

IDENTIFICATION OF METHICILLIN RESISTANT *STAPHYLOCOCCUS AUREUS* FROM SURGICAL WOUND SWAB AND ITS ANTIMICROBIAL SENSITIVITY

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## Abstract

Methicillin-resistant *Staphylococcus aureus* (MRSA) colonization is known to increase the risk of surgical site infections and currently there is no proper recommendation for postoperative *S. aureus* screening and decolonization because the data from surgical settings is still unclear. A prospective, cross-sectional investigation was carried out in a lab at Lady Reading Hospital in Peshawar, Pakistan. The Clinical and Laboratory Standards Institute's (CLSI) recommendations were followed for isolating pathogens from wound and pus swabs. Wound swabs were taken from 146 patients, admitted in the surgical ward and were then cultured on Mannitol

Salt Agar and CLED Agar. Using microbiological methods, *Staphylococcus aureus* was identified, and the disk diffusion method was used to screen for antibiotic susceptibility. By using cefoxitin (30 µg) disk testing, methicillin-resistant *Staphylococcus aureus* has been identified. Further confirmed by gram staining and biochemical catalase and coagulase tests. The study comprised 146 wound and pus samples, in which 79 (54.10%) of the patients were males and 67 (45.89) were females. The age group of 11–20 years old had the highest incidence of surgical site infections (SSIs) (22.1%). Of the patients that were enrolled, 114 (78.08%) had *Staphylococcus aureus* isolated from them. Methicillin-resistant *S. aureus* (MRSA) made up 103 cases (70.54%) of these, whilst methicillin-susceptible *S. aureus* (MSSA) made up 7.53%. High levels of resistance to cefoxitin (90.35%) and erythromycin (77.19%) were found in antimicrobial susceptibility testing, but vancomycin exhibited 100% sensitivity. This study emphasizes the high incidence of surgical site infections (SSIs) and the rising concern over antibiotic resistance in *Staphylococcus aureus*. The significant rates of antibiotic resistance that have been found highlight the urgent need for strong antimicrobial stewardship initiatives and stringent infection control procedure.

## INTRODUCTION

Bacteria are a noteworthy aspect of public health as they are the causative agents of many infectious diseases (Y. Huang *et al.*, 2021). Bacteria are amongst the oldest and most successful forms of life, enduring as single-celled prokaryotes capable of enduring extreme environments. Having succeeded in evolution, their simplistic structural morphology reflects a cell size range of 0.5-2.0 micrometers. Most common shapes are cocci (spherical), bacilli (rod), and filamentous. Bacteria exist as single cells, as well as in an ordered collection of cells, termed a colony (Stevenson, FJ *et al.*, 1986). The structural and chemical composition of cell envelopes of gram positive and gram-negative bacteria are different, particularly in the arrangement of membranes and cell walls (Epanand, R.M *et al.*, 2016), (Quintiliani, R.J *et al.*, 1998). Bacterial infections which hinge on thoroughly mechanistic understandings of lethality, continue to plague the practice of medicine and public health and thus continue to kill millions, in the twenty first century, the infections and international losing health is bordered by ischemic heart disease as the number two killer in the world as found in the research of The Lancet dated 21 November, 2022 (GBD *et al.*, 2019). Out of the many species of bacteria that live on or come in contact with human skin, very few are capable of causing human infections. The onset of skin infections from bacteria can start small and develop to cover large areas of skin. These infections can however start small and develop to cover large areas of skin. While there are numerous skin infecting bacteria, the most common are *Staphylococcus* and *Streptococcus*. There are also other, less common, infecting bacteria that skin or wounds, particularly among patients that are hospitalized or in long term care facilities (Ruggiero, M.A *et al.*, 2015). *Staphylococcus* is derived from the Greek words “staphyle” meaning branch or bunch and “kokkos” meaning berry. This is in reference to how the bacterium looks under a microscope, resembling a cluster of grapes. *Staphylococcus aureus* or “Golden cluster seed” is where “golden staph” originated from (Ogston, A *et al.*, 1881). Coccus shaped, gram positive, spore-less and non-migrating opportunistic *Staphylococcus aureus* is a non-spore forming, gram positive, coccus shaped, non-migrating and opportunistic bacterium. It is catalase positive and produces lipases, coagulases, proteases, nucleases, collagenases and beta lactamase, among other enzymes. *Staphylococcus aureus* has colonies of: pink on chromogenic agar, golden on

blood agar, grayish white on blood agar, and yellow on mannitol salt agar, in addition to other colors on various culture media (Carter *et al.*, 1995). It is important to note that *Staphylococcus aureus* is a sphere, gram positive and facultative anaerobe.

