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ORIGINAL ARTICLE

Frequency of Staphylococcus Aureus Infection Associated with Femoral Double Lumen Catheter in Kidney Failure Patients

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ABSTRACT

Introduction: The most frequent pathogens responsible for renal failure in hospitalised patients are Staphylococcus species, particularly in those receiving hemodialysis. The aim was to isolate and identified the Staphylococcus species from hemodialysis patients.

Methodology: The research was carried out between November 2021 and February 2022. This study included participants who were receiving maintenance hemodialysis for end-stage renal illness. Each patient's demographic information was gathered using a structured questionnaire that had been evaluated beforehand. Blood samples from both sexes were taken (28 males and 22 females). The blood samples were sent directly to the research lab for processing after being inoculated into the Brain Heart Infusion broth under aseptic circumstances. Gram stain and biochemical assays were used to determine the bacteria's identity.

Result : Overall, there were 44 percent female patients and 56 percent male patients. Out of all the blood samples analyse, 26% of samples showed evidence of bacterial development in the Brain Heart Infusion broth. S. aureus species (62 percent), S. epidermidis (15 percent), and S. saprophyticus were the Staphylococcus species that were detected 23 percent).

Conclusion

According to the findings, coagulase-positive Staphylococci were the most often isolated species from hemodialysis patients who had bacteremia, but coagulase-negative strains were identified far less frequently. The findings of the current study need to be confirmed by more research.

Keywords: S. aureus, Kidney Failure, Hemodialysis, Femoral Double Lumen Catheter

INTRODUCTION

Ogston initially used the name Staphylococcus to describe a class of micrococci that produced pus and caused irritation in 1883. They are non-motile, do not produce facultative anaerobe spores, and proliferate through fermentation or aerobic respiration[1]. Staphylococci are Gram-positive bacteria that range in diameter from 0.5 to 1.5 mm and form grape-shaped clusters that are separated into multiple planes when the individual cocci are combined. There are more than 30 species and 8 subspecies in the genus Staphylococcus, many of which are beneficial and hinder the human body. The three most prevalent strains are S. aureus, S. saprophyticus, and S. epidermidis[2, 3].

Coagulase-negative Staphylococci are part of the typical human skin and mucous membrane microbiome. Staphylococcus bacteria can spread from person to person by direct and indirect physical contact, and when the organism is pathogenic, it can be a major issue. In surgical incisions, S. aureus is a significant source of nosocomial infection[4].

A condition known as chronic kidney disease (CKD) occurs when the kidneys lose their ability to remove electrolytes and excess water from the blood as waste products. Renal transplantation and dialysis are potential treatments for CKD. Dialysis is preferable to a kidney transplant. Dialysis may sometimes be administered for the rest of one's life due to the lack of kidney donors[5]. Dialysis is a process that takes the place of the kidneys' usual function. The two main methods of dialysis are hemodialysis and peritoneal dialysis. For at-home therapy, peritoneal dialysis (PD) is preferred over hemodialysis (HD), which is done in healthcare facilities[6].

End-stage renal disease patients' quality of life is improved with hemodialysis (HD) (ESRD). By eliminating wastes, controlling bodily fluids, and preserving chemical equilibrium, HD machines filter blood. Central venous catheters (CVCs) are frequently employed in HD. The CVC is the ideal option since it is immediately available, especially when urgent HD is required, such as when starting renal replacement treatment or when permanent vascular access ceases to work[2]. Due to the prolonged access to the circulatory system during HD operations, patients who require chronic hemodialysis are at a greater risk of infections. Patients who need multiple sessions of dialysis have a higher risk of infection either directly or indirectly through contaminated tools or devices in a dialysis unit[7].However, in dialysis patients, it is a frequent risk factor for Staphylococcus aureus infection and bacteremia[8]. CVCs are the ideal hemodialysis alternative for individuals with uremia and other blood diseases[9]. Both the proximal central vein and the peripheral vein can be used to place catheters. The internal jugular, subclavian, and femoral veins are the most often utilised veins for double lumen catheters[10].

