ABSTRACT

Objective: The present study is to determine the prevalence and antibiotic susceptibility of Acinetobacter species in samples collected from patients in tertiary care hospital in Chennai.

Methods: A total of 17,827 patient's clinical samples were collected from various wards and ICUs of Saveetha Medical College and Hospital, Chennai, Tamilnadu over a period of 7 mo between January 2020 and July 2020. All samples were tested in the microbiology lab of Saveetha Medical College and Hospital using standard operating procedures.

Results: Out of 17,827 samples, 2,816 were culture positive. 122 of the isolates tested positive for Acinetobacter spp. and 81.1% of the isolates belonged to Acinetobacter baumannii. Most of the infection occurred in the age group of 21-40 y and predominantly in female patients (female: male ratio 1.9:1). General wards contributed to 54.9% of the Acinetobacter infection, followed by ICU (27%) and OPD (18%). Maximum isolates were recovered from urine (34.4%) and endotracheal secretions (29.5%). 60.7% of the Acinetobacterspp were multidrug-resistant (MDR) i.e. resistant to more than 3 antibiotic group.

Conclusion: Our study observed a high incidence of MDR in Acinetobacterspp, which is in line with most of the research findings in recent times. Most of Acinetobacterspp were resistant to penicillin, third and fourth generation cephalosporins, quinolones, carbapenems, which is alarming as it leaves fewer options for the line of treatment. Some strains were sensitive to cefepime, ceftazidime, piperacillin-tazobactam, levofloxacin, imipenem and meropenem. Considering the increasing MDR nature of Acinetobacter spp a combination of the former along with colistin, tigecycline, amikacin (which have shown more than 85% sensitivity) would need to be studied. Also, strict measures to control the spread of Acinetobacter infection, better management of antibiotics usage and newer therapeutic option for treatment need to be looked at.

Keywords: Acinetobacter, Multidrug-resistant, Prevalence, Antibiotics, Resistance, Sensitive, Carbapenem, Cephalosporin, Quinolones, Infection, Hospital-acquired infection

INTRODUCTION

Acinetobacter is a group of gram-negative coccobacilli that are non-motile, strictly aerobic, catalase-positive, and oxidase-negative, which are commonly found in the environment, like soil and water [1]. In humans, Acinetobacter can colonize in skin, wounds, and the respiratory and gastrointestinal tracts. It can cause serious conditions like sepsis, meningitis, pneumonia, necrotizing fasciitis [2]. It has become a pathogen of increasing significance because over the decades it has grown to become a major cause of hospital acquired infection, a major problem confronting ICU clinicians [3]. One of the reasons is due to its ability to survive for long periods on hospital surface and equipment. Also, Acinetobacterspp has the extraordinary ability to develop multiple resistance against major antibiotic classes which has made it even more difficult to treat the infection. They have become highly resistant to broad spectrum of antibiotics like penicillin, third-generation cephalosporins, carbapenem, and carbapenem [4]. Most strains are resistant to fluoroquinolones as well. Acinetobacterspp produce a wide range of aminoglycoside-inactivating enzymes which also is one of the causes for increased resistance to antibiotics [5]. There is a significant difference in the behavior and spread of multi-drug resistant Acinetobacter spp. recovered in various geographic locations. As several factors cause resistance in Acinetobacterspp, treatment of infections caused by this organism should be based on antibiotic susceptibility tests. Therefore, having information regarding the prevalence and pattern of bacterial resistance to these drugs is important. Keeping these above facts in view, we have analyzed the frequency, risk factors, and resistance pattern of Acinetobacter spp. that were isolated from different clinical samples in a tertiary care hospital in Chennai, India.

MATERIALS AND METHODS

A retrospective, hospital record-based, cross-sectional study was carried out from Jan 2020 to July 2020 in the Department of Clinical Microbiology at a tertiary care hospital in Chennai. A total of 17,827 clinical samples like pus/swab, urine, sputum, blood, body fluid, tracheal aspirate, endotracheal tube, and intravenous (IV) catheter tips were collected from the patients and transferred to the laboratory without delay for further processing. The study was conducted after due approval was obtained from the institutional ethical committee.

Sample processing and antibiogram

In the laboratory, all the collected samples were cultured aerobically on blood agar and MacConkey agar. Blood specimen was cultured in trypticase soy broth (TSB) and subcultured in blood agar and chocolate agar. The isolation identification, and speciation were done according to the standard procedure [5]. All the isolates were tested for antimicrobial susceptibility testing by the standard Kirby-Bauer disk diffusion method. Samples were processed for culture by standard conventional methods and susceptibility testing were determined by Kirby Baurs disc diffusion method. Antibiotics and their strength used was according to the Clinical and Laboratory Standards Institute (CLSI) guidelines [6].

