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Prevalence of Methicillin Resistant Staphylococcus Aureus in Pus Sample

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Abstract
Methicillin-Resistant Staphylococcus aureus (MRSA) has become a significant problem worldwide, causing infections in both hospitals and the wider community. In Pakistan, MRSA is a serious public health issue, especially when it involves skin and wound infections. This study looked at how common MRSA is in pus samples and examined how it resists common medications. The aim of this research is to find out how frequently MRSA occurs in pus samples drawn from clinical sources. A descriptive cross-sectional study was conducted in selected healthcare facilities in Lahore. A total of 250 pus samples were collected from patients presenting with wound or skin infections. The samples were processed for the isolation and identification of *Staphylococcus aureus* using blood and MacConkey AGAR. Methicillin resistance was determined by antimicrobial susceptibility testing performed using the Disk diffusion method on Muller Hinton agar according to guideline of CLSI. Out of 250 clinical samples analyzed, 49% tested positive for Methicillin-Resistant *Staphylococcus aureus* (MRSA). Antibiotic susceptibility testing was performed on 55% of the samples. Among the tested isolates, resistance was observed to Vancomycin (31%), Methicillin (24%), and Ceftriaxone (22%). The highest resistance was noted for Vancomycin. Gender-wise distribution showed a slightly higher infection rate in males (54%) compared to females (46%). The increasing number of MRSA cases found in Staphylococcus aureus is a serious health issue that we need to address. This situation indicates that crucial medicines like methicillin and vancomycin are becoming less effective against these infections. Therefore, it is vital to conduct regular tests and be cautious with the use of antibiotics. Additionally, there is a need to enhance laboratory testing methods since not all samples are being examined. To combat the spread of MRSA, we must focus on improving infection control practices, using antibiotics wisely, and continuously monitoring the situation. By taking these steps, we can better manage and reduce the risks associated with MRSA.

Key words: Staphylococcus Aureus, Antibiotic Resistance, Vancomycin, Methicillin, Ceftriaxone

Introduction

A lethal and versatile bacteria, Staphylococcus aureus may cause a variety of diseases. It most frequently leads to respiratory infections and skin disease. Skin disease is usually contracted in the community, but most nosocomial S. aureus infection is lung infection. S. aureus is the most common infection people get in hospitals, and it can make people very sick or be life-threatening. This bacterium often causes pneumonia in patients already in the hospital for other reasons, particularly if they have weak immune systems or viral infections. S. aureus is responsible for other serious illnesses too. It can

cause infective endocarditis, affecting the heart, and osteomyelitis, affecting the bones. It may also lead to scalded skin syndrome and toxic shock syndrome, which can sometimes be fatal. Occasionally, S. aureus is linked to very severe diseases such as necrotizing fasciitis, which destroys tissue beneath the skin, and necrotizing pneumonia, a severe infection of the lungs.[1] Staphylococcus aureus is a type of round-shaped bacteria that doesn't move by itself and has a thick cell wall, identified as Gram-positive. It belongs to the larger group called Firmicutes. Among all the bacteria in the Staphylococcus family, which includes 52 species and 28 subspecies, S. aureus is particularly important for medicine and healthcare. This bacterium is often found in the noses of about 20–40% of people, living there harmlessly as part of the normal mix of microbes in that area.[2] Antibiotic resistance is a big concern around the world today. Staphylococcus aureus is a common kind of bacteria causing infections in hospitals and communities. It is known as a Gram-positive cocci, referring to its shape and test reaction. A resistant form of this bacteria called Methicillin-resistant Staphylococcus aureus (MRSA) is tough to treat because it's not affected by most standard antibiotics. MRSA developed this resistance in the UK just two years after methicillin was first used. Because of this resistance, doctors usually rely on vancomycin and other glycopeptide antibiotics to successfully treat MRSA infections.[3] MRSA is a type of bacteria resistant to oxacillin when levels are above 4 μg/mL. This resistance occurs because oxacillin is part of β-lactam antibiotics, which usually work by blocking proteins called PBPs in bacteria. These proteins are important for bacteria to survive. If an antibiotic sticks well to PBPs, it's good at fighting the bacteria. But MRSA has a special gene called mecA. This gene makes a different kind of PBP that doesn't hold onto the antibiotic well. So MRSA keeps growing even when treated with methicillin and similar antibiotics. All MRSA strains produce this special PBP, known as PBP2a, which helps some cells avoid being killed by these drugs.[4] In 2000, a type of methicillin-resistant Staphylococcus aureus (MRSA) called USA300 was first discovered in the United States. This strain, also known by specific codes like multilocus sequence type 8 and clonal complex 8, as well as staphylococcal cassette chromosome mec type IV, became known for causing skin and soft tissue infections. These infections were initially found among football players in Pennsylvania and also among inmates in Missouri prisons. While USA300 mainly causes infections on the skin and in soft tissues, it can also lead to more severe health problems. These include a serious lung infection called fulminant necrotizing pneumonia, a bone infection known as osteomyelitis, an infection of the bloodstream known as bacteremia, and an infection of the heart lining called endocarditis.[5] Staphylococcus aureus (SA), which belongs to clonal complex 398 (CC398), is important because it is found everywhere in the world. There are two main types of SA CC398. The first type, methicillin-susceptible SA (MSSA), is sensitive to methicillin and is usually found in humans. The second type, methicillin-resistant SA (MRSA), is resistant to methicillin and is often found in livestock. In this discussion, we are focusing on MSSA CC398. This type of SA is often linked to serious health problems like endocarditis, infections in the bloodstream, and issues with bones and joints. Even though the presence of MSSA CC398 is increasing around the globe, the number of cases is not the same everywhere. MSSA CC398 is often found to resist erythromycin, an antibiotic.[6] Vancomycin resistance was first found in the 1950s, but reports of vancomycin-intermediate S. aureus (VISA) and heterogenous-VISA (hVISA) did not appear until the mid-1990s. Scientists have struggled to understand the complex gene and cell wall changes that cause S. aureus to develop low-level resistance to vancomycin. VISA seems to have gradually developed from vancomycin-susceptible S. aureus (VSSA). Research reveals that the transformation from VSSA to VISA is linked not just to antibiotic resistance but also to other changes that may help the infection persist. Studies suggest that hVISA and VISA represent stages in the evolution of bacteria that improve their ability to survive against medical treatments and conditions inside the human body. Research into hVISA and VISA has provided important new insights into how staphylococcus bacteria function and grow. These studies have helped