The most frequent causes of infections associated with dialysis are catheter loss and transition to hemodialysis. It's crucial to modify the catheter's surface to prevent infections[7]. Mechanical issues or infections, which cause significant morbidity and mortality in hemodialysis patients, frequently impede the use of temporary hemodialysis catheters. Catheter-related bacteremia is often caused by Gram-positive and Gram-negative bacteria, which can lead to serious systemic infections such osteomyelitis, endocarditis, septic arthritis, epidural abscess, and even mortality[11]. The frequency of catheter manipulation, the kind of catheter being used, and the patient's health, such as any underlying illnesses, all affect the incidence of catheter-related bloodstream infections. Peripheral venous catheters are the most frequently used catheter[6].

Arteriovenous fistula (AVF), double lumen catheter, and Permacath are the three methods that hemodialysis can be performed. Three different variants of the double lumen catheter procedure—inter jugular, femoral, and subclavian—are frequently utilised for hemodialysis. All of these catheterization procedures are carried out under the care of critically unwell patients. In order to prevent venous stenosis, the US Centers for Disease Control and Prevention recommends placing the catheter in either the femoral or jugular vein when necessary[12]. Because there is a lower risk of infection and mechanical problems, internal jugular locations are typically preferred. When a patient has complex anatomy, such as a scar at the insertion site or any other indication that might make an insertion difficult, only professionals should perform catheter

insertions[13]. Infectious, mechanical, and thrombotic problems from this invasive treatment can raise morbidity, mortality, and expense during dialysis. Femoral venous catheterization is carried out in an emergency to provide vascular access, although it should be avoided to reduce hospital-acquired infections. Although the subclavian site is the initial option, bigger catheters like uncuffed temporary dialysis catheters are less appropriate for this location. Central venous catheterization carries risks for infections, as well as mechanical and thrombotic problems. It is necessary to take a chest radiograph to verify placement and check for problems. There are three different ways that catheter infections might happen: hub colonisation followed by infection through the intraluminal pathway; local insertion site infection; and hematogenous seeding of the catheter[14].

The measurement of central venous pressure, fluid infusion, and long-term intravenous nourishment are now all made possible by the catheterization of central veins. yet susceptible to illnesses like septicemia. Hemodialysis is used to treat patients with renal disease. A significant source of morbidity and mortality is bloodstream infection associated with central venous catheters. Staphylococci species, of which Staphylococcus aureus is the most dangerous, are mostly to blame for septicemia, which can lead to serious consequences and even death. Nasal S. aureus colonisation, prolonged catheter usage, greater total intravenous iron dose, advanced age, prior bacteremia, lower haemoglobin diabetes mellitus, serum albumin levels, and most recently admitted to the hospital are risk factors[15]. Clinically suspected signs of chronic hemodialysis include fever and chills. Blood culture testing and the administration of broad-spectrum intravenous antibiotics should be done when catheter-related illness is suspected. When the same microbe is identified from quantitative culture and peripheral blood of a patient with clinical symptoms of infection and no alternative sources of infection, the diagnosis is considered to be definitive. Gram-positive cocci, most frequently S. aureus and S. epidermidis, produce bacteremia in two-thirds of cases[16]. A paediatrics research from Pakistan The most significant infection in hospitalised renal failure patients, particularly those receiving hemodialysis, is Staphylococcus species[2]. Kind II diabetics are more susceptible to endogenous S. aureus linked with vascular access septicemia (VAS), a dependent type of vascular access used for hemodialysis, due to relatively greater nasal carriage rates. Cardiovascular disease is a secondary cause of death for individuals with renal failure who get bloodstream infections in hospitals. Hemodialysis catheters are to blame for the bulk of these infections[17]. S. aureus was isolated from 21.2 percent of samples in Pakistan, according to another study[18].

The incidence of methicillin-resistant S. aureus (MRSA) and methicillin-susceptible S. aureus (MSSA) nasal passage and its impact on vancomycin-resistant Staphylococcus (VRS) were examined in order to determine the most vulnerable group and develop a strategy for prospective preventive measures in accordance. The likelihood of acquiring MSSA and MRSA nasal carriage-related VRS in type II diabetics was 35 and 38 times greater with central venous catheters (CVCs) compared to arterio-venous fistula (AVF). It was discovered that MSSA and MRSA nasal carriage-related VRS had the lowest risk.The necessity to maximise AVF in the risk group while simultaneously restricting the use of CVCs makes it difficult to reduce the number of S. aureus nasal passage-related VRS[19].