RESULTS

During the period of study from Jan 2020 to July 2020 a total of 17,827 samples were examined from different age group, admitted
in various departments of the hospital. Of the total samples processed, 2,816 samples were culture positive and showed growth of different microorganisms. Out of these positive isolates, 122 (4.3%) were confirmed as Acinetobacter spp. as represented in fig. 1.

![Fig. 1: Percentage of acinetobacter spp isolates in total positive samples](image1)

Of the total 122 Acinetobacter spp isolates, 99 (81.1%) were Acinetobacter baumannii. Other species included Acinetobacter junii 9 (7.4%), Acinetobacter lwoffii 5 (4.1%), Acinetobacter calcoaceticus 4 (3.3%), and Acinetobacter ursingii and haemolyticus together 5 (4.1%) isolates. Fig. 2 shows the distribution of the various Acinetobacter spp.

![Fig. 2: Species-wise distribution of Acinetobacter isolates](image2)

Most of the infection occurred in the population age group of 21-40 y followed by the age group 41-60 y as shown in fig. 3 below.

![Fig. 3: Age-wise distribution of Acinetobacter isolates](image3)

The female patients predominated over male patients in the ratio was 1.9:1 as shown in fig. 4 below.

![Fig. 4: Ratio of Acinetobacter isolates in female vs male](image4)
Acinetobacter infection was seen predominantly in general wards 67 (54.9%) followed by ICU 33 (27%) and OPD 22 (18%) (fig. 5).

Maximum number of isolates were recovered from Urine 42 (34.4%) and Endotracheal secretions 36 (29.5%) as shown in table 1 below.

### Table 1: Distribution of study isolates in various specimens

| Clinical samples   | No. of isolates (%) |
|--------------------|---------------------|
| Urine              | 42 (34.4%)          |
| Endotracheal       | 36 (29.5%)          |
| Blood              | 22 (18.0%)          |
| Exudate            | 16 (13.1%)          |
| Pleural Fluid      | 3 (2.5%)            |
| Sputum             | 2 (1.6%)            |
| BAL                | 1 (0.8%)            |

Antibiotics susceptibility testing of 17 antibiotics belonging to 7 antibiotic group was done and the results are tabulated in table 2 below.
Acinetobacter species showed high resistance to ampicillin (100%), nitrofurantoin 107 (87.7%), ertapenem 101(82.8%), ceftazidime 75(61.5%), gentamicin 64(52.5%), ciprofloxacin 61(50.0%), ceftriaxone 78(63.9%), and cefepime 67(54.9%).

Most of the 122 positive Acinetobacter spp were resistant to multiple antibiotics as seen in the table 3 below.

Table 3: MDR distribution in the study isolates

| Isolates resistant to 'n' of antibiotic groups | Resistant No. |
|----------------------------------------------|---------------|
| n=2                                          | 0(0.0%)       |
| n=3                                          | 80(65.6%)     |
| n=4                                          | 74(60.7%)     |
| n=5                                          | 65(53.3%)     |
| n>5                                          | 54(44.3%)     |

As observed in table 2, none of the isolates were resistant to colistin. Other sensitive drugs were tigecycline 114(93.4%) and amikacin 107(87.7%).

**DISCUSSION**

In the present study, from the 2,816 isolates, 122(4.3%) of the isolates tested positive for Acinetobacter spp. Similar prevalence of 4.5% and 3.4% of the total organism isolated was reported by Rit K. et al. in Odisha and Gupta et al. in Pune.[7,8]

Among the Acinetobacterspp,99 (81.1%) of the strains were confirmed as Acinetobacterbaumannii and remaining 23(18.9%) as other Acinetobacter species(fig. 2). Study by Sharma et al[9] also reported a similar value of 230(83.3%) positive Acinetobacterbaumannii, while Rit K. et al[7] reported slightly lower incidence of 74.02%. Only a study in West Bengal in 2012 by Bhattacharyya. et al. reported a very low value of 5.4% baumannii species[10].

Most infections occurred in age group 21-40 y (52) followed by 41-60 age group(23) (fig. 3). Similar prevalence was reported by most studies [9,11,12].

Gender ratio was 1.9:1, which shows a female preponderance in our study. This was not the case with most studies that reported a slight male preponderance [7-9]. Saha et al. and Sivaranjaniet al.[11,12] have reported slight predominance of female patients over male. One reason for was predominance of female in our study was the high number of female in-patients in obstetrics wards, which accounted for 20% of the isolates.

In the present study maximum isolates were isolated from general wards 67(54.9%) followed by ICU 33(27%). Refer fig. 5 above. Most Acinetobacterspp studies have pointed to high incidence of Acinetobacter infection in patients admitted in hospital especially in ICUs as they are capable of rapid adaptation to the hospital environment [13-15].