us understand more about the biology of this bacteria.[7] Vancomycin is considered the top medicine for treating infections caused by meticillin-resistant Staphylococcus aureus (MRSA). However, in 1996, a vancomycin-resistant MRSA was found in Japan. The patient had a surgical wound infection that didn't improve with extended vancomycin treatment. This problem isn't just in Japan; vancomycin-resistant Staphylococcus aureus (VRSA) has been found in the USA, France, Korea, South Africa, and Brazil, showing it's a global issue. There's a specific type of S. aureus called hetero-VRSA. When it is exposed to vancomycin, it often becomes VRSA. Infections from hetero-VRSA might not respond well to vancomycin, which suggests that this medicine is gradually becoming less effective in hospitals.Vancomycin resistance occurs when bacteria change and their cell walls become thicker due to having too much peptidoglycan. This type of resistance is seen in all VRSA samples collected worldwide up to now.[8] Toxins that result in toxic shock syndrome is most common in hospitalised and disabled individuals. Healthier patients with a good immune system are mostly the ones who develop toxic shock syndrome due to either Staphylococcus aureus or Streptococcus pyogenes. Both the infections share some similarities, such as shock in early stages and organ failure, but they also show a great deal of variation based on clinical features and symptoms, morbidity, and mortality. S. aureus and S. pyogenes both infect humans with a range of distinctive infections which may or may not be related to toxic shock syndrome.[9] Staphylococcus aureus is a leading cause of infections both in hospitals and among the general public. A major challenge in treating these infections is the bacteria's ability to resist methicillin, which means it won't respond to any β-lactam antibiotics. When this resistance occurs, it's referred to as MRSA, short for methicillin-resistant Staphylococcus aureus. MRSA infections are increasingly common and difficult to treat due to their resistance to multiple drugs. The bacteria can also spread beyond hospitals, leading to outbreaks in the community. This makes it crucial to implement strong infection control measures. Therefore, studying how these infections spread and understanding the bacteria are vital.[10] Staphylococcus aureus, or S. aureus, becomes resistant to methicillin because it makes a special protein known as PBP2a. This protein is not the same as another protein called PBP2. PBP2a can take over the role of linking parts of the bacterial cell wall, a job typically done by the host's PBPs, and it can do this without being affected by methicillin. Usually, the amount of PBP2a made by the bacteria is low because its production is carefully controlled. However, if there are changes in the genes that regulate this process, the bacteria might start producing more PBP2a, increasing its resistance.[11] Around 25% of people have a common bacterium called Staphylococcus aureus, or S. aureus, in the front part of their nose. S. aureus is known for causing many different infections. Some of these infections are mild and include things like pimples, impetigo, boils, cellulitis, inflamed hair follicles, carbuncles, scalded skin syndrome, and abscesses.[12] However, it can also lead to more serious conditions like pneumonia, meningitis, bone infections (osteomyelitis), heart infections (endocarditis), blood infections (bacteremia), and sepsis. S. aureus is the most aggressive type in the Staphylococcus family. It can cause severe issues such as toxic shock syndrome (TSS) because of a specific toxin it releases. S. aureus is frequently responsible for infections after surgeries because it can resist many antibiotics used in medical settings.[13] MRSA is a big issue worldwide, and it's getting worse. It affects between 23.3% and 73% of people, making it the leading cause of infections in the blood, lungs, and skin. The Centers for Disease Control and Prevention report that in the U.S., MRSA accounts for 59.5% of all healthcare-related S. aureus infections. Additionally, MRSA infections are on the rise in India.[14] Pakistan faces a significant challenge with MRSA, a bacteria resistant to many antibiotics. A study conducted in Islamabad revealed that 39% of individuals had MRSA. Specifically, 35% of patients in surgical wards and 65% in intensive care units were affected. Over the years, various cities have reported different rates of MRSA. In 1999, Sargodha had a rate of 23%, while in 2001, Lahore reported 38.5%. By 2005, Karachi's rate was 43%, Rawalpindi showed 50% in 2014, and between 2015 and 2017, Karachi again

had a rate of 52%. Our own study discovered an even higher rate of 66.03%. In five major government hospitals in Rawalpindi, MRSA was found in 60.40% of patients, as reported by Parveen et al. Another study indicated that the rate in two significant hospitals in Rawalpindi and Islamabad was 53.3%. The National Institute of Health in Islamabad reported a high incidence of 71.1% in 2015. In contrast, Peshawar had much lower rates, between 1% and 5.2%, during 2012-2013. Worldwide, MRSA is becoming more common, though rates vary. Europe has a rate of 8.7%, the US 6%, and Japan 2.6%. These figures are lower than the 13% incidence observed locally. Among healthcare workers, MRSA was found in 10.5% of nurses, 3.8% of doctors, 1.9% of paramedics, and 3.6% of administrative staff. These local rates are relatively high compared to international figures, with MRSA being more prevalent in intensive care unit staff in larger hospitals. Among healthcare workers, 13% had MRSA, with the highest rate among nurses at 8.69%. The regimen for reducing MRSA in carriers proved to be 83.33% effective.[15] When S. aureus gets into an open wound, it often causes infections. However, if there is a viral infection in the upper airways, it can damage the mucosal linings and potentially lead to S. aureus pneumonia about a week after the flu begins. When S. aureus first reaches areas other than skin or mucosal surfaces, its harmful genes start to work more actively. The body's immune system responds to this threat. It is triggered by special skin and mucosal cells reacting to bacterial substances or tissue damage. Molecules in the body recognize parts of S. aureus and start inflammation. This process activates local immune cells and attracts white blood cells like neutrophils and macrophages to fight the infection.S. aureus can survive both inside and outside body cells. The bacteria must defend itself against the body's defenses that use complement and antibodies, which try to destroy or engulf it. S. aureus has a capsule and proteins like clumping factor A, protein A, and other inhibitors that help it avoid being marked and destroyed by the immune system, making it harder for the body to eliminate the bacteria.A specific type, MRSA, is particularly important because of its impact on health. In 2005, MRSA was responsible for around 94,360 serious illnesses and 18,650 deaths in the US, surpassing those caused by HIV. There are two main types of MRSA: HA-MRSA, usually found in hospitals, and CA-MRSA, which often affects people with weakened immune systems or those exposed to healthcare settings. Recently, CA-MRSA has led to an increase in skin and soft tissue infections. HA-MRSA carries genetic elements with antibiotic resistance genes, which make them more challenging to treat.[16] We identified how the infection started in the body. The S. aureus bacteria broke through the body's defenses and got into the bloodstream, which could happen due to an infection or a surgery. We divided these entry points into two main categories 1. Vascular devices 2. Intravenous drug use, infections of the skin and soft tissue, or other types of injections.[17] Its contributing factors are the extensive and unscientific use of antibiotics. Therefore, MRSA has been a major hazard and treatment barrier for laboratory and clinical staff as well.One of the primary sites where MRSA stain spreads and results in epidemic and endemic MRSA outbreaks is hospitals. Other risk factors for MRSA transmission are prolonged hospitalisation, extensive antibiotic use, exposure in nursing homes, immunosuppression, inappropriate dosing of antibiotics, indwelling catheterization, invasive medical devices, drug addicts, and unsterilized instruments. Since MRSA is infectious and difficult to treat, it is a serious health issue in industrialised as well as underdeveloped countries.[18] We collected the samples normally and used a BACTEC 9240 and 9050 BD machine to study bacterial growth after finishing the blood cultures. Samples were placed on special media dishes like Mac-Conkey and Blood agar, among others. These dishes were then kept at 37°C with air for 24 to 48 hours to let bacteria grow. To identify the bacteria, we used common steps such as Gram staining, checking the colonies, and performing tests like the coagulase and catalase tests. To see how bacteria reacted to different antibiotics, we used the Kirby-Bauer disc diffusion method following CLSI guidelines.[19] If someone develops a severe skin infection that involves redness, swelling, or other signs suggesting the infection is spreading, doctors need to investigate if it could be MRSA, which is a type of resistant bacteria. This is especially important if

there have been multiple infections among a group of people, known as an outbreak. In such cases, medical professionals should collect samples from the infected area to identify the bacteria and determine which antibiotics can effectively treat it. Important samples to collect include fluid from skin abscesses, mucus or fluid from the respiratory system, blood from critically ill patients, and material from bones or joints if an infection is suspected there. To detect MRSA in laboratories, traditional methods such as disc diffusion, broth microdilution, and oxacillin agar screen tests are commonly used. These methods are recommended by the National Committee for Clinical Laboratory Standards. However, some research has reported issues with accuracy when using these methods, especially with automated or machine-based systems. MRSA is resistant to several antibiotics, including gentamicin, trimethoprim/sulfamethoxazole, clindamycin, erythromycin, and tetracycline. Fortunately, an antibiotic called vancomycin is generally effective against MRSA. Although MRSA might seem susceptible to certain other antibiotics, like cephalosporins, in laboratory settings, these do not work well against MRSA in practice.[20] The study only emphasized pus and wound specimens. Isolation of MRSA can also be done using other clinical specimens like blood, respiratory secretions, urine, and soft tissue infections. Failure to Analyze Risk Factors: The risk factors for MRSA infections, including prior antibiotic use, hospital stay duration, and immunosuppression, were not extensively studied in this research. Expand the sample size and study horizon: Include other clinical samples (e.g., blood or urine) to derive a broader picture of MRSA's role in hospital-acquired infections. For determining the prevalence of MRSA, conduct a multi-center study with larger sample sizes. Analysis of Risk Factors: Integrate review of risk factors associated with the patient, such as hospital stay length, utilization of invasive devices (e.g., catheters), immunocompromised state, and previous use of antibiotics.