Methicillin-resistant *S. aureus* (MRSA) was discovered in the 1960s and has since been a significant threat to global public health (Harkins, C.P., Pichon, B *et al.*, 2017). Initially, the infections of Methicillin Resistant *S. Aureus* were confined to the hospital environment (HA-MRSA) but the first community associated outbreaks (CA-MRSA) during the 1990s in the USA and Australia were reported for the very first time and then later on, the world (Chambers, H *et al.*, 2001, Maira, A *et al.*, 2020). . Initially, community associated methicillin resistant *S. aureus* (CA-MRSA) strains occurred in the community and rested predominantly in the young and healthy population (Naimi *et al.*, 2003). A now, infections of CA-MRSA has been reported to increase in the hospital setting, as well as, the community (Huang *et al.*, 2007, Nakaminami *et al.*, 2018, David *et al.*, 2014 ). An MRSA infection is termed community-acquired (CA-MRSA) and does not require any surgical procedures, hospitalization, or residence in a long-term care facility in the 12 months prior. Indwelling catheters or percutaneous devices and dialysis in the year before hospitalization for less than 48 hours preceding MRSA culture are also unrequired. Finally, the patient needs no prior history of MRSA colonization/infection (Buck *et al.*, 2005). HA-MRSA is most often linked with nosocomial infections and greatly retains aminoglycoside, macrolide, and fluoroquinolone non-beta-lactam antibiotic resistance (Rossato *et al.*, 2020). PVL is gene-deficient and is considered HA-MRSA, SCCmec types I, II, and III (A Nichol, K.; Adam *et al.*, 2019). One of the most important and harmful antibiotic-resistant bacteria, (MRSA), is distinguished by resistance to the beta-lactam antimicrobial Methicillin, penicillin, and oxacillin, and amoxicillin. His is the first case of the prevalence of community-acquired MRSA (CA-MRSA) which, over the past decade, has risen as a result to the unchecked spread of these strains (Bloom *et al.*, 2017). *Staphylococcus aureus* strains (MRSA) have emerged as central to the global unease in public health. With the wide spread of MRSA, Vancomycin became the new therapy of choice. Since the 1980s, Vancomycin has dominated as the medication of choice for the most severe and serious of MRSA infections in several

hospital settings (Nezhad RR *et al.*, 2017). The focus of this study was to evaluate the frequency and the spread of antibiotic resistance in strains of MRSA obtained from patients of the surgical ward.

## METHODOLOGY

### Population and Area Under Study:

This study was carried out at Lady Reading Hospital (LRH) which is one of the largest facilities in the province of Khyber Pakhtunkhwa, Pakistan.

This study involved a total of 146 patients from various surgical wards. Their age ranged from children to adults and both genders were included in the study. Samples obtained from patients with postoperative wound infections and discharging pus from the surgical site. The surgical wards involved in this study included the following: General Surgery, Orthopedic Surgery, Pediatric Surgery, Ear, Nose, and Throat (ENT) Surgery, Intensive Care Unit (ICU), Oral and Maxillofacial Surgery, Gynecology and Obstetrics Surgery

### STUDY DESIGN:

The present study was designed as a prospective, cross-sectional, laboratory-based investigation and performed on 146 patients with surgical wound infections in surgical wards of Lady Reading Hospital Peshawar Pakistan.

### SAMPLING AND ISOLATION PROCEDURE:

#### Sample Collection:

The specimens in this study were wound swabs and pus discharges collected from patients presenting with clinical signs of infection, such as abscesses, inflamed surgical wounds, or purulent discharges. Samples were collected using sterile cotton swabs moistened with sterile saline and were carefully rolled over the infected area to obtain adequate microbial load. In the case of deep wounds or abscesses, pus was aspirated using a sterile syringe to avoid contamination with skin flora. All patients were informed about the aseptic collection process, and proper personal protective equipment (PPE)

was used by the healthcare workers. The swabs were immediately placed in sterile transport media and labeled accurately before being transported to the microbiology laboratory within 1-2 hours of collection. If immediate transport was not possible, samples were refrigerated at 4-6°C for no more than 24 hours, in line with standard protocols (Kateete DP *et al.*,2010).



Figure 3.1 labeled samples isolated from surgical wound swab.

#### Culture and Isolation:

As soon as the laboratory was reached, all samples were cultured within the next two hours. Specimens were inoculated onto Cysteine Lactose Electrolyte Deficient (CLED) Agar as well as Mannitol Salt Agar (MSA) plates for the isolation of the *Staphylococcus aureus*. The obtained plates were kept for incubation at 37°C for 18-24 hours under aerobic conditions. Suspected *S. aureus* colonies (golden yellow,  $\beta$ -hemolytic) were further confirmed by Biochemical test. MRSA strains were identified using cefoxitin (30  $\mu$ g) disc diffusion as per CLSI guidelines (Adam *et al.*, 2019).

