








Immune System Evasion Mechanisms in *Staphylococcus aureus*: Current Understanding

Hesham A. Malak^{1,2} , Hussein H. Abulreesh^{1,2*} , Sameer R. Organji^{1,2},
Khaled Elbanna^{1,2,3} , Mohammed R. Shaaban^{1,2,4}, Samreen⁵, Iqbal Ahmad⁵ ,
Ashwag Shami⁶ , Wafa A. Alshehri⁷ , Ashjan Khalel⁸ , Hiyam H. Abureesh⁹,
Fatimah H. Asiri⁹, Mohammad S. Aldosari⁹ and Meshal H.K. Almalki^{1,2}

¹Department of Biology, Faculty of Applied Science, Umm Al-Qura University, Makkah, Saudi Arabia.

²Research Laboratories Unit, Faculty of Applied Science, Umm Al-Qura University, Makkah, Saudi Arabia.

³Department of Agricultural Microbiology, Faculty of Agriculture, Fayoum University, Fayoum, Egypt.

⁴Department of Chemistry, Faculty of Science, Cairo University, Cairo, Egypt.

⁵Department of Agricultural Microbiology, Faculty of Agricultural Sciences, Aligarh Muslim University, Aligarh - 202 002, India.

⁶Biology Department, College of Sciences, Princess Nourah bint Abdulrahman University, Riyadh, Saudi Arabia.

⁷Department of Biology, College of Science, University of Jeddah, Jeddah, Saudi Arabia.

⁸Department of Biology, Faculty of Science, King Abdulaziz University, Jeddah, Saudi Arabia.

⁹King Abdulaziz Hospital, Makkah, Saudi Arabia.

Abstract

Staphylococcus aureus is a major human pathogen that may cause a wide range of infections and is a frequent cause of soft tissue and bloodstream infections. It is a successful pathogen due to its collective virulence factors and its ability to evade the host immune systems. The review aims to highlight how *S. aureus* destroys and damage the host cells and explains how immune cells can respond to this pathogen. This review may also provide new insights that may be useful for developing new strategy for combating MRSA and its emerging clones such as community-associated methicillin-resistant *S. aureus* (CA-MRSA).

Keywords: *Staphylococcus aureus*, pathogenesis, immune system, virulence, immune evasion

*Correspondence: hhabulreesh@uqu.edu.sa

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INTRODUCTION

Initially described in 1878, staphylococci; Gram-positive cocci that have been implicated in infections involving multiple systems of the human body, including the skin and soft tissue, the skeletal system, the respiratory system, the bloodstream, and more recently, infections involving implanted medical devices^{1,2}. Staphylococci are further classified as being coagulase-positive, primarily identifying *Staphylococcus aureus* (*S. aureus*), or coagulase-negative (CoNS)³. *Staphylococcus epidermidis* (*S. epidermidis*) is the most important and best-studied member of the CoNS group⁴.

S. aureus is facultative, non-spore-forming, and non-motile bacteria that have the ability to grow well on most cultural media. *S. aureus* is a common inhabitant of the human and animal body, particularly the nasal cavity and the skin⁴. Despite being a commensal in human and animals, *S. aureus* is a virulent bacterium that are able to produce number of toxins and enzymes which allow this bacterium to be able to invade human body and cause wide range of infections, some of which may be considered as life-threatening diseases (Table 1) in immunocompromised and immunocompetent individuals^{5,6}. Moreover, some strains of *S. aureus* are resistant to a wide range of antibacterial agents, some of which show multiple resistance (resistant to three or more classes of antibacterial agent), The most clinically important strains are the methicillin-resistant *Staphylococcus aureus* (MRSA) which are implicated in a large number of hospital and community acquired infections. The resistance towards antibacterial agents and at times multiple resistance make the treatment of

these infections complicated and more challenging to achieve⁷. This review aims to provide and updated literature survey on epidemiology, virulence and pathogenicity as well as interaction mechanisms with host immune system by *S. aureus*.

Staphylococcus aureus Epidemiology

S. aureus infections may not be all reported, given the wide variety of community and hospital-acquired of infection, however, the number of reported cases is increasing worldwide, for instance, in the USA around 80461 cases of MRSA infections in 2011, resulting in a 11000 deaths⁶. Reported MRSA infections in Australia increased from 10.3% in 2000 to 16% in six years⁸. In Taiwan, dramatic increase of MRSA infection rose from 9.8% in 2000 to 56% in 2005, similar increases of *S. aureus* infections were also reported in Saudi Arabia, Lebanon, England, India^{6,8,9}. It is important to mention that these figures represent over all infections, the epidemiological data of every type of Tong et al.⁵ has well documented *S. aureus* infections concluding that there is an increase of hospital-acquired infections particularly in the cases of infective endocarditis and infections related to prosthetic devices, moreover, an epidemic of community-associated skin and soft tissue infections controlled by certain virulence factors and resistance to antibiotics particularly β -lactam group⁵. Molecular epidemiology of *S. aureus* infections showed diversity of clones responsible for infections in every content, with the existence of predominant clones such as ST-80 which was found in North and South America and Europe. In Asia, high heterogeneity in terms of the clones associated with infection was highlighted.

