UNIVERSITY OF CAPE COAST

ANTIBIOTIC SENSITIVITY PROFILES AND RESISTANT GENES OF *STAPHYLOCOCCUS AUREUS* ISOLATED FROM THE NASOPHARYNX OF CHILDREN IN CAPE COAST, GHANA

PRINCE SEDEM SEMANSHIA

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BY

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Thesis submitted to the Department of Microbiology and Immunology of School of Medical Sciences, College of Health and Allied Sciences, University of Cape Coast, in partial fulfillment of the requirements for the award of Master of Philosophy degree in Infection and Immunity

AUGUST 2023

DECLARATION

Candidate's Declaration

I hereby declare that this thesis is the result of my own original research and that no part of it has been presented for another degree in this university or elsewhere.

Candidate's Signature …… Date ……

Name: Prince Sedem Semanshia

Supervisors' Declaration

We hereby declare that the preparation and presentation of the thesis were supervised in accordance with the guidelines on supervision of thesis laid down by the University of Cape Coast.

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ABSTRACT

Staphylococcus aureus, a bacterium with global significance, is responsible for substantial morbidity and mortality, sparking considerable interest in understanding its interaction with humans. As a significant opportunistic pathogen, it can cause mild and severe infections. Up to 50% of individuals may asymptomatically carry *S. aureus* in their anterior nares, serving as a reservoir for contact transmission and endogenous infections. Children, especially those in overcrowded school and preschool environments, play a crucial role in the community dispersal of *S. aureus*, including methicillin-resistant strains. Unfortunately, treating staphylococcal infections has become challenging due to multiple drug resistance (MDR). This research, conducted in Cape Coast, Ghana, aims to assess the carriage rate and antibiotic resistance profiles of *S. aureus* isolated from 880 nasopharyngeal samples from children. Results showed a carriage rate of 16.5%, with age being a significant risk factor. Of the isolates, 37.2% exhibited MDR, including 29.7% methicillin-resistant *S. aureus* (MRSA). Molecular analyses revealed genotypic-phenotypic correlations for antibiotic resistance, and specific resistance genes were identified. These findings provide critical data on *S. aureus* carriage in children and inform empirical treatment approaches for *S. aureus* infections. The study emphasizes the importance of prudent antibiotic use in combating antibiotic resistance.

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DEDICATION

To Almighty God; my lovely parents, Mr. Raymond Semanshia and Mrs. Evelyn Ziggah; and my uncle, Mr. Ashiboe-Mensah.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ACRONYMS

β-lactam	Beta-lactam
CA-MRSA	Community-Associated Methicillin Resistant *Staphylococcus aureus*
CDC	Centers for Disease and Control
CLSI	Clinical and Laboratory Standards Institute
CONs	Coagulase-Negative Staphylococci
DNA	Deoxyribonucleic Acid
HAI	Hospital Acquired Infections
HA-MRSA	Healthcare-Associated Methicillin Resistant *Staphylococcus aureus*
MDR	Multi-drug Resistant
MRSA	Methicillin Resistant *Staphylococcus aureus*
MSSA	Methicillin Susceptible *Staphylococcus aureus*
PBP	Penicillin binding protein
PCR	Polymerase Chain Reaction
PFGE	Pulse-field Gel Electrophoresis
PVL	Panton-Valentine Leukocidin
S. aureus	*Staphylococcus aureus*
SCCMEC	Staphylococcal Cassette Chromosome mec
spa	Staphylococcal Protein A
TAE	Tris-Acetate EDTA
TSST-1	Toxic Shock Syndrome Toxin -1
VISA	Vancomycin Intermediate *Staphylococcus aureus*
VRSA	Vancomycin Resistant *Staphylococcus aureus*
WHO	World Health Organization
γ hemolysin	Gamma Hemolysins

CHAPTER ONE

INTRODUCTION

The human body's normal flora plays a crucial role in promoting human health by preventing the colonization of harmful microorganisms, thus impeding the development of diseases. However, it is essential to acknowledge that normal flora can also be involved in disease development and complications. A notable example is Staphylococcus aureus, residing in the nasopharynx, which can cause localized and systemic diseases, particularly in individuals with compromised immune systems. S. aureus is a leading cause of various ailments, including skin and soft tissue infections, community-acquired pneumonia, and outbreaks. The rising antimicrobial resistance poses significant challenges in managing S. aureus-related diseases, necessitating a comprehensive understanding to address knowledge gaps and inform policies on the prudent use of antimicrobial agents in treating infections caused by this microorganism.

Background

Staphylococcus aureus is part of the normal microbial flora of humans. It can dwell in diverse ecological niches within the human body, where it can flourish as an unhazardous microbe or cause infections (Sollid et al., 2014; Tong et al., 2015). In recent years, the relationship between *S. aureus* and humans has earned much interest because the bacterium is liable for substantial morbidity and mortality globally (Clegg et al., 2021). According to Kourtis et al., in the United States of America, *S. aureus* has been approximated to be the reason for 20,000 mortalities and amounts to an overall bill of $15 billion on the health service annually (Kourtis

et al., 2019). *S. aureus* is a major opportunistic pathogen that causes a wide range of clinical infections. These infections include boils, impetigo, cellulitis, bacteremia, pneumonia, and toxic shock syndrome (Geoghegan & Foster, 2017). *Staphylococcus aureus* is a regular instigator of infections in the community and hospital. Despite advances in healthcare treatment, *S. aureus* remains a leading cause of nosocomial infections among hospital patients (Pendleton et al., 2013; Tong et al., 2015).

S. aureus is a facultative anaerobe, Gram-positive cocci, non-motile and non-spore-forming microbe (Clarke & Foster, 2006). *S. aureus* is coagulase-positive, catalase-positive and causes haemolysis when grown on blood agar plates. Some *S. aureus* strains are coated with a polysaccharide layer called the capsule, enveloping their cell surface. Others also form biofilms that promote the persistence of the *S. aureus* at an infection site or surface (Visansirikul et al., 2020). For the development of biofilms, *S. aureus* makes a self-produced extracellular matrix (ECM) composed of carbohydrates, proteins, and extracellular DNA, which sheathes the bacterial cells within a sticky matrix that enables survival in hostile or extreme environments (Periasamy et al., 2012).

S. aureus primarily colonizes the nose of humans; however, it can colonize other body sites, including the skin, perineum, vagina, axillae, pharynx, gastrointestinal tract, urinary tract, and throat (Zhao et al., 2021).

The ability of *S. aureus* to survive in different ecological niches of the human body demonstrates its versatility and diversity in colonizing its host (Mulcahy et al., 2012; Van Belkum et al., 2009). According to Raineri et al., a higher

rate of nasal colonization is discovered in children, estimated to be about 45% in the first weeks of birth (Raineri et al., 2022). Nonetheless, *S. aureus* nasal carriage declines with aging (Wertheim et al., 2005). *S. aureus* prefers the human nose because of the moist squamous epithelium of the anterior nares (Van Belkum et al., 2009). In a recent study, Mulcahy et al. (2012) demonstrated that loricrin, the most abundant protein in the cornified envelope, was the principal target ligand for a surface protein called Clumping factor B (ClfB) during *S. aureus* nasal colonization (Mulcahy et al., 2012). Sakr et al. also reported the interaction between ClfB and cytokeratin 10, cytokeratin 8, and fibrinogen (Sakr et al., 2018). Conversely, *S. aureus* also colonizes the mid regional nares to the deeper regions of the nose. Interaction with the resident nasal microflora also influences *S. aureus* nasal colonization and persistence (Yan et al., 2013). *Corynebacterium pseudodiphtheriticum*, *S. epidermidis*, and *S. lugdunensis* adversely influence *S. aureus* colonization, while *C. accolens* promote *S. aureus* growth (Zipperer et al., 2016). This requires a balance between efficient attachment at the colonization site and resisting the mechanical forces that attempt to dislodge it from those niches. For successful colonization, *S. aureus* should proliferate and overwhelm the defense mechanisms in the nasal cavity (Edwards et al., 2012). Even though nasal colonization is asymptomatic, it remains a significant risk factor for infection (Wertheim et al., 2005). Up to 50% of a given population could asymptomatically carry *S. aureus* in their anterior nares, consequently aiding as a source for contact transmissions and endogenous infections (Warnke et al., 2014). Habits such as nose-picking could be a possibility for transferring *S. aureus* pathogens carried in

the nose to other portions of the human body and also to non-colonized individuals (Wertheim et al., 2006). Methicillin-resistant *Staphylococcus aureus* (MRSA), which also colonizes the nasal cavity of healthy individuals, can sometimes be transmitted to other persons. MRSA carriage in the nose and its ability to cause infection in immunocompromised persons has led to the development of effective nasal decolonization strategies. Intranasal application of antibiotics such as mupirocin has been reported as a means of decolonizing the anterior nares of MRSA (Liu et al., 2011).

Apart from decolonizing asymptomatic carriers, staphylococcal infections are treated with antibiotics. The European Union assessed that MRSA infections affect more than 150,000 patients annually with a cost of €380 million (Köck et al., 2010). In the United States of America, it was published that 80,461 MRSA infections and 11,285 associated deaths ensued in 2011, and an approximate annual burden of $1.4 billion and 13.8 billion was accredited to community-acquired MRSA (CDC, 2013; B. Y. Lee et al., 2013). Antibiotics used to treat staphylococcal infections include ampicillin, penicillin, tetracycline, flucloxacillin, dicloxacillin, cefazolin, clindamycin, doxycycline, and erythromycin (Rayner & Munckhof, 2005a). *S. aureus* resistance to commonly used antibiotics such as penicillin, methicillin, gentamicin, and erythromycin have increased over the years. A study conducted in the Eastern Cape Province of South Africa by Akanbi et al. showed *S. aureus* susceptibility towards doxycycline (50%), tetracycline (56.7%), gentamycin (63.3%), and ciprofloxacin (66.7%) were low. Higher resistance to erythromycin

(70%) and clindamycin (80%, 24/30) was identified, with resistance to penicillin and ampicillin being the highest (each recording 96.7%) (Akanbi et al., 2017)

Increasing incidences of antibiotics resistance to *S. aureus* demand the development of alternative strategies to combat *S. aureus* infections. Unfortunately, the treatment of staphylococcal infections is becoming progressively challenging due to the emergence of multiple drug resistance (MDR). Forbearance to antibiotics is a crucial public-health challenge, and antibiotic patronage is increasingly acclaimed as the principal push to resistance (Goossens et al., 2005a). The global spread of staphylococcal MDR strains has complicated the treatment outcomes of *S. aureus* infections (Holden et al., 2013). Reports of Methicillin resistance in *S. aureus* have surpassed 20% in all World Health Organization (WHO) regions and above 80% in some regions (WHO, 2014). According to Abdulgader et al., Africa has limited epidemiological data on MDR, particularly MRSA (Abdulgader et al., 2015a). National data from 9 African countries shows MRSA rates of approximately between 12% and 80%, with some countries exceeding 82% (Falagas et al., 2013; Gelband et al., 2015). Recorded in Uganda, the MRSA prevalence rates are between 31.5% and 42% among patients and healthcare workers (Kateete et al., 2011), in Rwanda 31 to 82% (Masaisa et al., 2018), and 34% in Dakar – Senegal (Breurec et al., 2011). In Ghana, there is no routine patient or specimen screening for MRSA; there is barely any information on the epidemiology of MRSA and effective surveillance (Donkor et al., 2019).

The genetic basis for antibiotic resistance to commonly used antibiotics has been studied in some locations (Falagas et al., 2013; Wangai et al., 2019). The

excessive use of antimicrobials has caused resistance either by the development of point mutations or by the gaining of foreign resistance genes, which has led to modifications of the antimicrobial target and the degradation of the antimicrobial or reduction of the cell's internal antimicrobial concentration (Akanbi et al., 2017). Alterations in some genes, such as the *pbp* gene, have lowered affinity to β-lactams drugs, thereby conferring resistance in *S. aureus*. For example, methicillin resistance is typically due to the *mecA* gene, borne on the staphylococcal cassette chromosome mec (SCCmec) that codes for a 78-kDa penicillin-binding protein (PBP2a), with decreased affinity to methicillin and all beta-lactam antibiotics (Chambers, 1997). The association between methicillin resistance and resistance to other antibiotics is well documented (Fluit et al., 2001). Kirby first revealed that penicillin was inactivated by penicillin-resistant strains of *S. aureus* (Kirby, 1944). About 90% of all staphylococcal isolates make penicillinase, irrespective of the clinical setting, and the dispersal of penicillin resistance occurs primarily by spreading resistant strains (Lowy, 2003). Staphylococcal resistance to penicillin is facilitated by *blaZ*, the gene that codes β-lactamase. This enzyme is synthesized when staphylococci are subjected to β-lactam antibiotics; it hydrolyzes the β-lactam ring, leaving the β-lactam inactive (Lowy, 2003). Expression of resistance in some MRSA strains is also regulated by homologues of the regulatory genes for *blaZ*. Erythromycin resistance in staphylococcal isolates is mainly facilitated by the *erm* genes, coding for erythromycin-resistant methylase. Usually, there is either a ribosomal modification by 23S rRNA methylases mediated primarily by *ermA, ermB,* or *ermC* or an active efflux of the antimicrobial agent by an ATP-dependent

pump mediated by *msrA* (Nicola et al., 1998). Tetracycline-resistant bacteria acquire tetracycline resistance genes (*tet*). The two main mechanisms of resistance to tetracycline in *S. aureus* are the active efflux, resulting from acquiring the plasmid-located *tetK* gene and ribosomal protection by elongation factor-like proteins encoded by chromosomal or transposonal *tetM* determinants (Esposito et al., 2009). The *tet* genes are contained within conjugative transposons that can be transferred horizontally and expressed in Gram-positive and Gram-negative bacteria (Chopra & Roberts, 2001). In a study conducted in Iran, Emaneini et al. published that the coexistence rate of *mecA* and *tet* genes was 61%, which is slightly higher than similar reports from Europe (57.1%) and lower than those from Japan (100%) (Emaneini et al., 2013; C. H. Jones et al., 2006; Lohan et al., 2021).

S. aureus expresses large, diverse, and robust virulence factors that facilitate the bacteria's ability to interrelate with host tissue and the extracellular matrix components. *S. aureus* virulence factors can be classified into secreted and cell surface factors. The goal of these virulence factors is to (1) adhere to the host cell surface, (2) disperse throughout the host, (3) evade host immune defense, and (4) produce toxins and other products, which can cause damage to the host's cells (Geoghegan & Foster, 2017). Many factors, such as enzymes, superantigens, and membrane damaging toxins, are secreted into the extracellular milieu by *S. aureus* (A. R. Costa et al., 2014). Staphylococcal toxic shock syndrome, which can be fatal, is caused by the activation of the host's T cells by superantigen factors such as toxic shock syndrome toxin (TSST), leading to excessive proliferation and production of cytokines (Otto, 2014). Membrane damaging toxins include proteins such as

Hemolysin-α (α- toxin), Panton-Valentine leukocidin (PVL), Phenol-soluble modulins (PSMs), and gamma-toxin (gamma-hemolysin, HlgA, HlgB, HlgC) (Otto, 2014). *S. aureus* also secretes Staphylokinase, Staphylocoagulase, and Von Willebrand factor (vWF), further influencing its virulence. The exact functions of discrete staphylococcal factors in nasty life-threatening infections are challenging to assess, but PVL is linked with soft tissue infections (SSTI) and severe necrotising pneumonia. It has been revealed to be a distinguishing feature of community-acquired (CA) -MRSA clones disseminated in Europe and Middle East (ST-80), Australia and South America (ST30-IV), and United States (ST8-IV) (David et al., 2011; R. Deurenberg & Stobberingh, 2009; Lina et al., 1999). PVL, γ-hemolysin, and other leukocidins belong to the family of synergohymenotropic toxins (Dey et al., 2013). These synergohymenotropic toxins injure membranes of host immune cells and erythrocytes by the synergetic action of two non-associated classes of secretory proteins. According to Lina et al., gamma-hemolysin is yielded by >99% of *S. aureus* clinical strains, and PVL is secreted by less than 5% of *S. aureus* strains (Lina et al., 1999). However, the prevalence of *S. aureus* strains that produce Panton-Valentine Leukocidin (PVL) is 0.3% in South Africa and 100% in Tunisia (Abdulgader et al., 2015).

Understanding the content and structure of *S. aureus* in respiratory samples will be critical for determining illness risk and developing probiotics that can colonize the respiratory system and contribute to infection prevention (Camelo-Castillo et al., 2019). Antibiotics are commonly prescribed to young children to treat bacterial infections such as *Staphylococcus aureus* invasive staphylococcal

infections. Despite their apparent benefits, little is understood about antibiotics' potential side effects on children's nasopharyngeal microbiota (Henares, Rocafort, et al., 2021). *Staphylococcus aureus* carriage in the nasopharynx essentially ensues in children attending gatherings in schools, churches, mosques, and playgrounds where there is usually crowding. Crowding supports the horizontal spread of bacteria. These children become the primary reservoirs for infections developing in other susceptible individuals, such as individuals with a weak immunity (Kovács et al., 2020). Furthermore, hand carriage and nasal carriage of *S. aureus* are sturdily correlated, proposing that contaminated hands usually initiate the colonization of the nares (Wertheim et al., 2006). The colonization begins at birth, with volatile initial communities influenced heavily by an infant's environmental exposures. The respiratory microbiome changes continuously during the first years of life, eventually achieving an adult-like stabilized structure (Henares, Brotons, et al., 2021). Among European children, studies have shown a declining nasal carriage rate during the first year of life, continuing steady at 20–30% until it upsurges again to 40–50% between the age of 6 to 12 years (Bogaert et al., 2004; Lebon et al., 2008). Several determinants have been recommended to inspire the carriage rate in healthy children. The number of older siblings, family size, breastfeeding, and passive smoking has been found to be associated with *S. aureus* nasal carriage (Bogaert et al., 2004). Additionally, some studies have publicized that carriage of *S. aureus* is inversely correlated to the carriage of *S. pneumoniae* in healthy children (Bogaert et al., 2004; Regev-Yochay et al., 2004). Clarifying the interactions between these different pathogens is essential to assess the overall impact on the

pneumococcal conjugate vaccine (Adegbola et al., 2014). In case of autoinfection or transmission to other children, those highly virulent isolates may cause severe infections in pediatric populations (Eibach et al., 2017). Acute respiratory infections (ARIs) are the second leading cause of death in children under the age of five, accounting for approximately 15% of all deaths in this age group (Henares, Brotons, et al., 2021; WHO, 2020). ARIs are also a significant component of morbidity and one of the primary reasons for antibiotic administration in children. Streptococcus pneumoniae, Haemophilus influenzae, and *Staphylococcus aureus* are common bacterial pathobionts that cause ARIs and are frequent colonizers of healthy children's nasopharynx (de Steenhuijsen Piters et al., 2015).

Problem statement

The nasal carriage of *S. aureus* plays a vital role in the epidemiology and pathogenesis of staphylococcal infection. Carriers have a 3-6-fold increased risk of developing *S. aureus* infection compared to non-carriers (Wertheim, Vos, Ott, et al., 2004). Individuals can be colonized persistently or intermittently (J. Kluytmans et al., 1997). Nasal carriage with *Staphylococcus aureus* is a common risk factor for invasive infections, indicating the necessity to monitor prevalent strains, particularly in the vulnerable pediatric population. According to Dey et al., children with nasal carriage of *S. aureus* form an essential part in community dispersal of *S. aureus* and methicillin-resistant *S. aureus* (MRSA) (Dey et al., 2013). Some studies from India have shown that the main risk factors for nasal carriage of *S. aureus* were connected to overcrowding, mostly among children going to school and preschool (Dey et al., 2013; Pathak et al., 2010).

Studies from West and Central Africa show carriage rates ranging from 21%, 29%, and 36% in Ghana, Gabon, and Senegal, respectively (Ateba Ngoa et al., 2012; Egyir et al., 2014; Fall et al., 2014). Additionally, *S. aureus* infection is an urgent medical problem due to its growing frequency and poor associated outcome due to the paucity of high-quality data on the virulence gene. Also, the resistance gene, infection prevalence, and antibiotic susceptibility pattern of *S. aureus* present a medical problem in Ghana. *S. aureus* infection is one of the most common infections reported in Ghanaian health centers (Donkor, 2017).

The available studies on *S. aureus* in Ghana have concentrated on the adult population and patients (Egyir et al., 2014); however, there is a paucity of data on *S. aureus* carriage in children. Furthermore, most of these *S. aureus* studies that have been conducted in Ghana have been limited to Ashanti, Greater Accra, and the Northern regions of Ghana (Egyir et al., 2014; Eibach et al., 2017; Walana et al., 2020). Moreover, the determination of virulence genes and antibiotic resistance genes in *S. aureus* studies in Ghana are limited. Therefore, this study aimed to provide valuable data on *S. aureus*, their antibiotic resistance profile, and the prevalent virulence and resistance genes in children living in Cape Coast.

Aims

The research aims to determine the carriage rate, antibiotic resistance profiles, and antibiotic resistance of S. aureus isolated from the nasopharynx of children living in Cape Coast, Ghana.

Objectives

The study intends to:

1. Determine the prevalence of nasopharyngeal carriage of *S. aureus*
2. Investigate antibiotic susceptibility patterns
3. Determine antibiotic resistance genes among the *S. aureus* isolates

Justification

The increasing prevalence of Staphylococcal infections coupled with the emergence of antibiotic resistance staphylococcal strains necessitates the development of alternative treatment options. To develop alternative treatment options, it is essential to understand the genetic basis of *S. aureus* resistance among strains collected in Ghana. In addition, it is necessary to identify the common virulence genes present among the *S. aureus* strains, and this is because recent studies have identified a relationship between antibiotic resistance and virulence genes (Cepas & Soto, 2020; Pan et al., 2020). Therefore, this study will provide primary data on *S. aureus* carriage among children in Cape Coast. It will further describe the different antibiotic resistance profiles in addition to the genetic basis of the identified antibiotic resistance. Data on the antibiotic resistance profiles can guide the use of empirical treatment for *S. aureus* infections. Comprehensive data will be generated from the virulence genes analysis from carriage strains and can set the foundation for detailed genomic studies on *S. aureus* in the future.

Delimitations

This cross-sectional research focuses on the Cape Coast South and North Constituencies in the Cape Coast Metropolis. Only two of the four circuits (the Aboom and Ola circuits) were covered. Akotokyir, Kwaprow, Ewim, Bakaano,

Amamoma, and the University of Cape Coast neighborhood were chosen as immunization sites.

The study included characteristics such as gender, religion, age, and facility type, but variables such as signs of respiratory disorders and parents' academic qualifications were omitted.

Limitations

This study collected samples and data at selected schools and immunization centers to cover children aged 72 months to as low as 2 months. However, the sample collection covered only some parts of the Cape Coast Metropolis, so the data obtained might not be an exact representation of the entire population. The study is a cross-sectional study and not a longitudinal study. Because of that, the study cannot determine the precise dynamics of carriage and acquisition rates or estimate the carriage duration of *staphylococcus aureus*. Moreover, the samples were archived in a -80 freezer for four years. The molecular analysis was performed using conventional PCR to identify 20 genes out of the 21 targeted genes. The most significant shortcoming in the current study was the need for more information from the investigations that further constrained analysis and regression.

Definition of Terms

Staphylococcal infections: This is an infection brought on by bacteria from the Staphylococcus genus.

Antibiotics: These are the medication that intercepts the development of microorganisms or eliminates them.

Antibiotic resistance: When bacteria adapt to the usage of antibiotics, antibiotic resistance develops.

Antimicrobial: A natural or synthetic medication that extinguishes or restricts the development of microorganisms, including bacteria, fungi, and viruses.

Antimicrobial Resistance (AMR): This refers to the ability of microorganisms, such as bacteria, viruses, fungi, and parasites, to survive exposure to antimicrobial drugs, such as antibiotics, antivirals, and antifungals.

Multiple drug resistance (MDR): It is the resistance of a microbe to three or more different classes of antibiotics.

Nasopharyngeal carriage: It is when a microorganism establishes a colony in a person's nasopharynx.

Colonisation: It is a microorganism's existence on or inside a host, along with the organism's development and proliferation,

but without any clinical manifestations or an immune reaction.

Decolonization: This is a medical strategy that strives to clear a patient of a pathogen resistant to antibiotics.

Organisation of the Study

Except for the abstract and references, this thesis is divided into six chapters. With an emphasis on terms like *Staphylococcus aureus*, Staphylococcal infections, virulence factors, antibiotics, and antibiotic resistance, chapter one provides background information on the study. Along with the specific topics the research aims to address, the research's primary goal is also indicated. This chapter examines the topics that the research attempts to solve in more detail, including research

knowledge gaps, inconsistencies, and the utilization that will be made of the study's results.

Similar investigations carried out all around the world were examined in chapter two and described using a conceptual framework. The need to fill the gaps left by the examined literature and justify any methodology adoption or modification was emphasized. This chapter also includes global information on the prevalence of nasopharyngeal carriage, pathogenesis, immunology, and the occurrence of resistant *S. aureus* isolates. relationship between infection and immune response

The third chapter examines the resources and procedures utilized to carry out the study. The target population, area, and research design are all provided, along with some details. The chapter also goes into great depth on how the samples and data were gathered and processed, analyzed, and evaluated once they were acquired.

Chapter four depicted the processed and analyzed data in tables and graphs. Chapter five entailed the discussions. The obtained results were discussed based on the arrangement of the specific objectives.

Chapter six wraps up the entire thesis by summarizing the preceding chapters and then providing probable reasons for any disparities that may have occurred with the findings. Based on the significant and ambiguous results, recommendations were also offered.

Chapter Summary

In conclusion, this chapter provided an overview of the study's background, with particular attention paid to staphylococcal infection, the use of antibiotics, and antibiotic resistance. Along with the specific topics the study aims to address, the study's primary goal is also indicated. This chapter examines the topics that the study attempts to solve in more detail, including research knowledge gaps, inconsistencies, and the use that will be made of the study's results.

CHAPTER TWO

LITERATURE REVIEW

Introduction

In the 1880s, *S. aureus* was first illustrated by Sir Alexander Ogston, who observed a grape-like cluster of bacteria from slide preparations of pus from post-operative wounds and abscess patients (Ogston, 1882). Rosenbach, in 1884, was able to successfully sequester and cultivate the bacteria on a solid medium, which he named *Staphylococcus aureus* because of the distinguishing yellowish pigmentation of their colonies (Hardie & Whiley, 1995). Carotenoids called staphyloxanthin are responsible for the yellow pigmentation of the colonies. Evidence for its virulence was demonstrated in 1941 when Skinner and Keefer reported that the increasing antibiotic resistance indicates that its prevalence will continue to rise. Given the number and severity of *S. aureus* bacteremia in 122 patients at Boston City Hospital, the rate was 82% (Skinner & Keefer, 1941) .

Gram-positive pathogens' cell wall envelopes serve as a scaffold for the attachment of virulence factors and a sieve that prevents molecule diffusion. During infection, the envelope of Gram-positive bacteria is the site of interaction between microbes and their host environment (Hazmanian et al., 2003). The cell wall of *S. aureus* exhibits the characteristics of gram-positive bacterial cell walls, in which it maintains cellular integrity. It appears as a relatively thick (about 20 to 40 nm) homogeneous structure under the electron microscope (Giesbrecht et al., 1998). Peptidoglycan makes up 50% of the staphylococcal cell wall. Peptidoglycan is made up of alternating polysaccharide subunits of N -acetylglucosamine and N -acetylmuramic acid with 1,4-b linkages, the average chain length is between 10 and 20 disaccharides. Tetrapeptide chains bound to N-acetylmuramic acid and a pentaglycine bridge specific for *S. aureus* cross-link the peptidoglycan chains. N-acetylmuramic acid is attached to tetrapeptides containing l-alanine, d-glutamine, l-

lysine, and d-alanine. A pentaglycine group connects approximately 90% of these stem peptides to the stem peptides of another glycan chain (Labischinski, 1992). This pentaglycine is a staphylococcal peptidoglycan feature that connects the ε-amino group of one stem peptide to the d-alanine of the other. The non-cross-linked stem peptides contain an extra d-alanine cleaved during the cross-linking reaction (Giesbrecht et al., 1998). Transpeptidases, which are penicillin-binding proteins, catalyze the process of cell wall cross-linking (PBPs). Peptidoglycan may have endotoxin-like activity, stimulating macrophage cytokine release, complement activation, and platelet aggregation. Variations in staphylococcal strains' peptidoglycan structure may contribute to differences in their ability to cause disseminated intravascular coagulation (Kessler et al., 1991).

The teichoic acid attached to the cell membrane of the nasal epithelium is one of the most significant *S. aureus* colonization factors (Weidenmaier et al., 2004). In a study conducted by Weidenmaier et al., surface-exposed staphylococcal polymer called wall teichoic acid (WTA) is crucial for nasal colonization and mediates contact with human nasal epithelial cells. WTA-deficient mutants had trouble adhering to nasal epithelial cells and had no possibility of colonizing cotton rat nares at all. The study described the initial essential component necessary for *S. aureus* nasal colonization (Weidenmaier et al., 2004). Interestingly, WTA structures across Gram-positive bacteria are extraordinarily variable and frequently strain- or species-specific (Neuhaus & Baddiley, 2003). In most *S. aureus* bacteria the ribitol teichoic acids are covalently bound to peptidoglycan and are essential components of the cell wall. Teichoic acid, a polymer covalently linked to muramic acid via phosphodiester bonds, accounts for approximately 50% of the total mass of the cell wall. Teichoic acids are made up of lengthy chains of ribitol phosphate units (Lowy, 1998). Other literature also highlights that WTA polymer is made up of repeating units of ribitol-phosphate (RboP) with 11– 40 substitutions for d-alanine (d-ala) and N-acetylglucosamine (GlcNAc). In peptidoglycan, the 6-OH group of N-acetylmuramic acid residues is

covalently bonded to the WTA polymer by a disaccharide made up of GlcNAc-1-P and N-actelymannosamine, followed by two units of glycerol-phosphate (GroP) (Weidenmaier & Peschel, 2008; Winstel et al., 2013). D-ala residues on WTA and lipoteichoic acids (LTA) lead to resistance to glycopeptide antibiotics like vancomycin or teicoplanin and cationic antimicrobial peptides like defensins or cathelicidins (Collins et al., 2002; Peschel et al., 2000). Additionally, the elision of tagO, which encodes the first enzyme of the WTA biosynthesis pathway, showed that WTA is not necessary for *S. aureus* viability in a lab setting but affects several crucial cellular functions, such as autolysis, cell division, localization of penicillin-binding protein 4 (PBP4), and survival at high temperatures (Atilano et al., 2010; S. Brown et al., 2012; Vergara-Irigaray et al., 2008; Weidenmaier et al., 2004). Surprisingly, *S. aureus* WTA is crucial for severe invasive infections like endocarditis and helps build biofilms by adhering to epithelial and endothelial cells (Vergara-Irigaray et al., 2008; Weidenmaier et al., 2004). The human complement system is activated by *S. aureus* WTA via both the classical and mannose-binding lectin pathways, which results in opsonophagocytosis (Jung et al., 2012; Kurokawa et al., 2013). *S. aureus* and most other Gram-positive bacteria produce membrane-bound lipoteichoic acids (LTA) in addition to WTA. Lipoteichoic acid is a polymer of glycerol phosphate linked to a glycolipid terminus anchored in the cytoplasmic membrane (Lowy, 1998).

According to Neuhaus & Baddiley, wall teichoic acid (WTA) and lipoteichoic acid (LTA) make up teichoic acids (TAs) (Neuhaus & Baddiley, 2003). WTA and LTA, along with peptidoglycan, comprise a polyanionic network or matrix that performs tasks related to the envelope's elasticity, porosity, tensile strength, and electrostatic steering (Buckland & Wilton, 2000). In addition to its role in cation homeostasis, the polyanionic matrix also plays essential roles in the transport of ions, nutrients, proteins, and antibiotics, the regulation of autolysins, and the presentation of envelope proteins. It is possible to alter the

net anionic charge, ascertain the cationic binding capacity, and see the cations in the wall by esterifying TAs with D-alanyl esters (Neuhaus & Baddiley, 2003).

O-acetylation of muramic acid is an essential feature of staphylococcal peptidoglycan. As a result, staphylococcal cell walls are rarely degraded by lysozyme, which acts sterically (Giesbrecht et al., 1998).

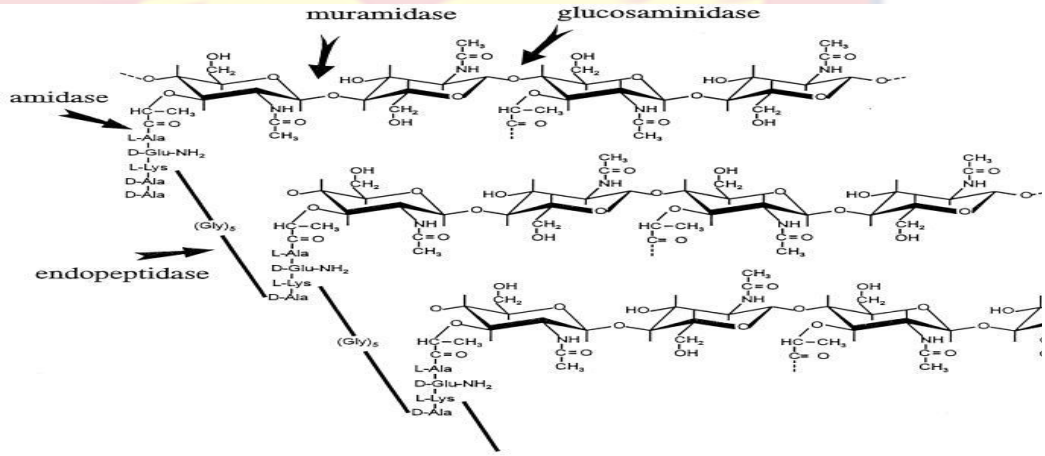


Figure 1: The structure of peptidoglycan and the sites where peptidoglycan may be attacked by cell wall hydrolases.

Extracellular capsular polysaccharides are commonly produced by microorganisms that cause invasive diseases. Capsules boost microbial virulence by making bacteria resistant to phagocytosis. Gilbert described capsule production by *S. aureus* in 1931 (O'Riordan & Lee, 2004). The majority of staphylococci produce microcapsules. Types 5 and 8 account for 75 percent of human infections identified among the 11 microcapsular polysaccharide serotypes (O'Riordan & Lee, 2004). The majority of methicillin-resistant *S. aureus* isolates are type 5.

The chemical composition of four antiphagocytic polysaccharides has been determined, including types 5 and 8, and all four are chemically related (Lowy, 1998). In study a conducted by Melly et al., only a few strains of *S. aureus* were found to be capsule positive. These highly encapsulated strains produced mucoid colonies that were resistant to

phagocytosis and were virulent to mice (Melly et al., 1974; Wiley & Maverakis, 1974). Karakawa and Vann proposed a new capsular polysaccharide typing scheme for *S. aureus* in 1982, using absorbed rabbit antiserum to prototype *S. aureus* strains. These researchers were the first to report that most *S. aureus* strains were encapsulated, and they identified eight capsular serotypes. Serotypes 1 and 2 were assigned to the heavily encapsulated strains M and Smith diffuse, respectively. Strains of these two serotypes form mucoid colonies on solid medium and are uncommon among clinical isolates (Arbeit et al., 1984). On a solid medium, isolates from the remaining serotypes form nonmucoid colonies with colony morphology indistinguishable from strains lacking a capsule. Some researchers have referred to nonmucoid. Encapsulated *S. aureus* isolates as microencapsulated to distinguish them from atypical mucoid strains (O'Riordan & Lee, 2004).

Many staphylococcal surface proteins share structural characteristics. At the carboxyl-terminal, these features include a secretory signal sequence, positively charged amino acids that extend into the cytoplasm, a hydrophobic membrane-spanning domain, and a cell-wall–anchoring region. Some of these proteins can function as adhesins because of a ligand-binding domain at the N terminal exposed on the bacterial cell's surface. Protein A, the prototype of these proteins, possesses antiphagocytic properties due to its ability to bind the Fc portion of an immunoglobulin (Foster & McDevitt, 1994; Lowy, 1998). Several related proteins bind extracellular matrix molecules and have been designated as microbial-surface components recognizing adhesive matrix molecules (MSCRAMM). Recent research suggests that these proteins are essential to staphylococci's ability to colonize host tissue (Patti et al., 1994).

Ribosomes, the universal cellular multicomponent RNA/protein particles that convert the genetic code into proteins in all living cells, can be paralyzed by a significant range of antibiotics (Eyal et al., 2015). The structures of ribosomes from pathogens such as *S. aureus* resemble those of other eubacteria due to their similar ribosomal RNA (rRNA)

and ribosomal protein sequences (Eyal et al., 2015). The phases of initiation, elongation, termination and recycling make up the ribosome cycle in bacteria. Ribosomes can proceed in one of two ways after the recycling stage, either to the subsequent initiation stage or to the production of 100S (Yoshida et al., 2009).

A study conducted by Ueta et al., shows that even though *S. aureus* lacks ribosomal modulation factor (RMF), it can still generate 100S ribosomes since it only contains *S. aureus* hibernation promoting factor (SaHPF). All growth phases of *S. aureus* are characterized by the presence of 100S ribosomes and SaHPF, whose levels rise and fall during development in ways distinct from those observed in E. coli. (Ueta et al., 2010). During an infection, *S. aureus* uses heme as a source of iron because it needs it for growth. Hemoglobin is taken up by staphylococcal surface proteins, which then liberate the heme from it and move the complex across the bacterial cell wall envelope and plasma membrane into the cytoplasm (Skaar et al., 2004).

The absence of iron is a significant barrier to colonization. Iron is needed by the majority of live microorganisms in the 0.4–4.0 μM range (Bullen, 1981). However, in mammals, the quantity of free ionic iron is kept constant at around 10^{-9} M (Chipperfield & Ratledge, 2000). The low solubility of iron at physiological pH, the intracellular location of iron (99.9% of total body iron is found within mammalian cells), and the sequestration of this ion within the iron-binding glycoproteins transferrin and lactoferrin or heme-containing proteins like hemoglobin are the leading causes of iron sequestration in host tissues (Robert I. Handin et al., 2003). To use hemoglobin as an iron source, bacteria must bind the hemoglobin polypeptide, remove and transport the heme molecule, and open the heme porphyrin ring to remove the single iron atom. All heme-degrading enzymes that have been identified are monooxygenases known as heme oxygenases (Skaar et al., 2004). Heme oxygenases are discovered throughout nature and are accountable for the oxidative breakdown of heme to biliverdin, CO, and free iron

(Maines, 1997). Heme degradation is required in bacteria in order to access the iron atom for use as a nutrient source. Bacterial pathogens from over fifteen genera can use heme as their sole iron source; however, a comparatively small number of bacterial heme-degrading enzymes have been identified (Ratliff et al., 2001; Zhu et al., 2000). Because there are few identifiable heme-degrading enzymes in bacteria that use heme as an iron source, it is possible to speculate on how these bacteria access the iron atom of the heme porphyrin ring. In a study conducted in 2003 by Hazmanian et al., heme-uptake procedure in the pathogenic bacterium *Staphylococcus aureus* and iron-regulated surface determinants (isd)(Hazmanian et al., 2003). The isd appears to be an import apparatus, relaying heme-iron across the bacterial envelope via cell wall-anchored proteins. This cluster contains three transcriptional units, isdA, isdB, and isdCDEFsrtBisdG, which specify three hemin-binding cell-wall anchored proteins: IsdA, IsdB, and IsdC. Furthermore, IsdB binds hemoglobin-like receptor-ligand interactions. IsdD (a membrane protein), IsdE (a lipoprotein ATPase), and IsdF (a polytopic transmembrane protein) all have homology to Gram-positive and Gram-negative heme-iron transporters, whereas SrtB is a sortase that anchors IsdC to the cell wall (Mazmanian et al., 2002). *S. aureus*, involving a large number of gene products.

S. aureus initially produces a variety of toxins, including hemolysins capable of lysing red blood cells. This causes hemoglobin to be disengaged from erythrocytes for use as a potential iron source. When Fur-mediated repression is released in humans, a lack of available iron causes transcription of the isd genes. Sortase A then sorts IsdB and IsdA to the cell wall, while SrtB sorts IsdC to a different portion of the cell wall. Free hemoglobin binds to IsdB, causing the heme molecule to be removed in an IsdB and IsdA-dependent manner (Skaar et al., 2004). The heme molecule is subsequently transferred to the IsdC cell wall transport protein and moved through the IsdDEF-based membrane transport pathway. IsdG and IsdI can carry out oxidative destruction of the

heme molecule upon entry to the cytoplasm, releasing free iron for usage as an iron source. Additional heme consumption proteins are still to be discovered in the *S. aureus* genome because the inactivation of the Isd heme transport system's components does not effectively prevent the development of heme as a sole iron source (Skaar et al., 2004). This discovery highlights the redundant nature of *S. aureus*'s iron uptake and demonstrates the organism's capacity to obtain iron from a variety of iron sources.

According to Sciara et al., *S. aureus* proteins with sequence signatures that induct them in a family of monooxygenases involved in the oxidation of aromatic intermediates, a process consistent with heme degradation (Sciara et al., 2003). Purified IsdG and IsdI can both bind heme in a 1:1 ratio, displaying binding characteristics similar to known heme oxygenases. IsdG was found to be cytoplasmically localized, implying that heme degradation occurs in the cytoplasm of *S. aureus* (Skaar et al., 2004).

Biofilms are created by a collection of bacteria, a complex assembly of proteins, polysaccharides, DNAs, and an extracellular polymeric matrix on various living and nonliving surfaces (Dincer et al., 2020). The ability to create biofilms is one of *S. aureus*' defense mechanisms. Standard antibiotic regimens frequently fail to eliminate bacteria buried in biofilms because they are naturally immune-resistant (R. Patel, 2005). Transient adhesion to a surface is the first step in the multi-phase process that leads to biofilm development. Microbial surface components recognizing adhesive matrix molecules (MSCRAMMS), particularly bacterial adhesins, subsequently facilitate the actual adhesion (Foster & Höök, 1998). Bacteria in a biofilm are much more resistant to antimicrobial agents than bacteria in planktonic forms because bacteria that are resistant to antimicrobial agents can become resistant after forming a biofilm. Bacterial biofilm growth provides several benefits, including protection against hostile environmental conditions such as osmotic stress, metal toxicity, and antibiotic exposure. The bacterium's multiphased defense consists of low antibiotic penetration into the biofilm, slow reproduction, and the

presence of an adaptive stress response (Dincer et al., 2020). The ability of bacteria to form biofilms appears to be an essential virulence factor in ensuring colonization on living tissues or medical devices, making treatment challenging. A biofilm is a sessile community made up of cells derived from microbes. These cells are irreversibly attached to a surface or interface or each other and are inserted in a matrix of extracellular polymeric substances (EPSs) that they have produced, resulting in a phenotype that is altered in terms of growth rate and gene transcription (Flemming et al., 2016). Proteins, cellulose, alginates, extracellular teichoic acid, poly-N-acetyl, and other organic compounds make up EPSs, which play an essential role in the formation of glucosamine, lipids, nucleic acids, phospholipids, polysaccharides, and extracellular DNA (eDNA), as well as physical interactions (Flemming et al., 2016; Jolivet-Gougeon & Bonnaure-Mallet, 2014). The stages of biofilm development include the initial attachment of the planktonic cell to the surface, cell differentiation, EPS secretion, maturation, and biofilm dispersion. It can be divided into three stages: irreversible adhesion to the surface, bacterial division and extracellular matrix production, and finally, matrix disassembly and bacterial dispersion (Mangwani et al., 2016).

Cells in mature biofilms release substances that might cause a transition from a biofilm to a planktonic mode of life as part of the natural biofilm growth cycle. The process of cell dispersal from biofilms depends on this change (McDougald et al., 2011; Penesyan et al., 2015). In *S. aureus*, the quorum-sensing system Agr regulates biofilm detachment. According to specific theories, this mechanism serves as a crucial regulatory switch between planktonic and biofilm regimes, facilitating the spread and colonization of *S. aureus* in new environments (Boles & Horswill, 2008; Yarwood et al., 2004).

Genetics of *S. aureus*

S. aureus genome comprises a single circular chromosome of approximately 2800 bp and a variety of extrachromosomal accessory genetic elements, including conjugative

and nonconjugative plasmids, mobile elements (IS, Tn, Hi), prophages, transposons and other variable elements (Lowy, 1998; Młynarczyk et al., 1998a). Bacterial genomes are typically made up of stable regions known as the core genome and variable regions known as the flexible gene pool.

Bacteriophages, plasmids, transposons, and unstable large regions known as genomic islands make up the flexible part. Pathogenicity islands are genomic islands that encode pathogenic bacteria virulence factors (Hacker et al., 2003). The chromosome contains genes that control virulence, antibiotic resistance, and extrachromosomal elements. The core genes encode the essential metabolic functions of *S. aureus*. Aside from the core genes, numerous mobile genetic elements (MGE) acquired through horizontal gene transfer may be advantageous under certain environmental conditions. These MGE, including bacteriophages, transposons, plasmids, and pathogenicity islands, account for approximately 15% of the *Staphylococcus aureus* genome (Alibayov et al., 2014). Mobile genetic elements are DNA fragments that can encode one or more virulence and resistance determinants and enzymes that facilitate their transfer and integration into other DNA (Frost et al., 2005). MGE is referred to as "mobilomes" when they can move both within and between cells. MGE can insert various sizes of DNA sequences such as phages, transposons, pathogenicity islands, plasmids, and chromosome cassettes via vertical gene transfer (Alibayov et al., 2014). Most MGE is acquired through horizontal genomic islands (GEI), which are defined as discrete DNA segments shared by closely related strains.

The GEI contributes to the spread of microorganisms by significantly impacting their genome plasticity and evolution. The GEI is also involved in spreading antibiotic resistance and virulence genes (Alibayov et al., 2014).

The core genome is the collection of all genes responsible for cell survival (genes encoding metabolism molecules, nucleic acids synthesis, and replication). The accessory is a collection of genes that exhibit the diversity observed within bacterial species and

encode proteins required for bacteria's adaptation to various environmental conditions (mainly through the acquisition of resistance and the production of virulence factors) (Alibayov et al., 2014; Lindsay & Holden, 2004).

Exogenous genetic information from other cells or the surrounding environment can be obtained by *S. aureus* in three ways: (1) transformation (uptake of free DNA from the environment), (2) transduction, and (3) conjugation (direct contact between bacterial cells) (Alibayov et al., 2014).

S. aureus extra-chromosomal DNA, such as MGE, can play an essential role in genome plasticity, facilitating the bacterium's adaptation to harsh environmental conditions. The epidemiological reports of resistance and virulence genes in natural *S. aureus* strains, on the other hand, are not reflected. Horizontal gene transfer (HGT) processes can spread virulence and resistance genes from plasmids through *S. aureus* populations (McCarthy & Lindsay, 2012).

The majority of naturally occurring *S. aureus* strains contain one or more plasmids ranging from 1 to 60 kbp. *S. aureus* plasmids have been classified into three categories. Class I plasmids are small (1.3–4.6 kbp), multicopy (10–55 copies per cell) plasmids with cryptic or caring a single (rarely two) resistance determinant (pT181, pC194, pSN2, and pE194). Class II plasmids are larger (15–46 kbp) and have fewer copies (4–6 per cell), but they contain the majority of penicillinase and amino glycol side/ trimethoprim resistance plasmids (e.g., pSK 1 and pIP630). Class III plasmids have a determinant of transfer (tra) by conjugation and, in most cases, a combination of resistance markers; this group also contains glycoside-resistance plasmids (pGO1, pG0400, and pCRG1600). These plasmids typically include one or two transposons and multiple copies of insertion sequences(Alibayov et al., 2014; Młynarczyk et al., 1998a; Sharma et al., 1994; Weigel et al., 2003). Phages, plasmids, pathogenicity islands, and the staphylococcus cassette chromosome encode virulence factors. A transposon (Tn 1546) inserted into a conjugated

plasmid-encoded resistance to other things, including disinfectants, encoded increased antibiotic resistance (Clark et al., 2005).

The regulatory genetic locus staphylococcal accessory regulator (sarA), which regulates the intracellular adhesin (ica) operon and agr regulated pathways, is crucial for *S. aureus* biofilm production. According to some research, the development of biofilms in methicillin-resistant *S. aureus* (MRSA) is primarily controlled by surface adhesins that are suppressed under agr expression. In contrast, the development of biofilms in methicillin-susceptible *S. aureus* (MSSA) is more dependent on cell-to-cell adhesion through the production of polysaccharide intercellular adhesin (PIA), also known as poly-N-acetylglucosamine (PNAG) or slime. However, the *Staphylococcus aureus*' ica locus does not appear to have the same role as Staphylococcus epidermidis.(O'Gara, 2007; O'Neill et al., 2007). Extracellular DNA (eDNA) is a significant and expected component of the bacterial biofilm matrix. Endogenous eDNA can be obtained from the outer membrane or cell integrity-degraded biofilm microorganisms without quorum sensing-mediated release. DNA can boost biofilm resistance to antimicrobial agents (Hall & Mah, 2017). In addition to playing a physical role in antibiotic resistance, eDNA has provided horizontal transfer of antibiotic resistance genes between microorganism cells, forming a biofilm (Hall & Mah, 2017). Because DNA is an anionic molecule, it can chelate cations such as magnesium ions and cause a decrease in Mg2+ concentration in the membrane, which is one of the mechanisms by which it increases biofilm resistance (Dincer et al., 2020).

General Epidemiology of *S. aureus*

Infections frequently begin locally, such as in the dermis of the nose, but if the local immune system fails to recognize the infection and defend against it, germs can spread and result in systemic infections. Such infections are linked to high total disease burden and mortality, even with prompt treatment and clinical management. *Staphylococcus aureus* has been identified as an important pathogen in terms of

epidemiology. Despite antibiotic therapy, staphylococcal infections are common in hospitalized patients and can be fatal (M. Askarian et al., 2009). As a result, staphylococcal infection deterrence has become more critical. *S. aureus* carriage appears to be crucial in the epidemiology and pathogenesis of infection (J. Kluytmans et al., 1997). Several studies around the world have reported rates of nasal carriage of *S. aureus* strains ranging from 16.8% to 90% (Alghaithy et al., 2000; M. Askarian et al., 2009; Bolyard et al., 1998). In different studies, the prevalence of *S. aureus* nasal carriage among hospital personnel and patients in Asia, South America and Africa ranged from 28.2% to 60.5% (Rahbar et al., 2003; Shrestha et al., 2009; Tewodros & Gedebou, 1984). However, the prevalence of *S. aureus* varies by country, profession, and demographic group. There is little data on the prevalence, population structure, and molecular epidemiology of *S. aureus* carriage in healthy Africans (Schaumburg et al., 2014). Although the prevalence and correlates of *S. aureus* nasal carriage have been documented in hospitalized adult patients and health workers in certain African countries, carriage rates in the community, particularly among children more vulnerable to staphylococcal infections, are unknown (Bebell et al., 2017; Kateete et al., 2019). The limited epidemiological data suggest that *S. aureus* clone distribution is remarkably heterogeneous in Africa, possibly due to the enormous cultural and geographical diversity. Toxinogenic strains are also unequally disseminated across countries. In South Africa, for instance, the prevalence of *S. aureus* strains that produce Panton-Valentine Leukocidin (PVL) is 0.3%, while in Tunisia it is 100% (Abdulgader et al., 2015b; Schaumburg et al., 2014). The prevalence of MRSA in resource-limited settings is unknown, and risk factors for carriage and invasive infection may differ from those in high-resource areas. *Methicillin-resistant Staphylococcus aureus* (MRSA) infections in the United States are linked to recent hospitalization and colonization of the anterior nares, though community-associated MRSA infections are common (Klevens et al.,

2007; Safdar & Bradley, 2008). According to epidemiological studies, the nasal carriage is a risk factor for staphylococcal infections and is usually the source of the infection (Wertheim, Vos, Ott, et al., 2004b). *S. aureus*, as a pathogenic agent, causes a wide range of infections, ranging from folliculitis and furunculosis to potentially fatal conditions such as sepsis, deep abscesses, pneumonia, osteomyelitis, and infective endocarditis. *S. aureus* colonizes the skin and mucosae of humans as well as a variety of animal species (Lowy, 1998). Over the last two decades, there have been two distinct shifts in the epidemiology of *S. aureus* infections: first, an increase in health-care-associated infections, particularly in infective endocarditis and prosthetic device infections; and second, an epidemic of community-associated skin and soft tissue infections caused by strains with specific virulence factors and resistance to beta-lactam antibiotics(Tong et al., 2015). Individuals with *S. aureus* bacteremia can develop a broad range of complications that can be difficult to detect at first and increase morbidity. Mortality rates of 20 to 40% have been reported (Mylotte et al., 1987; Shurland et al., 2007). Although the prevalence of methicillin-resistant *S. aureus* (MRSA) is still very low in northern European countries such as the Netherlands, the number of MRSA infections is increasing globally (Wertheim, Vos, Boelens, et al., 2004).

Methicillin-resistant *S. aureus* bacteremia appears more lethal than methicillin-sensitive *S. aureus* bacteremia (van Hal et al., 2012). Several other studies have found that bedside consultation with an infectious diseases specialist is associated with more profitable outcomes, such as fewer mortality, fewer relapses, and lower readmission rates, compared to telephone consultation or no consultation. These findings highlight the importance of performing serial examinations to look for metastatic infection and clues to the source of bacteremia (Forsblom et al., 2013; Fowler et al., 1998; Rieg et al., 2009). The population incidence of *S. aureus* bacteremia in developed nations ranges from 10 to 30 per 100,000 person-years. Longitudinal data from Denmark shed light on

the influence of changes in access to health-care interventions on *S. aureus* bacteremia incidence. *S. aureus* bacteremia increased from 3 per 100,000 person-years to 20 per 100,000 person-years between 1957 and 1990(Frimodt-Møller et al., 1997). Hospital admissions and invasive medical interventions increased exponentially in Denmark during the same period. As a result, nosocomial acquisition played a significant role in the overall increase in the incidence of SAB. However, the overall *S. aureus* bacteremia incidence in Denmark has remained relatively stable since 1990, at 21.8 per 100,000 person-years (Mejer et al., 2012). While across-the-board *S. aureus* bacteremia rates may have leveled off in the last two decades, the contribution of methicillin-resistant *S. aureus* has fluctuated. In Quebec, Canada, for example, the incidence of MRSA bacteremia increased from 0 to 7.4 per 100,000 person-years from 1991 to 2005, despite stable rates of methicillin-susceptible *S. aureus* (MSSA) bacteremia (Allard et al., 2008). Increasing MRSA bacteremia incidence was observed in Minnesota from 1998 to 2005, Calgary, Canada, from 2000 to 2006, and Oxfordshire, United Kingdom, from 1997 to 2003 (El Atrouni et al., 2009; Laupland et al., 2008; Wyllie et al., 2005).

Not so much is known about the prevalence and burden of *S. aureus* bacteremia in the world's non-industrialized and newly industrialized regions. Although the overall incidence of community-acquired *S. aureus* bacteremia in northeast Thailand was 2.5 per 100,000 person-years from 2004 to 2010, this study only reported incidence rates for community-acquired *S. aureus* bacteremia. Incomplete case identification may have also contributed to the low reported incidence (Kanoksil et al., 2013). In Kilifi, Kenya, the incidence of *S. aureus* bacteremia was 27 per 100,000 person-years among children under age 5. In Manhica District, Mozambique, children under the age of 15 had a mortality rate of 48 per 100,000 person-years, while children under the age of 13 had a mortality rate of 26 per 100,000 person-years in Soweto, South Africa (Berkley et al., 2005; Groome et al., 2012; Sigaúque et al., 2009)

Individuals whom *S. aureus* strains have colonized are more likely to become infected with these strains. Most nosocomial infections are acquired through contact with health care workers who have been transiently colonized with staphylococci from their reservoir or through contact with an infected patient. Outbreaks may also result from exposure to a single long-term carrier or environmental sources, but these modes of transmission are less common (Lowy, 1998; Sanford et al., 1994).

There are several options for reducing the prevalence of nosocomial *S. aureus* infections and treating MRSA infections that are also resistant to other antimicrobials. Implementing more effective infection control procedures. The new Centers for Disease Control and Prevention guidelines for preventing nosocomial pathogen transmission may aid in the prevention of staphylococci transmission within the hospital. Although this practice is unlikely to reduce the reservoir, it may reduce the number of *S. aureus* infections patients acquire (Garner, 1996). Lessening nasal colonization, many wound infections are caused by autoinoculation; reducing nasal carriage with systemic or topical agents has been shown to reduce *S. aureus* infections (J. A. J. W. Kluytmans & Wertheim, 2005). *S. aureus* vaccine development, vaccination with various staphylococcal antigens or whole organisms has been used to prevent infections in animals (such as bovine mastitis) for some time, but it has had little success in preventing infections in humans. A conjugate vaccine containing *S. aureus* types 5 and 8 capsular polysaccharides and Pseudomonas exotoxin A is immunogenic in humans and provides some protection against animal challenges. Additional antigens, including toxoids, should be investigated (Fattom et al., 1993). *Development of new or improved antimicrobial agents.* Chemotherapy for *S. aureus* infections is becoming increasingly difficult as the prevalence of multidrug-resistant iso-lates rises. The pharmaceutical industry is responding by either broadening the spectrum of existing compounds or developing new ones. Modified tetracyclines (glycylcyclines), fluoroquinolones, glycopeptides, and B-lactam agents are among the former (cephems and

carbapenems). Oxazolidinones and a combination drug containing semisynthetic derivatives of streptogramin A (dalfopristin) and streptogramin B are among the latter (quinupristin). Efforts are also being made to find other unique compounds that could attack new or novel bacterial targets (Archer, 1998).

Structure and anatomy of the nares

In 30-70% of the population, *S. aureus* lives as a commensal of the human nose (Peacock et al., 2001). The surface of the anterior nares is shielded with stratified squamous epithelium that is continuous with the external skin (Hanssen et al., 2017). The epidermis is separated into four strata: stratum basale and stratum spinosum, stratum granulosum, and stratum corneum. The epidermis matures, dividing into several layers starting at the basal layer, a process known as epidermal differentiation. Overall, the many layers combine to make the nasal epithelium resilient to pathogens and robust enough to fend off environmental attacks (Fuchs & Raghavan, 2002; Proksch et al., 2008). The basal layer continually supplies new cells, while the epidermis is constantly being desquamated to maintain the maturation and regeneration of the skin (Candi et al., 2005). At some point, basal layer keratinocytes change and separate from the basement membrane, halt cell division, and start differentiating (Fuchs & Raghavan, 2002; Simpson et al., 2011). Keratinocytes highly express keratins 5, 14, and 15 in the basal layer (Fuchs & Green, 1980; Porter & Lane, 2003). Cells of the granular layer include lamella bodies (LBs). LBs contain lipids such as cholesterol, sphingomyelin, glucosylceramides, and phospholipids (Feingold, 2012; Porter & Lane, 2003). Lipids (LBs) discharge their contents into the intercellular space after fusing with the plasma membrane (Feingold, 2012). At the stratum corneum, the cytoplasmic membrane of cells is replaced by a cornified envelope. The CE includes K1 and K10 and the proteins filaggrin, involucrin, and loricrin (Steinert & Marekov, 1995). The lipids produced by the LBs' contents give these cells their particular features (Proksch et al., 2008; Simpson

et al., 2011). Collagen, elastin fibers and other materials like connective tissue are found in the dermis. The dermis serves as a home for immune cells such as dendritic cells, T helper cells, and macrophages (Nestle et al., 2009).

Sites of Infection and colonization

Staphylococcus aureus is an incredibly adaptable pathogen that can survive in harsh external environments, colonize mucous membranes and skin, and cause severe non-purulent toxin-mediated disease or severe invasive purulent infections in humans. According to Huttenhower et al, *S. aureus* is a persistent environmental threat to humans, and our bodies offer a variety of biological niches for it and other microorganisms to flourish. Not all *S. aureus* exposures, though, result in effective colonization (Huttenhower et al., 2012; Van Belkum et al., 2009). van Belkum et al stated that, only about 30% of all humans carry staphylococcal cells chronically but asymptomatically in their nasopharynx and other body regions, despite *Staphylococcus aureus* being a bacterial species of significant medical importance. This is mainly ignored by the host, demonstrating that the human ecology is naturally friendly to the bacteria or at least receptive to them and that through a process of co-evolution, this has resulted in a condition of acceptance or tolerance.(van Belkum et al., 2009). The process by which *S. aureus* colonizes its host involves intricate interactions between the bacteria and its host (Huttenhower et al., 2012; van Belkum et al., 2009). The adherence of bacteria to nasal epithelial surfaces, a process that depends on precise interactions between adhesins on the bacterial cell surface and their target ligands in the epithelium, is believed to determine how *S. aureus* and the host interact during nasal colonization (Peacock et al., 2001).

S. aureus nasal carriage seems to be caused by several different factors. Staphylococcal toxins and cell wall-associated proteins are examples of bacterial factors. Hospitalization and crowding are examples of environmental variables. Host

susceptibility factors, such as immune suppression or other severe underlying disorders, also play a significant role (Bogaert et al., 2004; Weidenmaier et al., 2004). The presence of the normal flora can also affect *S. aureus*' capacity to colonize the nares (Uehara et al., 2000). To be able to colonize an ecological niche, *S. aureus* must be able to adhere to the receptors there. It must also be able to survive and avoid being wiped out by the host's defense mechanisms or the local microbiota (Mulcahy & McLoughlin, 2016).

A critical and significant risk factor for developing *S. aureus* infection and hospital-acquired illnesses is *S. aureus* colonization. In 1932, Danbolt made the first connection between nasal carriage and skin infection caused by furunculosis (Sollid et al., 2014). The nose serves as the main ecological niche for Staph aureus colonization in humans. It has been documented that *S. aureus* can also colonize the skin, perineum, vagina, axillae, pharynx, gastrointestinal system, urinary tract, and throat in humans (Acton et al., 2009; J. A. J. W. Kluytmans & Wertheim, 2005). Exclusive *S. aureus* colonization of the throat, pharynx, and intestinal tract without nasal transport has been documented (Nakamura et al., 2010). Habits like picking one's nose could be a way for *S. aureus* to spread from the nose to other body parts. This suggests that the nasal carriage of *S. aureus* most likely acts as a repository for the organism's dissemination into the environment or colonization of other body parts (Mermel et al., 2011).

The moist squamous epithelium of the anterior nares of healthy adults in the general population is the main ecological niche of *S. aureus* in the human nose (Van Belkum et al., 2009). In vitro cell studies confirmed this, revealing increased adherence of *S. aureus* to desquamated epithelial cells isolated from the anterior nares (Mulcahy et al., 2012). *S. aureus*, on the other hand, colonizes other regions of the nose, from the mid-region nares to the deeper regions of the nose (Yan et al., 2013). Interestingly, Kaspar et al. found that the posterior portion of the nose was consistently colonized compared to the anterior nares in the sample population of their investigation (Kaspar et al., 2016). The middle

and back of the nose are lined with pseudostratified columnar ciliated epithelium, whereas the surface of the anterior nares is lined with an epithelium that resembles skin. It has been proposed that these various surface cellular components influence the nasal microbiota. The epithelial component does not impact the nasal microbial diversity, according to another study examining the human nasal microbiome. However, many of the study subjects had chronic nasal inflammation (Kaspar et al., 2016; Yan et al., 2013).

Nasal colonization of *S. aureus* may be complex due to various barriers in the nasal cavity. The nose acts as a filter for air entering the system and as an entrance into the olfactory and respiratory systems (Geurkink, 1983). The mucus produced in the nasal cavity traps small molecules, including germs. The nasal epithelium also sheds cells, clearing the nose of bacteria. In addition, the nasal environment includes antimicrobial substances such as lysozyme, lactoferrin, and secretory IgA (Cohen, 2006; Eng et al., 2008). *S. aureus* must be able to grow and get past the defenses present in the nose in order to colonize the human nasal cavity successfully. This is accomplished via various surface-attached, secreted proteins and other elements like wall teichoic acid (A. M. Edwards et al., 2012). In vitro and in vivo, *S. aureus* attaches to the mucus components, affecting how effectively the mucus is cleared from the nasal cavity (Shuter et al., 1996). Despite the increased concentration of α-defensins and β-defensins seen in the nasal secretions of *S. aureus* carriers, *S. aureus* is more resilient in carriers' nasal fluids than in non-carriers. Additionally, *S. aureus* carriers' nasal secretions contain hemoglobin, encouraging bacteria to colonize surfaces (Pynnonen et al., 2011; van Belkum et al., 2007). Additionally, there is mounting evidence that *S. aureus* can survive inside nasal tissue cells. Because *S. aureus* lives inside cells, it can cause recurrent infections like rhinosinusitis (Hanssen et al., 2017; Sinha & Fraunholz, 2010).

According to a recent study by Weidenmaier et al., a mutant that lacks cell wall-associated teichoic acid (WTA) makes it difficult for bacteria to colonize cotton rat nares.

WTA-coated beads stuck firmly to primary epithelial cell cultures, while pure WTA prevented colonization in an animal model (Weidenmaier et al., 2004). In vitro tests involving developing epithelial and endothelial cells should be interpreted with caution, nevertheless, because *S. aureus* appears to adhere to terminally differentiated squamous cells in vivo (Peacock et al., 2001).

The bacterial cell wall-associated proteins known as MSCRAMMs facilitate the adhesion of *S. aureus* cells to host proteins such as fibrinogen, fibronectin, collagen, elastin, von Willebrand factor, vitronectin, and bone sialoprotein (microbial surface components recognizing adhesive matrix molecules). Following sortase-mediated cleavage of the C-terminal LPXTG motif, *Staphylococcus aureus* can express up to 20 potential MSCRAMMs anchored to the cell wall peptidoglycan (Mazmanian et al., 1999). Only the fibrinogen-binding proteins ClfA (clumping factor A) and ClfB (clumping factor B), the fibronectin-binding proteins FnBPA and FnBPB (which also bind fibrinogen), the collagen-binding protein Cna, and protein A, which binds IgG and von Willebrand factor, have been thoroughly studied (Hartleib et al., 2000; Wann et al., 2000).

In vivo, squamous cells change shape, increasing keratinization and losing the nucleus as they move from the basal layer to the exposed surface. In vitro, *S. aureus* adheres firmly to the most mature cell type. Unknown variables contribute to bacterial adhesion on the surface of squamous epithelial cells. The heterodimeric protein keratin, which appears to be exposed on the surface of epithelial cells, is one potential contender (Sajjan et al., 2002). Keratins are essential structural proteins that belong to the intermediate filament protein superfamily. Based on isoelectric point and sequencing, they are classified into one of two categories. K1 and K8 are type II keratins, which are more prominent (53–67 kDa) and more fundamental than type I molecules. K10 and K18 are type I keratins, typically smaller (40–63 kDa) and more acidic. Type I and type II heterodimeric multimers of keratin make up keratin filaments (O'Brien et al., 2002). As a

result, type I keratin K10 pairs with type II keratin K1 or K1. A considerable keratin expression changes as epithelial cells develop and move towards the epidermis' surface. The more developed cells of the supra-basal layer contain significant levels of K1 and K10, whereas the cells in the basal layer express K5 and K14. K8 and K18 are not connected to stratified squamous epithelia, even though they are typical of simple epithelial cells (O'Brien et al., 2002). In a paper published by O'Brien et al., ClfB was expressed in the heterologous host Lactococcus lactis, and a mutant of *S. aureus* lacking ClfB expression was used to demonstrate that ClfB promoted adherence to squamous nasal epithelial cells, most likely through a specific interaction with the type I cytokeratin molecule, K10. They demonstrate that K10 is expressed on the surface of desquamated human nasal epithelial cells and that *S. aureus* transcribes the ClfB gene in the human nares in vivo. They also show that ClfB stimulates bacterial adhesion to immobilized purified epidermal keratin. They contend that ClfB is a crucial factor in *S. aureus* nasal colonization (O'Brien et al., 2002).

