Staphylococcus aureus and Coagulase-Negative Staphylococci in Bacteraemia: The Epidemiology, Predisposing Factors, Pathogenicity and Antimicrobial Resistance

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Abstract

Staphylococcus species are the predominant Gram-positive organisms obtained from blood culture samples. Its incidence in bloodstream infection among children and adults varies among. *Staphylococcus aureus* is regarded as pathogenic with high morbidity and mortality while coagulase-negative staphylococci (CoNS) are often regarded as a contaminant and not a true cause of bacteremia despite its rising occurrence. Predisposing factors of staphylococcal bacteremia include malnutrition, malaria, HIV/AIDS and nosocomial infections. Methicillin-resistance in *Staphylococcus aureus* and CoNS in bacteremia is associated with an increase in multidrug-resistant virulent strains when compared to methicillin-sensitive *S. aureus* or CoNS. The impact of coagulase-negative staphylococci in bacteremia is on the increase with marked clinical complications. However, this review summarizes the prevalence and epidemiology of *S. aureus* and CoNS in bacteremia based on research outcomes in Africa, Asia, Europe, and North America.

Keywords: *Staphylococcus aureus*; Coagulase-negative staphylococci; Bacteraemia

Introduction

Staphylococci species are members of the family Micrococcaceae known to be Gram-positive cocci occurring singly, in pairs, short chains and as rough grape-like clusters. They are catalase-positive and also differentiated based on their ability to produce coagulase, causing visible agglutination on slide and tube coagulase test [1].

Staphylococcus aureus bacteremia is defined as the microbiological presence of *S. aureus* in the bloodstream. It is characterized by high morbidity, mortality and associated with frequent use of intravascular devices, invasive surgeries and immunodeficiency in patients. It has become a leading cause of hospital and community-acquired bacteremia [2]. In intensive care units, *S. aureus* is the most common Gram-positive pathogen isolated in bloodstream infections [3].

Coagulase-negative staphylococci (CoNS) are found as normal flora on the skin and mucous membrane of humans [4]. It is commonly reported in very-low-birth-weight neonates, immune-compromised patients, and individuals with indwelling intravascular devices or implanted medical devices [5]. Being a commensal on the human skin, CoNS can be transmitted from the hands of medical staff and caregivers especially in children [6].

In neonates, CoNS accounts for about 70% of the positive blood culture, although they are mostly regarded as contaminants [7]. True bacteremia with CoNS occurs mostly in preterm neonates below 32 weeks gestation based on their under-developed immunity. Obolski et al. pointed out that before CoNS are considered as contaminants, they should be tested for resistance [8].

Clinical diagnosis of CoNS bacteremia involves a positive blood culture, a medical picture of health deterioration and an acute phase inflammatory marker usually C-reactive protein or two positive CoNS blood cultures collected consecutively within 48 hours or a positive blood culture in mixed growth with significant pathogenic bacteria [7,9]. In a study by Elzi et al., the clinical significance of CoNS as a true pathogen using a single positive blood culture was evaluated based on the presence of a central venous catheter and two Systemic Inflammatory Response Syndromes (SIRS) or at least three SIRS [10]. Some reports from sub-Saharan Africa brings to light that CoNS bacteremia is having an upsurge in occurrence and clinical significance [11].

Some study assumes that the high incidence of CoNS is due to the usage of 70% alcohol alone in skin cleaning before venipuncture instead of in combination with 10% povidone iodine [12]. Archibald et al. indicated an increased contamination rate with povidone-iodine [11].

DISCUSSION

Epidemiology of staphylococci colonization

Staphylococcus aureus can be obtained from human and animal hosts. It is highly clonal in nature with variants that can colonize specific or multiple hosts [13]. Molecular typing aids in the understanding of *S. aureus* epidemiology. Coagulase-negative staphylococci species are normal flora on the skin. Differentiating CoNS bacteremia infection from contamination in blood culture specimen poses some difficulty since CoNS is a widespread skin commensal [5]. There are about 38 recognized CoNS species with 13 colonizing humans e.g. *Staphylococcus saprophyticus, Staphylococcus*

haemolyticus, Staphylococcus hominis, Staphylococcus warneri, Staphylococcus xylosus, Staphylococcus epidermidis, Staphylococcus schleferi, and *Staphylococcus lugdunensis* [14]. The common CoNS species isolated clinically and often considered for its virulence and pathogenicity is *Staphylococcus epidermidis* [5,15,16].