Table 1. Human infections caused by *Staphylococcus aureus*

Infection site	Clinical syndrome
Skin and soft tissues	Boils, abscesses, impetigo, wound infection, scalded skin syndrome, necrotizing fasciitis, cellulitis
Bone	Osteomyelitis
Joints	Septic arthritis
Blood	Bacteremia, Toxic shock syndrome, septic thrombophlebitis
Lung	Pneumonia
Brain	Brain abscess, meningitis
Heart	Endocarditis
Urinary tract	Urinary tract infection
Intestine	Food poisoning

In general, it was suggested that low restrictions in the sales of antibiotics and/or prescribing antibiotics with proper laboratory analysis in some countries aided the increase of *S. aureus* infections as well the diversity of clones responsible for these infections⁸.

***Staphylococcus aureus* virulence factors**

S. aureus infections occur when the mucosal or skin barriers are breached, following the successful invasion of the bacterium into the host tissues and its ability to escape the defensive barriers of the immune system to enter the bloodstream. *S. aureus* have an arsenal of virulence factors that play significant roles in the wide variety of infections and diseases in humans including animals. These factors can also support and provide protection for *S. aureus* to evade the host immune system recognition and their actions. One of the most important virulence factors is surface proteins which promote and encourage the binding and the attachment processes of this bacterium to the host cells surfaces. To avoid recognition from the host immune cells, the surface proteins combined with the blood proteins that lead to aid the bacterium to survive and cause damage to host tissues¹⁰. Another vital factor is protein A, which is located in the cell wall of *S. aureus* which accurately anchored to

the peptidoglycan pentaglycine bridges of the bacteria. It is also known as an IgG-binding protein which combines with the Fragment crystallisable (Fc) region of the antibody in order to cover the surface of the bacterium with IgG antibody to make the recognition of this organism by the immune cells difficult and hard to detect^{11,12}.

The clumping factor A (ClfA) is another *S. aureus* virulence factor that is expressed by the surface of the bacteria cells. Also, it is known as fibrinogen binding protein that promotes the clotting process of blood cells and the damage process of tissues¹³. In addition, the polysaccharide capsule is considered as one of the most essential virulence factor which can contribute to assist the *S. aureus* surviving within the host cells by inhibiting the phagocytosis process by macrophage and dendritic cells¹⁰, moreover, *S. aureus* cells are capable of secreting several important toxins and enzymes such as; coagulase; DNAase; leukocidin; hemolysins; exfoliative toxin in order to promote bacterial penetration and help the bacterium to evade in to the host tissues (Table 2)¹⁰.

Moreover, *S. aureus* can generate another harmful type of toxins such as Panton-Valentine leukocidin (PVL) which cause pneumonia in children and Toxic Shock Syndrome Toxin-1 (TSST-1) which is associated with some cases of

Table 2. *Staphylococcus aureus* virulence factors

Factor	Effect / function
Cell surface proteins	Helping attachment to host tissues
Polysaccharide microcapsule	Resist phagocytosis
Protein A	Limits the host immune response
Panton-Valentine leukocidin (PVL)	Forming porins in cell membrane of host cells, resulting in cell death by leakage of its contents
Alpha hemolysin	Red blood cells lysis
Chemotaxis inhibitory proteins	Inhibit the chemotaxis of neutrophil and monocytes
Extracellular adherence protein	Play a role in adherence and invasion of host tissues and also has immune-modulatory activity
Proteases, lipases, nucleases, hyaluronatylase, phospholipase C, elastase and Staphylokinase	These enzymes contribute to host tissue destruction that facilitate the invasion and penetration of the bacterium into host tissues
Toxic shock syndrome toxin-1 (TSST-1)	Causes toxic shock syndrome particularly in menstrual women
Exfoliative toxin	Causes scalded skin syndrome particularly affecting infants
Enterotoxins	Several types of enterotoxins (SEA, SEB, SEC, SED, SEG, SEH & SEI) causes food poisoning
Clumping factor	Clots blood plasma

Adapted from^{10, 11, 12, 13}

septicaemia due to the use of particular types of tampons (Table 2)^{13,10}. A number of *S. aureus* strains have the ability to produce a pigment that known as staphyloxanthin which acts as an important virulence factor. This strain has an antioxidant role against reactive oxygen species used by the host immune cells, in order to help the bacterium to escape from killing action of immune cells¹⁴. Furthermore, *S. aureus* are capable of forming biofilms of various surfaces which associated generally with most indwelling medical devices problems such as heart valves and knee replacements and via this biofilm community resistance can be acquired to antibacterial agents through horizontal gene transfer¹².

How Does *S. aureus* subvert immune responses?

When *S. aureus* invade the host tissues, the innate immune system responds rapidly as an early defense against the bacterial invasion. This system consists of three important parts; (i) the complement system, (ii) phagocytes and (iii) antimicrobial peptides. It also play many significant roles against the pathogen for instance; it summons the immune cells to the infection sites by producing chemical signaling molecules called cytokines, activates the complement system to recognize the microbe to encourage the clearance process of dead cells, and it activates the second line of the host defense which is known as the adaptive (specific) immune system through antigen presentation process, in order to help the body to eliminate the microbe and to generate long lasting immunity against it to prevent any challenge in the future by the same organism^{15,16}.

