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Prevalence of Staphylococcus Aureus Infection in Blood Culture of New Borne among Kirkuk City

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Abstract: Background: Staphylococcus aureus is a common cause of infection of the bloodstream in new borne, there is insufficient based on population's evidence available.

Objectives: This study aims to investigate the distribution of S aureus bacteremia in new borne.

Materials and Methods: They conducted based on population's surveillance of every S. aureus bacteremia as in neonates with blood culture-proven neonatal sepsis. Blood samples a prospective, cross-sectional study was carried out on from 80 different sources (newborns) have been utilized in the present study with blood culture-proven neonatal sepsis. They were collected from new borne that frequented hospital in Kirkuk City. They were categorized according to the age of onset of symptoms as having early-onset sepsis and late-onset sepsis. The bacterial profile that resulted from their blood cultures was analyzed.

Results:

This study included eighty clinical samples from new borne who visited the Kirkuk City hospital. Including 50 (62.5%) staphylococcus aureus detected on mannitol salt agar. While 20 (25%) isolate other bacterial spp. and 10 (12.5%) show no growth on blood agar. Each of the fifty S. aureus isolates appeared resistance to Augmentin, Tetracycline, Trimethoprim, and Azithromycin, where as 80% were responsive to Cefotaxime and Chloramphenicol.

Conclusions: S aureus bacterial infections is a significant source of mortality in the pediatric population. Healthcare disorders and devices that are inserted pose major risks. Methicillin-resistant S. aureus has an extremely low rate of mortality.

Keywords: Staphylococcus aureus, newly born specimens, Blood culture, Bloodstream infection.

Introduction:

Staphylococcus aureus is known as the primary cause of aggressive illness in children. *Staphylococcus aureus* is a naturally occurring bacteria found on saturated portions of the epidermis and intestinal the membrane, primarily in the anterior nares of humans. If the organism is transferred to a sterile place on the body, it can produce an isolated or extensive widespread infection. The isolates of S. aureus were initially discovered to be tolerant of beta-lactam antibiotics; however, resistance to these antibiotics developed over time (1). The main steps are beta-lactamase manufacturing and the acquisition of the mecA gene. The incidence of bacteria with antibiotic resistance and genotype in severe infections has increased substantially in the past few years (2).

After the development of penicillin in 1940, bacterial infections decreased. However, Staphylococcus aureus produced β-lactamase, which eliminates the penicillin β-lactam

fundamental ring (3). Methicillin medicines were developed to combat resistance to penicillin due to genetic differences in the β-lactamase enzyme. Methicillin were effective in treating *S. aureus* infections through 1961, after the first variant of S. aureus that was resistant to methicillin (MRSA) was discovered. MRSA has since migrated to the world's nursing homes and hospitals (4). New borne immune systems are weakened, making them susceptible to infection, especially infections of the skin and soft tissues such as New borne area impotence, and severe infections. As therefore, hospital-associated infections caused by MRSA have arisen as a major concern, mostly due to treatment failure. S. aureus was found in the skin and mucosa of up to 40% of patients with New borne, with 30% suffering from severe dangerous shock. (5). Neonatal sepsis isa systemic infection that typically affects sterile body fluids and is accompanied by hemodynamic abnormalities and other clinical symptoms. It results insignificant morbidity and mortality and is primarily caused by bacteria and viruses;though,it can also be triggeredby fungi and parasites.1,2Because of the increasing number of premature babies, who are more vulnerable to neonatal sepsis because of their weakened innate immune function and the necessity for invasive procedures to maintain their life support, the incidence of neonatal sepsis is anticipated to rise internationally. (6). The present research investigates particular epidemiological features and trends in the frequency of Staphylococcus aureus circulatory infection in newborns, with the goal of finding techniques to further lower the possibility of infection.

Aims of study

1-Identification and isolation of *Staph. aureus* from newly born specimens of patients in Kirkuk City.

2- Examination the susceptibility of *Staph. aureus* to a variety of the antibiotics**.**

Materials and methods

Collecting blood samples

Blood samples from 80 various sources (new born) were used in the current investigation. They were obtained from new born who visited Hospitals in Kirkuk City between end of April 2025 and the end of August 2025 by blood sample with media, and grown on agar from the blood and infusions of brain hearts agar. Cultivated samples were incubated under aerobic conditions at 37 °C for 24 hours, and isolated growing colonies demonstrated characteristic development, color, and hemolysis, while suspicious colonies were cultivated for further analysis.

Isolation and Identification of Staphylococci

Colonies with a homogeneous, round, convex shape were selected and subjected to the following tests:

Microscopic Examination: The morphological characteristics of the cells were studied by taking smears stained with Gram stain and prepared for direct microscopic examination to observe the shape, color, regularity, and reaction of the cells to the stain.