Clumping factor B (ClfB) and iron-regulated surface determinant A (IsdA) of *S. aureus* have been linked to nasal colonization. ClfB and IsdA were found to promote adhesion to squames in vitro when wild-type strains were compared to isogenic mutants lacking the proteins (Clarke et al., 2006). *S. aureus* surface protein G (SasG) and the serine-aspartate repeat proteins SdrC and SdrD also stimulate bacterial adhesion to squames in vitro, in addition to boosting colonization of rodent nares and in the case of ClfB. However, their effects, if any, in vivo have not yet been investigated (Corrigan et al., 2009). The cell wall-anchored (CWA) protein clumping factor B (ClfB) facilitates *Staphylococcus aureus* attachment to the anterior nares during colonization via high-affinity interactions with the cornified envelope. ClfB has been shown to promote nasal colonization in both rodents and humans via this interaction. ClfB is only expressed during the early exponential phase of cell growth and is absent during the late and stationary phases (Mulcahy et al., 2012). ClfB

binds to the most prevalent protein in the cornified envelope of squamous cells, loricrin, and plasma fibrinogen, cytokeratin 10, which makes up most of the inside of squamous cells. Loricrin is an essential ligand for ClfB in vivo, at least in mice, as evidenced by the much lower rates of *S. aureus* nasal colonization in loricrin-knockout mice compared to wild-type mice (Mulcahy et al., 2012).

In the basal layer, keratinocytes divide continually. In order to differentiate into squames when cells move toward the stratum corneum, proteins that will eventually form the cornified envelope (CE), which takes the place of the cytoplasmic membrane in these cells, are expressed. The CE is made up of covalently and non-covalently bonded ceramides, as well as cross-linked proteins such as loricrin, involucrin, and tiny proline-rich proteins (Candi et al., 2005). The CE is a highly robust structure, crucial to barrier function due to its extensive cross-linking and conformational characteristics. The CE's most prevalent protein, loricrin, makes for about 80% of the protein mass. Squames contain cytokeratins 1 and 10, which are visible on their surface (O'Brien et al., 2002; Roop, 1995). Loricrin is surprisingly non-essential despite the CE's critical role in barrier function and the fact that squames' CE contains much of it. A loricrin knock-out mouse has been created, and while these mice show a delay in the development of the skin barrier during embryonic development, the skin phenotype vanishes by days 4-5 after birth. Loricrin-deficient mice reproduce correctly and share identical phenotypes as their wild-type litter mates (Koch et al., 2000). These mice have a compensatory loricrin backup mechanism, which prevents them from developing a more severe phenotype.

In the CE of mice lacking Loricrin, there has been an increased expression of tiny proline-rich proteins. Interestingly, the expression of other CE parts, including involucrin, filaggrin, and Cytokeratin 10 (K10), was similar in mice with and without Loricrin deficiency (Jarnik et al., 2002).

The healthy adult population of *S. aureus* nasal carriers has been divided into two categories: persistent carriers and non-persistent carriers. The conventional *S. aureus* nasal carriers kinds of persistent carriers, intermittent carriers, and non-carriers were superseded by this categorization (Van Belkum et al., 2009; Williams, 1963). There have been disagreements about the appropriate definition and standards to apply when identifying a person as a persistent carrier of *S. aureus*. However, a global standard based on the "culture rule" has been adopted. Non-persistent carriers have one positive *S. aureus* culture, while persistent carriers have at least two positive cultures from nasal samples obtained one week apart (J. L. Nouwen et al., 2004). It has been found that persistent carriers have greater *S. aureus* loads and are more vulnerable to *S. aureus* infection. Persistent carriers can also act as a reservoir for the later spread of *S. aureus* to other people in the population. This could be due to *S. aureus* having a longer lifespan in persistent nasal carriers than in non-persistent carriers (J. Nouwen et al., 2004; Yan et al., 2013). While various *S. aureus* strains can colonize non-persistent carriers throughout their lifetime, persistent carriers are primarily colonized by a single strain over time. Additionally, there are differences in the antibody profile responses between persistent and non-persistent carriers (Van Belkum et al., 2009). In comparison to nonpersistent carriers, persistent carriers of *S. aureus* are said to have higher immunoglobulin G (IgG) titers and IgA against the pathogen (Verkaik et al., 2009). Additionally, it is acknowledged that chronic carriers benefit from having *S. aureus* around constantly (Kolata et al., 2011). This makes sense because the strains that infect the host are typically endogenous. It is interesting to note that persistent *S. aureus* nasal carriers reacquired their endogenous strain from the mixture after being artificially infected with a mix of *S. aureus* inoculum (J. L. Nouwen et al., 2005).

Pathogenesis of *S. aureus*

Staphylococcus aureus is a crucial diagnostic focus for the routine microbiology laboratory and one of the most important bacterial pathogens in clinical practice. It is carried as a harmless commensal in up to two-thirds of the population at any given time, primarily in the anterior nares. It colonizes skin breach sites like ulcers and wounds, causing superficial and deep skin and soft tissue infections and life-threatening deep infections, including endocarditis and osteomyelitis (Aryee & Edgeworth, 2016). The organism's vast virulence factors account for its success as a pathogen and its capacity to cause various illnesses. It is a model for researching the pathophysiology of infectious diseases and has more virulence mechanisms than practically any other human pathogen (Archer, 1998). Global regulatory systems, such as agr and sar, control which virulence factors are produced at different stages of growth and in response to the local environment (Archer, 1998). Controlling infections caused by a pathogen as virulent as *S. aureus* is critical. The organism is initially able to avoid opsonophagocytosis because of certain conditions. Invading through tissue from the site of infection's initial infection; causing the sepsis syndrome by encouraging a significant release of cytokines; exiting the bloodstream and entering underlying tissue by adhering to and invading endothelial cells, and specifically increasing this pathogen's susceptibility to antibacterial syndromes through the production of toxins (Archer, 1998).

A wide variety of structural, enzymatic, and poisonous substances produced by staphylococci are associated with adhesion, invasion, toxicity, and evading host defense mechanisms. The Staphylococcus genus' most common human pathogen, *S. aureus*, is the root of several human diseases. Based on the site and mechanism of infection, staphylococcal infections can be categorized into four groups: (1) local infections linked to skin and soft tissue infections (SSTIs), (2) systemic infections linked to bacteremia, sepsis, and pneumonia, (3) invasive device entry infections linked to patients on dialysis,

and intravascular catheters, and (4) toxin-associated diseases like toxic shock syndrome and staphylococcal scalded skin syndrome (SSSS) (Kurlenda & Grinholc, 2012; Tong et al., 2015).

Numerous substances produced by *S. aureus* are released into the extracellular environment. These secreted substances include enzymes, superantigens, and poisons that harm membranes (Otto, 2014). Toxic shock syndrome toxin (TSST), a superantigen factor, activates the host's T cells, causing them to proliferate excessively and produce cytokines, ultimately resulting in the deadly outcome such as Staphylococcal toxic shock syndrome. Toxins that damage membranes pierce the host cell's cytoplasmic membrane, causing cell lysis and the release of intracellular contents (Otto, 2014). Toxins that cause membrane damage include proteins like Hemolysin-α (α - toxin), Panton-Valentine leukocidin (PVL), phenol-soluble modulins (PSMs), and gamma-toxin (gamma-hemolysin, HlgA, HlgB, HlgC) (Grumann et al., 2014; Otto, 2014). These proteins' modes of action are diverse. For instance, PVL interacts with the C5aR and C5L2 receptors on neutrophils, but it is believed that PSMs have receptor-independent effects on host cells (Otto, 2014; Spaan, Henry, et al., 2013). Additionally, A disintegrin and metalloproteinase 10 bind to the α- toxin (ADAM10), and the interaction causes loss of epithelial integrity by impairing focal adhesion and degrading E-cadherins. Additionally, *S. aureus* secretes enzymes that further affect bacterial pathogenicity, including staphylokinase, staphylocoagulase, and von Willebrand factor (vWF) (Inoshima et al., 2011; Wilke & Wardenburg, 2010). Also implicated in the pathophysiology of staphylococcal scalded skin disease is *S. aureus* Exfoliative Toxin (ET) (SSSS) (Nishifuji et al., 2008). Toxins can cause a weak response in the colonized organism because they can degrade some host cells, manipulate innate and adaptive immune responses, and degrade inter-cellular junctions, all of which contribute to *S. aureus* proliferation (Grumann et al., 2014). Toxins, for example, are suspected of causing diseases like toxic shock syndrome (TSS), staphylococcal scalded skin syndrome

(SSSS), necrotizing pneumonia, and deep-seated infections. As a result, a strong relationship between virulence genes and specific disease symptoms has been established (Holtfreter & Bröker, 2005; Ladhani, 2003). This apparent linkage between certain diseases and toxins has prompted urgent research into understanding the pathogenesis, etiology, epidemiology, pathophysiology, and, more precisely, the highly controlled mechanism of toxin production of *S. aureus*. Effective preventive measures will increase the ability to control staphylococcal infections because this bacterium's virulence machinery is mainly responsible for its tenacity as a pathogen (A. H. Bartlett & Hulten, 2010; Otto, 2010).

Many of the *S. aureus* virulence factors that are released can be classified as toxins because they are frequently called harmful compounds. Toxins, however, differ from other virulence factors in that they are produced by the organism that produces them and directly impact the host (Otto, 2014). Research on the role that various toxins play in forming biofilms has shown that some of them are particularly important for developing biofilms. Scheer et al., for instance, showed how leukotoxin AB (LukAB) and hemolysin (Hla or -toxin) affect the persistence of biofilms (Scherr et al., 2015). Utilizing a murine orthopedic implant biofilm infection model, the significance of these toxins was evaluated, and the results showed a synergistic role for Hla and LukAB in encouraging macrophage dysfunction and inducing cell death. When *S. aureus* was arranged as a biofilm, the decrease in macrophage phagocytosis facilitated its capacity to evade adverse host reactions (Scherr et al., 2015). Another study illustrating the role of toxins in the development of biofilms was carried out by Dastgheyb, who discovered that phenol-soluble modulins (PSMs) prevent the formation of biofilms by interfering with the interactions of matrix molecules with bacterial cell surfaces. Agglomeration is started by the contact of *S. aureus* surface proteins with host matrix proteins like fibrin, and it develops to enormous dimensions because PSM expression is not present in the conditions of the study. (Dastgheyb et al., 2015). Similar findings were found in an in vitro

investigation by Periasamy on the role of *S. aureus* PSMs in biofilm development. PSMs destroyed biofilms and caused their dispersal due to the toxin's surfactant capabilities (Periasamy et al., 2012).

Ninety five percent (95%) of clinical *S. aureus* strains release Hla, a 33 kDa polypeptide. *Hla* is a water-soluble monomer that is released as a beta-barrel-forming toxin. This toxin is not harmful on its own; instead, the danger comes from its capacity to bind to other molecules and oligomerize into a heptameric form on the host cell membrane (Gouaux et al., 1994; Grumann et al., 2014). In order to attack the cell membrane, Hla attaches to its target cell, forms an oligomer to a pre-pore structure, and then extrudes a β-barrel through the lipid bilayer to create a hydrophilic transmembrane channel (Seilie & Bubeck Wardenburg, 2017). This toxin is the model for the class of tiny cytotoxins that produce β-barrel pores (PFTs). Over time, pore development and cellular lysis were thought to be the main effects of Hla activity. Recent investigation has highlighted the significance of changing the cell signaling pathways, which control cell growth, inflammation, cytokine release, and cell-cell communication. Epithelial, endothelial, T cells, monocyte, and macrophage are a few human cell types that this toxin has been shown to harm (Berube & Wardenburg, 2013; M. W. Parker & Feil, 2005). Depending on the exposed cell type and the relative concentration of toxin, Hla causes several signaling processes in the target cell. The small pore created when the toxin enters the cell permits the quick release of ATP and K^+ ions while preventing the passage of large molecules through the cell membrane (Berube & Wardenburg, 2013; Lizak & Yarovinsky, 2012). There is an inflow of extracellular calcium into the cell following the pore-formation stage. The hydrolysis of membrane phospholipids and the conversion of arachidonic acid to leukotrienes, prostanoids, and thromboxane A2 are stimulated by increased intracellular calcium. Additionally, the nuclear translocation of NF-B has induced the activation of protein kinase C (Rose et al., 2002; Suttorp et al., 1993). The occurrence of all these things

and the IL-1, IL-6, and IL-8 production together represents the pro-inflammatory stimuli. E-cadherin is a primary substrate for ADAM10 in epithelial cells, which are essential targets of Hla (Maretzky et al., 2008). Cellular Hla receptor ADAM10 is a zinc-dependent metalloprotease expressed as a type I transmembrane protein on the surface of the majority of host cells. After the Hla bond compromises the function of the epithelial tissue barrier, the enzyme that breaks down E-cadherin is triggered, allowing *S. aureus* penetration (D. R. Edwards et al., 2008; Grumann et al., 2014). A more thorough investigation of the toxin's effects on a particular cell population is now possible thanks to identifying ADAM10 as a cellular receptor for Hla. Two pieces of evidence support the idea that this toxin is crucial to the pathogenesis of *S. aureus*. The first relates to individuals with *S. aureus* illness and shows serum antibody responses to the toxin that is consistent with the production of the toxin (Adhikari et al., 2012; Fritz et al., 2013). The additional supporting information relates to bacterial genetic and protein profile analysis (Berube & Wardenburg, 2013). The capacity to express Hla was discovered by examining the Hla and agr loci of the *S. aureus* strains engaged in epidemic occurrences in the 1950s and 1960s. This ability was further supported by the presence of a highly virulent phenotype in these isolates in animal studies of Hla-mediated illness. Together, these data suggest that the pathophysiology of invasive illness in healthy individuals may depend on the expression of hla (Berube & Wardenburg, 2013; DeLeo et al., 2011).

Glenny and Stevens initially identified hemolysin in 1935, while Projan et al. discovered the hlb gene sequence in 1989 (Glenny & Stevens, 1935; Projan et al., 1989). This toxin has been proven in sheep to be enormously hemolytic to erythrocytes, but not in rabbits. Since the toxin is also known as sphingomyelinase, the difference in erythrocyte susceptibility may be caused by the varied sphingomyelin contents of these cells. The most prevalent sphingolipid in eukaryotic membranes, sphingomyelins, are hydrolyzed by phosphoric diester hydrolases known as sphingomyelinases (Dinges et al., 2000; Doery et

al., 1965; Flores-Díaz et al., 2016). It is unclear how this toxin contributes to sickness. Animal isolates are known to produce vast amounts of β-hemolysin, and this high expression may suggest that β-hemolysin producers have some selective advantage due to toxin production (Dinges et al., 2000). Large amounts of this toxin are produced by strains seen in chronic skin diseases and bovine mastitis(Aarestrup et al., 1999; Katayama et al., 2013). Inhibiting the expression of interleukin-8 (IL-8) by endothelial cells, β-hemolysin is cytotoxic to human keratinocytes, polymorphonuclear leukocytes, monocytes, and T lymphocytes. These may help *S. aureus* escape from phagosomes and cause the production of biofilms (Huseby et al., 2010; Katayama et al., 2013). The significance of this toxin for *S. aureus* pathogenicity has been authenticated by some researches. In models of pneumonia and mouse ear skin infections, the virulence of a mutant *S. aureus* strain lacking hlb was reduced (Katayama et al., 2013). Additionally, a mutant strain that expressed a β-hemolysin with reduced biofilm formation demonstrated lower pathogenicity in a rabbit endocarditis model (Huseby et al., 2010).

Panton and Valentine first identified a potent leukocidal toxin generated by various *Staphylococcus aureus* isolates at the beginning of the 1930s. This leukocidin, now known as Panton-Valentine leukocidin (PVL), is lymphocyte-insensitive but cytotoxic to neutrophils, monocytes, and macrophages to a lesser extent (Gauduchon et al., 2001; Meyer et al., 2009). Woodin first purified Panton-Valentine Leukocidin (PVL) from *S. aureus* culture supernatants. Two protein constituents of 32 kDa and 38 kDa, referred to as S for slow-eluted and F for fast-eluted, were separated from this purification phase based on their migration on a carboxymethyl cellulose column (WOODIN, 1960). The class S and class F proteins specific for PVL are yielded by all PVL-producing isolates (LukS-PV and LukF-PV), and all PVL genes (LukS-PV and LukF-PV) are encoded in various bacteriophages that express Sa2 integrase (Goerke et al., 2009).

The protein subunits LukS-PV and LukF-PV make up the bicomponent, pore-forming exotoxin PVL, which is encoded by prophages (Diep et al., 2006). Secondary binding of LukF-PV is triggered by the initial binding of LukS-PV, which is a crucial step to the surface of target cells.

This causes the assembly of lytic hetero-octamers that generate pores (Jayasinghe & Bayley, 2005). PVL improves host responses by priming sensitive cells at sublytic concentrations (Graves et al., 2012). According to Gauduchon et al., although the participation of a myeloid-specific receptor has been hypothesized, the molecular basis for PVL's selectivity for particular cell types and species remains unknown (Gauduchon et al., 2001). Methicillin-resistant *S. aureus* (MRSA) was discovered to infect previously healthy people in the 1990s, according to Vandenesch et al. Community-associated (CA) MRSA strains have since spread quickly throughout the world. Most CA-MRSA isolates include PVL-encoding prophages, partly because the PVL-carrying clone USA300 has successfully disseminated throughout the United States (DeLeo et al., 2010; Vandenesch et al., 2003). According to epidemiological research, notably in nations with low CA-MRSA prevalence, PVL is linked to severe necrotizing pneumonia and skin and soft tissue infections in otherwise healthy people (Hidron et al., 2009; Shallcross et al., 2013). Species specificity is a typical feature of various immune modulators released by staphylococci, according to Rooijakkers et al. Depending on the distinctive amino acid sequences of the host targets, this selectivity can frequently be reduced to high-affinity protein-protein interactions (Rooijakkers, Van Kessel, et al., 2005). The human C5a receptor (C5aR, CD88), a seven-transmembrane G protein-coupled receptor, is targeted by the chemotaxis inhibitory protein of *S. aureus* (CHIPS) (GPCR). A potent anaphylatoxin called C5a is released when the complement system is activated. One of the earliest innate recognition processes is the C5aR-mediated detection of invading bacteria by phagocytes (De Haas et al., 2004; Postma et al., 2005). CHIPS effectively prevents neutrophil activation and

recruitment in vitro by blocking the human C5aR (De Haas et al., 2004). Another seven-transmembrane receptor called C5L2 is liganded by C5a (GPR77). A large portion of the later receptor's function is yet unknown. C5L2, despite being closely related to C5aR, is not linked to a G protein and lacks direct intracellular signaling activity as a result (Monk et al., 2007).

In a study conducted by Spaan et al., LukS-PV binds the human C5a receptors C5aR and C5L2. PVL, a functional bicomponent toxin, induces pore formation in the target cell membrane via C5a receptors (Spaan, Henry, et al., 2013). The C5a receptor expression profiles on different cell populations explain 80-year-old observations that PVL targets neutrophils, monocytes, and macrophages but not lymphocytes because the latter are negative for expression of both C5aR and C5L2 (Monk et al., 2007; Woodruff et al., 2011). A more recent study estimated that the number of LukS-PV receptors matches the combined expression level of both C5a receptors on neutrophils. Although some nonmyeloid cells express both C5a receptors, their levels of expression are lower than in myeloid cells (Gauduchon et al., 2001; Haviland et al., 1995). The toxin may easily target the C5aR because of its widespread expression, especially on neutrophils, which enables it to discriminate between phagocytic cells and other cells. The protection of human neutrophils against PVL-induced pore formation by the selective C5aR inhibitor CHIPS in the experimental setting by Spaan et al. suggests that the C5aR is the predominant receptor for PVL on neutrophils, and the C5L2, the minor receptor. The fact that C5aR-silenced THP-1 macrophages are less susceptible to PVL cytotoxicity further supports the idea that the C5aR is the primary PVL receptor and suggests the presence of any additional major PVL receptors is extremely unlikely (Spaan, Henry, et al., 2013). TLR2 and CD14 were described as essential receptor complexes for PVL-induced lung inflammation in mice in a recent study. However, in a different study, TLR2, TLR4, and CD14-specific antibodies

did not prevent PVL from priming human neutrophils for improved function (Graves et al., 2012; Zivkovic et al., 2011).

Mehlin et al. identified phenol-soluble modulins (PSMs) in an S. epidermidis culture in 1999. In that investigation, three peptides—PSMα, PSMβ, and PSMγ—were discovered and described as a pro-inflammatory complex extracted using hot phenol from the culture filtrate (Mehlin et al., 1999; Wang et al., 2007). Due to its similarity to the δ-toxin produced by S. epidermidis, PSMγ is also known as a δ-toxin (McKevitt et al., 1990). PSMs in *S. aureus* were also discovered through research. PSMs are encoded in three different places in the genome of *S. aureus* (Wang et al., 2007). PSMα peptides (PSMα1-PSMα4) and PSMβ (PSMβ1, PSMβ2) are encoded in the psmα operon and psmβ operon respectively. The coding sequence for RNAIII contains the information for δ-toxin (Peschel & Otto, 2013). The α-helix amphipathic structure of PSM peptides spans almost the complete length of the peptide in shorter α-type PSMs and is found in the carboxy-terminal region in longer β-type PSMs. PSMs are thought to connect non-specifically to the cytoplasmic membrane, resulting in membrane disintegration. According to some theories, phospholipid content and membrane charge play a significant role in a cell's vulnerability to PSMs (Otto, 2014). The charge properties of PSMs vary. PSMα has positive and PSMβ negative charges, whereas δ-toxin has a neutral charge. The primary elements in pathogenesis seem to be PSMs' propensity to cluster in oligomers, generating short-lived pores, and their ability to assist in spreading on surfaces or creating biofilms (Peschel & Otto, 2013). According to research by Wang et al., PSMα peptides have a significant role in the ability of community-associated MRSA to induce bacteremia and cutaneous infections, while δ-toxin and PSMβ have little to no impact on mice models of *S. aureus* infection (Wang et al., 2007). PSM peptides in *S. aureus* show biofilm-structuring actions on a global scale, indicating that they impact the formation of biofilms through a routine set of physicochemical characteristics. Moreover, because PSMs have the power to lyse

human neutrophils and trigger inflammatory responses, it is thought that their synthesis is closely tied to the staphylococcal species' propensity to produce invasive infections (Periasamy et al., 2012; Rautenberg et al., 2011).

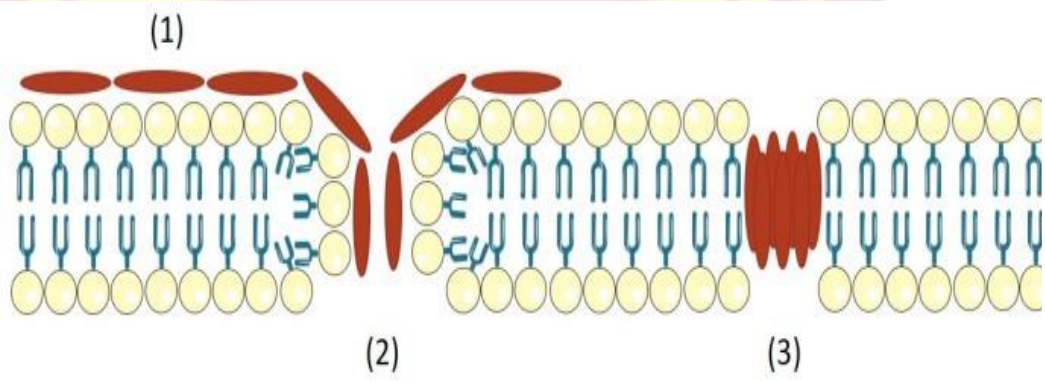


Figure 2: Model of pore-formation mechanism for *S. aureus* PSMs. PSMs adhere to the cytoplasmic membrane in an unspecific manner (1), which may cause membrane breakdown (2). PSMs have a propensity to assemble into oligomers and generate a transient pore (3).

The serine proteases known as exfoliative toxins (ETs), commonly referred to as epidermolytic toxins, are released by *S. aureus*. In the epidermal surface layers of the skin, these proteases detect and hydrolyze desmosome cadherins (Mariutti et al., 2017). ETs are exotoxins that can cause blistering and skin peeling by cleaving keratinocyte connections and cell-cell adhesion in the host's epidermis (Nishifuji et al., 2008). Von Rittershain documented the clinical characteristics of epidermal exfoliation in newborns in 1878. However, Lyell did not unearth the connection between exfoliation and *S. aureus* until 1967 (Lyell, 1967; Rittershain, 1878). Since the toxin is disseminated from far-off sites of infection through the bloodstream, Ritter's and Lyell's discoveries were delayed as a result of the fact that the blister fluid and exfoliated regions are frequently devoid of cultivable staphylococci (Bukowski et al., 2010). Lyell had earlier proposed a hypothetical toxin,

which Melish et al. identified in 1972 and showed had a detrimental effect on newborn mice, which were used as exploratory models (Melish et al., 1972). Exfoliative toxins A, B, C, and D are the most common ETs that are currently recognized (ETA, ETB, ETC, ETD). While ETC was only discovered from a horse infection and was not linked to any human disease, ETA and ETB are the most likely to cause human skin injury (C. Y. Lee et al., 1987; Mariutti et al., 2017). In a clinical sample of *S. aureus*, ETD was first discovered in 2002. About 5% of *S. aureus* strains produce these ETs, with ETA being more frequent in Europe, Africa, and America and ETB being more common in Japan (Ladhani, 2001). Certain *S. aureus* strains produce ETs responding to localized epidermal infections like bullous impetigo and widespread illnesses like SSSS (Mariutti et al., 2017). Skin exfoliation is the hallmark of the SSSS syndrome, but its early symptoms include fever, skin hypersensitivity, and erythema. These are followed by the development of superficial fluid-filled blisters and skin separation. Large portions of the body may be impacted by SSSS. Bullous impetigo, on the other hand, is a condition that affects a smaller area. The only etiological difference between the two disorders is the proportion of skin affected (Bukowski et al., 2010; Hardwick et al., 1995). Due to their undeveloped immune systems and inadequate renal toxin clearance, babies and young children are most commonly affected by SSSS. However, the mortality rate is typically relatively low, about 5%, when children receive the proper care. In contrast, this illness has a fatality rate of about 59% when it affects adults, particularly those with immunosuppressed systems (Lyell, 1979; Popov et al., 2014).

Superantigens (SAgs) were initially known as staphylococcal enterotoxins (SEs) because they induce symptoms parallel to *Staphylococcus aureus* food poisonings, such as vomiting and diarrhea. Nevertheless, the International Nomenclature Committee issued SE, this new terminology, in 2004, as some of the most recently identified toxins in this group did not exhibit these emetic properties (Grumann et al., 2014). Superantigens (SAgs)

are bacterial proteins that have strong immunological effects. They interact with a significant number of T cells that exhibit certain T cell receptor V beta domains after being presented by MHC class II molecules.

When a SAg is encountered, reactive T cells are initially stimulated and clonally deleted (Maillard et al., 1997). Many different microbes produce SAgs, which they use to take advantage of the immune system. Bacterial exotoxins, known as SAgs, have been related to several illnesses in humans and animals. The toxic shock syndrome is a well-known example where SAg-reactive cells are assumed to play a significant pathogenic role through the large production of cytokines (Maillard et al., 1997). More than 23 staphylococcal SAgs toxins have been identified, including the toxic shock syndrome toxin (TSST-1) and staphylococcal enterotoxins (SEA to SEE, SEG to SEJ, SEL to SEQ, and SER to SET), as well as 11 staphylococcal superantigen-like (SSL) toxins (SEIK to SEIQ, SEIU to SEIX) (Grumann et al., 2011, 2014; Holtfreter & Bröker, 2005). Bernhard Fleischer and Hubert Schrezenmeier first described the SAgs action mechanism in 1988. Since then, it has been thought that SAgs directly cross-link specific T cell receptor V domains with conserved regions on major histocompatibility complex class II (MHC II) molecules to activate a significant portion of T lymphocytes at once (Grumann et al., 2011; Holtfreter et al., 2006). The ability of macrophages to control T cell responsiveness to SAgs appears to be impacted by MHC II molecules.

The systemically acting SAgs cause a significant number of T-cells to create enormous amounts of pro-inflammatory cytokines (IL-2, IFN- and TNF), which present as symptoms including high fever, rash, desquamation, vomiting, diarrhea, hypotension, and commonly can lead to multiple organ failure (McCormick et al., 2001). Following this cytokine storm, T-cells either fail to respond by failing to multiply or release IL-2, or they can die as a result. This indicates that SAgs are potent immunogens capable of triggering and neutralizing antibody responses (Grumann et al., 2011; McCormick et al., 2001). It is

debatable if SAgs play a part in conditions like sepsis, skin allergies, or respiratory allergies. For instance, it was discovered from the analysis of patients with chronic rhinosinusitis, which is frequently accompanied by intrinsic asthma, that they have high titers of IgE antibodies specific to SAgs in their serum or their polyps, indicating that SAgs cause or at least intensify chronic airway inflammation (Bachert et al., 2010; N. Zhang et al., 2011). The ability of the human immune system to identify and destroy viruses and their antigens has developed over time. SAgs, on the other hand, are the only recognized microbial virulence factor whose immediate function is to purposefully trigger the activation of the adaptive immune system. This defies logic, given the various staphylococcal virulence elements that appear to be created for immune evasion and subversion (Nizet, 2007). Thus, the question of why *S. aureus* produces SAgs arises. Given the diversity and ubiquity of SAg genes, it is likely that these genes would disappear without affecting the fitness of *S. aureus*, especially since they are primarily encoded on mobile genetic elements. In this case, a detailed introspection of the architecture and impact of mobile genetic elements that encode SAg has been published (McGavin et al., 2012).

A secretion signal is present in SAg pro-toxins and is cleaved off the N-terminus following export through the common Sec-dependent secretion pathway. The ultimate toxin product of SAgs is a non-enzymatic, relatively small protein that ranges in size from 22 to 29 kDa. A core α-helix connects the two structurally related domains that make up every SAg. While the smaller C-terminal domain has the β-grasp motif, which is comparable to immunoglobulin-binding domains, the enormous N-terminal domain has a β-barrel motif akin to an oligosaccharide-binding fold (Mitchell et al., 2000). Despite minor or significant variations among the many SAg-mediated T cell activation complexes, each member of the SAg family of toxins can effectively activate vast numbers of T cells. However, the involvement of the Vβ CDR2 loop, the only structural characteristic shared by all described SAgs other than the Vα-specific SEH, appears to be the critical determinant

for Vβ-specificity (Nur-ur Rahman et al., 2011). Staphylococcal enterotoxins B in compound with the human leukocyte antigen (HLA)-DR1 and SEC3 in association with the mouse TCR V8.2-chain were used in early crystallographic investigations to identify a molecular mechanism by which SAgs can activate such a large number of T cells (Jardetzky et al., 1994), (Fields et al., 1996) (Li et al., 1999). These investigations revealed that SAgs attach to the lateral surfaces of both TCRs and pMHC class II molecules to "distort" the typical TCR-pMHC II interaction and push the CDR3 loops of both the TCR α- and β-chains away from the antigenic peptide. This mechanism explains how SAgs are Vβ-specific, as activation of the T cell is no longer antigen-specific but depends on which that specific SAg can bind Vβs (Li et al., 1999). The subsequent release of a large number of proinflammatory cytokines by SAg-activated T cells may, under extreme circumstances, result in the "cytokine storm" phenomena distinctive to TSS (McCormick et al., 2001). Because MyD88 is involved, which upregulates NF-κB and causes the generation of proinflammatory molecules, the activation of antigen-presenting cells (APCs) by SAgs also leads to the release of cytokines (Kissner et al., 2011). There have been numerous new developments in the structural characterization of staphylococcal SAgs in recent years, and it is now clearer how SAgs from various evolutionary groups affect the normal process of T cell activation. For instance, the Group I SAg TSST-1, which is highly specific for human Vβ2+ T cells, binds to the MHC II α-chain through a relatively low-affinity interface strongly impacted by various antigenic peptides within MHC II to produce a distinct T cell activation complex (Choi et al., 1989; J. Kim et al., 1994).

S. aureus allocates a significant portion of its resources to the production of virulence factors to subvert the host's immune system and obtain vital nutrients for its survival. The mechanisms of toxin-mediated host immune evasion and their functions in *S. aureus* virulence were reviewed in the section above. *S. aureus* also generates a significant amount of virulence factors with enzymatic characteristics in addition to the

toxins. Enzymes for the breakdown of tissue components and cofactors that activate host zymogens make up the two basic categories of enzymatic properties. Although the substrates and modes of action of these cofactors and secreted enzymes (exoenzymes) differ, they all work to cleave bacterial and host molecules in order for the bacteria to absorb nutrients, survive, and spread (Tam & Torres, 2019).

Human plasma coagulation caused by *S. aureus* was first described in 1903 (L. Loeb, 1903). The causative agents, coagulase (Coa) and von Willebrand factor binding protein (vWbp), are very active in coagulating human and rabbit plasma (Bjerketorp et al., 2004). One of the primary criteria employed in current medical microbiology for species classification in the genus Staphylococcus is the ability to produce coagulation, distinguishing "coagulase-positive" and "coagulase-negative" species. Most staphylococci are coagulase-negative, but a few, like *S. aureus* and S. intermedius, are coagulase-positive; however, S. schleiferi has both coagulase-positive and coagulase-negative subspecies (Becker et al., 2014).

An old natural defense process known as coagulation captures and immobilizes invasive germs in a clot to protect the body from microbial infections. However, coagulation is also the target of bacterial immunological evasion tactics, as seen with many other host defense processes (Loof et al., 2011; McAdow et al., 2012). Staphylokinase (Sak), von Willebrand factor binding protein (vWbp), and coagulase (Coa) are cofactors made by *Staphylococcus aureus* that do not have enzymatic functions on their own but can activate host zymogens. These three proteins hijack various components of the host coagulation system, modifying the host's natural defenses to increase bacterial survival and spread (Tam & Torres, 2019).Coagulase (Coa) and von Willebrand factor binding protein (vWbp) are two coagulation-promoting proteins released by *S. aureus*, according to Kroh et al., 2009. Both of these proteins activate prothrombin in a non-proteolytic manner. (Kroh et al., 2009).

At their N-terminal ends, coa and vWbp each associate with the prosite of prothrombin to create an active site that can only be created by thrombin. Notably, during Coa- and vWbp-mediated activation, fVa, and fXa do not cleave prothrombin (Friedrich et al., 2003). The length of coa, a protein with roughly 670 amino acids, varies amongst various strains. The α-helical D1-D2 domains of Coa's N-terminal 282 amino acids attach to the C-terminus of the β-chain of prothrombin (Kawabata et al., 1985; Watanabe et al., 2005). A 153-residue linker region follows the D1-D2 domains, although its purpose is unclear. The fibrinogen-binding domain, which is also present at the C-terminal end of another staphylococcal fibrinogen-binding protein, is a 27-residue peptide discovered in the C-terminal end of coa (Friedrich et al., 2003).

At the N-terminus, Coa and vWbp have around 30% protein sequence identity (A. G. Cheng et al., 2010). A D1D2 domain for prothrombin binding is present in both of them. At the C-terminus, they diverge dramatically, however. The 188-residue linker region of Coa's C-terminus is followed by a repeat region made up of tandem repeats of 27 residues that bind fibrinogen. In contrast, the von Willebrand factor (vWF) domain and the fibrinogen binding domain are present in the C-terminus of vWbp (Bjerketorp et al., 2002; McAdow et al., 2012). Coa and vwb are both chromosomally encoded genes. Currently, 12 distinct isoforms of coa have been discovered; most of the variability is related to the significant sequence variability (>50%) of the N-terminus coding region between different strains (McCarthy & Lindsay, 2010; Watanabe et al., 2005). With only two known alleles, vwb, which encodes vWbp, is relatively conserved. However, a recent study discovered many vWbps that coagulate ruminant and horse plasma carried by *Staphylococcus aureus* pathogenicity islands (SaPIs) (Viana et al., 2010; Watanabe et al., 2005).

Coa facilitates the coagulation of soluble fibrinogen, plasma, or blood due to its interaction with fibrinogen and prothrombin (A. G. Cheng et al., 2010). The role of coagulases in disease has been the subject of extensive inquiry. After discovering that *S.*

aureus isolates can cause blood or plasma to clot, microbiologists discovered a link between virulence and a clinical staphylococcal isolate's ability to infect humans (McAdow et al., 2012). Even when the bacteria were cleaned before injection, mice that received coagulase-negative staphylococci cultured with pure coagulase died more frequently. Animal survival, however, was unaffected if coagulase was administered before a challenge with coagulase-negative staphylococci. These findings revealed that the closeness of staphylococci to the resulting coagulation product was necessary for coagulase to play its harmful role (Ekstedt & Yotis, 1960). However, the outcomes of studies looking at the function of coagulase during *S. aureus* infection have not been clear-cut. In mouse models of mastitis or subcutaneous infection, as well as in a rat model of infective endocarditis, chromosomal deletion of coa did not affect *S. aureus* pathogenicity (Phonimdaeng et al., 1990). Contrary to Streptococcus gordonii lacking these genes, heterologous expression of clfA, which encodes the fibrinogen-binding clumping factor of *S. aureus*, increased bacterial adhesion to fibrin platelet thrombi and increased incidence of infective endocarditis in rats (Stutzmann Meier et al., 2001). The screening of several *S. aureus* isolates for coagulase activity and pathogenicity, in contrast, after the intravenous challenge of mice, demonstrated a positive connection between coagulase titer and bacterial burden in the lung. Furthermore, in research by Sawai et al., the wild-type strain recovered much more live bacteria from the lung seven days after infection than the coa deletion mutant did (Sawai et al., 1997). Prior studies used chemical mutagenesis to isolate coagulase-negative *S. aureus* variants, but the mutational lesions were not identified (Haraldsson & Jonsson, 1984). The finding that both coa and vwb encode secreted factors (coagulases) that cause blood to clot gave the field a vital boost. The coa-vwb double mutant showed a considerable reduction in the capacity to generate abscesses or deadly sepsis in mice, in contrast to the targeted deletion of either coa or vwb alone, which only resulted in a moderate drop in virulence (Bjerketorp et al., 2002; A. G. Cheng et al., 2010).

The in vivo activity of coagulase was examined by injecting rabbits with pure coagulase. Fibrinogen levels plummeted at a dose of 2-5 mg Coa, and blood obtained from these rabbits failed to clot, showing that the coagulation system was engaged and fibrinogen stores were exhausted (McAdow et al., 2012). Rabbits died within 20 minutes of receiving a 20 mg Coa injection.

Fibrin thrombi were discovered in the vasculature of the kidneys, adrenal glands, and lung tissues after necropsy (McAdow et al., 2012). The experiment demonstrates the ability of coagulase to clot blood in vivo, even though such high concentrations of coagulase are probably nonphysiological. Coagulases undoubtedly aid in the development of abscesses, a characteristic of *S. aureus* infection (A. G. Cheng et al., 2009). When mice are implanted with collodion bags containing either live or sterile staphylococcal extracts, a fibrinous capsule with a polymorphonuclear infiltrate forms around the bag. Encapsulation is not noticeable when the staphylococcal extracts have lost their coagulase activity. Additionally, animals that were subcutaneously injected with wild-type *S. aureus* instead of the coa mutant developed abscesses (LAM et al., 1963). Immunohistochemical staining demonstrates that prothrombin and fibrin surround and contain Coa and vWbp in abscesses created during *S. aureus* infection. When mice are infected intravenously with *S. aureus* that lacks both coagulases, abscess development in any organ system is virtually eliminated (A. G. Cheng et al., 2010). During an infection, coagulases play an antiphagocytic function. In one investigation, clusters of staphylococci encircled by eosinophilic material were found in the peritoneal lavage fluid after intraperitoneal *S. aureus* infection. Despite the fact that neutrophil recruitment does take place in these circumstances, relatively few of the leukocytes that are recruited can phagocytose staphylococci. These findings inspired the idea that staphylococcal coagulation/clumping allows for staphylococcal escape from phagocytic clearance (Kapral, 1966; Sawai et al., 1997).

Staphylokinase (Sak) is a cofactor that uses host plasmin as a vehicle to activate plasminogen and break down fibrin clots while facilitating bacterial spread. Staphylococci lysogenic strains produce Sak; the prophage carrying the enterotoxin A and chemotaxis inhibitory protein-encoding genes for Sak frequently also carries other virulence factor genes (Coleman et al., 1989; De Haas et al., 2004). The sak gene is carried by three different types of phages (Tam & Torres, 2019). Serotype B phages positively convert Sak without other genes being affected. Some serotype F phages can also facilitate the positive conversion of sak, although phage integration damages the hlb gene (Bokarewa et al., 2006; Winkler et al., 1965). Additionally, it has been noted that the sak-carrying phage alters the coding sequences of peptidoglycan hydrolase and N-acetylmuramyl-L-alanine amidase (Borchardt et al., 1993; Horii et al., 2000). Sak is a protein with a single domain that consists of a core α-helix, a 5-strand β-sheet, and two shorter β-strands (Rabijns et al., 1997). In the serum, Sak and plasmin combine to create Sak-plasmin in a 1:1 combination (LIJNEN et al., 1993). This complex has a very high conversion efficiency for plasminogen to plasmin. Although Sak can also attach to plasminogen, this complex is inert and must be transformed into Sak-plasmin to have enzymatic activity (Collen et al., 1993). The first ten residues at the N-terminus of mature Sak are eliminated in an active Sak-plasmin combination to reveal the charged residue - Lys11. Sak is inactivated when Lys11 is deleted(Gase et al., 1996; Schlott et al., 1997). Sak binding to plasmin directs the active site of plasmin to favor breakage of the activation loop in plasminogen and increases plasminogen to plasmin conversion by improving substrate presentation to plasmin(Parry et al., 1998). Although fibrin-bound complexes are shielded against inactivation, circulating Sak-plasmin complexes are vulnerable to dissociation by α_2-antiplasmin (Lijnen et al., 1991). The fibrin-bound complexes cut human C3b and IgG, preventing the complement system from opsonizing the bacteria. The matrix metalloprotease 1 (MMP-1) can also be activated by Sak-plasmin complexes, which are crucial for leukocyte movement

and activation (Rooijakkers, Van Wamel, et al., 2005; Santala et al., 1999). Sak is significant because it inhibits the bactericidal effects of LL-37 and α-defensins, the two crucial human anti-microbial peptides (AMPs) (Braff et al., 2007; Jin et al., 2004). Sak has a strong species preference. Sak is active for the plasminogens of humans, dogs, goats, rabbits, and sheep but inactive for the plasminogens of mice, pigs, cows, and buffalo (Kwiecinski et al., 2016). Studies using transgenic mice that produce human plasminogen showed that Sak makes it easier for *S. aureus* to penetrate the epidermal barrier and cause big, open sores (Kwiecinski et al., 2013; Peetermans et al., 2014). Plasmin activation, on the other hand, is known to accelerate wound healing and lessen inflammation. Sak may therefore act as a vanguard to establish the primary infection during skin infection, but once the infection is established, Sak reduces the severity of infections to facilitate dispersal (Kwiecinski et al., 2013). Additionally, by activating plasminogen, Sak lowers biofilm synthesis and makes it easier for mature biofilm to be detached. High Sak-producing strains are frequently linked to reduced biofilm formation in vitro and non-invasive infections in humans, supporting these studies (Kwiecinski et al., 2013, 2016).

Cunningham et al. discovered staphylococcal nuclease, formerly known as micrococcal DNase, in the culture supernatants of *S. aureus* in 1956 (Cunningham et al., 1956). Ca^{2+} ions activate nuclease but not by other divalent cations. Thermonuclease is another name for staphylococcal nuclease, which was given to it because it can withstand heat inactivation. Staphylococcal nuclease cleaves the 5'-phosphoryl ester link to degrade DNA and RNA substrates, acting as both an endonuclease and an exonuclease (Cuatrecasas et al., 1967; Cunningham et al., 1956). The sequence of the Staph aureus genome became available with the advent of whole genome sequencing in the late 1990s, which allowed for the discovery of two distinct staphylococcal nuclease genes, nuc (SA0746) and nuc2 (SA1160) (Kuroda et al., 2001; J. Tang et al., 2008). The two genes are controlled by different promoters and are situated in different parts of the genome. The two nucleases

have 42% similarity in the catalytic domain and 34% overall similarity in amino acids (Kiedrowski et al., 2014). Both nucleases are Ca^{2+} dependent, heat resistant, and can employ DNA and RNA as substrates (Kiedrowski et al., 2014). The cellular localization of Nuc and Nuc2 is a crucial distinction. The enzyme Nuc, which has two distinct isoforms, NucB and NucA, is secreted. Nuc2, on the other hand, is surface-bound (Kiedrowski et al., 2011, 2014). The majority of the current understanding of nucleases comes from research done on Nuc. Nuc controls the development of biofilms during infections and facilitates bacterial escape from neutrophil extracellular traps (NETs) (Tam & Torres, 2019). Nuc disperses biofilm by destroying extracellular DNA (eDNA). In strains that do not generate Nuc, biofilm formation is accelerated. Nuclease expression is suppressed during biofilm development, indicating that *S. aureus* regulates nuclease expression to regulate biofilm formation. Furthermore, the nuc mutant shows lower fitness in vivo during intraperitoneal infection (Kiedrowski et al., 2011; Olson et al., 2013). The second function of Nuc is to facilitate bacterial egress from NETs. Invading pathogens are immobilized and made easier to eliminate by the innate immune defense process known as NET, which uses DNA released from dead neutrophils. In order to let *S. aureus* escape, Nuc degrades NETs (Berends et al., 2010; Brinkmann et al., 2004). Additionally, adenosine synthase A (AdsA) uses the monophosphate nucleotides produced when Nuc breaks down DNA in the abscess or NETs as a substrate. AdsA transforms the damaged DNA into deoxyadenosine, which activates caspase-3 and causes the macrophages around the abscess or NET to apoptose, enhancing *S. aureus* survival (Thammavongsa et al., 2013). Due to Nuc2's lower expression levels than Nuc, its role in *S. aureus* pathogenicity is less evident. In vitro experiments have shown that purified Nuc2 can disperse biofilms (Kiedrowski et al., 2014). The nuclease is produced during intramuscular infections in mice, albeit at a much lower level, according to a mutant expressing only Nuc2 and not Nuc. The discovery of Nuc2 in living organisms

raises the possibility that it contributes to *S. aureus* pathogenicity, maybe acting similarly to secreted Nuc on the bacterial surface (Kiedrowski et al., 2014; Tam & Torres, 2019).

A linear polysaccharide called hyaluronic acid (HA) is made up of repeating units of N-acetylglucosamine and glucuronic acid that are joined by alternating β−1,3 and β−1,4 glycosidic linkages (Laurent & Fraser, 1992). In vertebrates, HA is an essential part of the extracellular matrix (ECM), which give cells and tissues homeostasis and structural integrity. It is also essential for the control of the immune system. HA-degrading enzymes are collectively referred to as hyaluronate lyases or hyaluronidase (Lee-Sayer et al., 2015; Monslow et al., 2015). Hyaluronidases are present in bacteria, invertebrates, and vertebrates in nature. Both invertebrates and vertebrates have hyaluronidases, which convert HA to tetrasaccharides. However, bacterial hyaluronidases function as endo-N-aceylhexoaminidases and cleave the β−1,4 linkage in a method known as β−elimination, converting HA into unsaturated disaccharides (Hynes & Walton, 2000).

The only staphylococci that have been found to produce hyaluronidase are *S. aureus* and S. hyicus. Duran-Reynals first identified staphylococcal hyaluronidase activity as a "spreading agent" that enlarged lesions in a rabbit skin infection model in 1933(Duran-Reynal, 1933; Hart et al., 2009). Chain and Duthie later identified hyaluronidase as this "spreading agent" in 1940. The staphylococcal hyaluronidase gene, hysA, would eventually be cloned and the associated protein purified in 1995, in any case (Chain & Duthie, 1940; Farrell et al., 1995). By dissolving HA in ECMs and biofilms, hyaluronidase is considered a "spreading factor" in the spread of bacteria. ECMs are particularly prevalent in the skin and lungs. In skin and lung infection models, deletion of hysA decreased skin and lung pathology and decreased bacterial burden, respectively (Ibberson et al., 2014; Makris et al., 2004). Additionally, it has been shown that hysA deletion results in increased biofilm formation and decreased bacterial spread (Ibberson et al., 2016).

Both β-toxin and phosphatidylinositol-specific phospholipase C (PI-PLC) are phospholipases that *S. aureus* is capable of producing. In *S. aureus* culture supernatants, the staphylococcal phospholipase, PI-PLC, was first identified in the 1960s. PI-PLC degraded phosphatidyl inosito(J. Cheng et al., 2012)l (PI) to diglyceride and inositol phosphate (Doery et al., 1965; Magnusson et al., 1962). The only staphylococci known to manufacture PI-PLC now are still *S. aureus*. Since the membrane of *S. aureus* does not contain PI, it is believed that to adapt to the host's environment, *S. aureus* evolved PI-PLC (Beining et al., 1975; Daugherty & Low, 1993). The staphylococcal PI-PLC, like other bacterial PI-PLCs, has an incomplete $(\beta\alpha)_8$-barrel structure, also known as the TIM barrel (Goldstein et al., 2012). The PI-PLC active site is conserved and is positioned at the C-terminus of the β-strands that comprise the β-barrel (Goldstein et al., 2012). The crystal structure of staphylococcal PI-PLC was solved, which explained many of its metabolic features. The optimal pH range for PI-PLC is stated to be 5.5-6.0. The free substrate access to the active site under acidic circumstances explains this characteristic. Under basic conditions, however, the substrate's accessibility is limited (Daugherty & Low, 1993; Goldstein et al., 2012). NaCl, HgCl2, and Cu2SO4 all inhibit PI-PLC activity (Daugherty & Low, 1993). The strong electropositivity of the barrel rim region and the active site can be used to explain the PI-salt PLC's sensitivity. Phosphocholine (PC) boosts PI-PLC activity when it is present (Goldstein et al., 2012). Phosphocholine (PC) boosts PI-PLC activity when it is present.The presence of PC, according to structural studies of PI-PLC, permits the transitory dimerization of two PI-PLC monomers, which increases enzyme activity (J. Cheng et al., 2012; Goldstein et al., 2012). Phosphatidylinositol (PI) is digested by bacterial PI-PLC in two phases. The first process results in the intermediate products myo-inositol 1,2-cyclic phosphate and diacylglycerol (DAG) (cIP). Secondly, slower hydrolysis of cIP to myo-inositol 1-phosphate (IP) occurs after that (O. H. Griffith & Ryan, 1999). In order to activate intracellular pathways in mammalian cells for growth and

survival, DAG is a crucial secondary messenger (Flores-Díaz et al., 2016). Additionally, PI-PLC has the capacity to liberate proteins attached to cell membranes via glycosylphosphatidylinositol (GPI) (Daugherty & Low, 1993). The decay-accelerating factor (DAF) and the C8 binding protein are two examples of such proteins. Both proteins serve as complement regulators that prevent complements from activating on their cells when they are typically present on host cells. Recent research has shown that PI-PLC helps *S. aureus* survive in human blood and neutrophils (Walter et al., 1992; White et al., 2014).

S. aureus possesses two lipases: *S. aureus* lipase 1 and *S. aureus* lipase 2. (SAL1 & SAL2). SAL1 and SAL2 genes are often annotated as gehA and gehB for glycerol ester hydrolase, respectively (Cadieux et al., 2014; Götz et al., 1998). In the literature, SAL1 is also known as lip1. The two genes are encoded in different parts of the *S. aureus* genome, although they have protein sequence similarities with each other and other staphylococcal lipases (Götz et al., 1998; Rosenstein & Götz, 2000). Lipases are synthesized as pre-pro-enzymes. Signal peptidase I cleaves the signal peptide and secretes it from the pre-pro-enzyme. The mature lipase is produced when aureolysin cleaves the secreted pro-enzyme. On the other hand, cleavage of the pro-peptide is unnecessary and has no influence on enzyme activity (Cadieux et al., 2014; Rollof & Normark, 1992). The lipase pro-peptides were discovered to be significant for the translocation of the lipases to the extracellular milieu and stabilizing the proteins to avoid degradation using chimeric lipases of S. hyicus produced in S. carnosus (Demleitner & Götz, 1994; Liebl & Götz, 1986). The conserved catalytic trio of serine, aspartate, and histidine gives lipases their enzymatic properties. SAL1 and SAL2 vary biochemically and prefer distinct substrates while having a similar catalytic mechanism. SAL1 is most effective at a pH of 6.0 and stable in acidic environments, although it is inactivated at a pH greater than 10 (Cadieux et al., 2014; Simons et al., 1996). Analyses using biochemical and molecular methods revealed that Ca2+ promotes the activity of SAL1 and stabilizes the structure of SAL1. Chelators like

EDTA or EGTA, therefore, prevent SAL1 action. Short-chain triglycerides are strongly preferred by SAL1, whereas long-chain triglycerides cannot be hydrolyzed by SAL1 (Simons et al., 1996). SAL2, in contrast, is inert in acidic environments and operates best at a pH of 8.0. Ca2+ is not required for increased SAL2 activity (Rollof, Hedström, et al., 1987). As a result, chelators have little influence on activity. The preferred substrate for SAL2 is long-chain triglycerides (Cadieux et al., 2014). SAL2, on the other hand, has been demonstrated to hydrolyze short-chain triglycerides, mono- and di-glycerides with lesser efficiency and no apparent positional selectivity (Cadieux et al., 2014).

Lipases are conserved in staphylococcal species, suggesting their evolutionary relevance. However, the role of lipases in illness is unknown. Lipases are produced in more significant quantities by *S. aureus* clinical isolates from deep tissue infections than isolates from superficial infections (Rollof, HedstrÖM, et al., 1987). At high concentrations, purified lipases produce granulocyte aggregation and impair phagocytosis (Rollof et al., 1992). In a mouse intraperitoneal infection model, SAL2 was demonstrated to be crucial for the development of biofilms and the pathogenicity of *S. aureus* strain RN4220.These findings imply that lipases play a role in *S. aureus* pathogenicity and increase bacterial survival in biofilms and abscesses.

Paradoxically, antibacterial free fatty acids are released during lipase-mediated triglyceride hydrolysis, which can interfere with pathogenicity. Most lipase-producing bacteria can detoxify these bactericidal fatty acids through fatty acid-modifying enzymes (C. Hu et al., 2012; Shryock et al., 1992).

Mortensen et al. discovered fatty acid modifying enzyme (FAME) in 1992 after discovering that *S. aureus* culture filtrates hindered the bactericidal actions of host lipids in abscesses. FAME activity has been well-documented in several staphylococcal species since its discovery (Chamberlain & Brueggemann, 1997; Mortensen et al., 1992). This enzyme is produced by approximately 80% of *S. aureus* and S. epidermidis. Despite its

frequency, neither the matching gene nor the protein for FAME has been found (Lu et al., 2012). The bactericidal-free lipids are esterified with an alcohol substrate by FAME to produce alcohol esters, which aid in the survival of staphylococci. FAME likes to esterify cholesterol, which is particularly prevalent in abscesses, even though it can also do so with other free lipids such as methanol, ethanol, 1-propanol, 2-propanol, and 1-butanol (Mortensen et al., 1992). FAME readily esterifies saturated and unsaturated fatty acids with 15-19 carbons; nevertheless, esterification of fatty acid chains with 11-24 carbons is also reported (Kapral et al., 1992). The enzyme's ideal pH varies from 5.0 to 5.5, and its optimal temperature is 40°C. Di- and tri-glycerides containing unsaturated fatty acid side chains decrease enzyme activity (Kapral et al., 1992; Mortensen et al., 1992). Lipases and FAME are hypothesized to work together to help staphylococci survive abscesses. While lipases degrade triglycerides, which impede FAME activity, FAME processes the free fatty acids released by lipases to protect staphylococci. This notion is supported by the discovery that most *S. aureus* strains carrying genes for lipases have FAME activities, which are connected to the bacteria's invasiveness in vivo (J. P. Long & Kapral, 1993; Lu et al., 2012).

S. aureus expends energy extensively on producing virulence factors to defend itself from host immune surveillance and increase bacterial survival in unfavorable conditions. The relevance of these virulence factors during infection has been thoroughly proven in several ex vivo and in vivo infection models. Pathogenic *S. aureus* is typically found in adverse host habitats with limited resources; hence, creating several virulence factors that serve the same function can be a waste of scarce resources and detrimental to survival (Tam & Torres, 2019). In contrast, this redundancy can protect the bacteria if one of the virulence factors becomes ineffective. Alternatively, the bacteria might have evolved these redundant virulence characteristics to better adapt to diverse types of infections or colonization locations. With antibiotic resistance in bacteria, especially *S. aureus*, there is an urgent need to develop innovative treatments and vaccines to tackle this lethal infection.

Understanding the functions of these essential virulence factors throughout illness progression can offer the knowledge needed to create better therapies and find vaccination targets (Tam & Torres, 2019).

Resistance of *S. aureus*

Staphylococcus aureus is continually evolving, thanks to mutations and the acquisition of mobile genetic elements that provide more excellent resistance and virulence. Since the 1960s, hospitals have had to deal with the advent of methicillin-resistant *S. aureus* (MRSA) strains, which spread faster and are more challenging to treat than methicillin-susceptible *S. aureus* (MSSA) (Aryee & Edgeworth, 2016). Distinct community MRSA strains have also emerged, since the 1980s, causing severe skin and respiratory infections. Antibiotic resistance is a significant public health issue, and antibiotic use is increasingly being identified as the primary selective pressure driving this resistance. (Goossens et al., 2005b). The spread of multi-resistant bacterial strains in the community is exacerbated by a paucity of new antibiotic classes in development (Boucher et al., 2009). According to Reygaert, the medical community believed that the war against infectious illnesses ended once antibiotics were developed. The battle has, however, appeared to turn in favor of the bacteria due to the widespread development of antimicrobial agent resistance among bacteria. Currently, infectious diseases are a substantial cause of morbidity and mortality in the world. (W. C. Reygaert, 2018). According to a World Health Organization (WHO) evaluation of these illnesses, the maximum causalities of morbidity and death are lower respiratory infections, diarrheal illnesses, HIV/AIDS, and malaria (World Health Organization, 2014). Antimicrobial resistance has dramatically exacerbated the effects of infectious illnesses, increasing the frequency of infections and the expense of treatment. There is established antimicrobial resistance to every one of the many antimicrobial medicines we have at our disposal for possible infection therapy, and this resistance develops quickly when a new medication

is given the go-ahead. The WHO's Global Action Plan on Antimicrobial Resistance was introduced in 2015 due to these worries (World Health Organization, 2015).

The mechanism of antimicrobial action allows for the grouping of antimicrobial agents. The primary categories include substances that prevent the formation of bacterial cell walls, depolarize cell membranes, prevent protein synthesis, prevent the synthesis of nucleic acids, and prevent the synthesis of certain enzymes (W. C. Reygaert, 2018). With such a diverse range of processes, humans would appear to have more control over the microorganisms. Unfortunately, poor management of antimicrobial drugs has contributed to the massive problem of resistance we currently encounter. Increased intake of antimicrobial medications by humans and animals, as well as poor antimicrobial therapy prescription, are two factors that have contributed to the problem of increasing resistance. Physicians may overprescribe certain popular antimicrobial drugs because they select their medications based on cheap cost and low toxicity (M. Griffith et al., 2012). Antimicrobials may occasionally be prescribed incorrectly, such as when a broad-spectrum medication is first prescribed even when it is not essential or is subsequently determined to be ineffective for the microorganisms causing the infirmity (Yu, 2011). Furthermore, a patient who has previously used antimicrobial medications runs the risk of contracting a drug-resistant organism, and individuals who have used antimicrobials the most often are more likely to have resistant bacterial infections (M. Griffith et al., 2012; Tacconelli, 2009). When antimicrobials are used excessively, resistance develops either as a result of point mutations or the acquisition of foreign resistance genes, which modify the antimicrobial target and cause it to degrade or decrease the concentration of antimicrobials inside the cell (Alekshun & Levy, 2007).

The classes and dosages of antibiotics administered to the animals are reflections of the antimicrobial resistance patterns witnessed in the animals. There are several potential routes by which antimicrobial resistance might spread from animals to people,

with the direct oral route being the most prevalent. Another typical route is direct human interaction with animals (Wegener, 2012). Antibiotics have been used for many years for veterinary purposes to remedy or prevent infection in livestock raised for food. The antibiotics used in animal feed originate from the majority of the antimicrobial groups utilized in human medicine, and they are frequently present in dosages ranging from below therapeutic levels to total therapeutic levels. Evidence supports the hypothesis that administering antibiotics to livestock may lead to the development of antimicrobial-resistant microbes, which may circulate to individuals who consume these livestock (Landers et al., 2012; Wegener, 2012). There are now fewer alternatives for treating patients due to ongoing increases in antibiotic resistance, and morbidity and death have risen as a result. As a result, humanity is now dealing with more severe infections requiring extensive treatment, and lengthier illness courses frequently necessitate prolonged hospitalization (W. C. Reygaert, 2018).

Pontes et al. state that virtually all the antibiotic families on the pharmaceutical market are resistant to Gram-positive and Gram-negative bacteria, which are responsible for human and animal infections. In addition to having a significant negative socioeconomic and ecological impact, antibiotic resistance has become a major issue for contemporary medicine and the pharmaceutical business.(Pontes et al., 2018). In the most recent decades, there has been an increase in the number of clinical and particularly among the most prevalent common human infections from the ESKAPE group (Enterococcus faeciumi, *Staphylococcus aureus*, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa, Enterobacter spp.), and environmental multi-resistant isolates have been seen (Boucher et al., 2009; Carlet et al., 2011). The problem of antibiotic resistance is made worse by this growth. The medical community's concern over drug resistance development has recently increased due to the failure of current antibiotic medicines to treat bacterial illnesses (Clatworthy et al., 2007; Tillotson &

Zinner, 2017). Clinical resistance is strongly linked to the indiscriminate use of antibiotics, unrestricted medication sales in healthcare, livestock farming, and poultry production. This abuse has spread to the environment, with unnecessary additives added to toothpaste and hand soap as preventative measures. Promiscuous Horizontal Genetic Transfers (HGT) between bacteria of various species or genera and bacterial exposure to other selection agents such as contaminants, heavy metals, and disinfectants may have also played a key role (Barriere, 2015; Fernandez-Lopez et al., 2016; Shafaati et al., 2016). Comprehension of the biochemical and molecular functions; thus, the molecules involved in antibiotic resistance is not a simple exercise. This is because the mode of action of antibiotics and the resultant mechanisms of resistance are intimately associated (Džidić et al., 2008; Hughes & Andersson, 2017). Genetic and environmental factors influence phenotypic resistance changes, and on occasion, genotypes are found that do not match expected phenotypes (Hughes & Andersson, 2017).

Antimicrobial drug effectiveness in the treatment of Staph aureus infections may depend on a variety of other factors, including the growth phase of the bacterium, such as the logarithmic growth phase or stationary phase; the localization of the infection, as drugs have varying concentrations in different parts of the body; biofilm formation; and many others. The type of infection, such as an epidemic, nosocomial, or chronic infection, which may be associated with the presence or absence of a specific pathogenicity factor, should also be considered when developing a treatment plan (Mlynarczyk-Bonikowska et al., 2022).

Before delving into the many components of antimicrobial resistance, it is essential to distinguish between resistance and persistence. Without further mutations, all progeny of a bacterium resistant to one antimicrobial agent would similarly be resistant to that agent. However, persistence refers to bacterial cells that lack resistance genes yet are not drug-susceptible. Since most antimicrobial medicines have little impact

on cells that are not actively growing and dividing, the persistence is undoubtedly caused by the possibility that some bacteria may be in a period of stalled growth known as dormancy. In a culture in a stationary phase, these persister cells appear at a rate of around 1% (Keren et al., 2004; Wood et al., 2013).

Bacteria as species are not always equally vulnerable to or resistant to a given antibiotic. Within bacterial groups that are closely related, resistance levels might vary substantially. The minor medication concentration essential to stop bacterial growth is known as the minimum inhibitory concentration or MIC, and it is used to determine both susceptibility and resistance. Susceptibility is a range of average MICs for any given drug across the same bacterial species. A species is said to have an inherent resistance to therapy if its average MIC falls within the spectrum of resistance.

The degree of resistance will vary based on the species and the genes gained, as bacteria may also obtain resistance genes from other related organisms (Coculescu, 2009; Martinez, 2014). Inherent or induced natural resistance are both possible. A feature that is uniformly present within a bacterial species, unaffected by prior antibiotic exposure, and unrelated to horizontal gene transfer is referred to as intrinsic resistance (Cox & Wright, 2013; Martinez, 2014). Reduced outer membrane permeability, most notably caused by lipopolysaccharide and naturally occurring efflux pumps, are two of the most frequent bacterial processes implicated in intrinsic resistance. Induced resistance is frequently caused by multidrug-efflux pumps as well (Cox & Wright, 2013; Fajardo et al., 2008).

Bacteria can acquire genetic material that imparts resistance through transformation, transposition, and conjugation—the three primary ways bacteria acquire any genetic material. The bacterium can get chromosomal DNA alterations (W. C. Reygaert, 2018). The acquisition might be either temporary or permanent. The most frequent way to acquire exogenous genetic material is the plasmid-mediated transfer of

resistance genes; bacteriophage-borne transmission is uncommon. Stressors, including starvation, UV radiation, and chemicals on the bacterium, are typical sources of genetic changes like substitutions and deletions. Internally, insertion sequences and integrins may shift genetic material around. Bacteria typically experience one mutation per 10^6 to 10^9 cell divisions, and the majority of these mutations are detrimental to the cell (Coculescu, 2009; J & D, 2010). Genes encoding drug targets, drug transporters, regulators that govern drug transporters, and antibiotic-modifying enzymes are often the only ones to experience mutations that contribute to antimicrobial resistance (Martinez, 2014). Furthermore, many of the changes that confer antimicrobial resistance come at a cost to the organism. For instance, when *S. aureus* acquires methicillin resistance, the growth rate of the bacteria is considerably reduced (W. Reygaert, 2009).

One major dilemma of antimicrobial resistance is that using these medications increases resistance. Using these medications increases resistance is a major problem with antimicrobial resistance. Bacteria can develop significant resistance levels even in response to low doses of antimicrobials. Also, the bacteria's hypermutable strains may be chosen, their capacity to develop resistance to additional antimicrobial agents may improve, and mobile genetic components may be encouraged to migrate (Blázquez et al., 2012).

Antimicrobial resistance mechanisms may be divided into four primary groups: (1) decreasing medication absorption, (2) altering drug targets, (3) rendering drugs inactive, and (4) active drug efflux. Limiting drug uptake, drug inactivation, and drug efflux are intrinsic resistance strategies; drug target modification, drug inactivation, and drug efflux are examples of acquired resistance strategies. The methods utilized by gram-negative and gram-positive bacteria differ due to structural variations. Gram-positive bacteria lack the capacity for some drug efflux mechanisms, while gram-negative

bacteria frequently restrict medication absorption and use all four primary methods (Chancey et al., 2012; Mahon et al., 2015).