#### Culturing of Specimens (Wound Swabs and Pus):

Clinical samples, including wound swabs and pus specimens, were cultured under aseptic conditions inside a laminar flow hood. The samples were streaked onto previously solidified media (MSA and

CLED agar). This was done to isolate and identify pathogenic organisms, particularly *Staphylococcus aureus*. Each specimen was labeled with patient information, including ID number, age, sex, date, and time of collection. After inoculation, the culture plates were incubated at 37°C for 18–24 hours to allow bacterial growth and colony formation (Cheesbrough et al., 2006; Tille et al., 2017).

#### Identification of *Staphylococcus aureus*:

After inoculation using the streak plate method with a wound swab, the plates were incubated at 37°C for 18–24 hours. Colonies showing yellow coloration of the medium indicated mannitol fermentation, a presumptive characteristic of *S. aureus* (Forbes et al., 2007).

#### Further Confirmation using the Biochemical tests:

The combination of mannitol fermentation, positive catalase and coagulase tests, and typical Gram stain morphology confirmed the identity of *Staphylococcus aureus* (Tille et al., 2017).

#### 3.9.2.1 Gram Staining:

Bacterial morphology is determined by Gram staining. Gram-positive bacteria are 90% coated in cellulose and a thick covering of peptidoglycan. Gram-positive bacteria exhibit crystal violet or purple coloration following the gram staining procedure, and acid alcohol did not decolorize them. Gram-negative bacteria have lipids and a thin coating of peptidoglycan covering 10% of their cell walls. After staining, Gram-negative bacteria had a red or pink color. Acid alcohol decolorizes these bacteria, which are then stained with crystal violet. When counter-stained with neutral red or safranin, the stain becomes red or pink. A sterile loop is used to select the colony, and a smear is created on a glass slide, let too dry in the air, and then fixed with heat. Crystal violet was used as a basic stain for 30 to 60 seconds on glass slides that had smears on them. Water may be used to clean the slide. On the slide, the iodine solution was applied for 30 to 60 seconds. With the aid of water, the slide was removed. Gram-positive bacteria maintained their blue hue whereas gram-negative bacteria were completely decolorized after a few seconds of acid alcohol

treatment. The slide was once more removed with the aid of water. For 120 seconds, either safranin or neutral red stain was used. On the other hand, the slide was cleaned using water and tape. The slide was examined under a microscope using an oil immersion at a 100x magnification after it had air dried (Rohde, 2019).

#### **Catalase Test:**

This evaluation differentiates those staphylococcus species which possess the catalase enzyme from those streptococcus species which do not possess it. A drop of 3% hydrogen peroxide was added to a colony, and immediate bubbling indicated a positive result (Cheesbrough et al., 2006).

#### **Coagulase Test:**

The tube coagulase test is the gold-standard method for detecting free coagulase produced by *Staphylococcus aureus*, which converts fibrinogen in plasma into a fibrin clot (Microbe Online, n.d.). Free coagulase enzyme secreted by *S. aureus* reacts with the coagulase-reacting factor (CRF) in plasma to form thrombin, which catalyzes the conversion of fibrinogen to fibrin, resulting in clot formation (Rakotovoao-Ravahatra, Randriatsarafara, Ranaivosoa, Rakotovoao, & Rasamindrakotroka, 2019).

#### **Antimicrobial Susceptibility Testing:**

To determine the resistance and sensitivity patterns of *Staphylococcus aureus* isolates against selected antibiotics, antibiotic susceptibility testing was conducted by the *Staphylococcus Bacteremia Study*. Craftsman and Associates's reports on the Standard and Guidelines Followed by the National Committee for Clinical Laboratory Standards, were adhered too (Wayne, PA: CLSI; 2023).

#### **Preparation of Bacterial Culture for Antibiotic Sensitivity:**

From pure culture, bacterial culture suspension was made. A sterile test tube was taken, numbered, and filled with one to two milliliters of regular saline. Colonies from a pure bacterial culture were selected using a sterilized wire loop and moved to a test tube filled with broth to create a bacterial

suspension. For identification purposes, plates containing media (MHA) were labeled when the bacterial suspension was ready. A sterile cotton swab and bacterial suspension were taken. The swab was dipped into the tube that held the bacterial culture suspension, and the swab was pressed against the tube walls to remove the fluid. The saline suspension solution was smeared across the plate. For the bacteria to proliferate uniformly, consistent streaking was carried out (Patra, Das, Das, & Thatoi, 2020).