Literature Review

Noshad ali sajid, *et al*., (2022) counducted a study on The study aimed to understand how often Staphylococcus aureus is found in pus culture samples and how it reacts to antimicrobial treatments. From 447 pus samples, 225 (50.4%) showed bacterial growth, with 79 (35.1%) confirmed as Staphylococcus aureus.The study observed high resistance rates to antibiotics, mainly penicillin. Out of 79 samples, 74 (93.6%) were resistant to penicillin, and 15.9% showed resistance to vancomycin, a concerning discovery due to increasing methicillin-resistant strains.Methicillin-resistant Staphylococcus aureus (MRSA) is becoming more resistant to common antibiotics, complicating treatment options. This research focused on both the prevalence of Staphylococcus Aureus and its resistance patterns.The study covered male and female patients of all ages with abscesses at infection sites. Pus samples were collected, but only those with Staphylococcus Aureus were included in the study. Samples without this bacteria were excluded. This study underscores the critical need for continuous monitoring of antimicrobial resistance patterns. Such ongoing analysis is vital to improving treatment decisions and effectively countering resistance challenges.[21] Kajal Parmar, *et al*., (2024) conducted a study on Staphylococcus aureus and Its Antibiotic Sensitivity in Hospital Samples This study focused on how the bacteria MRSA react to different antibiotics and looked for clindamycin resistance. It was conducted from January to December 2022 in the Microbiology Department at SBKS MI and RC.During the research, 1470 samples were collected, such as pus, blood, urine, high vaginal swabs, and sputum. From these, 114 coagulase-positive staphylococci (COPS) were discovered.To identify these bacteria, several tests were used: Gram staining, the Catalase test, Mannitol fermentation, and tube coagulase tests. The tube coagulase test was crucial for identification.The study reveals the serious issue of MRSA in hospital settings and emphasizes the urgent need for proper use of antibiotics to tackle this growing concern.[22] Subash chaudhary, *et al*., (2023) conducted a study on The study at the hospital in Mullana looked into how common a bacteria called Methicillin-Resistant Staphylococcus aureus (MRSA) is and how it interacts with antibiotics. The goal was to find out how

often MRSA and Methicillin-Sensitive Staphylococcus aureus (MSSA) appear in patient samples and how they respond to antibiotics. This information is crucial for choosing effective treatments and preventing the spread of infections. Researchers collected 200 pus samples from patients who visited the clinic or were hospitalized between 2019 and 2020. Out of these, 102 samples, or 51%, were identified as MRSA, showing that this resistant bacteria is quite common among the studied patients. The findings emphasize the urgent need for effective strategies to understand and manage the prevalence of MRSA and its susceptibility to antibiotics. Addressing these issues is vital to combat the increasing threat posed by MRSA.[23] Raghabendra Adhikar, *et al*., (2017) conducted study on MRSA and Vancomycin MIC This study examines Methicillin Resistant Staphylococcus aureus (MRSA) and measures the levels of vancomycin needed to inhibit S. aureus. The research was conducted using samples from pus and wounds at a major hospital in Kathmandu, Nepal. The rise in MRSA cases and its resistance to antibiotics is a serious public health issue, underlining the need for improved detection and treatment approaches. The study aimed to assess the effectiveness of two methods for detecting MRSA: the cefoxitin disc diffusion method and the oxacillin broth microdilution method. The mecA gene served as a reference standard for checking the accuracy of these methods. Researchers collected 711 pus and wound swab samples from various body areas. S. aureus was identified through colony appearance, Gram staining, and specific biochemical tests. Among 110 isolated S. aureus strains, 39 (35.50%) were determined to be MRSA using the cefoxitin disc method, and 31.82% were identified by the oxacillin broth method. The mecA gene was detected in only 29.1% of the MRSA samples. The vancomycin levels needed to stop S. aureus growth ranged from 0.016 µg/mL to 1 µg/mL, indicating different susceptibility levels among the bacteria.[24] Varsha S Puranik, *et al*.,(2024) focused on The study examined how common methicillin-resistant Staphylococcus bacteria are in pus samples from people with skin and soft tissue infections (SSTIs). It showed that 17% of Staphylococcus aureus samples were methicillin-resistant, which are known as MRSA. This indicates a growing issue with antibiotic resistance. During the study, they analyzed 50 pus samples, and 96% had bacterial growth. Of these, 45.83% were Staphylococcus aureus, while 22.91% were Coagulase-negative Staphylococci (CoNS). MRSA was notably found in both men and women aged 25-45 and 45-65, with each group showing a 40% prevalence rate. The study also revealed that MRSA strains are resistant to several types of antibiotics, including β-lactam antibiotics, aminoglycosides, and fluoroquinolones, which complicates treatment. The rise of MRSA and CoNS in hospitals presents major challenges for infection control and treatment planning. Therefore, it's crucial to test for antibiotic resistance before starting treatment, especially in infections that produce pus. This practice helps to manage the infection effectively and reduces healthcare costs by preventing extended hospital stays.[25] Hussaini (2018) conducted study carried out in several hospitals in Sokoto city, researchers examined 230 wound samples. They found that 63 samples contained coagulase positive Staphylococcus aureus, which means approximately 27.4% of the samples tested positive for this infection. The infection rates varied across hospitals. At the Specialist Hospital, the rate was very low, whereas Maryam Abacha Women and Children Hospital had a rate of 27.5%. Notably, Noma Hospital had no positive cases.Among the 63 cases of Staphylococcus aureus, 38 (60.3%) were found to be methicillin-resistant, known as MRSA (Methicillin-Resistant Staphylococcus aureus). A higher percentage of women were affected by MRSA, with 76.7% compared to 45.5% of men. The age group most affected was 41-50 years, with a 100% rate of MRSA infection. The antibiotic tests revealed that MRSA had high resistance to several antibiotics: 100% resistance to Ampicillin, Ceftazidime, and Cefuroxime, and significant resistance to Augmentin. However, resistance was lower to antibiotics like Gentamicin (34.2%), Nitrofurantoin (36.8%), and Mupirocin (36.8%). This high resistance to common antibiotics is alarming and highlights the urgent need for responsible antibiotic use and the development of better infection control methods in hospitals. The study underlines the necessity of ongoing monitoring of MRSA to guide healthcare