A study on establishing the significance of bacteremia in 170 Ethiopian children with severe malnutrition showed that about 20.6% of the children yielded bacterial isolates with *S. aureus* (28.6%) and coagulase-negative staphylococci (22.9%) having a high frequency among other isolates (Figure 1). The finding indicates that CoNS species are posing great health problems and it is consistent with the statement by von Eiff et al, that the fraction of bloodstream infections caused by CoNS and the total incidence of true CoNS bacteremia are increasing [17].

Most bloodstream infection yielding coagulase-negative staphylococci are often regarded as contaminants despite their predominance as Gram-positive isolates in blood culture. A study on bacteremia among 140 Gambian children aged under 5 years with severe acute malnutrition showed that out of the 38 positive blood culture isolates obtained, CoNS yielded 50% and *S. aureus* 0% apart from other Gram-positive and negative bacterial isolates observed [18]. Despite the high percentage of CoNS, they were regarded as contaminants resulting in no further species identification and antibiotic sensitivity analysis. But a study by Onipede et al. considered CoNS as a true isolate in bacteremia and did antimicrobial susceptibility testing, regardless of the low percentage of CoNS (0.5%) in the blood cultures [19].

This varies from the study by Obolski et al., where CoNS (31.5%) was tested for antibiotic resistance before being regarded as contaminants [8]. *Staphylococcus aureus* yielded 4.2% among other significant bacteria. Their findings on CoNS resistance suggested that it could predict antibiotic resistance in subsequent bacteremia with a known pathogen, affect the mortality rate and also guide clinicians in their choice of antibiotic therapy.

A collaborative retrospective study was conducted on blood culture samples from different hospitals in Tanzania, Malawi, and the United States. Out of 54 blood cultures obtained in a medical facility in the United States that were positive for bacterial growth, CoNS and *S. aureus* yielded 44.4% and 27.8% respectively; although, CoNS was regarded as contaminants [11]. High aseptic standard maintained during venipuncture and blood culture inoculation in the study at Tanzania resulted in decreased contaminant, with *S. aureus* and CoNS occurring as 2.5% and 0.6% respectively [20]. The study in Malawi had CoNS, specifically *S. epidermidis* as 0.4%, which was indicated as contaminants [21].

The detailed study on staphylococcal isolates carried out in 6 healthcare facilities in Ghana indicated that about 12.4% were *S. aureus* from Bloodstream Infections (BSI) while 65.9% signifying coagulase-negative staphylococci originated from bacteremia, skin and soft tissue infection (SSTI) and other infections [22]. Elzi et al. assessed the clinical importance of CoNS bacteremia using the Systemic Inflammatory Response Syndrome (SIRS) with 35% identified as true bacteremia out of 654 CoNS isolates [10]. The SIRS criteria used include temperature >38°C or <36°C, respiratory rate >20/min, heart rate >90/min and white blood cell count >12 G/L or <4 G/L.

Predisposing factors

Malnutrition in sub-Saharan African and South Asia is one of the widespread causes of childhood morbidity and mortality, with severe acute malnutrition affecting roughly one-tenth of children under 5 years globally. Resultant death of over 50% is associated with children living in abject poverty and unbalanced diet, and disease prognosis worsens with HIV/AIDS infection [4]. Growth retardation, muscle wastage and weight loss are common features of childhood malnutrition. Susceptibility to infection increased due to lowered immune defense mechanism and often makes clinical diagnosis and treatment difficult by concealing symptoms of other infectious diseases.