#### Placing the antibiotic disks on the agar surface

Individual antibiotic disks were applied on the agar surface of the plate with disc dispenser, or sterile forceps. The disks were lightly pressed down to ensure full coverage on contact with the agar. Disc spacing was kept at a minimum distance of 25 mm. Typically, about six discs were placed on 90mm plates. The plates were then sealed up and incubated overnight at 37°C and inverted. After incubation for 18-24 hours at 37 degrees Celsius, we checked the plates for the presence of inhibition zones connected to each antibiotic disc. Using a ruler, each zone was assessed and measured. The results obtained were analyzed according to the Clinical and Laboratory Standards Institute (CLSI) chart. The interrogated specimen was categorized through the sensitive, intermediate, and resistant classification according to the zone size and through the Kirby-Bauer disk diffusion method (Bauer AW et al., 1966).

Table 1: Antibiotic Disks Used for Susceptibility Testing of Staphylococcus aureus:

S. No.	Antibiotic	Abbreviation	Potency (µg)
1	Vancomycin	VA	30ug
2	Linezolid	LZD	30ug
3	Clindamycin	DA	2ug
4	Fusidic Acid	FD	10ug

5	Cefoxitin	FOX	30ug
6	Levofloxacin	LEV	5ug
7	Ciprofloxacin	CIP	5ug
8	Erythromycin	ERY	15ug
9	Clarithromycin	CLR	15ug
10	Doxycycline	DO	30ug
11	Moxifloxacin	MOX	5ug

## RESULTS

### Study Area and Demographic Characteristics:

The present research was carried out at Lady Reading Hospital, Peshawar, which is one of the largest hospitals in that district that serves a extensive population across Khyber Pakhtunkhwa and attached areas. Total 146 clinical samples were collected, comprising wound and pus swabs, were collected from patients admitted in the surgical wards. These specimens provided valuable insights into the prevalence of bacterial pathogens, with a particular focus on MRSA and their antibiotic susceptibility patterns within the hospital setting.

### Sample Distribution:

Samples were collected from a total of 146 hospitalized post-surgical patients admitted to the surgical wards of Lady Reading Hospital, Peshawar. All specimens were collected aseptically using sterile swabs from the infected surgical sites and immediately transported to the microbiology laboratory. The samples were then cultured under standard conditions to identify bacterial isolates, with particular emphasis on detecting MRSA and assessing their antibiotic susceptibility patterns.

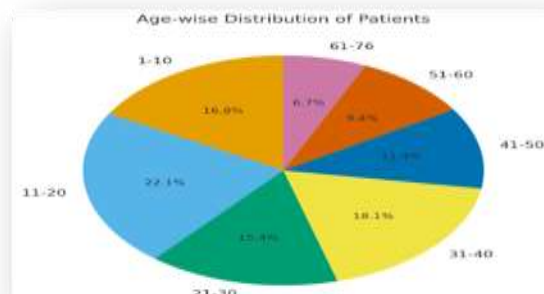


Figure 4.1 age-wise distribution of patients

Out of a total of 146 patients, 114 (78.08%) cases were culture-positive while 32 (21.91%) showed no bacterial growth. Among the positive cases, 103 (70.54%) were identified as MRSA and 11 (7.53%) as MSSA. The study population consisted of 79 (54.10%) male and 67 (45.89) female patients with ages ranging from 1 to 76 years, from whom 125 (85.61%) wound swabs and 21(14.38%) pus swabs were collected for microbiological analysis. This distribution highlights the predominance of MRSA among post-surgical infections and provides an overview of the patient demographics and sample types included in the study.

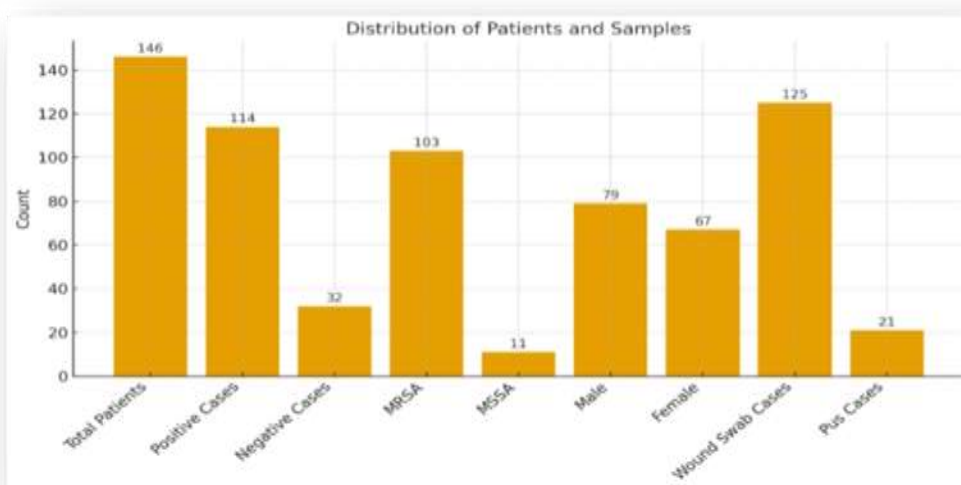


Figure 4.2 Distribution of patients and samples.

