INTRODUCTION

Non-fermenting Gram-negative bacilli (NFGNB) are ubiquitous in nature. They are primarily opportunistic pathogens [1]. It has an ability to survive in varying temperatures, pH, and humidity which acts as a major factor for being nosocomial pathogens. The infection posed by them is now a worldwide problem because of their multiple, intrinsic or acquired drug resistance that can lead to increased morbidity and mortality. They are capable of producing biofilms. Hence, there is a risk of colonization during the presence of underlying severe illnesses, long-term hospitalization, stays in intensive care units, selective antimicrobial pressure, and invasive interventions [2]. The relationship between antibiotic use and resistance is complex; a major driving factor for antibiotic resistance is antibiotic use/abuse [3]. In developing countries like India, where the infectious disease burden in high, these antimicrobial resistance is particularly pressing because of cost constraints also [4]. Only a few studies from India provide the antimicrobial susceptibility data of NFGNBs [5]. The emergence of multiresistant strains and pan-resistant strains of these organisms can even cause a sudden outbreak of infection in a clinical unit. High prevalence of multidrug resistance indicates a serious need for surveillance and planning of effective interventions to reduce multidrug resistance in such pathogens [6]. Multiple antibiotic resistance (MAR) indexing has been shown to be a cost-effective and valid method of resistance in such pathogens [7]. The multiple antibiotic resistance (MAR) indexing and finding multidrug-resistant (MDR) bacteria will help to indicate the origin from the high risk of contamination where the antibiotics are often used. Hence, this study was carried out to give the MAR index of non-fermenting Gram-negative bacilli in a tertiary care hospital which would help our infection control team also.

METHODS

Drug resistance was tested using Kirby-Bauer disc diffusion method. MAR index was calculated using the formula, a/b (where, a=number of antibiotics to which the organism was resistant and b=total number of antibiotics to which the organism was tested).

RESULTS

Of 240 Gram-negative non-fermenters isolated, 117 (49%) strains were >0.2 of MAR index, 95 (81%) was from inpatient department. 73 (62%) were hospitalized for more than 3 days, 44 (38%) was from surgery department. 49 (42%) was wound specimen. Out of 117 multiple antibiotic resistant isolates, 99 (85%) were MDR isolates.

Conclusion: Nearly 51% prevalence of isolates >0.2 MAR index shows that the source of contamination can still be brought down by proper surveillance and management with proper usage of surface and skin disinfectants, especially in surgery ward where the MAR index has indicated more usage of antibiotics.

Keywords: Multi-drug resistance, Multiple antibiotic resistance, Non-fermenters, Antibiotic susceptibility testing, Hospital acquired the infection.
Acinetobacter spp. and 93 (39%) were identified as Pseudomonas species. Species-level identification of these 137 organisms was not done. 117 (49%) of 240 NFGNB were found to have MAR index >0.2. MAR index distribution among NFGNB is shown in Table 1. Among these non-fermenters, P. aeruginosa exhibits 39%, Acinetobacter species 16%, and Pseudomonas species 45% of MAR index >0.2. This is explained in Table 2.

95 (81%) was from inpatient department. 73 (62%) were hospitalized for more than 3 days, 44 (38%) was from surgery department. 49 (42%) was wound specimen. The department-wise and sample-wise distribution is shown in Tables 3 and 4, respectively.

Out of 117 multiple antibiotic resistant isolates, 99 (85%) were MDR isolates. The anti-microbial susceptibility pattern of NFGNB with MAR index >0.2 is shown in Table 5.

The isolates were highly susceptible to IMP (70%) and AK (69%). The antimicrobial susceptibility profile of those 99 MDR isolates is shown in Table 6.