As previously stated, there is a natural variance in bacteria's capacity to inhibit antimicrobial agent uptake. In gram-negative bacteria, the structure and functions of the LPS layer operate as a barrier to specific types of chemicals. Due to this, certain bacteria have an inbuilt resistance to specific classes of powerful antimicrobial medicines(Blair et al., 2014). Since Gram-positive bacteria lack an outer barrier, limiting medication access is less common. The enterococci have an inherent resistance to aminoglycosides because polar molecules have trouble penetrating the cell wall. *S. aureus*, a different gram-positive bacterium, has recently gained vancomycin resistance. Out of the two defense mechanisms *S. aureus* employs against vancomycin, one causes the bacterium to create a thickened cell wall that makes it challenging for the medication to penetrate the cell and confers an intermediate level of resistance to vancomycin. These strains have the VISA designation (Lambert, 2002; Miller et al., 2014). Bacteria use porin channels with thick outer membranes to allow substances access to the cell. In gram-negative bacteria, hydrophilic molecules can typically be accessed through the porin channels(Blair et al., 2014). Porin alterations can restrict drug uptake in two main ways: by reducing the quantity of porins present and changing the porin channel's selectivity through mutations (A. Kumar & Schweizer, 2005). It is known that members of the Enterobacteriaceae family can develop resistance by having fewer porins and sometimes stopping production entirely of certain porins. These bacteria all work together to decrease the number of porins as a defense against carbapenems (Cornaglia et al., 1996). Due to mutations that change the porin channel, Neisseria gonorrhoeae and E. aerogenes are both resistant to imipenem, certain cephalosporins, β-lactam antibiotics, and tetracycline (Thiolas et al., 2004). The development of a biofilm by a bacterial community during bacterial colonization is another often observed event. The biofilm

community of the typical flora in the gut is an example of a biofilm community that contains a dominating organism and a diverse range of organisms (W. C. Reygaert, 2018). Antimicrobials that target maturing, diverging bacterial cells have a limited impact because the bacterial cells in the biofilm have a tendency to reduce metabolic rate and cell division. A significant finding concerning biofilms is that it is probable that the proximity of the bacterial cells facilitates horizontal gene transfer. This suggests that the exchange of genes for antimicrobial resistance among these bacterial groups may be more superficial (W. C. Reygaert, 2018; Van Acker et al., 2014).

There are several parts of the bacterial cell that antimicrobial substances may target, and there are just as many targets that the bacteria could modify to confer resistance to those medications. Modifications in the structure and quantity of penicillin-binding proteins (PBPs) are one method of resistance to the β-lactam medicines utilized almost exclusively by gram-positive bacteria. PBPs are transpeptidases that aid in forming peptidoglycan in the cell wall. The quantity of medication that can bind to that target is affected by changes in the number of PBPs. For instance, an increase in PBPs with a decreased capacity for drug binding or a decrease in PBPs with an average capacity. When *S. aureus* acquires the *mecA* gene, PBP2a undergoes structural changes that may reduce or completely prevent drug binding (Beceiro et al., 2013; W. Reygaert, 2009). In 2013, Randall et al. published research showing that while lipopeptides like daptomycin depolarize cell membranes, glycopeptides like vancomycin also function by blocking cell wall construction. These medications are intrinsically resistant to Gram-negative bacteria with a thick LPS coating (Randall et al., 2013). *S. aureus* and enterococci now face a severe challenge with vancomycin resistance. Due to the acquisition of van genes by resistant species, vancomycin's affinity for peptidoglycan precursors is diminished (Beceiro et al., 2013; Cox & Wright, 2013). Calcium is necessary for the binding of daptomycin. Gene mutations cause the cell membrane

surface to become positively charged, which prevents calcium from attaching to the surface (Stefani et al., 2015; Yang et al., 2009). The erm genes are primarily affected by drugs that target the ribosomal subunits. However, ribosomal mutation, methylation of the ribosomal subunits, or ribosomal protection can also result in resistance. These procedures stop the drug from binding to the ribosome. The level of drug interference varies significantly between these systems (S. Kumar et al., 2013; Roberts, 2004). Resistance to medications that target the synthesis of nucleic acids, such as fluoroquinolones, is caused by changes in DNA gyrase or topoisomerase IV. Gyrase and topoisomerase undergo modifications, altering their structural properties and reducing or completely abolishing the drug's capacity to bind to these components (Hawkey, 2003; Redgrave et al., 2014). The folate biosynthesis pathway's enzymes can change, leading to increased production of resistant DHPS (dihydropteroate synthase) and DHFR (dihydrofolate reductase) enzymes, which are the primary mechanisms by which medications that inhibit metabolic pathways become resistant. The sulfonamides and trimethoprim bind to the relevant enzymes as structural analogs of the corresponding natural substrates. These drugs connect to the enzymes' active site and compete with the enzymes to suppress their activity (Huovinen et al., 1995; Vedantam et al., 1998).

One of two ways bacteria can render a medicine inactive is by physically degrading it or adding a chemical group. "β-lactamases" refers to a broad class of enzymes that hydrolyze drugs. Another drug that can be rendered inactive by hydrolysis due to the tetX gene is tetracycline (Blair et al., 2015; S. Kumar et al., 2013). Acetyl, phosphoryl, and adenyl are the three chemical groups most frequently employed to impart chemical properties to the medication. A significant amount of transferases have been discovered. The most popular technique is acetylation, which works well against fluoroquinolones, chloramphenicol, streptogramins, and aminoglycosides. It is generally

known that phosphorylation and adenylation primarily target aminoglycosides (Blair et al., 2015; Schwarz et al., 2004).

In bacteria, the genes for efflux pumps are chromosomally encoded. Others are triggered or overexpressed in response to specific environmental stimuli or when a suitable substrate is present, while some are expressed by nature. The efflux pumps carry a wide range of molecules, and their primary purpose is to purge the bacterial cell of hazardous toxins. How readily available a carbon source is affecting the resistance capacity of several of these pumps (Blair et al., 2014; Villagra et al., 2012). A wide variety of efflux pumps are found in most microorganisms. Bacteria's efflux pumps can be divided into five significant families according to their structure and energy source: the ATP-binding cassette (ABC) family, the multidrug and toxic compound extrusion (MATE) family, the small multidrug resistance (SMR) family, the major facilitator superfamily (MFS), and the resistance-nodulation-cell division (RND) family. These efflux pumps are typically single-component pumps transporting substrates through the cytoplasmic membrane. The OMP-porin and the periplasmic membrane fusion protein (MFP) cooperate with the RND family of multi-component pumps, which are virtually exclusively found in gram-negative bacteria, to efflux substrate over the whole cell envelope (Blair et al., 2014; A. Kumar & Schweizer, 2005; Piddock, 2006a). In some instances, additional members of the efflux family work with other cellular components as multicomponent pumps in gram-negative bacteria. A tripartite pump called MacB, one of the ABC family members is used to extrude macrolide medicines. EmrB, an MFS member, works as a tripartite pump in E. coli to extrude nalidixic acid (Jo et al., 2017; Tanabe et al., 2009). The chromosomal encoding of efflux pumps raises the possibility of conferring intrinsic resistance on gram-positive bacteria. These pumps include members of the MATE and MFS families and fluoroquinolone efflux pumps. Additionally, gram-positive efflux pumps have been discovered in plasmids. Currently,

gram-positive bacteria have provided descriptions of MFS family pumps (S. S. Costa et al., 2013; Jonas et al., 2001). All five families of efflux pumps may be found in gram-negative bacteria, with the RND family having the most therapeutically relevant pumps (Blair et al., 2014; Kourtesi et al., 2013).

A sequence of waves can be used to visualize the development of antibiotic resistance in *S. aureus*. As the percentage of infections brought on by penicillin-resistant *S. aureus* rose in hospitals, the first wave started to emerge in the middle of the 1940s (Chambers & DeLeo, 2009). These strains produced a penicillinase required to hydrolyze the beta-lactam ring of penicillin, which gives penicillin its antibacterial characteristics. A plasmid encodes this penicillinase. Penicillin-resistant strains were a factor in several diseases then; by the early 1950s and 1960s, they had taken off like wildfire(Rountree & Freeman, 1955). Phage-type 80/81 of the *S. aureus* clone was predominantly responsible for these infections in hospitals and the general population (Rountree & Freeman, 1955). Following the development of methicillin, pandemic phage-type 80/81 *S. aureus* infections mostly disappeared, although penicillinase-producing strains of other *S. aureus* lineages have remained ordinary (Chambers & DeLeo, 2009). The second wave of resistance starts to emerge with the introduction of methicillin. *S. aureus* strains resistant to methicillin were first reported in 1961 (BARBER, 1961). Even though the specific gene, *mecA*, the methicillin resistance determinant that codes for the low-affinity penicillin-binding protein, PBP 2a, was not found for over 20 years, it was understood early on that the resistance mechanism was different from penicillinase-mediated resistance because there was no drug inactivation (Chambers & DeLeo, 2009). Contrary to penicillinase-mediated resistance, which has a narrower scope, methicillin resistance is a broad beta-lactam antibiotic class resistance to penicillins, cephalosporins, and carbapenems. The archetypal strain COL, a member of the so-called "archaic" clone of MRSA and possibly the most extensively studied MRSA strain, was found in a patient in

Colindale, United Kingdom, in 1960. It is one of the very first MRSA clinical isolates (Jevons, 1961). The most productive lineage of MRSA, which includes strains linked to hospitals and the general public, includes COL. Until the 1970s, these antiquated MRSA strains were common in hospitals across Europe (Chambers & DeLeo, 2009).

The majority of antimicrobial medications used in therapy are now resistant to practically all Staph aureus strains. The most significant of these is resistance to the tetracycline, beta-lactams, glycopeptides, and oxazolidinones, which are the most often prescribed medications for treating Gram-positive infections (Mlynarczyk-Bonikowska et al., 2022). In the past, penicillin was the antibiotic of choice for treating *S. aureus* infections; however, penicillin resistance is quite prevalent in most nations (Levy & Bonnie, 2004). As a result, as first-line therapy, a penicillinase-resistant beta-lactam antibiotic having the exact mechanism of action as penicillin, such as flucloxacillin or oxacillin, is routinely used (Alanis, 2005; S. S. Tang et al., 2014). In addition to being resistant to methicillin, *S. aureus* strains are also resistant to penicillins (penicillin V, penicillin G, ampicillin, oxacillin, carbenicillin, and amoxicillin), carbapenems (imipenem-cilastatin [Primaxin]), cephalosporins (cephalothin), and monobactams (Rayner & Munckhof, 2005b). Critical bacterial processes that are targeted by these antimicrobials include cell wall synthesis (beta-lactams and glycopeptides), protein synthesis (aminoglycosides, tetracyclines, macrolides, lincosamides, chloramphenicol, mupirocin, and fusidic acid), nucleic acid synthesis (quinolones), RNA synthesis (rifampin), and metabolic pathways like folic (sulphonamides and trimethoprim)(Alekshun & Levy, 2007; Tenover et al., 2006).

In the 1940s, Fleming made the discovery of penicillin and launched the use of antibiotics to treat infections (Klein et al., 2017). *S. aureus*-related infectious illnesses were under control then, but penicillin resistance emerged in the 1950s with its widespread use. *S. aureus* penicillin-resistant strains first appeared quickly after the

antibiotic was developed in the early 1940s (Pichereau & Rose, 2010; C. Walsh & Wencewicz, 2016). According to Lowy, in 2003, Kirby was the first to show that *S. aureus* strains with penicillin resistance rendered penicillin inactive (Lowy, 2003). They produced a beta-lactamase that hydrolyzed the essential beta-lactam link and rendered the medication inactive against bacteria. Semi-synthetic penicillin versions that were not substrates for beta-lactamase were produced by replacing the native aminoadipoyl chain with bulkier moieties (Foster, 2017). Methicillin, novel semi-synthetic penicillin that is resistant to the hydrolysis of beta-lactamase was later created by scientists (Khoshnood et al., 2019). Infection caused by penicillin-resistant *S. aureus* was successfully managed by methicillin after it was introduced to the clinic in 1959 (Jokinen et al., 2017). Methicillin-sensitive Staph aureus has a gene encoding the penicillin-binding protein 2a, or 2′ (PBP2a or PBP2′) (*mecA*) integrated into its chromosomal element (SCCmec), but only two years after the medication was released, in 1961, a British researcher by the name of Jevons reported the isolation of an MRSA strain (Schulte & Munson, 2019). The transferable enzyme beta-lactamase, expressed by bacterial chromosomal genes, is responsible for catalyzing the hydrolysis of a variety of beta-lactam antibiotics, including broad-spectrum antibiotics like carbapenem (Y. D. Lee & Park, 2016). According to current research, beta-lactam antibiotics extinguish bacteria primarily through two mechanisms: first, by binding to penicillin-binding protein, which inhibits the production of cell wall mucin, disrupts the cell wall, and induces bacterial expansion and lysis; second, by inducing the activity of the bacteria's autolytic enzyme, which results in autolysis and death (Matono et al., 2018). Antibiotic effectiveness is primarily decreased by excessive MRSA beta-lactamase release via two processes, which results in MRSA resistance (Khan et al., 2014). Beta-lactamase hydrolyzes and renders inactive beta-lactam antibiotics, which is the first mechanism. The second method is known as "pinching," whereby a significant amount of beta-lactamase binds quickly and firmly to

extracellular antibiotics to prevent the antibiotics from entering the intracellular space and, as a result, from reaching the target site. This ultimately results in MRSA resistance to antibiotics (Harada et al., 2014; Hashizume et al., 2017).

Bondi and Dietz later discovered Penicillinase's precise function. Regardless of the clinical environment, more than 90% of staphylococcal isolates now generate Penicillinase. A transposable element carrying the beta-lactamase gene is part of a giant plasmid that frequently contains other antimicrobial resistance genes (Bondi & Dietz, 1945). The process of producing beta-lactamases is one that frequently occurs in staphylococci. Ambler and Bush categorized the beta-lactamases produced by Staph aureus as belonging to class A and 2a, respectively (Bush & Jacoby, 2010). MSSA and most MRSA both synthesize them regularly. The blaI-blaR1-*blaZ* operon, which is present in many plasmids and transposons, often contains the *blaZ* gene, which encodes these enzymes (García-Álvarez et al., 2011; Młynarczyk et al., 1998b). The beta-lactamase-coding *blaZ* gene is responsible for the staphylococcal resistance to penicillin. This primarily extracellular enzyme, which hydrolyzes the beta-lactam ring and renders the beta-lactam inactive, is produced by staphylococci when exposed to beta-lactam antibiotics. Two adjacent regulatory genes that control *blaZ* are the repressor blaI and the antirepressor blaR1 (Lowy, 2003). According to recent research, the regulatory proteins BlaR1 and BlaI must be cleaved one after the other for the signaling pathway that leads to the creation of beta-lactamases. A transmembrane sensor transducer called BlaR1 cleaves itself after being exposed to beta-lactams (Gregory et al., 1997; H. Z. Zhang et al., 2001). According to Zhang et al., the cleaved protein acts as a protease that cleaves the repressor BlaI directly or indirectly, enabling *blaZ* to produce an enzyme (H. Z. Zhang et al., 2001).

The revelation that *mecA* is always located inside a mobile cassette element by Hiramatsu and colleagues was a significant step forward in our knowledge of the biology

of methicillin resistance and gave us another tool for identifying evolutionary links among MRSA (Ito et al., 2001). SCCmec, a mobile genetic element, carries the functional gene *mecA*, which results in methicillin resistance and other genes (Chambers & DeLeo, 2009). According to Zong et al., the mec gene complex and the ccr gene complex are two crucial parts of SCCmec. The cassette chromosome recombinase (ccr) genes (ccrC or the pair of ccrA and ccrB) encoding recombinases that mediate the integration and excision of SCCmec into and from the chromosome have been grouped into six different classes (A, B, C1, C2, D, and E) as part of the mec gene complex, which is made up of *mecA*, the regulatory (Saber et al., 2017; Zong et al., 2011). A small number of additional genes, including insertion sequences, transposons, and plasmids, are also present in SCCmec (Zong et al., 2011). The *mecA* gene is typically a component of the broader, distinctive SCCmec genetic element, as was already mentioned. Additionally, these islands may contain additional genes for antimicrobial resistance, insertion sequences, and genes with unknown functions, according to research by Katayama et al. The invertase/resolvase family's two recombinases, ccrA and ccrB are found in the four SCCmecs and are in charge of the site-specific integration and excision from the chromosome at attBscc, a region of an open reading frame close to the origin of replication. Uncertain genetic pathways underlie the movement of these substantial mobile components.(Ito et al., 1999; Katayama et al., 2000).

According to Turlej et al., the first SCCmec element was discovered in 1999, and two more SCCmec elements were discovered after that (Turlej et al., 2011). The *mecA* gene, which codes for PBP2a, a low-affinity penicillin-binding protein, is linked to staphylococci's resistance to methicillin and all beta-lactam antibiotics. PBP2a is lacking in susceptible staphylococci. In other words, the *mecA* gene causes staphylococci to become resistant to drugs similar to penicillin (Petinaki et al., 2001). The sole carrier described for the *mecA* gene, which codes for methicillin resistance in staphylococci, up

to this point has been SCCmec, according to Hanssen and Sollid in 2006 (Hanssen & Ericson Sollid, 2006). The International Working Group on the Staphylococcal Cassette Chromosome Elements states that SCCmec types are identified by Roman numerals followed by the ccr and mec gene complex; type I (1B) designates a SCCmec that possesses a type 1 ccr and a class B mec gene complex. Other recognized designated SCCmec types are type II (2A), type III (3A), type IV (2B), type V (5C2), type VI (4B), type VII (5C1), and type VIII (4A) (Ito et al., 2009). SCC elements, which lack the *mecA* gene but possess features like the capsule gene cluster, fusidic acid resistance, or the mercury-resistance operon, have also been found in staphylococci. Staphylococci have been divided into eleven SCCmec types (I-XI) and subtypes (Ito et al., 2009; Mongkolrattanothai et al., 2004). SCCmec types I through VI were identified molecularly by Deurenberg et al. The findings of their research indicate that only categories I (34.3 kb), IV (20.9 to 24.3 kb), V (28 kb), and VI (20.9 Kb) encode for resistance to beta-lactam antibiotics.SCCmec types II (53.0 kb) and III (66.9 kb) have the ability to resist multiple drugs because they also contain a transposon (Tn554) and additional drug resistance genes on integrated plasmids like pUB110, pI258, and pT181; Kanamycin, tobramycin, and bleomycin resistance are encoded by plasmid pUB110; pI258 encodes penicillin and heavy metals resistance; pT181 encodes tetracycline resistance; and inducible macrolide, lincosamide, and streptogramin (MLS) resistance is encoded by transposon Tn554 (carrying the ermA gene) (R. H. Deurenberg et al., 2007; Saber et al., 2017).

The SCCmec of *S. aureus* strain BA01611, which showed 62.6%-69.4% sequence similarity to all published ccrC1 sequences, had a novel ccr gene, ccrC2, according to Wu et al.'s research from September 2015. In staphylococcal isolates from the USA, France, Germany, and China, the ccrC2 gene was discovered to be primarily present in CoNS (Wu et al., 2015). Wu et al. classified the SCCmec of BA01611 as a

unique type defined as type XII, while Wu et al. classified the ccr gene complex under type 9. (9C2). The pseudo-SCC element 22 (ΨSCCBA01611), which carries a truncated ccrA1, covered this unique SCCmec member in BA01611. Using the 24 extra-chromosomal circular intermediates as a basis, the SCC elements and a composite SCC were removed from the chromosome. These researchers suggested revising the SCCmec typing method to type the ccrC2 gene and nine ccr gene complexes (Wu et al., 2015).

Methicillin resistance can manifest itself in various ways, and each MRSA strain has a unique profile of the percentage of bacterial cells that proliferate in response to different methicillin concentrations (Tomasz et al., 1991). Some MRSA strains have regulatory genes for *blaZ* homologues that influence the expression of resistance. Similar to how *blaZ* is regulated by the genes blaR1 and blaI in response to penicillin exposure, *mecA* is regulated by these genes, mecI, and mecR1, in response to beta-lactam antibiotics. In order to stop gene activation, the repressor genes really bind to similar DNA regions (Archer & Bosilevac, 2001). *MecA* expression is induced from this leaky alternative system due to the sequence similarity between mecI-mecR1 and the blaR1-blaI regulatory genes. Instead of a variable expression, constitutive production of mec is produced by deletions or mutations in mecI or the promoter region of *mecA* (Niemeyer et al., 1996). Rosato et al. recently discovered that every MRSA must have functional mecI or blaI, and they hypothesize that this may be a defense mechanism to prevent the overproduction of a harmful protein. The fem genes, a different set of genes necessary for methicillin resistance, function in the cross-linking of peptidoglycan strands and contribute to the heterogeneity of methicillin resistance expression (Berger-Bächi, 1994).

One of the first antibiotics, vancomycin, has been used in healthcare for almost 60 years. In the dense rainforests of Borneo, Dr. Kornield, an organic chemist for Eli Lilly, discovered vancomycin in 1957 (Rubinstein & Keynan, 2014). Gram-positive bacteria, such as Clostridium, Listeria, Corynebacterium, Enterococci, Streptococci, and

Staphylococci, are effectively treated with vancomycin. Currently, the two conditions that vancomycin is most frequently used to treat are MRSA infections and people with allergies to semi-synthetic penicillin or cephalosporins (Rubinstein & Keynan, 2014).

In order to specifically connect to the bacterial cell wall, vancomycin must first bind to peptidoglycan precursor short peptides that are terminated with D-alanyl-D-alanine, as was revealed to be the leading cause of vancomycin resistance. The elongation and cross-linking of the peptidoglycans in the bacterial cell wall are prevented by this interaction, which represses the production of new cell walls and ultimately causes bacterial death (Haseeb et al., 2019; Micek, 2007). In addition to inhibiting transpeptidase, vancomycin also inhibits transglycosylase (glycosyltransferase), the second enzyme in charge of cross-linking sugar residues. However, this mechanism seems to be less significant than that of transpeptidase inhibition. Uncertainty surrounds the mechanism of transglycosylase inhibition. Beta-lactam antibiotics attain a similar result, although they attach to different sites and have distinct effects on different molecular targets (J. G. Bartlett et al., 2010). The terminal D-alanyl-D-alanine (d-Ala-d-Ala) moieties of the precursor lipid II can form hydrogen bond interactions with the hydrophilic vancomycin molecule. Vancomycin binding causes a conformational change that inhibits the precursor from being incorporated into the developing peptidoglycan chain and transpeptidation, resulting in bacterial lysis and cell wall disintegration (Q. Hu et al., 2016).

Prior to the appearance of vancomycin-resistant staphylococci, the use of vancomycin to treat infections brought on by methicillin-resistant staphylococci, Clostridium difficile, and enterococcal infections increased dramatically (Kirst et al., 1998). Given that both HA-MRSA and CA-MRSA can result in dangerous, invasive infections, including pneumonia and sepsis, vancomycin has long been regarded as the best medication for treating severe MRSA infections (Holmes et al., 2015). The medical

community has taken note of the first report of vancomycin intermediate-resistant *S. aureus* (VISA), which was discovered in 1997 and originated in Japan (Baseri et al., 2018; Hiramatsu, Hanaki, et al., 1997). Additional instances were later reported from various nations, including the United States and China (Howden et al., 2010). Vancomycin-resistant staphylococci were first identified in a clinical isolate of Staphylococcus haemolyticus (Schwalbe et al., 1987). As the final line of defense against gram-positive cocci infection, vancomycin has a long history of use. The medical society is enormously concerned about the fact that *Staphylococcus aureus* is becoming more resistant to vancomycin regularly (Haseeb et al., 2019; Micek, 2007). Vancomycin-resistant *Staphylococcus aureus* (VRSA), vancomycin-intermediate *Staphylococcus aureus* (VISA), and heterologous vancomycin resistant *Staphylococcus aureus*(hetero-VRSA) are now the three categories most commonly used by researchers to categorize vancomycin-resistant *Staphylococcus aureus* (Amberpet et al., 2019). The term "VRSA" refers to *Staphylococcus aureus* with a minimum inhibitory concentration (MIC) of <32 mg/L for vancomycin in clinically isolated strains. It was initially noted in the United States in 2002. The first strain of *Staphylococcus aureus*, designated VISA, was discovered in Japan in 1997; its MIC range for vancomycin is 8–16 mg/L. The primary *Staphylococcus aureus* culture recovered from clinical specimens is hetero-VRSA. The MIC of vancomycin is ≤ 4 mg/L, and VRSA can be identified using either the MH micro broth dilution method or the agar dilution method (Baseri et al., 2018; Severin et al., 2004). The phrase "heteroresistant staphylococci" further contributes to misunderstandings regarding vancomycin resistance in staphylococci. The diversity of vancomycin susceptibilities within subpopulations of a single isolate is known as this phenomenon, which is observed in coagulase-negative staphylococci and *S. aureus* (Srinivasan et al., 2002). Two cell populations comprise a heteroresistant isolate: the bulk cells are vancomycin-susceptible, while the minority are resistant. Given that it was

detected in as many as 20% of *S. aureus* samples in one Japanese hospital, heteroresistance is probably more prevalent than pure resistance or reduced susceptibility (Hiramatsu, Aritaka, et al., 1997).

Van gene clusters are present in pathogens, glycopeptide-producing actinomycetes, anaerobic bacteria of the human gut flora, and the biopesticide Paenibacillus popilliae are responsible for vancomycin resistance in bacteria (Ammam et al., 2012; Hong et al., 2008; Xu et al., 2010). The DNA sequence of the ligase van gene homologues encodes the essential enzyme for the synthesis of either d-alanyl-d-lactate (d-Ala-d-Lac) or d-alanyl-d-serine, allows for the division of vancomycin resistance into various gene clusters (d-Ala–d-Ser). Currently, at least 11 van gene clusters are known to correspond to the phenotypes VanA, VanB, VanD, Van F, VanI, VanM, VanC, VanE, VanG, VanL, and VanN (Kruse et al., 2014; Lebreton et al., 2011; Xu et al., 2010). VanA, vanB, vanD, van F, vanI, and vanM are examples of genes encoding d-Ala:d-Lac ligases that frequently cause high levels of vancomycin resistance with MICs > 256 mg/ml, while genes encoding d-Ala:d-Ser ligases, such as vanC, vanE, vanG, vanL, and vanN, typically cause low levels of resistance with MICs of 8–16 mg/ml (Hollenbeck & Rice, 2012).

The principal reservoir of acquired vancomycin resistance, the Enterococcus species, has shown the vancomycin-resistant mechanism. Despite discovering 11 van gene clusters that give vancomycin resistance, the isolated VRSA strains are solely caused by the vanA gene cluster (Werner et al., 2008). VanS, VanR, VanH, VanA, and VanX are five proteins encoded by the vanA gene cluster, which are critical for vancomycin resistance (Arthur & Courvalin, 1993). A transposon called Tn1546 carries the original vanA gene cluster. In the presence of vancomycin, a two-component system made up of VanS, and VanR upregulates the expression of the cluster genes. VanH, VanA, and VanX change the natural D-Ala-D-Ala precursors into the robust D-Ala-D-Lac.

VanH acts as a dehydrogenase to decrease pyruvate and creates d-Lac (Cong et al., 2020a). In order to stop the native d-Ala-d-Ala from being utilized in the manufacture of peptidoglycan, VanX functions as a d,d-dipeptidase and hydrolyzes it. In order to create the resistant D-Ala-D-Lac that replaces d-Ala-d-Ala in the manufacture of peptidoglycan, VanA ligates D-Lac to D-Ala (Cong et al., 2020a). Vancomycin's action target is the precursor lipid II's terminal d-Ala-d-Ala moieties, where it establishes hydrogen bond interactions to stop future transglycosylation and transpeptidation. However, the changed d-Ala-d-Lac causes a decrease in vancomycin affinity of about 1000-fold. As a result, vancomycin is no longer bactericidal against bacteria that have changed peptidoglycan precursors (Arias & Murray, 2012; Werner et al., 2008). These Van components are interesting targets for creating novel medications because deletion of any of them results in the recovery of vancomycin action. Examples of transition-state analogues that have been shown to be VanA inhibitors include hydroxyethylamines, phosphinate, and phosphonate (Sova et al., 2009; C. T. Walsh et al., 1996). Covalent inhibitors, phosphonate-based, and sulfur substances have all been investigated as VanX inhibitors. To stop vancomycin from losing its ability to bind to its target, these inhibitors can be administered in conjunction with vancomycin (A. Y. Chen et al., 2019).

Daptomycin (Dap) is a cyclic peptide antibiotic with a side chain made of decanoyl fatty acids. In addition to being approved to treat *S. aureus* bacteremia and endocarditis, it is effective against a wide range of Gram-positive bacteria. The mainstay of modern anti-MRSA treatment is daptomycin (Arbeit et al., 2004). Daptomycin, according to Gomez Casanova et al. in 2017, has a distinct mechanism of action that prevents it from developing cross-resistance with other antibiotics. It can be used to treat MRSA-caused bloodstream infections and skin soft tissue infections, but not MRSA-induced pneumonia because alveolar surfactant can inhibit its activity (Casanova, Natalia Gómez, María, Siller Ruiz, 2017).

The medication daptomycin is a cyclized lipopeptide taken from the fermentation broth of Streptomyces roseosporus. Daptomycin does not block lipoteichoic acid due to its action method, which destroys the plasma membranes' electric potential when calcium ions are present (Heidary et al., 2018; S. D. Taylor & Palmer, 2016). Anionic molecule is the native form. The drug's antibacterial effectiveness depends solely on calcium, creating a compound called Ca-DAP (Bayer et al., 2013). The Ca-Dap complex interacts with the cell and exhibits cationic peptide behavior. When Ca-Dap binds to the negatively charged phosphatidylglycerol (PG) head groups, it first oligomerises to produce micelles that enter cells and insert into the cytoplasmic membrane. This causes tension in the lipid bilayer, which ultimately leads to cell death by causing depolarization, permeabilization, and ion leakage, particularly K^+ (Foster, 2017).

Mutations in the genes turn on the bacterium's defenses against threats to the cell envelope, such as host cationic antimicrobial peptides, resulting in Daptomycin resistance (Dapr) (Bayer et al., 2013). Infections with a high bacterial density, such as endocarditis, are treated for a long time before resistance develops. Due to overlapping resistance mechanisms, the patient's previous vancomycin treatment is linked to a faster emergence of Dapr during therapy (Foster, 2017). In order for the bacteria to resist daptomycin, they must either stop the drug from getting to the cytoplasmic membrane or obstruct its entry. The MIC can pass the susceptibility breakpoint when several mutations occur in various genes (Foster, 2017). The multiple peptide resistance factor (mrpF) gene typically undergoes the first mutational changes when susceptible cells are exposed to the drug for an extended period of time and at increasing concentrations during in vitro passage (Bayer et al., 2013). An integral membrane protein called MrpF transforms PG (phosphatidylglycerol) into lysyl-phosphatidylglycerol by adding a positively charged lysine residue (L-PG) (Ernst & Peschel, 2011). This is produced in the cytosol and moved to the membrane's exterior after being synthesized. Hotspots are where the mrpF

mutations connected to Dapr are found. The level of L-PG in the membrane rises due to the protein undergoing gain-of-function changes. As a result, less negatively charged PG is required for drug binding to cause membrane damage, repels the Ca-Dap and increases the charge of the membrane's outer face (Foster, 2017). Cardiolipin may act as a defense mechanism against Dap. The ratio of PG to cardiolipin may change when cardiolipin synthase mutations in Dapr are present. It has also been noted that alterations in membrane fluidity and increased rigidity brought on by higher staphyloxanthin pigment levels can decrease Ca-Dap binding (Bayer et al., 2013).

Invasive infections frequently lead to the emergence of *S. aureus* mutants with defects in the global regulator Agr. Daptomycin resistance in Agr mutants results from the release of phospholipids that bind and neutralize the antibiotic before it reaches its intended target in the membrane (Pader et al., 2016). Organisms with the Agr gene release much less lipid. Furthermore, the interaction between phospholipid and daptomycin is blocked by phenol-soluble modulins that are only secreted by Agr-positive organisms (Foster, 2017). Beta-lactam antibiotics paradoxically make MRSA strains that have developed daptomycin resistance susceptible. Daptomycin and beta-lactam combinations work together to combat MRSA and have been used to treat MRSA-related persistent infections with good results (Dhand & Sakoulas, 2014; Sakoulas et al., 2014). The mrpF gain-of-function mutations increase the level of L-PG in the membrane at the expense of the substrate for anchoring the lipoprotein chaperone PrsA on the outer face of the cytoplasmic membrane are associated with the molecular basis of beta-lactam sensitivity in Dapr MRSA (Foster, 2017). PBP2a must be localized appropriately and stable, and PrsA is required for this. Cells' sensitivity to -lactams that target PBP2a is explained by the absence of PBP2a despite normal levels of its mRNA. In comparison to antibiotics that target PBP2, PBP3, or PB4, -lactams like imipenem that target PBP1 have a more

pronounced synergistic effect. When exposed to the mixture, the pbpA gene, which codes for PBP1, increased transcription, accelerating bactericidal activity (Berti et al., 2016).

The tetracyclines (Tet) were first reported in the scientific literature in 1948. They were found to be naturally occurring products from soil bacteria called actinomycetes. From the late 1940s to the early 1950s, they were commercialized with clinical success and were known for their broad-spectrum antibacterial activity. The development of the tetracycline scaffold toward derivatives with increased potency and efficacy against tetracycline-resistant bacteria and improved pharmacokinetic and chemical properties can be seen in the second-generation semisynthetic analogs and more recent third-generation compounds (Nelson & Levy, 2011). For many years, tetracyclines have been used extensively to treat various infections. In 2005, a third-generation semi-synthetic derivative of tigecycline was authorized for the treatment of infections brought on by pathogens that are multidrug-resistant, such as MRSA. Tigecycline is much more effective than tetracycline and can still work against bacteria that have developed a resistance to it (Foster, 2017).

Protein synthesis is an essential requirement for all cells. It uses ribosomes, which are responsible for translating an mRNA code into functional proteins. This occurs on eukaryotes' ribosomes containing the 40S and 60S subunits. Protein synthesis occurs in prokaryotes, such as bacteria, using ribosomes with the 30S and 50S subunits. The ribosome transfer RNA (tRNA), which is charged with an amino acid, binds to the mRNA template at these locations (Shutter & Akhondi, 2022a). Cellular proteins are formed and extended due to the subsequent binding of each tRNA that has been charged with an amino acid. The 30S ribosomal subunit is inhibited explicitly by tetracyclines, which prevents the aminoacyl-tRNA from binding to the acceptor site on the mRNA-ribosome complex. A cell can no longer maintain proper functioning and will not be able to expand or continue to replicate if this process stops. Tetracyclines are referred to as

"bacteriostatic" because of the way in which they impair(Shutter & Akhondi, 2022a). Nguyen et al. also reported in 2014; Tetracyclines bind to the 30S subunit near the location where the incoming aminoacyl (aa) tRNA's anticodon recognizes the codon in the mRNA, the 16S rRNA's six different residues are connected by a complex that forms when Tet and Mg2+ interact. It dissociates after the aa-stable tRNA's binding is broken (Nguyen et al., 2014; D. N. Wilson, 2009).

Two related Tet efflux pumps called TetA(K), and TetA(L) with 14 transmembrane helices have been identified in staphylococci. The chromosomal SCCmecIII cassette of MRSA strains contains the TetK gene, encoded by the small multicopy plasmid pT181 (Jensen & Lyon, 2009). They both belong to the major facilitator superfamily (MFS) of transporters, which have 14 transmembrane domains in contrast to the 12 transmembrane domains found in most MFS transporters, such as the TetA protein of Gram-negative bacteria (Chopra & Roberts, 2001). The Tet efflux proteins work against a concentration gradient to swap a proton for a tetracycline molecule (Piddock, 2006b). Using the structures of the YajR or LacY transporters from Escherichia coli, molecular modeling has been used to determine the molecular basis of efflux (Nguyen et al., 2014). Bacteria expressing TetK or TetL confer little to no resistance due to the bulky substitutions on ring D of tigecycline, preventing the molecule from accessing the drug binding site in the efflux protein (Chopra & Roberts, 2001).

Tn916 and Tn1545 are conjugative transposons with TetO/M determinants typically located on chromosomes (Jensen & Lyon, 2009). The resistance gene encodes a GTPase-active protein with substantial similarities to EF-G(Dönhöfer et al., 2012) . The TetO/M GTPase, however, is ineffective as an elongation factor. Tet is released from the A site by the TetO/M protein, which binds to the EF-G binding site on the ribosome in the post-translocation state (Connell et al., 2003). Translation can go on despite Tet doses that would typically be inhibitory because of the remarkable effectiveness of this

process. The TetO/M determinant does not significantly impact the effectiveness of tigecycline. The TetO/M GTPase, however, is ineffective as an elongation factor. Tet is released from the A site by the TetO/M protein, which binds to the EF-G binding site on the ribosome in the post-translocation state (Foster, 2017).

The oxazolidinones are a family of antibacterial substances with significant action against Gram-positive bacteria. They were initially found in the late 1980s, but research was stopped due to hepatotoxicity safety concerns (Johnson et al., 2000). In contrast, interest in oxazolidinones resurfaced in the 1990s when glycopeptide resistance growth became a significant concern for the hospital infection community, particularly in methicillin-resistant *Staphylococcus aureus* (MRSA). The US Food and Drug Administration consequently approved and dispersed linezolid as the first oxazolidinone in April 2000. In 2014 and 2015, the European Medicines Agency and the Food and Drug Administration authorized tedizolid, the second oxazolidinone, respectively, after a ten-year postponement (European Medicines Agency (EMA), 2021).

According to Roger et al. in 2017, oxazolidinones function as a protein synthesis inhibitor on the bacterial ribosomal 50S subunit, focusing on the start step of protein synthesis. Functional initiation and messenger RNA translation are then blocked as a result. Other protein synthesis inhibitors, such as macrolides, lincosamides, tetracyclines, chloramphenicol, and tetracyclines, do not have the same mode of action as this drug. Due to this unique mode of action, antimicrobials like lincosamides and macrolides are no longer effective against infections that carry ribosome-related genes.(Roger et al., 2017). Linezolid's activity is unaffected by the rRNA methylases that alter the 23S rRNA to impede the binding of macrolides, clindamycin, and group B streptogramins because the target that linezolid possesses does not overlap with that of current protein synthesis inhibitors (Roberts, 2008). Perhaps due to this method of action, linezolid appears to be particularly efficient in blocking the synthesis of staphylococcal and streptococcal

virulence factors (Bernardo et al., 2004). It is intriguing to note that oxazolidinones only bind to the mitochondrial 70S ribosome and not the cytoplasmic 80S ribosome, which accounts for the myelosuppression and toxic optic neuropathy seen in individuals receiving extended treatment with linezolid(Leach et al., 2007; Nagiec et al., 2005).

Linezolid has several advantages over frequently used antibiotics, one of which is that it is entirely synthetic. As a result, it appears to lack a natural model, and given its characteristics, it was thought that there would not be a natural reservoir of resistance genes that would promote the formation of clinical resistance. It is widely accepted that all other protein synthesis inhibitors come from naturally occurring antibiotics of microbial origin, whose producers serve as the natural repository of resistance genes that may be passed to clinical infections through horizontal gene transfer (Toh et al., 2007). Until recently, this advantage was preserved; the only mechanism of resistance revealed was brought on by modifications to the drug's target site, namely the rRNA of the essential ribosomal subunit. This kind of resistance, as detailed below, is rare, develops slowly, and cannot be transmitted between pathogenic species since bacterial rRNA genes are redundant. This resistance appears to have evolved de novo through spontaneous mutation rather than genetic exchange (Stefani et al., 2010). In line with Stefani et al., particularly concerning is the recent discovery of a linezolid resistance mechanism based on obtaining a naturally occurring, possibly transferable resistance gene that alters a particular rRNA nucleotide near the drug's site of action. In the future, it may fundamentally alter the image of linezolid susceptibility. Due to its association with mobile genetic components, this gene increases the risk of transmission to other pathogenic strains and intra-species organisms (Stefani et al., 2010).

According to Franceschi et al., more isolates with mutations in the 23S rRNA gene's domain V and the genes encoding the ribosomal proteins L4 and L22 have been discovered over the past ten years that are resistant to the antibiotics macrolide,

lincosamide, streptogramin, ketolide, and oxazolidinone (MLSKO) (Franceschi et al., 2004).

Most bacteria resistant to telithromycin (ketolide) and linezolid have a mutation in one of these three genes. These mutational changes, which impact how the 23S rRNA and proteins operate and somewhat lessen a bacterium's susceptibility to one or more of the MLSKO antibiotics, have been described in both Gram-positive and Gram-negative bacteria (Roberts, 2008).

Resistance has been linked to the domain V region's core loop changes. The presence of numerous copies of the 23S rRNA gene in almost all bacteria was once assumed to reduce the likelihood that these bacteria would acquire resistance to these drugs (Prystowsky et al., 2001).

Microorganisms, including *S. aureus* and CoNS, exhibit linezolid resistance due to the G2576T transversion (Hill et al., 2010). The first clinical strain of *S. aureus* resistant to the antibiotic linezolid was described by Tsiodras et al. in 2001. This isolate had a G2576T mutation in the 23S rRNA gene's domain V region (Tsiodras et al., 2001). Further investigation revealed that this isolate had five copies of this gene, each of which had the G2576T mutation (Pillai et al., 2002).

Linezolid resistance has been demonstrated to be influenced by the number of rRNA genes that become altered, which depends on the length of linezolid exposure and its dosage (Arias et al., 2008). A series of MRSA isolates that were progressively more resistant to linezolid and carried progressively more mutant (G2576T) copies of the 23S rRNA gene were reported by Pillai et al. In this investigation, it was shown that the number of copies of mutations in the 23S rRNA genes rose in direct proportion to the number of linezolid-resistant *S. aureus* isolates (Pillai et al., 2002). Clinical isolates of linezolid-resistant, enterococci and laboratory-derived oxazolidinone-resistant *S. aureus*

mutants have demonstrated similar effects of mutant-gene dosage (Marshall et al., 2002; Meka et al., 2004).

After being exposed to linezolid for several months, Meka et al. examined successive clinical *S. aureus* isolates that developed resistance to the medication. They discovered the T2500A mutation for the first time in the 23S rRNA gene's domain V region and the deletion of one copy of this gene in the more resistant isolates (Arias et al., 2008; Meka et al., 2004). Although there is evidence that some 23S rRNA mutations have fitness costs, extremely linezolid-resistant 23S rRNA homozygous mutant strains of *S. aureus*, S. epidermidis, and E. faecalis have been recovered clinically (Besier et al., 2008). Even though C2192T, G2447T, A2503G, T2504C, G2505A, G2766T, and G2576T have all been reported as 23S rRNA variants that confer resistance to linezolid, the most frequent alterations detected in clinical isolates to far are G2576T and T2500A (Howe et al., 2003; Livermore et al., 2007, 2009; Tsiodras et al., 2001).

Mutations in the ribosomal proteins L3, which the rplC gene encodes, and L4, which the rplD gene encodes, represent a less frequent mechanism of linezolid resistance. Recently, linezolid resistance in staphylococci with a clinical etiology was linked to these two previously discovered mechanisms in streptococci (Locke et al., 2009). Even though the L22 ribosomal protein expressed by the rplV gene is positioned distant from the Lin binding site, it has already been noted that it is in charge of quinupristin/dalfopristin resistance in S. pneumoniae and *S. aureus* (Malbruny et al., 2002). Due to the availability of "unknown" resistance mechanisms mentioned in the literature, this target, along with many other ribosomal proteins, cannot be ruled out a priori and requires more exploration (R. N. Jones et al., 2007b, 2007a).

A novel phenicol and clindamycin resistance phenotype has recently been produced by an RNA methyltransferase known as Cfr. A comprehensive examination using drug footprinting studies and matrix-assisted laser desorption-ionization time of

flight/tandem mass spectrometry revealed that Cfr inserts an extra methyl group at position A2503 of 23S rRNA. Because A2503 sits near the overlapping ribosome binding sites of phenicols and clindamycin, it was inferred that Cfr-mediated methylation imparts resistance to these two types of antimicrobial medicines by interfering with drug placement (Kehrenberg et al., 2005). This gene gave resistance to phenicols, lincosamides, oxazolidinones, pleuromutilins, and streptogramin$_A$ but not macrolides, and hence differentiates from erm rRNA methylase genes in which methylation occurs at position A2058 (Roberts, 2008; Smith & Mankin, 2008).

Surveillance research for florfenicol resistance among staphylococci from animals led to the initial discovery of the cfr gene in 2000. Bovine strains of Staphylococcus simulans and Staphylococcus sciuri have been discovered to have the 16.5-kb multiresistance plasmid that was first used to determine it (Kehrenberg et al., 2004; Schwarz et al., 2000). The chloramphenicol/florfenicol exporter gene fexA and the cfr gene were recently discovered on the 35.7-kb pSCFS3 from a porcine Staphylococcus plasmid aureus strain (Kehrenberg & Schwarz, 2004). The Cfr gene was cloned and expressed in Escherichia coli, revealing that Cfr provided resistance to Gram-positive and Gram-negative bacteria in addition to the initial Gram-positive hosts(K. S. Long et al., 2006). The Cfr protein was found to be a member of the Radical SAM (S-adenosylmethionine) superfamily, which includes a wide range of enzymes from a diverse set of bacteria involved in radical protein formation, isomerization, sulfur insertion, anaerobic oxidation, and unusual methylations. Comparison with other protein sequences deposited in databases revealed that the Cfr protein is unrelated to other known resistance-conferring rRNAs (Kehrenberg et al., 2005).

The cfr gene was discovered in Staphylococcus spp. of animal origin in Europe, as was previously mentioned. Recently, Staphylococcus isolated from human strains was also discovered to contain the cfr gene (Mendes et al., 2008; Schwarz et al., 2000). Two

strains of Staphylococcus resistant to linezolid were discovered during the 2007 LEADER program: *S. aureus* (004-737X) and S. epidermidis (426-3147L). Mendes et al. described the structure of the cfr gene, which was discovered in both samples. The *S. aureus* isolation had ΔtnpB downstream of the cfr gene, and its structure matched that of the pSCFS3 plasmid discovered in an *S. aureus* isolate isolated from a pig's respiratory system.(Mendes et al., 2008). The istAS and istBS genes are present in the DNA sequence in the *S. aureus* isolate upstream of the cfr gene, and their resemblance to those of the pSCFS3 plasmid suggests that these insertion sequences may be involved in the mobilization of the cfr gene (Kehrenberg et al., 2007). The tnpA gene, which was farther upstream of the cfr gene on the pSCFS3 plasmid, however, produced a negative result, indicating that the upstream region of cfr on this isolate considerably varied from that of the pSCFS3 plasmid. Only the cfr gene was discovered in this strain of Staph epidermidis (Mendes et al., 2008).

Recent research in Germany revealed the presence of the cfr in the MRSA ST398 and ST9 lineages, which have swine as their primary reservoir but may colonize and infect people (Kehrenberg et al., 2009). The discovery of cfr in an *S. aureus* strain isolated from a clinical human isolate (labeled CM-05) from Colombia was revealed in another report. In contrast to the animal isolates, the MRSA CM-05 isolate was described, and it was discovered that the gene was situated in the chromosome. However, the gene was likely a component of an integrated plasmid that may have been capable of excision and mobilization (Toh et al., 2007). The cfr gene and the erm(B) gene were located on the chromosome in MRSA CM-05, establishing a transcriptional unit known as the mlr operon regulated by the ermB promoter. Both genes are expressed constitutively despite putative regulation of short open reading frames. In order to make the MRSA isolate resistant to all antibiotics targeting the large ribosomal subunit, the methyltransferases expressed by the mlr operon worked together to modify two particular residues in 23S

rRNA, A2058 and A2053 (Arias et al., 2008; Toh et al., 2007). The failure of the plasmid from the CM05 isolate to induce linezolid resistance on recipient *S. aureus* cells lends credence to this view. On one side, the ermB and cfr cluster is flanked by the transposase and integrase gene istAS from the IS21-558 mobile genetic element. Using PCR analysis, the complete IS21-558 was found. The mobility of the cfr gene in animal isolates was linked to the IS21-558 element, which may have assisted the clinical strain in mobilizing the gene. Two specific residues in 23S rRNA, A2058 and A2053, were modified by the methyltransferases encoded by the mlr operon in order to confer resistance on the MRSA strain to all antibiotics that target the large ribosomal subunit (Kehrenberg et al., 2007). A 5' section of the gene repS can be found upstream of ermB. The RepS protein, which is one of its byproducts, helps plasmid replication get started. Integration of a plasmid containing the cfr gene into the chromosome of CM05 cells was the most plausible method by which the MRSA human strain acquired linezolid resistance due to the intimate linkage of the cfr gene with a distinctive plasmid gene (Toh et al., 2007). The discovery of the cfr gene in this strain, which was obtained from a patient who had just recently been exposed to linezolid, suggests that this microorganism acquired the gene under selection pressure that did not include exposure to oxazolidinones. A further hypothesis is that the strain was chosen in a patient who was not identifiable and was exposed to linezolid before being transferred to the case patient (Arias et al., 2008; Toh et al., 2007).

Due to the rise of MRSA and MDR-resistant Gram-positive cocci, oxazolidinone research is particularly active. The search for novel oxazolidinones with enhanced potency, water solubility, and decreased toxicity is an urgent medical priority. The A- B-C-rings of linezolid have reportedly undergone changes in these attempts. The potential therapeutic candidates among all of these compounds included several that were created through C-ring alterations (Vara Prasad, 2007).

Decolonization of *S. aureus*

Staphylococcus aureus is present in several reservoirs, but the anterior nasal passages are a primary anatomical source of colonization. A 20% to 55% rate of asymptomatic *S. aureus* nasal carriage in healthy persons has been documented, and an increasing number of these invaders exhibit antibiotic resistance (Street et al., 2009). Studies prove that the nose is the main route through which non-infected carriers transmit pathogens to both themselves and other people (von Eiff et al., 2001). In actuality, most MRSA infections are caused by endogenous sources, and nasal carriage has been shown to increase the risk of infection almost four times (von Eiff et al., 2001). As a way to lower transmission and the overall infection rate, decolonization treatment for *S. aureus* carriers has been studied.

Two broad strategies for preventing healthcare-associated infections (HAI) have arisen since colonization frequently results in infection: 1) broad reductions in the burden of all pathogens through horizontal tactics, and 2) reductions in particular pathogen colonization or infection through vertical strategies(Wenzel & Edmond, 2010). Vertical strategies target a specific pathogen and frequently use active surveillance testing. This is significant because there are many more asymptomatic patients than infected patients with multidrug-resistant organisms (MDROs), such as VRE, multidrug-resistant Gram-negative organisms, MRSA, and C. difficile. These MDROs are similar in that colonization occurs before infection, and transmission occurs through direct or indirect contact. Additionally, patients who have colonized but are unaware of it might transmit it (E. Septimus et al., 2014). All clinically significant healthcare-associated microorganisms, including Gram-negative bacteria, enterococci, Candida, and *S. aureus*, are susceptible to horizontal decolonization techniques.

Multiple pathogens may be eliminated from a colonized patient using horizontal decolonization techniques. Due to several risk variables for colonization being shared by

many MDROs, multi-bacterial colonization is widespread (Harris et al., 2004). In order to stop infection and transmission, decolonization techniques work to reduce the bacterial load. These tactics frequently involve vertical screening of patients for particular infections (like MRSA or VRE) and decolonizing them if such germs are shown to be present. Endogenous and exogenous infections could be avoided in this way (E. J. Septimus & Schweizer, 2016).

Decolonization aims to lessen or eliminate the bacterial load on the body. High bacterial load carriers have a higher risk of illness and are more likely to spread the germs to their surroundings (Kalmeijer et al., 2000). The amount of *S. aureus* that persistent *S. aureus* carriers carry in their nostrils is higher than that of intermittent carriers, as determined by measuring in $\log_{10}$ CFU per nares culture (J. L. Nouwen et al., 2004). Between 1.8 and 2.9 $\log_{10}$ CFU per nares culture, the average *S. aureus* bacterial load among nasal carriers is typical (Mermel et al., 2010). One investigation discovered a correlation between higher log counts of MRSA in the nose, higher log counts in other body locations, and a higher chance of colonization at other body sites. The research revealed that the mean extra nasal MRSA loads range was 0.87 log10 CFU per culture in the axilla to 1.65 log10 CFU per culture in the perineum to 1.70 log10 CFU per culture per culture culture in the groin (Mermel et al., 2011). Some decolonizing agents assert that they eradicate the bacterial load from the areas where they are applied, while others assert that they reduce the burden. However, there is limited data on the minimum bacterial load that must be decreased to stop transmission and infestations (E. J. Septimus & Schweizer, 2016).

The most successful patient groups for decolonization are those with short-term infection risks. These populations include those who have undergone surgery, who may have a decreased risk of infection following surgical wound healing and closure and those who have undergone intensive care unit treatment, who have a much lower risk following

ICU release. Due to worries about both recolonization and resistance to agents that colonize, this window of time is crucial. Decolonization can therefore have a short-term positive impact on patient groups at risk for brief periods (B. Y. Lee et al., 2010). After being decolonized, studies have shown that patients frequently get *S. aureus* again weeks or months later.(Holton et al., 1991; Immerman et al., 2012). *S. aureus* recolonization rates for healthcare professionals after one year were close to 50%, whereas 75% for peritoneal dialysis patients (M. B. Loeb et al., 2003). Similarly, one research discovered that 56% of hemodialysis patients had *S. aureus* recolonized after four months (Bommer et al., 1995).

According to several clinical investigations, decolonization of the nose and other body locations has been shown to reduce the risk of nosocomial infections. Nasal MRSA decolonization frequently involves applying 2% Mupirocin calcium ointment, a topical antibiotic solution (Street et al., 2009).Various decolonization techniques have been put through testing in an effort to lessen the burden of recurring infections. Most of the introspection has been on *S. aureus* decontamination techniques to manage nosocomial outbreaks and lessen invasive staphylococcal infections in high-risk patients such as those immunosuppressed. Combinations of mupirocin nasal ointment, oral antibiotics, chlorhexidine bath washes, diluted bleach baths, and attention to general hygiene and wound care are methods utilized for ambulatory patients to decolonize *S. aureus* (Fritz et al., 2012).

The most used topical antibacterial medication is nasal mupirocin. Twelve clinical investigations, including 23 clinical trials and a comprehensive review of the literature, assessed antibiotics used topically. The authors concluded that nasal mupirocin administered for a short time was the most successful treatment for MRSA decolonization, with success rates reaching 90% at one week following treatment and around 60% at longer intervals. Mupirocin was equally effective for carriers of MSSA

and MRSA (Ammerlaan et al., 2009). Buehlmann et al. evaluated a decontamination program that included systemic oral antibiotics, body wash/mouthwash with chlorhexidine, and mupirocin nasal therapy. Decolonization is defined as three consecutive sets of negative cultures, and this regimen resulted in an 87% decolonization rate in 62 individuals. Another team conducted a comprehensive evaluation of research that had already been done on the use of intranasal mupirocin in surgical patients and came to the conclusion that it considerably reduced the incidence of postoperative surgical infections (Buehlmann et al., 2008; van Rijen et al., 2008). Over the course of a 5-year trial period, nasal mupirocin and chlorhexidine body wash-based decolonization treatment was also found to lower the infection rate in a critical care unit (Sandri et al., 2006).

Short-term use of nasal mupirocin for postoperative prophylaxis to avoid *S. aureus* SSIs has not been linked to a rise in mupirocin resistance, in contrast to unrestricted usage. Only 6 of the 1,021 isolates tested positive for mupirocin resistance (0.6%) by Perl et al. after administering the drug to more than 2,000 individuals (Perl et al., 2002). Point-prevalence surveys were conducted repeatedly over four years in a different investigation to see if mupirocin resistance had developed in surgical units employing preoperative prophylaxis with five days of nasal mupirocin. They did not discover proof of mupirocin resistance's persistent appearance or expansion. There was no high-level mupirocin resistance (HL-MR) strains found (Fawley et al., 2006). An evaluation of more than 20,000 patients who received mupirocin prophylaxis before major cardiothoracic surgery was conducted in the Netherlands. There was no emergence of mupirocin resistance (van Rijen et al., 2008).

Presently, topical *S. aureus* nasal decontamination with mupirocin is the best alternative. However, mupirocin usage has resulted in mupirocin resistance and treatment failures, particularly when it has been used widely and for a long time. As a result, it is

critical to consider newer medicines or decontamination techniques to eradicate MRSA among patients colonized or infected with mupirocin-resistant strains (Poovelikunnel et al., 2015).

Bacillus subtilis serves as the source for the topical medication bacitracin. It fights MRSA and other Gram-positive bacteria by impeding the formation of bacterial cell walls. In order to treat *S. aureus* nasal colonization in healthcare professionals, Soto et al. conducted a randomized controlled trial (RCT) using a 5-day regimen of either mupirocin or bacitracin. Bacitracin was shown to be inferior to mupirocin for the removal of *S. aureus* after 30 days (23% versus 80%; $P<0.01$) (Soto et al., 1999).

A novel family of antibiotics known as pleuromutilins includes Retapamulin. Retapamulin interacts with the ribosome's 50S subunit to combat Gram-positive and Gram-negative bacteria (Rittenhouse et al., 2006). Due to its potent activity against *S. aureus* and S. pyogenes, which has MIC_{90}s values of 0.12 µg/ml and 0.03 µg/ml, respectively, it is recommended for the treatment of impetigo caused by Streptococcus pyogenes or MSSA. Additionally, it is effective against MRSA and staphylococci resistant to mupirocin (R. N. Jones et al., 2006).

A double-blinded, placebo-controlled RCT of nasal retapamulin was presented at a global conference in 2008, even though the U.S. Food and Drug Administration (FDA) has not yet licensed it for use in the nares. To ascertain if retapamulin nasal treatments given over three and five days may completely get rid of persistent *S. aureus* nasal carriage, this RCT investigated 43 individuals. The term "persistent carriers" describes those who tested positive for *S. aureus* at the three screening visits and just before the first treatment. At seven days, retapamulin resulted in the nasal decolonization of *S. aureus* in 92% to 94% of patients, and at 28 days, it occurred in 75% to 86% of patients. Sneezing, nosebleeds and headaches were the most frequent adverse reactions.

Nasal pain and rhinorrhea occurred in both groups at comparable rates (Naderer, 2008; E. J. Septimus & Schweizer, 2016).

A decontamination strategy that does not rely on the use of antibiotics would undoubtedly be beneficial in terms of bacterial resistance and patient compliance. In vitro research has shown that antimicrobial photodynamic therapy (aPDT) is a successful non-antibiotic antimicrobial method. In this method, a photosensitizer molecule is activated, and after that, it either transfers its energy directly to a substrate by electron abstraction (type I photoreaction) or reacts with molecular oxygen to form singlet oxygen (type II photoreaction)(M. Wilson, 1993)(Hamblin & Hasan, 2004). The idea that the antibacterial effectiveness of aPDT is directly connected to the production of singlet oxygen in the cellular environment and that the levels of singlet oxygen are proportional to the availability of ambient oxygen in the microenvironment is supported by several studies (Luksiene, 2003; Maisch et al., 2007). The effects of aPDT are largely confined since singlet oxygen has a very low half-life and very little ability to migrate away from the point of creation. Additionally, the preferential staining of bacterial cell wall constituents like LPS and peptidoglycan confers a degree of selectivity over eukaryotic host cells in the treatment site (Street et al., 2009). Due to its non-specific bactericidal action and lack of vulnerability to antibiotic resistance, aPDT is a crucial benefit over other antimicrobial therapy modalities. In addition, neither heat nor scarring damage to tissues is caused by the low-level laser light utilized to activate the dye agents. In order to cure bacterial infection or colonization, aPDT is a non-thermal, localized, topical therapy that may be applied to various anatomical sites. This technique has been used clinically on mucosal tissues inside the nasopharynx (Chondros et al., 2009). The current study by Street et al. describes how MRSA can be eradicated entirely by aPDT and how it may be used to treat asymptomatic carriers of the infection by decolonizing them (Street et al., 2009).

The majority of Gram-positive and Gram-negative bacteria, including multi-drug resistant pathogens, are susceptible to the bactericidal effects of alcohol. The most effective alcohol concentrations range from 60 to 90 percent. Alcohols may denature proteins, which makes them antibacterial. Isopropanol or ethanol makes up the majority of alcohol-based hand antiseptics (Boyce & Pittet, 2002). In a recent double-blinded, placebo-controlled RCT, Steed et al. examined how an alcohol-based nasal antiseptic reduced *S. aureus* nasal colonization in previously infected healthcare workers. Three times during the day, nasal alcohol-based antiseptics or placebo were administered to healthcare professionals who tested positive for nasal *S. aureus* colonization. The antiseptic mixture included benzalkonium chloride as a preservative, natural oil emollients, and 70% ethanol. Before and after a 10-hour shift, levels of nasal *S. aureus* and total bacterial colonization were assessed. Antiseptic therapy decreased the number of *S. aureus colony forming units (*CFU) from the starting point by 82% on average and 99% on median (P 0.001) (Steed et al., 2014). To ascertain if nasal ethanol antiseptic decolonization might lessen *S. aureus* infections, a substantially more extensive investigation involving patients with *S. aureus* colonization is required.

Numerous factors warrant the inclusion of universal alcohol-based nasal decolonization in all hospitals' infection control strategies, including the following:

Antimicrobial stewardship is a critical responsibility shared by all healthcare institutions. Multi-drug-resistant bacteria are becoming more prevalent, which puts patients in considerable danger. Alcohol-based nasal antiseptics efficiently lower bacterial prevalence while lowering the chance of developing antimicrobial resistance (E. J. Septimus & Schweizer, 2016).

Evidence suggests that universal decolonization is more successful in halting the spread of MRSA than conventional screening procedures and focused decolonization. Implementing universal eradication considerably lowers

"colonization pressure," which is an essential factor in MRSA transmission and measures the proportion of MRSA carriers in a hospital (E. Septimus et al., 2016). For decades, people all over the world have used chlorhexidine as a topical antiseptic. A cationic biguanide known as chlorhexidine gluconate (CHG) affects the osmotic balance of bacteria by attaching to their cell walls. Yeasts and bacteria of both the Gram-positive and Gram-negative varieties are susceptible to CHG. The safety record of CHG is outstanding. CHG side effects include minor skin irritation and infrequent severe allergic responses (Milstone et al., 2008). The effectiveness of CHG has been established for various reasons, including handwashing, skin preparation for procedures, vaginal antisepsis, dental care for preventing ventilator-associated pneumonia (VAP), gingivitis therapy, and body washes for infection prevention. CHG is available in various formulations and concentrations, ranging from 0.5% to 4%. CHG may be utilized alone or in mixtures with ethanol or isopropyl alcohol. There are also over-the-counter sales of several CHG goods. This review focuses solely on using CHG to prevent infections linked to healthcare-associated infections (HAIs) (E. J. Septimus & Schweizer, 2016). Compared to 70% alcohol, research from 1991 found that CHG alcohol disinfection of the central line site before insertion significantly reduced the risk of central line-associated infections. Regarding site preparation and upkeep, using CHG alcohol has become the norm (Maki et al., 1991; Marschall et al., 2014). In addition to increased hand hygiene compliance, a European quasi-experimental study conducted in 2014 looked at whether universal CHG cloth bathing may reduce the acquisition of MDROs. According to the results of that study, this intervention was linked to a considerable drop in MDROs. Then, in a follow-up cluster randomized study, they discovered that including quick screening and isolation had no additional impact on MDROs (Derde et al., 2014).

Gram-positive and Gram-negative bacteria are both sensitive to povidone-iodine. Applying povidone-iodine topically ranges in concentration from 4% to 10%. Although it is well accepted, it might result in slight skin irritations. Povidone-iodine has been demonstrated to be more effective in killing bacteria than CHG, although it does not have the same long-lasting effects (Block et al., 2000). In one research, a CHG preparation and a povidone-iodine preparation for surgical scrub usage were compared. The researchers discovered that CHG was more persistently active than povidone-iodine (Smylie et al., 1973). For catheter implantation, CHG is preferred over 10% iodine solutions because it has a decreased risk of infection. Despite having broad-spectrum qualities, povidone-iodine is not the best choice for topical decolonization since there is insufficient proof of its persistence and it produces worse results than CHG (Chaiyakunapruk et al., 2002).

Triclosan is effective against both Gram-positive and Gram-negative bacteria. Triclosan acts by concentrating on the numerous intracellular places where bacteria reside. Triclosan resistance occurs as a result of a single step shift in enoyl reductase (Al-Doori et al., 2003). Numerous toothpastes, liquid soaps, and acne treatments sold over-the-counter include triclosan. They range in concentration from 0.15 to 1 percent. But triclosan has also been found in the environment, in human blood, breast milk, and urine. The American Food and Drug Administration (FDA) and the American Environmental Protection Agency are now examining the safety of triclosan from a scientific and legal perspective (EPA)(C. A. Giuliano & Rybak, 2015). Giuliano and Rybak recently evaluated the use of triclosan as a bar of antibacterial soap and its connection to antimicrobial resistance. They came to the conclusion that triclosan had no advantage over non-antimicrobial soap, and that triclosan resistance had been established. They concluded that using triclosan is not worth the hazard (C. A. Giuliano & Rybak, 2015).

Altering cellular metabolism and destroying phospholipids are two effects of sodium hypochlorite. MRSA decolonization has been the primary usage of sodium hypochlorite. A recent study compared no intervention to one of three 5-day interventions: intranasal mupirocin alone, intranasal mupirocin with daily CHG bathing, or intranasal mupirocin plus daily bathing with diluted bleach (one quarter cup of 6% sodium hypochlorite per tub of water). At one month, only 38% of the control group had *S. aureus* completely eradicated compared to 56% with mupirocin alone (P = 0.03), 55% with mupirocin and CHG (P = 0.05), and 63% with mupirocin and bleach (P 0.01) (Fritz et al., 2011). Intranasal mupirocin and bleach baths produced with one-fourth cup of bleach in a one-quarter tub (about 13 gallons), which equals 2.5µl/ml, are suggested as treatments for children and adults with recurrent MRSA skin and soft tissue infections, according to the most recent IDSA guideline on MRSA. For three months, these baths must be taken twice a week for 15 minutes each (Liu et al., 2011).

In order to eliminate nasal *S. aureus*, systemic antibiotics typically cannot reach sufficient concentrations in secretions. As a result, oral antibiotics and topical treatments may be used in conjunction with decontamination regimens. Patients colonized at several locations or different nasal sites may benefit the most from oral medication. Rifampin (300 mg twice daily) and Novobiocin (500 mg twice daily) alone or Rifampin (300 mg twice daily) and Trimethoprim (160 mg)-Sulfamethoxazole (160 mg) were compared in a double-blinded RCT of 94 patients during seven days (800 mg twice daily) the combination reduced *S. aureus* colonization on the entire body (T. J. Walsh et al., 1993). It was discovered that the rifampin and novobiocin group had a decolonization rate of 67% (30 of 45 patients), while the rifampin and trimethoprim-sulfamethoxazole group had a decolonization rate of 53% (26 of 49 patients). Older age, MRSA-positive wound culture, and more than one contaminated site were risk factors for failed decontamination. A 7-day regimen of 2% CHG bathing once daily, 2% intranasal

mupirocin usage three times daily, 300 mg of oral rifampin twice daily, and 100 mg of doxycycline administered twice daily was compared to no therapy for MRSA decontamination at all body locations in an open-label RCT of hospitalized patients (Simor et al., 2007). One hundred twelve individuals were examined after three months out of the 146 patients who were randomly assigned. At three months, MRSA cultures were negative in 32% (8 of 25 patients in the control group) and 74% (64 of 87 patients) in the treatment group and control group, respectively (RR = 1.55; 95% CI, 1.17 to 2.04; P 0.01). However, adverse side effects, such as nausea, vomiting, and diarrhea, happened in 25% of the participants. In a recent study, Cluzet et al. evaluated the time it takes for MRSA colonization to clear up and the predictors of this process. They discovered that clindamycin treatment of skin and soft tissue infections led to earlier MRSA colonization clearance (Cluzet et al., 2015). Although various studies have assessed the use of oral medications in the decontamination of *S. aureus* patients, it is still unknown what the ideal dosage and length of therapy should be, as well as if combination therapy is preferable to monotherapy. Oral decolonizing drugs such as rifampin, quinolones, trimethoprim-sulfamethoxazole, novobiocin, clindamycin, doxycycline, and minocycline have all been studied; however, the most recent findings do not indicate a preferred agent (Muder et al., 1994; T. J. Walsh et al., 1993). Furthermore, it is not apparent if oral decolonizing drugs are more effective than topical ones. The possibility of resistance and the adverse effects must be considered when assessing these treatments. Current recommendations advise against using oral medicines for decolonization regularly (Liu et al., 2011).