**Morphological Character of Staphylococcus aureus:**

The identified bacterium belongs to the species *Staphylococcus aureus*, which is a Gram-positive organism. Upon staining, the cells appeared purple in color, confirming their Gram-positive nature due to the thick peptidoglycan layer in their cell wall. Morphologically, the cells were observed as spherical cocci, which is a typical feature of staphylococci. Under the microscope, the cocci were arranged in a grape-like cluster, reflecting their mode of binary fission in multiple planes. These morphological features are classical identifiers of *S. aureus* and play a critical role in distinguishing it from other related species.

**Table 4.1 Gram Staining Result of Staphylococcus aureus:**

Feature	Observation / Result	Interpretation / Notes
Species	<i>Staphylococcus aureus</i>	Identified pathogen from surgical samples
Size	0.5–1.5 $\mu\text{m}$ in diameter	Small spherical cells
Shape	Spherical (cocci)	Consistent with staphylococcal morphology
Color (Culture)	Golden yellow colonies	Typical pigmentation of <i>S. aureus</i>
Arrangement	Cluster / grape-like	Classic arrangement under the microscope
Gram Staining Color	Purple	Gram-positive organism

The isolated species was identified as *Staphylococcus aureus*, a Gram-positive bacterium frequently associated with surgical site infections. Culturally, it produced golden yellow colonies and appeared as spherical cocci measuring 0.5–1.5  $\mu\text{m}$  in diameter. Microscopically, the cells were arranged in grape-like clusters, which is a characteristic feature of this organism. Gram staining confirmed its Gram-positive nature, as the cells retained a purple coloration.

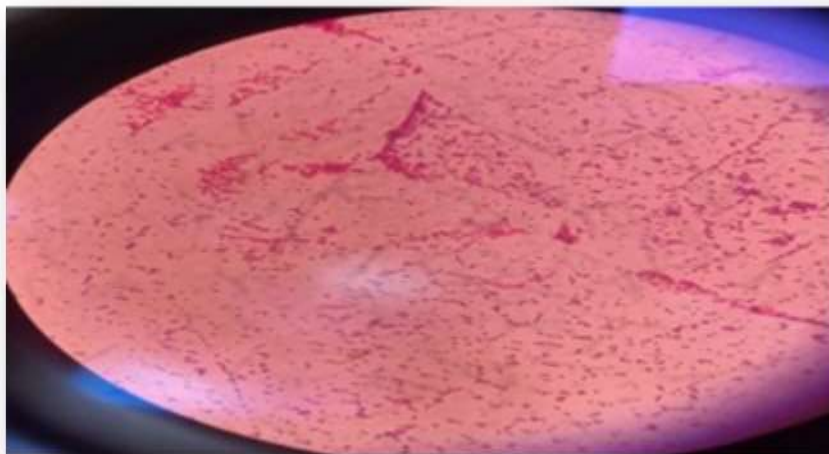


Figure 4.3: Gram Staining Result of Staphylococcus aureus under microscope.

#### Biochemical Confirmation of Staphylococcus aureus:

Biochemical confirmation was achieved through catalase and coagulase testing. The catalase test showed bubble formation on the slide, indicating the release of oxygen, while the coagulase test demonstrated clot formation, confirming its pathogenic potential. These cultural, morphological, and biochemical characteristics collectively validated the identification of the isolates as Staphylococcus aureus.

Table 4.2 Biochemical Results of Staphylococcus aureus

Feature / Test	Observation / Result	Interpretation / Notes
Catalase Test	Positive	Bubbles formed due to oxygen release
Coagulase Test	Positive	Clot formation observed due to presence of an enzyme called coagulase.

### Biochemical Test Results of *Staphylococcus aureus*:

#### Catalase Test Result:

The biochemical characterization of the isolates confirmed their identity as *Staphylococcus aureus*. The catalase test was performed by adding hydrogen peroxide to the culture, which resulted in the immediate formation of bubbles. This positive catalase reaction indicated the presence of the catalase enzyme that breaks down hydrogen peroxide into water and oxygen, a characteristic feature of staphylococci.



Figure 4.4 Catalase test (positive) result for *Staphylococcus aureus*.

