Prevalence and Recent Trend in the Antibiogram of Staphylococcus Species Isolated from Clinical Specimens in a Rural Tertiary Care Hospital in South India

Ruby Thomas*, K. Shilpa and Allavarapu Ramyashree

Department of Microbiology, Akash Institute of Medical Sciences and Research Center, Prasannahalli road, Devanahalli, Bengaluru rural-562110, India

*Corresponding author

ABSTRACT

The present study was carried out to determine the prevalence of Staphylococcus species in clinical samples and to find out the trend of susceptibility to antibiotics in a rural setting. Standard microbiological and biochemical methods were used to screen 503 clinical specimens comprising of sputum, pus, urine and blood for Staphylococcus species. One thirty nine isolates were obtained from these samples. Of the 64 positive isolates from pus, 19 were Methicillin sensitive Coagulase negative Staphylococcus species (MS-CoNS), 17 Methicillin resistant Coagulase negative Staphylococcus species (MR-CoNS), 24 Methicillin sensitive Staphylococcus aureus (MSSA) and 4 Methicillin resistant Staphylococcus aureus (MRSA). Out of the 30 isolates from urine 12 MS-CoNS, 10 MR-CoNS, 6 MSSA and 2 were MRSA. Blood grew 17 MS-CoNS, 12 MR-CoNS ,6 MSSA and no MRSA. The 10 sputum positive samples had 9 MSSA and 1 MRSA. Antibiotic susceptibility testing was done for all the positive isolates. Linezolid has been found to be the most sensitive to all the four categories followed by Gentamicin. For MSSA, Clindamycin and Erythromycin has been found to be equally sensitive (53%). Penicillin is the least effective drug for all the 4 categories. All the MRSA were sensitive to Vancomycin. Also it was noted that there was a reduction in the prevalence of MRSA (5.03%) from this area.

Keywords
Staphylococcus species, antibiotic susceptibility, prevalence, resistance, MRSA.

Introduction

Staphylococcus is a common bacterium found in clinical samples (Diekema et al., 2001) and also found to cause human infections (Lowy, 1998). Staphylococcus aureus (S. aureus) being the most important member of this group (Javid et al., 2006) usually produces skin infections and if left untreated it may progress to a wide range of complications like wounds, tissue infections, pneumonia, joint and /or bone infections (Delorme et al., 2009). It is also the most frequently encountered species in the hospitals (Emmerson et al., 2004). The hospital acquired infections includes wound infections, ventilator associated pneumonia, blood stream infections caused by intravenous devices and prosthetic materials. The major reservoir of Staphylococcus...
Staphylococcus aureus in hospitals are colonised and infected patients. Health care workers and patients are the main source of Staphylococcus aureus infections. Methicillin was introduced in 1961 and was the first semi synthetic penicillinase resistant drug. S. aureus after that showed a wide spread resistance to Penicillin (Jevons et al., 1961). This further lead to the emergence of Methicillin resistance spreading globally across hospitals. This Methicillin resistance to S. aureus is spreading not only in hospitals but also in community settings. Methicillin resistance is also seen in several other species like S.epidermidis, S.haemolyticus, S.hominis and S.capitis. (Peacock et al., 2005).

Materials and Methods

This study was carried out at Akash hospital, Bangalore Rural North between January 2015 to October 2016 which serves as a tertiary care hospital in a rural area. From a total of 503 samples processed from pus, urine, blood and sputum, we have obtained 139 clinical isolates of all species of Staphylococcus. S.aureus (both Methicillin resistant and Methicillin sensitive) and CoNS (both Methicillin resistant and sensitive) were obtained from the above mentioned samples. These clinical samples were inoculated on Blood agar and Mac Conkey agar incubated at 37°C aerobically for 24 hours.

The isolates were subjected to antibiotic testing by the Kirby Bauer disc diffusion method on Mueller Hinton agar plates as per Clinical Laboratory Standards Institute guidelines (CLSI) 2014. The antibiotics used were Cefoxitin(30µg), Penicillin(10units), Erythromycin (15µg), Clindamycin (2µg), Gentamicin(10µg), Linezolid (30µg), Cotrimoxazole (25µg), Ciprofloxacin (30µg) and Vancomycin (30µg) for MRSA. MRSA was determined showing resistance to Cefoxitin disc while inducible Clindamycin resistance was detected by performing the D test following CLSI 2014.

Results and Discussion

The distribution pattern of Staphylococcal species from all clinical samples is shown in Table 1. Methicillin resistance to S.aureus was seen in 5.03% of the isolates (5 from pus, 1 from urine and 1 from sputum) all were Vancomycin sensitive. MR-CoNS made up 28.05% of the isolates (17 from pus, 10 from urine, and 12 from blood samples), out of which some may be contaminants which were also Vancomycin sensitive. Vancomycin tested by the disc diffusion method however can produce erratic results. D test was positive in four MSSA isolates that indicates inducible resistance to Clindamycin (ICR).

Of the 139 isolates there was female preponderance in age group between, 16 to 30 yrs as shown in Table 3.

Drug resistance pattern to several antibiotics was different among the isolates as shown in Figures 1, 2, 3 & 4. There was also a significant reduction in the number of MRSA isolates (5.03%) as shown in Table 1.

In case of MRSA as shown in Figure 4, out of the 9 antibiotics used Linezolid and Vancomycin works the best with 100% sensitivity. Vancomycin is the drug of choice for MRSA.