Biochemical Tests: Before conducting the tests, the bacteria were activated on the liquid brain and heart infusion medium or nutrient broth. The following are the tests performed for diagnosis:

1- **Catalase test**: The test was performed by transferring a portion of the bacterial colony grown on solid nutrient medium at 24 hours of age using sterile wooden sticks to A dry, clean transparent slide containing a drop of 3% hydrogen peroxide solution. The presence of bubbles in the air signifies a positive test (7).

2- Growth on mannitol saline medium: The bacterial suspension was cultured on solid mannitol saline medium and incubated at 37°C for 24 hours. Growth indicates the bacteria's ability to tolerate 6.5% sodium chloride. A positive result for mannitol fermentation is indicated by a change in the color of the medium from pink to yellow (Macfaddin, 2000) (8).

Coagulase and Clumping Factor Detection: The test tube method was used to detect the coagulase in blood plasma. Bacteria were grown in a liquid brain-heart infusion medium at 37°C for 24 hours. 0.1 ml of the bacterial culture was mixed with 0.5 ml of undiluted normal human plasma in a sterile glass test tube and incubated in a water bath at 37°C for 4 hours, with monitoring every 15 minutes. Positive results were recorded when a clot formed and the plasma solidified (8). While the clumping factor was investigated using the slide test method, two drops of plasma were placed on both sides of the slide, and bacterial growth was added to one of them, while a drop of normal saline was added to the other for comparison (control). The occurrence of any clotting of plasma compared to the control is evidence of a positive test (8).

Antibiotic Susceptibility Test:-

Preparation of Culture Media:-

The Muller-Hinton medium was used for the purpose of this study. The medium used had been reduced to 45-50 degrees Celsius and a suitable amount of autoclaved medium was poured into petri dishes.When the medium solidified, the petri dishes underwent incubation at 37°C to remove any surplus humidity.

Ethical approval

This study followed the ethical principles outlined in the document known as the Declaration of Helsinki. While taking the sample, the participant provided informed written and verbal consent after the local ethics committee reviewed and approved the study methodology and subject information. According to the mission facilitation letter issued by the Kirkuk Health Department No. 309 dated 4/28/2025 to Azadi Teaching Hospital.

Analytical statistics

SPSS 22 was used for statistical analysis. The continuous variable format was means ± SE. The link between categorical factors was examined. P-values under or equle 0.05 were significant.

Results

Bacteriological results

Identification of *Staphylococcus spp.* aerobic bacteria (gram positive)

Staphylococcus aureus strains could grow on mannitol salt agar medium, as illustrated in Figure (1). Because it could sustain a high sodium chloride concentration of approximately 7.5%. In addition from staphylococci, other kinds of bacteria will be inhibited, and the phenol red indicator showed that Staph. aureus metabolizes mannitol and produces yellow regions in the reddish in color agar because of to the the fermentation process and generation of acidic compounds that are accountable for decreasing the pH of the medium, causing the the phenol red to turn yellow. Staph. aureus generates large, smooth, spherical clusters with a full border. aureus responded favorably to Gram stain, while cocci were clustered in couples or groups, resembled grapes, and produced no spores. All the staph aureus isolates were positive for catalase. Mannitol salt agar (positive reaction), motility (negative reaction), beta-hemolysis, and coagulase (all positive). Oxidase (negative response).

Figure (1): *Staphylococcus aeures* bacteria on (A) blood ager (B) Mannitol salt agar at 37˚C for 24hr

Demographic and clinical characteristics of the neonate's no.80

Eighty cases were included in the study. Table 1 details the demographic and clinical characteristics

Table 1 Distribution of neonate according to different variables

Variables	Values
Age, days Mean±SD	15.76±5.2
Sex Male Female	 40(50%) 40(50%)
Gestational age Term Preterm	 50 (62.5%) 30(37.5%)
Mode of delivery Vaginal Cesarean section	 20(25%) 60(75%)
Birth weight Normal Low	 50 (62.5%) 30(37.5%)

Identification and Characterization of Staphylococcus aureus.

This investigation included eighty clinical specimens from people who frequented Kirkuk City hospitals. Containing 50 (62.5%) staphylococcus aureus detected on mannitol salt agar. While 20 (25%) identify additional bacterial spp. and 10 (12.5%) show no growth on blood agar The source of microorganisms transmitted as a sample of blood.

Table 2 Prevalence of different isolates percentage

bacterial isolates	Number of Positive isolates (%)
S. aureus	50(62.5%)
Other bacterial spp.	20(25%)
No growth	10(12.5%)
Total	80(100%)

***S. aureus* samples are susceptible to six various antibiotics.**

Of the total S. aureus isolates (n = 50), 100%, 100%, 80%, and 60% were resistant to Augmentin, Tetracycline, Trimethoprim, and azithromycin, respectively, while 80% were susceptible to Cefotaxime and Chloramphenicol. As demonstrated in the Table.