There have only been a few analyses of how successful horizontal and vertical decolonization measures are in terms of cost. According to a number of economic computer models, screening and nasal decontamination are cost-effective in some patient populations but not in others (E. J. Septimus & Schweizer, 2016). Murthy et al.

investigated a combined strategy that includes contact isolation for patients who tested positive for MRSA, PCR-based MRSA screening prior to surgery, and mupirocin and CHG decontamination of patients who tested positive for MRSA. According to their findings, the expenses that would have been incurred had MRSA infections not been reduced were not offset enough by the costs of screening, making this not enormously cost-effective (Murthy et al., 2010). However, the data used to create this model came from a hospital in Geneva, which could have lower rates of MRSA colonization than hospitals in the United States. On the other hand, several studies utilizing data from the United States discovered that MRSA screening and disinfection prior to cardiac, vascular, orthopedic, or heart-lung transplant was cost-effective from the standpoint of the third-party payer and the hospital (Clancy et al., 2014; B. Y. Lee et al., 2010). However, Lee et al. discovered that screening and decontaminating expectant mothers before cesarean birth were not financially advantageous (B. Y. Lee, Wiringa, et al., 2011). Furthermore, various economic models have determined that MRSA screening and decolonization are economical for all hospitalized patients, including those receiving hemodialysis and those in intensive care units (S. S. Huang et al., 2014; B. Y. Lee, Song, et al., 2011). Two separate investigations determined the cost-effectiveness of universal decolonization in the ICU environment (S. S. Huang et al., 2014; Ziakas et al., 2015). One economic model examined seven potential approaches to stop MRSA from spreading and infecting ICU patients and discovered that the approaches that included decontamination were more affordable and efficient than the alternatives (Gidengil et al., 2015).

The capacity of *S. aureus* to acquire resistance to antibiotic therapy is one of the most striking and challenging elements of managing the bacteria clinically. This effect was demonstrated during the rise of methicillin-resistant *S. aureus* (MRSA) in the 1960s and more recently with strains demonstrating intermediate resistance to vancomycin, one of the few remaining treatments for MRSA infection, and on infrequent occasions, total

resistance to it (J. W. Park et al., 2019). There is a pressing need for alternative treatments for *S. aureus*. Since the human immune system has so many different functions, immunotherapies are a desirable alternative since there is less chance that resistance will arise. The scientific community has worked hard over the past two decades to create a vaccine that can shield against *S. aureus* infection, but no vaccine candidates are effective in clinical trials (Clegg et al., 2021).

The infection caused by *S. aureus* currently has no vaccination. Innovation in the creation of *S. aureus* vaccines is praiseworthy. As shown via the vast range of antigen selection and innovative adjuvants and delivery technologies intended to harness particular humoral and cellular responses, an ever-increasing variety in vaccination platforms is being noticed. In order to increase the chance of success while producing new vaccines, it is crucial that facts explaining the historical failure of vaccinations be adequately studied and considered (Clegg et al., 2021). The development of a vaccine has been delayed for several important reasons, one of which is the failure to properly convert the vaccination protectivity seen in preclinical infection models to protective effects shown in human individuals. In this case, Clegg et al. contend that the use of more relevant animal models, more accurate in vitro models, and ex vivo human subjects to study the pathogenicity of *S. aureus* would increase the validity of data obtained at the preclinical stage and thereby increase the likelihood that vaccines moving into clinical trials will be effective (Clegg et al., 2021; Warren et al., 2015).

Additionally, protein A is produced by *S. aureus*. In order to restrict the adaptive immune response by decreasing the synthesis of B cell antibodies, Protein A, a surface protein with a large number of interactions with immunoglobulin's Fc domain, is necessary (D. Parker & Prince, 2012). Evidence suggests that protein A is a deterrent to antibody development and activity against *S. aureus*. Deletion or mutation of the IgG binding region of protein A to nullify this effect has also shown promise in vaccination

experiments in mice (H. K. Kim et al., 2015). The lack of an *S. aureus* vaccine on the market might be attributed to several factors. Due to the fact that *S. aureus* produces a wide range of virulence factors, vaccination against any of them could not be successful. For this reason, recent studies are concentrating on several antigen formulations. Clinical studies are now being conducted on these formulations, which include a variety of *S. aureus* virulence factors, including clumping factor A (ClfA), manganese transport protein C (MntC), fibronectin-binding protein B (FnbB), and capsular polysaccharides (Anderson et al., 2012; Rauch et al., 2014). Purified proteins of *S. aureus* virulence factors or surface proteins have been used in all vaccines created to date rather than attenuated live strains (D. Parker, 2018). Clegg et al. also consider some of the complexity of the host-pathogen connection between humans and *S. aureus* to comprehend better the difficulties involved in developing an effective vaccine. In addition to being a significant human pathogen with several virulence factors targeted at neutralizing crucial immune system components, *S. aureus* also creates colonization relationships, leading to the majority, if not all, people having pre-existing immunity (Albrecht et al., 2015; Dryla et al., 2005). The enhancement of natural immunity and the resulting provision of infection protection will consequently be necessary for effective vaccination. There are now various approaches being researched for targeting *S. aureus*, in addition to the creation of vaccines. This is due to the advent of innovative therapeutic and short-term prophylactic therapies for *S. aureus* ailment. Exciting new experimental therapies for *S. aureus* include the revival of techniques like bacteriophage therapy, monoclonal antibody therapy, and antibiotics, as well as the creation of novel therapeutic proteins like centyrins (Clegg et al., 2021).

Immunology of *S. aureus*

In order to successfully colonize the mammalian host, bacterial cells must get past a variety of innate and adaptive host defense mechanisms (Skaar et al., 2004). The

host immune system is able to identify, combat, and get rid of *S. aureus* (Karauzum & Datta, 2017). Innate and adaptive immune systems are subsets of the immune system. When a pathogen comes into touch with a host, the innate immune responses are the initial line of defense that goes into action. Although innate immune responses are quick and non-specific, they can distinguish between self and other helpful commensal bacteria and pathogens invading the body. The innate immune system's components trigger the adaptive immune system, which is a delayed, focused reaction. Immunological memory is developed by the adaptive immune system, enabling a quick reaction to later reinfection by the same pathogen. Later in the course of infection, adaptive immunity against *S. aureus* infection starts to develop. The adaptive immune system responds by activating B and T cells, producing antibodies, and releasing cytokines. This may further modulate and magnify the innate immunity's initial reaction (Karauzum & Datta, 2017). One of the principal methods for removing *S. aureus* from the body is thought to be phagocytosis by neutrophils (Bekeredjian-Ding et al., 2017).

The first line of defense against an environmental microbial invasion is the skin (Nestle et al., 2009). Keratinocytes that are closely packed together and the constant desquamation of the epidermal cells provide the skin's immunological defense (Proksch et al., 2008). Additionally, filaggrin component breakdown in the stratum corneum generates acidic components such as urocanic acid (UCA) and pyrrolidone carboxylic acid (PCA). These elements lower the pH of the skin's surface and prevent *S. aureus* CWA proteins ClfB, FnbpA, and protein A from being expressed (Miajlovic et al., 2010). Skin commensal microorganisms also provide defense against *S. aureus*. For instance, PSM and the serine protease Esp generated by S. epidermidis on the skin prevent *S. aureus* from colonizing (Cogen et al., 2010). Additionally, antimicrobial peptides produced by epidermal keratinocytes, such as beta-defensins, RNase7, and cathelicidin, have an inhibitory effect against *S. aureus*, hindering the successful colonization of the

host (Proksch et al., 2008; Schauber & Gallo, 2008). *S. aureus* is very susceptible to human beta-defensins (Kisich et al., 2007). It has been demonstrated that cathelicidins are exceptionally efficient in killing both external and intracellular *S. aureus* by creating holes in the pathogen's cell membrane (Noore et al., 2013; Xhindoli et al., 2016). RNase7 was identified in the skin's stratum corneum and prevented *S. aureus* from colonizing skin explants (Ryu et al., 2014). The presence of *S. aureus* or other elements like lipoteichoic acid (LTA) can stimulate the synthesis of these antimicrobial peptides (Hattar et al., 2006).

Pathogen recognition receptors (PRRs) are found in cells in the nasal cavity, skin, and other *S. aureus* colonization sites. These PRRs identify conserved microbial components in *S. aureus* and other pathogenic microorganisms known as pathogen-associated molecular patterns (PAMPs)(Janeway, 1989). Teichoic acid, LTA, and other surface-associated substances are some of the PAMPs for *S. aureus*. Toll-like receptors (TLRs) are an essential subset of PRRs. The extracellular domain, transmembrane area, and cytosolic Toll/IL-1 receptor (TIR) domain are all found in the transmembrane proteins known as TLRs (Kawai & Akira, 2011). TLR2 is a crucial TLR that detects *S. aureus* and its microbial component. A mouse missing TLR2 has shown its significance in reducing *S. aureus* infections (Yimin et al., 2013). In addition, it has been proposed that decreased TLR2 activation in people with atopic dermatitis may have a role in *S. aureus* skin infections. TLR2 interacts with LTAs and lipoproteins produced on the surface of *S. aureus* through the heterodimer complex; it forms with either TLR6 or TLR1 to become functionally active (Kawai & Akira, 2011; Niebuhr et al., 2011). TLR2 interaction with its ligands causes the intracellular signaling cascade to be activated, which activates the transcription factor nuclear factor-κB (NF-κB), which in turn causes the synthesis of inflammatory substances such as chemokines and cytokines (Niebuhr et al., 2010). Additionally, adhesion molecules, including E-selectin, Intercellular Adhesion

Molecule 1 (ICAM1), and Vascular Cell Adhesion Molecule 1 (VCAM1), are expressed more often as a result of NF-κB (Aggarwal, 2004). These adhesion molecules draw blood-borne immune cells like neutrophils into circulation. The development of epidermal tight junctions is also promoted by TLR2 activation, which improves the skin's barrier function. Additionally, commensal skin bacteria that activate TLR2 increase the generation of antimicrobial peptides that can prevent *S. aureus* colonization and infection (Kuo et al., 2013; Wanke et al., 2011).

More than 30 proteins in tissues and blood comprise the complement system. Prior to being cleaved, complement proteins are inactive. After activation, they interact and set off a series of events that aid in battling the infection. The lectin pathway (LP), the alternative pathway (AP), and the classical pathway (CP) are the three pathways that can activate the complement system. The molecules that can trigger these pathways vary. The direct binding of C1q to the bacterial surface or C1q binding to antibody complexes (IgM or IgG) existing on the bacterial surface are the two mechanisms by which the classical route is activated. The alternative pathway is activated when the spontaneously produced C3b binds to bacteria. The lectin pathway is triggered by attaching ficolin or a mannose-binding lectin to the mannose-containing carbohydrates on the bacterial surface. Regardless of the mechanisms, complement activation causes the synthesis of C3 convertases (E. J. Brown, 1991; Kawai & Akira, 2011; Noris & Remuzzi, 2013). There are three functions for complement activation.

First, the activated complement components attach to the pathogen surfaces, opsonizing them and increasing the efficiency of pathogen phagocytosis. Second, during complement activation, effector proteins like C5a and C3a operate as chemoattractants to draw circulating immune cells (phagocytes) into the body. Additionally, when complement is activated, a membrane attack complex (MAC) is produced, which lyses the pathogen's membrane, notably in the case of Gram-negative bacteria (E. J. Brown,

1991; Kawai & Akira, 2011; Noris & Remuzzi, 2013) . The higher mortality shown in complement-depleted mice following *S. aureus* bacteremia serves as evidence of the necessity of complements in the fight against *S. aureus* (Cunnion et al., 2001). Additionally, it has been demonstrated that activating complements on *S. aureus* surfaces decreased the bacteria's adhesion to the surfaces of endothelial cells (Cunnion & Frank, 2003).

The first to move to the site of staphylococcal infections are neutrophils. The fact that those with impaired neutrophil activities are more susceptible to *S. aureus* infections shows how crucial neutrophils are in preventing *S. aureus* infection (Lekstrom-Himes & Gallin, 2000; Rigby & DeLeo, 2012). Neutrophils' primary function in fighting infection is to phagocytose pathogens that the PRRs have identified. Additionally, they are crucial in developing an abscess upon staphylococcal infection (Kobayashi et al., 2015). A gradient of chemotactic signals, such as Interleukin-8 (IL-8), complement components C3a and C5a, aids in drawing circulating neutrophils to the infection site (Rigby & DeLeo, 2012; Spaan, Surewaard, et al., 2013). Rolling adhesion, integrin activation, firm adhesion, and transmigration are the four phases of the recruitment process (McGuinness et al., 2016). The process of capturing circulating neutrophils begins when they adhere to adhesion molecules on endothelial cells, such as E-selectin, P-selectin, and intracellular adhesion molecule (ICAM) (McEver & Cummings, 1997). Neutrophil-expressed receptors such as P-selectin glycoprotein ligand 1 (PSGL-1) make it easier for cells to bind to these adhesion molecules. They then stop moving through the bloodstream and move along the endothelium walls in the direction of the diseased tissue location (Hidalgo et al., 2007; Ley et al., 2007).

The presence of opsonins such as complement factors and immunoglobulins on the pathogen's surface promotes more effective phagocytosis by neutrophils (Rigby & DeLeo, 2012). The opsonins interact with the receptors present on the cell surface of

neutrophils and include the Fc and complement receptors (Futosi et al., 2013; McGuinness et al., 2016). However, it has also been noted that pathogen phagocytosis by neutrophils occurs more slowly in the absence of opsonization (Vandenbroucke-Grauls et al., 1984). These interactions trigger the pathogen's phagocytosis, which results in the development of phagosomes. Phagosomes go through several stages of maturity, which ultimately destroy bacteria (W. L. Lee et al., 2003). To assure bacterial eradication, neutrophils create reactive oxygen species (ROS), proteinases, and AMPs, among other substances (Segal, 2005). Additionally, by using neutrophil extracellular traps (NETs) that are coated with antimicrobials, neutrophils can capture and kill *S. aureus* (Brinkmann et al., 2004).

Bacterial immunoglobulin (IgG)-binding proteins are protein A. Protein A is made by *Staphylococcus aureus*. Staphylococcus-derived Protein A is notable for its capacity to attach to various species' constant (Fc) region of immunoglobulin molecules. Most IgG molecules bind protein A, albeit the affinity varies between IgG subclasses and species. This is the sole condition that must be met without exception. Although the sensitivity of the protein A-peroxidase and similar protein A-PAP methods is inferior to that of the PAP, ABC, or streptavidin-based approaches, they have advantages that can justify their usage in some situations. (C. R. Taylor et al., 2006).

In order to combat host immunological defenses against them, *S. aureus* has evolved evasion mechanisms or tactics. These tactics are made possible by secreted or surface-bound virulence, which aids *S. aureus* in interfering with the operation of the host's natural defenses, fostering an environment that is more favorable to the bacteria's survival and growth. Different methods of immune evasion are used by *S. aureus*. The capacity of *S. aureus* to avoid the phagocytic and intracellular killing functions of neutrophils is the most noticeable of these evasion techniques. Additionally, *S. aureus* retains the capability to disrupt TLR signaling, complement activation, and opsonization

and decrease the potency of antimicrobial peptides (F. Askarian et al., 2018; Foster, 2005).

By blocking the cytotoxic mechanisms that result in bacterial destruction, *S. aureus* can persist after being consumed within phagocytes. Staphyloxanthin and superoxide dismutases, two compounds produced by *S. aureus*, shield the bacteria from the phagosomes of neutrophils' reactive oxygen species (Foster, 2005; Guerra et al., 2017).

S. aureus nasal carriage in Children

The respiratory microbiome is a complex network of bacteria, viruses, fungi, bacteriophages, archaea, and eukaryotes that colonize the mucosal surfaces of the respiratory tract and constantly interact with each other, the host, and the environment (Henares, Brotons, et al., 2021). It has been postulated that the degree of stability of microbiota is largely determined by the initial colonization by keystone species during the early days of life (Relman, 2012). Although community-associated [CA] *S. aureus* [CA-SA] is the kind of *S. aureus* most frequently found in children, it has long been known that CA-SA is a significant contributor to nosocomial infections in children who are hospitalized (healthcare-associated [HA] *S. aureus*)(Rojo et al., 2010).

Over the past ten years, infections with *S. aureus* have dramatically increased. Compared to other age groups, children have a much higher prevalence of infection from methicillin-resistant SA strains (MRSA). Children are a crucial source of SA transmission in the community and hospital settings. They are also a significant SA reservoir (Gorwitz, 2008; J. A. Patel et al., 2015). The assessment of MRSA nasal colonization in children has been the subject of numerous studies in the last few years, both in hospitals and among the general public. In comparison to adults, persistent carriage rates are higher in children and adolescents under 20 (Armstrong-Esther & Smith, 1976; Gorwitz, 2008). It is well known that infants become colonized with *S. aureus* soon after delivery. Breastfeeding, the

number of people living in the home, low birth weight, early gestational age at birth, indwelling catheters, and the number of days spent on antibiotics or a ventilator are all recognized risk factors for newborn *S. aureus* colonization (Chatzakis et al., 2011; Shiri et al., 2013).

Given the enormous microbial diversity in colostrum, transition, and mature milk, it is envisaged that human milk will be a primary supply of keystone species for human microbiota development, particularly in the respiratory tract (Hunt et al., 2011). Human milk is widely regarded as the best source of nourishment for practically all healthy newborns due to the numerous health advantages it provides. Breastfeeding, for example, protects newborns against diarrheal and respiratory infections and is linked to a lower long-term risk of obesity, particularly in developing nations (Hunt et al., 2011). Culture-dependent techniques have long-established the presence of bacteria, including Staphylococcus and Streptococcus species, in aseptically collected milk (Heikkilä & Saris, 2003). Recent research, however, has revealed that human milk contains colonies of bacteria that may have health effects (Heikkilä & Saris, 2003). While these studies show that bacteria are present in aseptically collected milk, very little research has looked at the plausibility of a core milk microbiome among nursing mothers or the stability of these communities inside a person over time. These sorts of studies are essential because they allow researchers to understand the functions these communities may play in sustaining mammary gland health, bacterial colonization of the newborn's gastrointestinal tract, and other short- and long-term mother and infant health indicators. As a result, the current study was meant to delve deeper into human milk bacterial populations' long-term stability and diversity. We expected that human milk includes a wider variety of bacterial phylotypes than had previously been observed and that these communities would remain stable over time inside each nursing woman (Hunt et al., 2011).

The rates of persistent carriage appear to be greater in children and adolescents under 20 than in adults. The prevalence of persistent carriers was reported to be 10% in 0 to 9-year-old children and 24% in 10 to 19-year-old youngsters. Infants 3 months old or less were found to have the most remarkable *S. aureus* carriage rate (Lebon et al., 2008; Regev-Yochay et al., 2004). According to research by Ciftci et al., 23% of school-aged children had *S. aureus* in their nasal passages. This *S. aureus* nasal carriage result was consistent with Ghanaian data (22.6%) (Ciftci et al., 2007; Eibach et al., 2017). Nevertheless, this study found a greater frequency of *S. aureus* nasal carriage in Serbia (2.59%), China (2.4%), Iraq (17.75%), Vietnam (10.4%), and Nigeria (18.3%).(Hussein et al., 2015; Tigabu et al., 2018). However, compared to findings from Italy (39.2%), India (25%), Iran (28%), Iraq (30%), Nigeria (56.3%), Jimma (47.34%), and Bahir Dar (41%), this study demonstrated a reduced prevalence of *S. aureus* nasal carriage (Kejela & Bacha, 2013; Nsofor et al., 2015; Shetty et al., 2014; Tigabu et al., 2018). This discrepancy in the nasal carriage rate of *S. aureus* from previous studies may result from several factors, including the characteristics of the research population, the caliber of the sample, the culturing methods, the geographic distribution, and the diagnostic methods.

According to research by Tigabu et al., a significant proportion of MRSA isolation in urban primary schools was identified among students. The high prevalence of MRSA in urban schools compared to rural schools may be caused by several factors, including the proximity of health care facilities to urban schools, the high antibiotic selective pressure in urban settings, and the prevalence of overcrowding in urban as opposed to rural schools (Tigabu et al., 2018).

The study by Tigabu et al. established that the number of pupils in the classroom was a significant risk factor for *S. aureus* nasal colonization. Most pupils with high nasal *S. aureus* carriage belonged to students who had between 41 and 60 peers in the classroom, demonstrating that as class size increased, nasal *S. aureus* carriage in schoolchildren

became more common. This outcome was in line with research by Jimma et al. in Ethiopia. The likelihood of frequent interaction between children in a classroom, close quarters, and increased nasal flora sharing, all of which contribute to the transmission of pathogens, are likely explanations (Kejela & Bacha, 2013; Tigabu et al., 2018).

S. aureus interaction with other nasal commensals

The aggregate genomes of commensal, symbiotic, and pathogenic microbes in the human body make up the human microbiome, which is essential for immunity and health. Every bodily environment is home to a distinct bacterial population, which is not stable throughout life but instead shifts with aging (Abt et al., 2012; Thomas et al., 2017). Commensal bacteria are present in the human nasal cavity, where they prevent the colonization of opportunistic pathogens by competing for available resources and space. Some commensal bacteria can even produce toxins that directly inhibit or eliminate rival pathogens (Hardy et al., 2019). Numerous environmental variables have been proven to influence the nasal microbiome's makeup. Some microorganisms, like *Staphylococcus aureus*, maybe both commensal and versatile opportunistic pathogens, causing severe illness and even mortality, so it is difficult to distinguish between commensal and pathogenic microbes (Otto, 2010; Rawls & Ellis, 2019). Therefore, dysbiosis, a malfunction or imbalance in microbial populations, can significantly influence human health (Degruttola et al., 2016). The development of conditions like allergic rhinitis (AR), chronic rhinosinusitis (CRS), acute respiratory tract infections (ARTI), otitis media (OM), and asthma are thought to be influenced by the presence of opportunistic pathogens in the nasal cavity, according to research into the bacterial communities of the nasal cavity (Esposito & Principi, 2018; Y. J. Huang, 2017).

The habitat of commensals may be threatened when they share an ecological niche with pathogenic bacteria that might cause pathophysiological reactions. Therefore, the capacity to directly compete with the invader may be advantageous. The anterior nares are

the primary location of *S. aureus* colonization, and this body region was the focus of many of the earliest research on direct microbial interactions with *S. aureus* (J. Kluytmans et al., 1997; Krismer et al., 2017). According to some reports, artificial implantation of Corynebacterium sp. and S. epidermidis prior to administration in a murine nasal colonization paradigm of colonization prevented subsequent *S. aureus* colonization, and S. epidermidis strains releasing a serine protease (Esp) cleared *S. aureus* nasal colonization in human volunteers (Iwase et al., 2010; B. Park et al., 2011). Other research exhibited that the Esp protease breaks down the colonization-related host and bacterial surface proteins (C. Chen et al., 2013; Sugimoto et al., 2013). It is evident that many commensal CoNS strains generate antibiotic-like chemicals when researchers collect a larger sample of these organisms from nasal locations, which may facilitate commensal colonization in a hostile environment (Janek et al., 2016). One well-known instance is the discovery that Staphylococcus lugdunensis produces lugdunin, a thiazoline-containing cyclic peptide antibiotic that inhibits *S. aureus* in the well-known cotton rat nasal colonization paradigm. S. lugdunensis nasal carriage in humans was adversely associated with *S. aureus* colonization, albeit this study did not prove direct competition (Zipperer et al., 2016). Some bacterial species can secrete anti-staphylococcal compounds that can control the population of *S. aureus*. For instance, *S. aureus* can be terminated by Streptococcus pneumoniae's in vitro generation of H_2O_2 (Regev-Yochay et al., 2006; Selva et al., 2009).

The "ecological niche" of the anterior nares has a limited area that may be occupied, and many genotypes compete with one another for this space, according to Dall'Antonia et al.

In one research, 8% of patients had nasal MRSA colonization, while 17% had MSSA colonization. The researchers concluded that although various organisms might fight for the same niche, MSSA has superior fitness, suggesting that it may hinder colonization by MRSA since just 0.6% of the samples co-colonized with both. The

additional resistance mechanisms of MRSA, which have costs associated with viability and competitiveness, can be used to explain the fitness advantage of MSSA over MRSA.(Dall'Antonia et al., 2005; Hurdle et al., 2004).

Chapter Summary

This chapter serves as a comprehensive literature review, focusing on similar carriage studies of Staphylococcus aureus and its identification, therapeutic approaches, and challenges with antibiotic resistance. The review emphasizes the strengths and weaknesses of existing treatment methods for S. aureus infections. Special attention was given to exploring various identification techniques and molecular methods employed in this study, accompanied by a rationale for their selection. By analyzing previous research, this chapter lays the groundwork for the current study's objectives and contributes valuable insights into S. aureus carriage and its management.

CHAPTHER THREE

RESEARCH METHODS

Introduction

This study examined the distribution of *Staphylococcus aureus* among healthy children in Cape Coast and its antibiotic resistance profiles. Resistance patterns to a few selected antimicrobials and the prevalence of staphylococcal carriage were discovered. The resources and procedures described in this chapter were employed to meet the various goals of the study. The laboratory processes, data analysis, research design, study population, sample, and sampling procedure are all covered in detail.

Study area

Cape Coast is a city, fishing port, and the capital of Cape Coast Metropolitan District and Central Region of south Ghana. One of the country's most historic cities, it is the location of Cape Coast Castle, a World Heritage Site, with the Gulf of Guinea situated to its south. According to the 2010 census, Cape Coast had a settlement population of 169,894 people. The language of the people of Cape Coast is Fante (GSS, 2012). Cape Coast is a humid area with mean monthly relative humidity varying between 85% and 99% (Cape Coast Metropolitan Assembly, 2012). In Cape Coast, the population census for children under five years is 20441. Of these, the girls are 10003, and the boys are 10438 (*Cape Coast, Ghana — Statistics 2022*, 2022).

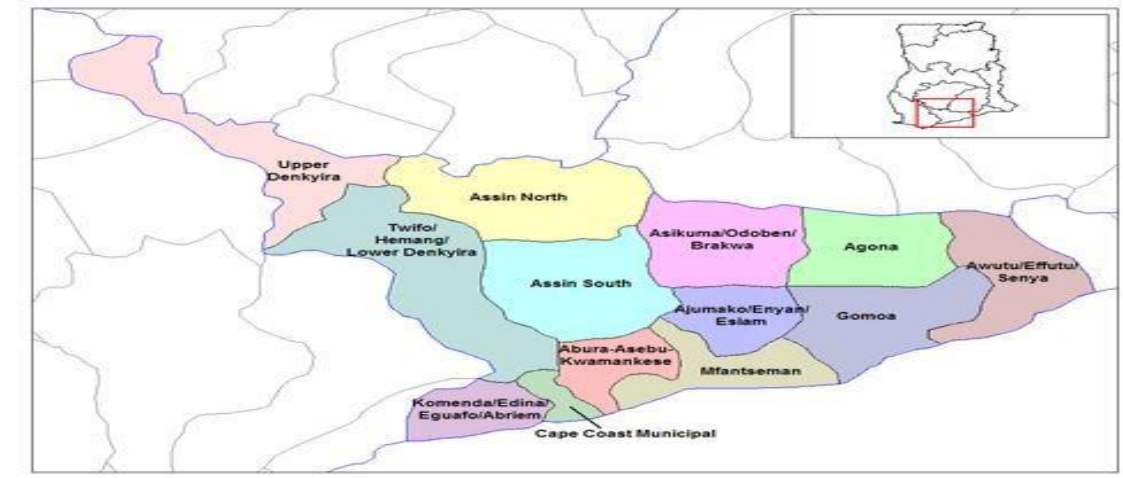


Figure 2: A map showing the study area (Ghana districts.com)

Study design

A retrospective and cross-sectional study of carriage isolates of *Staphylococcus aureus* collected in February 2018.

Study population

Isolates were obtained from healthy children under five years old attending kindergartens and immunization centers within the Cape Coast metropolis.

Specimen and Data Collection

Healthcare personnel underwent training on the proper technique for obtaining nasopharyngeal swabs from the study participants. The swabs used were Copan plastic mini-tip flocked swabs, and the guidelines recommended by the World Health Organization (WHO) were followed (Satzke et al., 2013). The swab stick was inserted gently into the back of the nasopharynx, rotated, and then removed. It was then placed in a vial labeled with 1ml of sterilized skimmed milk-tryptone-glucose-glycerine (STGG) transport medium. The collected specimen was immediately transported on ice to the laboratory within eight hours, following the

WHO protocol for nasopharyngeal carriage evaluation. In the laboratory, the swab was vortexed for 10-20 seconds to disperse the microbes, and the specimen was stored at -80°C until it was ready for further characterization.

Data was also collected from consenting parents or guardians of participating children at the immunization centres with the aid of a pretested questionnaire. The questionnaire explored areas of socio-demographic information such as age, sex, religion, and type of facility.

Bacterial isolation and identification

S. aureus identification was based on the guidelines of the Infectious Disease Society of America (IDSA). In brief, frozen nasopharyngeal (NP) samples were thawed, vortexed for 10–20 seconds, and 10 µL of every sample was inoculated on 5% defibrinated sheep blood agar. Inoculated isolates were incubated at 37°C for 24 hours. Single colonies were tested using the tube coagulase method and growth on Mannitol Salt agar (MSA) to confirm *S. aureus* isolates. To confirm the fermentation of mannitol, the growth of yellow colonies on MSA surrounded by yellow zones after 24 hours of incubation at 37°C indicated a positive result. Furthermore, molecular identification of *S. aureus* was made using designed primers.

Antibiotic susceptibility testing

In accordance with Giuliano et al. description, the disc diffusion method was employed to assess the antimicrobial susceptibility of *Staphylococcus aureus* isolates on Mueller-Hinton agar (C. Giuliano et al., 2019). Antibiotics discs containing 30 µg cefoxitin (FOX), 30 µg cefuroxime (CXM), 30 µg

chloramphenicol (CHL), 15 µg erythromycin (ERY), 25 µg cotrimoxazole (COT), 30 µg tetracycline (TET), 2 µg clindamycin (DA), 30 µg ceftriaxone (CRO), 30 µg amikacin (AMK), 25 µg amoxicillin (AMX), 10 µg amoxicillin clavulanic acid (AMC), 10 µg gentamicin (GEN) and 5 µg levofloxacin (LEV) were used. *S. aureus* suspension equivalent to 0.5 McFarland standard was inoculated on Müller-Hinton agar. Antibiotic discs were applied on the agar plates followed by incubation at 37ºC for 18–24 hours The diameter of the inhibition zones were measured and compared with the Clinical and Laboratory Standards Institute guidelines (CLSI) breakpoint to determine if an isolate was susceptible or resistant to an antibiotic (Clinical and Laboratory Standards Institute (CLSI), 2021). Isolates showing resistance to ≥3 classes of antibiotics were classified as multidrug-resistant (MDR).

Resistance Genes Identification

DNA extraction

Genomic DNA from *S. aureus* was extracted and purified by vortexing a solution containing colonies of *S. aureus* in distilled water for about 2 min; then, the solution was boiled for 10 min at 95°C and frozen in the freezer for 10min; this procedure was repeated three times. Afterward, the solution was centrifuged at 4000 rpm for 5 min at 4ºC. The supernatant containing the DNA was pipetted into Eppendorf tubes (Dashti et al., 2009).

Molecular identification of *S. aureus*

Polymerase chain reaction (PCR) amplification of virulence and resistant genes was carried out. Specific primers were used to amplify the virulence and resistant genes in all *S. aureus* isolates. For gene amplification, 25 µl of the reaction

mixture was prepared to contain 3µl of template DNA, 1µl of primers, 1 µl of 5mM dNTPs, 1µl Taq polymerase, 2.5µl of 10x Taq buffer, and 1µl of 25 mM $MgCl_2$. All the isolates were amplified individually for the *nuc* gene using the *nuc* specific primer, initial denaturation of 94°C for 5 min followed by 40 cycles of denaturation at 94 °C for 1 min, annealing at 58 °C for 1 min, extension at 72 °C for 1 min, and a final extension at 72 °C for 8 min, on a DNA engine.

Molecular Detection of resistant genes

The PCR primers were used to detect the staphylococcal virulence genes, including the *vanB, ermC, mecA, tetK,* and *blaZ*. Primer length, guanine, and cytosine content was kept as uniform as possible to minimize differences in annealing temperatures. For the gene amplifications, 25 µl of the reaction mixture was prepared to contain 3µl of template DNA, 1 µl of each primer, 1 µl of 5mM dNTP mix, 2 µl Taq polymerase, 2.5 µl of 10x Taq buffer, and 1µl of 25 mM $MgCl_2$. The PCR conditions were: initial denaturation at 94°C for 5 min, followed by 30 cycles of denaturation at 94°C for 3min, 30s with the respective annealing temperature, extension at 72°C for respective extension time, and a final extension at 72°C for 10 min on a DNA engine. The PCR mixture and conditions were similar to those described by Li et al. (Li et al., 2018).

Table 1: List of Primer sequences used in the Study

Primer	Primer sequences	Product size (bp)	References
nuc	F: GCGATTGATGGTGATACGGTT R: AGCCAAGCCTTGACGAACTAAAGC	279	Hussain et al., 2016
tetK	F: GTA GCG ACA ATA GGT AAT AGT	360	Strommenger et al., 2003

	R: GTA GTG ACA ATA AAC CTC CTA		
blaZ	F: ACT TCA ACA CCT GCT GCT TTC	173	Martineau et al., 2000
	R: TGA CCA CTT TTA TCA GCA ACC		
vanB	F: AGACATTCCGGICGAGGAAC	220	Akanbi et al., 2017
	R: GCTGTCAATTAGTGOGGGAA		
mecA	F: GTAGAAATGACTGAACGTCCGATAA	310	Netsvyetayeva et al.,2014
	R: CCAATTCCACATTGTTTCGGTCTAA		
ermC	F: CTT GTT GAT CAC GAT AAT TTC C	190	Martineau et al., 2000
	R: ATC TTT TAG CAA ACC CGT ATT C		

Polymerase chain reaction product detection

Amplicons were combined with 6X loading dye (Thermo Scientific) in a 5:1 ratio, and 5µL of the mixture was loaded onto a 1.5% agarose gel produced from 1X TAE (Tris-HCL Acetic acid EDTA) and stained with HD GreenTM DNA Stain solution (0.5 g/ml), along with 5µL of 100 bp and 1000 bp Gene rule (Life Sciences Biotechnology, China). At 100 V for 30 minutes, the amplicons were electrophoresed. By comparing them to the gene ruler (Thermo Scientific), the sizes of the amplicons were ascertained by viewing the gel pictures using a UV transilluminator (Cleaver Scientific Ltd).

Quality Control

To minimize cross-contamination, standard microbiological procedures were strictly followed. Positive and negative controls were included in the test reactions. DNA extraction and PCR amplifications were carried out in molecular laboratories separate from the clinical microbiology laboratory, where cultures

were grown. The PCR laboratory designated sections for pre-amplification, amplification, and post-amplification, with a unidirectional movement of staff.

Data Processing And Analysis

Data in Excel format was exported to SPSS version-25 for analysis. All data were tabulated for descriptive statistics. Categorical variables were summarized in frequency (n) and percentage (%). The characteristics of the study populations were summarized using frequencies, mean, and standard deviation or median and interquartile range (IQR) depending on the normality of distribution. Categorical variables were expressed as proportions and compared using the Chi-square test (two-tailed). The confidence interval (CI) was set at 95% for univariate and multivariate analysis, and a p-value of <0.05 was considered statistically significant.

Chapter Summary

The target demographic for this study was children under five, and Cape Coast Metropolis was chosen as the study location. In this investigation, archived samples from 2018 were used. The statistics may not accurately represent the total population, though, given that the sample only included a small portion of the Cape Coast Metropolis. The research is cross-sectional. Due to this, the study was unable to assess the *staphylococcus aureus* carriage duration or pinpoint the dynamics of carriage and acquisition rates.

CHAPTER FOUR

RESULTS AND DISCUSSION

Introduction

This chapter includes data analysed on the sociodemographic characteristics of participants, antimicrobial susceptibility testing. This research aims to identify the carriage rate, antibiotic resistance profiles, and antibiotic resistance and virulence genes of *S. aureus* isolated from the nasopharynx of children living in Cape Coast, Ghana. Therefore, this study will provide primary data on *S. aureus* carriage among children on Cape Coast. A cross-sectional study was performed with 880 isolates. Cultures from nasal samples were obtained and characterized according to their antimicrobial susceptibility profile and resistance factors-encoding genes. Potential risks for *S. aureus* and MRSA carriage were analysed. Data obtained were entered into Microsoft Excel, and analysis and graphs were plotted using SPSS v25.

Description of the study participants

This study enrolled 880 apparently healthy children from 11 communities across Cape Coast. Of the 880 participants, 50.2 % (N= 442) were males, and the remaining 49.8 % (N=438) were females. The majority of the participants (221, 25.1 %) were between the ages of 25 and 36 months. There were 11 localities included in the sampling, with Abura providing the majority of samples (27.7%) and Pedu providing the least (1.1%). Most participants were Fantes (369, 41.9%) and Christians (756, 85.9 %). Of the 880 participants included in this study, 145 were culture positive for *S. aureus*. Thus, the prevalence of *S. aureus*

nasopharyngeal carriage was 16.5% (95% Confidence Interval [CI]: 0.141% – 0.191%).

Table 2: Socio-demographic Characteristic of Participants

Criteria	Frequency	Percentage (%)
Age in months		
0 – 12	149	16.9
13 – 24	215	24.4
25 – 36	221	25.1
37 – 48	97	11.0
49 – 60	160	18.2
61 - 72	38	4.3
Sex		
Male	442	50.2
Female	438	49.8
Religion		
Islam	124	14.1
Christianity	756	85.9
Facility		
Private Schools	441	50.1
Public Schools	156	17.7
Immunization centers	283	32.2
Location of facility		
Amamoma	84	9.5
Kwaprow	73	8.3
Akotokyir	31	3.5
UCC	161	18.3
Aboom Wells	71	8.1
Pedu	10	1.1
Abura	244	27.7
Ansapetu	60	6.8
Bakano	65	7.4
Ewim	53	6.0
Kukuado	28	3.2
Ethnicity		
Fante	369	41.9
Other Akan	260	29.5
Ga-Dangme	49	5.6
Northern tribes	128	12.5
Ewe	60	6.8
Foreign tribe	14	1.6

From Table 3, the number of isolates confirmed to be *S. aureus* was 145 samples, representing 16.5%. *S. aureus* carriage prevalence was highest among 49–60 months, in which 25.6% were carrying *S. aureus* in their nasopharynx. Association was found between *S. aureus* and age in months (p = 0.003) as well as *S. aureus* and facility (p = 0.001). However, no significant association between *S. aureus* and religious affiliation (p = 0.093), ethnicity (p = 0.534), and sex (p = 0.693) was found.

Table 3: Bivariate Analysis Related to *Staphylococcus aureus* Colonization

Criteria	*Staphylococcus aureus* Colonization				X^2 value	p-value
	No		Yes			
	Frequency	Percentage	Frequency	Percentage		
Age in months					18.147	0.003
0 – 12	137	18.6	12	8.2		
13 – 24	185	25.2	30	20.7		
25 – 36	186	25.3	35	24.1		
37 – 48	74	10.1	23	15.9		
49 – 60	123	16.7	37	25.6		
61 – 72	30	4.1	8	5.5		
Sex					0.156	0.693
Male	367	49.9	75	51.7		
Female	368	50.1	70	48.3		
Facility					21.213	< 0.001
Private Schools	352	47.9	89	61.4		
Public Schools	123	16.7	33	22.8		
Immunization centers	260	35.4	23	15.8		
Location of facility					42.519	< 0.001
Amamoma	65	8.9	19	13.1		
Kwaprow	67	9.1	6	4.1		
Akotokyir	28	3.8	3	2.1		
UCC	142	19.3	19	13.1		
Aboom Wells	57	7.8	14	9.6		
Pedu	9	1.2	1	0.7		
Abura	184	25.0	60	41.4		
Ansapetu	50	6.8	10	6.9		
Bakano	61	8.3	4	2.8		
Ewim	53	7.2	0	0.0		
Kukuado	19	2.6	9	6.2		

Criteria	*Staphylococcus aureus* Colonization				X^2 value	p-value
	No		Yes			
	Frequency	Percentage	Frequency	Percentage		
Religion					2.822	0.093
Islam	110	15.0	14	9.7		
Christianity	625	85.0	131	90.3		
Ethnicity					0.156	0.534
Fante	301	40.9	68	46.9		
Other Akan	215	29.3	45	31.0		
Ga-Dangme	42	5.7	7	4.8		
Northern tribes	112	15.2	16	11.0		
Ewe	52	7.1	8	5.6		
Foreign tribes	13	1.8	1	0.7		
Total	**735**	**83.5**	**145**	**16.5**		

Table 4: Multivariate Analysis Related to *Staphylococcus aureus* Colonization

Criteria	OR	p-value	95% CI
Age in months			
0 – 12	0.497	0.241	0.154 - 1.599
13 – 24	0.837	0.721	0.316 - 2.220
25 – 36	0.815	0.658	0.330 - 2.014
37 – 48	1.176	0.734	0.463 - 2.988
49 – 60	1.108	0.821	0.455 - 2.699
Sex			
Female	1.195	0.342	0.827 - 1.727
Facility			
Private Schools	0.680	0.054	0.460 - 1.007
Location of facility	1.009	0.763	0.952 - 1.069
Pedu			
Religion			
Christianity	2.170	0.201	0.661 - 7.125
Ethnicity			
Fante	2.555	0.378	0.318 - 20.545
Other Akan	2.482	0.395	0.306 - 20.130
Ga-Dangme	1.525	0.709	0.166 - 14.036
Northern tribes	2.524	0.424	0.260 - 24.471
Ewe	1.866	0.578	0.207 - 16.824

Antimicrobial Susceptibility of *S. aureus* isolates

The figure 1 below illustrates antimicrobial susceptibility. In this study, 145 *Staphylococcus aureus* were isolated and tested against some antibiotics (Penicillins, Cephalosporins, Aminoglycosides, Glycopeptides, Tetracyclines, and Macrolides). The outcome of the antimicrobial susceptibility testing is further categorized as susceptible, intermediate, and resistant.

The majority of the *S. aureus* isolates exhibited resistance against the penicillin classes. Thus, penicillin (n=141, 97.2%), and ampicillin (n=142, 97.9%). A few *S. aureus* developed resistance against Gentamicin (n=5, 3.4%) and Vancomycin (n=5, 3.4%), which were aminoglycosides and glycopeptides, respectively. Majority of the *S. aureus* isolates were susceptible to Gentamicin (n=140, 96.6%), Vancomycin (n=136, 93.8%) and Cloxacillin (n=126, 86.9%). The isolates resistant to more than three different classes of antibiotics were classified as multidrug-resistant (MDR). Of the 145 isolates, 54, representing 37.2%, were MDR, and 91(62.8%) were not multidrug-resistant isolates

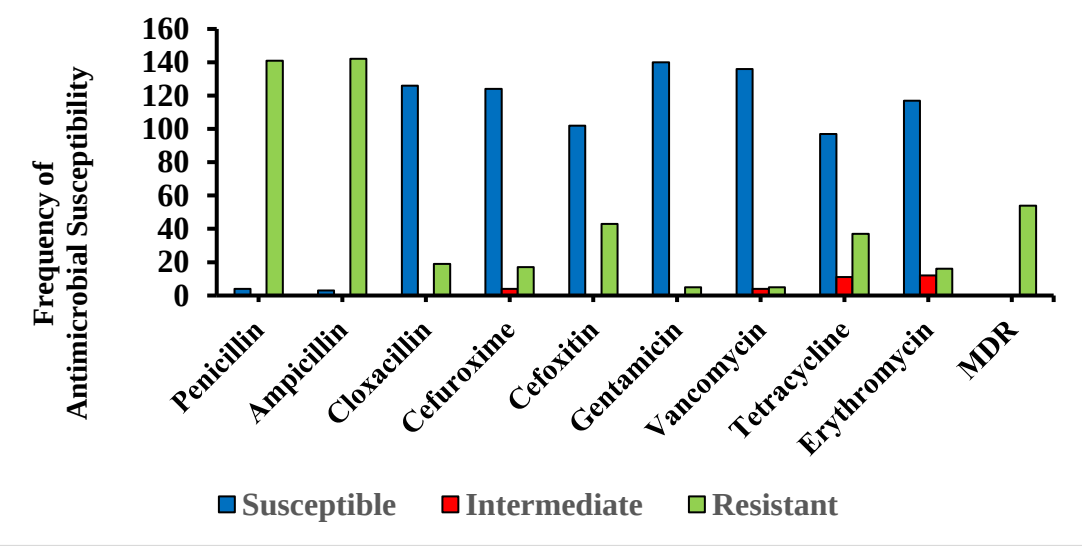


Figure 3: Antimicrobial Resistance Profile of *Staphylococcus aureus* Isolates

Antimicrobial Susceptibility testing for some multidrug-resistant *S. aureus* isolates

After the initial antimicrobial susceptibility testing exercise, some *S. aureus* (n=50, 34.5%) isolates were resistant to the selected antibiotics. This led to a further antimicrobial susceptibility testing process against new classes of antibiotics. These include; Cephalosporins, Aminoglycosides, Lincosamides, and Amphenicols. The table below illustrates further antimicrobial susceptibility testing conducted for *S. aureus* (n=50, 34.5%) isolates, which were initially resistant to multiple antibiotics.

A total of 50 *S. aureus* isolates resistant to more than three classes of antibiotics out of 145 Staphylococcus isolates were isolated randomly and tested against some selected antibiotics. Majority of these fifty (50) multidrug-resistant *S. aureus* isolates were resistant to Ceftazidime (n=50, 100%), Chloramphenicol (n=30, 60.0%). Most of those susceptible to these antibiotics were Amikacin (n=31, 62.0%) and Clindamycin (n=22, 44.0%).

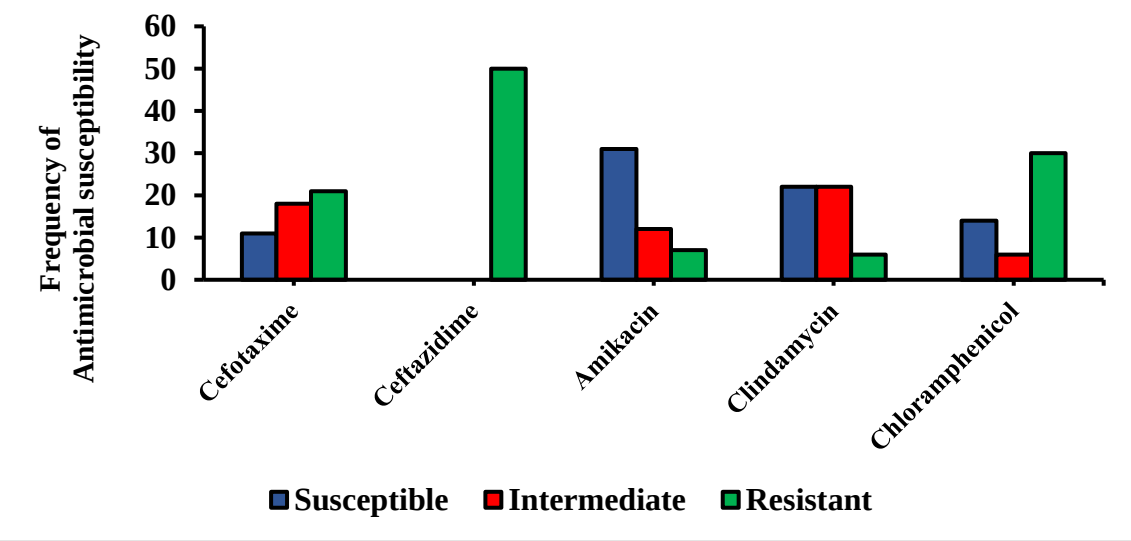


Figure 4: Antimicrobial Resistance Profile of 50 randomly selected MDR *Staphylococcus aureus* Isolates.

From Table 5, the number of isolates confirmed to be MDR *S. aureus was* 54, representing 37.2%, while the isolates not MDR *S. aureus*, were 91 (62.8%). MDR *S. aureus* carriage prevalence was highest among 49–60 months, in which 27.7% carried MDR *S. aureus* in their nasopharynx, whereas 5.5% in the 61–72 months age group showed the least prevalence. The standard deviation was 16.8, and the median age was 36.29 months. Association was found between MDR *S. aureus* and age in months type of facility (p = 0.054). However, no significant association between MDR *S. aureus* and religious affiliation (p = 0.901), ethnicity (p = 0.584), and sex (p = 0.749) was found.

Table 5: Bivariate Analysis Related to *Multidrug-resistant Staphylococcus aureus* Colonization.

Criteria	*Multidrug-resistant Staphylococcus aureus* Colonization				X^2 value	p-value
	Not Multidrug-resistant		Multidrug-resistant			
	Frequency	Percentage	Frequency	Percentage		
Age in months					1.155	0.949
0 – 12	7	7.7	5	9.3		
13 – 24	20	21.8	10	18.5		
25 – 36	21	23.1	14	26.0		
37 – 48	15	16.5	8	14.8		
49 – 60	22	24.2	15	27.7		
61 – 72	6	6.6	2	3.7		
Sex					0.102	0.749
Male	48	52.7	27	50.0		
Female	43	47.3	27	50.0		
Facility					5.853	0.054
Public Schools	19	20.9	14	26.0		
Private Schools	62	68.1	27	50.0		
Immunization centers	10	11.0	13	24.0		
Location of facility					13.118	0.157
Amamoma	9	9.9	10	18.5		
Kwaprow	3	3.3	3	5.5		
Akotokyir	2	2.2	1	1.9		
UCC	10	11.0	9	16.7		
Aboom Wells	7	7.7	7	13.0		
Pedu	0	0	1	1.9		
Abura	47	51.6	13	24.0		

Criteria	*Multidrug-resistant Staphylococcus aureus* Colonization				X^2 value	p-value
	Not Multidrug-resistant		Multidrug-resistant			
	Frequency	Percentage	Frequency	Percentage		
Ansapetu	5	5.5	5	9.3		
Bakano	2	2.2	2	3.7		
Kukuado	6	6.6	3	5.5		
Religion					0.015	0.901
Christianity	82	90.1	49	90.7		
Islam	9	9.9	5	9.3		
Ethnicity					3.763	0.584
Fante	45	49.5	23	42.6		
Other Akan	25	27.4	20	37.0		
Ga-Dangme	5	5.5	2	3.7		
Northern tribes	10	11.0	6	11.1		
Ewe	6	6.6	2	3.7		
Foreign tribes	0	0	1	1.9		
Total	**91**	**62.8**	**54**	**37.2**		

Table 6: Multivariate Analysis Related to *Multidrug-resistant Staphylococcus aureus* Colonization.

Criteria	OR	p-value	95% CI
Age in months			
0 – 12	0.402	0.472	0.034 - 4.810
13 – 24	0.626	0.650	0.082 - 4.749
25 – 36	1.280	0.800	0.191 - 8.587
37 – 48	1.116	0.912	0.161 - 7.728
49 – 60	1.160	0.883	0.161 - 8.370
Sex			
Female	0.643	0.250	0.303 - 1.363
Facility	1.574	0.257	0.719 - 3.445
Location of facility	0.897	0.089	0.792 - 1.016
Religion			
Christianity	2.094	0.641	0.093 - 46.940

Antimicrobial Resistance Profile of MRSA and MSSA

From Table 7, the number of isolates confirmed to be MSSA *was* 102, representing 70.3%, while MRSA was 43 (29.7%). MRSA carriage prevalence was

highest among 49–60 months, in which 25.5% carried MRSA in their nasopharynx, and 25.5% of that age group were MSSA carriers. 4.7% in the 61–72 months age group showed the least MRSA prevalence. From the table, it was observed that MRSA colonization ascended with increasing age. Association was found between MRSA and type of facility and the locations of the facilities (p = 0.002) and (p=0.010), respectively. However, no significant association between MRSA and religious affiliation (p = 0.602), ethnicity (p = 0.562), and sex (p = 0.123) were found.

Table 7: Bivariate Analysis Related to *Methicillin-resistant Staphylococcus aureus* Colonization.

Criteria	*Methicillin-resistant Staphylococcus aureus* Colonization				X2 value	p-value
	MRSA		MSSA			
	Frequency	Percentage	Frequency	Percentage		
Age in months					4.621	0.464
0 – 12	5	11.6	7	6.9		
13 – 24	6	14.0	24	23.5		
25 – 36	9	20.9	26	25.5		
37 – 48	10	23..3	13	12.7		
49 – 60	11	25.5	26	25.5		
61 – 72	2	4.7	6	5.9		
Sex					2.382	0.123
Male	18	41.9	57	55.9		
Female	25	58.1	45	44.1		
Facility					12.34	0.002
Private Schools	15	34.9	18	17.6		
Public Schools	17	39.5	72	70.6		
Immunization centers	11	25.6	12	11.8		
Location of facility					21.681	0.010
Amamoma	9	20.8	10	9.8		
Kwaprow	2	4.7	4	3.9		
Akotokyir	1	2.3	2	1.9		
UCC	7	16.3	12	11.8		
Aboom Wells	8	18.6	6	5.9		
Pedu	1	2.3	0	0		
Abura	7	16.3	53	52.0		
Ansapetu	4	9.3	6	5.9		
Bakano	2	4.7	2	1.9		
Kukuado	2	4.7	7	6.9		

Criteria	*Methicillin-resistant Staphylococcus aureus* Colonization				X2 value	p-value
	MRSA		MSSA			
	Frequency	Percentage	Frequency	Percentage		
Religion					0.273	0.602
Islam	5	11.6	9	8.8		
Christianity	38	88.4	93	91.2		
Ethnicity					3.912	0.562
Fante	19	44.2	49	48.0		
Other Akan	15	34.9	30	29.4		
Ga-Dangme	2	4.7	5	4.9		
Northern tribes	5	11.6	11	10.8		
Ewe	1	2.3	7	6.9		
Foreign tribes	1	2.3	0	0		
Total	**43**	**29.7**	**102**	**70.3**		

Table 8: Multivariate Analysis Related to *Methicillin-resistant Staphylococcus aureus* Colonization.

Criteria	OR	p-value	95% CI
Age in months			
0 – 12	1.788	0.661	0.133 - 23.981
13 – 24	4.417	0.179	0.505 - 38.601
25 – 36	1.900	0.526	0.262 - 13.792
37 – 48	0.752	0.773	0.109 - 5.213
49 – 60	2.506	0.384	0.316 - 19.854
Sex			
Female	2.074	0.083	0.908 - 4.739
Facility	0.892	0.788	0.389 - 2.045
Location of facility	1.214	0.006	1.057 - 1.395
Religion			
Christianity	0.735	0.603	0.231 - 2.338

Table 9: Antibiotic Susceptibility Patterns of *Methicillin-resistant Staphylococcus aureus* (n = 43) and *Multidrug-resistant Staphylococcus aureus* (n = 54) to some Selected Antimicrobials.

Antimicrobial agents	Methicillin-resistant *Staphylococcus aureus* Colonization MRSA		p-value	Multidrug-resistant *Staphylococcus aureus* Colonization MDR		p-value
	N	%		N	%	
Penicillin			0.188			0.118
Resistant	43	100.0		54	100.0	
Susceptible	0	0.0		0	0.0	
Ampicillin			0.256			0.178
Resistant	43	100.0		54	100.0	
Susceptible	0	0.0		0	0.0	
Cefoxitin			-			**< 0.001**
Resistant	-	-		41	76.0	
Susceptible	-	-		13	24.0	
Cloxacillin			**< 0.001**			**< 0.001**
Resistant	18	41.9		18	33.3	
Susceptible	25	58.1		36	66.7	
Cefuroxime			**< 0.001**			< 0.001
Resistant	17	39.6		17	31.5	
Susceptible	22	51.2		33	61.1	
Gentamicin			0.131			< 0.001
Resistant	3	7.0		4	7.4	
Susceptible	40	93.0		50	92.6	
Vancomycin			**< 0.001**			< 0.001
Resistant	4	9.3		5	9.3	
Susceptible	35	81.4		45	83.3	
Tetracycline			**0.047**			< 0.001
Resistant	14	32.6		24	44.4	
Susceptible	23	53.5		21	38.9	
Erythromycin			**0.001**			< 0.001
Resistant	10	23.3		13	24.1	
Susceptible	27	62.8		31	57.4	
MDR	41	95.3	**< 0.001**	-	-	-
Total	**43**	**29.7**		**54.0**	**37.2**	

Molecular detection of antibiotic-resistant genes

PCR analysis showed a relationship between antibiotic resistance's phenotypic and genotypic expression. Of the 16 erythromycin-resistant isolates, 10.0% harboured the *ermC*. However, the *ermB* gene was not detected in this study. Among the 141 penicillin-resistant isolates, 94 (67.1%) possessed the *blaZ* gene.

Of the 37 isolates that tested positive for tetracycline resistance, 24(17.1%) showed the *tetM* gene's presence. Fourteen (10%) of the isolates possessed the *mecA*.

Table 10: Prevalence of Genes encoding Antibiotic Resistance

Antibiotic Resistance Genes	Prevalence		Total number of Samples run based on phenotype resistance
	Present N (%)	Absent N (%)	
tetK	24 (17.1)	116 (82.9)	140
mecA	14 (10)	126 (90.0)	140
blaZ	94 (67.1)	46 (32.9)	140
ermC	7 (10)	63 (90)	70
vanB	1 (2.4)	40 (97.6)	41

Discussion

In this study, we aimed to identify the carriage rate, antibiotic resistance profiles, and antibiotic resistance and virulence genes of *Staphylococcus aureus* isolated from the nasopharynx of children living in Cape Coast, Ghana. The current study shows the distribution of *S. aureus* among healthy children in Cape Coast and its antibiotic resistance profiles. Resistance patterns to selected antibiotics and the prevalence of multi-drug resistance carriage were discovered.

Staphylococcus aureus infections are widespread in both adult and paediatric populations. After decades of rising prevalence, the percentage of *S. aureus* infections caused by methicillin-resistant *S. aureus* is declining in adults (Sutter et al., 2016). Children's populations also have fewer statistics available. *S. aureus* is an important pathogen that infects both humans and animals. It is also becoming frighteningly more resistant to most antimicrobial drugs now on the market. Due to its proficiency in gaining mechanisms for developing antibiotic resistance and harmful determinants, *S. aureus* is one of the most effective and

adaptable human infections. As a result, it has emerged in both nosocomial and community settings (Akanbi et al., 2017).

As far as we know, this research is the first to apply a combination of phenotypic and genotypic techniques concurrently to ascertain the prevalence and antibiotic resistance profiles of *S. aureus* in the study region. Eight hundred and eighty apparently healthy children aged 2 to 72 months were included in this research. The children were recruited from 11 communities across Cape Coast. Of the 880 participants, 50.2 % were males, and the remaining 49.8 % were females similar to a study by Dayie et al. in Ghana (Dayie et al., 2021). The majority of the participants, 25.1 %, were between the ages of 25 and 36 months. Of the 880 participants included in this study, 145 were culture positive for *S. aureus*. Thus, the prevalence of *S. aureus* nasopharyngeal carriage was 16.5% (95% Confidence Interval [CI]: 0.141% – 0.191%), which, compared to prior research by Nsofor et al. in 2015 and Garoy et al. in 2019, is much lower (Garoy et al., 2019; Nsofor et al., 2015). In the present investigation, *S. aureus* was primarily found in males (51.7% of cases). The sex of the recruited individuals and the prevalence of *S. aureus* isolation in the research region at the time of the investigation were not correlated. These results agree with that from Nsofor et al. in Nigeria. (51.3%) of the (56.3%) of children in their research who tested positive for *S. aureus* were male, compared to (48.7%) of the females. Additionally, the findings showed that age ($P< 0.05$) was a significant risk factor for *S. aureus* carriage, but sex was not (Nsofor et al., 2015). Age and gender, however, substantially impacted the colonization of *S. aureus* in Germany (Neidhart et al., 2018). *S. aureus* carriage

varies according to place, race, sex, anatomical site, population under study, and the presence of underlying diseases (Sollid et al., 2014). Additionally, this study discovered a relationship ($p < 0.001$) between *S. aureus* colonization, the facilities where samples were collected, and their locations at the time of this investigation

The prevalence of *S. aureus* nasopharyngeal carriage appeared to increase with age in this study. However, in a study conducted in Nigeria, a contrary observation was made that nasal carriage declined with age (Nsofor et al., 2015). A respiratory pathogen's colonization may reach its apex at a younger age (Nsofor et al., 2015). *Pneumococci, Haemophilus influenzae, Moraxella catarrhalis,* and *S. aureus* are a few examples of pathogens competing for colonization of the anterior nares at this stage. Bacterial interference, which is a phenomenon wherein the colonization of one bacterial strain blocks the colonization of another, is crucial in favoring or eradicating a particular bacterial strain (Sivaraman et al., 2009).

To help guide the treatment of patients with infections, one of the goals of this study is to identify the antibiotic susceptibility patterns of the recovered isolates. Numerous studies have demonstrated regional differences in resistance to routinely used antibiotics (Guo et al., 2020). Nevertheless, it has been determined that over 90% of *S. aureus* strains are penicillin-resistant (Appiah et al., 2020; Gurung et al., 2020). *S. aureus* has exhibited a high degree of penicillin resistance, for example, in Africa. However, low penicillin sensitivity has been discovered in India (Bhave et al., 2016; Schaumburg et al., 2014). Individual *S. aureus* resistance to penicillin (97.2%) and ampicillin (97.9%) was high among the isolates tested in this study. High resistance to these β-lactam drugs was not unexpected given that

penicillin has been losing its effectiveness against *S. aureus* since the 1960s, and ampicillin is one of the most frequently used antibiotics for treating infections in people and animals (Akanbi et al., 2017). Additionally, isolates resistant to ampicillin may also be resistant to other beta-lactams (Akindolire, 2013). Our study showed this when resistance to ampicillin and penicillin was equally prevalent. Penicillin was earlier introduced in Ghana for the treatment of bacterial illnesses. However, this medication is no longer effective because of the emergence of resistance. This demonstrates the frequent usage of this antibiotic, which can be ascribed to the ease with which antibiotics can be acquired from pharmacies, whether with or without a prescription (Donkor et al., 2012; Labi et al., 2018). Consequently, they may have been severely overused over a period of time.

The erythromycin, chloramphenicol, and tetracycline resistances found in this study were comparable to those that Kotey et al. had previously reported in 2022 (Kotey et al., 2022). The high levels of sensitivity to erythromycin, gentamicin, and tetracycline, 80.7%, 96.6%, and 66.9%, respectively, are consistent with the outcomes of previous research accomplished in Ghana. It is comparable to what was discovered among *S. aureus* nasal carriers by Dayie et al. in 2021 and Eibach et al. in 2017 (Dayie et al., 2021; Eibach et al., 2017).

Since 1966, clindamycin has been used extensively to treat Staphylococcal infections. It offers several benefits, including excellent bone penetration and long-lasting efficacy against bacterial adhesion and biofilm development (Bonnaire et al., 2021). The development of antibiotic resistance is impeding the effective treatment of *S. aureus*. Clindamycin is a superior option since it is less expensive

and has fewer side effects (Thapa et al., 2021). Clindamycin sensitivity was shown to be relatively high in this study, similar to findings from Egypt and India (Ahmed et al., 2020; Bajaj et al., 2015). The findings from the investigations by Dayie et al. and Eibach et al., which found that *S. aureus* was entirely susceptible to clindamycin, contradicted this study's findings (Dayie et al., 2021; Eibach et al., 2017). As a result, it may be inferred that clindamycin is still effective in Ghana for treating staphylococcal infections. Nevertheless, clindamycin resistance was present in 22 out of 50 isolates (44.0%), which is evident in the drug's infrequent usage. A research conducted in Nepal revealed a comparable frequency of 41.8% (Adhikari et al., 2017). However, this indicates that a significant resistance might emerge fast, and it is crucial to use this antibiotic carefully to avoid this. Thapa et al. claim that the isolates might progressively produce constitutively resistant mutations during clindamycin therapy, both in vitro and in vivo. Some patients experience therapy failure as a result of this. Therefore, identifying these resistant phenotypes is crucial to reducing treatment failure (Thapa et al., 2021). The majority of bacteria are known to acquire resistance to very affordable and frequently used antibiotics. A possible reason for the observed heterogeneity is that different nations have different antibiotic prescribing policies.

From *Streptomyces orientalis*, vancomycin was first isolated in 1957 (Rubinstein & Keynan, 2014). Vancomycin is used as a last resort for treating MRSA infections, despite reports of diminished susceptibility in a small number of instances (Cong et al., 2020b). Additionally, it is utilized to treat severe gram-positive infections in people with semi-synthetic penicillin allergies (Rubinstein &

Keynan, 2014). Vancomycin exhibits a "slow bactericidal" effect against MSSA compared to beta-lactams, claim Rubinstein & Keynan in 2014. The poor clinical outcomes of MSSA bacteremia and pneumonia cases treated with vancomycin are also a consequence of this delayed action (Rubinstein & Keynan, 2014). The *S. aureus* isolates in this study had a 93.8% vancomycin susceptibility rate, consistent with reports from Ghana, India, and China, where all of their isolates had a high vancomycin susceptibility rate (Eibach et al., 2017; Gu et al., 2015; Gurung et al., 2020). However, vancomycin resistance among MRSA isolates was shown to be more prevalent (44.5%) in Nigeria (Maharjan et al., 2021). Vancomycin, sulphonamide, and polymyxin were among the few antibiotics Donkor et al. said were seldom used for self-medication. This outcome is in line with Ghana's limited supply and the rare usage of this prescribed antibiotic (Donkor et al., 2012; Labi et al., 2018). The study by Serra-Burriel et al. found that it is also challenging to eradicate vancomycin-resistant *S. aureus* after it has colonized a hospital and a community. In the future, the hospital setting may potentially be a source of nosocomial illnesses. (Serra-Burriel et al., 2020).