The Complement System

The complement system is a group of

proteins and proteolytic molecules that are found and circulate in the blood (Table 3). With *S. aureus*, complement employs some significant molecules to: (1) mark the microbe with C3b and iC3b to make its parts ready for phagocytosis through important immune cells such as; neutrophil and macrophage, and also to (2) seize the attention of phagocytes by small chemoattractant molecules such as: C3a and C5a which are produced during the activation of complement to facilitate phagocytosis as well^{15,17}.

This system is initiated and activated by three significant pathways: (a) the classical, (b) the alternative and (c) and the lectin pathways (Fig. 1). All the three pathways come together at the formation of a surface bound biomolecular enzyme known as C3 convertases. This enzyme stimulates and enhances the activation of complement system by cleaving the complement protein C3 to form C3a and C3b which are required to promote and facilitate any additional essential activation events such as; opsonization and phagocytosis¹⁵⁻¹⁷.

S. aureus Complement avoidance mechanisms

It has been shown that *S. aureus* developed different mechanisms to avoid the action of the complement system by producing a number of proteins that can change and affect the stages of the complement cascade, these mechanisms include; preventing complement identification; cleavage of complement proteins; and/or inhibit the interaction of complement receptors on phagocytes^{18,19}.

Although, the classical pathway C1 complex has the ability to recognize the microbe bound IgG and IgM antibodies and the lectin pathway (mannose-binding lectins and ficolins) bind the saccharide elements of microbe, *S.*

Table 3. Functional protein classes in the complement system

Types of protein classes	Examples
Binding to antigen: antibody complexes and pathogen surfaces	C1q
Binding to mannose on bacteria	MBL
Activating enzymes	C1r, C1s, C2b, Bb, D, MASP-1 and MASP-2
Membrane-binding proteins and opsonins	C4b and C3b
Peptide mediators of inflammation	C5a, C3a and C4a
Membrane-attack proteins	C5b, C6, C7, C8 and C9
Complement receptors	CR1, CR2, CR3, CR4 and C1qR
Complement-regulatory proteins	C1INH, C4bp, CR1 and MCP

Adapted from²⁵

aureus produce two surface proteins that can damage and harm the IgG function (Fig 2.a), the first protein is the Staphylococcal protein A (SpA) and the second protein is known as the Staphylococcal immunoglobulin binding protein (Sbi). Staphylococcal protein (SpA) is a protein

located on the surface of the bacterium that consists of four or five immunoglobulin binding domains. Every domain is able to combine with the Fc parts of IgG antibody, thus inhibiting the interaction with Fc receptors on neutrophils in vitro¹⁷⁻¹⁹.

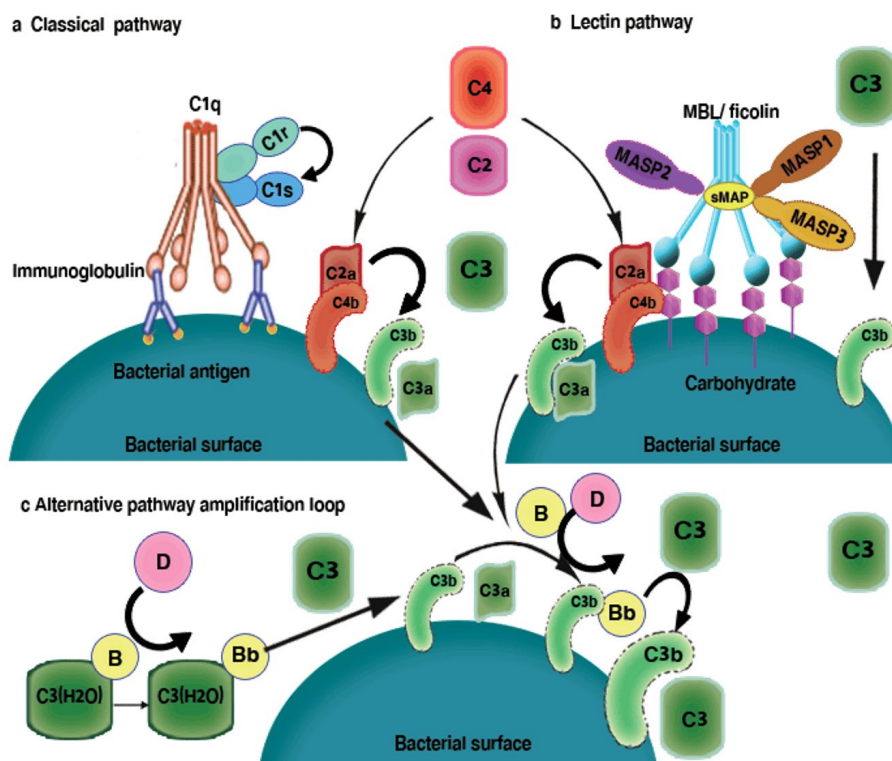


Fig. 1. Complement activation pathway: (A) The classical pathway is initiated by the binding of the C1 complex to antibodies that are bound to antigens on the surface of bacteria. The C1 complex consists of C1q and two molecules each of C1r and C1s. The binding of the recognition subcomponent C1q to the Fc portion of immunoglobulins results in autoactivation of the serine protease C1r. Then C1r cleaves and activates C1s, which translates the activation of the C1 complex into complement activation through the cleavage of C4 and C2 to form C4bC2a enzyme complex. C4bC2a acts as a C3 convertase and cleaves C3 which results in products that bind to, and cause the destruction of, invading bacteria.

