IOSR Journal of Dental and Medical Sciences (IOSR-JDMS)
e-ISSN: 2279-0853, p-ISSN: 2279-0861.Volume 21, Issue 10 Ser.3 (October. 2022), PP 57-60
www.iosrjournals.org

Prevalence of Staphylococcus aureus resistance from the isolates of clinical cases obtained from a hospital: A prospective study

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Abstract
Background: *The current study aims to evaluate biofilm production among clinical isolates of Methicillin-Resistant Staphylococcus aureus (MRSA) in a Tertiary Care Hospital in Central India. This goal was accomplished by focusing on the isolation and prevalence of S. aureus in various clinical samples. as well as the detection and prevalence of Methicillin-Sensitive Staphylococcus aureus (MSSA) and MRSA isolated from clinical samples.* ***Methods:*** *This is an observation-and-description-based research project. Index Medical College Hospital & Research Centre in Indore approved the research project's ethics.Pus, urine, blood, sputum, urethral swab, CSF, and pleural fluid were obtained and processed at the hospital's microbiology laboratory for culture and antibiotic susceptibility testing.* ***Results:*** *Highest number of samples we procured are from the Pus. The next samples we procured from the urine.The number of cases susceptible and resistant to Methicillin bacteria through Cefoxitin (30µg) disc diffusion method when performed on total 170 samples.Anti-microbial susceptibility as well as resistant in MRSA subjects. We observed 100% susceptibility in terms with cefoxitin and 0% resistant in MRSA subjects. Next highest susceptibility was seen in Vancomycin and Linezolid anti-microbials. The least susceptibility was observed in anti-microbials such as Cotrimoxazole and Penicillin.*
Conclusion: *Cefoxitin was discovered to be useful in providing accurate detection of MRSA. Hospitals absolutely need to have antibiotic policies and infection control programs in place if they want to prevent and get a handle on hospital-acquired infections like MRSA.*

Date of Submission: 26-09-2022 Date of Acceptance: 11-10-2022

I. Introduction:

In hospitals, penicillin-resistant *Staphylococcus aureus* strains began to show up in the 1940s and have since reached anywhere from 75 to 95 percent prevalence [1-3]. MRSA strains have been linked to infections of the skin, soft tissue, endovascular tissue, urinary tract infections, lung infections (such as pneumonia), heart infections (such as endocarditis), and septic shock [2,3]. MRSA has spread to a significant number of hospitals and other health care facilities around the world [3]. It is the pathogen that is found the most frequently all over the world, making it the most resistant to antimicrobial treatment. In India, the rate of MRSA infection is anywhere between 30 and 70 percent of the population [4,5]. With the discovery of methicillin in the 1960s, it became possible to effectively treat infections brought on by *Staphylococcus aureus*, which is responsible for the production of the penicillinase enzyme [6.7]. In 1961, MRSA strains containing PBP-Pencillin Binding protein were found, altered, and eventually appeared. These strains rapidly spread across the world and the resistance can be traced back to the modified form of PBP2a [8,9].

Community Acquired MRSA is a strain of MRSA that has emerged in hospitals and the general public alike, and it is the source of problems in a large number of Asian countries as well as the United States and the United Kingdom (HA-MRSA and community-acquired MRSA) [1,2]. Over the past twenty years, there has been a rise in the incidence of MRSA in many countries and it is contingent on a number of epidemiological factors and variables [9]. It is currently more difficult and expensive to treat infections caused by MRSA, which has been caused by a small number of epidemic strains of MRSA, than it is to treat infections caused by MSSA [5-8].The current study aims to evaluate biofilm production among clinical isolates of Methicillin-Resistant Staphylococcus aureus (MRSA) in a Tertiary Care Hospital in Central India. This goal was accomplished by focusing on the isolation and prevalence of S. aureus in various clinical samples. as well as the detection and prevalence of Methicillin-Sensitive Staphylococcus aureus (MSSA) and MRSA isolated from clinical samples.

II. Materials & Methods:

This is an observation-and-description-based research project. Index Medical College Hospital & Research Centre in Indore approved the research project's ethics.Only 170 specimens received at the microbiology laboratory of Index Medical College Hospital & Research Centre in Indore were included in this study. Standard microbiological sampling procedures were used [10].