#### Coagulase Test Result:

In addition, the coagulase test showed a positive result, as visible clot formation occurred due to the action of the coagulase enzyme on plasma. This reaction is a key diagnostic marker that differentiates *S. aureus* from other coagulase-negative staphylococci (CoNS). Together, these biochemical test results strongly support the identification of the isolates as *Staphylococcus aureus*.



Figure 4.5: Coagulase test (positive) result for *Staphylococcus aureus*.

#### Anti-Biogram Analysis:

The antibiotic sensitivity and resistivity were measured against *Staphylococcus aureus* for 11 different antibiotics (Table. 5). MHA media was used. The percentage-based analysis of antibiotic susceptibility revealed that *Staphylococcus aureus* isolates showed the highest sensitivity to Vancomycin 100% and Linezolid 96.49%, making them the most reliable treatment options. Moderate sensitivity was observed for Clindamycin 57.01% and Fusidic Acid 57.01%. In contrast highest resistivity was observed against Cefoxitin 90.35%, and then resistivity was observed for Erythromycin 77.19%, and fluoroquinolones such as Ciprofloxacin 71.05%, Levofloxacin 71.92%, and Moxifloxacin 70.17% (all above 50% resistance). These findings highlight the alarming rise in resistance among commonly used antibiotics and reinforce the need to rely on Vancomycin and Linezolid for managing MRSA-related surgical infections.

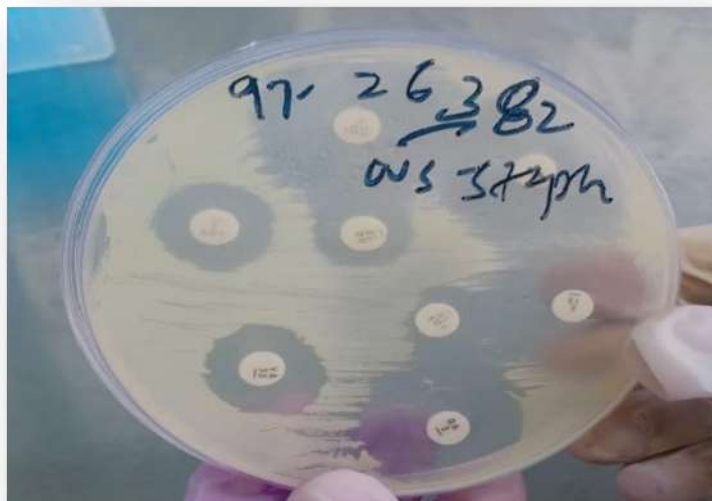


Figure 4.6 Antibiotic susceptibility testing showing zone of inhibition.

Table 4.3 Antibiotic Sensitivity and Resistance Pattern of Staphylococcus aureus (n = 114) positive cases:

Antibiotic	Sensitive (n)	Sensitive (%)	Resistant (n)	Resistant (%)
Clindamycin (DA)	65	57.01%	49	42.11%
Cefoxitin (FOX)	11	9.64%	103	90.35%
Erythromycin (ERY)	21	18.42%	88	77.19%
Doxycycline (DO)	64	56.14%	50	43.85%
Vancomycin (VA)	114	100%	0	0%
Clarithromycin (CLR)	24	21.05%	84	73.68%
Linezolid (LZD)	110	96.49%	4	3.50%
Fusidic Acid (FD)	65	57.01%	43	37.71%
Levofloxacin (LEV)	28	24.56%	82	71.92%
Moxifloxacin (MXF)	27	23.68%	80	70.17%

Ciprofloxacin (CIP)	29	25.43%	81	71.05%
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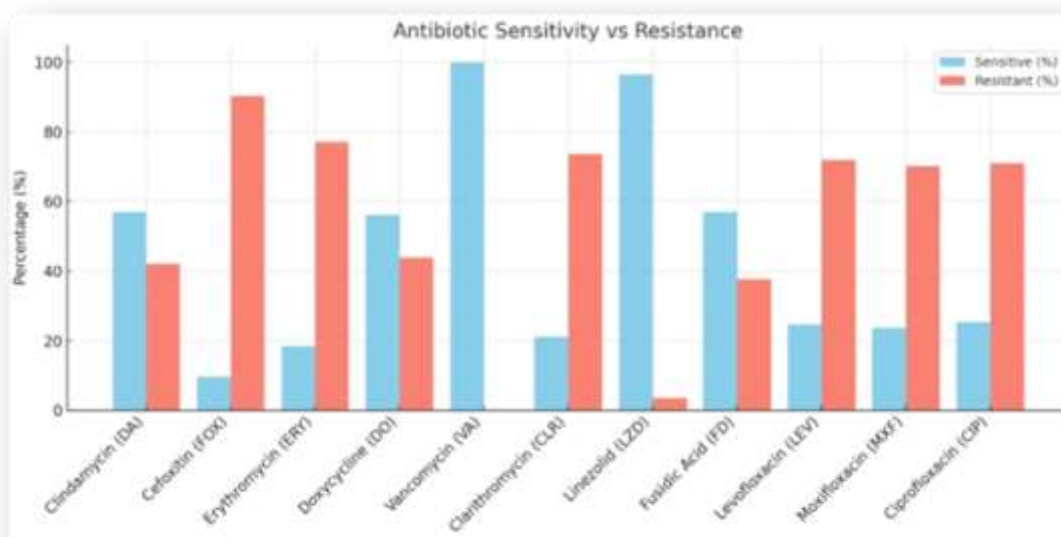