(B) The lectin pathway is initiated by the binding of either mannose binding lectin (MBL) or ficolin -associated with MBL-associated serine protease1 (MASP2), MASP2, MASP3 and small MBL-associated protein (sMAP) – to an array of carbohydrate groups on the surface of a bacteria cell. Similar to C1s, MASP2 is responsible for the activation of C4 and C2, which leads to the generation of the same C3 convertase (C4bC2a). As in the classical pathway, C3 convertase cleaves C3 to C3b and the chemo attractant peptide C3a. The C3b-C2a-C4b complex then cleaves C5 to C5a and the chemo attractant peptide C5b, which stimulates assembly of factors C6, C7, C8 and C9. MASP1 is able to cleave C3 directly.

(C) The alternative pathway is initiated by the low grade activation of the C3 by hydrolysed C3 (C3(H₂O)) and activated factor B (Bb). The activated C3b binds factor B (B), which is then cleaved into Bb by factor D (D) to form the alternative pathway C3 convertase, C3bBb. Once C3b is attached to the cell surface, the amplification loop consisting of the alternative pathway components is activated, and the C3 convertase enzyme cleaves many molecules of C3 to C3b, which bind covalently around the site of complement activation [15, 17, 48, 49].

Staphylococcal Sbi is made of four parts of Sbi-I and Sbi-II which are also able to bind with IgG. Besides blocking Fc-receptor-mediated phagocytosis, Sbi has been shown to play some roles in blocking the binding of C1q and subsequent activation of the complement classical pathway¹⁸. Furthermore, *S. aureus* uses another approach to evade the recognition of the complement system by removing and degrading the opsonic molecules (the molecules that are able to bind with both antigen and receptors of phagocytic cells like C3b component of complement system) from its surface through proteolytic process. In addition, *S. aureus* produce a staphylokinase (SAK) which

is an anti-opsonic protein, in order to stimulate and encourage surface bound plasminogen into plasmin, which make the bacteria able to invade and infect the host tissues¹⁷. In fact, the presence of the C3 convertases is significant for the activation of complement system and for the response of host immune cells. *S. aureus* uses three ways to affect and change this vital step in the complement cascade (Fig. 2.b)^{17,18}.

The Cleavage of C3 convertase

The *S. aureus* clumping factor A (ClfA) is a surface protein which can join the human C3b protease factor I (fI), thereby enhancing cleavage of surface-bound C3b into iC3b *in vitro*²⁰.

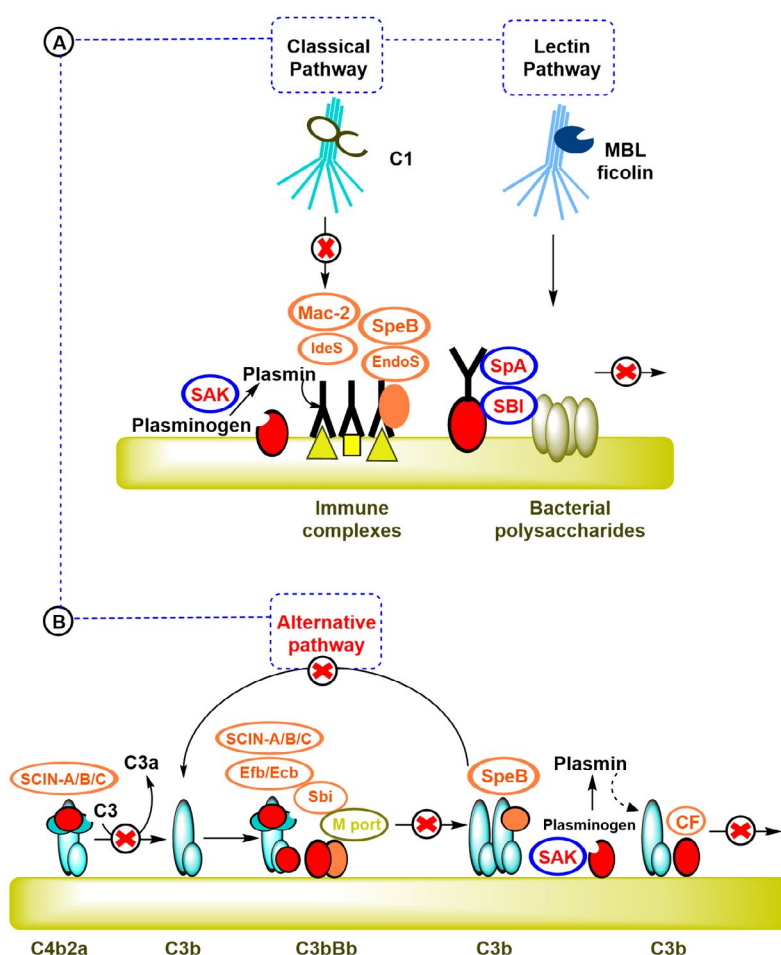


Fig. 2. *Staphylococcus* evasion of complement recognition: (A)- *S. aureus* produce two surface proteins that can damage and harm IgG function; Staphylococcal protein A (SpA) and Staphylococcal immunoglobulin binding protein (Sbi). (B)- Presence of the C3 convertases is significant for the activation of complement system and for the response of host immune cells. *S. aureus* use three ways to affect and change this vital step in the complement cascade^[17, 18, 19].