In Ethiopia, about one in two children below five years have stunted growth, muscle wastage and reduced weight for age especially among children in rural areas [4]. Staphylococcal species were the main cause of bacteremia in the malnourished children with CoNS species occurrence being possibly linked to immune suppression in severely malnourished children. Standard use of antibiotics for empirical therapy is proposed by WHO for children with severe acute malnutrition since they are predisposed to bacteremia, diarrhea, urinary tract infection and pneumonia [18].

Malaria in African children below 5 years is about 90% on a scale of approximately 500 million cases reported yearly globally. The medical presentation of malaria is closely similar to septicemia but having limited local data on its correlation [23]. A study in Nairobi on admitted febrile children with malaria in the age range of 3 months to 12 years indicated that 18 out of 158 children with malaria had bacteremia while 14 out of 106 had bacteremia but without malaria. Coagulase-negative staphylococci (34.4%) were the predominant Gram-positive bacteria but were regarded as contaminants while *Staphylococcus aureus* yielded 12.5% [23]. However, co-infection of bacteremia with or without malaria appeared to be similar in this study occurring as 11.4% and 13.2% respectively.

Nosocomial infections, which is particularly common in intensive care units are often associated with *S. aureus* bacteremia [5]. It is characterized by high virulence with a mortality rate between 20%-50% (2). Nosocomial *S. aureus* bacteremia is often considered if it appears after 72 hours of hospital admission.

The German nosocomial infection surveillance system (KISS) reported that *S. aureus* yields about 16.3% of the nosocomial infections in the intensive care unit [24]. Methicillin-Resistant *S. aureus* (MRSA) was observed in about 42.9% of hospital-acquired bloodstream infection when compared to 7.9% community-acquired MRSA [12].

Recent trends have revealed the importance of CoNS infection as nosocomial pathogens, depicted by the significant rise in morbidity, mortality and healthcare cost in pediatric and neonatal care unit [25]. The study by Blot et al., showed that 0.38% were diagnosed with *S. aureus* bacteremia [2]. Approximately 17.6% of the admitted patients developed *S. aureus* bacteremia within 72 hours and did not have their main diagnosis as septicemia at the time of admission.
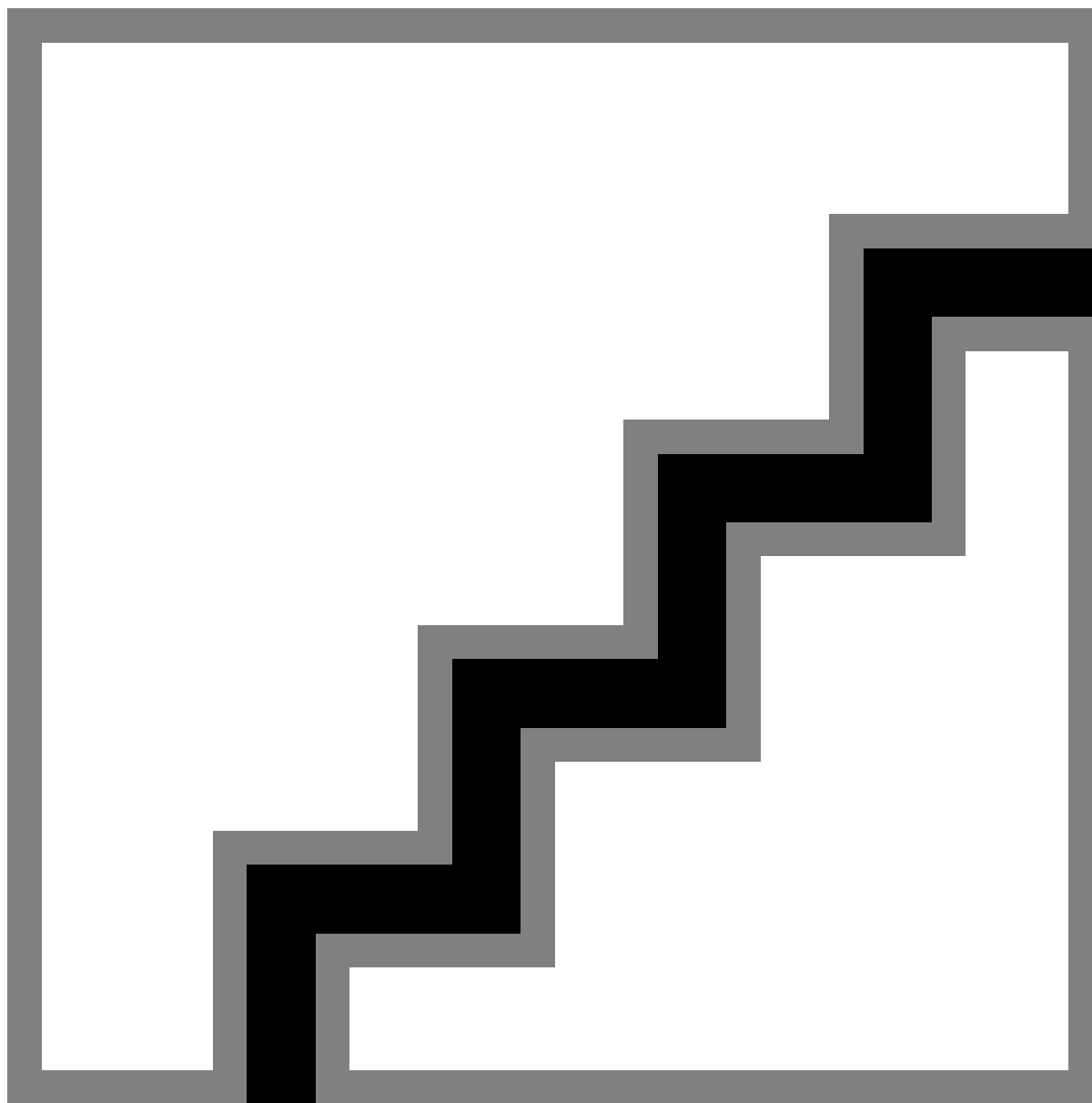
Figure 1: Frequency of *S. aureus* and CoNS in bacteremia.