Figure: 4.7 Antibiotic sensitivity vs resistance.

DISCUSSION

One of the main reasons for hospital-acquired infections, particularly in developing nations, is MRSA. MRSA is becoming a more widespread issue in healthcare institutions these days (Darboe S et al., 2019). Staphylococcus aureus was found in 18% of surgical site infection (SSI) cases that were reported to the National Healthcare Safety Network (NHSN) between 2015 and 2017, with methicillin-resistant S. aureus (MRSA) accounting for 53% of these infections (Weiner-Lastinger et al., 2020). When compare to surgical site infections (SSIs) produced by other bacteria, those caused by methicillin-resistant Staphylococcus aureus (MRSA) are associated with greater death rates, longer hospital admissions, and higher healthcare expenses. As a result, healthcare professionals are extremely cautious about the possibility of SSIs linked to MRSA (Weigelt, JA et al., 2020). The frequency of antibiotic resistance in S. aureus isolates, which frequently calls for the adoption of more advanced treatment options, further emphasizes the infectious nature of these bacteria (Klevens RM et al., 2007). Methicillin Resistant Staphylococcus Aureus (MRSA) bacteria are

resistant to a wide range of antibiotics, especially those in the beta-lactam class, they pose a significant clinical concern. Methicillin, oxacillin, and ceftazidime are among them; these are all commonly used in laboratory testing to identify methicillin resistance (Wayne et al., 2017).

Ceftazidime is one of these antibiotics, and because it can cause high expression of the *mecA* gene and clearly distinguish between resistant and susceptible isolates, it has been utilized extensively as a phenotypic detecting reagent for the identification of MRSA. Following several reports of ceftazidime resistance in Pakistan, the ceftazidime disc diffusion method has emerged as a standard diagnostic microbiology technique for accurately identifying MRSA isolates (Ullah, A., Qasim, 2016). In terms of clinical significance, MRSA is more important than beta-lactam resistance. Treatment options for these strains are getting more restricted, and they are frequently resistant to many drugs. Their resistance to lincosamides like clindamycin, fluoroquinolones, amino glycosides, and macrolides like erythromycin in addition to beta-lactam antibiotics, suggests that they have adapted to other antimicrobial processes (David & Daum 2010). The current investigation carried out on MRSA isolated from patients hospitalized in the surgical departments of Lady Reading Hospital Peshawar. Over the period of time three months, from July to September 2025. The research study was conducted purely for isolation and identification of MRSA from surgical wound infections and to check its susceptibility towards different antibiotics. Total 146 sample of wound and pus swabs were collected from patients hospitalized in the surgical departments of both genders, male and female with ages ranging from 1 to 76. All these samples were processed under Standard laboratory techniques. Out of which 114 (78.08%) samples were identified with positive, in which 103(70.54%) were MRSA isolates and 11(7.53%) were MSSA isolates. The percentage of infected males 54.10% was found greater than females 45.89% from whom 125 (85.61%) pus swabs and 21(14.38%). There is no gender-specific restriction on the spread of *Staphylococcus aureus* infection. While some researches have revealed no significant correlation between gender and the risk of MRSA infection, other studies have found that the illness is more prevalent in young people, and still other studies have suggested that the elderly are more likely to contract it (Park DC et al., 2008, Gopal Rao G et al., 2007). Despite the possibility of isolated MRSA in different places, the percentage report varies

slightly because of regional, cultural, health, and hospital referral type variances. However, every finding points to the high frequency of MRSA in clinical samples.

