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Original article

Frequency of MRSA among Random Clinical Specimens Collected from Different Clinics in Surt city

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Abstract

Gram-positive cocci that resemble grape clusters are called staphylococcus aureus. It is mostly a pathogen that affects humans. Methicillin, a semisynthetic antibiotic, was developed, and methicillin-resistant S. aureus (MRSA) was clinically detected; MRSA is mediated by mecA. Normally, S. aureus coexists peacefully with a healthy person. It is also categorized as an opportunistic microorganism, and it can cause abscesses and life-threatening diseases. The study's objectives are to identify MRSA in Surt patients and assess antibiotic sensitivity to MRSA. During the course of a year, from January 1, 2022, to December 31, 2023, 15 clinical isolates of MRSA were obtained from various samples that were submitted to the Diagnostic Medical Microbiology laboratory of various clinics. This includes testing for antibiotic susceptibility as well as isolating and identifying MRSA. 247 positive clinical specimens were used in this investigation; MRSA was found in 6% of the 247 patients. The greatest isolation rate was seen in wound swab specimens (7: 46.6%), followed by urine (4: 26.6%). All MRSA isolates (100%) were sensitive to chloramphenicol. Vancomycin shown susceptibility in 14 isolates (93.3%). A 6% MRSA isolation incidence in Surt City clinical samples is found in the study, underscoring the necessity of better infection control procedures. In order to stop emerging resistance. The study also emphasizes the necessity of consistent monitoring and prudent antibiotic usage. It is essential to educate people about good hygiene and preventative measures.

Keywords: *MRSA, Staphylococcus Aureus, Antibiotic, Infection, Isolation.*

Introduction

Staphylococcus aureus is a gram-positive, cocci, arranged in a grape-clustered shape. It is primarily a human pathogen [1]. In 1881 Sir Alexander Ogston discovered that Staphylococcus could cause wound infections in living organisms [2]. Normally, S. aureus can also be found in healthy individuals [3]. It cannot cause infections on healthy skin, but, if it enters the internal tissues or bloodstream, it may cause severe infections [4]. S. aureus can cause mild skin infections such as impetigo, scalded skin, boils, and abscesses. It also causes life-threatening diseases such as meningitis, pneumonia, endocarditis, bacteremia, osteomyelitis, and sepsis [5].

According to studies, 20% of individuals are nasal carriers of S. aureus, and 30% are intermittent carriers [6]. In recent years, S. aureus has been considered one of the major reasons for the spread of hospital and community-acquired infections, which brings about serious consequences and life-threatening diseases [7].

The discovery of antibiotics helped to treat the infectious diseases caused by S. aureus. In 1928, penicillin was discovered [8]. 2 years after introducing penicillin, S. aureus resistance strains emerged, then in 1942, the first penicillin-resistant S. aureus strain was identified, which was mediated by the β-lactamase gene blaZ [8] in 1950, methicillin, a semisynthetic antibiotic, was designed, and methicillin-resistant S. aureus (MRSA) was clinically detected in the 1960s [9,10]. The first MRSA strains were found in the United Kingdom, and this epidemic was primarily constrained to Europe. Soon after, MRSA was identified in the United States, Japan, and Australia [9].

Methicillin resistance is mediated by mecA that is acquired by horizontal transfer of a mobile genetic element called staphylococcal cassette chromosome mec (SCCmec). The gene mecA encodes penicillin-binding protein 2a (PBP2a), an enzyme responsible for cross-linking the peptidoglycans in the bacterial cell wall. PBP2a has a low affinity for β-lactams, resulting in resistance to this entire class of antibiotics [11].

Colonization with MRSA is a main risk factor for MRSA infection in adults and children, this is especially for patients who acquire MRSA colonization in the nosocomial setting,

where the risk of developing an MRSA infection as a result is around 30%. MRSA is usually spread by direct skin-to-skin contact and this may occur during hospital admission, transfer, or other healthcare-related contact [12]. So, it is considered a major cause of hospital-acquired infections (HA-MRSA) and community-acquired infections (CA-MRSA). CA-MRSA is genetically different from HA-MRSA by possessing a small type of SCCmec, and the frequent production of Panton-Valentine leukocidin, and cytotoxin. CA-MRSA strains are restricted to people outside the health care practice, and associated with the increasing use of outpatient intravenous (parenteral) antimicrobial therapy [13]. The reported incidence of MRSA infection ranges from 7% to 60% [14].