This investigation sought to determine the incidence of S. aureus infections in renal failure patients using a femoral double catheter.

MATERIALS AND METHODS

This four-month cross-sectional study was carried out at Hayatabad Medical Complex in Hayatabad Peshawar (from November 2021 to February 2022). Convenient sampling, a non-probability technique, was used for this investigation.

The research comprised 50 patients with renal failure receiving hemodialysis using a femoral double lumen catheter. The research included all patients (both sexes) with renal failure who were receiving hemodialysis using a femoral double lumen catheter and were between the ages of 15 and 75. Patients of OPD, or hospitalised patients affected with other diseases than chronic kidney failure were excluded from the research. Patients who were 15 years of age or older were also not included.

Before each subject was accepted into the research, their parents or legal guardians provided written informed permission. The statistical analysis was performed using Microsoft Excel 2016.

The treatment was carried out by skilled and knowledgeable medical professionals pursuing their residency in nephrology. Patients receiving hemodialysis had their blood drawn using a vein puncture procedure in an environment that was completely sterile. This was accomplished using iodine and 70% alcohol sterilising the collecting location. Each patient gave 5 ml of their blood using sterilised syringes.

The 5 ml of specimens were rapidly inoculated into 70 ml of Brain Heart Infusion broth blood culture containers aseptic conditions and cultivated aerobically at 37 ° c for up to seven days. The blood culture containers were sterilised using 70 percent alcohol and iodine, and an aseptic one replaced the needle of a syringe, which was used in specimen collection. Before performing the subculture, all blood culture bottles were inspected for indications of bacterial development, such as (turbidity, presence of clot, and hemolysis). Then, mannitol salt agar was subcultured by streaking out a little quantity of infected broth onto the upper border of the agar until 1/3 of the plate was covered, while rotating the plate at 60 degrees. To produce a pure culture, the sample was evenly distributed by regular streaking from the end of the first region into the second one. Later, the blood culture bottles underwent a second 370C incubation. After each 24 hour incubation, the subcultured method was carried out twice more.

Growth produced after the first, second, or third subculture was tested for colony morphology and mannitol-fermenting capacity. The sample was destroyed and labelled as sterile or negative blood culture after failing to develop for up to a week. Based on size, colour, density, edges, side view, and fermentation of mannitol sugar, the colonial characteristic is chosen as the initial identifying stage. Following an overnight incubation at 370°C, the isolated organisms were purified in sterile nutrient agar slop, and the identified organisms were then utilised. The sterile nutritional glycerol broth medium was used to store the purified organisms at -200C in sterile crayon tubes.

By analysing pathogens' Gram responses, cell configuration, and organisation in accordance with the Gram technique, the Gram stain was utilised to identify them in cultures. The procedure was carried out as follows. The smear was created on a spotless slide, which was then quickly passed over the Bunsen flame to correct it. The smear was floated with mordant (Lugol's iodine) for two minutes after being coated with crystal violet for one minute. The smear was then gently washed with water. The smear was then gently washed again for a little while after being decolored with alcohol. Finally, safranine was applied to the smears and allowed to sit for 2 minutes before being washed again and dried by blotting on filter paper.

For example, the following biochemical assays were used to identify the isolated strains of microorganisms: (catalase, DNAse, coagulase, sensitivity to polymixin B and novobiocinpolymixin , and sugar fermentation tests). In the conversion of hydrogen peroxide into oxygen and water, catalase functions as a catalyst. Numerous colonies of tested microorganisms were scraped using sterile wooden sticks, submerged in the hydrogen peroxide solution, and checked for quick bubbling. Three millilitres of hydrogen peroxide was then added to the sterile test tubes. A catalase test that is bubbling actively is positive; one in which there are no bubbles is negative.