Methicillin-resistant *S. aureus* (MRSA) infections are now linked to high mortality and morbidity and place a tremendous financial strain on the world's constrained healthcare resources because they are more challenging and expensive to treat than infections caused by methicillin-susceptible *S. aureus* (MSSA) strains (Kong et al., 2016). In this study, 70.3% of the *S. aureus* isolates were susceptible to Cefoxitin, while 29.7% were resistant. In contrast to classical methicillin resistance, caused by the *mecA* gene producing an altered penicillin-binding protein

(PBP2a), cefoxitin resistance is used in this study to assess MRSA, given that cefoxitin has a strong inducing effect on the *mecA* regulatory mechanism (Skov et al., 2014). The reported resistance to Cefoxitin (MRSA) is higher than the reports of 1.5% by Gorwitz et al. in the United States and Olonitola et al. 14.85% in Zaria, Nigeria, from anterior nares of a healthy population, adults, and schoolchildren, respectively (Gorwitz et al., 2008; Olonitola et al., 2007). According to MRSA research that has already been published, there is a significant regional difference in the prevalence of the infection within and across nations (Garoy et al., 2019). Hence, according to Schaumburg et al., the prevalence of MRSA among *S. aureus* isolates in Africa has been found to be less than 50% (Schaumburg et al., 2014) which is consistent with the current study. MRSA carriage was 21% among children living with HIV (CLWH) in the Botswana research, with two nasal swabs collected at different times (Reid et al., 2017). However, substantially greater MRSA prevalence rates have been noted in various sub-Saharan African settings: in Algeria, 75% of sample types came from surgical wounds (Rebiahi et al., 2011), 78% from cancer patients in Egypt (Ashour & El-Sharif, 2007), 73.8% from Nigeria (Udobi et al., 2013), and 82% from Rwanda (Ntirenganya et al., 2015), where multiple sample types were collected. Other regions of the world have also reported experiencing high MRSA prevalence, including Peru (80%) and Colombia (90%) (Guzmán-Blanco et al., 2009; Jiménez et al., 2012). There are a number of variables that contribute to the intra- and inter-national heterogeneity in MRSA prevalence. These include variations in research design, types of the specimen, laboratory techniques, study population, infection control practices in the various settings, and

duration of the study, among other things (Deyno et al., 2017). For instance, some studies solely employ DNA-based approaches, such as multilocus sequence typing (MLST), whole genome sequencing, microarrays, conventional PCR, and spa-typing. In contrast, others only use phenotypic methods to identify MRSA (Garoy et al., 2019).

Beta-lactam medications, in particular, have been recognized to be ineffective against MRSA worldwide (Kahsay et al., 2014). It is not unexpected that numerous isolates have been found to be multi-drug resistant (MDR). In this study, the prevalence of multi-drug resistant *S. aureus* isolates from the anterior nares of healthy individuals was relatively high, at 37.2%, of which 76.0% were MRSA. In comparison to the study done in Nigeria's Niger Delta, a greater prevalence of 52.5% was found in their research (Onanuga & Temedie, 2011). These findings raise serious concerns since other investigations have demonstrated that *S. aureus* carriage in the nasal cavity plays a significant role in the pathogenesis of infection (von Eiff et al., 2001). This raises concerns since these MDR isolates could spread to other organisms and non-carriers, resulting in further dispersal in the communities (Sakr et al., 2018). Pharmacies in Ghana and many other African nations selling antibiotics without a prescription may contribute to the increase of MDR. Self-medication is, therefore, a widespread behavior among people (Donkor et al., 2012). Additionally, the majority of individuals do not finish their medication, which results in antibiotic resistance. These findings provide a clear picture of the overuse of antibiotics in Ghana and offer insight into the country's developing antibiotic resistance trend. Currently, in Ghana, there is a pressing need for a

national commitment to address the issue of antibiotic abuse and resistance. This would necessitate extensive health education focusing on behavior modification and stringent guidelines about the inappropriate use of antibiotics when necessary (Donkor et al., 2010, 2012).

In several investigations carried out in Ghana by Egyir et al. and Sampane-Donkor et al., low multidrug resistance prevalence rates were noted, with values of 6.0% and 16.7%, respectively (Egyir et al., 2014; Sampane-Donkor et al., 2017). Their finding was in line with that made in the research by Dayie et al. at a rate of 3.2%, which is notably low in the same nation (Dayie et al., 2021). Dayie et al. assert that the observed low proportions of MRSA and MDR in the above studies can be attributed to the low consumption of antimicrobials like fluoroquinolones such as levofloxacin or ciprofloxacin and third-generation cephalosporins such as ceftazidime in the community setting in Ghana, as a result of their relatively high costs and typical prescription as therapeutic agents for acute infections. MRSA isolation is accelerated by exposure to these classes of antibiotics (Dayie et al., 2021; Egyir et al., 2014).

The *nuc* gene served as the housekeeping gene in this study for identifying S. *aureus* molecularly using PCR. The *nuc* gene serves as the standard for identifying and classifying *S. aureus,* and in the current study, the *nuc* gene fragment measured 270 bp. The isolates of *S. aureus* in this investigation demonstrated increased resistance to various antibiotics, including the beta-lactam medications tetracycline, gentamycin, and erythromycin.

The *blaZ* gene generates the beta-lactamase enzyme, which provides resistance to beta-lactam drugs like penicillin and ampicillin (Siu et al., 2022). Mutations in the *blaZ* gene were present in 1 percent of the 141 isolates that showed phenotypic penicillin resistance. The *mecA* gene was identified by molecular validation in only 14 MRSA isolates, representing 10% of those discovered. When this gene is present, a protein called penicillin-binding protein 2a (PBP2a), essential for methicillin resistance in staphylococci, is produced. This protein has a decreased affinity for beta-lactam antibiotics (Akanbi et al., 2017).

The efficacy of macrolides in treating staphylococcal infections can be questioned because of the existence of *erythromycin ribosomal methylase (erm)* genes. The *ermA* and *ermC* genes have been described as the most often identified *erm* genes linked with staphylococci in human infections. They code for erythromycin-resistant methylase, which primarily facilitates erythromycin resistance in staphylococci (Miklasińska-Majdanik, 2021). The distribution of genes affecting resistance to macrolide antibiotics, according to Miklasiska-Majdanik, is location-dependent. Of interest, China and Egypt are the two countries where the *ermB* gene is isolated the most frequently. Conversely, the *ermA* gene is more prevalent in South America. The *ermA* gene is isolated with a frequency equivalent to that of the *ermC* gene, which is the dominant gene in Europe (Miklasińska-Majdanik, 2021). However, the outcomes of our analysis identified *ermC* as the sole gene encoding erythromycin resistance. The *ermC* gene was found in 10% of the 16 isolates that were phenotypically resistant to erythromycin.

In many regions of the world, tetracyclines continue to be the first-line therapy for numerous illnesses (Emaneini et al., 2013) such as pneumonia, actinomycosis, chlamydial infections, syphilis, and traveler's diarrhea (Shutter & Akhondi, 2022b). Tetracycline resistance genes (*tet*) are present in most tetracycline-resistant bacteria. This study's findings indicate that *tetK* mediates the prevalence of the tetracycline resistance pathway. Through a resistance mechanism called tetracycline efflux, the *tetK* gene guard's bacteria against tetracycline. By generating a cytoplasmic-membrane protein that pumps tetracycline out of the cell more quickly than it enters, this procedure stops tetracycline from accumulating inside bacterial cells (Hatem et al., 2022). Of the 37 isolates that tested positive for tetracycline resistance, 24 (17.1%) showed the *tetK* gene presence. Similar studies assessing the prevalence of *tetK* genes in Malaysia showed a rate of 38.4%, which is greater than the present research (Ong et al., 2017). With a significantly greater prevalence of 78%, a similar trend was observed in Pakistan (Ullah et al., 2013).

Only a few cases of *vancomycin-resistant Staphylococcus aureus* (VRSA) were reported over the course of the previous decade, and it did not spread significantly. There have been 33 instances of van-type VRSA recorded globally as of the end of 2012 (Saadat et al., 2014). In the current study, the presence of *vanA* in one of the samples was discovered using PCR screening for vancomycin resistance loci. The study by Banerjee & Anupurba revealed the presence of *vanA* in two of the isolates. Additionally, quantitative real-time PCR results indicated that these isolates did not express the *vanA* gene (Banerjee & Anupurba, 2012).

Study limitation

A range of virulence factors in *S. aureus* may allow it to colonize its host and infect it with the disease. The genes for enterotoxin, leukocidin, and toxin, which allow invasion and tissue obliteration in hosts, have been discovered through several research (Cheung et al., 2021; Egyir et al., 2021). Leukocidins (E and D), PVL, *tst* gene, and γ toxins (*hlgA, hlgB,* and *hlgC*) have all been linked to increased virulence, predicting poorer clinical outcomes. As a result, it is essential to appropriately identify genes in order to limit and monitor their dissemination and guide infection prevention strategies (Egyir et al., 2021). One possible limitation of this research was that it could not determine how virulence was distributed throughout the population under study.

Chapter Summary

The study revealed that the prevalence of *S. aureus* nasopharyngeal carriage was 16.5%, which was measured in 880 apparently healthy school children from 11 communities across Cape Coast. There was a significant association between *S. aureus* and age in months ($p = 0.003$) and *S. aureus* and facility ($p = 0.001$). The majority of *S. aureus* isolates exhibited resistance to the penicillin classes. Of the 145 isolates, 54, representing 37.2%, were MDR, and the MRSA was 43 representing 29.7%. *Staphylococcus aureus* isolates that tested positive for genotypes had more excellent resistance rates to related phenotypes than negative isolates, suggesting that *S. aureus* isolates might be disseminated with both phenotypic and genotypic markers (He et al., 2020).

CHAPTER FIVE

SUMMARY, CONCLUSION AND RECOMMENDATION

Introduction

Staphylococcus aureus has garnered significant attention as a prominent bacterial pathogen with widespread implications for human health. It functions as a significant opportunistic pathogen, causing a range of mild to severe infections. Among carriers, children with nasopharyngeal carriage play a critical role in community transmission, particularly for methicillin-resistant strains. Overcrowding in schools and preschools has been identified as a prominent risk factor for nasal carriage in children, making them primary reservoirs for infections in susceptible individuals. Unfortunately, treating staphylococcal infections presents challenges due to the prevalence of multiple drug resistance (MDR), which has been observed in both carriage and clinical isolates in previous studies conducted in Ghana. The global dissemination of MDR strains further complicates the outcomes of *S. aureus* infections. To address these concerns, this research evaluated the carriage rate, antibiotic resistance profiles, and antibiotic susceptibility of *S. aureus* strains isolated from the nasopharynx of children. This was achieved using nasopharyngeal swabs from children of consenting parents in the archive. The *S. aureus* was detected using the appropriate culture methods and confirmed by the molecular detection of the *nuc* gene. The bacteria were later taken through antibiotic susceptibility testing using the disk diffusion method and further tested for resistance genes.

Summary

This is the first in-depth study of *S. aureus* prevalence, antibiotic sensitivity patterns and antibiotic resistance in children under the age of five from Cape Coast. The prevalence of *S. aureus* was high, according to our findings. The nasopharyngeal carriage rate was established at 16.5%. The age range with the most significant *S. aureus* carriage prevalence was 49–60 months. A correlation between *S. aureus* and facility, as well as age in months, was discovered.

The antimicrobial susceptibility of *S. aureus* infections has been found to have significantly risen, with enhanced sensitivity to erythromycin, gentamicin, and vancomycin and decreased sensitivity to penicillin, ampicillin, tetracycline, and chloramphenicol. This suggests that gentamicin can be used to treat *S. aureus* infections, and vancomycin is a potential option for usage as a last-resort medication to treat *S. aureus* and MRSA infections.

Isolates identified as MRSA and MSSA both displayed multidrug resistance. MRSA colonization increased with advancing age, and the prevalence of MRSA was 29.7%. There was; however, no conclusive evidence correlating MRSA to sex, ethnicity, or religion. MDR *S. aureus* prevalence was recorded to be 37.2%. Additionally, PCR results showed that *S. aureus* isolates in this study area have *ermC, blaZ,* and *mecA* resistance genes in their genetic makeup. PCR revealed an association between the phenotypic and genotypic expressions of antibiotic resistance. Thus, the results of this investigation provide critical information about the spread of *S. aureus* isolates from children's nasopharynxes in Cape Coast, Ghana, as well as profiles of antibiotic resistance.

One of the most significant factors influencing the development of antibiotic resistance is antibiotic use. Consequently, programs encouraging the prudent use of antibiotics may prove more effective than targeted screening based on risk factors, isolating carriers, and decolonizing populations. Simple hygiene practices like hand washing successfully stop the spread of resistant organisms in the population. Finally, to obliterate severe multidrug-resistant *S. aureus* infections in our communities, it is necessary to put mechanisms in place to eradicate the nasopharyngeal carriage of the bacteria.

Recommendations

This research, the first in the Central region of Ghana, provides a starting point for subsequent studies. In order to monitor and restrict the spread of staphylococcal infections in communities and hospitals, ongoing monitoring and infection control procedures must be implemented. This research recommends that the government and various institutions take action to raise public awareness of antibiotics, particularly among those without a health professional or academic background in various communities. We also recommend that parents, especially those with young children, be targeted in antimicrobial stewardship initiatives. Elementary and secondary school curricula should incorporate educational objectives concerning antibiotics and antibiotic resistance. Clinical practitioners and prescribers should adhere strictly to treatment guidelines to avert antibiotic resistance in *S. aureus*. Hospitals must routinely screen for MRSA using phenotypic testing and antibiotic susceptibility evaluations, which can help decide how to treat infections effectively. Studies comparing *S. aureus* carriage in the nose and isolates

found in surgical site infections should also be conducted to ascertain whether these are the same strains and whether nasal decolonization is required prior to surgery in children.

Suggestions for further research

In order to provide a thorough examination of the dynamics of carriage, incidence rates, and length of carriage of sequence types, further work should take the form of a longitudinal study. Future research should evaluate the risk variables associated with carriage, pathogenicity, and resistance concerning *S. aureus* distribution. It is essential to examine the molecular causes of drug resistance to tetracycline, clindamycin, and erythromycin, mainly since erythromycin resistance necessitates sequencing the *ermC, ermB,* and *mef g*enes. The molecular features of circulating *S. aureus* genotypes should be investigated further utilizing whole genome sequencing. Finally, research studies should be carried out to understand better the state of virulence factors and the prevalent strains in hospitals and the community, including more communities, hospitals, a bigger sample size, and different organisms.

REFERENCES

Aarestrup, F. M., Larsen, H. D., Eriksen, N. H. R., Elsberg, C. S., & Jensen, N. E. (1999). Frequency of α- and β-haemolysin in Staphylococcus aureus of bovine and human origin. *APMIS*, *107*(1–6), 425–430. https://doi.org/10.1111/J.1699-0463.1999.TB01576.X

Abdulgader, S. M., Shittu, A. O., Nicol, M. P., & Kaba, M. (2015a). Molecular epidemiology of Methicillin-resistant Staphylococcus aureus in Africa: a systematic review. *Frontiers in Microbiology*, *6*(APR). https://doi.org/10.3389/FMICB.2015.00348

Abdulgader, S. M., Shittu, A. O., Nicol, M. P., & Kaba, M. (2015b). Molecular epidemiology of Methicillin-resistant Staphylococcus aureus in Africa: a systematic review. *Frontiers in Microbiology*, *6*(APR). https://doi.org/10.3389/FMICB.2015.00348

Abt, M. C., Osborne, L. C., Monticelli, L. A., Doering, T. A., Alenghat, T., Sonnenberg, G. F., Paley, M. A., Antenus, M., Williams, K. L., Erikson, J., Wherry, E. J., & Artis, D. (2012). Commensal Bacteria Calibrate the Activation Threshold of Innate Antiviral Immunity. *Immunity*, *37*(1), 158. https://doi.org/10.1016/J.IMMUNI.2012.04.011

Acton, D. S., Tempelmans Plat-Sinnige, M. J., Van Wamel, W., De Groot, N., & Van Belkum, A. (2009). Intestinal carriage of Staphylococcus aureus: how does its frequency compare with that of nasal carriage and what is its clinical impact? *European Journal of Clinical Microbiology & Infectious Diseases : Official Publication of the European Society of Clinical Microbiology*, *28*(2),

115–127. https://doi.org/10.1007/S10096-008-0602-7

Adegbola, R. A., DeAntonio, R., Hill, P. C., Roca, A., Usuf, E., Hoet, B., & Greenwood, B. M. (2014). Carriage of Streptococcus pneumoniae and other respiratory bacterial pathogens in low and lower-middle income countries: a systematic review and meta-analysis. *PloS One*, *9*(8). https://doi.org/10.1371/JOURNAL.PONE.0103293

Adhikari, R. P., Ajao, A. O., Aman, M. J., Karauzum, H., Sarwar, J., Lydecker, A. D., Johnson, J. K., Nguyen, C., Chen, W. H., & Roghmann, M. C. (2012). Lower antibody levels to Staphylococcus aureus exotoxins are associated with sepsis in hospitalized adults with invasive S. aureus infections. *The Journal of Infectious Diseases*, *206*(6), 915–923. https://doi.org/10.1093/INFDIS/JIS462

Adhikari, R. P., Shrestha, S., Barakoti, A., & Amatya, R. (2017). Inducible clindamycin and methicillin resistant Staphylococcus aureus in a tertiary care hospital, Kathmandu, Nepal. *BMC Infectious Diseases*, *17*(1), 1–5. https://doi.org/10.1186/S12879-017-2584-5/TABLES/4

Aggarwal, B. B. (2004). Nuclear factor-κB: The enemy within. *Cancer Cell*, *6*(3), 203–208. https://doi.org/10.1016/j.ccr.2004.09.003

Ahmed, E. F., Gad, G. F. M., Soliman, W. E., El-Asady, R. S., Hasaneen, A. M., & Abdelwahab, S. F. (2020). Prevalence of methicillin-resistant coagulase-negative staphylococci among Egyptian patients after surgical interventions. *Https://Doi.Org/10.1177/0049475520962740*, *51*(1), 40–44. https://doi.org/10.1177/0049475520962740

Akanbi, O. E., Njom, H. A., Fri, J., Otigbu, A. C., & Clarke, A. M. (2017). Antimicrobial Susceptibility of Staphylococcus aureus Isolated from Recreational Waters and Beach Sand in Eastern Cape Province of South Africa. *International Journal of Environmental Research and Public Health*, *14*(9). https://doi.org/10.3390/IJERPH14091001

Akindolire, M. A. (2013). *Detection and Molecular characterisation of Virulence Genes in Antibiotic Resistant Staphylococcus aureus from milk in the North West Province, South Africa*. https://repository.nwu.ac.za/handle/10394/16104

Al-Doori, Z., Morrison, D., Edwards, G., & Gemmell, C. (2003). Susceptibility of MRSA to triclosan. *The Journal of Antimicrobial Chemotherapy*, *51*(1), 185–186. https://doi.org/10.1093/JAC/DKG013

Alanis, A. J. (2005). Resistance to antibiotics: are we in the post-antibiotic era? *Archives of Medical Research*, *36*(6), 697–705. https://doi.org/10.1016/J.ARCMED.2005.06.009

Albrecht, V. S., Limbago, B. M., Moran, G. J., Krishnadasan, A., Gorwitz, R. J., McDougal, L. K., & Talan, D. A. (2015). Staphylococcus aureus Colonization and Strain Type at Various Body Sites among Patients with a Closed Abscess and Uninfected Controls at U.S. Emergency Departments. *Journal of Clinical Microbiology*, *53*(11), 3478. https://doi.org/10.1128/JCM.01371-15

Alekshun, M. N., & Levy, S. B. (2007). Molecular mechanisms of antibacterial multidrug resistance. *Cell*, *128*(6), 1037–1050.

https://doi.org/10.1016/J.CELL.2007.03.004

Alghaithy, A. A., Bilal, N. E., Gedebou, M., & Weily, A. H. (2000). Nasal carriage and antibiotic resistance of Staphylococcus aureus isolates from hospital and non-hospital personnel in Abha, Saudi Arabia. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, *94*(5), 504–507. https://doi.org/10.1016/S0035-9203(00)90066-X

Alibayov, B., Baba-Moussa, L., Sina, H., Zdeňková, K., & Demnerová, K. (2014). Staphylococcus aureus mobile genetic elements. *Molecular Biology Reports*, *41*(8), 5005–5018. https://doi.org/10.1007/S11033-014-3367-3

Allard, C., Carignan, A., Bergevin, M., Boulais, I., Tremblay, V., Robichaud, P., Duperval, R., & Pepin, J. (2008). Secular changes in incidence and mortality associated with Staphylococcus aureus bacteraemia in Quebec, Canada, 1991-2005. *Clinical Microbiology and Infection : The Official Publication of the European Society of Clinical Microbiology and Infectious Diseases*, *14*(5), 421–428. https://doi.org/10.1111/J.1469-0691.2008.01965.X

Amberpet, R., Sistla, S., Sugumar, M., Nagasundaram, N., Manoharan, M., & Parija, S. (2019). Detection of heterogeneous vancomycin-intermediate Staphylococcus aureus: A preliminary report from south India. *Indian Journal of Medical Research*, *150*(2), 194–198. https://doi.org/10.4103/IJMR.IJMR_1976_17

Ammam, F., Marvaud, J. C., & Lambert, T. (2012). Distribution of the vanG-like gene cluster in Clostridium difficile clinical isolates. *Canadian Journal of Microbiology*, *58*(4), 547–551. https://doi.org/10.1139/W2012-002

Ammerlaan, H. S. M., Kluytmans, J. A. J. W., Wertheim, H. F. L., Nouwen, J. L., & Bonten, M. J. M. (2009). Eradication of methicillin-resistant Staphylococcus aureus carriage: a systematic review. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *48*(7), 922–930. https://doi.org/10.1086/597291

Anderson, A. S., Scully, I. L., Timofeyeva, Y., Murphy, E., McNeil, L. K., Mininni, T., Nuñez, L., Carriere, M., Singer, C., Dilts, D. A., & Jansen, K. U. (2012). Staphylococcus aureus manganese transport protein C is a highly conserved cell surface protein that elicits protective immunity against S. aureus and Staphylococcus epidermidis. *The Journal of Infectious Diseases*, *205*(11), 1688–1696. https://doi.org/10.1093/INFDIS/JIS272

Appiah, V. A., Pesewu, G. A., Kotey, F. C. N., Boakye, A. N., Duodu, S., Tette, E. M. A., Nyarko, M. Y., & Donkor, E. S. (2020). Staphylococcus aureus Nasal Colonization among Children with Sickle Cell Disease at the Children's Hospital, Accra: Prevalence, Risk Factors, and Antibiotic Resistance. *Pathogens (Basel, Switzerland)*, *9*(5). https://doi.org/10.3390/PATHOGENS9050329

Arbeit, R. D., Karakawa, W. W., Vann, W. F., & Robbins, J. B. (1984). Predominance of two newly described capsular polysaccharide types among clinical isolates of Staphylococcus aureus. *Diagnostic Microbiology and Infectious Disease*, *2*(2), 85–91. https://doi.org/10.1016/0732-8893(84)90002-6

Arbeit, R. D., Maki, D., Tally, F. P., Campanaro, E., & Eisenstein, B. I. (2004).

The safety and efficacy of daptomycin for the treatment of complicated skin and skin-structure infections. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *38*(12), 1673–1681. https://doi.org/10.1086/420818

Archer, G. L. (1998). Staphylococcus aureus: A Well-Armed Pathogen. *Clinical Infectious Diseases*, *26*(5), 1179–1181. https://doi.org/10.1086/520289

Archer, G. L., & Bosilevac, J. M. (2001). Signaling antibiotic resistance in staphylococci. *Science (New York, N.Y.)*, *291*(5510), 1915–1916. https://doi.org/10.1126/SCIENCE.1059671

Arias, C. A., & Murray, B. E. (2012). The rise of the Enterococcus: beyond vancomycin resistance. *Nature Reviews. Microbiology*, *10*(4), 266–278. https://doi.org/10.1038/NRMICRO2761

Arias, C. A., Vallejo, M., Reyes, J., Panesso, D., Moreno, J., Castañeda, E., Villegas, M. V., Murray, B. E., & Quinn, J. P. (2008). Clinical and microbiological aspects of linezolid resistance mediated by the cfr gene encoding a 23S rRNA methyltransferase. *Journal of Clinical Microbiology*, *46*(3), 892–896. https://doi.org/10.1128/JCM.01886-07

Armstrong-Esther, C. A., & Smith, J. E. (1976). Carriage patterns of Staphylococcus aureus in a healthy non-hospital population of adults and children. *Annals of Human Biology*, *3*(3), 221–227. https://doi.org/10.1080/03014467600001381

Arthur, M., & Courvalin, P. (1993). Genetics and mechanisms of glycopeptide resistance in enterococci. *Antimicrobial Agents and Chemotherapy*, *37*(8),

1563–1571. https://doi.org/10.1128/AAC.37.8.1563

Aryee, A., & Edgeworth, J. D. (2016). Carriage, Clinical Microbiology and Transmission of. *Current Topics in Microbiology and Immunology*, *409*, 1–19. https://doi.org/10.1007/82_2016_5

Ashour, H. M., & El-Sharif, A. (2007). Microbial spectrum and antibiotic susceptibility profile of gram-positive aerobic bacteria isolated from cancer patients. *Journal of Clinical Oncology : Official Journal of the American Society of Clinical Oncology*, *25*(36), 5763–5769. https://doi.org/10.1200/JCO.2007.14.0947

Askarian, F., Wagner, T., Johannessen, M., & Nizet, V. (2018). Staphylococcus aureus modulation of innate immune responses through Toll-like (TLR), (NOD)-like (NLR) and C-type lectin (CLR) receptors. *FEMS Microbiology Reviews*, *42*(5), 656–671. https://doi.org/10.1093/FEMSRE/FUY025

Askarian, M., Zeinalzadeh, A., Japoni, A., Alborzi, A., & Memish, Z. A. (2009). Prevalence of nasal carriage of methicillin-resistant Staphylococcus aureus and its antibiotic susceptibility pattern in healthcare workers at Namazi Hospital, Shiraz, Iran. *International Journal of Infectious Diseases : IJID : Official Publication of the International Society for Infectious Diseases*, *13*(5). https://doi.org/10.1016/J.IJID.2008.11.026

Ateba Ngoa, U., Schaumburg, F., Adegnika, A. A., Kösters, K., Möller, T., Fernandes, J. F., Alabi, A., Issifou, S., Becker, K., Grobusch, M. P., Kremsner, P. G., & Lell, B. (2012). Epidemiology and population structure of Staphylococcus aureus in various population groups from a rural and semi

urban area in Gabon, Central Africa. *Acta Tropica*, *124*(1), 42–47. https://doi.org/10.1016/J.ACTATROPICA.2012.06.005

Atilano, M. L., Pereira, P. M., Yates, J., Reed, P., Veiga, H., Pinho, M. G., & Filipe, S. R. (2010). Teichoic acids are temporal and spatial regulators of peptidoglycan cross-linking in Staphylococcus aureus. *Proceedings of the National Academy of Sciences of the United States of America*, *107*(44), 18991–18996. https://doi.org/10.1073/PNAS.1004304107/SUPPL_FILE/PNAS.201004304SI.PDF

Bachert, C., Zhang, N., Holtappels, G., De Lobel, L., Van Cauwenberge, P., Liu, S., Lin, P., Bousquet, J., & Van Steen, K. (2010). Presence of IL-5 protein and IgE antibodies to staphylococcal enterotoxins in nasal polyps is associated with comorbid asthma. *The Journal of Allergy and Clinical Immunology*, *126*(5). https://doi.org/10.1016/J.JACI.2010.07.007

Bajaj, A., Kukanur, S., Kotigadde, S., & Meundi, M. (2015). Gram Positive Cocci Causing Surgical Site Infection: Identification and Antibiotic Susceptibility Pattern. *IOSR Journal of Dental and Medical Sciences*, *14*(8), 62–67. https://doi.org/10.9790/0853-14846267

Banerjee, T., & Anupurba, S. (2012). Colonization with vancomycin-intermediate Staphylococcus aureus strains containing the vanA resistance gene in a tertiary-care center in north India. *Journal of Clinical Microbiology*, *50*(5), 1730–1732. https://doi.org/10.1128/JCM.06208-11

BARBER, M. (1961). Methicillin-resistant staphylococci. *Journal of Clinical*

Pathology, *14*(4), 385–393. https://doi.org/10.1136/JCP.14.4.385

Barriere, S. L. (2015). Clinical, economic and societal impact of antibiotic resistance. *Expert Opinion on Pharmacotherapy*, *16*(2), 151–153. https://doi.org/10.1517/14656566.2015.983077

Bartlett, A. H., & Hulten, K. G. (2010). Staphylococcus aureus pathogenesis: secretion systems, adhesins, and invasins. *The Pediatric Infectious Disease Journal*, *29*(9), 860–861. https://doi.org/10.1097/INF.0B013E3181EF2477

Bartlett, J. G., Auwaerter, P. G., Pham, P. A., Johns Hopkins Medicine., & Johns Hopkins POC-IT Center. (2010). *The Johns Hopkins ABX guide : diagnosis and treatment of infectious diseases*. 856.

Baseri, N., Najar-Peerayeh, S., & Bagheri Amiri, F. (2018). Prevalence of vancomycin-intermediate Staphylococcus aureus among clinical isolates in Iran: A systematic review and meta-analysis. *Journal of Global Antimicrobial Resistance*, *15*, 178–187. https://doi.org/10.1016/J.JGAR.2018.06.018

Bayer, A. S., Schneider, T., & Sahl, H. G. (2013). Mechanisms of daptomycin resistance in Staphylococcus aureus: role of the cell membrane and cell wall. *Annals of the New York Academy of Sciences*, *1277*(1), 139–158. https://doi.org/10.1111/J.1749-6632.2012.06819.X

Bebell, L. M., Ayebare, A., Boum, Y., Siedner, M. J., Bazira, J., Schiff, S. J., Metlay, J. P., Bangsberg, D. R., Ttendo, S., & Firth, P. G. (2017). Prevalence and correlates of MRSA and MSSA nasal carriage at a Ugandan regional referral hospital. *Journal of Antimicrobial Chemotherapy*, *72*(3), 888–892.

https://doi.org/10.1093/JAC/DKW472

Beceiro, A., Tomás, M., & Bou, G. (2013). Antimicrobial resistance and virulence: a successful or deleterious association in the bacterial world? *Clinical Microbiology Reviews*, *26*(2), 185–230. https://doi.org/10.1128/CMR.00059-12

Becker, K., Heilmann, C., & Peters, G. (2014). Coagulase-negative staphylococci. *Clinical Microbiology Reviews*, *27*(4), 870–926. https://doi.org/10.1128/CMR.00109-13

Beining, P. R., Huff, E., Prescott, B., & Theodore, T. S. (1975). Characterization of the lipids of mesosomal vesicles and plasma membranes from Staphylococcus aureus. *Journal of Bacteriology*, *121*(1), 137–143. https://doi.org/10.1128/JB.121.1.137-143.1975

Bekeredjian-Ding, I., Stein, C., & Uebele, J. (2017). The Innate Immune Response Against Staphylococcus aureus. *Current Topics in Microbiology and Immunology*, *409*, 385–418. https://doi.org/10.1007/82_2015_5004

Berends, E. T. M., Horswill, A. R., Haste, N. M., Monestier, M., Nizet, V., & Von Köckritz-Blickwede, M. (2010). Nuclease expression by Staphylococcus aureus facilitates escape from neutrophil extracellular traps. *Journal of Innate Immunity*, *2*(6), 576–586. https://doi.org/10.1159/000319909

Berger-Bächi, B. (1994). Expression of resistance to methicillin. *Trends in Microbiology*, *2*(10), 389–393. https://doi.org/10.1016/0966-842X(94)90617-3

Berkley, J. A., Lowe, B. S., Mwangi, I., Williams, T., Bauni, E., Mwarumba, S.,

Ngetsa, C., Slack, M. P. E., Njenga, S., Hart, C. A., Maitland, K., English, M., Marsh, K., & Scott, J. A. G. (2005). Bacteremia among children admitted to a rural hospital in Kenya. *The New England Journal of Medicine*, *352*(1), 39–47. https://doi.org/10.1056/NEJMOA040275

Bernardo, K., Pakulat, N., Fleer, S., Schnaith, A., Utermöhlen, O., Krut, O., Müller, S., & Krönke, M. (2004). Subinhibitory concentrations of linezolid reduce Staphylococcus aureus virulence factor expression. *Antimicrobial Agents and Chemotherapy*, *48*(2), 546–555. https://doi.org/10.1128/AAC.48.2.546-555.2004

Berti, A. D., Theisen, E., Sauer, J. D., Nonejuie, P., Olson, J., Pogliano, J., Sakoulas, G., Nizet, V., Proctor, R. A., & Rose, W. E. (2016). Penicillin Binding Protein 1 Is Important in the Compensatory Response of Staphylococcus aureus to Daptomycin-Induced Membrane Damage and Is a Potential Target for β-Lactam–Daptomycin Synergy. *Antimicrobial Agents and Chemotherapy*, *60*(1), 451. https://doi.org/10.1128/AAC.02071-15

Berube, B. J., & Wardenburg, J. B. (2013). Staphylococcus aureus α-toxin: nearly a century of intrigue. *Toxins*, *5*(6), 1140–1166. https://doi.org/10.3390/TOXINS5061140

Besier, S., Ludwig, A., Zander, J., Brade, V., & Wichelhaus, T. A. (2008). Linezolid resistance in Staphylococcus aureus: gene dosage effect, stability, fitness costs, and cross-resistances. *Antimicrobial Agents and Chemotherapy*, *52*(4), 1570–1572. https://doi.org/10.1128/AAC.01098-07

Bhave, P., Kartikeyan, S., Ramteerthakar, M., & Patil, N. (2016). Bacteriological

study of surgical site infections in a tertiary care hospital at Miraj, Maharashtra state, India. *International Journal of Research in Medical Sciences*, 2630–2635. https://doi.org/10.18203/2320-6012.IJRMS20161922

Bjerketorp, J., Jacobsson, K., & Frykberg, L. (2004). The von Willebrand factor-binding protein (vWbp) of Staphylococcus aureus is a coagulase. *FEMS Microbiology Letters*, *234*(2), 309–314. https://doi.org/10.1016/J.FEMSLE.2004.03.040

Bjerketorp, J., Nilsson, M., Ljungh, Å., Flock, J. I., Jacobsson, K., & Frykberg, L. (2002). A novel von Willebrand factor binding protein expressed by Staphylococcus aureus. *Microbiology (Reading, England)*, *148*(Pt 7), 2037–2044. https://doi.org/10.1099/00221287-148-7-2037

Blair, J. M. A., Richmond, G. E., & Piddock, L. J. V. (2014). Multidrug efflux pumps in Gram-negative bacteria and their role in antibiotic resistance. *Future Microbiology*, *9*(10), 1165–1177. https://doi.org/10.2217/FMB.14.66

Blair, J. M. A., Webber, M. A., Baylay, A. J., Ogbolu, D. O., & Piddock, L. J. V. (2015). Molecular mechanisms of antibiotic resistance. *Nature Reviews. Microbiology*, *13*(1), 42–51. https://doi.org/10.1038/NRMICRO3380

Blázquez, J., Couce, A., Rodríguez-Beltrán, J., & Rodríguez-Rojas, A. (2012). Antimicrobials as promoters of genetic variation. *Current Opinion in Microbiology*, *15*(5), 561–569. https://doi.org/10.1016/J.MIB.2012.07.007

Block, C., Robenshtok, E., Simhon, A., & Shapiro, M. (2000). Evaluation of chlorhexidine and povidone iodine activity against methicillin-resistant Staphylococcus aureus and vancomycin-resistant Enterococcus faecalis using

a surface test. *The Journal of Hospital Infection*, *46*(2), 147–152. https://doi.org/10.1053/JHIN.2000.0805

Bogaert, D., Van Belkum, A., Sluijter, M., Luijendijk, A., De Groot, R., Rümke, H. C., Verbrugh, H. A., & Hermans, P. W. M. (2004). Colonisation by Streptococcus pneumoniae and Staphylococcus aureus in healthy children. *Lancet (London, England)*, *363*(9424), 1871–1872. https://doi.org/10.1016/S0140-6736(04)16357-5

Bokarewa, M. I., Jin, T., & Tarkowski, A. (2006). Staphylococcus aureus: Staphylokinase. *The International Journal of Biochemistry & Cell Biology*, *38*(4), 504–509. https://doi.org/10.1016/J.BIOCEL.2005.07.005

Boles, B. R., & Horswill, A. R. (2008). Agr-mediated dispersal of Staphylococcus aureus biofilms. *PLoS Pathogens*, *4*(4). https://doi.org/10.1371/JOURNAL.PPAT.1000052

Bolyard, E. A., Tablan, O. C., Williams, W. W., Pearson, M. L., Shapiro, C. N., & Deitchman, S. D. (1998). Guideline for infection control in healthcare personnel, 1998. Hospital Infection Control Practices Advisory Committee. *Infection Control and Hospital Epidemiology*, *19*(6), 407–463. https://doi.org/10.1086/647840

Bommer, J., Vergetis, W., Andrassy, K., Hingst, V., Borneff, M., & Huber, W. (1995). Elimination of Staphylococcus aureus in hemodialysis patients. *ASAIO Journal (American Society for Artificial Internal Organs : 1992)*, *41*(1), 127–131. https://doi.org/10.1097/00002480-199501000-00021

Bondi, A., & Dietz, C. C. (1945). Penicillin resistant staphylococci. *Proceedings*

of the Society for Experimental Biology and Medicine. Society for Experimental Biology and Medicine (New York, N.Y.), *60*(1), 55–58. https://doi.org/10.3181/00379727-60-15089

Bonnaire, A., Vernet-Garnier, V., Lebrun, D., Bajolet, O., Bonnet, M., Hentzien, M., Ohl, X., Diallo, S., & Bani-Sadr, F. (2021). Clindamycin combination treatment for the treatment of bone and joint infections caused by clindamycin-susceptible, erythromycin-resistant Staphylococcus spp. *Diagnostic Microbiology and Infectious Disease*, *99*(1), 115225. https://doi.org/10.1016/J.DIAGMICROBIO.2020.115225

Borchardt, S. A., Babwah, A. V., & Jayaswal, R. K. (1993). Sequence analysis of the region downstream from a peptidoglycan hydrolase-encoding gene from Staphylococcus aureus NCTC8325. *Gene*, *137*(2), 253–258. https://doi.org/10.1016/0378-1119(93)90016-V

Boucher, H. W., Talbot, G. H., Bradley, J. S., Edwards, J. E., Gilbert, D., Rice, L. B., Scheld, M., Spellberg, B., & Bartlett, J. (2009). Bad bugs, no drugs: no ESKAPE! An update from the Infectious Diseases Society of America. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *48*(1), 1–12. https://doi.org/10.1086/595011

Boyce, J. M., & Pittet, D. (2002). Guideline for Hand Hygiene in Health-Care Settings: Recommendations of the Healthcare Infection Control Practices Advisory Committee and the HICPAC/SHEA/APIC/IDSA Hand Hygiene Task Force. *Infection Control & Hospital Epidemiology*, *23*(S12), S3–S40. https://doi.org/10.1086/503164

Braff, M. H., Jones, A. L., Skerrett, S. J., & Rubens, C. E. (2007). Staphylococcus aureus exploits cathelicidin antimicrobial peptides produced during early pneumonia to promote staphylokinase-dependent fibrinolysis. *The Journal of Infectious Diseases*, *195*(9), 1365–1372. https://doi.org/10.1086/513277

Breurec, S., Zriouil, S. B., Fall, C., Boisier, P., Brisse, S., Djibo, S., Etienne, J., Fonkoua, M. C., Perrier-Gros-Claude, J. D., Pouillot, R., Ramarokoto, C. E., Randrianirina, F., Tall, A., Thiberge, J. M., Laurent, F., & Garin, B. (2011). Epidemiology of methicillin-resistant Staphylococcus aureus lineages in five major African towns: emergence and spread of atypical clones. *Clinical Microbiology and Infection*, *17*(2), 160–165. https://doi.org/10.1111/J.1469-0691.2010.03219.X

Brinkmann, V., Reichard, U., Goosmann, C., Fauler, B., Uhlemann, Y., Weiss, D. S., Weinrauch, Y., & Zychlinsky, A. (2004). Neutrophil extracellular traps kill bacteria. *Science (New York, N.Y.)*, *303*(5663), 1532–1535. https://doi.org/10.1126/SCIENCE.1092385

Brown, E. J. (1991). Complement receptors and phagocytosis. *Current Opinion in Immunology*, *3*(1), 76–82. https://doi.org/10.1016/0952-7915(91)90081-B

Brown, S., Xia, G., Luhachack, L. G., Campbell, J., Meredith, T. C., Chen, C., Winstel, V., Gekeler, C., Irazoqui, J. E., Peschel, A., & Walker, S. (2012). Methicillin resistance in Staphylococcus aureus requires glycosylated wall teichoic acids. *Proceedings of the National Academy of Sciences of the United States of America*, *109*(46), 18909–18914. https://doi.org/10.1073/PNAS.1209126109

Buckland, A. G., & Wilton, D. C. (2000). The antibacterial properties of secreted phospholipases A(2). *Biochimica et Biophysica Acta*, *1488*(1–2), 71–82. https://doi.org/10.1016/S1388-1981(00)00111-6

Buehlmann, M., Frei, R., Fenner, L., Dangel, M., Fluckiger, U., & Widmer, A. F. (2008). Highly Effective Regimen for Decolonization of Methicillin-Resistant Staphylococcus aureus Carriers. *Infection Control & Hospital Epidemiology*, *29*(6), 510–516. https://doi.org/10.1086/588201

Bukowski, M., Wladyka, B., & Dubin, G. (2010). Exfoliative toxins of Staphylococcus aureus. *Toxins*, *2*(5), 1148–1165. https://doi.org/10.3390/TOXINS2051148

Bullen, J. J. (1981). The significance of iron in infection. *Reviews of Infectious Diseases*, *3*(6), 1127–1138. https://doi.org/10.1093/CLINIDS/3.6.1127

Bush, K., & Jacoby, G. A. (2010). Updated functional classification of beta-lactamases. *Antimicrobial Agents and Chemotherapy*, *54*(3), 969–976. https://doi.org/10.1128/AAC.01009-09

Cadieux, B., Vijayakumaran, V., Bernards, M. A., McGavin, M. J., & Heinrichs, D. E. (2014). Role of lipase from community-associated methicillin-resistant Staphylococcus aureus strain USA300 in hydrolyzing triglycerides into growth-inhibitory free fatty acids. *Journal of Bacteriology*, *196*(23), 4044–4056. https://doi.org/10.1128/JB.02044-14

Camelo-Castillo, A., Henares, D., Brotons, P., Galiana, A., Rodríguez, J. C., Mira, A., Muñoz-Almagro, C., Andres-Franch, M., Bautista, C., Díaz, A., Esteva, C., De Sevilla, M. F., Fernandez, A., Garcia-Fernandez, M., Galles, C.,

Garcia, L., Garcia-Garcia, J. J., Hernandez-Bou, S., Launes, C., … Sellares, E. (2019). Nasopharyngeal microbiota in children with invasive pneumococcal disease: Identification of bacteria with potential disease-promoting and protective effects. *Frontiers in Microbiology*, *10*(JAN), 11. https://doi.org/10.3389/FMICB.2019.00011/BIBTEX

Candi, E., Schmidt, R., & Melino, G. (2005). The cornified envelope: a model of cell death in the skin. *Nature Reviews. Molecular Cell Biology*, *6*(4), 328–340. https://doi.org/10.1038/NRM1619

Cape Coast, Ghana — statistics 2022. (2022).

Carlet, J., Collignon, P., Goldmann, D., Goossens, H., Gyssens, I. C., Harbarth, S., Jarlier, V., Levy, S. B., N'Doye, B., Pittet, D., Richtmann, R., Seto, W. H., Van Der Meer, J. W., & Voss, A. (2011). Society's failure to protect a precious resource: antibiotics. *Lancet (London, England)*, *378*(9788), 369–371. https://doi.org/10.1016/S0140-6736(11)60401-7

Casanova, Natalia Gómez, María, Siller Ruiz, J. L. M. B. (2017). Mechanisms of resistance to Glycopeptides in Staphylococcus aureus. *Kasmera*, *38*(6), 36–44. http://seq.es/wp-content/uploads/2017/12/gomez25oct2017.pdf

CDC. (2013). *ANTIBIOTIC RESISTANCE THREATS in the United States*. Centers for Disease Control and Prevention.

Cepas, V., & Soto, S. M. (2020). Relationship between Virulence and Resistance among Gram-Negative Bacteria. *Antibiotics*, *9*(10), 1–11. https://doi.org/10.3390/ANTIBIOTICS9100719

Chain, E., & Duthie, E. S. (1940). Identity of Hyaluronidase and Spreading

Factor. *British Journal of Experimental Pathology*, *21*(6), 324. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2065381/

Chaiyakunapruk, N., Veenstra, D. L., Lipsky, B. A., & Saint, S. (2002). Chlorhexidine compared with povidone-iodine solution for vascular catheter-site care: a meta-analysis. *Annals of Internal Medicine*, *136*(11), 792–801. https://doi.org/10.7326/0003-4819-136-11-200206040-00007

Chamberlain, N. R., & Brueggemann, S. A. (1997). Characterisation and expression of fatty acid modifying enzyme produced by Staphylococcus epidermidis. *Journal of Medical Microbiology*, *46*(8), 693–697. https://doi.org/10.1099/00222615-46-8-693

Chambers, H. F. (1997). Methicillin resistance in staphylococci: molecular and biochemical basis and clinical implications. *Clinical Microbiology Reviews*, *10*(4), 781–791. https://doi.org/10.1128/CMR.10.4.781

Chambers, H. F., & DeLeo, F. R. (2009). Waves of Resistance: Staphylococcus aureus in the Antibiotic Era. *Nature Reviews. Microbiology*, *7*(9), 629. https://doi.org/10.1038/NRMICRO2200

Chancey, S. T., Zähner, D., & Stephens, D. S. (2012). Acquired inducible antimicrobial resistance in Gram-positive bacteria. *Future Microbiology*, *7*(8), 959–978. https://doi.org/10.2217/FMB.12.63

Chatzakis, E., Scoulica, E., Papageorgiou, N., Maraki, S., Samonis, G., & Galanakis, E. (2011). Infant colonization by Staphylococcus aureus: role of maternal carriage. *European Journal of Clinical Microbiology & Infectious Diseases : Official Publication of the European Society of Clinical*

Microbiology, *30*(9), 1111–1117. https://doi.org/10.1007/S10096-011-1199-9

Chen, A. Y., Adamek, R. N., Dick, B. L., Credille, C. V., Morrison, C. N., & Cohen, S. M. (2019). Targeting Metalloenzymes for Therapeutic Intervention. *Chemical Reviews*, *119*(2), 1323–1455. https://doi.org/10.1021/ACS.CHEMREV.8B00201

Chen, C., Krishnan, V., Macon, K., Manne, K., Narayana, S. V. L., & Schneewind, O. (2013). Secreted Proteases Control Autolysin-mediated Biofilm Growth of Staphylococcus aureus. *The Journal of Biological Chemistry*, *288*(41), 29440. https://doi.org/10.1074/JBC.M113.502039

Cheng, A. G., Hwan, K. K., Burts, M. L., Krausz, T., Schneewind, O., & Missiakas, D. M. (2009). Genetic requirements for Staphylococcus aureus abscess formation and persistence in host tissues. *FASEB Journal : Official Publication of the Federation of American Societies for Experimental Biology*, *23*(10), 3393–3404. https://doi.org/10.1096/FJ.09-135467

Cheng, A. G., McAdow, M., Kim, H. K., Bae, T., Missiakas, D. M., & Schneewind, O. (2010). Contribution of coagulases towards Staphylococcus aureus disease and protective immunity. *PLoS Pathogens*, *6*(8), 19–20. https://doi.org/10.1371/JOURNAL.PPAT.1001036

Cheng, J., Goldstein, R., Stec, B., Gershenson, A., & Roberts, M. F. (2012). Competition between anion binding and dimerization modulates Staphylococcus aureus phosphatidylinositol-specific phospholipase C enzymatic activity. *The Journal of Biological Chemistry*, *287*(48), 40317–

40327. https://doi.org/10.1074/JBC.M112.395277

Cheung, G. Y. C., Bae, J. S., & Otto, M. (2021). Pathogenicity and virulence of Staphylococcus aureus. *Https://Doi.Org/10.1080/21505594.2021.1878688*, *12*(1), 547–569. https://doi.org/10.1080/21505594.2021.1878688

Chipperfield, J. R., & Ratledge, C. (2000). Salicylic acid is not a bacterial siderophore: a theoretical study. *Biometals : An International Journal on the Role of Metal Ions in Biology, Biochemistry, and Medicine*, *13*(2), 165–168. https://doi.org/10.1023/A:1009227206890

Choi, Y., Kotzin, B., Herron, L., Callahan, J., Marrack, P., & Kappler, J. (1989). Interaction of Staphylococcus aureus toxin “superantigens” with human T cells. *Proceedings of the National Academy of Sciences of the United States of America*, *86*(22), 8941–8945. https://doi.org/10.1073/PNAS.86.22.8941

Chondros, P., Nikolidakis, D., Christodoulides, N., Rössler, R., Gutknecht, N., & Sculean, A. (2009). Photodynamic therapy as adjunct to non-surgical periodontal treatment in patients on periodontal maintenance: a randomized controlled clinical trial. *Lasers in Medical Science*, *24*(5), 681–688. https://doi.org/10.1007/S10103-008-0565-Z

Chopra, I., & Roberts, M. (2001). Tetracycline Antibiotics: Mode of Action, Applications, Molecular Biology, and Epidemiology of Bacterial Resistance. *Microbiology and Molecular Biology Reviews*, *65*(2), 232. https://doi.org/10.1128/MMBR.65.2.232-260.2001

Ciftci, I. H., Koken, R., Bukulmez, A., Ozdemir, M., Safak, B., & Cetinkaya, Z. (2007). Nasal carriage of Staphylococcus aureus in 4–6 age groups in healthy

children in Afyonkarahisar, Turkey. *Acta Paediatrica*, *96*(7), 1043–1046. https://doi.org/10.1111/J.1651-2227.2007.00315.X

Clancy, C. J., Bartsch, S. M., Nguyen, M. H., Stuckey, D. R., Shields, R. K., & Lee, B. Y. (2014). A computer simulation model of the cost-effectiveness of routine Staphylococcus aureus screening and decolonization among lung and heart-lung transplant recipients. *European Journal of Clinical Microbiology & Infectious Diseases : Official Publication of the European Society of Clinical Microbiology*, *33*(6), 1053–1061. https://doi.org/10.1007/S10096-013-2046-Y

Clark, N. C., Weigel, L. M., Patel, J. B., & Tenover, F. C. (2005). Comparison of Tn1546-Like Elements in Vancomycin-Resistant Staphylococcus aureus Isolates from Michigan and Pennsylvania. *Antimicrobial Agents and Chemotherapy*, *49*(1), 470. https://doi.org/10.1128/AAC.49.1.470-472.2005

Clarke, S. R., Brummell, K. J., Horsburgh, M. J., McDowell, P. W., Syed Mohamad, S. A., Stapleton, M. R., Acevedo, J., Read, R. C., Day, N. P. J., Peacock, S. J., Mond, J. J., Kokai-Kun, J. F., & Foster, S. J. (2006). Identification of In Vivo–Expressed Antigens of Staphylococcus aureus and Their Use in Vaccinations for Protection against Nasal Carriage. *The Journal of Infectious Diseases*, *193*(8), 1098–1108. https://doi.org/10.1086/501471

Clatworthy, A. E., Pierson, E., & Hung, D. T. (2007). Targeting virulence: a new paradigm for antimicrobial therapy. *Nature Chemical Biology*, *3*(9), 541–548. https://doi.org/10.1038/NCHEMBIO.2007.24

Clegg, J., Soldaini, E., McLoughlin, R. M., Rittenhouse, S., Bagnoli, F., &

Phogat, S. (2021). Staphylococcus aureus Vaccine Research and Development: The Past, Present and Future, Including Novel Therapeutic Strategies. *Frontiers in Immunology*, *12*, 2693. https://doi.org/10.3389/FIMMU.2021.705360/BIBTEX

Clinical and Laboratory Standards Institute (CLSI). (2021). M100 Performance Standards for Antimicrobial Susceptibility Testing. In *Clinical and Laboratory Standards Institute (CLSI)* (Vols. M100-Ed32).

Cluzet, V. C., Gerber, J. S., Nachamkin, I., Metlay, J. P., Zaoutis, T. E., Davis, M. F., Julian, K. G., Royer, D., Linkin, D. R., Coffin, S. E., Margolis, D. J., Hollander, J. E., Mistry, R. D., Gavin, L. J., Tolomeo, P., Wise, J. A., Wheeler, M. K., Bilker, W. B., Han, X., … Lautenbach, E. (2015). Duration of Colonization and Determinants of Earlier Clearance of Colonization With Methicillin-Resistant Staphylococcus aureus. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *60*(10), 1489–1496. https://doi.org/10.1093/CID/CIV075

Coculescu, B. I. (2009). Antimicrobial resistance induced by genetic changes. *Journal of Medicine and Life*, *2*(2), 114. /pmc/articles/PMC3018982/

Cogen, A. L., Yamasaki, K., Sanchez, K. M., Dorschner, R. A., Lai, Y., MacLeod, D. T., Torpey, J. W., Otto, M., Nizet, V., Kim, J. E., & Gallo, R. L. (2010). Selective antimicrobial action is provided by phenol-soluble modulins derived from Staphylococcus epidermidis, a normal resident of the skin. *The Journal of Investigative Dermatology*, *130*(1), 192–200. https://doi.org/10.1038/JID.2009.243

Cohen, N. A. (2006). Sinonasal mucociliary clearance in health and disease. *The Annals of Otology, Rhinology & Laryngology. Supplement*, *196*(9 II), 20–26. https://doi.org/10.1177/00034894061150S904

Coleman, D. C., Sullivan, D. J., Russell, R. J., Arbuthnott, J. P., Carey, B. F., & Pomeroy, H. M. (1989). Staphylococcus aureus bacteriophages mediating the simultaneous lysogenic conversion of beta-lysin, staphylokinase and enterotoxin A: molecular mechanism of triple conversion. *Journal of General Microbiology*, *135*(6), 1679–1697. https://doi.org/10.1099/00221287-135-6-1679

Collen, D., Schlott, B., Engelborghs, Y., Van Hoef, B., Hartmann, M., Lijnen, H. R., & Behnke, D. (1993). On the mechanism of the activation of human plasminogen by recombinant staphylokinase. *Journal of Biological Chemistry*, *268*(11), 8284–8289. https://doi.org/10.1016/S0021-9258(18)53094-5

Collins, L. V., Kristian, S. A., Weidenmaier, C., Faigle, M., Van Kessel, K. P. M., Van Strijp, J. A. G., Goütz, F., Neumeister, B., & Peschel, A. (2002). Staphylococcus aureus strains lacking D-alanine modifications of teichoic acids are highly susceptible to human neutrophil killing and are virulence attenuated in mice. *The Journal of Infectious Diseases*, *186*(2), 214–219. https://doi.org/10.1086/341454

Cong, Y., Yang, S., & Rao, X. (2020a). Vancomycin resistant Staphylococcus aureus infections: A review of case updating and clinical features. *Journal of Advanced Research*, *21*, 169. https://doi.org/10.1016/J.JARE.2019.10.005

Cong, Y., Yang, S., & Rao, X. (2020b). Vancomycin resistant Staphylococcus aureus infections: A review of case updating and clinical features. *Journal of Advanced Research*, *21*, 169. https://doi.org/10.1016/J.JARE.2019.10.005

Connell, S. R., Trieber, C. A., Dinos, G. P., Einfeldt, E., Taylor, D. E., & Nierhaus, K. H. (2003). Mechanism of Tet(O)-mediated tetracycline resistance. *The EMBO Journal*, *22*(4), 945–953. https://doi.org/10.1093/EMBOJ/CDG093

Cornaglia, G., Mazzariol, A., Fontana, R., & Satta, G. (1996). Diffusion of carbapenems through the outer membrane of enterobacteriaceae and correlation of their activities with their periplasmic concentrations. *Microbial Drug Resistance (Larchmont, N.Y.)*, *2*(2), 273–276. https://doi.org/10.1089/MDR.1996.2.273

Corrigan, R. M., Miajlovic, H., & Foster, T. J. (2009). Surface proteins that promote adherence of Staphylococcus aureus to human desquamated nasal epithelial cells. *BMC Microbiology*, *9*(1), 1–10. https://doi.org/10.1186/1471-2180-9-22/FIGURES/5

Costa, A. R., Batistão, D. W. F., Ribas, R. M., Sousa, A. M., Pereira, M. O., & Botelho, C. M. (2014). Staphylococcus aureus virulence factors and disease. *Microbial Pathogens and Strategies for Combating Them: Science, Technology and Education Volume 3*, 1668–1674.

Costa, S. S., Viveiros, M., Amaral, L., & Couto, I. (2013). Multidrug Efflux Pumps in Staphylococcus aureus: an Update. *The Open Microbiology Journal*, *7*(1), 59–71. https://doi.org/10.2174/1874285801307010059

Cox, G., & Wright, G. D. (2013). Intrinsic antibiotic resistance: mechanisms, origins, challenges and solutions. *International Journal of Medical Microbiology : IJMM*, *303*(6–7), 287–292. https://doi.org/10.1016/J.IJMM.2013.02.009

Cuatrecasas, P., Fuchs, S., & Anfinsen, C. B. (1967). Catalytic Properties and Specificity of the Extracellular Nuclease of Staphylococcus aureus. *Journal of Biological Chemistry*, *242*(7), 1541–1547. https://doi.org/10.1016/S0021-9258(18)96126-0

Cunningham, L., Catlin, B. W., & De Garilhe, M. P. (1956). A Deoxyribonuclease of Micrococcus pyogenes. *Journal of the American Chemical Society*, *78*(18), 4642–4645. https://doi.org/10.1021/JA01599A031/ASSET/JA01599A031.FP.PNG_V03

Cunnion, K. M., & Frank, M. M. (2003). Complement Activation Influences Staphylococcus aureus Adherence to Endothelial Cells. *Infection and Immunity*, *71*(3), 1321. https://doi.org/10.1128/IAI.71.3.1321-1327.2003

Cunnion, K. M., Lee, J. C., & Frank, M. M. (2001). Capsule production and growth phase influence binding of complement to Staphylococcus aureus. *Infection and Immunity*, *69*(11), 6796–6803. https://doi.org/10.1128/IAI.69.11.6796-6803.2001

Dall'Antonia, M., Coen, P. G., Wilks, M., Whiley, A., & Millar, M. (2005). Competition between methicillin-sensitive and -resistant Staphylococcus aureus in the anterior nares. *The Journal of Hospital Infection*, *61*(1), 62–67. https://doi.org/10.1016/J.JHIN.2005.01.008

Dashti, A. A., Jadaon, M. M., Abdulsamad, A. M., & Dashti, H. M. (2009). Heat Treatment of Bacteria: A Simple Method of DNA Extraction for Molecular Techniques. *Kuwait Medical Journal*, *41*(2), 117–122. http://www.ncbi.nih.

Dastgheyb, S. S., Villaruz, A. E., Le, K. Y., Tan, V. Y., Duong, A. C., Chatterjee, S. S., Cheung, G. Y. C., Joo, H. S., Hickok, N. J., & Otto, M. (2015). Role of Phenol-Soluble Modulins in Formation of Staphylococcus aureus Biofilms in Synovial Fluid. *Infection and Immunity*, *83*(7), 2966–2975. https://doi.org/10.1128/IAI.00394-15

Daugherty, S., & Low, M. G. (1993). Cloning, expression, and mutagenesis of phosphatidylinositol-specific phospholipase C from Staphylococcus aureus: a potential staphylococcal virulence factor. *Infection and Immunity*, *61*(12), 5078–5089. https://doi.org/10.1128/IAI.61.12.5078-5089.1993

David, M. Z., Boyle-Vavra, S., Zychowski, D. L., & Daum, R. S. (2011). Methicillin-Susceptible Staphylococcus aureus as a Predominantly Healthcare-Associated Pathogen: A Possible Reversal of Roles? *PLOS ONE*, *6*(4), e18217. https://doi.org/10.1371/JOURNAL.PONE.0018217

Dayie, N. T. K. D., Osei, M. M., Opintan, J. A., Tetteh-Quarcoo, P. B., Kotey, F. C. N., Ahenkorah, J., Adutwum-Ofosu, K. K., Egyir, B., & Donkor, E. S. (2021). Nasopharyngeal Carriage and Antimicrobial Susceptibility Profile of Staphylococcus aureus among Children under Five Years in Accra. *Pathogens*, *10*(2), 1–12. https://doi.org/10.3390/PATHOGENS10020136

De Haas, C. J. C., Veldkamp, K. E., Peschel, A., Weerkamp, F., Van Wamel, W. J. B., Heezius, E. C. J. M., Poppelier, M. J. J. G., Van Kessel, K. P. M., &

Van Strijp, J. A. G. (2004). Chemotaxis Inhibitory Protein of Staphylococcus aureus, a Bacterial Antiinflammatory Agent. *Journal of Experimental Medicine*, *199*(5), 687–695. https://doi.org/10.1084/JEM.20031636

de Steenhuijsen Piters, W. A. A., Sanders, E. A. M., & Bogaert, D. (2015). The role of the local microbial ecosystem in respiratory health and disease. *Philosophical Transactions of the Royal Society of London. Series B, Biological Sciences*, *370*(1675). https://doi.org/10.1098/RSTB.2014.0294

Degruttola, A. K., Low, D., Mizoguchi, A., & Mizoguchi, E. (2016). Current understanding of dysbiosis in disease in human and animal models. *Inflammatory Bowel Diseases*, *22*(5), 1137. https://doi.org/10.1097/MIB.0000000000000750

DeLeo, F. R., Kennedy, A. D., Chen, L., Wardenburg, J. B., Kobayashi, S. D., Mathema, B., Braughton, K. R., Whitney, A. R., Villaruz, A. E., Martens, C. A., Porcella, S. F., McGavin, M. J., Otto, M., Musser, J. M., & Kreiswirth, B. N. (2011). Molecular differentiation of historic phage-type 80/81 and contemporary epidemic Staphylococcus aureus. *Proceedings of the National Academy of Sciences of the United States of America*, *108*(44), 18091–18096. https://doi.org/10.1073/PNAS.1111084108

DeLeo, F. R., Otto, M., Kreiswirth, B. N., & Chambers, H. F. (2010). Community-associated meticillin-resistant Staphylococcus aureus. *The Lancet*, *375*(9725), 1557–1568. https://doi.org/10.1016/S0140-6736(09)61999-1

Demleitner, G., & Götz, F. (1994). Evidence for importance of the

Staphylococcus hyicus lipase pro-peptide in lipase secretion, stability and activity. *FEMS Microbiology Letters*, *121*(2), 189–197. https://doi.org/10.1111/J.1574-6968.1994.TB07098.X

Derde, L. P. G., Cooper, B. S., Goossens, H., Malhotra-Kumar, S., Willems, R. J. L., Gniadkowski, M., Hryniewicz, W., Empel, J., Dautzenberg, M. J. D., Annane, D., Aragão, I., Chalfine, A., Dumpis, U., Esteves, F., Giamarellou, H., Muzlovic, I., Nardi, G., Petrikkos, G. L., Tomic, V., … Bonten, M. J. M. (2014). Interventions to reduce colonisation and transmission of antimicrobial-resistant bacteria in intensive care units: an interrupted time series study and cluster randomised trial. *The Lancet. Infectious Diseases*, *14*(1), 31–39. https://doi.org/10.1016/S1473-3099(13)70295-0

Deurenberg, R. H., Vink, C., Kalenic, S., Friedrich, A. W., Bruggeman, C. A., & Stobberingh, E. E. (2007). The molecular evolution of methicillin-resistant Staphylococcus aureus. *Clinical Microbiology and Infection : The Official Publication of the European Society of Clinical Microbiology and Infectious Diseases*, *13*(3), 222–235. https://doi.org/10.1111/J.1469-0691.2006.01573.X

Deurenberg, R., & Stobberingh, E. (2009). The Molecular Evolution of Hospital- and Community-Associated Methicillin-Resistant Staphylococcus aureus. *Current Molecular Medicine*, *9*(2), 100–115. https://doi.org/10.2174/156652409787581637

Dey, S., Rosales-Klintz, S., Shouche, S., Pathak, J. P. N., & Pathak, A. (2013). Prevalence and risk factors for nasal carriage of Staphylococcus aureus in

children attending anganwaries (preschools) in Ujjain, India. *BMC Research Notes*, *6*(1). https://doi.org/10.1186/1756-0500-6-265

Deyno, S., Fekadu, S., & Astatkie, A. (2017). Resistance of Staphylococcus aureus to antimicrobial agents in Ethiopia: A meta-analysis. *Antimicrobial Resistance and Infection Control*, *6*(1), 1–15. https://doi.org/10.1186/S13756-017-0243-7/FIGURES/9

Dhand, A., & Sakoulas, G. (2014). Daptomycin in combination with other antibiotics for the treatment of complicated methicillin-resistant Staphylococcus aureus bacteremia. *Clinical Therapeutics*, *36*(10), 1303–1316. https://doi.org/10.1016/J.CLINTHERA.2014.09.005

Diep, B. A., Gill, S. R., Chang, R. F., Phan, T. H. Van, Chen, J. H., Davidson, M. G., Lin, F., Lin, J., Carleton, H. A., Mongodin, E. F., Sensabaugh, G. F., & Perdreau-Remington, F. (2006). Complete genome sequence of USA300, an epidemic clone of community-acquired meticillin-resistant Staphylococcus aureus. *The Lancet*, *367*(9512), 731–739. https://doi.org/10.1016/S0140-6736(06)68231-7

Dincer, S., Uslu, F. M., & Delik, A. (2020). Antibiotic Resistance in Biofilm. *Bacterial Biofilms*. https://doi.org/10.5772/INTECHOPEN.92388

Dinges, M. M., Orwin, P. M., & Schlievert, P. M. (2000). Exotoxins of Staphylococcus aureus. *Clinical Microbiology Reviews*, *13*(1), 16–34. https://doi.org/10.1128/CMR.13.1.16

Doery, H. M., Magnusson, B. J., Gulasekharam, J., & Pearson, J. E. (1965). The properties of phospholipase enzymes in staphylococcal toxins. *Journal of*

General Microbiology, *40*(2), 283–296. https://doi.org/10.1099/00221287-40-2-283

Dönhöfer, A., Franckenberg, S., Wickles, S., Berninghausen, O., Beckmann, R., & Wilson, D. N. (2012). Structural basis for TetM-mediated tetracycline resistance. *Proceedings of the National Academy of Sciences of the United States of America*, *109*(42), 16900–16905. https://doi.org/10.1073/PNAS.1208037109/-/DCSUPPLEMENTAL/SAPP.PDF

Donkor, E. S. (2017). A genomic infection control study for Staphylococcus aureus in two Ghanaian hospitals. *Journal of Clinical & Experimental Pathology*, *07*(01), 1. https://doi.org/10.4172/2161-0681.C1.031

Donkor, E. S., Dayie, N. T. K. D., & Tette, E. M. A. (2019). Methicillin-Resistant Staphylococcus aureus in Ghana: Past, Present, and Future. *Microbial Drug Resistance (Larchmont, N.Y.)*, *25*(5), 717–724. https://doi.org/10.1089/MDR.2018.0115

Donkor, E. S., Newman, M. J., Oliver-Commey, J., Bannerman, E., Dayie, N. T. K. D., & Badoe, E. V. (2010). Invasive disease and paediatric carriage of Streptococcus pneumoniae in Ghana. *Scandinavian Journal of Infectious Diseases*, *42*(4), 254–259. https://doi.org/10.3109/00365540903490000

Donkor, E. S., Tetteh-Quarcoo, P. B., Nartey, P., & Agyeman, I. O. (2012). Self-Medication Practices with Antibiotics among Tertiary Level Students in Accra, Ghana: A Cross-Sectional Study. *International Journal of Environmental Research and Public Health*, *9*(10), 3519.

https://doi.org/10.3390/IJERPH9103519

Dryla, A., Prustomersky, S., Gelbmann, D., Hanner, M., Bettinger, E., Kocsis, B., Kustos, T., Henics, T., Meinke, A., & Nagy, E. (2005). Comparison of Antibody Repertoires against Staphylococcus aureus in Healthy Individuals and in Acutely Infected Patients. *Clinical and Diagnostic Laboratory Immunology*, *12*(3), 387. https://doi.org/10.1128/CDLI.12.3.387-398.2005

Duran-Reynal, F. (1933). STUDIES ON A CERTAIN SPREADING FACTOR EXISTING IN BACTERIA AND ITS SIGNIFICANCE FOR BACTERIAL INVASIVENESS. *The Journal of Experimental Medicine*, *58*(2), 161–161. https://doi.org/10.1084/JEM.58.2.161

Džidić, S., Šušković, J., & Kos, B. (2008). Antibiotic resistance mechanisms in bacteria: Biochemical and genetic aspects. *Food Technology and Biotechnology*, *46*(1), 11–21.