From the 1980s onward, several antibiotics appeared on the market, allowing for the better management of infections. On the other hand, leads to the emergence of additional resistance mechanisms as mutations and acquired resistance determinants that lead to the emergence and spread of resistance to other classes of antibiotics such as aminoglycosides, lincosamides, fluoroquinolones, and macrolides [15]. Vancomycin is one of the first-line drugs to treat MRSA infections for decades, most MRSA isolates are susceptible to it, and to the newly introduced antibiotics such as daptomycin, linezolid, tedizolid, ceftaroline, and quinopristin/dalfopristin [16].

The emergence of vancomycin-resistant S. aureus (VRSA) is the most worrying S. aureus genetic adaptation due to the reliance on this antibiotic in the treatment of MRSA infections. VRSA has been shown to emerge through plasmid transfer of the vanA, vanB, vanD, vanE, vanF, and vanG operon from vancomycin-resistant Enterococcus faecalis. [17] In 2002, the first VRSA strain was recovered in Michigan, USA. [18] According to several studies, the prevalence rates of VRSA range from as low as 1.3 % to 20% [19]. The study aims to detect MRSA among Surt patients, and measure antibiotic sensitivity to MRSA.

Methods

Study setting

The study was conducted on 15 clinical isolates of MRSA collected from different samples submitted to the Diagnostic Medical Microbiology laboratory of different Clinics, throughout one year starting from the first of January 2022 to the end of December 2023.

Isolation and identification of MRSA

All the clinical specimens (blood, urine, sputum, minibal, CSF) delivered to the microbiology laboratory of different Clinics throughout one year (from the 1st of January 2022 till the 31st of December 2023), were processed according to standard operating procedures of AMUH lab.

Patients' specimens were inoculated onto Blood agar and MacConkey's agar plates (Oxoid), (chocolate and Sabouraud dextrose agars, upon required) and incubated aerobically at 37°C for 24 hours [20]. Preliminary identification of the Staph spp isolates was performed according to the standard microbiological techniques [21]. The organisms were presumptively identified by conventional methods as Staphylococcus spp.

Antimicrobial susceptibility testing

The susceptibility of all Methicillin-resistant Staphylococcus aureus (MRSA) isolates to different antibiotics was determined by the Bauer-Kirby disk diffusion technique, and interpreted according to the guidelines of the CLSI for all tested antibiotic agents [22].

All confirmed S. aureus isolates were screened for methicillin resistance by inoculation of Mueller Hinton agar supplemented with 4% NaCl. The 10µg cefoxitin discs were aseptically placed on the surface of the inoculated plates and incubated aerobically at 35oC for 18- 24 hours. The isolates were similarly inoculated onto the surfaces of plain Mueller-Hinton agar plates and Gentamicin (10µg), Amoxycillin/clavulanate (30µg), Erythromycin (15µg), Chloramphenicol (30µg), Cotrimoxazole (25µg), Tetracycline (30µg), Penicillin (10iu), Ciprofloxacin (5µg), Ofloxacin (5µg), Levofloxacin (5µg), Ceftriaxone (30µg), Clindamycin (2ug), 221Vncomycin (30ug) and Linozolide (30µg) discs were placed and incubated at 37oC for 24hrs.

Results

This study was carried out on 247 positive clinical specimens (blood, sputum, urine, and pus) delivered to the microbiology laboratory of Sirt Specialized Clinic throughout one year (from the 1st of January 2021 till the 31st of December 2022), were processed according to standard operating procedures.

Isolation percentage of MRSA.
Out of which 247 clinical isolates, 83 (33.6%) were identified as Staphylococcus. Among the 83 Staphylococcus isolates, 24 were identified as Staphylococcus aureus, while the remaining 59 were identified as coagulase-negative Staphylococcus spp. by conventional methods. Among the total 24 S. aureus isolates 15(6%) isolates were MRSA, and among 59 noncoagulase-negative Staphylococcus spp, 24 (9.7%) isolates were MR-Coagulase-negative Staphylococcus spp.