Pus, urine, blood, sputum, urethral swab, CSF, and pleural fluid were obtained and processed at the hospital's microbiology laboratory for culture and antibiotic susceptibility testing [11]. Kirby–Bauer disk diffusion was used to test each isolate's antibiotic susceptibility. CLSI recommends this method. The Mueller-Hinton agar surface was swabbed with a sterile cotton swab and compared to the McFarland 0.5 turbidity standard. Antibiotic discs included tetracycline, ciprofloxacin, gentamycin, clindamycin, cotrimoxazole, erythromycin, vancomycin, linezolid, and penicillin. The plates were incubated overnight at 37°C. After a set time, the zone diameter organism was classified as resistant, intermediate, or sensitive. Multidrug-resistant (MDR) bacteria were resistant to three or more antibiotic classes.

MRSA detection using Cefoxitin Disc-Diffusion: S. aureus susceptibility to cefoxitin (30 g) was determined using a modified Kirby–Bauer disc-diffusion method. MRSA-resistant S. aureus strains (isolates with a 21-mm zone of inhibition) were screened for MRSA[12].

Statistical Analysis:

All gathered data was entered into SPSS (22.0) and will be analyzed. Between MRSA and MSSA, antibiotic susceptibility, MRSA, and biofilm formation were compared.

III. Results:

Table 1 depicts the specimen distribution from which tissue we got the samples. Highest number of samples we procured are from the Pus. The next samples we procured from the urine.Table 2 show the number of cases susceptible and resistant to Methicillin bacteria through Cefoxitin (30µg) disc diffusion method when performed on total 170 samples.Table 3 shows the anti-microbial susceptibility as well as resistant in MRSA subjects. We observed 100% susceptibility in terms with cefoxitin and 0% resistant in MRSA subjects. Next highest susceptibility was seen in Vancomycin and Linezolid anti-microbials. The least susceptibility was observed in anti-microbials such as Cotrimoxazole and Penicillin.

IV. Discussion:

It has emerged as a significant pathogen that can be found in hospitals as well as the general public[13,14]. The infections that it can cause range from superficial skin and soft-tissue infections to systemic infections that can be fatal. The increasing prevalence of bacteria that are resistant to methicillin is making an already challenging situation even more so. Two of the most important antibiotic classes for treating staphylococcal infection, cephalosporins and monobactams, have been found to be resistant to this antibiotic resistance [15].Studies [14-17] found that preventing the spread of MRSA in hospitals was associated with significant financial costs as well as concerns regarding patient safety. As a consequence of this, it is absolutely necessary for clinical microbiology laboratories to accurately identify the organism. With the help of this data, we will be able to determine the most effective antimicrobial therapy.

Index Medical College & Hospital in Indore had a 38 percent prevalence of Methicillin-resistant *S. aureus* in our prospective study. Both Amritsar-based Oberoi et al. (2012) [18] and Kathmandu-based Khanal et al. (2018) [19] found similar rates of isolation. While Kumari et al. (2008) [20] reported a prevalence rate of 26.4%, the current study found a lower prevalence rate of 26.3 percent. Anupurba et al. [21] conducted a study in Uttar Pradesh in 2003 and found that MRSA prevalence was 54.85 percent.

Among the participants in this study, MRSA was found in those older than 60 years (30 percent) and MSSA was found in those between the ages of 51 and 60 years. In other words, the majority of those affected were between the ages of 31 and 40. However, Sharma et al. (2011) [22] and Dar et al. (2006) [23] report that people between the ages of 45 and 65 are most likely to experience symptoms.

Females (36%) were more affected than males (64%) in a comparative sex study of MRSA prevalence, with a female:male ratio of 29:34. Cugati et al., 2017 reported a different female-to-male prevalence ratio (62:38), as did Sharma et al., 2006. MRSA isolates were primarily isolated from pus (56%) and urine (18%); similar findings were reported by Anupurba S et al., 2003 [21]. However, Sasirekha et al. (2012) [24] reported 71.42 percent.