Edwards, A. M., Massey, R. C., & Clarke, S. R. (2012). Molecular mechanisms of Staphylococcus aureus nasopharyngeal colonization. *Molecular Oral Microbiology*, *27*(1), 1–10. https://doi.org/10.1111/J.2041-1014.2011.00628.X

Edwards, D. R., Handsley, M. M., & Pennington, C. J. (2008). The ADAM metalloproteinases. *Molecular Aspects of Medicine*, *29*(5), 258–289. https://doi.org/10.1016/J.MAM.2008.08.001

Egyir, B., Bentum, J., Attram, N., Fox, A., Obeng-Nkrumah, N., Appiah-Korang, L., Behene, E., Kumordjie, S., Yeboah, C., Agbodzi, B., Bentil, R. E., Tagoe, R., Tabi, B. K. A., Owusu, F., Dayie, N. T. K. D., Donkor, E. S., Nsaful, J.,

Asah-Opoku, K., Nyarko, E., … Letizia, A. G. (2021). Whole Genome Sequencing and Antimicrobial Resistance of Staphylococcus aureus from Surgical Site Infections in Ghana. *Pathogens 2021, Vol. 10, Page 196*, *10*(2), 196. https://doi.org/10.3390/PATHOGENS10020196

Egyir, B., Guardabassi, L., Esson, J., Nielsen, S. S., Newman, M. J., Addo, K. K., & Larsen, A. R. (2014). Insights into Nasal Carriage of Staphylococcus aureus in an Urban and a Rural Community in Ghana. *PLOS ONE*, *9*(4), e96119. https://doi.org/10.1371/JOURNAL.PONE.0096119

Eibach, D., Nagel, M., Hogan, B., Azuure, C., Krumkamp, R., Dekker, D., Gajdiss, M., Brunke, M., Sarpong, N., Owusu-Dabo, E., & May, J. (2017). Nasal Carriage of Staphylococcus aureus among Children in the Ashanti Region of Ghana. *PloS One*, *12*(1). https://doi.org/10.1371/JOURNAL.PONE.0170320

Ekstedt, R. D., & Yotis, W. W. (1960). Studies on staphylococci. II. Effect of coagulase on the virulence of coagulase negative strains. *Journal of Bacteriology*, *80*(4), 496–500. https://doi.org/10.1128/JB.80.4.496-500.1960

El Atrouni, W. I., Knoll, B. M., Lahr, B. D., Eckel-Passow, J. E., Sia, I. G., & Baddour, L. M. (2009). Temporal trends in the incidence of Staphylococcus aureus bacteremia in Olmsted County, Minnesota, 1998 to 2005: a population-based study. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *49*(12). https://doi.org/10.1086/648442

Emaneini, M., Bigverdi, R., Kalantar, D., Soroush, S., Jabalameli, F., Noorazar

Khoshgnab, B., Asadollahi, P., & Taherikalani, M. (2013). Distribution of genes encoding tetracycline resistance and aminoglycoside modifying enzymes in Staphylococcus aureus strains isolated from a burn center. *Annals of Burns and Fire Disasters*, *26*(2), 76.

Eng, H. O., Wormald, P. J., & Lor, W. T. (2008). Innate Immunity in the Paranasal Sinuses: A Review of Nasal Host Defenses: *Https://Doi.Org/10.2500/Ajr.2008.22.3127*, *22*(1), 13–19. https://doi.org/10.2500/AJR.2008.22.3127

Ernst, C. M., & Peschel, A. (2011). Broad-spectrum antimicrobial peptide resistance by MprF-mediated aminoacylation and flipping of phospholipids. *Molecular Microbiology*, *80*(2), 290–299. https://doi.org/10.1111/J.1365-2958.2011.07576.X

Esposito, S., Leone, S., Petta, E., Noviello, S., & Ianniello, F. (2009). Treatment options for skin and soft tissue infections caused by meticillin-resistant Staphylococcus aureus:oralvs.parenteral; home vs. hospital. *International Journal of Antimicrobial Agents*, *34 Suppl 1*(SUPPL. 1). https://doi.org/10.1016/S0924-8579(09)70547-3

Esposito, S., & Principi, N. (2018). Impact of nasopharyngeal microbiota on the development of respiratory tract diseases. *European Journal of Clinical Microbiology & Infectious Diseases : Official Publication of the European Society of Clinical Microbiology*, *37*(1). https://doi.org/10.1007/S10096-017-3076-7

European Medicines Agency (EMA). (2021). *An overview of Libtayo and why it*

is authorised in the EU What is Libtayo and what is it used for ? 44(0), 1–2. https://www.ema.europa.eu/en/documents/overview/mylotarg-epar-summary-public_en.pdf

Eyal, Z., Matzov, D., Krupkin, M., Wekselman, I., Paukner, S., Zimmerman, E., Rozenberg, H., Bashan, A., & Yonath, A. (2015). Structural insights into species-specific features of the ribosome from the pathogen Staphylococcus aureus. *Proceedings of the National Academy of Sciences of the United States of America*, *112*(43), E5805–E5814. https://doi.org/10.1073/PNAS.1517952112/-/DCSUPPLEMENTAL/PNAS.201517952SI.PDF

Fajardo, A., Martínez-Martín, N., Mercadillo, M., Galán, J. C., Ghysels, B., Matthijs, S., Cornelis, P., Wiehlmann, L., Tümmler, B., Baquero, F., & Martínez, J. L. (2008). The neglected intrinsic resistome of bacterial pathogens. *PloS One*, *3*(2). https://doi.org/10.1371/JOURNAL.PONE.0001619

Falagas, M. E., Karageorgopoulos, D. E., Leptidis, J., & Korbila, I. P. (2013). MRSA in Africa: Filling the Global Map of Antimicrobial Resistance. *PLOS ONE*, *8*(7), e68024. https://doi.org/10.1371/JOURNAL.PONE.0068024

Fall, C., Richard, V., Dufougeray, A., Biron, A., Seck, A., Laurent, F., & Breurec, S. (2014). Staphylococcus aureus nasal and pharyngeal carriage in Senegal. *Clinical Microbiology and Infection*, *20*(4), O239–O241. https://doi.org/10.1111/1469-0691.12385

Farrell, A. M., Taylor, D., & Holland, K. T. (1995). Cloning, nucleotide sequence

determination and expression of the Staphylococcus aureus hyaluronate lyase gene. *FEMS Microbiology Letters*, *130*(1), 81–85. https://doi.org/10.1016/0378-1097(95)00187-A

Fattom, A., Schneerson, R., Watson, D. C., Karakawa, W. W., Fitzgerald, D., Pastan, I., Li, X., Shiloach, J., Bryla, D. A., & Robbins, J. B. (1993). Laboratory and clinical evaluation of conjugate vaccines composed of Staphylococcus aureus type 5 and type 8 capsular polysaccharides bound to Pseudomonas aeruginosa recombinant exoprotein A. *Infection and Immunity*, *61*(3), 1023–1032. https://doi.org/10.1128/IAI.61.3.1023-1032.1993

Fawley, W. N., Parnell, P., Hall, J., & Wilcox, M. H. (2006). Surveillance for mupirocin resistance following introduction of routine peri-operative prophylaxis with nasal mupirocin. *The Journal of Hospital Infection*, *62*(3), 327–332. https://doi.org/10.1016/J.JHIN.2005.09.022

Feingold, K. R. (2012). Lamellar bodies: the key to cutaneous barrier function. *The Journal of Investigative Dermatology*, *132*(8), 1951–1953. https://doi.org/10.1038/JID.2012.177

Fernandez-Lopez, R., de Toro, M., Moncalian, G., Garcillan-Barcia, M. P., & de la Cruz, F. (2016). Comparative Genomics of the Conjugation Region of F-like Plasmids: Five Shades of F. *Frontiers in Molecular Biosciences*, *3*(NOV). https://doi.org/10.3389/FMOLB.2016.00071

Fields, B. A., Malchiodi, E. L., Li, H., Ysern, X., Stauffacher, C. V., Schlievert, P. M., Karjalainen, K., & Mariuzza, R. A. (1996). Crystal structure of a T-cell receptor beta-chain complexed with a superantigen. *Nature*, *384*(6605), 188–

192. https://doi.org/10.1038/384188A0

Flemming, H. C., Wingender, J., Szewzyk, U., Steinberg, P., Rice, S. A., & Kjelleberg, S. (2016). Biofilms: an emergent form of bacterial life. *Nature Reviews Microbiology 2016 14:9*, *14*(9), 563–575. https://doi.org/10.1038/nrmicro.2016.94

Flores-Díaz, M., Monturiol-Gross, L., Naylor, C., Alape-Girón, A., & Flieger, A. (2016). Bacterial Sphingomyelinases and Phospholipases as Virulence Factors. *Microbiology and Molecular Biology Reviews : MMBR*, *80*(3), 597–628. https://doi.org/10.1128/MMBR.00082-15

Fluit, A. C., Visser, M. R., & Schmitz, F. J. (2001). Molecular Detection of Antimicrobial Resistance. *Clinical Microbiology Reviews*, *14*(4), 836. https://doi.org/10.1128/CMR.14.4.836-871.2001

Forsblom, E., Ruotsalainen, E., Ollgren, J., & Järvinen, A. (2013). Telephone consultation cannot replace bedside infectious disease consultation in the management of Staphylococcus aureus Bacteremia. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *56*(4), 527–535. https://doi.org/10.1093/CID/CIS889

Foster, T. J. (2005). Immune evasion by staphylococci. *Nature Reviews. Microbiology*, *3*(12), 948–958. https://doi.org/10.1038/NRMICRO1289

Foster, T. J. (2017). Antibiotic resistance in Staphylococcus aureus. Current status and future prospects. *FEMS Microbiology Reviews*, *41*(3), 430–449. https://doi.org/10.1093/FEMSRE/FUX007

Foster, T. J., & Höök, M. (1998). Surface protein adhesins of Staphylococcus

aureus. *Trends in Microbiology*, *6*(12), 484–488. https://doi.org/10.1016/S0966-842X(98)01400-0

Foster, T. J., & McDevitt, D. (1994). Surface-associated proteins of Staphylococcus aureus: their possible roles in virulence. *FEMS Microbiology Letters*, *118*(3), 199–205. https://doi.org/10.1111/J.1574-6968.1994.TB06828.X

Fowler, V. G., Sanders, L. L., Sexton, D. J., Kong, L., Marr, K. A., Gopal, A. K., Gottlieb, G., McClelland, R. S., & Corey, G. R. (1998). Outcome of Staphylococcus aureus bacteremia according to compliance with recommendations of infectious diseases specialists: experience with 244 patients. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *27*(3), 478–486. https://doi.org/10.1086/514686

Franceschi, F., Kanyo, Z., Sherer, E. C., & Sutcliffe, J. (2004). Macrolide resistance from the ribosome perspective. *Current Drug Targets. Infectious Disorders*, *4*(3), 177–191. https://doi.org/10.2174/1568005043340740

Friedrich, R., Panizzi, P., Fuentes-Prior, P., Richter, K., Verhamme, I., Anderson, P. J., Kawabata, S. I., Huber, R., Bode, W., & Bock, P. E. (2003). Staphylocoagulase is a prototype for the mechanism of cofactor-induced zymogen activation. *Nature*, *425*(6957), 535–539. https://doi.org/10.1038/NATURE01962

Frimodt-Møller, N., Espersen, F., Skinhøj, P., & Rosdahl, V. T. (1997). Epidemiology of Staphylococcus aureus bacteremia in Denmark from 1957

to 1990. *Clinical Microbiology and Infection : The Official Publication of the European Society of Clinical Microbiology and Infectious Diseases*, *3*(3), 297–305. https://doi.org/10.1111/J.1469-0691.1997.TB00617.X

Fritz, S. A., Camins, B. C., Eisenstein, K. A., Fritz, J. M., Epplin, E. K., Burnham, C.-A., Dukes, J., & Storch, G. A. (2011). Effectiveness of measures to eradicate Staphylococcus aureus carriage in patients with community-associated skin and soft-tissue infections: a randomized trial. *Infection Control and Hospital Epidemiology*, *32*(9), 872–880. https://doi.org/10.1086/661285

Fritz, S. A., Hogan, P. G., Hayek, G., Eisenstein, K. A., Rodriguez, M., Epplin, E. K., Garbutt, J., & Fraser, V. J. (2012). Household versus individual approaches to eradication of community-associated Staphylococcus aureus in children: a randomized trial. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *54*(6), 743–751. https://doi.org/10.1093/CID/CIR919

Fritz, S. A., Tiemann, K. M., Hogan, P. G., Epplin, E. K., Rodriguez, M., Al-Zubeidi, D. N., Bubeck Wardenburg, J., & Hunstad, D. A. (2013). A serologic correlate of protective immunity against community-onset Staphylococcus aureus infection. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *56*(11), 1554–1561. https://doi.org/10.1093/CID/CIT123

Frost, L. S., Leplae, R., Summers, A. O., & Toussaint, A. (2005). Mobile genetic elements: the agents of open source evolution. *Nature Reviews Microbiology*

2005 3:9, *3*(9), 722–732. https://doi.org/10.1038/nrmicro1235

Fuchs, E., & Green, H. (1980). Changes in keratin gene expression during terminal differentiation of the keratinocyte. *Cell*, *19*(4), 1033–1042. https://doi.org/10.1016/0092-8674(80)90094-X

Fuchs, E., & Raghavan, S. (2002). Getting under the skin of epidermal morphogenesis. *Nature Reviews. Genetics*, *3*(3), 199–209. https://doi.org/10.1038/NRG758

Futosi, K., Fodor, S., & Mócsai, A. (2013). Neutrophil cell surface receptors and their intracellular signal transduction pathways. *International Immunopharmacology*, *17*(3), 638–650. https://doi.org/10.1016/J.INTIMP.2013.06.034

García-Álvarez, L., Holden, M. T. G., Lindsay, H., Webb, C. R., Brown, D. F. J., Curran, M. D., Walpole, E., Brooks, K., Pickard, D. J., Teale, C., Parkhill, J., Bentley, S. D., Edwards, G. F., Girvan, E. K., Kearns, A. M., Pichon, B., Hill, R. L. R., Larsen, A. R., Skov, R. L., … Holmes, M. A. (2011). Meticillin-resistant Staphylococcus aureus with a novel mecA homologue in human and bovine populations in the UK and Denmark: A descriptive study. *The Lancet Infectious Diseases*, *11*(8), 595–603. https://doi.org/10.1016/S1473-3099(11)70126-8

Garner, J. S. (1996). Guideline for isolation precautions in hospitals. The Hospital Infection Control Practices Advisory Committee. *Infection Control and Hospital Epidemiology*, *17*(1), 53–80. https://doi.org/10.1086/647190

Garoy, E. Y., Gebreab, Y. B., Achila, O. O., Tekeste, D. G., Kesete, R., Ghirmay,

R., Kiflay, R., & Tesfu, T. (2019). Methicillin-Resistant Staphylococcus aureus (MRSA): Prevalence and Antimicrobial Sensitivity Pattern among Patients-A Multicenter Study in Asmara, Eritrea. *The Canadian Journal of Infectious Diseases & Medical Microbiology = Journal Canadien Des Maladies Infectieuses et de La Microbiologie Medicale*, *2019*. https://doi.org/10.1155/2019/8321834

Gase, A., Hartmann, M., Gührs, K. H., Röcker, A., Collen, D., Behnke, D., & Schlott, B. (1996). Functional significance of NH2- and C00H-terminal regions of staphylokinase in plasminogen activation. *Thrombosis and Haemostasis*, *76*(5), 755–760. https://doi.org/10.1055/S-0038-1650656/ID/JR_11

Gauduchon, V., Werner, S., Prévost, G., Monteil, H., & Colin, D. A. (2001). Flow cytometric determination of Panton-Valentine leucocidin S component binding. *Infection and Immunity*, *69*(4), 2390–2395. https://doi.org/10.1128/IAI.69.4.2390-2395.2001

Gelband, H., Miller-Petrie, M., … P. S.-T. state of the world's, & 2015, U. (2015). The state of the world's antibiotics, 2015. *Cabdirect.Org*.

Geoghegan, J. A., & Foster, T. J. (2017). Cell wall-anchored surface proteins of Staphylococcus aureus: Many proteins, multiple functions. *Current Topics in Microbiology and Immunology*, *409*, 95–120. https://doi.org/10.1007/82_2015_5002

Geurkink, N. (1983). Nasal anatomy, physiology, and function. *The Journal of Allergy and Clinical Immunology*, *72*(2), 123–128.

https://doi.org/10.1016/0091-6749(83)90518-3

Gidengil, C. A., Gay, C., Huang, S. S., Platt, R., Yokoe, D., & Lee, G. M. (2015). Cost-effectiveness of strategies to prevent MRSA transmission and infection in an intensive care unit. *Infection Control and Hospital Epidemiology*, *36*(1), 17. https://doi.org/10.1017/ICE.2014.12

Giesbrecht, P., Kersten, T., Maidhof, H., & Wecke, J. (1998). Staphylococcal Cell Wall: Morphogenesis and Fatal Variations in the Presence of Penicillin. *Microbiology and Molecular Biology Reviews*, *62*(4), 1371. https://doi.org/10.1128/MMBR.62.4.1371-1414.1998

Giuliano, C. A., & Rybak, M. J. (2015). Efficacy of triclosan as an antimicrobial hand soap and its potential impact on antimicrobial resistance: a focused review. *Pharmacotherapy*, *35*(3), 328–336. https://doi.org/10.1002/PHAR.1553

Giuliano, C., Patel, C. R., & Kale-Pradhan, P. B. (2019). A Guide to Bacterial Culture Identification And Results Interpretation. *Pharmacy and Therapeutics*, *44*(4), 192. /pmc/articles/PMC6428495/

Glenny, A. T., & Stevens, M. F. (1935). Staphy-lococcus Toxins and Antitoxins. *Journal of Pathology and Bacteriology*, *40*, 201–210.

Goerke, C., Pantucek, R., Holtfreter, S., Schulte, B., Zink, M., Grumann, D., Bröker, B. M., Doskar, J., & Wolz, C. (2009). Diversity of prophages in dominant Staphylococcus aureus clonal lineages. *Journal of Bacteriology*, *191*(11), 3462–3468. https://doi.org/10.1128/JB.01804-08

Goldstein, R., Cheng, J., Stec, B., & Roberts, M. F. (2012). Structure of the S.

aureus PI-specific phospholipase C reveals modulation of active site access by a titratable π-cation latched loop. *Biochemistry*, *51*(12), 2579–2587. https://doi.org/10.1021/BI300057Q

Goossens, H., Ferech, M., Vander Stichele, R., & Elseviers, M. (2005a). Outpatient antibiotic use in Europe and association with resistance: a cross-national database study. *Lancet (London, England)*, *365*(9459), 579–587. https://doi.org/10.1016/S0140-6736(05)17907-0

Goossens, H., Ferech, M., Vander Stichele, R., & Elseviers, M. (2005b). Outpatient antibiotic use in Europe and association with resistance: a cross-national database study. *Lancet (London, England)*, *365*(9459), 579–587. https://doi.org/10.1016/S0140-6736(05)17907-0

Gorwitz, R. J. (2008). A review of community-associated methicillin-resistant Staphylococcus aureus skin and soft tissue infections. *The Pediatric Infectious Disease Journal*, *27*(1), 1–7. https://doi.org/10.1097/INF.0B013E31815819BB

Gorwitz, R. J., Kruszon-Moran, D., McAllister, S. K., McQuillan, G., McDougal, L. K., Fosheim, G. E., Jensen, B. J., Killgore, G., Tenover, F. C., & Kuehnert, M. J. (2008). Changes in the prevalence of nasal colonization with Staphylococcus aureus in the United States, 2001-2004. *The Journal of Infectious Diseases*, *197*(9), 1226–1234. https://doi.org/10.1086/533494

Götz, F., Verheij, H. M., & Rosenstein, R. (1998). Staphylococcal lipases: molecular characterisation, secretion, and processing. *Chemistry and Physics of Lipids*, *93*(1–2), 15–25. https://doi.org/10.1016/S0009-3084(98)00025-5

Gouaux, J. E., Braha, O., Hobaugh, M. R., Song, L., Cheley, S., Shustak, C., & Bayley, H. (1994). Subunit stoichiometry of staphylococcal alpha-hemolysin in crystals and on membranes: a heptameric transmembrane pore. *Proceedings of the National Academy of Sciences of the United States of America*, *91*(26), 12828–12831. https://doi.org/10.1073/PNAS.91.26.12828

Graves, S. F., Kobayashi, S. D., Braughton, K. R., Whitney, A. R., Sturdevant, D. E., Rasmussen, D. L., Kirpotina, L. N., Quinn, M. T., & DeLeo, F. R. (2012). Sublytic concentrations of Staphylococcus aureus Panton-Valentine leukocidin alter human PMN gene expression and enhance bactericidal capacity. *Journal of Leukocyte Biology*, *92*(2), 361–374. https://doi.org/10.1189/JLB.1111575

Gregory, P. D., Lewis, R. A., Curnock, S. P., & Dyke, K. G. H. (1997). Studies of the repressor (BlaI) of beta-lactamase synthesis in Staphylococcus aureus. *Molecular Microbiology*, *24*(5), 1025–1037. https://doi.org/10.1046/J.1365-2958.1997.4051770.X

Griffith, M., Postelnick, M., & Scheetz, M. (2012). Antimicrobial stewardship programs: methods of operation and suggested outcomes. *Expert Review of Anti-Infective Therapy*, *10*(1), 63–73. https://doi.org/10.1586/ERI.11.153

Griffith, O. H., & Ryan, M. (1999). Bacterial phosphatidylinositol-specific phospholipase C: structure, function, and interaction with lipids. *Biochimica et Biophysica Acta*, *1441*(2–3), 237–254. https://doi.org/10.1016/S1388-1981(99)00153-5

Groome, M. J., Albrich, W. C., Wadula, J., Khoosal, M., & Madhi, S. A. (2012).

Community-onset Staphylococcus aureus bacteraemia in hospitalised African children: high incidence in HIV-infected children and high prevalence of multidrug resistance. *Paediatrics and International Child Health*, *32*(3), 140–146. https://doi.org/10.1179/1465328111Y.0000000044

Grumann, D., Nübel, U., & Bröker, B. M. (2014). Staphylococcus aureus toxins--their functions and genetics. *Infection, Genetics and Evolution : Journal of Molecular Epidemiology and Evolutionary Genetics in Infectious Diseases*, *21*, 583–592. https://doi.org/10.1016/J.MEEGID.2013.03.013

Grumann, D., Ruotsalainen, E., Kolata, J., Kuusela, P., Järvinen, A., Kontinen, V. P., Bröker, B. M., & Holtfreter, S. (2011). Characterization of infecting strains and superantigen-neutralizing antibodies in Staphylococcus aureus bacteremia. *Clinical and Vaccine Immunology : CVI*, *18*(3), 487–493. https://doi.org/10.1128/CVI.00329-10

Gu, F. F., Han, L. Z., Chen, X., Wang, Y. C., Shen, H., Wang, J. Q., Tang, J., Zhang, J., & Ni, Y. X. (2015). Molecular characterization of Staphylococcus aureus from surgical site infections in orthopedic patients in an orthopedic trauma clinical medical center in Shanghai. *Surgical Infections*, *16*(1), 97–104. https://doi.org/10.1089/SUR.2014.027

Guerra, F. E., Borgogna, T. R., Patel, D. M., Sward, E. W., & Voyich, J. M. (2017). Epic immune battles of history: Neutrophils vs. Staphylococcus aureus. *Frontiers in Cellular and Infection Microbiology*, *7*(JUN), 286. https://doi.org/10.3389/FCIMB.2017.00286/BIBTEX

Guo, Y., Song, G., Sun, M., Wang, J., & Wang, Y. (2020). Prevalence and

Therapies of Antibiotic-Resistance in Staphylococcus aureus. *Frontiers in Cellular and Infection Microbiology*, *10*, 107. https://doi.org/10.3389/FCIMB.2020.00107

Gurung, R. R., Maharjan, P., & Chhetri, G. G. (2020). Antibiotic resistance pattern of Staphylococcus aureus with reference to MRSA isolates from pediatric patients. *Future Science OA*, *6*(4). https://doi.org/10.2144/FSOA-2019-0122

Guzmán-Blanco, M., Mejía, C., Isturiz, R., Alvarez, C., Bavestrello, L., Gotuzzo, E., Labarca, J., Luna, C. M., Rodríguez-Noriega, E., Salles, M. J. C., Zurita, J., & Seas, C. (2009). Epidemiology of meticillin-resistant Staphylococcus aureus (MRSA) in Latin America. *International Journal of Antimicrobial Agents*, *34*(4), 304–308. https://doi.org/10.1016/J.IJANTIMICAG.2009.06.005

Hacker, J., Blum-Oehler, G., Hochhut, B., & Dobrindt, U. (2003). The molecular basis of infectious diseases: pathogenicity islands and other mobile genetic elements. A review. *Acta Microbiologica et Immunologica Hungarica*, *50*(4), 321–330. https://doi.org/10.1556/AMICR.50.2003.4.1

Hall, C. W., & Mah, T. F. (2017). Molecular mechanisms of biofilm-based antibiotic resistance and tolerance in pathogenic bacteria. *FEMS Microbiology Reviews*, *41*(3), 276–301. https://doi.org/10.1093/FEMSRE/FUX010

Hamblin, M. R., & Hasan, T. (2004). Photodynamic therapy: a new antimicrobial approach to infectious disease? *Photochemical & Photobiological Sciences :*

Official Journal of the European Photochemistry Association and the European Society for Photobiology, *3*(5), 436–450. https://doi.org/10.1039/B311900A

Hanssen, A. M., & Ericson Sollid, J. U. (2006). SCCmec in staphylococci: genes on the move. *FEMS Immunology and Medical Microbiology*, *46*(1), 8–20. https://doi.org/10.1111/J.1574-695X.2005.00009.X

Hanssen, A. M., Kindlund, B., Stenklev, N. C., Furberg, A. S., Fismen, S., Olsen, R. S., Johannessen, M., & Sollid, J. U. E. (2017). Localization of Staphylococcus aureus in tissue from the nasal vestibule in healthy carriers. *BMC Microbiology*, *17*(1), 1–11. https://doi.org/10.1186/S12866-017-0997-3/FIGURES/7

Harada, Y., Chong, Y., Shimono, N., Miyake, N., Uchida, Y., Kadowaki, M., Akashi, K., & Shimoda, S. (2014). Nosocomial spread of meticillin-resistant Staphylococcus aureus with β-lactam-inducible arbekacin resistance. *Journal of Medical Microbiology*, *63*(PART 5), 710–714. https://doi.org/10.1099/JMM.0.065276-0/CITE/REFWORKS

Haraldsson, I., & Jonsson, P. (1984). Histopathology and pathogenesis of mouse mastitis induced with Staphylococcus aureus mutants. *Journal of Comparative Pathology*, *94*(2), 183–196. https://doi.org/10.1016/0021-9975(84)90039-2

Hardie, J. M., & Whiley, R. A. (1995). The genus <Emphasis Type="Italic">Streptococcus</Emphasis>. *The Genera of Lactic Acid Bacteria*, 55–124. https://doi.org/10.1007/978-1-4615-5817-0_4

Hardwick, N., Parry, C. M., & Sharpe, G. R. (1995). Staphylococcal scalded skin syndrome in an adult. Influence of immune and renal factors. *The British Journal of Dermatology*, *132*(3), 468–471. https://doi.org/10.1111/J.1365-2133.1995.TB08686.X

Hardy, B. L., Dickey, S. W., Plaut, R. D., Riggins, D. P., Stibitz, S., Otto, M., & Merrell, D. S. (2019). Corynebacterium pseudodiphtheriticum Exploits Staphylococcus aureus Virulence Components in a Novel Polymicrobial Defense Strategy. *MBio*, *10*(1). https://doi.org/10.1128/MBIO.02491-18

Harris, A. D., Nemoy, L., Johnson, J. A., Martin-Carnahan, A., Smith, D. L., Standiford, H., & Perencevich, E. N. (2004). Co-carriage rates of vancomycin-resistant Enterococcus and extended-spectrum beta-lactamase-producing bacteria among a cohort of intensive care unit patients: implications for an active surveillance program. *Infection Control and Hospital Epidemiology*, *25*(2), 105–108. https://doi.org/10.1086/502358

Hart, M. E., Hart, M. J., & Roop, A. J. (2009). Genotypic and Phenotypic Assessment of Hyaluronidase among Type Strains of a Select Group of Staphylococcal Species. *International Journal of Microbiology*, *2009*. https://doi.org/10.1155/2009/614371

Hartleib, J., Köhler, N., Dickinson, R. B., Chhatwal, G. S., Sixma, J. J., Hartford, O. M., Foster, T. J., Peters, G., Kehrel, B. E., & Herrmann, M. (2000). Protein A is the von Willebrand factor binding protein onStaphylococcus aureus. *Blood*, *96*(6), 2149–2156. https://doi.org/10.1182/BLOOD.V96.6.2149

Haseeb, A., Ajit Singh, V., Teh, C. S. J., & Loke, M. F. (2019). Addition of ceftaroline fosamil or vancomycin to PMMA: An in vitro comparison of biomechanical properties and anti-MRSA efficacy. *Journal of Orthopaedic Surgery*, *27*(2). https://doi.org/10.1177/2309499019850324/ASSET/IMAGES/LARGE/10.1177_2309499019850324-FIG3.JPEG

Hashizume, H., Takahashi, Y., Masuda, T., Ohba, S. I., Ohishi, T., Kawada, M., & Igarashi, M. (2017). In vivo efficacy of β-lactam/tripropeptin C in a mouse septicemia model and the mechanism of reverse β-lactam resistance in methicillin-resistant Staphylococcus aureus mediated by tripropeptin C. *The Journal of Antibiotics 2018 71:1*, *71*(1), 79–85. https://doi.org/10.1038/ja.2017.88

Hatem, Z. A., Al-Dulaimi, A. A. F., & Al-Taai, H. R. R. (2022). Prevalence of tetracycline resistance genes in Staphylococcus aureus isolated from different clinical sources in Diyala, Iraq. *International Journal of Health Sciences*, *6*(April), 13200–13209. https://doi.org/10.53730/ijhs.v6ns2.8514

Hattar, K., Grandel, U., Moeller, A., Fink, L., Iglhaut, J., Hartung, T., Morath, S., Seeger, W., Grimminger, F., & Sibelius, U. (2006). Lipoteichoic acid (LTA) from Staphylococcus aureus stimulates human neutrophil cytokine release by a CD14-dependent, Toll-like-receptor-independent mechanism: Autocrine role of tumor necrosis factor-[alpha] in mediating LTA-induced interleukin-8 generation. *Critical Care Medicine*, *34*(3), 835–841. https://doi.org/10.1097/01.CCM.0000202204.01230.44

Haviland, D. L., McCoy, R. L., Whitehead, W. T., Akama, H., Molmenti, E. P., Brown, A., Haviland, J. C., Parks, W. C., Perlmutter, D. H., & Wetsel, R. A. (1995). Cellular expression of the C5a anaphylatoxin receptor (C5aR): demonstration of C5aR on nonmyeloid cells of the liver and lung. *Journal of Immunology (Baltimore, Md. : 1950)*, *154*(4), 1861–1869. http://www.ncbi.nlm.nih.gov/pubmed/7836770

Hawkey, P. M. (2003). Mechanisms of quinolone action and microbial response. *The Journal of Antimicrobial Chemotherapy*, *51 Suppl 1*(SUPPL. 1), 29–35. https://doi.org/10.1093/JAC/DKG207

Hazmanian, S. K., Skaar, E. P., Gaspar, A. H., Humayun, M., Gornicki, P., Jelenska, J., Joachmiak, A., Missiakas, D. M., & Schneewind, O. (2003). Passage of heme-iron across the envelope of Staphylococcus aureus. *Science (New York, N.Y.)*, *299*(5608), 906–909. https://doi.org/10.1126/SCIENCE.1081147

He, S., Lin, J., Li, Y., Zhang, W., Zhou, J., Han, Z., & Yao, Z. (2020). Insights into the epidemiology of methicillin-resistant coagulase-negative Staphylococci carriage in community-based drug users. *Journal of Infection and Public Health*, *13*(11), 1742–1748. https://doi.org/10.1016/J.JIPH.2020.09.011

Heidary, M., Khosravi, A. D., Khoshnood, S., Nasiri, M. J., Soleimani, S., & Goudarzi, M. (2018). Daptomycin. *Journal of Antimicrobial Chemotherapy*, *73*(1), 1–11. https://doi.org/10.1093/JAC/DKX349

Heikkilä, M. P., & Saris, P. E. J. (2003). Inhibition of Staphylococcus aureus by

the commensal bacteria of human milk. *Journal of Applied Microbiology*, *95*(3), 471–478. https://doi.org/10.1046/J.1365-2672.2003.02002.X

Henares, D., Brotons, P., de Sevilla, M. F., Fernandez-Lopez, A., Hernandez-Bou, S., Perez-Argüello, A., Mira, A., Muñoz-Almagro, C., & Cabrera-Rubio, R. (2021). Differential nasopharyngeal microbiota composition in children according to respiratory health status. *Microbial Genomics*, *7*(10). https://doi.org/10.1099/MGEN.0.000661

Henares, D., Rocafort, M., Brotons, P., de Sevilla, M. F., Mira, A., Launes, C., Cabrera-Rubio, R., & Muñoz-Almagro, C. (2021). Rapid Increase of Oral Bacteria in Nasopharyngeal Microbiota After Antibiotic Treatment in Children With Invasive Pneumococcal Disease. *Frontiers in Cellular and Infection Microbiology*, *11*, 929. https://doi.org/10.3389/FCIMB.2021.744727/BIBTEX

Hidalgo, A., Peired, A. J., Wild, M. K., Vestweber, D., & Frenette, P. S. (2007). Complete identification of E-selectin ligands on neutrophils reveals distinct functions of PSGL-1, ESL-1, and CD44. *Immunity*, *26*(4), 477–489. https://doi.org/10.1016/J.IMMUNI.2007.03.011

Hidron, A. I., Low, C. E., Honig, E. G., & Blumberg, H. M. (2009). Emergence of community-acquired meticillin-resistant Staphylococcus aureus strain USA300 as a cause of necrotising community-onset pneumonia. *The Lancet Infectious Diseases*, *9*(6), 384–392. https://doi.org/10.1016/S1473-3099(09)70133-1

Hill, R. L. R., Kearns, A. M., Nash, J., North, S. E., Pike, R., Newson, T.,

Woodford, N., Calver, R., & Livermore, D. M. (2010). Linezolid-resistant ST36 methicillin-resistant Staphylococcus aureus associated with prolonged linezolid treatment in two paediatric cystic fibrosis patients. *The Journal of Antimicrobial Chemotherapy*, *65*(3), 442–445. https://doi.org/10.1093/JAC/DKP494

Hiramatsu, K., Aritaka, N., Hanaki, H., Kawasaki, S., Hosoda, Y., Hori, S., Fukuchi, Y., & Kobayashi, I. (1997). Dissemination in Japanese hospitals of strains of Staphylococcus aureus heterogeneously resistant to vancomycin. *Lancet (London, England)*, *350*(9092), 1670–1673. https://doi.org/10.1016/S0140-6736(97)07324-8

Hiramatsu, K., Hanaki, H., Ino, T., Yabuta, K., Oguri, T., & Tenover, F. C. (1997). Methicillin-resistant Staphylococcus aureus clinical strain with reduced vancomycin susceptibility. *The Journal of Antimicrobial Chemotherapy*, *40*(1), 135–136. https://doi.org/10.1093/JAC/40.1.135

Holden, M. T. G., Hsu, L. Y., Kurt, K., Weinert, L. A., Mather, A. E., Harris, S. R., Strommenger, B., Layer, F., Witte, W., De Lencastre, H., Skov, R., Westh, H., Žemličková, H., Coombs, G., Kearns, A. M., Hill, R. L. R., Edgeworth, J., Gould, I., Gant, V., … Nübel, U. (2013). A genomic portrait of the emergence, evolution, and global spread of a methicillin-resistant Staphylococcus aureus pandemic. *Genome Research*, *23*(4), 653–664. https://doi.org/10.1101/GR.147710.112

Hollenbeck, B. L., & Rice, L. B. (2012). Intrinsic and acquired resistance mechanisms in enterococcus. *Virulence*, *3*(5), 421–569.

https://doi.org/10.4161/VIRU.21282

Holmes, N. E., Tong, S. Y. C., Davis, J. S., & Hal, S. J. V. (2015). Treatment of methicillin-resistant staphylococcus aureus: Vancomycin and beyond. *Seminars in Respiratory and Critical Care Medicine*, *36*(1), 17–30. https://doi.org/10.1055/S-0034-1397040/ID/JR01090-42

Holtfreter, S., & Bröker, B. M. (2005). Staphylococcal superantigens: do they play a role in sepsis? *Archivum Immunologiae et Therapiae Experimentalis*, *53*(1), 13–27. https://europepmc.org/article/med/15761373

Holtfreter, S., Roschack, K., Eichler, P., Eske, K., Holtfreter, B., Kohler, C., Engelmann, S., Hecker, M., Greinacher, A., & Bröker, B. M. (2006). Staphylococcus aureus carriers neutralize superantigens by antibodies specific for their colonizing strain: a potential explanation for their improved prognosis in severe sepsis. *The Journal of Infectious Diseases*, *193*(9), 1275–1278. https://doi.org/10.1086/503048

Holton, D. L., Nicolle, L. E., Diley, D., & Bernstein, K. (1991). Efficacy of mupirocin nasal ointment in eradicating Staphylococcus aureus nasal carriage in chronic haemodialysis patients. *The Journal of Hospital Infection*, *17*(2), 133–137. https://doi.org/10.1016/0195-6701(91)90177-A

Hong, H. J., Hutchings, M. I., & Buttner, M. J. (2008). Vancomycin resistance VanS/VanR two-component systems. *Advances in Experimental Medicine and Biology*, *631*, 200–213. https://doi.org/10.1007/978-0-387-78885-2_14

Horii, T., Yokoyama, K., Barua, S., Odagiri, T., Futamura, N., Hasegawa, T., & Ohta, M. (2000). The staphylokinase gene is located in the structural gene

encoding N-acetylmuramyl-L-alanine amidase in methicillin-resistant Staphylococcus aureus. *FEMS Microbiology Letters*, *185*(2), 221–224. https://doi.org/10.1111/J.1574-6968.2000.TB09065.X

Howden, B. P., Davies, J. K., Johnson, P. D. R., Stinear, T. P., & Grayson, M. L. (2010). Reduced vancomycin susceptibility in Staphylococcus aureus, including vancomycin-intermediate and heterogeneous vancomycin-intermediate strains: Resistance mechanisms, laboratory detection, and clinical implications. *Clinical Microbiology Reviews*, *23*(1), 99–139. https://doi.org/10.1128/CMR.00042-09/ASSET/3E28D4B9-B4DB-4740-AB24-EC4EC556CB59/ASSETS/GRAPHIC/ZCM0011023000009.JPEG

Howe, R. A., Wootton, M., Noel, A. R., Bowker, K. E., Walsh, T. R., & MacGowan, A. P. (2003). Activity of AZD2563, a novel oxazolidinone, against Staphylococcus aureus strains with reduced susceptibility to vancomycin or linezolid. *Antimicrobial Agents and Chemotherapy*, *47*(11), 3651–3652. https://doi.org/10.1128/AAC.47.11.3651-3652.2003

Hu, C., Xiong, N., Zhang, Y., Rayner, S., & Chen, S. (2012). Functional characterization of lipase in the pathogenesis of Staphylococcus aureus. *Biochemical and Biophysical Research Communications*, *419*(4), 617–620. https://doi.org/10.1016/J.BBRC.2012.02.057

Hu, Q., Peng, H., & Rao, X. (2016). Molecular events for promotion of vancomycin resistance in vancomycin intermediate Staphylococcus aureus. *Frontiers in Microbiology*, *7*(OCT). https://doi.org/10.3389/FMICB.2016.01601

Huang, S. S., Septimus, E., Avery, T. R., Lee, G. M., Hickok, J., Weinstein, R. A., Moody, J., Hayden, M. K., Perlin, J. B., Platt, R., & Ray, G. T. (2014). Cost savings of universal decolonization to prevent intensive care unit infection: implications of the REDUCE MRSA trial. *Infection Control and Hospital Epidemiology*, *35 Suppl 3*(S3), S23–S31. https://doi.org/10.1086/677819

Huang, Y. J. (2017). Nasopharyngeal microbiota: Gatekeepers or fortune tellers of susceptibility to respiratory tract infections? *American Journal of Respiratory and Critical Care Medicine*, *196*(12), 1504–1505. https://doi.org/10.1164/RCCM.201707-1470ED/SUPPL_FILE/DISCLOSURES.PDF

Hughes, D., & Andersson, D. I. (2017). Environmental and genetic modulation of the phenotypic expression of antibiotic resistance. *FEMS Microbiology Reviews*, *41*(3), 374–391. https://doi.org/10.1093/FEMSRE/FUX004

Hunt, K. M., Foster, J. A., Forney, L. J., Schütte, U. M. E., Beck, D. L., Abdo, Z., Fox, L. K., Williams, J. E., McGuire, M. K., & McGuire, M. A. (2011). Characterization of the Diversity and Temporal Stability of Bacterial Communities in Human Milk. *PLOS ONE*, *6*(6), e21313. https://doi.org/10.1371/JOURNAL.PONE.0021313

Huovinen, P., Sundstrom, L., Swedberg, G., & Skold, O. (1995). Trimethoprim and sulfonamide resistance. *Antimicrobial Agents and Chemotherapy*, *39*(2), 279–289. https://doi.org/10.1128/AAC.39.2.279

Hurdle, J. G., O'Neill, A. J., Ingham, E., Fishwick, C., & Chopra, I. (2004).

Analysis of Mupirocin Resistance and Fitness in Staphylococcus aureus by Molecular Genetic and Structural Modeling Techniques. *Antimicrobial Agents and Chemotherapy*, *48*(11), 4366. https://doi.org/10.1128/AAC.48.11.4366-4376.2004

Huseby, M. J., Kruse, A. C., Digre, J., Kohler, P. L., Vocke, J. A., Mann, E. E., Bayles, K. W., Bohach, G. A., Schlievert, P. M., Ohlendorf, D. H., & Earhart, C. A. (2010). Beta toxin catalyzes formation of nucleoprotein matrix in staphylococcal biofilms. *Proceedings of the National Academy of Sciences of the United States of America*, *107*(32), 14407–14412. https://doi.org/10.1073/PNAS.0911032107

Hussein, N. R., Basharat, Z., Muhammed, A. H., & Al-Dabbagh, S. A. (2015). Comparative evaluation of MRSA nasal colonization epidemiology in the urban and rural secondary school community of Kurdistan, Iraq. *PloS One*, *10*(5). https://doi.org/10.1371/JOURNAL.PONE.0124920

Huttenhower, C., Gevers, D., Knight, R., Abubucker, S., Badger, J. H., Chinwalla, A. T., Creasy, H. H., Earl, A. M., Fitzgerald, M. G., Fulton, R. S., Giglio, M. G., Hallsworth-Pepin, K., Lobos, E. A., Madupu, R., Magrini, V., Martin, J. C., Mitreva, M., Muzny, D. M., Sodergren, E. J., … White, O. (2012). Structure, function and diversity of the healthy human microbiome. *Nature 2012 486:7402*, *486*(7402), 207–214. https://doi.org/10.1038/nature11234

Hynes, W. L., & Walton, S. L. (2000). Hyaluronidases of Gram-positive bacteria. *FEMS Microbiology Letters*, *183*(2), 201–207. https://doi.org/10.1111/J.1574-6968.2000.TB08958.X

Ibberson, C. B., Jones, C. L., Singh, S., Wise, M. C., Hart, M. E., Zurawski, D. V., & Horswill, A. R. (2014). Staphylococcus aureus hyaluronidase is a CodY-regulated virulence factor. *Infection and Immunity*, *82*(10), 4253–4264. https://doi.org/10.1128/IAI.01710-14

Ibberson, C. B., Parlet, C. P., Kwiecinski, J., Crosby, H. A., Meyerholz, D. K., & Horswill, A. R. (2016). Hyaluronan Modulation Impacts Staphylococcus aureus Biofilm Infection. *Infection and Immunity*, *84*(6), 1917–1929. https://doi.org/10.1128/IAI.01418-15

Immerman, I., Ramos, N. L., Katz, G. M., Hutzler, L. H., Phillips, M. S., & Bosco, J. A. (2012). The persistence of Staphylococcus aureus decolonization after mupirocin and topical chlorhexidine: implications for patients requiring multiple or delayed procedures. *The Journal of Arthroplasty*, *27*(6), 870–876. https://doi.org/10.1016/J.ARTH.2012.01.010

Inoshima, I., Inoshima, N., Wilke, G. A., Powers, M. E., Frank, K. M., Wang, Y., & Wardenburg, J. B. (2011). A Staphylococcus aureus pore-forming toxin subverts the activity of ADAM10 to cause lethal infection in mice. *Nature Medicine*, *17*(10), 1310–1314. https://doi.org/10.1038/NM.2451

Ito, T., Hiramatsu, K., Oliveira, D. C., De Lencastre, H., Zhang, K., Westh, H., O'Brien, F., Giffard, P. M., Coleman, D., Tenover, F. C., Boyle-Vavra, S., Skov, R. L., Enright, M. C., Kreiswirth, B., Kwan, S. K., Grundmann, H., Laurent, F., Sollid, J. E., Kearns, A. M., … Soderquist, B. (2009). Classification of staphylococcal cassette chromosome mec (SCCmec): guidelines for reporting novel SCCmec elements. *Antimicrobial Agents and*

Chemotherapy, *53*(12), 4961–4967. https://doi.org/10.1128/AAC.00579-09

Ito, T., Katayama, Y., Asada, K., Mori, N., Tsutsumimoto, K., Tiensasitorn, C., & Hiramatsu, K. (2001). Structural comparison of three types of staphylococcal cassette chromosome mec integrated in the chromosome in methicillin-resistant Staphylococcus aureus. *Antimicrobial Agents and Chemotherapy*, *45*(5), 1323–1336. https://doi.org/10.1128/AAC.45.5.1323-1336.2001

Ito, T., Katayama, Y., & Hiramatsu, K. (1999). Cloning and nucleotide sequence determination of the entire mec DNA of pre-methicillin-resistant Staphylococcus aureus N315. *Antimicrobial Agents and Chemotherapy*, *43*(6), 1449–1458. https://doi.org/10.1128/AAC.43.6.1449

Iwase, T., Uehara, Y., Shinji, H., Tajima, A., Seo, H., Takada, K., Agata, T., & Mizunoe, Y. (2010). Staphylococcus epidermidis Esp inhibits Staphylococcus aureus biofilm formation and nasal colonization. *Nature*, *465*(7296), 346–349. https://doi.org/10.1038/NATURE09074

J, D., & D, D. (2010). Origins and evolution of antibiotic resistance. *Microbiology and Molecular Biology Reviews : MMBR*, *74*(3), 9–16. https://doi.org/10.1128/MMBR.00016-10

Janek, D., Zipperer, A., Kulik, A., Krismer, B., & Peschel, A. (2016). High Frequency and Diversity of Antimicrobial Activities Produced by Nasal Staphylococcus Strains against Bacterial Competitors. *PLoS Pathogens*, *12*(8). https://doi.org/10.1371/JOURNAL.PPAT.1005812

Janeway, C. A. (1989). Approaching the asymptote? Evolution and revolution in immunology. *Cold Spring Harbor Symposia on Quantitative Biology*, *54 Pt*

1(1), 1–13. https://doi.org/10.1101/SQB.1989.054.01.003

Jardetzky, T. S., Brown, J. H., Gorga, J. C., Stern, L. J., Urban, R. G., Chi, Y. I., Stauffacher, C., Strominger, J. L., & Wiley, D. C. (1994). Three-dimensional structure of a human class II histocompatibility molecule complexed with superantigen. *Nature*, *368*(6473), 711–718. https://doi.org/10.1038/368711A0

Jarnik, M., De Viragh, P. A., Schärer, E., Bundman, D., Simon, M. N., Roop, D. R., & Steven, A. C. (2002). Quasi-Normal Cornified Cell Envelopes in Loricrin Knockout Mice Imply the Existence of a Loricrin Backup System. *Journal of Investigative Dermatology*, *118*(1), 102–109. https://doi.org/10.1046/J.0022-202X.2001.01661.X

Jayasinghe, L., & Bayley, H. (2005). The leukocidin pore: Evidence for an octamer with four LukF subunits and four LukS subunits alternating around a central axis. *Protein Science*, *14*(10), 2550–2561. https://doi.org/10.1110/PS.051648505

Jensen, S. O., & Lyon, B. R. (2009). Genetics of antimicrobial resistance in Staphylococcus aureus. *Future Microbiology*, *4*(5), 565–582. https://doi.org/10.2217/FMB.09.30

Jevons, M. P. (1961). "Celbenin" - resistant Staphylococci. *British Medical Journal*, *1*(5219), 124. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC1952888/

Jiménez, J. N., Ocampo, A. M., Vanegas, J. M., Rodriguez, E. A., Mediavilla, J. R., Chen, L., Muskus, C. E., Vélez, L. A., Rojas, C., Restrepo, A. V.,

Ospina, S., Garcés, C., Franco, L., Bifani, P., Kreiswirth, B. N., & Correa, M. M. (2012). CC8 MRSA Strains Harboring SCCmec Type IVc are Predominant in Colombian Hospitals. *PLOS ONE*, *7*(6), e38576. https://doi.org/10.1371/JOURNAL.PONE.0038576

Jin, T., Bokarewa, M., Foster, T., Mitchell, J., Higgins, J., & Tarkowski, A. (2004). Staphylococcus aureus resists human defensins by production of staphylokinase, a novel bacterial evasion mechanism. *Journal of Immunology (Baltimore, Md. : 1950)*, *172*(2), 1169–1176. https://doi.org/10.4049/JIMMUNOL.172.2.1169

Jo, I., Hong, S., Lee, M., Song, S., Kim, J. S., Mitra, A. K., Hyun, J., Lee, K., & Ha, N. C. (2017). Stoichiometry and mechanistic implications of the MacAB-TolC tripartite efflux pump. *Biochemical and Biophysical Research Communications*, *494*(3–4), 668–673. https://doi.org/10.1016/J.BBRC.2017.10.102

Johnson, A. P., Warner, M., & Livermore, D. M. (2000). Activity of linezolid against multi-resistant gram-positive bacteria from diverse hospitals in the United Kingdom. *The Journal of Antimicrobial Chemotherapy*, *45*(2), 225–230. https://doi.org/10.1093/JAC/45.2.225

Jokinen, E., Laine, J., Huttunen, R., Rahikka, P., Huhtala, H., Vuento, R., Vuopio, J., & Syrjänen, J. (2017). Comparison of outcome and clinical characteristics of bacteremia caused by methicillin-resistant, penicillin-resistant and penicillin-susceptible Staphylococcus aureus strains. *Http://Dx.Doi.Org/10.1080/23744235.2017.1292046*, *49*(7), 493–500.

https://doi.org/10.1080/23744235.2017.1292046

Jolivet-Gougeon, A., & Bonnaure-Mallet, M. (2014). Biofilms as a mechanism of bacterial resistance. *Drug Discovery Today. Technologies*, *11*(1), 49–56. https://doi.org/10.1016/J.DDTEC.2014.02.003

Jonas, B. M., Murray, B. E., & Weinstock, G. M. (2001). Characterization of emeA, a NorA homolog and multidrug resistance efflux pump, in Enterococcus faecalis. *Antimicrobial Agents and Chemotherapy*, *45*(12), 3574–3579. https://doi.org/10.1128/AAC.45.12.3574-3579.2001

Jones, C. H., Tuckman, M., Howe, A. Y. M., Orlowski, M., Mullen, S., Chan, K., & Bradford, P. A. (2006). Diagnostic PCR analysis of the occurrence of methicillin and tetracycline resistance genes among Staphylococcus aureus isolates from phase 3 clinical trials of tigecycline for complicated skin and skin structure infections. *Antimicrobial Agents and Chemotherapy*, *50*(2), 505–510. https://doi.org/10.1128/AAC.50.2.505-510.2006

Jones, R. N., Fritsche, T. R., Sader, H. S., & Ross, J. E. (2006). Activity of retapamulin (SB-275833), a novel pleuromutilin, against selected resistant gram-positive cocci. *Antimicrobial Agents and Chemotherapy*, *50*(7), 2583–2586. https://doi.org/10.1128/AAC.01432-05

Jones, R. N., Fritsche, T. R., Sader, H. S., & Ross, J. E. (2007a). LEADER surveillance program results for 2006: an activity and spectrum analysis of linezolid using clinical isolates from the United States (50 medical centers). *Diagnostic Microbiology and Infectious Disease*, *59*(3), 309–317. https://doi.org/10.1016/J.DIAGMICROBIO.2007.06.004

Jones, R. N., Fritsche, T. R., Sader, H. S., & Ross, J. E. (2007b). Zyvox Annual Appraisal of Potency and Spectrum Program Results for 2006: an activity and spectrum analysis of linezolid using clinical isolates from 16 countries. *Diagnostic Microbiology and Infectious Disease*, *59*(2), 199–209. https://doi.org/10.1016/J.DIAGMICROBIO.2007.06.001

Jung, D.-J., An, J.-H., Kurokawa, K., Jung, Y.-C., Kim, M.-J., Aoyagi, Y., Matsushita, M., Takahashi, S., Lee, H.-S., Takahashi, K., & Lee, B. L. (2012). Specific serum Ig recognizing staphylococcal wall teichoic acid induces complement-mediated opsonophagocytosis against Staphylococcus aureus. *Journal of Immunology (Baltimore, Md. : 1950)*, *189*(10), 4951–4959. https://doi.org/10.4049/JIMMUNOL.1201294

Kahsay, A., Mihret, A., Abebe, T., & Andualem, T. (2014). Isolation and antimicrobial susceptibility pattern of Staphylococcus aureus in patients with surgical site infection at Debre Markos Referral Hospital, Amhara Region, Ethiopia. *Archives of Public Health = Archives Belges de Sante Publique*, *72*(1). https://doi.org/10.1186/2049-3258-72-16

Kalmeijer, M. D., Nieuwland-Bollen, E. van, Bogaers-Hofman, D., Baere, G. A. J. de, & Kluytmans, J. A. J. W. (2000). Nasal carriage of Staphylococcus aureus is a major risk factor for surgical-site infections in orthopedic surgery. *Infection Control and Hospital Epidemiology*, *21*(5), 319–323. https://doi.org/10.1086/501763

Kanoksil, M., Jatapai, A., Peacock, S. J., & Limmathurotsakul, D. (2013). Epidemiology, microbiology and mortality associated with community-

acquired bacteremia in northeast Thailand: a multicenter surveillance study. *PloS One*, *8*(1). https://doi.org/10.1371/JOURNAL.PONE.0054714

Kapral, F. A. (1966). Clumping of Staphylococcus aureus in the peritoneal cavity of mice. *Journal of Bacteriology*, *92*(4), 1188–1195. https://doi.org/10.1128/JB.92.4.1188-1195.1966

Kapral, F. A., Smith, S., & Lal, D. (1992). The esterification of fatty acids by Staphylococcus aureus fatty acid modifying enzyme (FAME) and its inhibition by glycerides. *Journal of Medical Microbiology*, *37*(4), 235–237. https://doi.org/10.1099/00222615-37-4-235

Karauzum, H., & Datta, S. K. (2017). Adaptive immunity against Staphylococcus aureus. *Current Topics in Microbiology and Immunology*, *409*, 419. https://doi.org/10.1007/82_2016_1

Kaspar, U., Kriegeskorte, A., Schubert, T., Peters, G., Rudack, C., Pieper, D. H., Wos-Oxley, M., & Becker, K. (2016). The culturome of the human nose habitats reveals individual bacterial fingerprint patterns. *Environmental Microbiology*, *18*(7), 2130–2142. https://doi.org/10.1111/1462-2920.12891

Katayama, Y., Baba, T., Sekine, M., Fukuda, M., & Hiramatsu, K. (2013). Beta-hemolysin promotes skin colonization by Staphylococcus aureus. *Journal of Bacteriology*, *195*(6), 1194–1203. https://doi.org/10.1128/JB.01786-12/ASSET/A77D6D64-9C44-4AC6-8D22-7E98AF906426/ASSETS/GRAPHIC/ZJB9990924630005.JPEG

Katayama, Y., Ito, T., & Hiramatsu, K. (2000). A new class of genetic element, staphylococcus cassette chromosome mec, encodes methicillin resistance in

Staphylococcus aureus. *Antimicrobial Agents and Chemotherapy*, *44*(6), 1549–1555. https://doi.org/10.1128/AAC.44.6.1549-1555.2000

Kateete, D. P., Asiimwe, B. B., Mayanja, R., Mujuni, B., Bwanga, F., Najjuka, C. F., Källander, K., & Rutebemberwa, E. (2019). Nasopharyngeal carriage, spa types and antibiotic susceptibility profiles of staphylococcus aureus from healthy children less than 5 years in eastern uganda. *BMC Infectious Diseases*, *19*(1), 1–10. https://doi.org/10.1186/S12879-019-4652-5/FIGURES/2

Kateete, D. P., Namazzi, S., Okee, M., Okeng, A., Baluku, H., Musisi, N. L., Katabazi, F. A., Joloba, M. L., Ssentongo, R., & Najjuka, F. C. (2011). High prevalence of Methicillin resistant Staphylococcus aureus in the surgical units of Mulago hospital in Kampala, Uganda. *BMC Research Notes*, *4*(1), 1–5. https://doi.org/10.1186/1756-0500-4-326/COMMENTS

Kawabata, S. ichiro, Morita, T., Iwanaga, S., & Igarashi, H. (1985). Staphylocoagulase-binding region in human prothrombin. *Journal of Biochemistry*, *97*(1), 325–331. https://doi.org/10.1093/OXFORDJOURNALS.JBCHEM.A135057

Kawai, T., & Akira, S. (2011). Toll-like receptors and their crosstalk with other innate receptors in infection and immunity. *Immunity*, *34*(5), 637–650. https://doi.org/10.1016/J.IMMUNI.2011.05.006

Kehrenberg, C., Aarestrup, F. M., & Schwarz, S. (2007). IS21-558 insertion sequences are involved in the mobility of the multiresistance gene cfr. *Antimicrobial Agents and Chemotherapy*, *51*(2), 483–487.

https://doi.org/10.1128/AAC.01340-06

Kehrenberg, C., Cuny, C., Strommenger, B., Schwarz, S., & Witte, W. (2009). Methicillin-resistant and -susceptible Staphylococcus aureus strains of clonal lineages ST398 and ST9 from swine carry the multidrug resistance gene cfr. *Antimicrobial Agents and Chemotherapy*, *53*(2), 779–781. https://doi.org/10.1128/AAC.01376-08

Kehrenberg, C., Ojo, K. K., & Schwarz, S. (2004). Nucleotide sequence and organization of the multiresistance plasmid pSCFS1 from Staphylococcus sciuri. *The Journal of Antimicrobial Chemotherapy*, *54*(5), 936–939. https://doi.org/10.1093/JAC/DKH457

Kehrenberg, C., & Schwarz, S. (2004). fexA, a novel Staphylococcus lentus gene encoding resistance to florfenicol and chloramphenicol. *Antimicrobial Agents and Chemotherapy*, *48*(2), 615–618. https://doi.org/10.1128/AAC.48.2.615-618.2004

Kehrenberg, C., Schwarz, S., Jacobsen, L., Hansen, L. H., & Vester, B. (2005). A new mechanism for chloramphenicol, florfenicol and clindamycin resistance: methylation of 23S ribosomal RNA at A2503. *Molecular Microbiology*, *57*(4), 1064–1073. https://doi.org/10.1111/J.1365-2958.2005.04754.X

Kejela, T., & Bacha, K. (2013). Prevalence and antibiotic susceptibility pattern of methicillin-resistant Staphylococcus aureus (MRSA) among primary school children and prisoners in Jimma Town, Southwest Ethiopia. *Annals of Clinical Microbiology and Antimicrobials*, *12*(1). https://doi.org/10.1186/1476-0711-12-11

Keren, I., Kaldalu, N., Spoering, A., Wang, Y., & Lewis, K. (2004). Persister cells and tolerance to antimicrobials. *FEMS Microbiology Letters*, *230*(1), 13–18. https://doi.org/10.1016/S0378-1097(03)00856-5

Kessler, C. M., Nussbaum, E., & Tuazon, C. U. (1991). Disseminated intravascular coagulation associated with Staphylococcus aureus septicemia is mediated by peptidoglycan-induced platelet aggregation. *The Journal of Infectious Diseases*, *164*(1), 101–107. https://doi.org/10.1093/INFDIS/164.1.101

Khan, S., Sallum, U. W., Zheng, X., Nau, G. J., & Hasan, T. (2014). Rapid optical determination of β-lactamase and antibiotic activity. *BMC Microbiology*, *14*(1), 1–14. https://doi.org/10.1186/1471-2180-14-84/TABLES/5

Khoshnood, S., Heidary, M., Asadi, A., Soleimani, S., Motahar, M., Savari, M., Saki, M., & Abdi, M. (2019). A review on mechanism of action, resistance, synergism, and clinical implications of mupirocin against Staphylococcus aureus. *Biomedicine & Pharmacotherapy*, *109*, 1809–1818. https://doi.org/10.1016/J.BIOPHA.2018.10.131

Kiedrowski, M. R., Crosby, H. A., Hernandez, F. J., Malone, C. L., McNamara, J. O., & Horswill, A. R. (2014). Staphylococcus aureus Nuc2 is a functional, surface-attached extracellular nuclease. *PloS One*, *9*(4). https://doi.org/10.1371/JOURNAL.PONE.0095574

Kiedrowski, M. R., Kavanaugh, J. S., Malone, C. L., Mootz, J. M., Voyich, J. M., Smeltzer, M. S., Bayles, K. W., & Horswill, A. R. (2011). Nuclease modulates biofilm formation in community-associated methicillin-resistant

Staphylococcus aureus. *PloS One*, *6*(11). https://doi.org/10.1371/JOURNAL.PONE.0026714

Kim, H. K., Falugi, F., Thomer, L., Missiakas, D. M., & Schneewind, O. (2015). Protein A suppresses immune responses during Staphylococcus aureus bloodstream infection in guinea pigs. *MBio*, *6*(1). https://doi.org/10.1128/MBIO.02369-14

Kim, J., Urban, R. G., Strominger, J. L., & Wiley, D. C. (1994). Toxic shock syndrome toxin-1 complexed with a class II major histocompatibility molecule HLA-DR1. *Science (New York, N.Y.)*, *266*(5192), 1870–1874. https://doi.org/10.1126/SCIENCE.7997880

Kirby, W. M. M. (1944). EXTRACTION OF A HIGHLY POTENT PENICILLIN INACTIVATOR FROM PENICILLIN RESISTANT STAPHYLOCOCCI. *Science (New York, N.Y.)*, *99*(2579), 452–453. https://doi.org/10.1126/SCIENCE.99.2579.452

Kirst, H. A., Thompson, D. G., & Nicas, T. I. (1998). Historical yearly usage of vancomycin. *Antimicrobial Agents and Chemotherapy*, *42*(5), 1303–1304. https://doi.org/10.1128/AAC.42.5.1303

Kisich, K. O., Howell, M. D., Boguniewicz, M., Heizer, H. R., Watson, N. U., & Leung, D. Y. M. (2007). The Constitutive Capacity of Human Keratinocytes to Kill Staphylococcus aureus Is Dependent on β-Defensin 3. *Journal of Investigative Dermatology*, *127*(10), 2368–2380. https://doi.org/10.1038/SJ.JID.5700861

Kissner, T. L., Ruthel, G., Alam, S., Ulrich, R. G., Fernandez, S., & Saikh, K. U.