Direct inactivation of C3 convertases

Convertases are the major complement target among *S. aureus* immune evasion strategies.

S. aureus produces five unlike molecules which work directly to prevent the action of these important enzyme mixtures. Staphylococcal complement inhibitor (SCIN) and its homologues

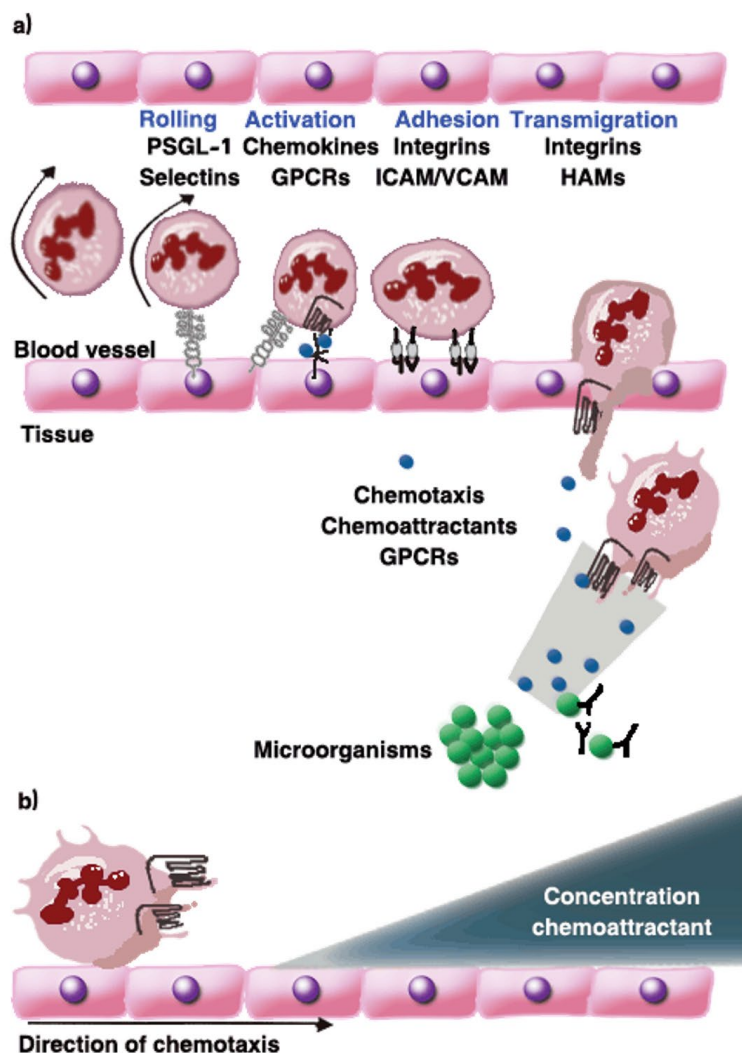


Fig. 3. Phagocyte extravasation: (a) for phagocytes to reach infection sites, they need to extravasate from the blood vessels into the tissue. The successful recruitment of circulating immune cells depends on the productive interaction of leukocytes with the endothelial cells lining the vessel wall and it is a multistep process. Phagocytes tether and roll on activated endothelium through transient interactions of PSGL-1 and selectins. Following stimulation of the cells by endothelium bound chemokines, integrins are activated that mediate the firm cellular adhesion to intercellular adhesion molecule-1 (ICAM-1) and vascular adhesion molecule-1 (VCAM-1) presented by the vessel wall. The phagocytes then transmigrate through the endothelium into the underlying tissue. The cells are directed to the site of infection through the sensing of chemoattractant gradients by GPCRs. Here, chemokines and bacteria and complement derived products are important. The pathogen is cleared by phagocytosis upon recognition of the bacterium opsonized by antibodies and/or complement components. (b) At the infection site, infected cells and the surrounding tissues release chemoattractants. These secreted chemoattractants form a concentration gradient and attract leukocytes, which move through the gradient towards the higher concentrations, a process called chemotaxis. Leukocytes move through the gradient toward the higher concentrations, a process called chemotaxis ^[25, 26, 17].

SCIN-B and SCIN-C are extremely efficient C3 convertase inhibitors which inhibit the alteration of C3, later phagocytosis and C5a formation *in vitro* at low concentrations¹⁸.

Besides that, the alternative pathway C3 convertase made of a vital cofactor called C3b which connected to the protease subunit (Bb) loosely¹⁸. The action of SCIN on the classical pathway convertase remains to be resolved but seems to be caused by a stabilizing mechanism as well²¹. In addition, the alternative pathway convertase can be changed via the extracellular fibrinogen-binding protein (Efb) and the extracellular complement-binding protein (Ecb) by binding the convertase to the C3b particle directly²⁰. The crystal structures of both molecules in complex with the C3d domain of C3 have revealed their exact binding sites²¹.

