolates

(-) indicates no study carried out.

As seen in Table 6, the study shows highest prevalence of infection was seen in Surgery (35.02%) followed by Medicine (30.28%) and in OPD (29.34%). Whereas least prevalence was seen in gynaecology ward (05.36%) and orthopaedics and paediatric ward with prevalence rate of 13.25% and 16.09% respectively. ICU prevalence rate was 12.62%.

The study done by Lt Col KK Lahiri et al, where highest prevalence rate seen in Surgery with 16.40%, which was comparable to our study. Study done by M.IDOMIR et al. has shown highest prevalence in orthopaedic ward (19.44%).

In the present study, 317 Acinetobacter spp. were sensitive to Colistin (97.80%), polymyxin (98.74%), imipenem (65.62%), followed by levofloxacin (47.63%), ampicillin-sulbactam (32.18%), piperacillin-tazobactam (47.95%), amikacin (46.37%), gentamicin (48.26%), co-trimoxazole (52.68%). In the study done by Neelam Taneja et al. Imipenem (67.4%) shows similar sensitivity pattern with present study (65,62%) but show higher susceptibility with Piperacillin Tazobactam (61.6%) as compared to present study (47.95%). Lower sensitivity pattern seen

with other antibiotics such as Amikacin. Ceftazidime. Gentamycin and Cotrimoxazole. In the study done by Preeti B. Mindolli et al. Piperacillin Tazobactam (83%) and Amikacin (56.5%) shows higher sensitivity pattern compared to present study (47.95% and 46.37% respectively). But average Gentamicin sensitivity pattern is similar with (43.5%) present study (48.26%). The study done by M.IDOMIR et al. ha shown higher sensitivity pattern with Imipenem (93.5%) as compared with present study (65.62%). However, Cotrimoxazole (26.9%) and Gentamicin (38%) shows lower sensitivity pattern then (52.68%, present study 48.26% respectively) and similar sensitivity pattern seen with amikacin.

Malini et al shows 100% sensitivity with Imipenem as compared with present study (65.62%) but lower sensitivity seen with Cotrimoxazole (20%) and Cefepime (10.8%) then present study (52.68%, 37.54% respectively) [Table:7] Resistance patterns among nosocomial bacterial pathogen may vary from one place to other and even within different areas of hospital, over the period of time. [14]

Antimicrobials	Present	Neelam Taneia Et	Preeti B. Mindolli Et	M. IDOMIR	Malini, Et Al ¹⁴
	N=59	Al ¹²	Al ¹³	Et Al ⁹	N=43
	11 37	N=224	N=200	N=108	11 45
Colistin	97.80%	-	-	-	-
Ceftazidime	36.28%	22.3%	-	-	-
Cefepime	37.54%	-	-	-	10.8%
Amp/Sul	32.18%	-	-	-	-
Gentamicin	48.26%	20.1%	43.5%	38.00%	-
Amikacin	46.37%	25.4%	56.5%	47.2%	53.5%
Levofloxacin	47.63%	-	-	-	-
Ciprofloxacin	48.90%	-	-	-	
Co-Tri	52.68%	16.1%	-	26.9%	20%
Pip-Taz	47.95%	61.6%	83.00%	-	-
Imipenem	65.62%	67.4%	-	93.5%	100%
Meropenem	64.98%	-	-	-	-

Table 7: Sensitivity Pattern of The Acinetobacter Spp. Isolated In Various Studies

(-) indicates individual drug is not reported.

Conclusion

Over the past decade, non-fermenter gram negative bacilli emerged as an important health care associated and community acquired high priority critical pathogen. Prevention of spread of this resistant organism critically important. is Preparation of antibiotic policies and following while it strictly doing surveillance programs for multidrug resistant organisms. Infection control practices need to be strictly implemented. Present study shows most common age group affected with Acinetobacter species causing urinary tract infection were 0-10, 11-12 and 21-30 (17.66%, 17.35%, and 18.30% respectively) & the highest number of isolates 111 (35.02%) from Surgery department. Highest sensitivity was seen with Colistin (97.80%) followed by Imipenem and Meropenem with 65.62% and 64.98% respectively. The antibiotic susceptibility pattern of bacterial pathogens Acinetobacter like species require specialized clinical units to be continuously monitored and the results readily made available to clinicians to minimize the resistance. Continuous and combined efforts of microbiologist, clinician. pharmacist, and community to promote

greater understanding of this problem and managing Acinetobacter infections meticulously.

Limitation: We cannot determine the molecular basis of resistance observed in present study due to non-availability of recent diagnostic facility.

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