Pathogenicity and virulence

Staphylococcal species have different classes of surface proteins based on their structural and functional differences. These proteins are covalently bound to the peptidoglycan layer of the cell wall. It is an important virulence factor necessary for their survival as commensals or pathogens [26]. The functions of surface proteins include cell adhesion, invasion, and colonization of host tissues, evading of the immune response and production of slime that aids their adherence to indwelling medical devices such as valvular prostheses, central and peripheral venous catheters, artificial heart valves, pacemakers, and orthopedic prosthesis. CoNS produce proteases, lipases and other exoenzymes that enable it to thrive and possibly degrade host tissues [27].

Staphylococcus aureus produces a vital and unique virulence factor known as thermostable nuclease (TNase), which is encoded by the *nuc* gene [28]. Conversely, other coagulase-negative staphylococci strains such as *S. simulans, S. caprae, S. intermidus, S. carnosus,* and *S. capitis* produce TNase but can be differentiated from TNase produced by *S. aureus* using the monoclonal antibody and nuclease-based test [29].

It is important to note that most MSSA isolates from Africa harbor the Panton-Valentine Leucocidin (PVL) genes: *lukS-pv* and *lukF-pv*, about 55% more than the rest of the world [22]. This may be linked to the interrelationship between the host, the microorganism and the environment [30]. The genetic material of a bacteriophage that infects *S. aureus* produces the PVL, thereby making it more virulent. A study on *S. aureus* isolates indicates that there are some genotypes and virulence factors that are widespread in Africa due to staphylococcal

infection than from colonization; e.g. PVL encoded by the *lukS-pv* and *lukF-pv* genes [31].

In a study by Giormezis et al, CoNS isolates were tested for antimicrobial susceptibility, biofilm formation, *mecA, ica* operon, adhesin (accumulation-associated protein aap and biofilm-associated protein bap) and toxin genes [32]. Approximately 58% produced biofilm; 7% carried the *ica* operon which encodes the production of polysaccharide intercellular adhesin; 30% carried the adhesin genes while 99% carried the *mecA* gene and showed significant multidrug resistance.

