PREVALENCE AND ANTIMICROBIAL SUSCEPTIBILITY PATTERN OF *ACINETOBACTER* SPECIES AMONG VARIOUS CLINICAL SAMPLES IN A TERTIARY CARE TEACHING HOSPITAL, SVIMS, TIRUPATHI.

Microbiology

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ABSTRACT

Multidrug resistance isolates of *Acinetobacter* sp. are increasing due to indiscriminate use of antibiotics in health care settings. The incidence of resistance to broad spectrum antibiotics is increasing and thus posing a therapeutic challenge. All samples were processed and isolates were identified as per standard protocol.⁵ Antibiotic susceptibility of all isolates was determined by the standard Kirby Bauer disc diffusion method as per Clinical laboratory standards institute (CLSI) guidelines 2017.⁶ Majority of the *Acinetobacter* sp. were isolated from E.T aspirates (47.3%). In our study *A. baumannii* (65.3%) was the most common species responsible for the infections. Among 150 *Acinetobacter* isolates, 68% of isolates were multidrug resistant organisms, 52% were extended spectrum beta-lactamase (ESBL) producers and 44% of isolates were carbapenem resistant. To abate antimicrobial resistance, antimicrobials ought to be prescribed sensibly and observational antimicrobial treatment ought to be resolved for every hospital as per institutional antibiogram. Strict adherence to the antibiotic policy and proper implementation of antibiotic stewardship program in coordination with clinicians can reduce emergence of multidrug resistant *Acinetobacter* species which is of great concern.

KEYWORDS

Acinetobacter, Antimicrobial resistance, infection.

INTRODUCTION:

The prevalence of *Acinetobacter* species (sp.) in India is ranging from 4.8 percent to 12.9 percent in clinical specimens.^{1,2} Multidrug resistance isolates of *Acinetobacter* sp. are increasing due to indiscriminate use of antibiotics in health care settings. The high level of resistance to most of the antibiotics is due to its capacity to accumulate resistant genes.³ The incidence of resistance to carbapenems, cephalosporins, aminoglycosides, quinolones and other broad spectrum antibiotics is increasing and thus posing a therapeutic challenge.⁴

This study might be helpful to know the prevalence of *Acinetobacter* sp. infections, which is useful to clinicians for better diagnosis and treatment, to formulate efficient hospital antibiotic policy and infection control protocol in order to control the incidence of *Acinetobacter* infection and ultimately for the holistic healthcare.

MATERIALS & METHODS:

Clinical specimens that were submitted to the department of clinical Microbiology laboratory from various departments of SVIMS, SPMC (W) hospital, Tirupathi were included in the present study.

Specimens such as blood, pus, endotracheal / tracheal aspirates (ET), urine, sputum, body fluids and cerebrospinal fluid were processed as per the standard protocol.⁵ Samples were processed on blood agar, nutrient agar, and MacConkey agar, further incubated up to 48 hours at 37°C before reporting as sterile. The isolates that showed non-lactose fermenting (NLF) colonies on MacConkey agar and failed to acidify the butts of triple sugar iron (TSI) agar were provisionally considered as Non Fermentative Gram negative bacilli (NFGNB) and they were further identified by using a standard protocol for identification.⁵

Acinetobacter genus was identified by standard methods like Gram staining where they appear as Gram negative coccobacilli, non motile, growth characteristics on culture media, biochemical tests like Catalase test, Oxidase test and Triple sugar iron test. Based on oxidation-fermentation test, growth at 37°C and 42°C, arginine hydrolysis, hemolytic property and chloramphenicol sensitivity, speciation was performed.

Antibiotic susceptibility of all isolates was determined by the standard Kirby Bauer disc diffusion method as per Clinical laboratory standards institute (CLSI) guidelines 2017.⁶ *Escherichia coli* ATCC 25922 and *Pseudomonas aeruginosa* ATCC 27853 were used as control strains. Antibiotics tested were amikacin (30 µg), gentamicin (10 µg), ciprofloxacin (5 µg), cefotaxime (30 µg), ceftriaxone (30 µg),

cefepime (30 µg), ceftazidime (30 µg), piperacillin-tazobactam (100/10 µg), imipenem (10 µg), polymyxin B (300U), cotrimoxazole (1.25/23.75 µg), meropenem (10 µg), and tigecycline (15 µg). (HiMedia, Mumbai).