professionals in choosing effective treatments.[26] J Ojulong, *et al*., (2008) studied carried out that the research focuses on the common presence of Methicillin-resistant Staphylococcus aureus, known as MRSA, in infections following surgeries at Mulago National Referral Hospital in Kampala, Uganda. The study was carried out from February to May in 2007. Altogether, 188 patients who had either planned or emergency surgeries participated. For each patient, a sample was taken from their infected wound. Out of these 188 samples, 54 were found to contain the bacteria S. aureus. Among these 54 S. aureus samples, 17 were identified as MRSA using a test called PCR. The results show that MRSA infections are highly prevalent in surgical wounds at this hospital. This highlights an urgent need to improve infection prevention methods to tackle this significant health challenge.[27] Osinupebi1 *et al*., (2018) Conduct A study in Abeokuta, Nigeria, investigated how common MRSA is in medical samples. MRSA stands for Methicillin-resistant Staphylococcus aureus, a bacterium hard to treat with regular antibiotics. The study discovered that 41% of the samples contained MRSA, highlighting a major health issue in the region. Over six months, researchers collected 338 samples, including pus, body fluids, ear, and wound swabs. Most samples, about 50.3%, were from male patients. Children aged 0 to 9 were the most affected, with 37.9% having MRSA.The study showed that MRSA rates were similar in different hospitals, indicating that the problem is widespread and not limited to a single location. These findings underscore the urgent need for better infection control in hospitals to safeguard public health. Given the rising MRSA cases, conducting more research and continuous monitoring is crucial to lessen its impact on communities.[28] Bahram khan kho, *et al*., (2022) The study focused on finding out how common Methicillin-Resistant Staphylococcus aureus (MRSA) is among people with skin infections called pyoderma within the community. Conducted with the Dermatology and Microbiology departments at Jinnah Postgraduate Medical Centre in Karachi, the research was carried out between August 1, 2022, and November 25, 2022. Researchers collected 232 pus samples from patients suspected of having pyoderma to examine the bacteria and check their resistance to antibiotics. Out of these samples, 196 (84.5%) showed positive bacterial growth. The main bacteria identified were Staphylococcus aureus with 119 cases (51.2%), Streptococcus pyogenes with 51 cases (22%), and Staphylococcus epidermidis with 3 cases (1.3%). Additionally, some samples had Gram-negative bacteria like Pseudomonas aeruginosa (13 cases) and E. coli. Among the Staphylococcus aureus cases, 73 strains (61.3%) were identified as MRSA. This study showed that many MRSA strains were resistant to different antibiotics such as Penicillin (89%), Cefoxitin (74.7%), and Ciprofloxacin (60.5%). The findings indicated that males were more affected, and the most common age group impacted was adolescents aged 11 to 20 years.[29] Kumari N, *et al*., (2008) study conduct on Methicillin-resistant Staphylococcus aureus, known as MRSA, is a serious issue in hospitals worldwide because it's difficult to treat with standard antibiotics. People can catch MRSA both in hospitals and out in the community, making it a significant global health concern. The cost for treating MRSA infections is high, ranging from $20,000 to $114,000 for outbreaks and as much as $1,600,000 for ongoing infections annually. A study conducted over a year found 750 strains of Staphylococcus aureus, and 196 of these were MRSA. Most cases (70%) came from hospital wards, with only 10% from intensive care units. The study identified several risk factors for MRSA, including the overuse of antibiotics, long hospital stays, use of medical devices inside the body, stays in the intensive care unit, and catching it from people in the community who carry the bacteria.Tests showed that more than 65% of MRSA strains are resistant to common antibiotics. The resistance rates are as follows: 100% for Penicillin, 72.45% for Cefotaxime, 62.24% for Ceftazidime, 67.35% for Ciprofloxacin, 31.63% for Gentamicin, 29.59% for Erythromycin, and 27.55% for Tetracycline. Also, 47.96% of the strains are resistant to Amikacin. Because MRSA is so hard to treat, hospitals must have effective methods to control and prevent it, aiming to decrease its impact on public health..[30] Hamama Islam But, *et al*., (2022)counducted the study in Lahore how common the bacteria Staphylococcus aureus (S. aureus) is

in pus and nasal swab samples from patients. They collected 368 samples from different hospitals to find out how often S. aureus appears and its resistance to antibiotics. They discovered that 90% of the bacterial samples were S. aureus, with a total of 330 cases identified. Most of these bacteria were found in pus samples, with 247 cases (74.84%), while nasal swabs contained 83 cases (25.15%). Other bacteria found included Pseudomonas spp. (8 cases, 2%), E. coli (5 cases, 1%), Streptococcus (1 case, 0.27%), Klebsiella spp. (1 case, 0.27%), and Candida spp. (1 case, 0.27%). There were also 22 samples (6%) that showed no bacterial growth.The study revealed that S. aureus in pus samples showed high resistance to several antibiotics: Clindamycin (23.49%), Erythromycin (21.82%), Amoxicillin (16.59%), Oxacillin (10.12%), Amikacin (8.96%), and Gentamycin (8.92%).The research emphasizes the significant presence of S. aureus in patient samples from Lahore and highlights the worryingly high rates of antibiotic resistance. This calls for urgent action to improve infection control and antibiotic management to combat the growing problem of resistant bacteria in hospitals..[31] Lakshmi, *et al.*, (2013) conducted a study to find out how common Methicillin Resistant Staphylococcus Aureus (MRSA) is in pus samples from a large hospital. The main goal was to see how often MRSA appears and understand its reaction to different antibiotics. This study took place in a hospital in Chennai between August 2012 and January 2013. They collected pus samples from various hospital departments to check for MRSA. A total of 160 pus samples were gathered from patients with different infections, using either sterile cotton swabs or direct needle aspiration. The study results highlighted a strong need for continuous monitoring and strategic actions to tackle the MRSA issue in hospital settings..[32] Dr Tansuri Biswas, *et al*., (2021)counducted a study to find out how often Methicillin Resistant Staphylococcus aureus (MRSA) is found in pus samples at a major hospital in eastern India. This research aimed to see how common MRSA is in these pus samples. The samples were taken from patients in the hospital. The study also checked how these MRSA strains responded to different antibiotics to understand their resistance patterns.The research took place between September 2019 and January 2020. A total of 179 pus samples were collected from patients in different hospital wards. The samples were taken from infected wounds and quickly sent to the lab for further study. The team used nutrient agar and blood agar to grow and study the bacteria.Out of the 179 samples, Staphylococcus aureus was found in 134, which is about 74.86%. Among these, 39 samples, or 29.10%, were identified as MRSA.The study concluded that there is a high rate of MRSA in pus samples. This shows the need for careful antibiotic use and strong infection control strategies..[33] Pradhan P, *et al*., (2021)A study was conducted in a large hospital in Nepal to find out how common methicillin-resistant Staphylococcus aureus (MRSA) is. There were three main goals in this study. The first goal was to determine the number of MRSA cases among people with Staphylococcus aureus infections from January 2018 to December 2020. The second goal was to identify which antibiotics were still effective against MRSA by examining resistance patterns. The third goal was to look at patients' ages, genders, and health details, as well as how treatments for those with MRSA infections turned out during this period.The study found that there are treatments that work well against MRSA, mostly using antibiotics to which MRSA was still sensitive. It pointed out the importance of continuously watching MRSA and its resistance to antibiotics, as it is important for public health.The study included 1,804 patients who were confirmed to have Staphylococcus aureus infections. Out of these patients, 1,027, or 57%, were found to have MRSA.In summary, the study shows an urgent need for better surveillance and infection control in healthcare settings to deal with the increasing MRSA cases. It stresses the necessity of monitoring antibiotic resistance to guide effective treatments and the use of suitable antibiotics.[34] Salma ghulam nabi, *et al*., (2019) conduct The study to understand how common Staphylococcus aureus is and how it responds to antibiotics in a large hospital in Islamabad, Pakistan. Researchers focused on checking the bacteria's reaction to antibiotics and the frequency of these infections among patients at HBS Hospital. They also compared infection rates between men and women.This research was detailed and