**DISCUSSION**

Although the NFGNB’s being the life-threatening pathogens, the proper surveillance and treatment will help to manage those organisms. In this study, the overall prevalence rate of NFGNB was only 7%. Which correlates with the prevalence rate of 10.5% with Olayinka and Onile [8]. NFGNB was of 18% with arora which was seen to be increased. This may be due to the difference in the study population. In this study, P. aeruginosa (42%) was the predominant organism followed by 39% of Pseudomonas species and 19% of Acinetobacter species. However in a study carried out by arora, Acinetobacter spp. (62%) was the most common followed by P. aeruginosa (18%), Burkholderia cepacia complex (5%) and S. maltophilia (3%). 12% (221/1781) of the NFGNBs could not be identified. The MAR index of >0.2 was seen more with Pseudomonas species, that is, 45%, followed by P. aeruginosa 39% then with Acinetobacter species 16%. In this study, the NFGNB with >0.2 MAR index was isolated more from the surgical unit (38%) and sample-wise they were isolated more from wound samples (42%) followed by urinary samples (25%). This was correlated with the study done by Stark and Maki, 1984 [10], where majority strains were isolated from wound swabs. However in the study conduction by Olayinka and Onile [8], P. aeruginosa strains were isolated more from urinary specimens. In this study, 70% were sensitive to IMP, 69% were sensitive to AK and 68% were sensitive to MR. However, 98.1% were sensitive to AK in the study done by Olayinka and Onile [8] being the first-line drug in our study AK exhibited only 69% susceptibility. Compared with the study done at the Lagos University teaching hospital, only 12.5% exhibited resistance to IMP. However in our study, 30% had shown resistance to IMP. Similarly, Olayinka and Onile [8] also had reported resistance to IMP . Similarly, Olayinka and Onile [8] also had reported resistance to IMP. However in our study, 30% had shown resistance to IMP. Similarly, Olayinka and Onile [8] also had reported resistance to IMP.

**Table 1: Distribution of MAR index among NFGNB**

| S.No. | Name of the organism | Total number of patient | MAR index |
|-------|-----------------------|-------------------------|-----------|
|       |                       |                         | 0.1 | 0.2 | 0.3 | 0.4 | 0.5 | 0.6 | 0.7 | 0.8 | 0.9 | 1 |
| 1.    | Pseudomonas aeruginosa | 102                     | 54  | 2   | 11 | 3  | 7  | Nil | 4   | 6  | 7  | 1  | 7 |
| 2.    | Acinetobacter species | 45                      | 20  | 1   | 1  | 1  | 1  | Nil | 1   | 4  | 3  | 1  | 6 |
| 3.    | Pseudomonas species   | 93                      | 40  | Nil | 18 | 5  | 5  | Nil | 2   | 7  | 7  | 3  | 6 |

MAR: Multiple antibiotic resistance, NFGNB: Non-fermenting Gram-negative bacilli

**Table 2: Percentage of NFGNB with MAR index >0.2**

| S.No. | Name of the organism | MAR index |
|-------|-----------------------|-----------|
|       |                       | 0.2 | 0.3 | 0.4 | 0.5 | 0.6 | 0.7 | 0.8 | 0.9 | 1 |
| 1.    | Pseudomonas aeruginosa | 11 | 3   | 7  | Nil | 4  | 6  | 7  | 1   | 7  | 39 |
| 2.    | Acinetobacter species | 1   | 1   | 1  | Nil | 1  | 4  | 3  | 1   | 6  | 16 |
| 3.    | Pseudomonas species   | 18 | 5   | 5  | Nil | 2  | 7  | 7  | 3   | 6  | 45 |
| Total |                       | 117 | 100 |

**Table 3: Department wise distribution of NFGNB with MAR index >0.2**

| Department | Inpatient | Outpatient | Total | Percentage |
|------------|-----------|------------|-------|------------|
| Medicine   | 20        | 5          | 25    | 22         |
| Surgery    | 39        | 5          | 44    | 38         |
| OG         | 6         | 1          | 7     | 6          |
| Orthopedics| 3         | 1          | 4     | 3          |
| Urology    | 2         | 2          | 4     | 3          |
| Pediatrics | 4         | 3          | 7     | 6          |
| ICU        | 18        | 3          | 21    | 18         |
| ENT        | 3         | 2          | 5     | 4          |
| Total      | 95        | 22         | 117   | 100        |

MAR: Multiple antibiotic resistance, NFGNB: Non-fermenting Gram negative bacilli

**Table 4: Sample-wise distribution of NFGNB with MAR index >0.2**

| Sample      | Inpatient | Outpatient | Total | Percentage |
|-------------|-----------|------------|-------|------------|
| Urine       | 26        | 4          | 30    | 25         |
| Wound       | 37        | 12         | 49    | 42         |
| Pus         | 16        | 3          | 19    | 16         |
| Sputum      | 9         | 3          | 12    | 10         |
| Tracheal aspirate | 2 | 0 | 2 | 2 |
| Tissue      | 1         | 0          | 1     | 1          |
| Ascitic fluid| 1         | 0          | 1     | 1          |
| Blood       | 3         | 0          | 3     | 3          |
| Total       | 95        | 22         | 117   | 100        |