(2011). Activation of MyD88 signaling upon staphylococcal enterotoxin binding to MHC class II molecules. *PloS One*, *6*(1). https://doi.org/10.1371/JOURNAL.PONE.0015985

Klein, E. Y., Mojica, N., Jiang, W., Cosgrove, S. E., Septimus, E., Morgan, D. J., & Laxminarayan, R. (2017). Trends in Methicillin-Resistant Staphylococcus aureus Hospitalizations in the United States, 2010-2014. *Clinical Infectious Diseases*, *65*(11), 1921–1923. https://doi.org/10.1093/CID/CIX640

Klevens, R. M., Morrison, M. A., Nadle, J., Petit, S., Gershman, K., Ray, S., Harrison, L. H., Lynfield, R., Dumyati, G., Townes, J. M., Craig, A. S., Zell, E. R., Fosheim, G. E., McDougal, L. K., Carey, R. B., & Fridkin, S. K. (2007). Invasive methicillin-resistant Staphylococcus aureus infections in the United States. *JAMA*, *298*(15), 1763–1771. https://doi.org/10.1001/JAMA.298.15.1763

Kluytmans, J. A. J. W., & Wertheim, H. F. L. (2005). Nasal Carriage of Staphylococcus aureus and Prevention of Nosocomial Infections. *Infection 2005 33:1*, *33*(1), 3–8. https://doi.org/10.1007/S15010-005-4012-9

Kluytmans, J., van Belkum, A., & Verbrugh, H. (1997). Nasal carriage of Staphylococcus aureus: epidemiology, underlying mechanisms, and associated risks. *Clinical Microbiology Reviews*, *10*(3), 505–520. https://doi.org/10.1128/CMR.10.3.505

Kobayashi, S. D., Malachowa, N., & Deleo, F. R. (2015). Pathogenesis of Staphylococcus aureus Abscesses. *The American Journal of Pathology*, *185*(6), 1518. https://doi.org/10.1016/J.AJPATH.2014.11.030

Koch, P. J., De Viragh, P. A., Scharer, E., Bundman, D., Longley, M. A., Bickenbach, J., Kawachi, Y., Suga, Y., Zhou, Z., Huber, M., Hohl, D., Kartasova, T., Jarnik, M., Steven, A. C., & Roop, D. R. (2000). Lessons from Loricrin-Deficient MiceCompensatory Mechanisms Maintaining Skin Barrier Function in the Absence of a Major Cornified Envelope Protein. *Journal of Cell Biology*, *151*(2), 389–400. https://doi.org/10.1083/JCB.151.2.389

Köck, R., Becker, K., Cookson, B., van Gemert-Pijnen, J. E., Harbarth, S., Kluytmans, J., Mielke, M., Peters, G., Skov, R. L., Struelens, M. J., Tacconelli, E., Torné, A. N., Witte, W., & Friedrich, A. W. (2010). Methicillin-resistant Staphylococcus aureus (MRSA): burden of disease and control challenges in Europe. *Euro Surveillance : Bulletin Europeen Sur Les Maladies Transmissibles = European Communicable Disease Bulletin*, *15*(41), 1–9. https://doi.org/10.2807/ESE.15.41.19688-EN

Kolata, J., Bode, L. G. M., Holtfreter, S., Steil, L., Kusch, H., Holtfreter, B., Albrecht, D., Hecker, M., Engelmann, S., Van Belkum, A., Völker, U., & Bröker, B. M. (2011). Distinctive patterns in the human antibody response to Staphylococcus aureus bacteremia in carriers and non-carriers. *PROTEOMICS*, *11*(19), 3914–3927. https://doi.org/10.1002/PMIC.201000760

Kong, E. F., Johnson, J. K., & Jabra-Rizk, M. A. (2016). Community-Associated Methicillin-Resistant Staphylococcus aureus: An Enemy amidst Us. *PLoS Pathogens*, *12*(10). https://doi.org/10.1371/JOURNAL.PPAT.1005837

Kotey, F. C. N., Awugah, S. A., Dayie, N. T. K. D., Tetteh-Quarcoo, P. B., Duodu, S., Osei, M. M., Bentum, J. N., Nyarko, M. Y., Neizer, M. L., Alsharif, K. F., Halawani, I. F., Alzahrani, K. J., & Donkor, E. S. (2022). High prevalence of methicillin-resistant Staphylococcus aureus carriage among infants at the Children's Hospital, Accra, Ghana. *Journal of Infection in Developing Countries*, *16*(9), 1450–1457. https://doi.org/10.3855/JIDC.14839

Kourtesi, C., Ball, A. R., Huang, Y.-Y., Jachak, S. M., Vera, D. M. A., Khondkar, P., Gibbons, S., Hamblin, M. R., & Tegos, G. P. (2013). Microbial efflux systems and inhibitors: approaches to drug discovery and the challenge of clinical implementation. *The Open Microbiology Journal*, *7*(1), 34–52. https://doi.org/10.2174/1874285801307010034

Kourtis, A. P., Hatfield, K., Baggs, J., Mu, Y., See, I., Epson, E., Nadle, J., Kainer, M. A., Dumyati, G., Petit, S., Ray, S. M., Ham, D., Capers, C., Ewing, H., Coffin, N., McDonald, L. C., Jernigan, J., & Cardo, D. (2019). Vital Signs: Epidemiology and Recent Trends in Methicillin-Resistant and in Methicillin-Susceptible Staphylococcus aureus Bloodstream Infections — United States. *MMWR. Morbidity and Mortality Weekly Report*, *68*(9), 214–219. https://doi.org/10.15585/MMWR.MM6809E1

Kovács, E., Sahin-Tóth, J., Tóthpál, A., van der Linden, M., Tirczka, T., & Dobay, O. (2020). Co-carriage of Staphylococcus aureus, Streptococcus pneumoniae, Haemophilus influenzae and Moraxella catarrhalis among three different age categories of children in Hungary. *PLoS ONE*, *15*(2).

https://doi.org/10.1371/JOURNAL.PONE.0229021

Krismer, B., Weidenmaier, C., Zipperer, A., & Peschel, A. (2017). The commensal lifestyle of Staphylococcus aureus and its interactions with the nasal microbiota. *Nature Reviews. Microbiology*, *15*(11), 675–687. https://doi.org/10.1038/NRMICRO.2017.104

Kroh, H. K., Panizzi, P., & Bock, P. E. (2009). Von Willebrand factor-binding protein is a hysteretic conformational activator of prothrombin. *Proceedings of the National Academy of Sciences of the United States of America*, *106*(19), 7786–7791. https://doi.org/10.1073/PNAS.0811750106

Kruse, T., Levisson, M., de Vos, W. M., & Smidt, H. (2014). vanI: a novel D-Ala-D-Lac vancomycin resistance gene cluster found in Desulfitobacterium hafniense. *Microbial Biotechnology*, *7*(5), 456–466. https://doi.org/10.1111/1751-7915.12139

Kumar, A., & Schweizer, H. P. (2005). Bacterial resistance to antibiotics: active efflux and reduced uptake. *Advanced Drug Delivery Reviews*, *57*(10), 1486–1513. https://doi.org/10.1016/J.ADDR.2005.04.004

Kumar, S., Mukherjee, M. M., & Varela, M. F. (2013). Modulation of Bacterial Multidrug Resistance Efflux Pumps of the Major Facilitator Superfamily. *International Journal of Bacteriology*, *2013*, 1–15. https://doi.org/10.1155/2013/204141

Kuo, I. H., Carpenter-Mendini, A., Yoshida, T., McGirt, L. Y., Ivanov, A. I., Barnes, K. C., Gallo, R. L., Borkowski, A. W., Yamasaki, K., Leung, D. Y., Georas, S. N., De Benedetto, A., & Beck, L. A. (2013). Activation of

epidermal toll-like receptor 2 enhances tight junction function: implications for atopic dermatitis and skin barrier repair. *The Journal of Investigative Dermatology*, *133*(4), 988–998. https://doi.org/10.1038/JID.2012.437

Kurlenda, J., & Grinholc, M. (2012). Alternative therapies in staphylococcus aureus diseases. *Acta Biochimica Polonica*, *59*(2), 171–184. https://doi.org/10.18388/abp.2012_2136

Kuroda, M., Ohta, T., Uchiyama, I., Baba, T., Yuzawa, H., Kobayashi, I., Kobayashi, N., Cui, L., Oguchi, A., Aoki, K. I., Nagai, Y., Lian, J. Q., Ito, T., Kanamori, M., Matsumaru, H., Maruyama, A., Murakami, H., Hosoyama, A., Mizutani-Ui, Y., … Hiramatsu, K. (2001). Whole genome sequencing of meticillin-resistant Staphylococcus aureus. *Lancet*, *357*(9264), 1225–1240. https://doi.org/10.1016/S0140-6736(00)04403-2

Kurokawa, K., Jung, D. J., An, J. H., Fuchs, K., Jeon, Y. J., Kim, N. H., Li, X., Tateishi, K., Park, J. A., Xia, G., Matsushita, M., Takahashi, K., Park, H. J., Peschel, A., & Lee, B. L. (2013). Glycoepitopes of staphylococcal wall teichoic acid govern complement-mediated opsonophagocytosis via human serum antibody and mannose-binding lectin. *The Journal of Biological Chemistry*, *288*(43), 30956–30968. https://doi.org/10.1074/JBC.M113.509893

Kwiecinski, J., Jacobsson, G., Karlsson, M., Zhu, X., Wang, W., Bremell, T., Josefsson, E., & Jin, T. (2013). Staphylokinase promotes the establishment of Staphylococcus aureus skin infections while decreasing disease severity. *The Journal of Infectious Diseases*, *208*(6), 990–999.

https://doi.org/10.1093/INFDIS/JIT288

Kwiecinski, J., Peetermans, M., Liesenborghs, L., Na, M., Björnsdottir, H., Zhu, X., Jacobsson, G., Johansson, B. R., Geoghegan, J. A., Foster, T. J., Josefsson, E., Bylund, J., Verhamme, P., & Jin, T. (2016). Staphylokinase Control of Staphylococcus aureus Biofilm Formation and Detachment Through Host Plasminogen Activation. *The Journal of Infectious Diseases*, *213*(1), 139–148. https://doi.org/10.1093/INFDIS/JIV360

Labi, A. K., Obeng-Nkrumah, N., Nartey, E. T., Bjerrum, S., Adu-Aryee, N. A., Ofori-Adjei, Y. A., Yawson, A. E., & Newman, M. J. (2018). Antibiotic use in a tertiary healthcare facility in Ghana: a point prevalence survey. *Antimicrobial Resistance and Infection Control*, *7*(1). https://doi.org/10.1186/S13756-018-0299-Z

Labischinski, H. (1992). Consequences of the interaction of beta-lactam antibiotics with penicillin binding proteins from sensitive and resistant Staphylococcus aureus strains. *Medical Microbiology and Immunology*, *181*(5), 241–265. https://doi.org/10.1007/BF00198846

Ladhani, S. (2001). Recent developments in staphylococcal scalded skin syndrome. *Clinical Microbiology and Infection : The Official Publication of the European Society of Clinical Microbiology and Infectious Diseases*, *7*(6), 301–307. https://doi.org/10.1046/J.1198-743X.2001.00258.X

Ladhani, S. (2003). Understanding the mechanism of action of the exfoliative toxins of Staphylococcus aureus. *FEMS Immunology and Medical Microbiology*, *39*(2), 181–189. https://doi.org/10.1016/S0928-

8244(03)00225-6

LAM, G. T., SWEENEY, F. J., WITMER, C. M., & WISE, R. I. (1963). ABSCESS-FORMING FACTOR(S) PRODUCED BY STAPHYLOCOCCUS AUREUS. II. *Journal of Bacteriology*, *86*(4), 87–91. https://doi.org/10.1128/jb.86.4.611-615.1963

Lambert, P. A. (2002). Cellular impermeability and uptake of biocides and antibiotics in Gram-positive bacteria and mycobacteria. *Journal of Applied Microbiology*, *92 Suppl*(1), 46S-54S. https://doi.org/10.1046/J.1365-2672.92.5S1.7.X

Landers, T. F., Cohen, B., Wittum, T. E., & Larson, E. L. (2012). A review of antibiotic use in food animals: perspective, policy, and potential. *Public Health Reports (Washington, D.C. : 1974)*, *127*(1), 4–22. https://doi.org/10.1177/003335491212700103

Laupland, K. B., Ross, T., & Gregson, D. B. (2008). Staphylococcus aureus bloodstream infections: risk factors, outcomes, and the influence of methicillin resistance in Calgary, Canada, 2000-2006. *The Journal of Infectious Diseases*, *198*(3), 336–343. https://doi.org/10.1086/589717

Laurent, T. C., & Fraser, J. R. E. (1992). Hyaluronan1. *The FASEB Journal*, *6*(7), 2397–2404. https://doi.org/10.1096/FASEBJ.6.7.1563592

Leach, K. L., Swaney, S. M., Colca, J. R., McDonald, W. G., Blinn, J. R., Thomasco, L. M. M., Gadwood, R. C., Shinabarger, D., Xiong, L., & Mankin, A. S. (2007). The site of action of oxazolidinone antibiotics in living bacteria and in human mitochondria. *Molecular Cell*, *26*(3), 393–402.

https://doi.org/10.1016/J.MOLCEL.2007.04.005

Lebon, A., Labout, J. A. M., Verbrugh, H. A., Jaddoe, V. W. V., Hofman, A., Van Wamel, W., Moll, H. A., & Van Belkum, A. (2008). Dynamics and determinants of Staphylococcus aureus carriage in infancy: The generation R study. *Journal of Clinical Microbiology*, *46*(10), 3517–3521. https://doi.org/10.1128/JCM.00641-08/ASSET/3F912442-FF8C-4E17-B76D-ABF1ABA92087/ASSETS/GRAPHIC/ZJM0100884590002.JPEG

Lebreton, F., Depardieu, F., Bourdon, N., Fines-Guyon, M., Berger, P., Camiade, S., Leclercq, R., Courvalin, P., & Cattoir, V. (2011). D-Ala-d-Ser VanN-type transferable vancomycin resistance in Enterococcus faecium. *Antimicrobial Agents and Chemotherapy*, *55*(10), 4606–4612. https://doi.org/10.1128/AAC.00714-11

Lee-Sayer, S. S. M., Dong, Y., Arif, A. A., Olsson, M., Brown, K. L., & Johnson, P. (2015). The where, when, how, and why of hyaluronan binding by immune cells. *Frontiers in Immunology*, *6*(MAR). https://doi.org/10.3389/FIMMU.2015.00150

Lee, B. Y., Singh, A., David, M. Z., Bartsch, S. M., Slayton, R. B., Huang, S. S., Zimmer, S. M., Potter, M. A., Macal, C. M., Lauderdale, D. S., Miller, L. G., & Daum, R. S. (2013). The economic burden of community-associated methicillin-resistant Staphylococcus aureus (CA-MRSA). *Clinical Microbiology and Infection : The Official Publication of the European Society of Clinical Microbiology and Infectious Diseases*, *19*(6), 528–536. https://doi.org/10.1111/J.1469-0691.2012.03914.X

Lee, B. Y., Song, Y., Mcglone, S. M., Bailey, R. R., Feura, J. M., Tai, J. H. Y., Lewis, G. J., Wiringa, A. E., Smith, K. J., Muder, R. R., Harrison, L. H., & Piraino, B. (2011). The economic value of screening haemodialysis patients for methicillinresistant Staphylococcus aureus in the USA. *Clinical Microbiology and Infection : The Official Publication of the European Society of Clinical Microbiology and Infectious Diseases*, *17*(11), 1717. https://doi.org/10.1111/J.1469-0691.2011.03525.X

Lee, B. Y., Wiringa, A. E., Bailey, R. R., Goyal, V., Lewis, G. J., Tsui, B. Y. K., Smith, K. J., & Muder, R. R. (2010). Screening Cardiac Surgery Patients for MRSA: An Economic Computer Model. *The American Journal of Managed Care*, *16*(7), e163. /pmc/articles/PMC3763192/

Lee, B. Y., Wiringa, A. E., Mitgang, E. A., McGlone, S. M., Afriyie, A. N., Song, Y., & Beigi, R. H. (2011). Routine Pre-cesarean Staphylococcus aureus Screening and Decolonization: A Cost-Effectiveness Analysis. *The American Journal of Managed Care*, *17*(10), 693. /pmc/articles/PMC3836821/

Lee, C. Y., Schmidt, J. J., Johnson-Winegar, A. D., Spero, L., & Iandolo, J. J. (1987). Sequence determination and comparison of the exfoliative toxin A and toxin B genes from Staphylococcus aureus. *Journal of Bacteriology*, *169*(9), 3904–3909. https://doi.org/10.1128/JB.169.9.3904-3909.1987

Lee, W. L., Harrison, R. E., & Grinstein, S. (2003). Phagocytosis by neutrophils. *Microbes and Infection*, *5*(14), 1299–1306. https://doi.org/10.1016/j.micinf.2003.09.014

Lee, Y. D., & Park, J. H. (2016). Phage Conversion for β-Lactam Antibiotic

Resistance of Staphylococcus aureus from Foods. *J. Microbiol. Biotechnol.*, *26*(2), 263–269. https://doi.org/10.4014/JMB.1508.08042

Lekstrom-Himes, J. A., & Gallin, J. I. (2000). Immunodeficiency diseases caused by defects in phagocytes. *The New England Journal of Medicine*, *343*(23), 1703–1714. https://doi.org/10.1056/NEJM200012073432307

Levy, S. B., & Bonnie, M. (2004). Antibacterial resistance worldwide: causes, challenges and responses. *Nature Medicine*, *10*(12 Suppl), S122–S129. https://doi.org/10.1038/NM1145

Ley, K., Laudanna, C., Cybulsky, M. I., & Nourshargh, S. (2007). Getting to the site of inflammation: the leukocyte adhesion cascade updated. *Nature Reviews Immunology 2007 7:9*, *7*(9), 678–689. https://doi.org/10.1038/nri2156

Li, H., Llera, A., Malchiodi, E. L., & Mariuzza, R. A. (1999). The structural basis of T cell activation by superantigens. *Annual Review of Immunology*, *17*, 435–466. https://doi.org/10.1146/ANNUREV.IMMUNOL.17.1.435

Liebl, W., & Götz, F. (1986). Studies on lipase directed export of Escherichia coli beta-lactamase in Staphylococcus carnosus. *Molecular & General Genetics : MGG*, *204*(1), 166–173. https://doi.org/10.1007/BF00330205

LIJNEN, H. R., VAN HOEF, B., & COLLEN, D. (1993). Interaction of staphylokinase with different molecular forms of plasminogen. *European Journal of Biochemistry*, *211*(1–2), 91–97. https://doi.org/10.1111/J.1432-1033.1993.TB19873.X

Lijnen, H. R., Van Hoef, B., De Cock, F., Okada, K., Ueshima, S., Matsuo, O., &

Collen, D. (1991). On the mechanism of fibrin-specific plasminogen activation by staphylokinase. *Journal of Biological Chemistry*, *266*(18), 11826–11832. https://doi.org/10.1016/S0021-9258(18)99031-9

Lina, G., Piémont, Y., Godail-Gamot, F., Bes, M., Peter, M. O., Gauduchon, V., Vandenesch, F., & Etienne, J. (1999). Involvement of Panton-Valentine leukocidin-producing Staphylococcus aureus in primary skin infections and pneumonia. *Clinical Infectious Diseases*, *29*(5), 1128–1132. https://doi.org/10.1086/313461/2/29-5-1128-TBL001.GIF

Lindsay, J. A., & Holden, M. T. G. (2004). Staphylococcus aureus: superbug, super genome? *Trends in Microbiology*, *12*(8), 378–385. https://doi.org/10.1016/J.TIM.2004.06.004

Liu, C., Bayer, A., Cosgrove, S. E., Daum, R. S., Fridkin, S. K., Gorwitz, R. J., Kaplan, S. L., Karchmer, A. W., Levine, D. P., Murray, B. E., Rybak, M. J., Talan, D. A., & Chambers, H. F. (2011). Clinical practice guidelines by the infectious diseases society of america for the treatment of methicillin-resistant Staphylococcus aureus infections in adults and children: executive summary. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *52*(3), 285–292. https://doi.org/10.1093/CID/CIR034

Livermore, D. M., Mushtaq, S., Warner, M., & Woodford, N. (2009). Activity of oxazolidinone TR-700 against linezolid-susceptible and -resistant staphylococci and enterococci. *The Journal of Antimicrobial Chemotherapy*, *63*(4), 713–715. https://doi.org/10.1093/JAC/DKP002

Livermore, D. M., Warner, M., Mushtaq, S., North, S., & Woodford, N. (2007). In vitro activity of the oxazolidinone RWJ-416457 against linezolid-resistant and -susceptible staphylococci and enterococci. *Antimicrobial Agents and Chemotherapy*, *51*(3), 1112–1114. https://doi.org/10.1128/AAC.01347-06

Lizak, M., & Yarovinsky, T. O. (2012). Phospholipid scramblase 1 mediates type i interferon-induced protection against staphylococcal α-toxin. *Cell Host & Microbe*, *11*(1), 70–80. https://doi.org/10.1016/J.CHOM.2011.12.004

Locke, J. B., Hilgers, M., & Shaw, K. J. (2009). Mutations in ribosomal protein L3 are associated with oxazolidinone resistance in staphylococci of clinical origin. *Antimicrobial Agents and Chemotherapy*, *53*(12), 5275–5278. https://doi.org/10.1128/AAC.01032-09

Loeb, L. (1903). The Influence of certain Bacteria on the Coagulation of the Blood. *The Journal of Medical Research*, *10*(3), 407. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2105975/

Loeb, M. B., Main, C., Eady, A., & Walkers-Dilks, C. (2003). Antimicrobial drugs for treating methicillin-resistant Staphylococcus aureus colonization. *The Cochrane Database of Systematic Reviews*, *2010*(4). https://doi.org/10.1002/14651858.CD003340

Lohan, K., Sangwan, J., Mane, P., & Lathwal, S. (2021). Prevalence pattern of MRSA from a rural medical college of North India: A cause of concern. *Journal of Family Medicine and Primary Care*, *10*(2), 752–757. https://doi.org/10.4103/jfmpc.jfmpc_1527_20

Long, J. P., & Kapral, F. A. (1993). Host response to coagulase-negative

staphylococci in abscesses induced within mice. *Journal of Medical Microbiology*, *39*(3), 191–195. https://doi.org/10.1099/00222615-39-3-191

Long, K. S., Poehlsgaard, J., Kehrenberg, C., Schwarz, S., & Vester, B. (2006). The Cfr rRNA methyltransferase confers resistance to Phenicols, Lincosamides, Oxazolidinones, Pleuromutilins, and Streptogramin A antibiotics. *Antimicrobial Agents and Chemotherapy*, *50*(7), 2500–2505. https://doi.org/10.1128/AAC.00131-06

Loof, T. G., Mörgelin, M., Johansson, L., Oehmcke, S., Olin, A. I., Dickneite, G., Norrby-Teglund, A., Theopold, U., & Herwald, H. (2011). Coagulation, an ancestral serine protease cascade, exerts a novel function in early immune defense. *Blood*, *118*(9), 2589–2598. https://doi.org/10.1182/BLOOD-2011-02-337568

Lowy, F. D. (1998). Staphylococcus aureus infections. *The New England Journal of Medicine*, *339*(8), 520–532. https://doi.org/10.1056/NEJM199808203390806

Lowy, F. D. (2003). Antimicrobial resistance: the example of Staphylococcus aureus. *Journal of Clinical Investigation*, *111*(9), 1265. https://doi.org/10.1172/JCI18535

Lu, T., Park, J. Y., Parnell, K., Fox, L. K., & McGuire, M. A. (2012). Characterization of fatty acid modifying enzyme activity in staphylococcal mastitis isolates and other bacteria. *BMC Research Notes*, *5*. https://doi.org/10.1186/1756-0500-5-323

Luksiene, Z. (2003). Photodynamic therapy: mechanism of action and ways to

improve the efficiency of treatment. *Medicina (Kaunas, Lithuania)*, *39*(12), 1137–1150. https://europepmc.org/article/med/14704501

Lyell, A. (1967). A review of toxic epidermal necrolysis in Britain. *The British Journal of Dermatology*, *79*(12), 662–671. https://doi.org/10.1111/J.1365-2133.1967.TB11434.X

Lyell, A. (1979). Toxic epidermal necrolysis (the scalded skin syndrome): a reappraisal. *The British Journal of Dermatology*, *100*(1), 69–86. https://doi.org/10.1111/J.1365-2133.1979.TB03571.X

Magnusson, B. J., Doery, H. M., & Gulasekharam, J. (1962). Phospholipase Activity of Staphylococcal Toxin. *Nature 1962 196:4851*, *196*(4851), 270–271. https://doi.org/10.1038/196270b0

Maharjan, M., Sah, A. K., Pyakurel, S., Thapa, S., Maharjan, S., Adhikari, N., Rijal, K. R., Ghimire, P., & Thapa Shrestha, U. (2021). Molecular Confirmation of Vancomycin-Resistant Staphylococcus aureus with vanA Gene from a Hospital in Kathmandu. *International Journal of Microbiology*, *2021*. https://doi.org/10.1155/2021/3847347

Mahon, C., Lehman, D., & Manuselis, G. (2015). Textbook of Diagnostic Microbiology, 5th edition. *Clinical Research and Leadership Faculty Publications*, 254–273. https://hsrc.himmelfarb.gwu.edu/smhs_crl_facpubs/37

Maillard, I., Luthi, F., Diggelmann, H., & Acha-Orbea, H. (1997). Role of the immune response induced by superantigens in the pathogenesis of microbial infections. *Parasitology*, *115 Suppl*(7), 67–78.

https://doi.org/10.1017/S0031182097001807

Maines, M. D. (1997). The heme oxygenase system: a regulator of second messenger gases. *Annual Review of Pharmacology and Toxicology*, *37*, 517–554. https://doi.org/10.1146/ANNUREV.PHARMTOX.37.1.517

Maisch, T., Baier, J., Franz, B., Maier, M., Landthaler, M., Szeimies, R. M., & Bäumler, W. (2007). The role of singlet oxygen and oxygen concentration in photodynamic inactivation of bacteria. *Proceedings of the National Academy of Sciences of the United States of America*, *104*(17), 7223–7228. https://doi.org/10.1073/PNAS.0611328104/ASSET/B84BFC36-CF1B-47A7-BF03-09B490A42195/ASSETS/GRAPHIC/ZPQ01707-5763-M02.JPEG

Maki, D. G., Alvarado, C. J., & Ringer, M. (1991). Prospective randomised trial of povidone-iodine, alcohol, and chlorhexidine for prevention of infection associated with central venous and arterial catheters. *Lancet (London, England)*, *338*(8763), 339–343. https://doi.org/10.1016/0140-6736(91)90479-9

Makris, G., Wright, J. D., Ingham, E., & Holland, K. T. (2004). The hyaluronate lyase of Staphylococcus aureus - a virulence factor? *Microbiology (Reading, England)*, *150*(Pt 6), 2005–2013. https://doi.org/10.1099/MIC.0.26942-0

Malbruny, B., Canu, A., Bozdogan, B., Fantin, B., Zarrouk, V., Dutka-Malen, S., Feger, C., & Leclercq, R. (2002). Resistance to quinupristin-dalfopristin due to mutation of L22 ribosomal protein in Staphylococcus aureus. *Antimicrobial Agents and Chemotherapy*, *46*(7), 2200–2207.

https://doi.org/10.1128/AAC.46.7.2200-2207.2002

Mangwani, N., Kumari, S., & Das, S. (2016). Bacterial biofilms and quorum sensing: fidelity in bioremediation technology. *Biotechnology & Genetic Engineering Reviews*, *32*(1–2), 43–73. https://doi.org/10.1080/02648725.2016.1196554

Maretzky, T., Scholz, F., Köten, B., Proksch, E., Saftig, P., & Reiss, K. (2008). ADAM10-mediated E-cadherin release is regulated by proinflammatory cytokines and modulates keratinocyte cohesion in eczematous dermatitis. *The Journal of Investigative Dermatology*, *128*(7), 1737–1746. https://doi.org/10.1038/SJ.JID.5701242

Mariutti, R. B., Tartaglia, N. R., Seyffert, N., Castro, T. L. de P., Arni, R. K., Azevedo, V. A., LeLoir, Y., & Nishifuji, K. (2017). Exfoliative Toxins of Staphylococcus aureus. *The Rise of Virulence and Antibiotic Resistance in* Staphylococcus Aureus. https://doi.org/10.5772/66528

Marschall, J., Mermel, L. A., Fakih, M., Hadaway, L., Kallen, A., O'Grady, N. P., Pettis, A. M., Rupp, M. E., Sandora, T., Maragakis, L. L., & Yokoe, D. S. (2014). Strategies to Prevent Central Line–Associated Bloodstream Infections in Acute Care Hospitals: 2014 Update. *Infection Control & Hospital Epidemiology*, *35*(7), 753–771. https://doi.org/10.1086/676533

Marshall, S. H., Donskey, C. J., Hutton-Thomas, R., Salata, R. A., & Rice, L. B. (2002). Gene dosage and linezolid resistance in Enterococcus faecium and Enterococcus faecalis. *Antimicrobial Agents and Chemotherapy*, *46*(10), 3334–3336. https://doi.org/10.1128/AAC.46.10.3334-3336.2002

Martineau, F., Picard, F. J., Lansac, N., Ménard, C., Roy, P. H., Ouellette, M., & Bergeron, M. G. (2000). Correlation between the resistance genotype determined by multiplex PCR assays and the antibiotic susceptibility patterns of Staphylococcus aureus and Staphylococcus epidermidis. *Antimicrobial Agents and Chemotherapy*, *44*(2), 231–238. https://doi.org/10.1128/AAC.44.2.231-238.2000/ASSET/AE80FDBB-1708-4E84-AB3A-858CE6D74398/ASSETS/GRAPHIC/AC0200232001.JPEG

Martinez, J. L. (2014). General principles of antibiotic resistance in bacteria. *Drug Discovery Today. Technologies*, *11*(1), 33–39. https://doi.org/10.1016/J.DDTEC.2014.02.001

Masaisa, F., Kayigi, E., Seni, J., Bwanga, F., & Muvunyi, C. M. (2018). Antibiotic Resistance Patterns and Molecular Characterization of Methicillin-Resistant Staphylococcus aureus in Clinical Settings in Rwanda. *The American Journal of Tropical Medicine and Hygiene*, *99*(5), 1239–1245. https://doi.org/10.4269/AJTMH.17-0554

Matono, T., Nagashima, M., Mezaki, K., Motohashi, A., Kutsuna, S., Hayakawa, K., Ohmagari, N., & Kaku, M. (2018). Molecular epidemiology of β-lactamase production in penicillin-susceptible Staphylococcus aureus under high-susceptibility conditions. *Journal of Infection and Chemotherapy*, *24*(2), 153–155. https://doi.org/10.1016/j.jiac.2017.10.014

Mazmanian, S. K., Liu, G., Ton-That, H., & Schneewind, O. (1999). Staphylococcus aureus sortase, an enzyme that anchors surface proteins to the cell wall. *Science (New York, N.Y.)*, *285*(5428), 760–763.

https://doi.org/10.1126/SCIENCE.285.5428.760

Mazmanian, S. K., Ton-That, H., Su, K., & Schneewind, O. (2002). An iron-regulated sortase anchors a class of surface protein during Staphylococcus aureus pathogenesis. *Proceedings of the National Academy of Sciences of the United States of America*, *99*(4), 2293–2298. https://doi.org/10.1073/PNAS.032523999

McAdow, M., Missiakas, D. M., & Schneewind, O. (2012). Staphylococcus aureus Secretes Coagulase and von Willebrand Factor Binding Protein to Modify the Coagulation Cascade and Establish Host Infections. *Journal of Innate Immunity*, *4*(2), 141. https://doi.org/10.1159/000333447

McCarthy, A. J., & Lindsay, J. A. (2010). Genetic variation in Staphylococcus aureus surface and immune evasion genes is lineage associated: implications for vaccine design and host-pathogen interactions. *BMC Microbiology*, *10*. https://doi.org/10.1186/1471-2180-10-173

McCarthy, A. J., & Lindsay, J. A. (2012). The distribution of plasmids that carry virulence and resistance genes in Staphylococcus aureus is lineage associated. *BMC Microbiology*, *12*, 104. https://doi.org/10.1186/1471-2180-12-104

McCormick, J. K., Yarwood, J. M., & Schlievert, P. M. (2001). Toxic shock syndrome and bacterial superantigens: an update. *Annual Review of Microbiology*, *55*, 77–104. https://doi.org/10.1146/ANNUREV.MICRO.55.1.77

McDougald, D., Rice, S. A., Barraud, N., Steinberg, P. D., & Kjelleberg, S.

(2011). Should we stay or should we go: mechanisms and ecological consequences for biofilm dispersal. *Nature Reviews. Microbiology*, *10*(1), 39–50. https://doi.org/10.1038/NRMICRO2695

McEver, R. P., & Cummings, R. D. (1997). Perspectives series: cell adhesion in vascular biology. Role of PSGL-1 binding to selectins in leukocyte recruitment. *Journal of Clinical Investigation*, *100*(3), 485. https://doi.org/10.1172/JCI119556

McGavin, M. J., Arsic, B., & Nickerson, N. N. (2012). Evolutionary blueprint for host- and niche-adaptation in Staphylococcus aureus clonal complex CC30. *Frontiers in Cellular and Infection Microbiology*, *2*, 48. https://doi.org/10.3389/FCIMB.2012.00048

McGuinness, W. A., Kobayashi, S. D., & DeLeo, F. R. (2016). Evasion of Neutrophil Killing by Staphylococcus aureus. *Pathogens (Basel, Switzerland)*, *5*(1). https://doi.org/10.3390/PATHOGENS5010032

McKevitt, A. I., Bjornson, G. L., Mauracher, C. A., & Scheifele, D. W. (1990). Amino acid sequence of a deltalike toxin from Staphylococcus epidermidis. *Infection and Immunity*, *58*(5), 1473–1475. https://doi.org/10.1128/IAI.58.5.1473-1475.1990

Mehlin, C., Headley, C. M., & Klebanoff, S. J. (1999). An inflammatory polypeptide complex from Staphylococcus epidermidis: isolation and characterization. *The Journal of Experimental Medicine*, *189*(6), 907–917. https://doi.org/10.1084/JEM.189.6.907

Mejer, N., Westh, H., Schønheyder, H. C., Jensen, A. G., Larsen, A. R., Skov, R.,

& Benfield, T. (2012). Stable incidence and continued improvement in short term mortality of Staphylococcus aureus bacteraemia between 1995 and 2008. *BMC Infectious Diseases*, *12*. https://doi.org/10.1186/1471-2334-12-260

Meka, V. G., Pillai, S. K., Sakoulas, G., Wennersten, C., Venkataraman, L., DeGirolami, P. C., Eliopoulos, G. M., Moellering, R. C., & Gold, H. S. (2004). Linezolid resistance in sequential Staphylococcus aureus isolates associated with a T2500A mutation in the 23S rRNA gene and loss of a single copy of rRNA. *The Journal of Infectious Diseases*, *190*(2), 311–317. https://doi.org/10.1086/421471

Melish, M. E., Glasgow, L. A., & Turner, M. D. (1972). The staphylococcal scalded-skin syndrome: isolation and partial characterization of the exfoliative toxin. *The Journal of Infectious Diseases*, *125*(2), 129–140. https://doi.org/10.1093/INFDIS/125.2.129

Melly, M. A., Duke, L. J., Liau, D. F., & Hash, J. H. (1974). Biological properties of the encapsulated Staphylococcus aureus M. *Infection and Immunity*, *10*(2), 389–397. https://doi.org/10.1128/IAI.10.2.389-397.1974

Mendes, R. E., Deshpande, L. M., Castanheira, M., DiPersio, J., Saubolle, M. A., & Jones, R. N. (2008). First report of cfr-mediated resistance to linezolid in human staphylococcal clinical isolates recovered in the United States. *Antimicrobial Agents and Chemotherapy*, *52*(6), 2244–2246. https://doi.org/10.1128/AAC.00231-08

Mermel, L. A., Cartony, J. M., Covington, P., Maxey, G., & Morse, D. (2011).

Methicillin-resistant Staphylococcus aureus colonization at different body sites: a prospective, quantitative analysis. *Journal of Clinical Microbiology*, *49*(3), 1119–1121. https://doi.org/10.1128/JCM.02601-10

Mermel, L. A., Eells, S. J., Acharya, M. K., Cartony, J. M., Dacus, D., Fadem, S., Gay, E. A., Gordon, S., Lonks, J. R., Perl, T. M., McDougal, L. K., McGowan, J. E., Maxey, G., Morse, D., & Tenover, F. C. (2010). Quantitative analysis and molecular fingerprinting of methicillin-resistant Staphylococcus aureus nasal colonization in different patient populations: a prospective, multicenter study. *Infection Control and Hospital Epidemiology*, *31*(6), 592–597. https://doi.org/10.1086/652778

Meyer, F., Girardot, R., Piémont, Y., Prévost, G., & Colin, D. A. (2009). Analysis of the specificity of panton-valentine leucocidin and gamma-hemolysin F component binding. *Infection and Immunity*, *77*(1), 266–273. https://doi.org/10.1128/IAI.00402-08

Miajlovic, H., Fallon, P. G., Irvine, A. D., & Foster, T. J. (2010). Effect of filaggrin breakdown products on growth of and protein expression by Staphylococcus aureus. *The Journal of Allergy and Clinical Immunology*, *126*(6), 1184. https://doi.org/10.1016/J.JACI.2010.09.015

Micek, S. T. (2007). Alternatives to Vancomycin for the Treatment of Methicillin-Resistant Staphylococcus aureus Infections. *Clinical Infectious Diseases*, *45*(Supplement_3), S184–S190. https://doi.org/10.1086/519471

Miklasińska-Majdanik, M. (2021). Mechanisms of Resistance to Macrolide Antibiotics among Staphylococcus aureus. *Antibiotics 2021, Vol. 10, Page*

1406, *10*(11), 1406. https://doi.org/10.3390/ANTIBIOTICS10111406

Miller, W. R., Munita, J. M., & Arias, C. A. (2014). Mechanisms of antibiotic resistance in enterococci. *Expert Review of Anti-Infective Therapy*, *12*(10), 1221–1236. https://doi.org/10.1586/14787210.2014.956092

Milstone, A. M., Passaretti, C. L., & Perl, T. M. (2008). Chlorhexidine: expanding the armamentarium for infection control and prevention. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *46*(2), 274–281. https://doi.org/10.1086/524736

Mitchell, D. T., Levitt, D. G., Schlievert, P. M., & Ohlendorf, D. H. (2000). Structural evidence for the evolution of pyrogenic toxin superantigens. *Journal of Molecular Evolution*, *51*(6), 520–531. https://doi.org/10.1007/S002390010116

Mlynarczyk-Bonikowska, B., Kowalewski, C., Krolak-Ulinska, A., & Marusza, W. (2022). Molecular Mechanisms of Drug Resistance in Staphylococcus aureus. *International Journal of Molecular Sciences*, *23*(15). https://doi.org/10.3390/IJMS23158088

Młynarczyk, A., Młynarczyk, G., & Jeljaszewicz, J. (1998a). The genome of Staphylococcus aureus: a review. *Zentralblatt Fur Bakteriologie : International Journal of Medical Microbiology*, *287*(4), 277–314. https://doi.org/10.1016/S0934-8840(98)80165-5

Młynarczyk, A., Młynarczyk, G., & Jeljaszewicz, J. (1998b). The Genome of Staphylococcus aureus: A Review. *Zentralblatt Für Bakteriologie*, *287*(4), 277–314. https://doi.org/10.1016/S0934-8840(98)80165-5

Mongkolrattanothai, K., Boyle, S., Murphy, T. V., & Daum, R. S. (2004). Novel non-mecA-containing staphylococcal chromosomal cassette composite island containing pbp4 and tagF genes in a commensal staphylococcal species: a possible reservoir for antibiotic resistance islands in Staphylococcus aureus. *Antimicrobial Agents and Chemotherapy*, *48*(5), 1823–1836. https://doi.org/10.1128/AAC.48.5.1823-1836.2004

Monk, P. N., Scola, A. M., Madala, P., & Fairlie, D. P. (2007). Function, structure and therapeutic potential of complement C5a receptors. *British Journal of Pharmacology*, *152*(4), 429–448. https://doi.org/10.1038/SJ.BJP.0707332

Monslow, J., Govindaraju, P., & Puré, E. (2015). Hyaluronan - a functional and structural sweet spot in the tissue microenvironment. *Frontiers in Immunology*, *6*(MAY). https://doi.org/10.3389/FIMMU.2015.00231

Mortensen, J. E., Shryock, T. R., & Kapral, F. A. (1992). Modification of bactericidal fatty acids by an enzyme of Staphylococcus aureus. *Journal of Medical Microbiology*, *36*(4), 293–298. https://doi.org/10.1099/00222615-36-4-293

Muder, R. R., Boldin, M., Brennen, C., Hsieh, M., Vickers, R. M., Mitchum, K., & Yee, Y. C. (1994). A controlled trial of rifampicin, minocycline, and rifampicin plus minocycline for eradication of methicillin-resistant Staphylococcus aureus in long-term care patients. *The Journal of Antimicrobial Chemotherapy*, *34*(1), 189–190. https://doi.org/10.1093/JAC/34.1.189

Mulcahy, M. E., Geoghegan, J. A., Monk, I. R., O'Keeffe, K. M., Walsh, E. J.,

Foster, T. J., & McLoughlin, R. M. (2012). Nasal colonisation by Staphylococcus aureus depends upon clumping factor B binding to the squamous epithelial cell envelope protein loricrin. *PLoS Pathogens*, *8*(12). https://doi.org/10.1371/JOURNAL.PPAT.1003092

Mulcahy, M. E., & McLoughlin, R. M. (2016). Host-Bacterial Crosstalk Determines Staphylococcus aureus Nasal Colonization. *Trends in Microbiology*, *24*(11), 872–886. https://doi.org/10.1016/J.TIM.2016.06.012

Murthy, A., De Angelis, G., Pittet, D., Schrenzel, J., Uckay, I., & Harbarth, S. (2010). Cost-effectiveness of universal MRSA screening on admission to surgery. *Clinical Microbiology and Infection : The Official Publication of the European Society of Clinical Microbiology and Infectious Diseases*, *16*(12), 1747–1753. https://doi.org/10.1111/J.1469-0691.2010.03220.X

Mylotte, J. M., McDermott, C., & Spooner, J. A. (1987). Prospective study of 114 consecutive episodes of Staphylococcus aureus bacteremia. *Reviews of Infectious Diseases*, *9*(5), 891–907. https://doi.org/10.1093/CLINIDS/9.5.891

Naderer, O. (2008). *Nasal Decolonization of Persistent* Staphylococcus aureus *(SA) Carriers with Twice Daily Application of Retapamulin Ointment, 1%, (RET) for 3 or 5 Days*.

Nagiec, E. E., Wu, L., Swaney, S. M., Chosay, J. G., Ross, D. E., Brieland, J. K., & Leach, K. L. (2005). Oxazolidinones inhibit cellular proliferation via inhibition of mitochondrial protein synthesis. *Antimicrobial Agents and Chemotherapy*, *49*(9), 3896–3902. https://doi.org/10.1128/AAC.49.9.3896-

3902.2005

Nakamura, M. M., McAdam, A. J., Sandora, T. J., Moreira, K. R., & Lee, G. M. (2010). Higher Prevalence of Pharyngeal than Nasal Staphylococcus aureus Carriage in Pediatric Intensive Care Units. *Journal of Clinical Microbiology*, *48*(8), 2957. https://doi.org/10.1128/JCM.00547-10

Neidhart, S., Zaatreh, S., Klinder, A., Redanz, S., Spitzmüller, R., Holtfreter, S., Warnke, P., Alozie, A., Henck, V., Göhler, A., Ellenrieder, M., AbouKoura, M., Divchev, D., Gümbel, D., Napp, M., Steinhoff, G., Nienaber, C., Ekkernkamp, A., Mittelmeier, W., … Bader, R. (2018). Predictors of colonization with Staphylococcus species among patients scheduled for cardiac and orthopedic interventions at tertiary care hospitals in north-eastern Germany-a prevalence screening study. *European Journal of Clinical Microbiology & Infectious Diseases : Official Publication of the European Society of Clinical Microbiology*, *37*(4), 633–641. https://doi.org/10.1007/S10096-017-3154-X

Nelson, M. L., & Levy, S. B. (2011). The history of the tetracyclines. *Annals of the New York Academy of Sciences*, *1241*(1), 17–32. https://doi.org/10.1111/J.1749-6632.2011.06354.X

Nestle, F. O., Di Meglio, P., Qin, J. Z., & Nickoloff, B. J. (2009). Skin immune sentinels in health and disease. *Nature Reviews. Immunology*, *9*(10), 679–691. https://doi.org/10.1038/NRI2622

Neuhaus, F. C., & Baddiley, J. (2003). A continuum of anionic charge: structures and functions of D-alanyl-teichoic acids in gram-positive bacteria.

Microbiology and Molecular Biology Reviews : MMBR, *67*(4), 686–723. https://doi.org/10.1128/MMBR.67.4.686-723.2003

Nguyen, F., Starosta, A. L., Arenz, S., Sohmen, D., Dönhöfer, A., & Wilson, D. N. (2014). Tetracycline antibiotics and resistance mechanisms. *Biological Chemistry*, *395*(5), 559–575. https://doi.org/10.1515/HSZ-2013-0292

Nicola, F. G., McDougal, L. K., Biddle, J. W., & Tenover, F. C. (1998). Characterization of Erythromycin-Resistant Isolates of Staphylococcus aureus Recovered in the United States from 1958 through 1969. *Antimicrobial Agents and Chemotherapy*, *42*(11), 3024. https://doi.org/10.1128/aac.42.11.3024

Niebuhr, M., Baumert, K., & Werfel, T. (2010). TLR-2-mediated cytokine and chemokine secretion in human keratinocytes. *Experimental Dermatology*, *19*(10), 873–877. https://doi.org/10.1111/J.1600-0625.2010.01140.X

Niebuhr, M., Heratizadeh, A., Wichmann, K., Satzger, I., & Werfel, T. (2011). Intrinsic alterations of pro-inflammatory mediators in unstimulated and TLR-2 stimulated keratinocytes from atopic dermatitis patients. *Experimental Dermatology*, *20*(6), 468–472. https://doi.org/10.1111/J.1600-0625.2011.01277.X

Niemeyer, D. M., Pucci, M. J., Thanassi, J. A., Sharma, V. K., & Archer, G. L. (1996). Role of mecA transcriptional regulation in the phenotypic expression of methicillin resistance in Staphylococcus aureus. *Journal of Bacteriology*, *178*(18), 5464–5471. https://doi.org/10.1128/JB.178.18.5464-5471.1996

Nishifuji, K., Sugai, M., & Amagai, M. (2008). Staphylococcal exfoliative toxins:

"molecular scissors" of bacteria that attack the cutaneous defense barrier in mammals. *Journal of Dermatological Science*, *49*(1), 21–31. https://doi.org/10.1016/J.JDERMSCI.2007.05.007

Nizet, V. (2007). Understanding how leading bacterial pathogens subvert innate immunity to reveal novel therapeutic targets. *The Journal of Allergy and Clinical Immunology*, *120*(1), 13–22. https://doi.org/10.1016/J.JACI.2007.06.005

Noore, J., Noore, A., & Li, B. (2013). Cationic Antimicrobial Peptide LL-37 Is Effective against both Extra- and Intracellular Staphylococcus aureus. *Antimicrobial Agents and Chemotherapy*, *57*(3), 1283. https://doi.org/10.1128/AAC.01650-12

Noris, M., & Remuzzi, G. (2013). Overview of complement activation and regulation. *Seminars in Nephrology*, *33*(6), 479–492. https://doi.org/10.1016/J.SEMNEPHROL.2013.08.001

Nouwen, J., Boelens, H., Van Belkum, A., & Verbrugh, H. (2004). Human Factor in Staphylococcus aureus Nasal Carriage. *Infection and Immunity*, *72*(11), 6685. https://doi.org/10.1128/IAI.72.11.6685-6688.2004

Nouwen, J. L., Fieren, M. W. J. A., Snijders, S., Verbrugh, H. A., & Van Belkum, A. (2005). Persistent (not intermittent) nasal carriage of Staphylococcus aureus is the determinant of CPD-related infections. *Kidney International*, *67*(3), 1084–1092. https://doi.org/10.1111/J.1523-1755.2005.00174.X

Nouwen, J. L., Ott, A., Kluytmans-Vandenbergh, M. F. Q., Boelens, H. A. M., Hofman, A., Van Belkum, A., & Verbrugh, H. A. (2004). Predicting the

Staphylococcus aureus nasal carrier state: Derivation and validation of a "culture rule." *Clinical Infectious Diseases*, *39*(6), 806–811. https://doi.org/10.1086/423376/2/39-6-806-FIG002.GIF

Nsofor, C. A., Nwokenkwo, V. N., & Nwaokpa, C. (2015). Nasal carriage of staphylococcus aureus among apparently healthy school children in owerri metropolis, Nigeria. *MOJ Cell Science & Report*, *Volume 2*(Issue 5). https://doi.org/10.15406/MOJCSR.2015.02.00038

Ntirenganya, C., Manzi, O., Muvunyi, C. M., & Ogbuagu, O. (2015). High Prevalence of Antimicrobial Resistance Among Common Bacterial Isolates in a Tertiary Healthcare Facility in Rwanda. *The American Journal of Tropical Medicine and Hygiene*, *92*(4), 865. https://doi.org/10.4269/AJTMH.14-0607

Nur-ur Rahman, A. K. M., Bonsor, D. A., Herfst, C. A., Pollard, F., Peirce, M., Wyatt, A. W., Kasper, K. J., Madrenas, J., Sundberg, E. J., & McCormick, J. K. (2011). The T cell receptor beta-chain second complementarity determining region loop (CDR2beta governs T cell activation and Vbeta specificity by bacterial superantigens. *The Journal of Biological Chemistry*, *286*(6), 4871–4881. https://doi.org/10.1074/JBC.M110.189068

O'Brien, M. M., Walsh, E. J., Massey, R. C., Peacock, S. J., & Foster, T. J. (2002). Staphylococcus aureus clumping factor B (ClfB) promotes adherence to human type I cytokeratin 10: implications for nasal colonization. *Cellular Microbiology*, *4*(11), 759–770. https://doi.org/10.1046/J.1462-5822.2002.00231.X

O'Gara, J. P. (2007). ica and beyond: biofilm mechanisms and regulation in Staphylococcus epidermidis and Staphylococcus aureus. *FEMS Microbiology Letters*, *270*(2), 179–188. https://doi.org/10.1111/J.1574-6968.2007.00688.X

O'Neill, E., Pozzi, C., Houston, P., Smyth, D., Humphreys, H., Robinson, D. A., & O'Gara, J. P. (2007). Association between methicillin susceptibility and biofilm regulation in Staphylococcus aureus isolates from device-related infections. *Journal of Clinical Microbiology*, *45*(5), 1379–1388. https://doi.org/10.1128/JCM.02280-06

O'Riordan, K., & Lee, J. C. (2004). Staphylococcus aureus Capsular Polysaccharides. *Clinical Microbiology Reviews*, *17*(1), 218–234. https://doi.org/10.1128/CMR.17.1.218-234.2004/ASSET/518D3425-9897-454B-9F0D-489E235B43BA/ASSETS/GRAPHIC/ZCM0010400360006.JPEG

Ogston, A. (1882). Micrococcus Poisoning. *Journal of Anatomy and Physiology*, *17*(Pt 1), 24. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC1310127/

Olonitola, O. S., INabo, H. I., Olayinka, B. O., & Bugo, I. D. (2007). Nasal carriage of methicillin-resistant *Staphylococcus aureus* by primary school pupils in a University Staff School, Zaria, Nigeria. *International Journal of Biological and Chemical Sciences*, *1*(1), 71–75. https://doi.org/10.4314/ijbcs.v1i1.39701

Olson, M. E., Nygaard, T. K., Ackermann, L., Watkins, R. L., Zurek, O. W., Pallister, K. B., Griffith, S., Kiedrowski, M. R., Flack, C. E., Kavanaugh, J.

S., Kreiswirth, B. N., Horswill, A. R., & Voyich, J. M. (2013). Staphylococcus aureus nuclease is an SaeRS-dependent virulence factor. *Infection and Immunity*, *81*(4), 1316–1324. https://doi.org/10.1128/IAI.01242-12

Onanuga, A., & Temedie, T. C. (2011). Nasal carriage of multi-drug resistant Staphylococcus aureus in healthy inhabitants of Amassoma in Niger delta region of Nigeria. *African Health Sciences*, *11*(2), 176. /pmc/articles/PMC3158512/

Ong, M. H. L., Ho, W. Y., Ng, W. W., & Chew, C. H. (2017). High Prevalence of tetM as Compared to tetK Amongst Methicillin-Resistant Staphylococcus aureus (MRSA) Isolates from Hospitals in Perak, Malaysia. *Jundishapur Journal of Microbiology 2017 10:6*, *10*(6), 13935. https://doi.org/10.5812/JJM.13935

Otto, M. (2010). Basis of virulence in community-associated methicillin-resistant Staphylococcus aureus. *Annual Review of Microbiology*, *64*, 143–162. https://doi.org/10.1146/ANNUREV.MICRO.112408.134309

Otto, M. (2014). Staphylococcus aureus toxins. *Current Opinion in Microbiology*, *17*(1), 32–37. https://doi.org/10.1016/J.MIB.2013.11.004

Pader, V., Hakim, S., Painter, K. L., Wigneshweraraj, S., Clarke, T. B., & Edwards, A. M. (2016). Staphylococcus aureus inactivates daptomycin by releasing membrane phospholipids. *Nature Microbiology*, *2*. https://doi.org/10.1038/NMICROBIOL.2016.194

Pan, Y., Zeng, J., Li, L., Yang, J., Tang, Z., Xiong, W., Li, Y., Chen, S., & Zeng,

Z. (2020). Coexistence of Antibiotic Resistance Genes and Virulence Factors Deciphered by Large-Scale Complete Genome Analysis. *MSystems*, *5*(3). https://doi.org/10.1128/MSYSTEMS.00821-19/SUPPL_FILE/MSYSTEMS.00821-19-S0001.DOC

Park, B., Iwase, T., & Liu, G. Y. (2011). Intranasal application of S. epidermidis prevents colonization by methicillin-resistant Staphylococcus aureus in mice. *PloS One*, *6*(10). https://doi.org/10.1371/JOURNAL.PONE.0025880

Park, J. W., Lee, H., Kim, J. W., & Kim, B. (2019). Characterization of Infections with Vancomycin-Intermediate Staphylococcus aureus (VISA) and Staphylococcus aureus with Reduced Vancomycin Susceptibility in South Korea. *Scientific Reports 2019 9:1*, *9*(1), 1–9. https://doi.org/10.1038/s41598-019-42307-6

Parker, D. (2018). A live vaccine to Staphylococcus aureus infection. *Virulence*, *9*(1), 700. https://doi.org/10.1080/21505594.2018.1426965

Parker, D., & Prince, A. (2012). Immunopathogenesis of Staphylococcus aureus pulmonary infection. *Seminars in Immunopathology*, *34*(2), 281. https://doi.org/10.1007/S00281-011-0291-7

Parker, M. W., & Feil, S. C. (2005). Pore-forming protein toxins: From structure to function. *Progress in Biophysics and Molecular Biology*, *88*(1), 91–142. https://doi.org/10.1016/j.pbiomolbio.2004.01.009

Parry, M. A. A., Fernandez-Catalan, C., Bergner, A., Huber, R., Hopfner, K. P., Schlott, B., Gührs, K. H., & Bode, W. (1998). The ternary microplasmin-staphylokinase-microplasmin complex is a proteinase-cofactor-substrate

complex in action. *Nature Structural Biology*, *5*(10), 917–923. https://doi.org/10.1038/2359

Patel, J. A., Alvarez-Fernandez, P., Jennings, K., Loeffelholz, M., Mccormick, D., & Chonmaitree, T. (2015). Factors Affecting Staphylococcus aureus Colonization of the Nasopharynx in the First Six Months of Life. *The Pediatric Infectious Disease Journal*, *34*(8), 826. https://doi.org/10.1097/INF.0000000000000744

Patel, R. (2005). Biofilms and antimicrobial resistance. *Clinical Orthopaedics and Related Research*, *437*(437), 41–47. https://doi.org/10.1097/01.BLO.0000175714.68624.74

Pathak, A., Marothi, Y., Iyer, R. V., Singh, B., Sharma, M., Eriksson, B., Macaden, R., & Lundborg, C. S. (2010). Nasal carriage and antimicrobial susceptibility of Staphylococcus aureus in healthy preschool children in Ujjain, India. *BMC Pediatrics*, *10*. https://doi.org/10.1186/1471-2431-10-100

Patti, J. M., Allen, B. L., McGavin, M. J., & Hook, M. (1994). MSCRAMM-mediated adherence of microorganisms to host tissues. *Annual Review of Microbiology*, *48*, 585–617. https://doi.org/10.1146/ANNUREV.MI.48.100194.003101

Peacock, S. J., De Silva, I., & Lowy, F. D. (2001). What determines nasal carriage of Staphylococcus aureus? *Trends in Microbiology*, *9*(12), 605–610. https://doi.org/10.1016/S0966-842X(01)02254-5

Peetermans, M., Vanassche, T., Liesenborghs, L., Claes, J., Velde, G. Vande, Kwiecinksi, J., Jin, T., De Geest, B., Hoylaerts, M. F., Lijnen, R. H., &

Verhamme, P. (2014). Plasminogen activation by staphylokinase enhances local spreading of S. aureus in skin infections. *BMC Microbiology*, *14*(1). https://doi.org/10.1186/S12866-014-0310-7

Pendleton, J. N., Gorman, S. P., & Gilmore, B. F. (2013). Clinical relevance of the ESKAPE pathogens. *Expert Review of Anti-Infective Therapy*, *11*(3), 297–308. https://doi.org/10.1586/ERI.13.12

Penesyan, A., Gillings, M., & Paulsen, I. T. (2015). Antibiotic discovery: combatting bacterial resistance in cells and in biofilm communities. *Molecules (Basel, Switzerland)*, *20*(4), 5286–5298. https://doi.org/10.3390/MOLECULES20045286

Periasamy, S., Joo, H. S., Duong, A. C., Bach, T. H. L., Tan, V. Y., Chatterjee, S. S., Cheung, G. Y. C., & Otto, M. (2012). How Staphylococcus aureus biofilms develop their characteristic structure. *Proceedings of the National Academy of Sciences of the United States of America*, *109*(4), 1281–1286. https://doi.org/10.1073/PNAS.1115006109/-/DCSUPPLEMENTAL/PNAS.201115006SI.PDF

Perl, T. M., Cullen, J. J., Wenzel, R. P., Zimmerman, M. B., Pfaller, M. A., Sheppard, D., Twombley, J., French, P. P., & Herwaldt, L. A. (2002). Intranasal mupirocin to prevent postoperative Staphylococcus aureus infections. *The New England Journal of Medicine*, *346*(24), 1871–1877. https://doi.org/10.1056/NEJMOA003069

Peschel, A., & Otto, M. (2013). Phenol-soluble modulins and staphylococcal infection. *Nature Reviews. Microbiology*, *11*(10), 667–673.

https://doi.org/10.1038/NRMICRO3110

Peschel, A., Vuong, C., Otto, M., & Gotz, F. (2000). The D-alanine residues of Staphylococcus aureus teichoic acids alter the susceptibility to vancomycin and the activity of autolytic enzymes. *Antimicrobial Agents and Chemotherapy*, *44*(10), 2845–2847. https://doi.org/10.1128/AAC.44.10.2845-2847.2000

Petinaki, E., Arvaniti, A., Dimitracopoulos, G., & Spiliopoulou, I. (2001). Detection of mecA, mecR1 and mecI genes among clinical isolates of methicillin-resistant staphylococci by combined polymerase chain reactions. *The Journal of Antimicrobial Chemotherapy*, *47*(3), 297–304. https://doi.org/10.1093/JAC/47.3.297

Phonimdaeng, P., O'Reilly, M., Nowlan, P., Bramley, A. J., & Foster, T. J. (1990). The coagulase of Staphylococcus aureus 8325-4. Sequence analysis and virulence of site-specific coagulase-deficient mutants. *Molecular Microbiology*, *4*(3), 393–404. https://doi.org/10.1111/J.1365-2958.1990.TB00606.X

Pichereau, S., & Rose, W. E. (2010). Invasive community-associated MRSA infections: epidemiology and antimicrobial management. *Http://Dx.Doi.Org/10.1517/14656566.2010.511614*, *11*(18), 3009–3025. https://doi.org/10.1517/14656566.2010.511614

Piddock, L. J. V. (2006a). Clinically relevant chromosomally encoded multidrug resistance efflux pumps in bacteria. *Clinical Microbiology Reviews*, *19*(2), 382–402. https://doi.org/10.1128/CMR.19.2.382-402.2006

Piddock, L. J. V. (2006b). Multidrug-resistance efflux pumps - not just for resistance. *Nature Reviews. Microbiology*, *4*(8), 629–636. https://doi.org/10.1038/NRMICRO1464

Pillai, S. K., Sakoulas, G., Wennersten, C., Eliopoulos, G. M., Moellering, R. C., Ferraro, M. J., & Gold, H. S. (2002). Linezolid resistance in Staphylococcus aureus: characterization and stability of resistant phenotype. *The Journal of Infectious Diseases*, *186*(11), 1603–1607. https://doi.org/10.1086/345368

Pontes, D. S., de Araujo, R. S. A., Dantas, N., Scotti, L., Scotti, M. T., de Moura, R. O., & Mendonca-Junior, F. J. B. (2018). Genetic Mechanisms of Antibiotic Resistance and the Role of Antibiotic Adjuvants. *Current Topics in Medicinal Chemistry*, *18*(1), 42–74. https://doi.org/10.2174/1568026618666180206095224

Poovelikunnel, T., Gethin, G., & Humphreys, H. (2015). Mupirocin resistance: clinical implications and potential alternatives for the eradication of MRSA. *The Journal of Antimicrobial Chemotherapy*, *70*(10), 2681–2692. https://doi.org/10.1093/JAC/DKV169

Popov, L., Kovalski, J., Grandi, G., Bagnoli, F., & Amieva, M. R. (2014). Three-Dimensional Human Skin Models to Understand Staphylococcus aureus Skin Colonization and Infection. *Frontiers in Immunology*, *5*(FEB). https://doi.org/10.3389/FIMMU.2014.00041

Porter, R. M., & Lane, E. B. (2003). Phenotypes, genotypes and their contribution to understanding keratin function. *Trends in Genetics : TIG*, *19*(5), 278–285. https://doi.org/10.1016/S0168-9525(03)00071-4

Postma, B., Kleibeuker, W., Poppelier, M. J. J. G., Boonstra, M., Van Kessel, K. P. M., Van Strijp, J. A. G., & De Haas, C. J. C. (2005). Residues 10–18 within the C5a Receptor N Terminus Compose a Binding Domain for Chemotaxis Inhibitory Protein of Staphylococcus aureus. *Journal of Biological Chemistry*, *280*(3), 2020–2027. https://doi.org/10.1074/JBC.M412230200

Projan, S. J., Kornblum, J., Kreiswirth, B., Moghazeh, S. L., Eisner, W., & Novick, R. P. (1989). Nucleotide sequence: the beta-hemolysin gene of Staphylococcus aureus. *Nucleic Acids Research*, *17*(8), 3305. https://doi.org/10.1093/NAR/17.8.3305

Proksch, E., Brandner, J. M., & Jensen, J. M. (2008). The skin: an indispensable barrier. *Experimental Dermatology*, *17*(12), 1063–1072. https://doi.org/10.1111/J.1600-0625.2008.00786.X

Prystowsky, J., Siddiqui, F., Chosay, J., Shinabarger, D. L., Millichap, J., Peterson, L. R., & Noskin, G. A. (2001). Resistance to linezolid: characterization of mutations in rRNA and comparison of their occurrences in vancomycin-resistant enterococci. *Antimicrobial Agents and Chemotherapy*, *45*(7), 2154–2156. https://doi.org/10.1128/AAC.45.7.2154-2156.2001

Pynnonen, M., Stephenson, R. E., Schwartz, K., Hernandez, M., & Boles, B. R. (2011). Hemoglobin Promotes Staphylococcus aureus Nasal Colonization. *PLOS Pathogens*, *7*(7), e1002104. https://doi.org/10.1371/JOURNAL.PPAT.1002104

Rabijns, A., De Bondt, H. L., & De Ranter, C. (1997). Three-dimensional structure of staphylokinase, a plasminogen activator with therapeutic potential. *Nature Structural Biology*, *4*(5), 357–360. https://doi.org/10.1038/NSB0597-357

Rahbar, M., Karamiyar, M., & Gra-Agaji, R. (2003). Nasal carriage of methicillin-resistant Staphylococcus aureus among healthcare workers of an Iranian hospital. *Infection Control and Hospital Epidemiology*, *24*(4), 236–237. https://doi.org/10.1086/503478

Raineri, E. J. M., Altulea, D., & van Dijl, J. M. (2022). Staphylococcal trafficking and infection—from 'nose to gut' and back. *FEMS Microbiology Reviews*, *46*(1), 1–22. https://doi.org/10.1093/FEMSRE/FUAB041

Randall, C. P., Mariner, K. R., Chopra, I., & O'Neill, A. J. (2013). The target of daptomycin is absent from Escherichia coli and other gram-negative pathogens. *Antimicrobial Agents and Chemotherapy*, *57*(1), 637–639. https://doi.org/10.1128/AAC.02005-12

Ratliff, M., Zhu, W., Deshmukh, R., Wilks, A., & Stojiljkovic, I. (2001). Homologues of neisserial heme oxygenase in gram-negative bacteria: degradation of heme by the product of the pigA gene of Pseudomonas aeruginosa. *Journal of Bacteriology*, *183*(21), 6394–6403. https://doi.org/10.1128/JB.183.21.6394-6403.2001

Rauch, S., Gough, P., Kim, H. K., Schneewind, O., & Missiakas, D. (2014). Vaccine Protection of Leukopenic Mice against Staphylococcus aureus Bloodstream Infection. *Infection and Immunity*, *82*(11), 4889.

https://doi.org/10.1128/IAI.02328-14

Rautenberg, M., Joo, H., Otto, M., & Peschel, A. (2011). Neutrophil responses to staphylococcal pathogens and commensals via the formyl peptide receptor 2 relates to phenol-soluble modulin release and virulence. *FASEB Journal : Official Publication of the Federation of American Societies for Experimental Biology*, *25*(4), 1254–1263. https://doi.org/10.1096/FJ.10-175208

Rawls, M., & Ellis, A. K. (2019). The microbiome of the nose. *Annals of Allergy, Asthma & Immunology : Official Publication of the American College of Allergy, Asthma, & Immunology*, *122*(1), 17–24. https://doi.org/10.1016/J.ANAI.2018.05.009

Rayner, C., & Munckhof, W. J. (2005a). Antibiotics currently used in the treatment of infections caused by Staphylococcus aureus. *Internal Medicine Journal*, *35 Suppl 2*(SUPPL. 2). https://doi.org/10.1111/J.1444-0903.2005.00976.X

Rayner, C., & Munckhof, W. J. (2005b). Antibiotics currently used in the treatment of infections caused by Staphylococcus aureus. *Internal Medicine Journal*, *35 Suppl 2*(SUPPL. 2). https://doi.org/10.1111/J.1444-0903.2005.00976.X

Rebiahi, S. A., Abdelouahid, D. E., Rahmoun, M., Abdelali, S., & Azzaoui, H. (2011). Emergence of vancomycin-resistant Staphylococcus aureus identified in the Tlemcen university hospital (North-West Algeria). *Médecine et Maladies Infectieuses*, *41*(12), 646–651.

https://doi.org/10.1016/J.MEDMAL.2011.09.010

Redgrave, L. S., Sutton, S. B., Webber, M. A., & Piddock, L. J. V. (2014). Fluoroquinolone resistance: mechanisms, impact on bacteria, and role in evolutionary success. *Trends in Microbiology*, *22*(8), 438–445. https://doi.org/10.1016/J.TIM.2014.04.007

Regev-Yochay, G., Dagan, R., Raz, M., Carmeli, Y., Shainberg, B., Derazne, E., Rahav, G., & Rubinstein, E. (2004). Association between carriage of Streptococcus pneumoniae and Staphylococcus aureus in Children. *JAMA*, *292*(6), 716–720. https://doi.org/10.1001/JAMA.292.6.716

Regev-Yochay, G., Trzciński, K., Thompson, C. M., Malley, R., & Lipsitch, M. (2006). Interference between Streptococcus pneumoniae and Staphylococcus aureus: In Vitro Hydrogen Peroxide-Mediated Killing by Streptococcus pneumoniae. *Journal of Bacteriology*, *188*(13), 4996. https://doi.org/10.1128/JB.00317-06

Reid, M. J. A., Fischer, R. S. B., Mannathoko, N., Muthoga, C., McHugh, E., Essigmann, H., Brown, E. L., & Steenhoff, A. P. (2017). Prevalence of Staphylococcus aureus Nasal Carriage in Human Immunodeficiency Virus–Infected and Uninfected Children in Botswana: Prevalence and Risk Factors. *The American Journal of Tropical Medicine and Hygiene*, *96*(4), 795. https://doi.org/10.4269/AJTMH.16-0650

Relman, D. A. (2012). The human microbiome: ecosystem resilience and health. *Nutrition Reviews*, *70*(suppl_1), S2–S9. https://doi.org/10.1111/J.1753-4887.2012.00489.X

Reygaert, W. (2009). Methicillin-Resistant Staphylococcus aureus (MRSA): Molecular Aspects of Antimicrobial Resistance and Virulence. *CLINICAL LABORATORY SCIENCE*, *22*. http://hwmaint.clsjournal.ascls.org/

Reygaert, W. C. (2018). An overview of the antimicrobial resistance mechanisms of bacteria. *AIMS Microbiology*, *4*(3), 482. https://doi.org/10.3934/MICROBIOL.2018.3.482

Rieg, S., Peyerl-Hoffmann, G., de With, K., Theilacker, C., Wagner, D., Hübner, J., Dettenkofer, M., Kaasch, A., Seifert, H., Schneider, C., & Kern, W. V. (2009). Mortality of S. aureus bacteremia and infectious diseases specialist consultation--a study of 521 patients in Germany. *The Journal of Infection*, *59*(4), 232–239. https://doi.org/10.1016/J.JINF.2009.07.015

Rigby, K. M., & DeLeo, F. R. (2012). Neutrophils in innate host defense against Staphylococcus aureus infections. *Seminars in Immunopathology*, *34*(2), 237–259. https://doi.org/10.1007/S00281-011-0295-3

Rittenhouse, S., Biswas, S., Broskey, J., McCloskey, L., Moore, T., Vasey, S., West, J., Zalacain, M., Zonis, R., & Payne, D. (2006). Selection of retapamulin, a novel pleuromutilin for topical use. *Antimicrobial Agents and Chemotherapy*, *50*(11), 3882–3885. https://doi.org/10.1128/AAC.00178-06

Rittershain, R. von. (1878). Die exfoliativa dermatitis jungerer Saulinge. *Centralz Kinderhheilk*, *2*, 3–23. https://cir.nii.ac.jp/crid/1570572701285138048

Robert I. Handin, Samuel E. Lux, & Thomas P. Stossel. (2003). *Blood: Principles and Practice of Hematology* (J. JW. Pine Jr (Ed.); Second Edition). https://books.google.com.gh/books?hl=en&lr=&id=H85dwxYTKLwC&oi=f

nd&pg=PA363&dq=Bridges+K.+Seligman+P.A.+Handin+R.I.+Lux+S.E.+Stossel+T.P.+Blood:+Principles+and+Practice+of+Hematology.+J.+B.+Lippincott+Company,%0A+Philadelphia1995&ots=AdrgBRsaDT&sig=hRFWqcU_-esfPLQhUMk2hZeHRJg&redir_esc=y#v=onepage&q&f=false

Roberts, M. C. (2004). Resistance to macrolide, lincosamide, streptogramin, ketolide, and oxazolidinone antibiotics. *Molecular Biotechnology*, *28*(1), 47–62. https://doi.org/10.1385/MB:28:1:47

Roberts, M. C. (2008). Update on macrolide-lincosamide-streptogramin, ketolide, and oxazolidinone resistance genes. *FEMS Microbiology Letters*, *282*(2), 147–159. https://doi.org/10.1111/J.1574-6968.2008.01145.X

Roger, C., Roberts, J. A., & Muller, L. (2017). Clinical Pharmacokinetics and Pharmacodynamics of Oxazolidinones. *Clinical Pharmacokinetics 2017 57:5*, *57*(5), 559–575. https://doi.org/10.1007/S40262-017-0601-X

Rojo, P., Barrios, M., Palacios, A., Gomez, C., & Chaves, F. (2010). Community-associated Staphylococcus aureus infections in children. *Expert Review of Anti-Infective Therapy*, *8*(5), 541–554. https://doi.org/10.1586/ERI.10.34

Rollof, J., Hedström, S. Å., & Nilsson-Ehle, P. (1987). Positional specificity and substrate preference of purified Staphylococcus aureus lipase. *Biochimica et Biophysica Acta*, *921*(2), 370–377. https://doi.org/10.1016/0005-2760(87)90039-7

Rollof, J., HedstrÖM, S., & Nilsson-Ehle, P. (1987). Lipolytic activity of Staphylococcus aureus strains from disseminated and localized infections. *Acta Pathologica, Microbiologica, et Immunologica Scandinavica. Section*

B, Microbiology, *95*(2), 109–113. https://doi.org/10.1111/J.1699-0463.1987.TB03096.X

Rollof, J., & Normark, S. (1992). In vivo processing of Staphylococcus aureus lipase. *Journal of Bacteriology*, *174*(6), 1844–1847. https://doi.org/10.1128/JB.174.6.1844-1847.1992

Rollof, J., Vinge, E., Nilsson-Ehle, P., & Braconier, J. H. (1992). Aggregation of human granulocytes by Staphylococcus aureus lipase. *Journal of Medical Microbiology*, *36*(1), 52–55. https://doi.org/10.1099/00222615-36-1-52

Rooijakkers, S. H. M., Van Kessel, K. P. M., & Van Strijp, J. A. G. (2005). Staphylococcal innate immune evasion. *Trends in Microbiology*, *13*(12), 596–601. https://doi.org/10.1016/J.TIM.2005.10.002

Rooijakkers, S. H. M., Van Wamel, W. J. B., Ruyken, M., Van Kessel, K. P. M., & Van Strijp, J. A. G. (2005). Anti-opsonic properties of staphylokinase. *Microbes and Infection*, *7*(3), 476–484. https://doi.org/10.1016/J.MICINF.2004.12.014

Roop, D. (1995). Defects in the barrier. *Science*, *267*(5197), 474–475. https://doi.org/10.1126/SCIENCE.7529942/ASSET/7AB23B21-787A-4C20-AB80-196FA9B2774C/ASSETS/SCIENCE.7529942.FP.PNG

Rose, F., Dahlem, G., Guthmann, B., Grimminger, F., Maus, U., Hänze, J., Duemmer, N., Grandel, U., Seeger, W., & Ghofrani, H. A. (2002). Mediator generation and signaling events in alveolar epithelial cells attacked by S. aureus alpha-toxin. *American Journal of Physiology. Lung Cellular and Molecular Physiology*, *282*(2).

https://doi.org/10.1152/AJPLUNG.00156.2001

Rosenstein, R., & Götz, F. (2000). Staphylococcal lipases: biochemical and molecular characterization. *Biochimie*, *82*(11), 1005–1014. https://doi.org/10.1016/S0300-9084(00)01180-9

Rountree, P. M., & Freeman, B. M. (1955). Infections caused by a particular Phage Type of Staphylococcus aureus. *Medical Journal of Australia*, *2*(5), 157–161.