planned in advance. It was conducted from December 2017 to April 2019 at the Pathology Department of HBS Medical & Dental College. Researchers gathered samples and used specific techniques to isolate the bacteria. They analyzed 150 clinical samples, providing crucial insights into how this bacteria reacts to antibiotics.The results revealed important information about the occurrence of methicillin-resistant Staphylococcus aureus (MRSA). Out of 108 infections, 48 were in males and 60 were in females. This indicates that infections were more frequent in females, with a ratio of 1.2 females to every male infected.The study discovered that 40.7% of the infections were resistant to methicillin. This discovery highlights the urgent need to improve infection control and use antibiotics responsibly to address the growing issue of antibiotic-resistant S. aureus in clinical environments..[35] Tariq anam, *et al*., (2019)counducted a study to investigate how common methicillin-resistant Staphylococcus aureus (MRSA) is in a large hospital and how it reacts to various antibiotics. Their goal was to uncover the genetic differences among MRSA strains and how these differences connect to different medical samples. There is a rising problem in Pakistani hospitals with bacteria that don't respond to antibiotics. This increase is mainly because antibiotics are often used incorrectly, which helps bacteria become resistant.The antibiotics tested included Oxacillin/Methicillin, Cefoxitin, and Vancomycin. The study isolated bacterial DNA using the CTAB method to better understand the genetic makeup.Researchers collected 96 strains of Staphylococcus aureus from different healthcare centers in Lahore. Of these, 67 strains (70%) were confirmed to be MRSA, while 29 strains (30%) were methicillin-sensitive Staphylococcus aureus (MSSA).The largest number of MRSA samples came from pus (57%), followed by sputum (21%), skin swabs (13%), and blood (9%). The prevalence of MRSA strains in Lahore was found to be as high as 70%, which is more than in other parts of Pakistan. This indicates a concerning rise in antibiotic resistance in local healthcare facilities. The high prevalence of MRSA poses a major health risk, highlighting the urgent need for better use of antibiotics and stricter infection control measures in hospitals..[36] Bala Kiran, *et al*., (2013) counducted study on Methicillin-resistant Staphylococcus aureus, or MRSA, is a serious health issue. It was discovered in 1960 and has become a major concern because it doesn't respond to many common antibiotics, like methicillin. MRSA is mostly found in hospitals, and its presence has been increasing significantly in recent years.A study looked at 4,271 pus samples over a year and found that 29.26% contained a bacterium called Staphylococcus aureus. Of those, 69.2% were MRSA. While most MRSA bacteria responded well to antibiotics like vancomycin, linezolid, and amikacin, a worrying 4.8% were resistant to all tested drugs, including vancomycin. This highlights the serious threat MRSA poses to patient health.Reports from the Centers for Disease Control and Prevention (CDC) state that MRSA causes over 90,000 severe infections and nearly 19,000 deaths each year in the United States. The study suggests that new rules for antibiotic use are necessary to stop MRSA from spreading.The growing number of MRSA cases and their resistance in hospitals demand prompt action for better infection control and improved antibiotic use management to safeguard patients and prevent the spread of this challenging bacterium.[37] Ankur kumar.et al(2015) counducted a study to examined how common methicillin-resistant Staphylococcus aureus (MRSA) infections are among patients in a large hospital in North India. The main focus was to identify and isolate Staphylococcus aureus from patient samples to find out the prevalence of MRSA in the hospital. MRSA is a serious health problem in India, with a prevalence rate of 29%. This situation stresses the need for effective measures to control its spread, such as practicing good hand hygiene and monitoring how the bacteria react to different treatments.The study spanned from January 2013 to September 2013, conducted at the Department of Microbiology, Teerthanker Mahaveer Medical College and Research Centre. It involved several crucial steps in identifying and detecting MRSA from various clinical samples. The research underscored the importance of implementing strategies to prevent MRSA infections in healthcare settings.[38] Kanthi pareera, *et al*., (1991) counducted study on Methicillin-resistant Staphylococcus aureus, known as

MRSA, is a major health issue, especially in hospitals. This study focused on the General Hospital in Colombo to discover how common MRSA is and to examine its resistance in different hospital areas. It's vital to know where MRSA is found and how it resists treatment to effectively control infections.Researchers collected 1,434 pus samples from patients using sterile tools from the Bacteriology Department. These samples were cultured on blood agar and MacConkey agar plates to grow the bacteria and were incubated at 37°C for 24 hours to promote bacterial growth.Out of the 672 positive samples, Staphylococcus aureus bacteria were identified. Among hospital inpatients, 30 out of 128 Staphylococcus aureus isolates (23.6%) were MRSA. For those visiting the hospital for outpatient care, 5 out of 47 isolates (10.6%) were MRSA.The findings showed that MRSA is a considerable problem at the General Hospital in Colombo, with many bacteria showing resistance to several antibiotics. Understanding this resistance is crucial for developing effective infection control strategies.[39] Piyush Rajbhandri, *et al*., (2024) counducted the study to focused on methicillin-resistant Staphylococcus aureus (MRSA), a type of bacteria that causes difficult-to-treat infections, especially in hospital settings. This research took place at Patan Hospital in Nepal, aiming to understand how widespread MRSA is in various samples.Conducted from January 1, 2021, to December 31, 2023, at Patan Academy of Health Sciences, the research was descriptive and cross-sectional, focusing on a snapshot of data collected over time. A total of 1,259 samples of the S. aureus bacteria were analyzed, and 570 of these, or 45.14%, were identified as MRSA. The study revealed consistently high MRSA rates over the three years: 44.5% in 2021, 46.5% in 2022, and 44.8% in 2023.The results showed that about 66.67% of MRSA cases came from samples collected during outpatient department visits. Additionally, MRSA demonstrated resistance to several other antibiotics: 39.57% to clindamycin, 36.23% to trimethoprim-sulfamethoxazole, 32.73% to azithromycin, 19% to gentamicin, 5.57% to ofloxacin, and 4.40% to ciprofloxacin.These findings highlight the growing threat of MRSA and underscore the urgent need for effective strategies to manage antibiotic resistance. Increasing awareness and developing better intervention practices are crucial for addressing this issue.[40] Ghaith alsabbagh, *et al*., (2023) This research was counducted to investigated how common Methicillin-Resistant Staphylococcus Aureus (MRSA) is at Rashid Hospital in Dubai, U.A.E. It includes patient samples collected over one year, from January 2021 to December 2021. total, 665 samples of S. aureus were collected. Out of these, 236 were identified as MRSA, making up 35.5%, and 429 were identified as MSSA, making up 64.5%. The samples came from various sources: 73 blood samples, 408 wound samples, 17 urine samples, 130 sputum samples, 6 body fluid samples, 16 biopsy samples, and 15 other samples.The MRSA samples showed 35.5% resistance to an antibiotic called oxacillin.However, they were not resistant to other antibiotics such as Vancomycin, Linezolid, Rifampicin, Tigecycline, and Teicoplanin.The findings indicate that MSSA is more prevalent than MRSA, particularly in wound infections. The analysis also shows that MSSA cases are higher in males across different age groups, with the exception of the youngest group..[41] Shafqat Husnain Khan, *et al*., in 2017A study focused on how common methicillin-resistant Staphylococcus aureus (MRSA) is in hospitals. MRSA is a major concern because it's resistant to some antibiotics, making it difficult to treat. The study's main goal was to find out how often MRSA appears in hospitals and how these bacteria respond to antibiotics.Researchers collected 100 samples of a type of bacteria called coagulase-positive Staphylococcus aureus from various sources inside the hospital. The findings revealed that 39 out of 100 samples were resistant to methicillin, indicating a 39% rate of MRSA. Fortunately, all these MRSA strains were sensitive to vancomycin. This means that vancomycin is still effective for treating MRSA infections.The study concluded that MRSA is a significant problem in the hospitals studied. It emphasized the urgent need for strong infection control practices. This includes regular monitoring, educating staff on infection prevention, and ensuring antibiotics are used wisely to manage MRSA effectively..[42] Shaika Farooq, *et al*., (2016)A study was carried out to determine how common

Methicillin-Resistant Staphylococcus aureus (MRSA) is and its antibiotic sensitivity in a major orthopaedic hospital in Kashmir, India.The study aimed to evaluate the antibiotic sensitivity patterns of MRSA and other bacteria in the Orthopaedic Department from November 2014 to November 2015. This evaluation helps in making informed decisions about which antibiotics to use, especially since some cases of vancomycin resistance have been reported.Researchers examined 160 pus samples from patients with different bone and surgical infections.The findings from the 160 samples were:84 samples (52%) showed no bacterial growth.43 samples (26.8%) contained Staphylococcus aureus.Of these, 32 samples (74.4%) had MRSA, and 11 samples (25.6%) had MSSA, which does not resist methicillin. Other bacteria were found, with E. coli being the most common among gram-negative types.Most MRSA cases were linked to sudden bone infections (acute osteomyelitis) and surgeries involving broken bones, highlighting a significant health concern.Overall, the study concluded that while traditional antibiotics like linezolid and vancomycin are still effective against most Staphylococcus aureus, the potential for vancomycin resistance exists. This discovery stresses the need for careful antibiotic selection and monitoring in managing bone and joint infections.[43]

Objective

The aim of this research is to find out how frequently MRSA occurs in pus samples drawn from clinical sources.

Problem Statement

One big health problem today is the increase in cases of MRSA, a type of bacteria resistant to methicillin. This resistance makes infections tough to treat. Despite improvements in medicine, MRSA still resists antibiotics, making it hard to control and treat. The study looks at why MRSA spreads and why it stays resistant to antibiotics. Understanding these reasons is key to dealing with this problem. This knowledge helps in creating special treatment plans and in using antibiotics more effectively, which are both important steps in fighting MRSA..