Modulating human convertase regulators

In order to protect from unnecessary action of the complement system, humans produce complement regulators which reduce and decrease the activity of the convertase²². On the other hand, a large number of microbes generate molecules which interfere with the function of these regulators. In addition to its two IgG binding fragments, Sbi-III and IV that can also bind to the C3, the staphylococcal IgG-binding molecule Sbi plays several important roles in the alteration of the complement²³. Moreover, Sbi has the ability to bind the human complement regulators such as; factor H (FH) and factor H-related proteins in order to make a constant tripartite compound with FH and C3. In general, these actions lead to inhibit the activity of the alternative pathway *in vitro*²⁴.

Phagocytosis

Phagocytosis is a common mechanism that involved in the immune response to eliminate pathogens or foreign particle may be a toxin invading the body. It is activated through the binding to pathogen-associated molecular patterns (PAMPS), which leads to the activation of nuclear factor kappa B (NF- κ B) that is important to control and regulate the response of the immune system²⁵. Furthermore, there are types of the immune cells that have the ability to engulf microbial pathogens in order to remove them from the host, these cells include; neutrophils, macrophages and dendritic cells²⁶. Moreover, complement system plays an important role in phagocytosis by facilitating the uptake of the pathogen by phagocyte cells. This process occurs through specific complement receptors (CRs) such as; CR1 and CR2 (Table 4), these receptors bind the microbe to make it ready for phagocytosis^{25,26}.

In addition, at the early stage of the immune response to the site of infection, the pathogen and its fragments are engulfed by antigen-presenting cells and other immune cells such as macrophages and neutrophils and transferred to the lymph nodes. This leads to the activation of the B cells to discriminate and to produce antibodies in order to reduce the effects of the bacterial toxins and to encourage and promote the phagocytosis of the pathogen²⁶. Furthermore, following the entry of *S. aureus* to the host tissue, the response of neutrophils and macrophages initiate which is critical and essential to promote and assist the body in

Table 4. Types and functions of complement receptors

Receptor	Specificity	Function	Target cells
CR1 (CD35)	C3b, C4b, iC3b	Promotes C3b and C4b decay Stimulates phagocytosis Erythrocyte transport of immune complexes	Erythrocyte, macrophage, monocytes, leukocytes, B cells
CR2 (CD21)	iC3b	Part of B-cell co-receptor	B cells
CR3 (Mac-1)	iC3b	Stimulates phagocytosis	Macrophages, monocytes, leukocytes
(CD11b/CD18)			
CR4 (gp150,95)	iC3b	Stimulates phagocytosis	Macrophages, monocytes, leukocytes, dendritic cells
(CD11c/CD18)			
C5a receptor	C5a	Binding of C5a activates G protein	Endothelial cells, mast cells, phagocytes
C3a receptor	C3a	Binding of C3a activates G protein	Endothelial cells, mast cells, phagocytes

Adapted from ²⁵

removing and eliminating the pathogens during the phagocytosis²¹. (Fig. 3.a).

These cells are called via an important chemoattractants such as C5a; leukotriene B₄; and chemokines such as CXCL8 which also known as interleukin-8 (IL-8) in order to direct their actions to the site of infection²⁷. This chemoattractants

formed through activated host cells and some of them produced as an activated component of the complement system (C5a) at the time of identification of a preserved structure present on the bacterium²⁸. All these chemoattractants trigger phagocytes by attaching to membrane bound receptors called G protein-coupled