Among the CoNS species, *Staphylococcus epidermidis* has had more research conducted on virulence factors such as biofilm formation, adhesion and aggregation. Adherence of CoNS to the host skin or indwelling medical device is the first step to colonization or infection [5]. This can occur directly or by means of extracellular matrix components and the host plasma protein components, specifically collagen, fibronectin, fibrinogen and heparin-related polysaccharides. Microbial surface components recognizing adhesive matrix molecules (MSCRAMMs) are adhesins expressed on bacteria cell surface that mediates adherence to the host [32].

Biofilm formation is an important factor in the pathogenesis of staphylococcal infections, which confers increased metabolic, mechanical, immune and antibiotic resistance. It is a microbial-derived sessile community comprising cells which are irreversibly attached to an interface or each other and embedded into the extracellular polymeric matrix that they produce [32-34]. The production of staphylococcal toxin such as enterotoxins and toxic shock syndrome, which act as superantigens toxin are vital to its pathogenesis. They bind the major histocompatibility complex class II directly, thereby triggering an increase in the T-cell stimulation and excess production of cytokine [32].

Antimicrobial resistance

The main public health problem associated with high mortality, longer hospitalization, and exorbitant treatment cost is antibiotic resistance. For effective analysis of bacteremia, the collection of blood cultures should precede empirical treatment and modified based on antibiotic susceptibility and resistance pattern [8]. Bacteremia caused by methicillin-resistant *S. aureus* results in greater mortality with an increased tendency of resistance to various antimicrobial agents when compared to Methicillin-sensitive *S. aureus* (MSSA) bacteremia [35]. The study by Gastmeier, et al., showed increased mortality [24] with MRSA (16.8%) when compared with MSSA (6.0%). It also results in increased healthcare cost with limited options for treatment due to multiple drug resistance [36].

Methicillin resistance depicted by the presence of the *mecA* gene in *S. aureus* and CoNS varies between different studies. This gene encodes the penicillin-binding protein with decreased affinity to β-lactam antibiotics [32].

In Africa, antimicrobial resistance in *S. aureus* is emerging, specifically methicillin-resistance in varying genetic backgrounds [30]. This genetic diversity has led to the emergence of 6 major clones globally with rising spread across to Africa. The clones are referred to as staphylococcal cassette chromosome SCCmec type I-VI [37]. A survey on bacteremia in South Africa reported about 23% prevalence of MRSA among the *S. aureus* isolates [36]. The majority of CoNS isolates have the *mecA* gene and an increase in multidrug-resistant virulent CoNS strains causing difficulty to treatment [32].

Vancomycin, a glycopeptide antibiotic is used as the main therapy but has shown resistance [5]. A typical African MRSA clone observed in some studies is the ST88-MRSA-III/IV.

Although a retrospective study by Blot et al., was carried out on critically ill adults aged between 49 to 70 years within a 7-year period in a Belgium hospital (2), the study showed that the mortality rate in patients with MRSA bacteremia was high (63.8%) when compared to MSSA (23.7%). However, the patients with MRSA bacteremia were elderly, sickly and more debilitated. There are conflicting data in comparing MRSA and MSSA bacteremia with varying effect on fatality rate.

A common feature in staphylococcal research in Africa is the high level of resistance to penicillin among other antimicrobial agents; this is encoded by the *blaZ* gene [31]. According to a study by Alem et al., all the *S. aureus* and CoNS isolates obtained showed resistance to penicillin, oxacillin, ampicillin, and chloramphenicol. Few of the isolates also showed resistance to erythromycin, tetracycline, ciprofloxacin, cotrimoxazole, vancomycin, and ceftriaxone in varying proportion [30]. This agrees with the high percentage of multi-drug resistant *S. aureus* observed by Onipede et al. [19].

Conclusion

Staphylococcal isolates, especially *S. aureus* are a major cause of bacteremia in malnourished children. The impact of CoNS in bloodstream infection is rising with major health complications, thereby indicating its possibility of clinical significance in bacteremia. The need to identify the CoNS species and determine their pathogenic potential instead of totally disregarding them as contaminants will give a better understanding of their role in true bacteremia and also aid clinical management.

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