Operational Definition(S)

Methicillin-Resistant Staphylococcus Aureus (MRSA):

A strain of *Staphylococcus aureus* that has acquired resistance to methicillin and other beta-lactam antibiotics, primarily due to the presence of the **mecA** gene which encodes altered penicillin-binding proteins (PBP2a).

Pus Sample:

Definition: A biological specimen consisting of exudate collected from infected wounds, abscesses, or surgical sites, typically containing dead cells, bacteria, and inflammatory cells.

Measurement: Collected aseptically from patients and submitted to the microbiology laboratory for culture and sensitivity testing.

Prevalence:

Definition: The proportion of pus samples that test positive for MRSA among the total number of *Staphylococcus aureus* isolates or all pus samples tested during the study period.

Measurement: Calculated by dividing the number of MRSA-positive isolates by the total number of relevant samples or *S. aureus* isolates, and expressed as a percentage.

Antibiotic Susceptibility Testing (AST):

Definition: A laboratory technique used to determine the sensitivity or resistance of bacterial isolates to various antibiotics.

Demographic Data:
Definition: Patient-related background information such as age, sex, clinical history, hospital ward, and type of infection.
Measurement: Recorded during patient registration or sample collection and used to analyze trends or risk factors associated with MRSA infection.

Clinical Diagnosis:
Definition: The medical identification of an infection site (e.g., abscess, surgical wound) based on signs, symptoms, and clinical examination by healthcare professionals.
Measurement: Documented by physicians in patient records and correlated with laboratory-confirmed MRSA results.

Material and Methods
Study Design: It will be cross sectional study.
Settings: This research is conducted in Chughtai Lab lahore.
Sample Size: Around 250 Random patient were Examine.
Sampling Technique: Pus sample were collected and performed on Vitex Ms.

Sample Selection:
Inclusion Criteria:
- Every pus sample that tested positive for Methicillin-Resistant Staphylococcus aureus (MRSA) will be included in this study.
- The study is focused on (HA-MRSA) strains.
- Demographic data such as patient age and gender, along with clinical details like hospital ward and antibiotic resistance patterns, was also analyzed.

Exclusion Criteria:
- Samples that tested negative for MRSA, as well as those that were not identified as Staphylococcus aureus or were found to be methicillin-sensitive Staphylococcus aureus (MSSA), were excluded.
- Additionally, community-acquired MRSA strains and entries with incomplete demographic or laboratory data were excluded to maintain accuracy and specificity of the findings

Equipment(s): BacT ALERT Vitro VITEK MS
Ethical Considerations (Where needed for clinical studies, animal study, experimental study)
The rules and regulations set by the ethical committee of Superior University, Lahore were followed while conducting the research and the rights of the research participants were respected.
- Written informed consent attached was taken from all the participants.
- All information and data collection were kept confidential.
- Participants were remained anonymous throughout the study.
- The subjects were informed that there are no disadvantages or risk on the procedure of the study.
- They were also be informed that they will be free to withdraw at any time during the process of the study.
- Data was kept in under key and lock while keeping keys in hand. In laptop it was kept under password.

Data Collection Procedure

Total Two hundred and fifty strains of Staphylococcus aureus that are resistant to antibiotics such as ceftriaxone, methicillin, and Vcancomycin were gathered from Chughtai Laboratory. A pus sample is used to obtain these strains. Patients who reported having a bacterial infection were the ones from whom these samples were taken.

Data Analysis Procedure

These samples were all gathered and transported to the microbiology lab in sterile containers.These samples are incubated for 24 hours at 37 C on nutrient agar, MacConkey agar, and blood agar. These methods, which include morphological appearance, Gram staining, and biochemical tests such the Catalase and Coagulase test, were used to identify the bacterial isolates.

Morphological Appearance:

When examined under a microscope following Gram staining, Staphylococcus aureus has a spherical, gram-positive morphological appearance that frequently forms a cluster resembling a grape.

Biochemical test:

Using a loop or sterile wooden stick, transfer a small amount of well-isolated bacterial colony development on the surface of a dry, clean glass slide. In the glass slide, put a drop of 3% hydrogen peroxide solution observe bubbling immediately

Preparation of Nutrient Agar and Sample Inoculation:

Sample numbers were assigned to label the nutrient agar plates. This medium is formed by mixing 200 milliliters of sterile nutritional agar with 100 milliliters of sterilized or fresh milk, which is later poured into plates. The staphyloxanthin pigment causes the S. aureus colonies in this medium to be golden-yellow after a 24-hour incubation at 37°C.

Preparation of Blood Agar and Sample Inoculation:

A representative number was used on the blood agar plates for identification. Blood agar was autoclaved for 15 minutes at 121°C after preparation. Prepared blood agar was placed aseptically over petri dishes. The materials (pus) were spread over the blood agar plates using the inoculating loop. The plates need to be incubated for 24 hours at 37°C. Verify the presence of bacterial growth after incubation. On blood agar, S. aureus displays beta hemolysis.after 24 hours incubation at 37°C.

Antimicrobial Susceptibility Testing:

Following the recommendations set out by the Clinical Laboratory Standards Institute (CLSI, 2011), the Kirby-Bauer disc diffusion method was used to evaluate the antimicrobial susceptibility of the detected Staphylococcus aureus isolates to vancomycin, methicillin, and ceftriaxone. For the disc diffusion assay, Oxoid (England) antibiotic discs were utilized. Vancomycin (30 µg), methicillin (5 µg), and ceftriaxone (30 µg) were among the antibiotic discs. As a stand-in marker for methicillin resistance (MRSA determination), cefoxitin (30 µg) discs were employed; a zone of inhibition <21 mm was deemed resistant, while ≥22 mm was deemed susceptible. Zone diameters interpreted in accordance with CLSI recommendations were used to classify isolates as susceptible or resistant.

Minimum Inhibitory Concentration (MIC):

According to CLSI recommendations, agar dilution technique was utilized for determining Minimum Inhibitory Concentration (MIC) of vancomycin, methicillin, and ceftriaxone against Staphylococcus aureus isolates. Molten Mueller-Hinton agar, which was cooled to approximately 50°C, was combined with consecutive two-fold serial dilutions of each drug. The recommended solvents of the respective medications were employed in producing antibiotic stock solutions and final dilutions. After solidification, standard bacterial suspensions were spot-inoculated on agar plates containing different doses of antibiotics, and the plates were incubated at 35°C for 18 to 24 hours. The minimum

concentration of antibiotics that completely inhibited visible bacterial growth was referred to as the minimum inhibitory concentration, or MIC.

Results:

In this study 250 pus sample were isolate , several parameters were evaluated through bar charts, pie charts, and tabular data to assess the prevalence of *Staphylococcus aureus*, MRSA detection, and antibiotic resistance patterns.

Gender-Wise Distribution

According to the gender-based pie chart, **54% of the samples were collected from male patients**, while **46% were from female patients**. This shows a slightly higher prevalence of infection among males compared to females.

Figure 5.1:Illustrating gender-wise percentage of staphylococcus aureus isolates

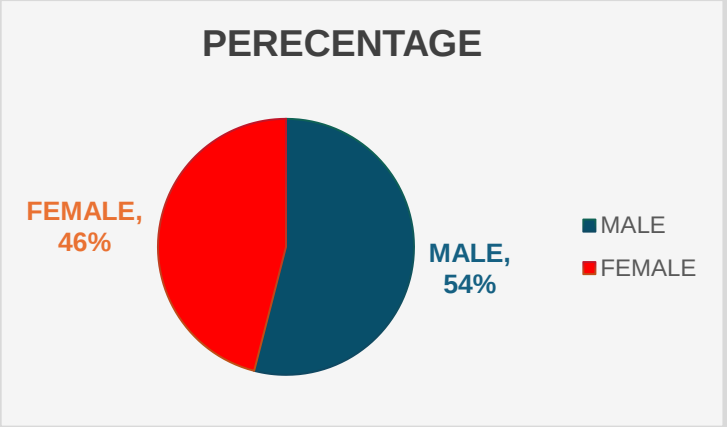

Table:5.1 Illustrating gender-wise percentage of staphylococcus aureus isolates

Gendrer	Percentage
Male	54%
Female	46%

The table shows the distribution of staphylococcus aureus bacteria by gender.This suggests a marginal male predominance in S. aureus infections among the study population it provide a breakdown of the number of positive sample from pus.The data in the table help to understand the ratio of gender in the pus sample

MRSA Prevlance

The MRSA data show that 49% of the Staphylococcus aureus isolates were resistant to Methicillin, confirming MRSA infection, while 51% were Methicillin-sensitive. This indicates a high MRSA burden, requiring targeted infection control strategies.