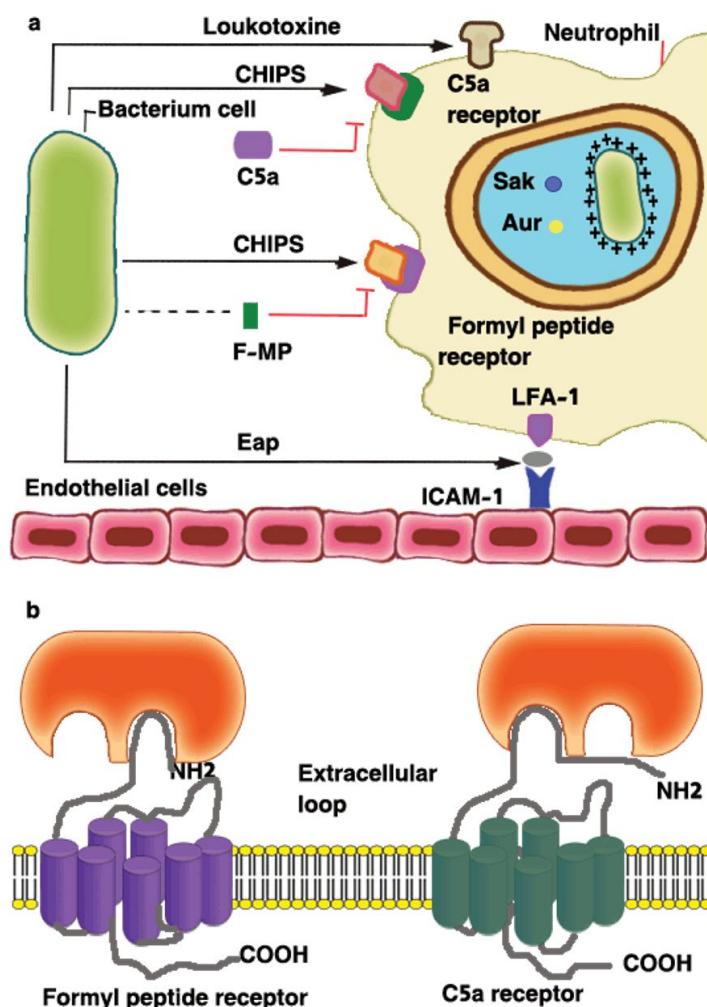


Fig. 4. Inhibition of the neutrophil response to infection: (a) the chemotaxis inhibitory protein of staphylococci (CHIPS) and the extracellular adherence protein (Eap) interfere with neutrophil chemotaxis and extravasation. Resistance to killing by antimicrobial peptides in the neutrophil phagosome is promoted by D-alanine and L-lysine modifications to cell-wall components (indicated by +), by secretion of staphylokinase (Sak) and aureolysin (Aur), and by the creation of 'spacious' phagosomes in which bacteria can survive. The pore-forming leukotoxins are shown by the mushroom-shaped insertion in the neutrophil membrane. (b) Model for interactions between CHIPS and the formyl peptide receptor (FPR) and C5a receptor. Two distinct but closely linked binding domains in CHIPS are indicated, one for the extreme N terminus of FPR involving residues F1 and F3, the second for a domain located between residues 10–20 of the C5a receptor. FMP, N formyl-methionyl peptide; ICAM-1, intercellular adhesion molecule-1; LFA-1, lymphocyte function associated antigen ^[19, 30, 48].

receptors (GPCRs). Both chemoattractants and GPCRs have an essential role which is directing the innate defence cells against the invading microbe (Fig. 3.b). As a result *S. aureus* has a wide range of strategies to evade and avoid phagocytic activity^{28,29}.

***S. aureus* phagocytosis avoidance mechanisms**

Inhibition of neutrophil chemotaxis

A large number of *S. aureus* strains (about 60%) secrete a chemotaxis inhibitory protein (CHIPS) which can combine with the C5a receptor (C5aR) and the formyl-peptide receptor (FPR) in order to block the binding of any related agonist (Fig. 4), the C5aR- and FPR-binding activities of CHIPS were separated by specific amino-acid substitutions and the specificity of blocking monoclonal antibodies^{19,30}. The intercellular adhesion molecule-1 (ICAM-1) on the endothelial cell surface is one of the several ligands recognized by the extracellular adherence protein Eap (or else called the main histocompatibility class II analogue protein Map)³⁰.

Resistance to phagocytosis

S. aureus produces many significant factors to avoid and evade from phagocytosis. It expresses anti-opsonic proteins bound to the surface and a capsule of polysaccharides which: (a) interfere with antibodies and with the formation of complement through classical and alternative pathways, (b) inhibit their interaction to neutrophil complement receptor and Fc receptor. Therefore, efficient phagocytosis by neutrophils that requires recognition of bound complement proteins and antibody is compromised²⁵. Moreover, *S. aureus* has some mechanisms which help and assist the organism to evade the killing of phagocytic cells, including interference with endosome fusion and release of antimicrobial substances by factors that are dependent on the global regulator SarA³¹.

Protein A

Is a surface protein that consists of four or five domains which can bind to the Fc region of IgG antibody. Protein A interact with IgG in order to cover the cell surface with IgG molecules which

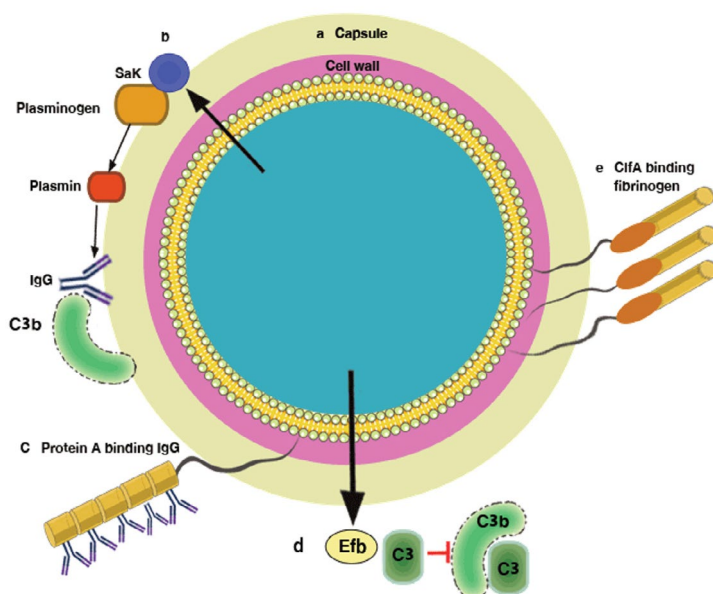


Fig. 5. Mechanisms by which *Staphylococcus aureus* avoids opsonophagocytosis: (a) the capsular polysaccharide, which can compromise neutrophil access to bound complement and antibody; (b) the extracellular staphylokinase (Sak), which activates cell-bound plasminogen and cleaves IgG and C3b; (c) protein A with 5 IgG Fc-binding domains; (d) fibrinogen-binding protein (Efb), which binds complement factor C3 and blocks its deposition on the bacterial cell surface. Complement activation beyond C3b attachment is prevented, thereby inhibiting opsonization. (e) ClfA, which binds the γ chain of fibrinogen ^[19, 21, 30, 33, 34].

are in the incorrect orientation easy to recognize by the Fc receptor of neutrophil (Fig. 5). This action can clarify the anti phagocytic role of protein A and its effects on *S. aureus* infections^{32,33}.