Statistical Analysis

Descriptive statistics were done by measuring proportion of each species of *Acinetobacter* from the clinical specimens. Proportion of *Acinetobacter* spp which were susceptible/ resistant to each antibiotic was done.

Above analysis was done by using SPSS Version 21 software.

No ethical issue was involved since the study did not involve any patient contact.

RESULTS:

During the study period from March to June 2019, a total of 150 *Acinetobacter* sp. were studied from various clinical samples. Majority of the *Acinetobacter* sp. were isolated from E.T aspirates (47.3%), followed by blood (32.6%), Sputum (4.7%), Pus (3.3%), Urine (2.7%), body fluids (2%), CSF (1.3%) and other samples (6%). According to the gender, out of 150 *Acinetobacter* species, 93 (62%) were isolated from male patients and 57 (38%) were isolated from female patients, showing male preponderance.

In our study, 47.3% isolates were reported from endotracheal aspirates followed by 32.6% from blood, 4.7% from sputum, 3.3% from pus, 2.7% from urine, 2% body fluids, 1.3% from CSF and remaining 6% from other samples.

A. baumannii (65.3%) was the most common species responsible for the infections followed by *A. lwoffii* (29.3%) and *A. hemolyticus* 8 (5.3%).

In the present study, more than 50% of *Acinetobacter* sp. was resistant to Piperacillin-Tazobactam, Cotrimoxazole, Ciprofloxacin, Ceftriaxone, Cefazolin and highly sensitive to Polymyxin B (99.3%), Cefepime (70.6%), Meropenem (56%), Amikacin (60.6%) and Chloramphenicol (57.3%).

Among 150 *Acinetobacter* isolates, 68% of isolates were multidrug resistant organisms, 52% were ESBL producers and 44% of isolates were carbapenem resistant.

DISCUSSION:

Normal inhabitants of *Acinetobacter* genus were soil and water. In people, *Acinetobacter* sp. can colonize skin, wounds, respiratory and gastrointestinal tracts.⁸ It is a pathogen of tropical and moist condition, yet a few species can endure ecological parching for a considerable length of time, a trademark that advances transmission through fomites tainting in emergency clinics.⁹

We have differentiated *Acinetobacter* species from various clinical samples including blood, urine, body liquids, ET suction, CSF, sputum, and different samples, however most predominantly from ET aspirates (47.3%). Similar results were obtained by Chakraborty et al. in West Bengal.¹⁰ Solitary et al. in Srinagar, India announced that dominant part (39.6%) of the *Acinetobacter* sp. were obtained from urine followed by wound discharge and exudates (29.5%) which was not in concordance with our observation.¹¹ *Acinetobacter* is omnipresent in the clinical setting. Its capacity to get by for extensive stretches combined with its capacity to exhibit various antimicrobial resistance patterns has made *Acinetobacter* a fruitful clinical pathogen.¹²

The majority of the patients who were admitted in our hospital had recently gone to essential and auxiliary consideration emergency clinics and normally received mix of β -lactam antibiotics like third and fourth generation cephalosporins (i.e., ceftazidime, ceftriaxone, and Cefepime) along with aminoglycosides (i.e., gentamicin and amikacin) or fluoroquinolones (ciprofloxacin, ofloxacin, and levofloxacin). Subsequently, major share of the confines in our study was impervious to regularly used antibacterial agents, for example, piperacillin-tazobactam, cotrimaxazole, ciprofloxacin, ceftriaxone, and cefazolin.

This implies *Acinetobacter* sp. was expanding step by step, most likely because of indiscriminate use of these antimicrobials in clinical setting. It is re-stressed that wide range antimicrobials ought to be prescribed with alert. We found that polymixin B (99.3%), cefepime-sulbactam (70.6%), amikacin (60.6%) and chloramphenicol (57.3%) were the most sensitive antimicrobials used against this pathogen which is in contrast to other studies conducted by Chakraborty et al and Rit k et al.^{13,10} Mostofi et al. in their observation mentioned that meropenem (31%) was the least safe medication followed by piperacillin/tazobactam (40%), however imipenem (76%) demonstrated high sensitivity for *Acinetobacter* sp.¹⁴, yet in our study meropenem resistance was 44%. Variation among studies could be because of the strategies and the study designs that are affected by the ecological variables and the antimicrobials tested. The lower resistance rate of *Acinetobacter* sp. to polymixin might be because of their confined use in our health care set up. Albeit antimicrobial resistance is an overall concern, it is as a matter of first importance a nearby issue, that is happening in health care settings, which would then be able to spread worldwide.¹⁵ There are numerous factors that may affect on antimicrobial resistance; lessening and limiting the utilization of antimicrobials to just those circumstances where they are justified, at legitimate portion and for the best possible span is the most proper solution.¹⁶ In this way, emergency departments, as the essential hatcheries of antimicrobial resistant pathogens, convey the most elevated obligation regarding appropriate stewardship of our current antimicrobial assets.