Rubinstein, E., & Keynan, Y. (2014). Vancomycin revisited - 60 years later. *Frontiers in Public Health*, *2*(OCT). https://doi.org/10.3389/FPUBH.2014.00217

Ryu, S., Song, P. I., Seo, C. H., Cheong, H., & Park, Y. (2014). Colonization and infection of the skin by S. aureus: immune system evasion and the response to cationic antimicrobial peptides. *International Journal of Molecular Sciences*, *15*(5), 8753–8772. https://doi.org/10.3390/IJMS15058753

Saadat, S., Solhjoo, K., Norooz-Nejad, M. J., & Kazemi, A. (2014). VanA and VanB Positive Vancomycin-resistant Staphylococcus aureus Among Clinical Isolates in Shiraz, South of Iran. *Oman Medical Journal*, *29*(5), 335. https://doi.org/10.5001/OMJ.2014.90

Saber, H., Jasni, A. S., Jamaluddin, T. Z. M. T., & Ibrahim, R. (2017). A Review of Staphylococcal Cassette Chromosome mec (SCCmec) Types in Coagulase-Negative Staphylococci (CoNS) Species. *The Malaysian Journal of Medical Sciences : MJMS*, *24*(5), 7. https://doi.org/10.21315/MJMS2017.24.5.2

Safdar, N., & Bradley, E. A. (2008). The risk of infection after nasal colonization with Staphylococcus aureus. *The American Journal of Medicine*, *121*(4), 310–315. https://doi.org/10.1016/J.AMJMED.2007.07.034

Sajjan, U., Ackerley, C., & Forstner, J. (2002). Interaction of cblA/adhesin-positive Burkholderia cepacia with squamous epithelium. *Cellular Microbiology*, *4*(2), 73–86. https://doi.org/10.1046/J.1462-5822.2002.00171.X

Sakoulas, G., Moise, P. A., Casapao, A. M., Nonejuie, P., Olson, J., Okumura, C. Y. M., Rybak, M. J., Kullar, R., Dhand, A., Rose, W. E., Goff, D. A., Bressler, A. M., Lee, Y., Pogliano, J., Johns, S., Kaatz, G. W., Ebright, J. R., & Nizet, V. (2014). Antimicrobial salvage therapy for persistent staphylococcal bacteremia using daptomycin plus ceftaroline. *Clinical Therapeutics*, *36*(10), 1317–1333. https://doi.org/10.1016/J.CLINTHERA.2014.05.061

Sakr, A., Brégeon, F., Mège, J. L., Rolain, J. M., & Blin, O. (2018). Staphylococcus aureus Nasal Colonization: An Update on Mechanisms, Epidemiology, Risk Factors, and Subsequent Infections. *Frontiers in Microbiology*, *9*(OCT). https://doi.org/10.3389/FMICB.2018.02419

Sampane-Donkor, E., Badoe, E. V., Annan, J. A., & Nii-Trebi, N. I. (2017). Colonisation of antibiotic resistant bacteria in a cohort of HIV infected children in Ghana. *The Pan African Medical Journal*, *26*. https://doi.org/10.11604/PAMJ.2017.26.60.10981

Sandri, A. M., Dalarosa, M. G., Alcântara, L. R. de, Elias, L. da S., & Zavascki,

A. P. (2006). Reduction in Inddence of Nosocomial Methicillin-Resistant Staphylococcus aureus (MRSA) Infection in an Intensive Care Unit: Role of Treatment With Mupirocin Ointment and Chlorhexidine Baths for Nasal Carriers of MRSA. *Infection Control & Hospital Epidemiology*, *27*(2), 185–187. https://doi.org/10.1086/500625

Sanford, M. D., Widmer, A. F., Bale, M. J., Jones, R. N., & Wenzel, R. P. (1994). Efficient detection and long-term persistence of the carriage of methicillin-resistant Staphylococcus aureus. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *19*(6), 1123–1128. https://doi.org/10.1093/CLINIDS/19.6.1123

Santala, A., Saarinen, J., Kovanen, P., & Kuusela, P. (1999). Activation of interstitial collagenase, MMP-1, by Staphylococcus aureus cells having surface-bound plasmin: a novel role of plasminogen receptors of bacteria. *FEBS Letters*, *461*(3), 153–156. https://doi.org/10.1016/S0014-5793(99)01440-4

Satzke, C., Turner, P., Virolainen-Julkunen, A., Adrian, P. V., Antonio, M., Hare, K. M., Henao-Restrepo, A. M., Leach, A. J., Klugman, K. P., Porter, B. D., Sá-Leão, R., Scott, J. A., Nohynek, H., & O'Brien, K. L. (2013). Standard method for detecting upper respiratory carriage of Streptococcus pneumoniae: Updated recommendations from the World Health Organization Pneumococcal Carriage Working Group. *Vaccine*, *32*(1), 165–179. https://doi.org/10.1016/J.VACCINE.2013.08.062

Sawai, T., Tomono, K., Yanagihara, K., Yamamoto, Y., Kaku, M., Hirakata, Y.,

Koga, H., Tashiro, T., & Kohno, S. (1997). Role of coagulase in a murine model of hematogenous pulmonary infection induced by intravenous injection of Staphylococcus aureus enmeshed in agar beads. *Infection and Immunity*, *65*(2), 466–471. https://doi.org/10.1128/IAI.65.2.466-471.1997

Schauber, J., & Gallo, R. L. (2008). Antimicrobial peptides and the skin immune defense system. *The Journal of Allergy and Clinical Immunology*, *122*(2), 261–266. https://doi.org/10.1016/J.JACI.2008.03.027

Schaumburg, F., Alabi, A. S., Peters, G., & Becker, K. (2014). New epidemiology of Staphylococcus aureus infection in Africa. *Clinical Microbiology and Infection : The Official Publication of the European Society of Clinical Microbiology and Infectious Diseases*, *20*(7), 589–596. https://doi.org/10.1111/1469-0691.12690

Scherr, T. D., Hanke, M. L., Huang, O., James, D. B. A., Horswill, A. R., Bayles, K. W., Fey, P. D., Torres, V. J., & Kielian, T. (2015). Staphylococcus aureus Biofilms Induce Macrophage Dysfunction Through Leukocidin AB and Alpha-Toxin. *MBio*, *6*(4). https://doi.org/10.1128/MBIO.01021-15

Schlott, B., Gührs, K. H., Hartmann, M., Röcker, A., & Collen, D. (1997). Staphylokinase requires NH2-terminal proteolysis for plasminogen activation. *The Journal of Biological Chemistry*, *272*(9), 6067–6072. https://doi.org/10.1074/JBC.272.9.6067

Schulte, R. H., & Munson, E. (2019). Staphylococcus aureus Resistance Patterns in Wisconsin. *Clinical Medicine & Research*, *17*(3–4), 72–81. https://doi.org/10.3121/CMR.2019.1503

Schwalbe, R. S., Stapleton, J. T., & Gilligan, P. H. (1987). Emergence of vancomycin resistance in coagulase-negative staphylococci. *The New England Journal of Medicine*, *316*(15), 927–931. https://doi.org/10.1056/NEJM198704093161507

Schwarz, S., Kehrenberg, C., Doublet, B., & Cloeckaert, A. (2004). Molecular basis of bacterial resistance to chloramphenicol and florfenicol. *FEMS Microbiology Reviews*, *28*(5), 519–542. https://doi.org/10.1016/J.FEMSRE.2004.04.001

Schwarz, S., Werckenthin, C., & Kehrenberg, C. (2000). Identification of a plasmid-borne chloramphenicol-florfenicol resistance gene in Staphylococcus sciuri. *Antimicrobial Agents and Chemotherapy*, *44*(9), 2530–2533. https://doi.org/10.1128/AAC.44.9.2530-2533.2000

Sciara, G., Kendrew, S. G., Miele, A. E., Marsh, N. G., Federici, L., Malatesta, F., Schimperna, G., Savino, C., & Vallone, B. (2003). The structure of ActVA-Orf6, a novel type of monooxygenase involved in actinorhodin biosynthesis. *The EMBO Journal*, *22*(2), 205–215. https://doi.org/10.1093/EMBOJ/CDG031

Segal, A. W. (2005). How neutrophils kill microbes. *Annual Review of Immunology*, *23*, 197–223. https://doi.org/10.1146/ANNUREV.IMMUNOL.23.021704.115653

Seilie, E. S., & Bubeck Wardenburg, J. (2017). Staphylococcus aureus pore-forming toxins: The interface of pathogen and host complexity. *Seminars in Cell & Developmental Biology*, *72*, 101–116.

https://doi.org/10.1016/J.SEMCDB.2017.04.003

Selva, L., Viana, D., Regev-Yochay, G., Trzcinski, K., Corpa, J. M., Lasa, Í., Novick, R. P., & Penadés, J. R. (2009). Killing niche competitors by remote-control bacteriophage induction. *Proceedings of the National Academy of Sciences of the United States of America*, *106*(4), 1234. https://doi.org/10.1073/PNAS.0809600106

Septimus, E., Hickok, J., Moody, J., Kleinman, K., Avery, T. R., Huang, S. S., Platt, R., & Perlin, J. (2016). Closing the Translation Gap: Toolkit-based Implementation of Universal Decolonization in Adult Intensive Care Units Reduces Central Line-associated Bloodstream Infections in 95 Community Hospitals. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *63*(2), 172–177. https://doi.org/10.1093/CID/CIW282

Septimus, E. J., & Schweizer, M. L. (2016). Decolonization in Prevention of Health Care-Associated Infections. *Clinical Microbiology Reviews*, *29*(2), 201. https://doi.org/10.1128/CMR.00049-15

Septimus, E., Weinstein, R. A., Perl, T. M., Goldmann, D. A., & Yokoe, D. S. (2014). Approaches for preventing healthcare-associated infections: go long or go wide? *Infection Control and Hospital Epidemiology*, *35 Suppl 2*(S2), S10–S14. https://doi.org/10.1017/S0899823X00193808

Serra-Burriel, M., Keys, M., Campillo-Artero, C., Agodi, A., Barchitta, M., Gikas, A., Palos, C., & López-Casasnovas, G. (2020). Impact of multi-drug resistant bacteria on economic and clinical outcomes of healthcare-associated

infections in adults: Systematic review and meta-analysis. *PLoS ONE*, *15*(1). https://doi.org/10.1371/JOURNAL.PONE.0227139

Severin, A., Tabei, K., Tenover, F., Chung, M., Clarke, N., & Tomasz, A. (2004). High Level Oxacillin and Vancomycin Resistance and Altered Cell Wall Composition in Staphylococcus aureus Carrying the Staphylococcal mecA and the Enterococcal vanA Gene Complex. *Journal of Biological Chemistry*, *279*(5), 3398–3407. https://doi.org/10.1074/jbc.M309593200

Shafaati, M., Boroumand, M., Nowroozi, J., Amiri, P., & Kazemian, H. (2016). Correlation Between qacE and qacEΔ1 Efflux Pump Genes, Antibiotic and Disinfectant Resistant Among Clinical Isolates of E.coli. *Recent Patents on Anti-Infective Drug Discovery*, *11*(2), 189–195. https://doi.org/10.2174/1574891X11666160815094718

Shallcross, L. J., Fragaszy, E., Johnson, A. M., & Hayward, A. C. (2013). The role of the Panton-Valentine leucocidin toxin in staphylococcal disease: a systematic review and meta-analysis. *The Lancet Infectious Diseases*, *13*(1), 43–54. https://doi.org/10.1016/S1473-3099(12)70238-4

Sharma, V. K., Johnston, J. L., Morton, T. M., & Archer, G. L. (1994). Transcriptional regulation by TrsN of conjugative transfer genes on staphylococcal plasmid pGO1. *Journal of Bacteriology*, *176*(12), 3445. https://doi.org/10.1128/JB.176.12.3445-3454.1994

Shetty, V., Trumbull, K., Hegde, A., Shenoy, V., Prabhu, R., Sumathi, K., Palavecino, E., & Shetty, A. K. (2014). Prevalence of Community-Acquired Methicillin-Resistant Staphylococcus aureus Nasal Colonization Among

Children. *Journal of Clinical and Diagnostic Research : JCDR*, *8*(12), DC12–DC15. https://doi.org/10.7860/JCDR/2014/9986.5276

Shiri, T., Nunes, M. C., Adrian, P. V., Van Niekerk, N., Klugman, K. P., & Madhi, S. A. (2013). Interrelationship of Streptococcus pneumoniae, Haemophilus influenzae and Staphylococcus aureus colonization within and between pneumococcal-vaccine naïve mother-child dyads. *BMC Infectious Diseases*, *13*(1), 483. https://doi.org/10.1186/1471-2334-13-483

Shrestha, B., Pokhrel, B. M., & Mohapatra, T. M. (2009). Staphylococcus aureus nasal carriage among health care workers in a Nepal hospital. *Brazilian Journal of Infectious Diseases*, *13*(5), 322. https://doi.org/10.1590/S1413-86702009000500001

Shryock, T. R., Dye, E. S., & Kapral, F. A. (1992). The accumulation of bactericidal lipids in staphylococcal abscesses. *Journal of Medical Microbiology*, *36*(5), 332–336. https://doi.org/10.1099/00222615-36-5-332

Shurland, S., Zhan, M., Bradham, D. D., & Roghmann, M.-C. (2007). Comparison of mortality risk associated with bacteremia due to methicillin-resistant and methicillin-susceptible Staphylococcus aureus. *Infection Control and Hospital Epidemiology*, *28*(3), 273–279. https://doi.org/10.1086/512627

Shuter, J., Hatcher, V. B., & Lowy, F. D. (1996). Staphylococcus aureus binding to human nasal mucin. *Infection and Immunity*, *64*(1), 310. https://doi.org/10.1128/IAI.64.1.310-318.1996

Shutter, M. C., & Akhondi, H. (2022a). Tetracycline. *Kucers the Use of*

Antibiotics: A Clinical Review of Antibacterial, Antifungal, Antiparasitic, and Antiviral Drugs, Seventh Edition, 1195–1203. https://doi.org/10.1201/9781315152110

Shutter, M. C., & Akhondi, H. (2022b). Tetracycline. *Kucers the Use of Antibiotics: A Clinical Review of Antibacterial, Antifungal, Antiparasitic, and Antiviral Drugs, Seventh Edition*, 1195–1203. https://doi.org/10.1201/9781315152110

Sigaúque, B., Roca, A., Mandomando, I., Morais, L., Quintó, L., Sacarlal, J., MacEte, E., Nhamposa, T., MacHevo, S., Aide, P., Bassat, Q., Bardají, A., Nhalungo, D., Soriano-Gabarró, M., Flannery, B., Menendez, C., Levine, M. M., & Alonso, P. L. (2009). Community-acquired bacteremia among children admitted to a rural hospital in Mozambique. *The Pediatric Infectious Disease Journal*, *28*(2), 108–113. https://doi.org/10.1097/INF.0B013E318187A87D

Simons, J. W. F. A., Adams, H., Cox, R. C., Dekker, N., Götz, F., Slotboom, A. J., & Verheij, H. M. (1996). The lipase from Staphylococcus aureus. Expression in Escherichia coli, large-scale purification and comparison of substrate specificity to Staphylococcus hyicus lipase. *European Journal of Biochemistry*, *242*(3), 760–769. https://doi.org/10.1111/J.1432-1033.1996.0760R.X

Simor, A. E., Phillips, E., McGeer, A., Konvalinka, A., Loeb, M., Devlin, H. R., & Kiss, A. (2007). Randomized controlled trial of chlorhexidine gluconate for washing, intranasal mupirocin, and rifampin and doxycycline versus no

treatment for the eradication of methicillin-resistant Staphylococcus aureus colonization. *Clinical Infectious Diseases : An Official Publication of the Infectious Diseases Society of America*, *44*(2), 178–185. https://doi.org/10.1086/510392

Simpson, C. L., Patel, D. M., & Green, K. J. (2011). Deconstructing the skin: cytoarchitectural determinants of epidermal morphogenesis. *Nature Reviews. Molecular Cell Biology*, *12*(9), 565–580. https://doi.org/10.1038/NRM3175

Sinha, B., & Fraunholz, M. (2010). Staphylococcus aureus host cell invasion and post-invasion events. *International Journal of Medical Microbiology : IJMM*, *300*(2–3), 170–175. https://doi.org/10.1016/J.IJMM.2009.08.019

Siu, S. W. I., Campbell-Valois, F.-X., Shoombuatong, W., Jusoh, S. A., Asyraf, M., Masimen, A., Harun, N. A., Maulidiani, M., Iryani, W., & Ismail, W. (2022). Overcoming Methicillin-Resistance Staphylococcus aureus (MRSA) Using Antimicrobial Peptides-Silver Nanoparticles. *Antibiotics 2022, Vol. 11, Page 951*, *11*(7), 951. https://doi.org/10.3390/ANTIBIOTICS11070951

Sivaraman, K., Venkataraman, N., & Cole, A. M. (2009). Staphylococcus aureus nasal carriage and its contributing factors. *Future Microbiology*, *4*(8), 999–1008. https://doi.org/10.2217/FMB.09.79

Skaar, E. P., Gaspar, A. H., & Schneewind, O. (2004). IsdG and IsdI, Heme-degrading Enzymes in the Cytoplasm of Staphylococcus aureus*. *Journal of Biological Chemistry*, *279*(1), 436–443. https://doi.org/10.1074/JBC.M307952200

Skinner, D., & Keefer, C. S. (1941). Significance of bacteremia caused by

staphylococcus aureus: A study of one hundred and twenty-two cases and a review of the literature concerned with experimental infection in animals. *Archives of Internal Medicine*, *68*(5), 851–875. https://doi.org/10.1001/ARCHINTE.1941.00200110003001

Skov, R., Larsen, A. R., Kearns, A., Holmes, M., Teale, C., Edwards, G., & Hill, R. (2014). Phenotypic detection of mecC-MRSA: cefoxitin is more reliable than oxacillin. *Journal of Antimicrobial Chemotherapy*, *69*(1), 133–135. https://doi.org/10.1093/JAC/DKT341

Smith, L. K., & Mankin, A. S. (2008). Transcriptional and translational control of the mlr operon, which confers resistance to seven classes of protein synthesis inhibitors. *Antimicrobial Agents and Chemotherapy*, *52*(5), 1703–1712. https://doi.org/10.1128/AAC.01583-07

Smylie, H. G., Logie, J. R. C., & Smith, G. (1973). From Phisohex to Hibiscrub. *British Medical Journal*, *4*(5892), 586–589. https://doi.org/10.1136/BMJ.4.5892.586

Sollid, J. U. E., Furberg, A. S., Hanssen, A. M., & Johannessen, M. (2014). Staphylococcus aureus: determinants of human carriage. *Infection, Genetics and Evolution : Journal of Molecular Epidemiology and Evolutionary Genetics in Infectious Diseases*, *21*, 531–541. https://doi.org/10.1016/J.MEEGID.2013.03.020

Soto, N. E., Vaghjimal, A., Stahl-Avicolli, A., Protic, J. R., Lutwick, L. I., & Chapnick, E. K. (1999). Bacitracin versus mupirocin for Staphylococcus aureus nasal colonization. *Infection Control and Hospital Epidemiology*,

20(5), 351–353. https://doi.org/10.1086/501633

Sova, M., Čadež, G., Turk, S., Majce, V., Polanc, S., Batson, S., Lloyd, A. J., Roper, D. I., Fishwick, C. W. G., & Gobec, S. (2009). Design and synthesis of new hydroxyethylamines as inhibitors of D-alanyl-D-lactate ligase (VanA) and D-alanyl-D-alanine ligase (DdlB). *Bioorganic & Medicinal Chemistry Letters*, *19*(5), 1376–1379. https://doi.org/10.1016/J.BMCL.2009.01.034

Spaan, A. N., Henry, T., Van Rooijen, W. J. M., Perret, M., Badiou, C., Aerts, P. C., Kemmink, J., De Haas, C. J. C., Van Kessel, K. P. M., Vandenesch, F., Lina, G., & Van Strijp, J. A. G. (2013). The staphylococcal toxin Panton-Valentine Leukocidin targets human C5a receptors. *Cell Host & Microbe*, *13*(5), 584–594. https://doi.org/10.1016/J.CHOM.2013.04.006

Spaan, A. N., Surewaard, B. G. J., Nijland, R., & Van Strijp, J. A. G. (2013). Neutrophils versus Staphylococcus aureus: a biological tug of war. *Annual Review of Microbiology*, *67*, 629–650. https://doi.org/10.1146/ANNUREV-MICRO-092412-155746

Srinivasan, A., Dick, J. D., & Perl, T. M. (2002). Vancomycin resistance in Staphylococci. *Clinical Microbiology Reviews*, *15*(3), 430–438. https://doi.org/10.1128/CMR.15.3.430-438.2002/ASSET/81FCE1EE-E559-4EEB-BB95-8EF299DDCA40/ASSETS/GRAPHIC/CM0320019001.JPEG

Steed, L. L., Costello, J., Lohia, S., Jones, T., Spannhake, E. W., & Nguyen, S. (2014). Reduction of nasal Staphylococcus aureus carriage in health care professionals by treatment with a nonantibiotic, alcohol-based nasal

antiseptic. *American Journal of Infection Control*, *42*(8), 841–846. https://doi.org/10.1016/J.AJIC.2014.04.008

Stefani, S., Bongiorno, D., Mongelli, G., & Campanile, F. (2010). Linezolid Resistance in Staphylococci. *Pharmaceuticals*, *3*(7), 1988. https://doi.org/10.3390/PH3071988

Stefani, S., Campanile, F., Santagati, M., Mezzatesta, M. L., Cafiso, V., & Pacini, G. (2015). Insights and clinical perspectives of daptomycin resistance in Staphylococcus aureus: A review of the available evidence. *International Journal of Antimicrobial Agents*, *46*(3), 278–289. https://doi.org/10.1016/J.IJANTIMICAG.2015.05.008

Steinert, P. M., & Marekov, L. N. (1995). The proteins elafin, filaggrin, keratin intermediate filaments, loricrin, and small proline-rich proteins 1 and 2 are isodipeptide cross-linked components of the human epidermal cornified cell envelope. *The Journal of Biological Chemistry*, *270*(30), 17702–17711. https://doi.org/10.1074/JBC.270.30.17702

Street, C. N., Pedigo, L., Gibbs, A., & Loebel, N. G. (2009). Antimicrobial photodynamic therapy for the decolonization of methicillin-resistant Staphylococcus aureus from the anterior nares. *Photodynamic Therapy: Back to the Future*, *7380*(July 2009), 73803B. https://doi.org/10.1117/12.828279

Stutzmann Meier, P., Entenza, J. M., Vaudaux, P., Francioli, P., Glauser, M. P., & Moreillon, P. (2001). Study of Staphylococcus aureus pathogenic genes by transfer and expression in the less virulent organism Streptococcus gordonii. *Infection and Immunity*, *69*(2), 657–664.

https://doi.org/10.1128/IAI.69.2.657-664.2001

Sugimoto, S., Iwamoto, T., Takada, K., Okuda, K. I., Tajima, A., Iwase, T., & Mizunoe, Y. (2013). Staphylococcus epidermidis Esp Degrades Specific Proteins Associated with Staphylococcus aureus Biofilm Formation and Host-Pathogen Interaction. *Journal of Bacteriology*, *195*(8), 1645. https://doi.org/10.1128/JB.01672-12

Sutter, D. E., Milburn, E., Chukwuma, U., Dzialowy, N., Maranich, A. M., & Hospenthal, D. R. (2016). Changing Susceptibility of Staphylococcus aureus in a US Pediatric Population. *Pediatrics*, *137*(4). https://doi.org/10.1542/PEDS.2015-3099

Suttorp, N., Fuhrmann, M., Tannert-Otto, S., Grimminger, F., & Bhakdi, S. (1993). Pore-forming bacterial toxins potently induce release of nitric oxide in porcine endothelial cells. *The Journal of Experimental Medicine*, *178*(1), 337–341. https://doi.org/10.1084/JEM.178.1.337

Tacconelli, E. (2009). Antimicrobial use: risk driver of multidrug resistant microorganisms in healthcare settings. *Current Opinion in Infectious Diseases*, *22*(4), 352–358. https://doi.org/10.1097/QCO.0B013E32832D52E0

Tam, K., & Torres, V. J. (2019). Staphylococcus aureus Secreted Toxins & Extracellular Enzymes. *Microbiology Spectrum*, *7*(2). https://doi.org/10.1128/MICROBIOLSPEC.GPP3-0039-2018

Tanabe, M., Szakonyi, G., Brown, K. A., Henderson, P. J. F., Nield, J., & Byrne, B. (2009). The multidrug resistance efflux complex, EmrAB from

Escherichia coli forms a dimer in vitro. *Biochemical and Biophysical Research Communications*, *380*(2), 338–342. https://doi.org/10.1016/J.BBRC.2009.01.081

Tang, J., Zhou, R., Shi, X., Kang, M., Wang, H., & Chen, H. (2008). Two thermostable nucleases coexisted in Staphylococcus aureus: evidence from mutagenesis and in vitro expression. *FEMS Microbiology Letters*, *284*(2), 176–183. https://doi.org/10.1111/J.1574-6968.2008.01194.X

Tang, S. S., Apisarnthanarak, A., & Hsu, L. Y. (2014). Mechanisms of β-lactam antimicrobial resistance and epidemiology of major community- and healthcare-associated multidrug-resistant bacteria. *Advanced Drug Delivery Reviews*, *78*, 3–13. https://doi.org/10.1016/J.ADDR.2014.08.003

Taylor, C. R., Shi, S. R., Barr, N. J., & Wu, N. (2006). Techniques of Immunohistochemistry: Principles, Pitfalls and Standardization. *Diagnostic Immunohistochemistry: Second Edition*, 1–42. https://doi.org/10.1016/B978-0-443-06652-8.50007-7

Taylor, S. D., & Palmer, M. (2016). The action mechanism of daptomycin. *Bioorganic & Medicinal Chemistry*, *24*(24), 6253–6268. https://doi.org/10.1016/J.BMC.2016.05.052

Tenover, F. C., McDougal, L. K., Goering, R. V., Killgore, G., Projan, S. J., Patel, J. B., & Dunman, P. M. (2006). Characterization of a strain of community-associated methicillin-resistant Staphylococcus aureus widely disseminated in the United States. *Journal of Clinical Microbiology*, *44*(1), 108–118. https://doi.org/10.1128/JCM.44.1.108-118.2006

Tewodros, W., & Gedebou, M. (1984). Nasal carrier rates and antibiotic resistance of Staphylococcus aureus isolates from hospital and non-hospital populations, Addis Ababa. *Transactions of the Royal Society of Tropical Medicine and Hygiene*, *78*(3), 314–318. https://doi.org/10.1016/0035-9203(84)90105-6

Thammavongsa, V., Missiakas, D. M., & Schneewind, O. (2013). Staphylococcus aureus degrades neutrophil extracellular traps to promote immune cell death. *Science (New York, N.Y.)*, *342*(6160), 863–866. https://doi.org/10.1126/SCIENCE.1242255

Thapa, D., Pyakurel, S., Thapa, S., Lamsal, S., Chaudhari, M., Adhikari, N., & Shrestha, D. (2021). Staphylococcus aureus with inducible clindamycin resistance and methicillin resistance in a tertiary hospital in Nepal. *Tropical Medicine and Health*, *49*(1), 1–7. https://doi.org/10.1186/S41182-021-00392-2/TABLES/3

Thiolas, A., Bornet, C., Davin-Régli, A., Pagès, J. M., & Bollet, C. (2004). Resistance to imipenem, cefepime, and cefpirome associated with mutation in Omp36 osmoporin of Enterobacter aerogenes. *Biochemical and Biophysical Research Communications*, *317*(3), 851–856. https://doi.org/10.1016/j.bbrc.2004.03.130

Thomas, S., Izard, J., Walsh, E., Batich, K., Chongsathidkiet, P., Clarke, G., Sela, D. A., Muller, A. J., Mullin, J. M., Albert, K., Gilligan, J. P., DiGuilio, K., Dilbarova, R., Alexander, W., & Prendergast, G. C. (2017). The Host Microbiome Regulates and Maintains Human Health: A Primer and

Perspective for Non-Microbiologists. *Cancer Research*, *77*(8), 1783. https://doi.org/10.1158/0008-5472.CAN-16-2929

Tigabu, A., Tiruneh, M., & Mekonnen, F. (2018). Nasal Carriage Rate, Antimicrobial Susceptibility Pattern, and Associated Factors of Staphylococcus aureus with Special Emphasis on MRSA among Urban and Rural Elementary School Children in Gondar, Northwest Ethiopia: A Comparative Cross-Sectional Study. *Advances in Preventive Medicine*, *2018*, 1–11. https://doi.org/10.1155/2018/9364757

Tillotson, G. S., & Zinner, S. H. (2017). Burden of antimicrobial resistance in an era of decreasing susceptibility. *Expert Review of Anti-Infective Therapy*, *15*(7), 663–676. https://doi.org/10.1080/14787210.2017.1337508

Toh, S. M., Xiong, L., Arias, C. A., Villegas, M. V., Lolans, K., Quinn, J., & Mankin, A. S. (2007). Acquisition of a natural resistance gene renders a clinical strain of methicillin-resistant Staphylococcus aureus resistant to the synthetic antibiotic linezolid. *Molecular Microbiology*, *64*(6), 1506–1514. https://doi.org/10.1111/J.1365-2958.2007.05744.X

Tomasz, A., Nachman, S., & Leaf, H. (1991). Stable classes of phenotypic expression in methicillin-resistant clinical isolates of staphylococci. *Antimicrobial Agents and Chemotherapy*, *35*(1), 124–129. https://doi.org/10.1128/AAC.35.1.124

Tong, S. Y. C., Davis, J. S., Eichenberger, E., Holland, T. L., & Fowler, V. G. (2015). Staphylococcus aureus Infections: Epidemiology, Pathophysiology, Clinical Manifestations, and Management. *Clinical Microbiology Reviews*,

28(3), 603. https://doi.org/10.1128/CMR.00134-14

Tsiodras, S., Gold, H. S., Sakoulas, G., Eliopoulos, G. M., Wennersten, C., Venkataraman, L., Moellering, R. C., & Ferraro, M. J. (2001). Linezolid resistance in a clinical isolate of Staphylococcus aureus. *Lancet (London, England)*, *358*(9277), 207–208. https://doi.org/10.1016/S0140-6736(01)05410-1

Turlej, A., Hryniewicz, W., & Empel, J. (2011). Staphylococcal Cassette Chromosome mec (SCCmec) classification and typing methods: An overview. *Polish Journal of Microbiology*, *60*(2), 95–103. https://doi.org/10.33073/pjm-2011-013

Udobi, C. E., Obajuluwa, A. F., & Onaolapo, J. A. (2013). Prevalence and antibiotic resistance pattern of methicillin-resistant staphylococcus aureus from an orthopaedic hospital in nigeria. *BioMed Research International*, *2013*. https://doi.org/10.1155/2013/860467

Uehara, Y., Nakama, H., Agematsu, K., Uchida, M., Kawakami, Y., Abdul Fattah, A. S. M., & Maruchi, N. (2000). Bacterial interference among nasal inhabitants: eradication of Staphylococcus aureus from nasal cavities by artificial implantation of Corynebacterium sp. *Journal of Hospital Infection*, *44*(2), 127–133. https://doi.org/10.1053/JHIN.1999.0680

Ueta, M., Wada, C., & Wada, A. (2010). Formation of 100S ribosomes in Staphylococcus aureus by the hibernation promoting factor homolog SaHPF. *Genes to Cells*, *15*(1), 43–58. https://doi.org/10.1111/J.1365-2443.2009.01364.X

Ullah, F., Malik, S. A., Ahmed, J., Ullah, F., Shah, S. M., Ayaz, M., Hussain, S., & Khatoon, L. (2013). Investigation of the Genetic Basis of Tetracycline Resistance in *Staphylococcus aureus* from Pakistan. *Tropical Journal of Pharmaceutical Research*, *11*(6), 925–931. https://doi.org/10.4314/tjpr.v11i6.8

Van Acker, H., Van Dijck, P., & Coenye, T. (2014). Molecular mechanisms of antimicrobial tolerance and resistance in bacterial and fungal biofilms. *Trends in Microbiology*, *22*(6), 326–333. https://doi.org/10.1016/J.TIM.2014.02.001

van Belkum, A., Emonts, M., Wertheim, H., de Jongh, C., Nouwen, J., Bartels, H., Cole, A., Cole, A., Hermans, P., Boelens, H., Toom, N. L. den, Snijders, S., Verbrugh, H., & van Leeuwen, W. (2007). The role of human innate immune factors in nasal colonization by Staphylococcus aureus. *Microbes and Infection*, *9*(12–13), 1471–1477. https://doi.org/10.1016/J.MICINF.2007.08.003

van Belkum, A., Melles, D. C., Nouwen, J., van Leeuwen, W. B., van Wamel, W., Vos, M. C., Wertheim, H. F. L., & Verbrugh, H. A. (2009). Co-evolutionary aspects of human colonisation and infection by Staphylococcus aureus. *Infection, Genetics and Evolution : Journal of Molecular Epidemiology and Evolutionary Genetics in Infectious Diseases*, *9*(1), 32–47. https://doi.org/10.1016/J.MEEGID.2008.09.012

Van Belkum, A., Verkalk, N. J., De Vogel, C. P., Boelens, H. A., Verveer, J., Nouwen, J. L., Verbrugh, H. A., & Wertheim, H. F. L. (2009).

Reclassification of Staphylococcus aureus Nasal Carriage Types. *The Journal of Infectious Diseases*, *199*(12), 1820–1826. https://doi.org/10.1086/599119

van Hal, S. J., Jensen, S. O., Vaska, V. L., Espedido, B. A., Paterson, D. L., & Gosbell, I. B. (2012). Predictors of mortality in Staphylococcus aureus Bacteremia. *Clinical Microbiology Reviews*, *25*(2), 362–386. https://doi.org/10.1128/CMR.05022-11

van Rijen, M. M. L., Bonten, M., Wenzel, R. P., & Kluytmans, J. A. J. W. (2008). Intranasal mupirocin for reduction of Staphylococcus aureus infections in surgical patients with nasal carriage: a systematic review. *The Journal of Antimicrobial Chemotherapy*, *61*(2), 254–261. https://doi.org/10.1093/JAC/DKM480

Vandenbroucke-Grauls, C. M., Thijssen, H. M., & Verhoef, J. (1984). Interaction between human polymorphonuclear leucocytes and Staphylococcus aureus in the presence and absence of opsonins. *Immunology*, *52*(3), 427–435. http://www.ncbi.nlm.nih.gov/pubmed/6086502%0Ahttp://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=PMC1454476

Vandenesch, F., Naimi, T., Enright, M. C., Lina, G., Nimmo, G. R., Heffernan, H., Liassine, N., Bes, M., Greenland, T., Reverdy, M. E., & Etienne, J. (2003). Community-Acquired Methicillin-Resistant Staphylococcus aureus Carrying Panton-Valentine Leukocidin Genes: Worldwide Emergence - Volume 9, Number 8—August 2003 - Emerging Infectious Diseases journal - CDC. *Emerging Infectious Diseases*, *9*(8), 978–984.

https://doi.org/10.3201/EID0908.030089

Vara Prasad, J. (2007). New oxazolidinones. *Current Opinion in Microbiology*, *10*(5), 454–460. https://doi.org/10.1016/J.MIB.2007.08.001

Vedantam, G., Guay, G. G., Austria, N. E., Doktor, S. Z., & Nichols, B. P. (1998). Characterization of mutations contributing to sulfathiazole resistance in Escherichia coli. *Antimicrobial Agents and Chemotherapy*, *42*(1), 88–93. https://doi.org/10.1128/AAC.42.1.88

Vergara-Irigaray, M., Maira-Litrán, T., Merino, N., Pier, G. B., Penadés, J. R., & Lasa, I. (2008). Wall teichoic acids are dispensable for anchoring the PNAG exopolysaccharide to the Staphylococcus aureus cell surface. *Microbiology (Reading, England)*, *154*(Pt 3), 865. https://doi.org/10.1099/MIC.0.2007/013292-0

Verkaik, N. J., De Vogel, C. P., Boelens, H. A., Grumann, D., Hoogenboezem, T., Vink, C., Hooijkaas, H., Foster, T. J., Verbrugh, H. A., Van Belkum, A., & Van Wamel, W. J. B. (2009). Anti-Staphylococcal Humoral Immune Response in Persistent Nasal Carriers and Noncarriers of Staphylococcus aureus. *The Journal of Infectious Diseases*, *199*(5), 625–632. https://doi.org/10.1086/596743

Viana, D., Blanco, J., Tormo-Más, M. Á., Selva, L., Guinane, C. M., Baselga, R., Corpa, J. M., Lasa, Í., Novick, R. P., Fitzgerald, J. R., & Penadés, J. R. (2010). Adaptation of Staphylococcus aureus to ruminant and equine hosts involves SaPI-carried variants of von Willebrand factor-binding protein. *Molecular Microbiology*, *77*(6), 1583–1594. https://doi.org/10.1111/J.1365-

2958.2010.07312.X

Villagra, N. A., Fuentes, J. A., Jofré, M. R., Hidalgo, A. A., García, P., & Mora, G. C. (2012). The carbon source influences the efflux pump-mediated antimicrobial resistance in clinically important Gram-negative bacteria. *The Journal of Antimicrobial Chemotherapy*, *67*(4), 921–927. https://doi.org/10.1093/JAC/DKR573

Visansirikul, S., Kolodziej, S. A., & Demchenko, A. V. (2020). Staphylococcus aureus capsular polysaccharides: a structural and synthetic perspective. *Organic & Biomolecular Chemistry*, *18*(5), 783–798. https://doi.org/10.1039/C9OB02546D

von Eiff, C., Becker, K., Machka, K., Stammer, H., & Peters, G. (2001). Nasal carriage as a source of Staphylococcus aureus bacteremia. Study Group. *The New England Journal of Medicine*, *344*(1), 11–16. https://doi.org/10.1056/NEJM200101043440102

Walana, W., Bobzah, B. P., Kuugbee, E. D., Acquah, S., Ezekiel, V. K., Yabasin, I. B., Abdul-Mumin, A., & Ziem, J. B. (2020). Staphylococcus aureus nasal carriage among healthcare workers, inpatients and caretakers in the Tamale Teaching Hospital, Ghana. *Scientific African*, *8*, e00325. https://doi.org/10.1016/J.SCIAF.2020.E00325

Walsh, C. T., Fisher, S. L., Park, I. S., Prahalad, M., & Wu, Z. (1996). Bacterial resistance to vancomycin: five genes and one missing hydrogen bond tell the story. *Chemistry & Biology*, *3*(1), 21–28. https://doi.org/10.1016/S1074-5521(96)90079-4

Walsh, C., & Wencewicz, T. A. (2016). *Antibiotics : challenges, mechanisms, opportunities*. 477. https://www.wiley.com/en-us/Antibiotics%3A+Challenges%2C+Mechanisms%2C+Opportunities%2C+2nd+Edition-p-9781555819309

Walsh, T. J., Standiford, H. C., Reboli, A. C., John, J. F., Mulligan, M. E., Ribner, B. S., Montgomerie, J. Z., Goetz, M. B., Mayhall, C. G., Rimland, D., Stevens, D. A., Hansen, S. L., Gerard, G. C., & Ragual, R. J. (1993). Randomized double-blinded trial of rifampin with either novobiocin or trimethoprim-sulfamethoxazole against methicillin-resistant Staphylococcus aureus colonization: prevention of antimicrobial resistance and effect of host factors on outcome. *Antimicrobial Agents and Chemotherapy*, *37*(6), 1334–1342. https://doi.org/10.1128/AAC.37.6.1334

Walter, E. I., Ratnoff, W. D., Long, K. E., Kazura, J. W., & Medof, M. E. (1992). Effect of glycoinositolphospholipid anchor lipid groups on functional properties of decay-accelerating factor protein in cells. *Journal of Biological Chemistry*, *267*(2), 1245–1252. https://doi.org/10.1016/s0021-9258(18)48421-9

Wang, R., Braughton, K. R., Kretschmer, D., Bach, T. H. L., Queck, S. Y., Li, M., Kennedy, A. D., Dorward, D. W., Klebanoff, S. J., Peschel, A., DeLeo, F. R., & Otto, M. (2007). Identification of novel cytolytic peptides as key virulence determinants for community-associated MRSA. *Nature Medicine*, *13*(12), 1510–1514. https://doi.org/10.1038/NM1656

Wangai, F. K., Masika, M. M., Maritim, M. C., & Seaton, R. A. (2019).

Methicillin-resistant Staphylococcus aureus (MRSA) in East Africa: Red alert or red herring? *BMC Infectious Diseases*, *19*(1), 1–10. https://doi.org/10.1186/S12879-019-4245-3/FIGURES/2

Wanke, I., Steffen, H., Christ, C., Krismer, B., Götz, F., Peschel, A., Schaller, M., & Schittek, B. (2011). Skin commensals amplify the innate immune response to pathogens by activation of distinct signaling pathways. *The Journal of Investigative Dermatology*, *131*(2), 382–390. https://doi.org/10.1038/JID.2010.328

Wann, E. R., Gurusiddappa, S., & Höök, M. (2000). The Fibronectin-binding MSCRAMM FnbpA ofStaphylococcus aureus Is a Bifunctional Protein That Also Binds to Fibrinogen *. *Journal of Biological Chemistry*, *275*(18), 13863–13871. https://doi.org/10.1074/JBC.275.18.13863

Warnke, P., Harnack, T., Ottl, P., Kundt, G., & Podbielski, A. (2014). Nasal Screening for Staphylococcus aureus – Daily Routine with Improvement Potentials. *PLoS ONE*, *9*(2). https://doi.org/10.1371/JOURNAL.PONE.0089667

Warren, H. S., Tompkins, R. G., Moldawer, L. L., Seok, J., Xu, W., Mindrinos, M. N., Maier, R. V., Xiao, W., & Davis, R. W. (2015). Mice are not men. *Proceedings of the National Academy of Sciences of the United States of America*, *112*(4), E345. https://doi.org/10.1073/PNAS.1414857111

Watanabe, S., Ito, T., Takeuchi, F., Endo, M., Okuno, E., & Hiramatsu, K. (2005). Structural comparison of ten serotypes of staphylocoagulases in Staphylococcus aureus. *Journal of Bacteriology*, *187*(11), 3698–3707.

https://doi.org/10.1128/JB.187.11.3698-3707.2005

Wegener, H. C. (2012). *ANTIBIOTIC RESISTANCE—LINKING HUMAN AND ANIMAL HEALTH*. https://www.ncbi.nlm.nih.gov/books/NBK114485/

Weidenmaier, C., Kokai-Kun, J. F., Kristian, S. A., Chanturiya, T., Kalbacher, H., Gross, M., Nicholson, G., Neumeister, B., Mond, J. J., & Peschel, A. (2004). Role of teichoic acids in Staphylococcus aureus nasal colonization, a major risk factor in nosocomial infections. *Nature Medicine 2004 10:3*, *10*(3), 243–245. https://doi.org/10.1038/nm991

Weidenmaier, C., & Peschel, A. (2008). Teichoic acids and related cell-wall glycopolymers in Gram-positive physiology and host interactions. *Nature Reviews. Microbiology*, *6*(4), 276–287. https://doi.org/10.1038/NRMICRO1861

Weigel, L. M., Clewell, D. B., Gill, S. R., Clark, N. C., McDougal, L. K., Flannagan, S. E., Kolonay, J. F., Shetty, J., Killgore, G. E., & Tenover, F. C. (2003). Genetic Analysis of a High-Level Vancomycin-Resistant Isolate of Staphylococcus aureus. *Science*, *302*(5650), 1569–1571. https://doi.org/10.1126/SCIENCE.1090956/SUPPL_FILE/WEIGEL_SOM.PDF

Wenzel, R. P., & Edmond, M. B. (2010). Infection control: the case for horizontal rather than vertical interventional programs. *International Journal of Infectious Diseases : IJID : Official Publication of the International Society for Infectious Diseases*, *14 Suppl 4*(SUPPL. 4). https://doi.org/10.1016/J.IJID.2010.05.002

Werner, G., Strommenger, B., & Witte, W. (2008). Acquired vancomycin resistance in clinically relevant pathogens. *Future Microbiology*, *3*(5), 547–562. https://doi.org/10.2217/17460913.3.5.547

Wertheim, H. F. L., Melles, D. C., Vos, M. C., Van Leeuwen, W., Van Belkum, A., Verbrugh, H. A., & Nouwen, J. L. (2005). The role of nasal carriage in Staphylococcus aureus infections. *The Lancet. Infectious Diseases*, *5*(12), 751–762. https://doi.org/10.1016/S1473-3099(05)70295-4

Wertheim, H. F. L., Vos, M. C., Boelens, H. A. M., Voss, A., Vandenbroucke-Grauls, C. M. J. E., Meester, M. H. M., Kluytmans, J. A. J. W., van Keulen, P. H. J., & Verbrugh, H. A. (2004). Low prevalence of methicillin-resistant Staphylococcus aureus (MRSA) at hospital admission in the Netherlands: the value of search and destroy and restrictive antibiotic use. *The Journal of Hospital Infection*, *56*(4), 321–325. https://doi.org/10.1016/J.JHIN.2004.01.026

Wertheim, H. F. L., Vos, M. C., Ott, A., Van Belkum, A., Voss, A., Kluytmans, J. A. J. W., Van Keulen, P. H. J., Vandenbroucke-Grauls, C. M. J. E., Meester, M. H. M., & Verbrugh, H. A. (2004a). Risk and outcome of nosocomial Staphylococcus aureus bacteraemia in nasal carriers versus non-carriers. *Lancet (London, England)*, *364*(9435), 703–705. https://doi.org/10.1016/S0140-6736(04)16897-9

Wertheim, H. F. L., Vos, M. C., Ott, A., Van Belkum, A., Voss, A., Kluytmans, J. A. J. W., Van Keulen, P. H. J., Vandenbroucke-Grauls, C. M. J. E., Meester, M. H. M., & Verbrugh, H. A. (2004b). Risk and outcome of nosocomial

Staphylococcus aureus bacteraemia in nasal carriers versus non-carriers. *Lancet (London, England)*, *364*(9435), 703–705. https://doi.org/10.1016/S0140-6736(04)16897-9

White, M. J., Boyd, J. M., Horswill, A. R., & Nauseef, W. M. (2014). Phosphatidylinositol-specific phospholipase C contributes to survival of Staphylococcus aureus USA300 in human blood and neutrophils. *Infection and Immunity*, *82*(4), 1559–1571. https://doi.org/10.1128/IAI.01168-13

WHO. (2014). *Antimicrobial resistance: global report on surveillance*.

WHO. (2020). *GHO | By category | Number of deaths by region - Acute lower respiratory infections*. https://apps.who.int/gho/data/view.main.CM1002015REG6-CH9?lang=en

Wiley, B. B., & Maverakis, N. H. (1974). Capsule production and virulence among strains of Staphylococcus aureus. *Annals of the New York Academy of Sciences*, *236*(0), 221–232. https://doi.org/10.1111/J.1749-6632.1974.TB41493.X

Wilke, G. A., & Wardenburg, J. B. (2010). Role of a disintegrin and metalloprotease 10 in Staphylococcus aureus α-hemolysin - Mediated cellular injury. *Proceedings of the National Academy of Sciences of the United States of America*, *107*(30), 13473–13478. https://doi.org/10.1073/PNAS.1001815107/SUPPL_FILE/PNAS.201001815SI.PDF

Williams, R. E. O. (1963). HEALTHY CARRIAGE OF STAPHYLOCOCCUS AUREUS: ITS PREVALENCE AND IMPORTANCE. *Bacteriological*

Reviews, *27*(1), 56. https://doi.org/10.1128/BR.27.1.56-71.1963

Wilson, D. N. (2009). The A-Z of bacterial translation inhibitors. *Critical Reviews in Biochemistry and Molecular Biology*, *44*(6), 393–433. https://doi.org/10.3109/10409230903307311

Wilson, M. (1993). Photolysis of oral bacteria and its potential use in the treatment of caries and periodontal disease. *The Journal of Applied Bacteriology*, *75*(4), 299–306. https://doi.org/10.1111/J.1365-2672.1993.TB02780.X

Winkler, K. C., de Waart, J., & Grootsen, C. (1965). Lysogenic conversion of staphylococci to loss of beta-toxin. *Journal of General Microbiology*, *39*(3), 321–333. https://doi.org/10.1099/00221287-39-3-321

Winstel, V., Xia, G., & Peschel, A. (2013). *Pathways and roles of wall teichoic acid glycosylation in Staphylococcus aureus ARTICLE IN PRESS*. https://doi.org/10.1016/j.ijmm.2013.10.009

Wood, T. K., Knabel, S. J., & Kwan, B. W. (2013). Bacterial persister cell formation and dormancy. *Applied and Environmental Microbiology*, *79*(23), 7116–7121. https://doi.org/10.1128/AEM.02636-13

WOODIN, A. M. (1960). Purification of the two components of leucocidin from Staphylococcus aureus. *The Biochemical Journal*, *75*(1), 158–165. https://doi.org/10.1042/BJ0750158

Woodruff, T. M., Nandakumar, K. S., & Tedesco, F. (2011). Inhibiting the C5–C5a receptor axis. *Molecular Immunology*, *48*(14), 1631–1642. https://doi.org/10.1016/J.MOLIMM.2011.04.014

World Health Organization. (2014). *World Health Statistics 2014*. World Health Organization. http://apps.who.int/iris/bitstream/handle/10665/112738/9789240692671_eng.pdf;jsessionid=7D7E8AD71E3388832E9122ADBD0747DC?sequence=1

World Health Organization. (2015). WHO Library Cataloguing-in-Publication Data Global Action Plan on Antimicrobial Resistance. *Microbe Magazine*, *10*(9), 354–355. www.paprika-annecy.com

Wu, Z., Li, F., Liu, D., Xue, H., & Zhao, X. (2015). Novel Type XII Staphylococcal Cassette Chromosome mec Harboring a New Cassette Chromosome Recombinase, CcrC2. *Antimicrobial Agents and Chemotherapy*, *59*(12), 7597–7601. https://doi.org/10.1128/AAC.01692-15

Wyllie, D. H., Peto, T. E. A., & Crook, D. (2005). MRSA bacteraemia in patients on arrival in hospital: a cohort study in Oxfordshire 1997-2003. *BMJ (Clinical Research Ed.)*, *331*(7523), 992–995. https://doi.org/10.1136/BMJ.38558.453310.8F

Xhindoli, D., Pacor, S., Benincasa, M., Scocchi, M., Gennaro, R., & Tossi, A. (2016). The human cathelicidin LL-37 — A pore-forming antibacterial peptide and host-cell modulator. *Biochimica et Biophysica Acta (BBA) - Biomembranes*, *1858*(3), 546–566. https://doi.org/10.1016/J.BBAMEM.2015.11.003

Xu, X., Lin, D., Yan, G., Ye, X., Wu, S., Guo, Y., Zhu, D., Hu, F., Zhang, Y., Wang, F., Jacoby, G. A., & Wang, M. (2010). vanM, a new glycopeptide resistance gene cluster found in Enterococcus faecium. *Antimicrobial Agents*

and Chemotherapy, *54*(11), 4643–4647. https://doi.org/10.1128/AAC.01710-09

Yan, M., Pamp, S. J., Fukuyama, J., Hwang, P. H., Cho, D. Y., Holmes, S., & Relman, D. A. (2013). Nasal microenvironments and interspecific interactions influence nasal microbiota complexity and S. aureus carriage. *Cell Host & Microbe*, *14*(6), 631–640. https://doi.org/10.1016/J.CHOM.2013.11.005

Yang, S. J., Kreiswirth, B. N., Sakoulas, G., Yeaman, M. R., Xiong, Y. Q., Sawa, A., & Bayer, A. S. (2009). Enhanced expression of dltABCD is associated with the development of daptomycin nonsusceptibility in a clinical endocarditis isolate of Staphylococcus aureus. *The Journal of Infectious Diseases*, *200*(12), 1916–1920. https://doi.org/10.1086/648473

Yarwood, J. M., Bartels, D. J., Volper, E. M., & Greenberg, E. P. (2004). Quorum sensing in Staphylococcus aureus biofilms. *Journal of Bacteriology*, *186*(6), 1838–1850. https://doi.org/10.1128/JB.186.6.1838-1850.2004

Yimin, Kohanawa, M., Zhao, S., Ozaki, M., Haga, S., Nan, G., Kuge, Y., & Tamaki, N. (2013). Contribution of Toll-Like Receptor 2 to the Innate Response against Staphylococcus aureus Infection in Mice. *PLOS ONE*, *8*(9), e74287. https://doi.org/10.1371/JOURNAL.PONE.0074287

Yoshida, H., Ueta, M., Maki, Y., Sakai, A., & Wada, A. (2009). Activities of Escherichia coli ribosomes in IF3 and RMF change to prepare 100S ribosome formation on entering the stationary growth phase. *Genes to Cells : Devoted to Molecular & Cellular Mechanisms*, *14*(2), 271–280.

https://doi.org/10.1111/J.1365-2443.2008.01272.X

Yu, V. L. (2011). Guidelines for hospital-acquired pneumonia and health-care-associated pneumonia: a vulnerability, a pitfall, and a fatal flaw. *The Lancet. Infectious Diseases*, *11*(3), 248–252. https://doi.org/10.1016/S1473-3099(11)70005-6

Zhang, H. Z., Hackbarth, C. J., Chansky, K. M., & Chambers, H. F. (2001). A proteolytic transmembrane signaling pathway and resistance to beta-lactams in staphylococci. *Science (New York, N.Y.)*, *291*(5510), 1962–1965. https://doi.org/10.1126/SCIENCE.1055144

Zhang, N., Holtappels, G., Gevaert, P., Patou, J., Dhaliwal, B., Gould, H., & Bachert, C. (2011). Mucosal tissue polyclonal IgE is functional in response to allergen and SEB. *Allergy*, *66*(1), 141–148. https://doi.org/10.1111/J.1398-9995.2010.02448.X

Zhao, N., Cheng, D., Jian, Y., Liu, Y., Liu, J., Huang, Q., He, L., Wang, H., Miao, F., Li, M., & Liu, Q. (2021). Molecular characteristics of Staphylococcus aureus isolates colonizing human nares and skin. *Medicine in Microecology*, *7*, 100031. https://doi.org/10.1016/J.MEDMIC.2020.100031

Zhu, W., Wilks, A., & Stojiljkovic, I. (2000). Degradation of heme in gram-negative bacteria: the product of the hemO gene of Neisseriae is a heme oxygenase. *Journal of Bacteriology*, *182*(23), 6783–6790. https://doi.org/10.1128/JB.182.23.6783-6790.2000

Ziakas, P. D., Zacharioudakis, I. M., Zervou, F. N., & Mylonakis, E. (2015). Methicillin-resistant Staphylococcus aureus prevention strategies in the ICU:

a clinical decision analysis*. *Critical Care Medicine*, *43*(2), 382–393. https://doi.org/10.1097/CCM.0000000000000711

Zipperer, A., Konnerth, M. C., Laux, C., Berscheid, A., Janek, D., Weidenmaier, C., Burian, M., Schilling, N. A., Slavetinsky, C., Marschal, M., Willmann, M., Kalbacher, H., Schittek, B., Brötz-Oesterhelt, H., Grond, S., Peschel, A., & Krismer, B. (2016). Human commensals producing a novel antibiotic impair pathogen colonization. *Nature 2016 535:7613*, *535*(7613), 511–516. https://doi.org/10.1038/nature18634

Zivkovic, A., Sharif, O., Stich, K., Doninger, B., Biaggio, M., Colinge, J., Bilban, M., Mesteri, I., Hazemi, P., Lemmens-Gruber, R., & Knapp, S. (2011). TLR 2 and CD14 Mediate Innate Immunity and Lung Inflammation to Staphylococcal Panton–Valentine Leukocidin In Vivo. *The Journal of Immunology*, *186*(3), 1608–1617. https://doi.org/10.4049/JIMMUNOL.1001665

Zong, Z., Peng, C., & Lü, X. (2011). Diversity of SCCmec elements in methicillin-resistant coagulase-negative staphylococci clinical isolates. *PloS One*, *6*(5). https://doi.org/10.1371/JOURNAL.PONE.0020191

APPENDICES

Appendix A: Ethical Clearance

UNIVERSITY OF CAPE COAST
INSTITUTIONAL REVIEW BOARD SECRETARIAT
TEL: 0558093143 / 0508878309
E-MAIL: irb@ucc.edu.gh
OUR REF: UCC/IRB/2B/352
YOUR REF:

18TH MAY, 2022

Dr. Richael Odarkor Mills
Department of Biomedical Sciences
School of Allied Health Sciences
University of Cape coast

Dear Dr. Mills,

RE: REQUEST FOR EXTENSION OF ETHICAL CLEARANCE TO COVER NEW STUDY (ETHICAL CLEARANCE ID – UCCIRB/EXT/2017/21)

The University of Cape Coast Institutional Review Board (UCCIRB) refers to your letter dated 26th April, 2022 with reference number SAHS/DBS/E.3/VOL.1/2 on the above subject matter. The University of Cape Coast Institutional Review Board (UCCIRB) has granted you an extension of your ethical clearance for your research titled ***Nasopharyngeal carriage of Bacteria Pathogens in the Nasopharynx of Children less than five years in Cape Coast Ghana.*** The approval is subject to the following conditions:

1. That you will continue to be the Principal Investigator of the proposed research;
2. That the approved research instruments would be the same instruments to be used by the three project implementing agencies;
3. That the approved category of respondents and study sites would remain the same;
4. That this extension is used to conduct further analysis on "nasopharyngeal carriage of bacteria pathogens in the nasopharynx of children less than five years in Cape Coast, Ghana" based on your archived samples from the study titled "Post-vaccination pneumococcal carriage in children less than five years of age in Cape Coast, Ghana."
5. That this extension will expire on 1st November, 2022.

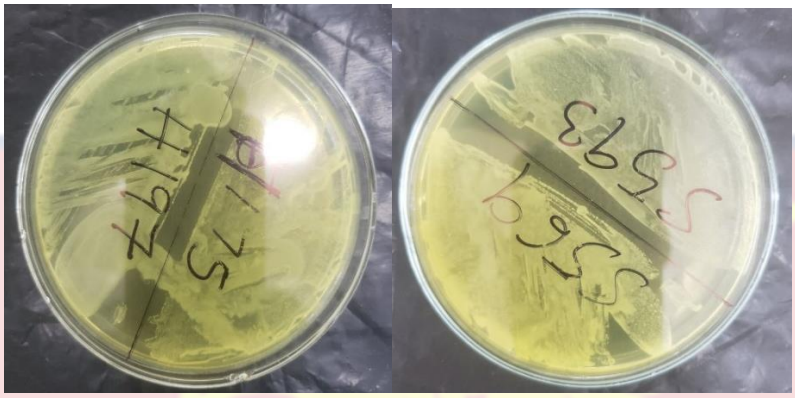
You may contact us for further information on this, if need be.

Yours faithfully,

Samuel Asiedu Owusu, PhD
UCCIRB Administrator

ADMINISTRATOR
INSTITUTIONAL REVIEW BOARD
UNIVERSITY OF CAPE COAST

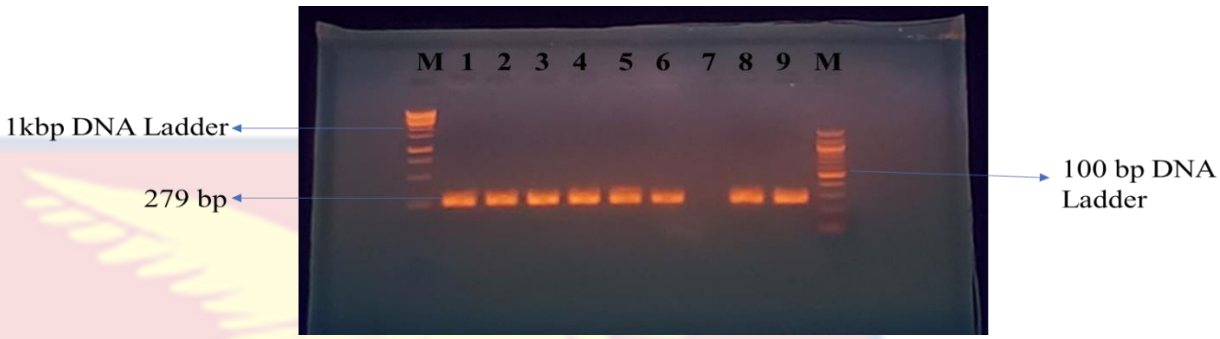
Appendix B: Tests for Identification of *S. aureus*



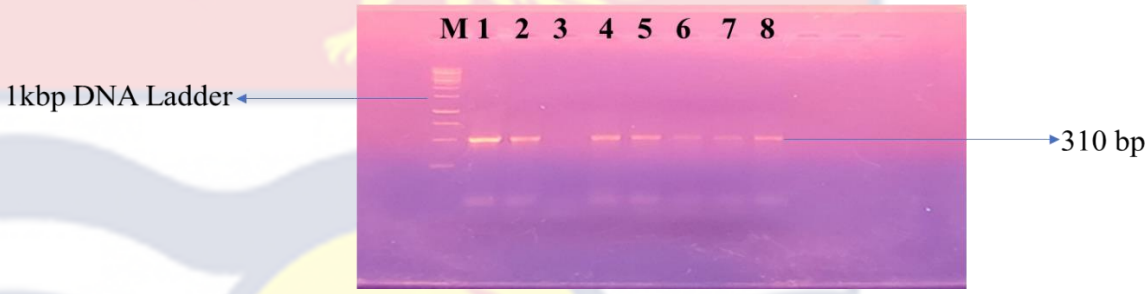
Appendix C: Susceptibility testing of *S. aureus*

Appendix D: DNA Quantification Using NanoDrop Microvolume Spectrophotometer

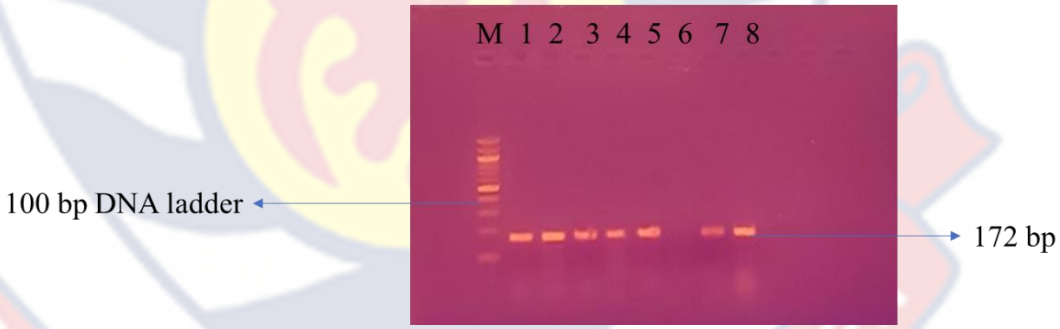
Appendix E: Image of Agarose gel showing *nuc* amplicons



Appendix F: Image of Agarose gel showing *mecA* amplicons



Appendix F: Image of Agarose gel showing *blaZ* amplicons



Appendix G: Image of Agarose gel showing *tetK* amplicons