Maximum number of isolates were from urine specimen 42(34.4%) followed by endotracheal 36(29.5%), 22(18%) in blood and exudate 16(13.1%). Bhattacharyya et al[10] also reported maximum isolates from urine samples (54%). WhileShanthi et al[16] reported maximum number of isolates from endotracheal (ET) secretion (41.8%), followed by urinary tract (25.5%), wound (20%) and blood (12.7%). Sharma et al[9] and Jaggi et al[14] also reported maximum isolates from respiratory secretions.

Out of the 122 isolates, 74(60.7%) isolates were Multi-Drug Resistant (MDR), defined as resistance to 3 antimicrobial agent groups. This is the similar finding of 62.1% reported by Saha S etal[11] in 2018. While the study in USA and Puerto Rico[17] reported MDR in 54% of its Acinetobacter isolates in 2012.

Our findings on resistance to the commonly prescribed cephalosporins namely ceftazidime and cefepime (36%-43.4%) are consistent with results from several previous studies in the Netherlands[18], USA[17] and India[19], which reported resistance rate of 16%-56% to ceftazidime and cefepime. Studies in Pakistan[20] have reported very high resistance to ceftazidime and cefepime (both 99.2%), which could be due to extensive use of antibiotics. A 55(45.1%) resistance to cefoperazonesulfactam was also recorded in our study, which is similar to 44.78% resistance reported by Saderet al[21] and Lai CC et al[40.0%][22].

Resistance towards carbapenems like imipenem, meropenem was recorded to be 48(39.3%) and 42(34.4%), respectively. A lower resistance to imipenem (25.3%) and meropenem (29.7%) was reported by Saha et al[11].

Studies as early as 1996 show that carbapenems were highly...
sensitive to Acinetobacterspp and were used as first line drug of choice to treat Acinetobacter infection[15,19,23]. Data collected from 37 centers in 11 European countries between 1997 and 2000 reported imipenem and meropenem as the most active agents against Acinetobacter, with resistance rates of 16% and 18% respectively[24]. However, the subsequent data from 12 European countries18 revealed a significant increase in the resistance rates against imipenem (42.5%) and meropenem (43.4%)[25]. Data of the antibiotic susceptibilities from different geographical regions revealed that the resistance of Acinetobacter spp. to imipenem was in the range of, no resistance to 40% (2000-2004)[26]. Other studies have also reported increasing resistance to carbapenems[9,23] which is reflected in our study as well.

Penicillin group of antibiotics showed high resistance to Acinetobacter spp. Ampicillin showed maximum resistance and for piperacillin-tazobactam 56(45.9%). Slightly higher value of resistance for piperacillin-tazobactam 50.5% has been reported by Saha et al[11]. Aminoglycosides resistance was observed with 64(52.5%) isolates resistant to gentamicin while Acinetobacter showed more sensitivity to amikacin 107(87.8%). Most studies[7,11] have shown similar high sensitivity (75%-85%) to amikacin and therefore it is considered as one of the drugs of choice in combination with others for treating Acinetobacter spp.[27]. None of the isolates were resistant to colistin. Most of the Acinetobacter spp. sensitivity study in India and US[9,11,12,14,28,29] have reported mostly zero or around 1% resistance to colistin. Studies in other countries have however shown increase in colistin resistance. Study in Western Pacific[30] region showed 3.3% resistance to colistin. Colistin resistance rates of Acinetobacterspp strains isolated in Germany[31] were 2.8%. In Korea[32], there was high resistance to colistin (30.6%). However, the resistance against colistin is not very high in our country it can be still be used in case of MDR Acinetobacter spp.

In our study, there was 6.6% resistance to tigecycline. Study in Germany[31] also reported 6% resistance. A study in Turkey reported considerably higher tigecycline resistance rates (25%) for Acinetobacter strains[33]. Sensitivity to colistin was 100% followed by tigecycline 114(93.4%) followed by Amikacin 107(87.7%).

Fluoroquinolone, namely ciprofloxacin, levofloxacin showed around 40%-50% resistance to Acinetobacter spp. Which was similar to data reported by Saha et al[11]. Fluoroquinolones have never been proved to be a mainstay antibiotic for Acinetobacterspp due to increased resistance over the years. However, usage of levofloxacin, along with colistin has shown better results[34].

CONCLUSION

Our study observed a high incidence of MDR in Acinetobacterspp, which is in line with most research in recent times. Most of them were resistant to penicillin, third and fourth generation cephalosporins, quinolones, carbapenems, which is alarming as it leaves fewer options for line of treatment. Some strains were sensitive to cepfepine, ceftazidime, piperacillin-tazobactam, levofloxacin, imipenem and meropenem. Considering the increasing MDR nature of Acinetobacterspp a combination of the former along with colistin, tigecycline, amikacin [which have shown more than 85% sensitivity] would need to be studied. Also, strict measures to control the spread of Acinetobacter infection, better management of antibiotics usage and newer therapeutic option for treatment need to be looked at.

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AUTHORS CONTRIBUTIONS

All the authors have contributed equally.

CONFLICTS OF INTERESTS

Declared none

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