Staphylococcus is ubiquitous and has a remarkable adaptability, and versatility to exist as a commensal and pathogen. It also is one of the most infectious agent with high prevalence in both hospital and community settings (Akindele et al., 2010).
**Table 1** Staphylococcus Species Isolated From Various Clinical Samples

| Specimens | MS-CoNS | MR-CoNS | MSSA | MRSA |
|-----------|---------|---------|------|------|
| Urine     | 12 (25%) | 10 (25.64%) | 6 (13.33%) | 2 (28.57%) |
| Pus       | 19 (39.58%) | 17 (43.58%) | 24 (53.33%) | 4 (57.14%) |
| Blood     | 17 (48.57%) | 12 (34.28%) | 6 (17.14%) | 0 |
| Sputum    | 0 | 0 | 9 (90%) | 1 (10%) |
| Total     | 48 (34.53%) | 39 (28.05%) | 45 (32.37%) | 7 (5.03%) |

**Table 2** Age Group of Patients Infected With MS-CoNS, MR-CoNS, MSSA, MRSA

| Characteristics | Total cases (%) (n=139) | MS-CoNS Cases (%) (n=48) | MR-CoNS Cases (%) (n=39) | MSSA Cases (%) (n=45) | MRSA Cases (%) (n=7) |
|-----------------|-------------------------|--------------------------|--------------------------|-----------------------|----------------------|
| Male            | 60 (43.16)              | 18 (37.5)                | 15 (38.46)               | 20 (44.44)            | 3 (42.85)            |
| Female          | 79 (56.83)              | 30 (62.5)                | 24 (61.53)               | 25 (55.55)            | 4 (57.14)            |
| Paediatrics (<18 years old) | 37 (6.61)              | 18 (37.5)                | 12 (30.76)               | 15 (33.33)            | 0 |
| Adults (>=18 years old) | 102 (73.38)            | 30 (62.5)                | 27 (69.23)               | 30 (66.66)            | 7 (100)              |

**Table 3** Frequency of Age and Sex Wise Distribution

| Age in years | Males (N=60) | Females (N=79) |
|--------------|--------------|----------------|
| 0-15         | 15           | 16             |
| 16-30        | 10           | 29             |
| 31-45        | 14           | 13             |
| 45-60        | 14           | 14             |
| >60          | 07           | 07             |

**Fig. 1** Methicillin sensitive Coagulase negative *Staphylococcus* (MS-CoNS)
Fig. 2 Methicillin resistant Coagulase negative *Staphylococcus* (MR-CoNS)

![Graph showing MR-CoNS sensitivity](image1)

Fig. 3 Methicillin sensitive *Staphylococcus aureus* (MSSA)

![Graph showing MSSA sensitivity](image2)

Figure 4 Methicillin resistant *Staphylococcus aureus* (MRSA)

![Graph showing MRSA sensitivity](image3)
This study reports the isolation and characterisation of *S. aureus* (both Methicillin sensitive and resistant) and CoNS (Methicillin sensitive and resistant) from clinical samples at the tertiary care hospital. A total of 139 isolates which were from pus, urine, blood and sputum were processed. The highest number of isolates of *S. aureus* was from pus which was 28 out of 139 (20.14%) followed by 10 from sputum (7.19%), 8 from urine (5.75%) and 6 from blood (4.31%). In our study there was an increase in isolation of MS-CoNS which was 48 out of 139 (34.53%) in comparison with 45 of MSSA (32.37%), findings were consistent with reports elsewhere (Orji *et al*., 2012). Some of the CoNS could have been contaminants or opportunistic pathogens (Nworie and Umeh, 2010). This could be due to poor personal hygiene of the patients from this locality who treat themselves or go elsewhere to quacks before seeking medical attention which could lead to colonisation of the wounds.

Methicillin sensitive *Staphylococcus aureus* (MSSA) was found to be 100% sensitive to Linezolid, 84% sensitive to Gentamicin, and 53% sensitive to both Erythromycin and Clindamycin, 49% sensitive to Ciprofloxacin, 44% sensitive to Cotrimoxazole and least susceptible to Penicillin (9%). Methicillin sensitive Coagulase negative *Staphylococcus* (MS-CoNS) was also found to be most sensitive to Linezolid (100%) followed by Gentamicin (84%) Ciprofloxacin (49%), Cotrimoxazole (44%) and least susceptibility to Penicillin (9%), Clindamycin sensitivity was found to be effective only for MSSA (53%). MRSA shows 100% sensitivity to Vancomycin. There was a reduction in MRSA (5.03%) as compared to other studies probably as this is a rural area where patients are less exposed to antibiotics compared to urban areas. Considering the limited number of antibiotics to which different species of *Staphylococcus* were sensitive it is mandatory to follow good infection control practices and also adopt an effective antimicrobial stewardship programme for the hospital.

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How to cite this article:

Ruby Thomas, K. Shilpa and Allavarapu Ramyashree. 2016. Prevalence and Recent Trend in the Antibiogram of Staphylococcus Species Isolated from Clinical Specimens in a Rural Tertiary Care Hospital in South India. Int.J.Curr.Microbiol.App.Sci. 5(12): 644-649.
doi: http://dx.doi.org/10.20546/ijcemas.2016.512.071