The test was used to distinguish between S. aureus and other Staphylococci as S. aureus produced the coagulase enzyme. Plasma clots as a result of coagulase's conversion of fibrinogen to fibrin. Coagulation factors come in two varieties: free and

bound coagulase. A positive result was shown by the development of a fibrin clot, whilst a negative result was indicated by the lack of a clot. The free coagulase, which converts fibrinogen to fibrin, was performed by adding 0.2 ml of plasma to 0.8 ml of test bacteria in a tube.

The test was carried out by spotting the test organism into DNA agar, after which all plates were incubated for an overnight period at 37℃. After the incubation time, the hydrochloric acid solution was poured onto the plates and let to sit for five minutes while researchers looked for a clear zone around the organism spot that would suggest DNA hydrolysis. If there is no clearing, the test was negative.

The ethics committee's and the heads of the microbiology and dialysis units' formal permissions were obtained. This information was acquired from the Microbiology Laboratory's laboratory record book at the Hayatabad Medical Complex in Hayatabad, Peshawar. The nursing staff took blood samples, which they then forwarded to the microbiology lab for blood culture. Then, these samples were processed by medical laboratory technicians. Seven days later, several bacterial growths were extracted, identified using microscopy, and the species were verified using a variety of biochemical assays. The research lab record book was updated with the reports. The patients participating in this study also provided their informed permission.

RESULTS

In this work, common Staphylococcus species in hemodialysis patients were isolated and identified. Fifty (n=50) hemodialysis patients using a femoral double lumen catheter were included in the study. Out of 50 hemodialysis patients, 28 (56%) men and 22 (44%) women made up the patient population (Table 1).

Table 1: Distribution of hemodialysis patients according to gender

Gender	Number	Percentage
Male	28	56%
Female	22	44%
Total	50	100%

Only 13 (n=26%) of the 50 blood samples under investigation showed positive bacterial growth, whereas the remaining 37 samples exhibited no growth at all, and 74% of the samples were negative. The Gram stain method was used to separate the isolated organisms into Gram-positive and Gram-negative types (Table 2).

Table 2: Bacterial growth in media

Culture Growth	Number N (%)
Growth	13 (26%)
No Growth	37 (74%)
Total	50 (100%)

Staphylococcus species were determined using a variety of biochemical techniques. The coagulase-negative Staphylococci were identified as S. epidermidis and S. saprophyticus by the results of the biochemical tests. They made up 38% of all the isolates combined. S. aureus had a 62 percent coagulase-positive Staphylococci proportion (Table 3).

Table 3: Frequency and percentage of isolated Staphylococcus species

Staphylococcus Specie	Female	Male	Total
Coagulase +ive	3	5	08
S. saprophyticus	2	1	03
S. epidermidis	0	2	02
Percentage(%)	38.46%	61.53%	100%

Eight of the 13 positive samples—05 in men and 03 in females—were coagulase-positive. 02 samples from men tested positive for S. epidermidis. S. saprophyticus was detected in one male sample and two female samples. Eight samples out of the 13 positives, or 62% of the total, were from male patients. 38 percent or 5 samples out of the 13 positive tests were obtained from female patients. While 17 of the 20 samples taken from patients who were female and 20 of those who were male were negative (54 and 46 percent, respectively) (Table 4 & 5).

Table 4: Distribution of patients into age groups.

Age	Females	Males	Total
15-30	5	8	13
31-45	4	7	11
46-60	11	10	21
61-75	2	3	5
Total	22	28	50

Table 5: Number and percentage of positive and negative blood cultures yielding Staphylococcus species for both males and females

Patients No	Blood culture results			
	Positive		Negative	
Male	8	61.5%	20	54%
Female	5	38.5%	17	46%
Total	13	100%	37	100%

Patients with culture growth had neutrophilic leukocytosis and an increased TLC count, whereas the 37 samples without culture growth displayed normal TLC levels (Table 6).