Figure 5.2: Representing MRSA prevlance showing percentage of MRSA positive and MRSA negative isolates

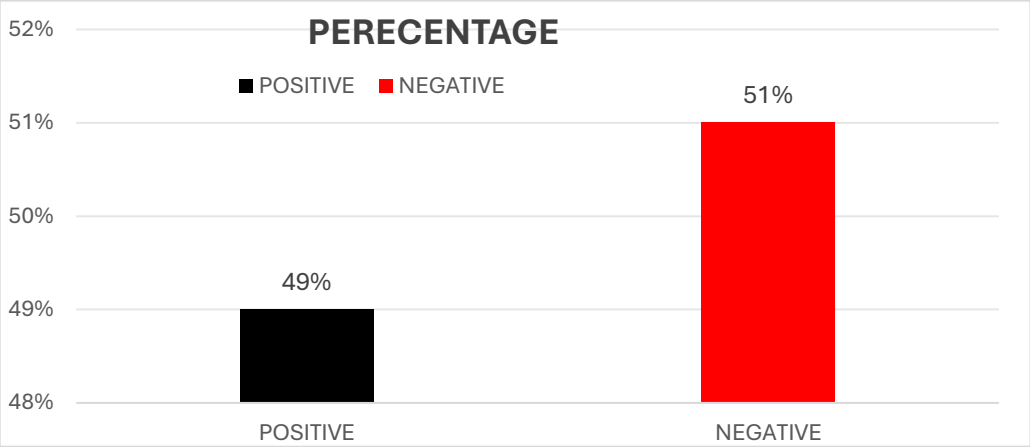

Table 5.2: Representing MRSA prevlance showing percentage of MRSA positive and MRSA negative isolates

MRSA Precences	Percentage
Positive	**49%**
Negative	**51%**

As shown in the MRSA prevalence in coloum chart , 49% of the samples tested positive for Methicillin-Resistant Staphylococcus aureus (MRSA), while 51% tested negative. These findings revealing that nearly half of the S. aureus cases were resistant to Methicillin, indicating a significant MRSA burden in the patient population studied.

Antibiotic Testing Status

Based on the antibiotic testing clustered column, it was observed that: Antibiotic testing was performed on 55% of the samples. 45% of the samples were not tested for antibiotic susceptibility.This indicates a need to improve laboratory practices to ensure all samples undergo proper antimicrobial susceptibility testing for effective treatment planning.

Figure 5.3:Depicting the percentage of samples performed VS not performed for antibiotic susceptibility

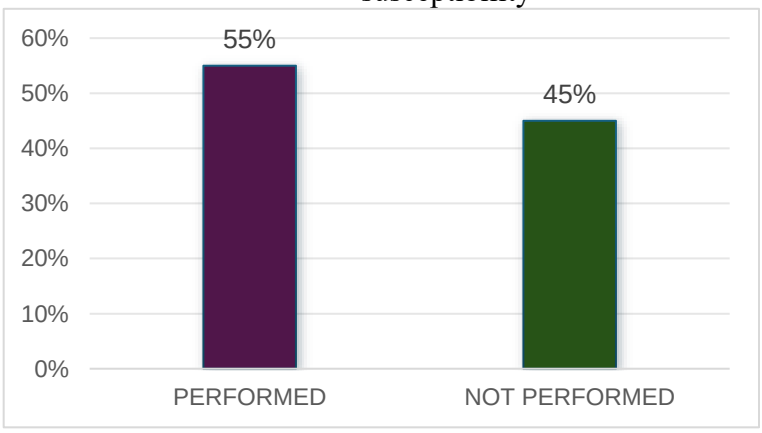

Table 5.3:Depicting the percentage of samples performed VS not performed for antibiotic susceptibility

Antibiotic Testing	Percentage
Performed	**55%**
Not Performed	**45%**

This column chart illustrate the proportion of samples that underwent antibiotic susceptibility testing. It was observed that: 55% of the total samples were tested for antibiotic sensitivity, while 45% were not tested. This finding highlights a gap in comprehensive antibiotic testing and underscores the need to ensure full laboratory evaluation of clinical samples for proper antibiotic stewardship and treatment planning.

Antibiotic Resistance Pattern

Resistance percentages for three commonly used antibiotics were presented using a dedicated Column chart: Vancomycin showed the highest resistance rate at 31%, followed by Methicillin at 24%,and Ceftriaxone at 22%. These results indicate that Vancomycin-resistant strains are emerging, which is of particular-concern due to its role as a last-resort antibiotic for Gram-positive infections. Methicillin resistance confirms the presence of MRSA, and resistance to Ceftriaxone highlights growing beta-lactam resistance among S. aureus isolates.

Figure 5.4: Showing resistance percentages to Vancomycin, Ceftriaxone, and Methicillin among S. aureus isolates

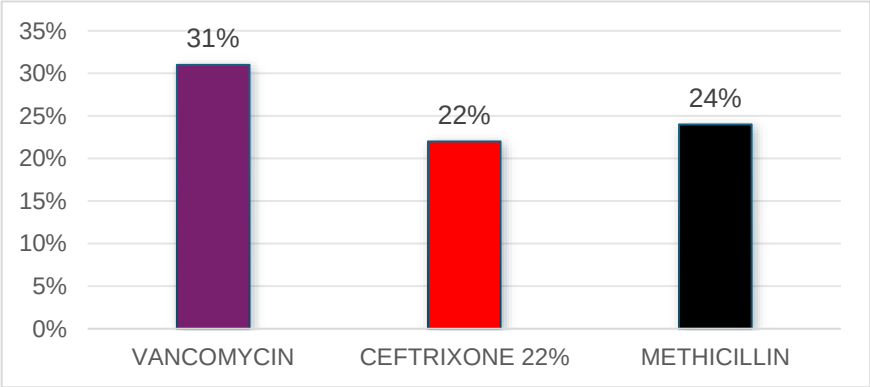

TABLE 5.4: Showing resistance percentages to Vancomycin, Ceftriaxone, and Methicillin among S. aureus isolates

Resistance Antibiotic	Percentage
Vancomycin	31%
Ceftrixone	22%
Methicillin	24%

This segment outlines the resistance levels to three commonly used antibiotics. The highest resistance was observed for Vancomycin (31%), followed by Methicillin (24%), and Ceftriaxone (22%). These values signal growing concerns about Vancomycin-resistant and Methicillin-resistant Staphylococcus aureus (MRSA) in the clinical setting.