Our study uncovered that majority (68%) were multidrug resistant (MDR) *Acinetobacter* sp., and among them, 52% were extended spectrum beta-lactamase (ESBL) producers. Different investigations led by Bhattacharyya et al. in West Bengal and Mostofi et al. in Tehran reported the MDR percentages to be 29% and 54% respectively.^{14,17} (Table 3)

Carbapenems have been the medication of choice for treating *Acinetobacter* infections; however tragically, carbapenem resistant *Acinetobacter* sp. due to carbapenemase enzyme is emerging rapidly around the world.¹⁸ In our study, 44% were carbapenemase producing *Acinetobacter* sp., which is alarming. Taneja et al. in Chandigarh, India considered 224 *A. baumannii* organisms, out of which 50 (22.3%) were resistant to carbapenems.

Polymyxin B and Tigecycline are new options in the treatment of *Acinetobacter* infections. Like our observation, Shareek et al. examined 44 isolates of *A. baumannii* and found that all were sensitive to colistin.¹⁹ The critical finding in their investigation was that eight (3.5%) isolates were resistant to both colistin and tigecycline.²⁰

Different investigators have revealed the resistance rate to colistin somewhere in the range of 1.8 and 2%, which is 0.7% in our study^{21,22} while sensitivity from tigecycline fluctuates from being nonexistent to 66%.^{23,24}

To conclude *Acinetobacter baumannii* species were the major pathogens which were accounted for major share from endotracheal suction and 68% of *Acinetobacter* isolates were MDR and the majority of these isolates were sensitive to Polymixin B, tigecycline and cefepime - sulbactam. To abate antimicrobial resistance, antimicrobials ought to be prescribed sensibly and observational antimicrobial treatment ought to be resolved for every hospital as per institutional antibiogram. Strict adherence to the antibiotic policy and proper implementation of antibiotic stewardship program in coordination with clinicians can reduce emergence of multidrug resistant *Acinetobacter* species which is of great concern.

Table1: Sample Wise Distribution Of *Acinetobacter* Species

S NO	Type of sample	Total no. isolated (n=150)	Percentage
1	ET aspirate	71	47.3%
2	Blood	49	32.6%
3	Sputum	7	4.7%
4	Pus	5	3.3%
5	Urine	4	2.7%
6	Body fluids	3	2.0%
7	Cerebrospinal fluid(CSF)	2	1.3%
8	others	9	6%

Species Wise Distribution Of *Acinetobacter* Spp.

Table3: Antimicrobial Susceptibility Pattern Of Isolated *Acinetobacter* Species

S.no	Antimicrobial agents	Sensitive (n)(%)	Resistant (n)(%)
1	Polymixin B(300U)	149(99.3%)	01(0.7%)
2	Cefoperazone-sulbactam(75/30 µg)	106(70.6%)	44(29.4%)
3	Amikacin(30 µg)	91(60.6%)	59(39.4%)
4	Chloramphenicol	86(57.3%)	64(42.7%)
5	Meropenem(10mcg)	85(56%)	65(44%)
6	Piperacillin-Tazobactam (100/10 µg)	67(44.6%)	83(55.4%)
7	Cotrimaxazole (1.25/23.75 µg)	58(38.6%)	92(61.4%)
8	Ciprofloxacin (5µg)	56(37.33%)	94(62.67%)
9	Ceftriaxone (30 µg)	39(26%)	111(74%)
10	Cefazolin	22(14.6%)	128(85.4%)
11	Tigecycline (15µg).	20(93.3%)	130(6.7%)

Percentage Of Multidrug Resistance Among *Acinetobacter* Isolates:

Extended spectrum beta lactamases (ESBL)	52%
Carbapenemase producing <i>Acinetobacter</i> spp.	44%
Multidrug resistant (MDR)	68%

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