Table 6: TLC count in infected patients

TLC	Number of patients N (%)
11000-15000	05 (39%)
16000-19000	06 (46%)
>190000	02 (15%)
Total	13 (100%)

DISCUSSION

Since the introduction of maintenance hemodialysis, nosocomial infections have been the most frequent side effect in patients who get dialysis. In a larger sense, dialysis patients are not the only ones who can have this problem. The rate of glomerular filtration for patients with infection problems that result in hospitalisation was less than 45 mL/min 1.73 m2[20]. Infection rates rise when renal functions deteriorate, clearly in individuals 60 years of age or older. This is brought on by the immune system's inefficient operation as well as the retention of too much uremic poison. Neutrophils, T-lymphocytes, and monocytes have been shown to have decreased effectiveness[21]. According to reports, in particular, the granulocytes' reduced phagocytic activity renders the environment more conducive to and less resistant to pathogenic bacteria[22]. Additionally, the majority of people with chronic kidney failure have diabetes or are on immunosuppressive medication for a variety of autoimmune diseases. All of these factors lead to a rise in genitourinary infections and a decline in renal function. However, bacteremia and lung infections are considerably more prevalent and make a considerable contribution[20].

Numerous studies' findings indicate that hemodialysis patients have significant incidence of blood infections. The majority of these studies demonstrated that S. aureus predominately serves as a risk factor for developing bacteremia[23].

The length of the catheter seen in our study is found to be comparable with the findings of other studies that have been published in this area and showed a substantial decrease in catheter colonisation[24]. It can also be contrasted with research presenting the findings of previous studies with transient dialysis catheters[25]. Our colonisation rate is lower than originally estimated. These differences may be explained by various research demographic or study design-related factors. For instance, a research by Harb et al. found that for the 79 dialysis catheters implanted in 47 critically sick patients with chronic renal disease, the colonisation incidence was only 5.4 per 1000 catheter days. Approximately 82% of implants were placed at the femoral site. Although it was not stated, it is probable that this cohort did not include bed-bound patients with recently diagnosed CKD[26]. In our

investigation, we discovered that the likelihood of catheter-related infections in dialysis patients is unaffected by gender. Other elements may have a substantial role in the transmission of diseases but have little or no bearing on the patient's gender. We were able to determine that individuals who had infections connected to catheters had significant levels of TLC. According to a published study, people on hemodialysis have considerably lower TLC counts[27].

When compared to alternative interpretations, this might also mean that using ultrasound during the process to introduce guidance would reduce the number of tries. By doing this, the patients' risk of developing septicemia would be reduced even further. According to our research, more efforts would lead to a higher frequency of infections in these people. To lower the prevalence of infections, more appropriate training and handling of the operation should be implemented. Our research has shown that these people can become infected by other pathogens in addition to S. aureas. There may be more germs that can infect these people and cause septicemia. The handler, instrument contamination, or the skin might be the source of the microorganisms. Antibiotic medication given as a preventive measure can lessen these individuals' risk of infection. For such individuals, the catheter with the antibiotic should be made available and utilised. This can lessen the likelihood that contaminated tools will transfer germs to such individuals. Hygiene practises can reduce bloodstream infections in dialysis patients[28]. The most significant of these precautions is hand cleanliness. Strict compliance and ongoing training can increase staff compliance, which helps to lower bacteremia rates[29]. Other straightforward procedures include cleaning a catheter's exit with chlorhexidine and decontaminating the catheter hub with 70% alcohol will lessen bloodstream infection and the transmission of these germs. These actions have already shown notable advantages for individuals with central venous catheters. Use of antimicrobial catheter closure solutions might be included as well[30].

CONCLUSION

This study found that hemodialysis patients had a moderate rate of Staphylococcus species isolation. Despite the fact that all of the HD patients included in this study had bacteremia symptoms, the majority of them had negative blood culture results, which suggests that a pathogen other than Staphylococci or an anaerobic pathogen may be to blame.

- To verify the accuracy of the study's findings, surveillance must be carried out and a sizable sample size should be enlisted.
- the implementation of more research to separate and pinpoint several BSI-causing substances in HD patients.
- It is advised to employ both aerobic and anaerobic blood culture medium when the examination of BSI in HD is asked to give a chance for the isolation of a variety of different bacteria.

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