Discussion

Methicillin-resistant S. aureus has been identified as a significant bacterial pathogen that causes infections in hospitals and the general population globally because of its heightened virulence and continuously increasing resistance to antibiotics. Epidermal tissue infections, bronchopneumonia, and

localized infections that migrate into the bloodstream worldwide are all caused by the common human infectious pathogen Staphylococcus aureus (360). Finding the pathogens and their drug-sensitivity patterns is crucial for treating bacterial infections and tackling the problem of antibiotic resistance. The prevalence of antibiotic resistance has increased over time. .The goal of the current investigation was to determine the bacteria found in patient samples and their pattern of antibiotic susceptibility. MRSA strains were identified in recent investigations of 250 samples. The fact that a greater percentage of isolates in the pus sample from patients in wound swabs (26%) could suggest that MRSA is more common in wound site infections. Males (52%) had a higher percentage of bacterial isolates in our study compared to females (46%). Similarly, the pus sample from an unidentified region in 2022 contained Staphylococcus aureus in 35.1 % of cases, while 50.4 % of samples displayed bacterial growth (Noshad Ali Sajid et al.).The emphasis is on the importance of constant monitoring of patterns of antibiotic resistance to guide proper treatment. In 2024, coagulase-positive Staphylococcus aureus was found in 7.75% (114 out of 1470) of clinical specimens at the SBKS MI & RC (Kajal Parmar et al.). In a 2019-2020 investigation at MMIMSR, Mullana, Ambala, 51% of Staphylococcus aureus isolates (102 out of 200) from pus samples were identified as MRSA (Subash Chaudhary et al.). This means that the load of MRSA is significant both in OPD and IPD patients. The prevalence of MRSA among Staphylococcus aureus isolates was 45.14% (570 out of 1259) in Nepal during 2021-2023 (Piyush Rajbhandari et al.)In 2021, 46.5% and 44.8% yearly prevalence were observed in 2022 and 2023, indicating a consistently high frequency. 66.67% of the isolates originated from outpatients, thereby suggesting population-specific transmission pathways. In a tertiary care hospital in eastern India, pus-purification samples (39/134) were found to be positive for Staphylococcus aureus, with 29.10% of them confirmed as MRSA (Dr. Tansuri Biswas et al.).S. aureus was isolated in 74.86% (134 samples) of 179 pus samples obtained between September 2019 and January 2020. This finding suggests that the bacterium is common in cases of wound infections. Based on the antibacterial resistance pattern of our study, 24% of the isolates of MRSA were resistant to methicillin, which is their characteristic difference. Additionally, 32% of the isolates had a resistance to vancomycin, which is very alarming since vancomycin is used only in the last instance. Furthermore, the drug ceftriaxone, a cephalosporin of the third generation that is used very often, did not show an efficacy for 22% of samples.The results suggest that resistance is increasingly being associated with the use of higher-generation β-lactam antibacterial agents. High resistances to multiple antibiotics also performed by the overall antimicrobial susceptibility testing. Penicillin revealed 100% resistance, a situation that is in line with the global pattern caused by the wide-spread synthesis of β-lactamase enzymes. The other high resistances were also exhibited by cephalosporins such cefotaxime (72.45%) and ceftazidime (74.55%). Notable resistances were also displayed by erythromycin (29,59%), tetracycline (27.55%), amikacin (47.96%), ciprofloxacin (67.35%) and gentamicin (31.63%). Those of other regional and international research were also identified.Consider, for instance, that there was a 83% resistance in 2015-2017 in Poland to ciprofloxacin, levofloxacin, erythromycin, and clindamycin (Kloos et al, 2019). For the 2013 study in the southwest of Ethiopia, all bacteria were 100% resistant to β-lactam antibiotics, such as ampicillin, cefoxitin, and penicillin (Kloos et al., 2019). The need to improve infection control practices and to regularly monitor antimicrobial resistance is urgent because resistance to important antibiotics, such as methicillin and vancomycin, is growing. For the prevention of new methicillin-resistant strains of S. aureus and the preservation of the effectiveness of last-resort antibiotics, we need antibiotic stewardship initiatives and the judicious use of antibiotics.

Conclusion(S)

This study examined how often Methicillin-Resistant Staphylococcus aureus (MRSA) appears in medical samples, especially in pus samples. It found that MRSA infections continue to be a significant issue in hospitals and clinics. These infections often do not respond to many common antibiotics, even

to those reserved for serious conditions, which is concerning because it means treatments are becoming less effective.The research highlighted that MRSA is particularly resistant to important antibiotics like Vancomycin and Methicillin. Additionally, many samples didn't undergo enough antibiotic testing, showing the need for better testing to choose the right treatments.The study showed a slight increase in MRSA infections among men, possibly due to behavior or environmental factors. The overall pattern of resistance shows the necessity for strict infection controls, careful use of antibiotics, and comprehensive lab testing.The findings add to the growing understanding of antibiotic resistance. They highlight the urgent need for healthcare systems to act. It's crucial to monitor MRSA closely, use antibiotics carefully, and support research efforts. These steps are essential to manage drug-resistant S. aureus and to keep our existing antibiotics effectiveThe data reveals that many routine tests checking if antibiotics are effective are lacking. As a result, numerous clinical samples aren't being evaluated for resistance. This poses a significant issue, as it can lead to treatments failing and germs spreading without control. Additionally, our findings indicate that male patients are affected more than females. This could be due to their work, habits, or lifestyles, which might expose them to infections more frequently.Traditional antibiotics are becoming less effective against S. aureus, making treatment options limited. This creates a major challenge for doctors and microbiologists, who now need to use advanced tests and newer, often more expensive, antibiotics. The current trends highlight the need for better infection prevention in healthcare settings. This includes improving hygiene, conducting thorough patient screenings, and isolating those at high risk.

Recommendations

Enhance Surveillance and Reporting of MRSA Cases

It is important to make better efforts to regularly track and record the levels of MRSA and how it resists treatment in both hospitals and labs. By using a centralized database, we can more effectively watch for new patterns and better manage the spread of strains that resist treatment.

Implement Strong Antibiotic Stewardship Programs

To ensure antibiotics are used correctly, we must have clear and strict rules. Healthcare professionals, including doctors, should receive proper training. They need to learn to prescribe antibiotics based on specific tests that identify the most effective medication. This approach is important to prevent the overuse of strong antibiotics, which can lead to resistance and other issues.

Ensure Routine Antimicrobial Susceptibility Testing

It's important to test how antibiotics affect a sample when there's a chance of a Staphylococcus aureus infection in pus. This testing helps choose the right treatment and quickly spot MRSA infections.

Improve Laboratory Infrastructure and Diagnostic Coverage

Laboratories need proper tools and trained staff for regular culture and sensitivity testing. Currently, 45% of samples are not tested, which is a gap that needs addressing.

Strengthen Infection Control Practices in Healthcare Settings

To avoid hospital-acquired infections and cross-contamination, strict cleanliness measures, appropriate wound care, and isolation of MRSA-positive patients should be put into place.

Raise Public and Clinical Awareness

Campaigns to educate the public and medical professionals about the risks posed by MRSA, the significance of finishing antibiotic regimens, and appropriate hygiene procedures should be launched.

Promote Research on Resistance Mechanisms

To comprehend the genesis and dissemination of resistance, more research is required to examine the molecular traits of MRSA, such as the presence of the mecA gene and SCCmec types.

Encourage Policy Support for Infection Control

Policies that prioritize infection control and antibiotic resistance monitoring should be supported by

health authorities, particularly in surgical and wound care units where pus samples are frequently collected.

Focus on Community-Acquired MRSA (CA-MRSA)

Hospitals should not be the only places under surveillance; community sources of MRSA should also be looked into because they are increasingly responsible for the development of resistant infections.

Regular Screening of Healthcare Workers

Hospital MRSA transmission can be reduced by routinely screening and decolonizing medical personnel in high-risk wards.

Limitation

Although the findings provide valuable insight into the local prevalence and resistance patterns of MRSA, several limitations should be acknowledged:

1. **Incomplete antibiotic testing** Out of all the samples, only 55% were tested to see how they react to antimicrobial treatments. This limited testing could mean that the data on resistance is not as complete as it could be.

2.Lack of molecular analysis Resistance was checked only with phenotypic methods. No genotypic confirmation, like finding the mecA gene, was done to prove the presence of MRSA.

3.Absence of clinical and demographic variables The information did not have important details about the patients, such as their age, other health conditions, past use of antibiotics, or their history of hospital stays. Without this information, it's difficult to directly relate the data to each patient's specific medical situation.

4.Single-center study All the samples came from one location, so they might not accurately represent how widespread MRSA is in the surrounding region or throughout the whole country.

5.Focus on only three antibiotics in resistance table The study checked several antibiotics, but the detailed resistance data in tables was only available for Vancomycin, Methicillin, and Ceftriaxone. This might not give us a complete picture of all the resistance patterns. There could be other important resistance information that we are missing because it wasn't included in the tables.

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