The study revealed a high level of antibiotic resistance against the cephamycin class of antibiotics, which are often categorized alongside second-generation cephalosporins, with resistance to Cefoxitin observed in 90.35% of isolates. In other studies, Cefoxitin resistance was high (70.54%) in *S. aureus* isolates, according to antibiotic susceptibility testing, suggesting a significant prevalence of MRSA. The majority of isolates also showed resistance to  $\beta$ -lactam antibiotics, which made them not very effective. These results are consistent with worldwide evidence, which indicates that *S. aureus* usually exhibits substantial penicillin resistance while being consistently vulnerable to vancomycin (Adhikari et al., 2017). On the other hand, *Staphylococcus aureus*'s antibiotic resistance profile in our study showed 100% sensitivity to Vancomycin, an antibiotic belonging to the glycopeptide class. Interestingly, none of the isolates in our investigation showed signs of Vancomycin resistance. It is crucial to educate healthcare professionals on MRSA and carriers' status in order to lessen its effects in hospital environments. Reducing the burden of MRSA transmission in hospital settings requires the consistent application of infection control measures (Holmes et al., 2005). Antibiotic resistance rates of 71.92% for levofloxacin, 70.17% for moxifloxacin, and 71.05% for ciprofloxacin were notable against the fluoroquinolone class of antibiotics (all above 50% resistance). In other study the moxifloxacin resistance of MRSA clinical isolates was assessed in relation to their genotype. Resistance to fluoroquinolones was more common than previously thought (Hashem, R. A et al., 2013, Mohamed, N. M. et al., 2019). The study found that 73.68% patients were resistant to clarithromycin and 77.19% were resistant to erythromycin, indicating a significant rate of resistance to macrolide antibiotics. Furthermore, 42.11% of isolates showed resistance to the lincosamide class of antibiotics, drug Clindamycin. Some other study said, Staphylococcal bacteria that were insensitive to macrolides first surfaced a few years after the antibiotics were introduced into the treatment. Macrolide resistance is now commonplace globally, and many bacteria are resistant to MLSB medicines (van Hoek, A.H.A.M et al., 2011). Due to their widespread usage in the treatment of Gram-positive bacterial infections, staphylococcal clinical strains are becoming less sensitive to

macrolides, which is typically linked to resistance to lincosamides and streptogramins B (Yao, W, Xu, G et al., 2019).

In the present study, resistance to the tetracycline class of antibiotics was observed in 43.85% of isolates for doxycycline. In other small cohort study the use of doxycycline for MRSA-related SSTIs showed little variation in treatment failure rates between isolates that were susceptible to tetracycline and those that were resistant to it. These results imply that even within tetracycline-resistant MRSA bacteria, doxycycline susceptibility might be maintained (Ashly Nham, et al., 2025). As per our investigation, 3.5% of MRSA isolates shown resistance to the antibiotic linezolid, which belongs to the oxazolidinone class. In previous study according to Khanam et al. in Bangladesh and Mamtora et al. in Mumbai, linezolid resistance was found in 2.6% and 2% of MRSA isolates, respectively, and our results are in agreement with their findings (Mamtora D et al., 2019, Khanam S, Haq JA et al., 2016). Resistance pattern of fusidic acid is 37.71% as per our study. In some other study of 2021, the average prevalence of fusidic acid resistance was estimated to be around 5% worldwide (Hajikhani, Goudarzi M et al., 2021). The differences observed in the frequency of isolation, specimen-wise distribution, and antimicrobial resistance patterns between this study and other related reports may be attributed to variations in gender, time period, geographic location, environmental conditions, educational and socioeconomic status, hygiene practices, availability of medical facilities, as well as methods of specimen collection and handling.

## CONCLUSION

In addition to the surgical wound swabs from the patients under observation, the study included the recovery and analysis of methicillin-resistant *Staphylococcus aureus* (MRSA). The objective of the study was to perform MRSA detection and determine patterns of antimicrobial susceptibility. The data collected demonstrated that the prevalence of MRSA along with surgical site infections poses severe public and surgical health problems. The existence of MRSA within surgical sites further demonstrates the lack of sufficient infection control and the ability of MRSA to causatively associate MRSA with disease. MRSA with antimicrobial susceptibility antibiogram demonstrates a class of

*staphylococci* of considerable clinical and other antibiotics resistance. The existence of multi antibiotic resistant MRSA strains indicates the need of more intensively developed and promptly executed policies of antibiotic stewardship on the balance of antibiotics used. The findings of this study once more prove that the lack of restraint and overly liberal policies on the administration of broad-spectrum antibiotics to patients, regardless of health being in the community or a health facility, has tremendously worsened the problem of resistance. The conclusion of this study emphasizes the necessity of policy reform to inappropriately and excessively limit antibiotic prescriptions. MRSA is a growing issue in surgical wards and surgical units of Pakistan. Policies in this case need timely monitoring of infection control practices, multidisciplinary control, training of practitioners, and community advocacy. This work deals with MRSA in Pakistan, focusing on surgical site infection and the need to address it with intensive clinical, laboratory, and epidemiology.

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