MAR: Multiple antibiotic resistance, NFGNB: Non-fermenting Gram negative bacilli

**Table 5: Antimicrobial susceptibility pattern of NFGNB with MAR index >0.2**

| Antibiotics | Sensitive (%) | Resistant (%) |
|-------------|---------------|---------------|
| AK          | 69 (59)       | 48 (41)       |
| G           | 41 (35)       | 76 (65)       |
| CAZ         | 13 (11)       | 104 (89)      |
| CPM         | 13 (11)       | 104 (89)      |
| CIP         | 21 (18)       | 96 (82)       |
| OF          | 27 (23)       | 90 (77)       |
| PIT         | 51 (44)       | 63 (56)       |
| IMP         | 70 (60)       | 47 (40)       |
| MR          | 68 (58)       | 49 (42)       |

MAR: Multiple antibiotic resistance, NFGNB: Non-fermenting Gram-negative bacilli, AK: Amikacin, G: Gentamycin, CAZ: Cefazidime, CPM: Cefepime, CIP: Ciprofloxacin, OF: Ofloxacin, PIT: Piperacillin tazobactam, IMP: Imipenem, MR: Meropenem
Table 6: The antimicrobial susceptibility profile of MDR isolates

| Susceptibility profile | Number of isolates |
|------------------------|--------------------|
| G, CAZ, CPM, OF, CIP, MR | 2 |
| G, CAZ, CPM, PIT, MR    | 2 |
| G, CAZ, CPM, CIP, OF, PIT | 2 |
| G, CAZ, CPM, CIP, OF, PIT, IMP, MR | 7 |
| AK, G, CIP, CIP         | 7 |
| AK, G, CIP, OF          | 5 |
| AK, G, CIP, OF, PIT, IMP, MR | 19 |
| AK, G, CPM, CIP, PIT, MR | 19 |
| AK, G, CIP, OF          | 1 |
| AK, G, CIP, OF, PIT     | 1 |
| AK, G, CIP, OF, PIT, IMP | 3 |
| CAZ, CPM, OF            | 3 |
| CAZ, CPM, OF, PIT, IMP  | 5 |
| CAZ, CPM, MR            | 10 |
| CAZ, CPM, MR, IMP       | 1 |
| CAZ, CPM, MR, IMP, MR   | 2 |
| CAZ, CPM, MR, MR        | 1 |
| CAZ, CPM, MR, MR, MR    | 3 |
| CAZ, CPM, MR, MR, MR, MR| 1 |
| CAZ, CPM, MR, MR, MR, PIT | 1 |
| Total                   | 99 |

MDR: Multi-drug resistant, AK: Amikacin, G: Gentamicin, CAZ: Ceftazidime, CPM: Cefepime, CIP: Ciprofloxacin, OF: Ofloxacin, PIT: Piperacillin-tazobactam, IMP: Imipenem, MR: Meropenem

more than 80% sensitivity to IMP in her study. In this study, 99 isolates exhibited multidrug resistance mechanism. Of which, 19 were pan-drug resistant. Susceptibility profile of MDR isolates revealed that the resistance pattern is equally distributed and none of the isolate is 100% susceptible to a drug. Whereas in a study conducted by Olayinka and Onile [8], 100% MDR strains were sensitive to IMP and 16 out of 18 were resistant to gentamicin. It has been said that there is generally an excess of resistance among isolates from hospitalized patients compared with those from outpatients [6]. This has been correlated well with this study, were 81% (98) were inpatient and 62% of which have been hospitalized for more than 3 days. MAR index higher than 0.2 has been said to be an indication of isolates originating from an environment where antibiotics were often used [7,11]. Analysis of the MAR index of the Pseudomonas strains in a study done by Olayinka and Onile [8], showed that 60.9% had MAR index of 0.3 and above. In this study, 49% exhibited >0.2 of MAR index. It shows that the use of antibiotics is still under control.

CONCLUSION

From this study, it is clear that the prevalence rate of NFGNB is less, that is, 7% in our set up. Of which only 49% exhibited MAR of >0.2. This can be controlled by proper management and surveillance. The isolates were more from surgery department. This shows that the chance of contamination of MDR isolates will be more in these units, which has to be taken into consideration. In this study, 85% were MDR isolates. None were 100% sensitive to any of the drugs because the resistance profile is equally distributed. Nearly 62% were hospitalized for more than 3 days which indicates that these organisms may be the probable source for nosocomial infection in future that need to be treated immediately. When strains have multiple antibiotic resistance, the choice of therapy is limited and difficult. Thus, it is important to have antibiotic policies and surveillance programs. Moreover, it is desirable to periodically monitor the susceptibility pattern of NFGNB as they were the common pathogens causing nosocomial infections worldwide. This will help to administer an effective therapeutic agent whenever there is a need to do so.

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