In this particular study, MRSA did not exhibit any resistance to the antibiotics vancomycin or linezolid. Both Oberoi et al. (2012) [18] and Arora et al. (2008) [25] discovered that these medications exhibited a sensitivity level of one hundred percent in the course of their research that was carried out in Punjab and Haryana.

An investigation was conducted to determine how resistant MRSA isolates are to a set of different antimicrobials. Isolates of MRSA demonstrated a high level of resistance to penicillin (91.8 percent),

cotrimoxazole (90.2 percent), ciprofloxacin (70.2 percent), and erythromycin (100 percent) (50.2 percent). According to research [21,22], there is also a high level of resistance to penicillin, erythromycin, and ciprofloxacin. In this study, a high percentage of amikacin sensitivity was discovered (98 percent), whereas in another study, a high percentage of amikacin resistance was discovered (78.57 percent) [24].

When molecular methods are not routinely used, the Cefoxitin disk diffusion test is an excellent substitute marker for detecting methicillin resistance [26]. Oxacillin screening agar and disc diffusion are the two most commonly recommended methods for MRSA testing, but this method outperforms both of them [27]. According to the results of this study, no strain was resistant to vancomycin or linezolid [28]. In contrast, vancomycin-resistant MRSA strains have recently been discovered in various parts of the country[29].

Penicillin, cotrimoxazole, and ciprofloxacin all showed statistical resistance patterns to MRSA. MRSA detection using the cefoxitin disc diffusion method was found to be both accurate and affordable. Compared to oxacillin as an MRSA indicator, cefoxitin outperforms oxacillin.

This study discovered a prevalence of MRSA that was lower than the average, falling between 40 and 69.8 percent [30,31], but it was comparable to a study conducted in Malaysia [32]. Eighty percent of strains of MRSA tested positive for resistance to at least one antibiotic, including clindamycin, erythromycin, penicillin, cotrimoxazole, and ciprofloxacin (90 percent). 92 percent of the total. The winner received 76.2 percent of the vote. The number given for the percentage is 76.2 percent. All of the pathogens were killed by the antibiotics penicillin (100 percent), vancomycin (100 percent), and linezolid (100 percent), respectively, at the same rate (100 per cent). Rani et al., 2014 [33] made the discovery that gram-positive bacteria have developed a complete resistance to the antibiotics Vancomycin, Linezolid, and Cefoxitin.

V. Conclusion:

The specimen showed that pus was the most common source of the isolated bacteria, followed by urine and least from semen. These strains exhibited a moderate level of resistance to cefoxitin, in contrast to their high level of resistance to cotrimoxazole, penicillin, and ciprofloxacin. Both vancomycin, and linezolid were found to have a one hundred percent success rate. Cefoxitin was discovered to be useful in providing accurate detection of MRSA. Hospitals absolutely need to have antibiotic policies and infection control programs in place if they want to prevent and get a handle on hospital-acquired infections like MRSA.

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Table 1: Specimen distribution of MSSA & MRSA in subjects of the study

Specimen	MSSA	MRSA	Total
Pus	57	38	95
Urine	15	16	31
Blood culture	8	3	11
High vaginal swab	3	2	5
Sputum	6	1	7
Swab culture	7	1	8
Tissue culture	8	1	9
Semen C/S	1	0	1
Pleural fluid	2	1	3
Total	107	63	170

Table 2: Results of Methicillin Resistance in *S.aureus* by Cefoxitin (30 µg) Disc Diffusion Method (N=170)

Pattern of Resistance	No. of Cases	Percentage
Susceptible	**107**	**63%**
Resistant	**63**	**37%**
Total	**170**	**100%**

Table 3: Anti-microbial Susceptibility & Resistant Pattern of MRSA Subjects

Antibiotics	Susceptible		Resistant	
	No. of Isolates	%	No. of Isolates	%
Tetracyclin	42	66.6	21	33.3
Ciprofloxacin	20	31.7	43	76.2
Gentamycin	60	95.2	3	4.8
Clindamycin	40	63.4	23	36.5
Cotrimoxazole	7	11.11	55	90.2
Erythromycin	32	50.7	31	50.2
Vancomycin	63	100	0	0
Linezolid	63	100	0	0
Penicillin	7	11.11	56	91.8
Cefoxitin	63	100	0	0