Table 1: Distribution of MRSA isolates among the total number of clinical isolates

Type of isolates	Number of isolates	Percentage
Total isolates	247	%100
Staphylococcus ssp	83	33.6%
MRSA	15	6%
MR-CONS	24	9.7%

Distribution of MRSA isolates according to the Type of specimen:
The highest isolation was from wound swab specimens (7: 46.6%), followed by urine (4: 26.6%) and sputum specimens (two isolates; 13.3%). And one isolate from vaginal swab (6.6%). Most (10) isolates were isolated from patients aged more than 50 years old. Out of 15 MRSA isolates, 11 (73.3%) were isolated from female patients, while four (26.6%) were isolated from male patients.

Table 2: Distribution of MRSA isolates according to the type of specimen.

Type of specimen	Number of MRSA isolates	%
Wound swab	7	46.6%
Urine	4	26.6%
Sputum	3	20%
Vaginal swab	1	6.6%
Total	15	100%

Results of antimicrobial susceptibility testing
All isolates of MRSA (100%) were susceptible to Chloramphenicol. 14 isolates (93.3%) were susceptible to Vancomycin. While 12(80%) isolates were susceptible to the following antibiotics Ciprofloxacin, Clindamycin, Amikacine, and Tetracycline. 5 isolates (33.3%) were susceptible to Gentamycine and sulphamethoxazole/ trimethoprim.

Table 3: Results of antimicrobial susceptibility test of MRSA isolates.

Antibiotics	Susceptible isolates N (%)	Intermediately susceptible isolates N (%)	Resistant isolates N (%)
Vancomycin	14 (93.3%)	0	1 (6.6%)
Tetracyclin	12 (80%)	0	3 (20%)
Sulfamethoxazole /Trimethoprim	5 (33.3%)	0	10 (66.6%)
Gentamycin	5 (33.3%)	0	10 (66.6%)
Amikacin	12 (80%)	0	3 (20%)
Chloramphenicol	15 (100%)	0	0%
Ciprofloxacin	12 (80%)	0	3 (20%)
Clindamycin	12 (80%)	0	3 (20%)

Discussion

S. aureus is commonly found on our skin and noses as normal flora without causing issues. But if there's a cut or scrape, it can enter the body and potentially cause infections. Humans can become ill with MRSA, which can be acquired from hospital settings and the environment. MRSA infections have a high rate of morbidity and death. Our findings indicate that 15 (62.5%) of the 24 samples of S. aureus possessed a rate of MRSA isolation.

Extensive research has also shown prevalence rates of 66.7% and 72%, Garoy et al., and Hussian et al., [23,24] found that they were equal in terms of overall resistance to AUG with a 72% higher incidence of prevalence. The specimen was mostly composed of wound swabs. On the other hand, Rehman et al., [25] observed a lower prevalence (18%) in Pakistan and 22.2% in India by Vijayamohan et al. [26], while the majority of MRSA isolates in the current study were from female patients. The Jorden study in 2015 reported the same prevalence in both male and female patients. This variation in MRSA prevalence could be caused by several factors, including infection control practices, healthcare facilities, and variations in antibiotic usage in different hospitals [27].

In a study by Mwangi and Maathai reported that chloramphenicol had the highest sensitivity, reaching 85.2%.[28]. In contrast, another study by Mohammad Qodrati in Iran found that 85% of participants had high resistance to clindamycin and 58.3% had resistance to ciprofloxacin, gentamicin, and SXT. The high sensitivity rate (100%) to vancomycin, however, is consistent with what we found [29].

Co-resistance rates against other antibiotics were 23 % to tetracycline and clindamycin, 21.0% to cotrimoxazole, and 17.0% to ciprofloxacin, and to amikacin 10.2 percent, chloramphenicol 23.1 percent, gentamycin 20.68 percent. Moreover, the proportion of vancomycin-sensitive strains was found to be very high (100%). It was documented in Nepal by Raja Ram. [30]

Conclusion

The study reveals a 6% isolation rate of Methicillin-resistant Staphylococcus aureus (MRSA) in clinical samples from Surt City, highlighting the urgent need for improved infection control practices in healthcare settings. MRSA isolates were responsive to vancomycin and chloramphenicol, but resistance to other antibiotics like trimethoprim/sulfamethoxazole and gentamicin was observed. The study also highlights the importance of regular monitoring and responsible antibiotic use to prevent new resistance. The study also highlights the influence of regional healthcare practices, community attributes, and antibiotic prescription methods on MRSA frequency and resistance patterns. Educating the public about MRSA is crucial for early detection and prevention, and training healthcare professionals and the community on hygiene practices can reduce transmission.

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