Table 3 : Antibiotic susceptible profile of methicillin-resistant *S. aureus* isolated from patient

Antibiotics	Resistance		Sensitive	
	NO.	%	NO.	%
Augmentin	50	100	0	0
Azithromycin	30	60	20	40
Cefotaxime	10	20	40	80
Chloramphenicol	10	20	40	80
Tetracycline	50	100	0	0
Trimethoprim	40	80	10	20

Leukocyte counts and differential WBC counts in the studied neonates no.80

Table 4 lists the leucocyte counts

Variables	Values
WBC count ×109/L Mean±SD	18.61±7.33
Neutrophil ×109/L Mean±SD	10.78±4.71
Lymphocyte×109/L Mean±SD	6.5±1.8
Mode of delivery Mean±SD	20(25%) 60(75%)
N/L ratio Mean±SD	3.17±1.90

Discussion

These variations could be attributed to the differences in sample size, age of the onset of sepsis, and general characteristics between different populations. The majority of neonates in this study were diagnosed with late-onset sepsis (70.11%), which was supported by Ferreira *et al.*(9) (82.2%), Almohammady *et al.*(10) (58.6%), Mohakud et al.(11) (72.2%), All these papers have chosen the same neonatal characteristics as used in the current study.

Regarding sex, the present study found that same females had and males. These outcomes are similar to Ferreira et al.'s findings (9).

In addition, Adane *et al.*(12) in Ethiopia found that The mean WBC count was 17.52 ± 6.58 × 109/L, which is slightly higher, while Abdul-Rahman et al.(13) in Erbil showed that elevated WBC counts more than 10 × 109/L were found in most cases (90%). This study also demonstrated that the absolute neutrophil and lymphocyte counts were 9.64 ± 5.68 × 109/L and 5.6 ± 2.4 × 109/L, respectively, which were higher than the results of Adane et al.(12).

S. aureus, which causes sepsis in newborns, has biochemical pathways that allow it to resist medicines. The bacteria can develop drug resistance through various methods (14). Some patients' resistance to all of these antimicrobial medicines against bacteria that are pathogenic is increasing as a result of their long-term usage of these medications. Bacteria are subjected to alterations in DNA/rRNA, which enable them to withstand. The cell wall of bacteria also plays a critical role as an entry point and aids in their survival; however, due to changes in DNA or

mutations in gene expression, the ingredients of the walls of cells or plasma membranes can alter, which supports the phenomena of resistance (15).

Makolo *et al.* (2022) investigated the resistance patterns of Staphylococcus aureus, which was collected from various sources in Nigerian health clinics. The bacteria were resistant to amoxicillin (63.0%) and chloramphenicol (52.4%). (16)

Mahfouz *et al.* (2023) identified susceptibility of the bacteria Staphylococcus aureus to amoxicillin (80.2%) and azithromycin (57.1%) (17).

The strong tolerance to these types of antibiotics may not be solely owing to a enzyme called enzyme synthesis, but also due to reduced attraction of target PBPs or drug penetration into cells. Beta lactam resistance can occur through a variety of methods. In resistant strains, a type of enzyme called beta-lactamase is present, which helps to 'break' the beta lactam ring, effectively nullifying the antibiotic's effectiveness. S. aureus, which traditionally produce penicillinase and ephalosporinase, eliminate the antibacterial agent before the situation can have a positive impact (17). Because of the increasing popularity of beta-lactam medicines, overuse, prescribing, and overdosing have allowed S. aureus isolates to evolve counter-measures to traditional pharmacological therapy, posing a development challenge.

The most prevalent clinical manifestations of disease in our pediatric patients with S aureus bacteremia were bones and joint inflammation, followed by epidermis and connective tissue infections. This is comparable to numerous other pediatric cohorts (18). In around one-third of our patients, no infection focus was found. An Australian case series (19) found musculoskeletal and joint infection in 59% of otherwise healthy neonates, with atherosclerosis occurring in only 1.4% of neonates versus 30% of adults. In our investigation, the population with no recognized focus of infection looked comparable to the New Zealand dataset if one combined rates for patients they treat with no determined focus with the individuals with intravenous catheter infections (27%). (20).

Conclusions

S. aureus infection represents a significant problem in the pediatric population. Infants with background risk factors such as having central venous devices, cancer, or chronic illness. People with illnesses are nevertheless more likely to get this deadly infection.

While tons of kids may have been diagnosed with *staphylococcal* a bacteremia without a specific cause of infection, it is critical to properly investigate the source because the majority of cases are secondary. Fortunately, the mortality rate from invasive *staphylococcal* illnesses in children is substantially lower than in adults. As MRSA becomes more ubiquitous, disease transmission may change, as demonstrated in several nations.

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Conflicts of interest

The authors have declared that no competing interests exist.

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