Clumping factor A (ClfA)

Is the dominant fibrinogen-binding protein present on the surface of *S. aureus* cells in the stationary phase of growth. In the murine model the ClfA is a virulence factor for sepsis and arthritis³⁴. Virulence was thought to increase during the bacteremic stage of the infection as well as during the growth of infected joints, since bacterial cells were coated with fibrinogen (Fig. 5), which in turn inhibited deposition of, or accessibility to opsonins^{30,34}. This notion is supported by the observation that ClfA protects *S. aureus* from phagocytosis by murine macrophages⁴⁶ and by

human neutrophils and that defense is at least partly dependent on fibrinogen^{21,34}. Furthermore, ClfB and fibronectin-binding proteins can also bind fibrinogen, to shield the bacterium through the exponential phase of growth, when the expression of such proteins is larger than ClfA³⁴.

Capsule

Is an important factor which expresses by *S. aureus* to inhibit the process of phagocytosis. The expression of type 5 and type 8 capsules is associated with increase virulence in animal infection models³⁵. It has been considered that the presence of the capsule lead to reduce the ability of neutrophils to uptake the pathogen cells in the presence of normal serum opsonins (Fig. 5), meaning that capsule act as anti-opsonic¹⁹.

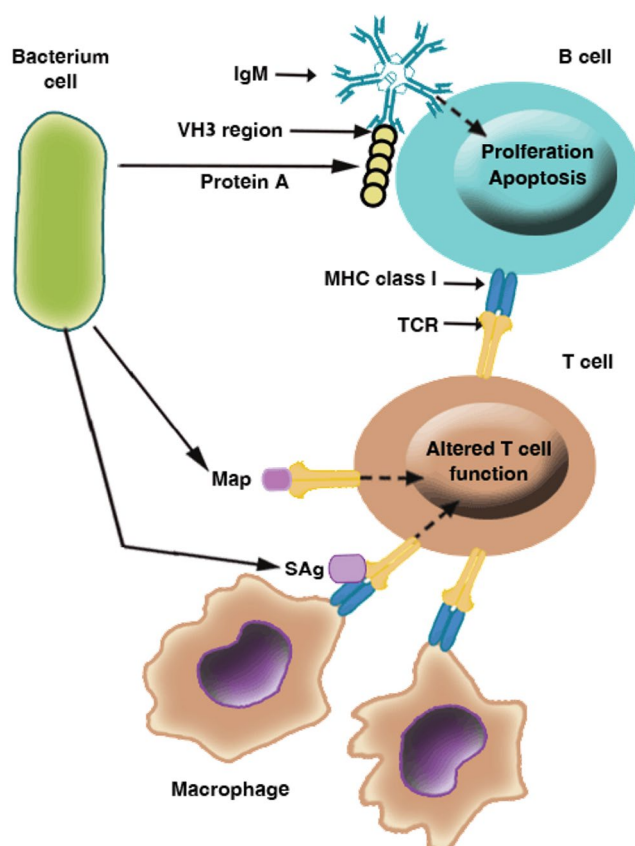


Fig. 6. Mechanisms of immunosuppression mediated by *Staphylococcus aureus*: This figure illustrates examples of immunomodulatory molecules used by *S. aureus* to alter the host immune response, including the superantigens (sAgs) enterotoxins and toxic shock syndrome toxin-1 that bind the MHC class II receptor to T-cell receptors; protein A, which binds immunoglobulin M (IgM) VH3 on B cells; and the MHC class II analogue protein Map, which binds the T-cell receptor (TCR) [36, 38, 39, 41].

Toxins that kill leukocytes

S. aureus is a common pathogen that has the ability to secrete and produce toxins which contribute to the damage of host cells membranes. The expression of cytolytic toxins that damage leukocytes contributes to development of abscesses by the killing of neutrophils that are attempting to engulf and kill the bacteria³⁶. Cytolytic toxins forming β -barrel pores in target cell cytoplasmic membranes cause leakage, and eventually lysis. The representative of this class is

the α -toxin, which is secreted as a monomer paired with a heptamer in the membrane, with β -strands assembled in a 14-stranded β -barrel pore from each monomer^{36,37}.

The two-component leukotoxins consist of two subunits, which are separately secreted and configured into hexameric or heptameric oligomers with a great leukocyte affinity. There are 4 different types of bicomponent leukotoxin, γ -toxin (Hlg), Pantone–Valentine leukocidin (PVL), leukocidin E / D, and leukocidin close to M / F-PV.

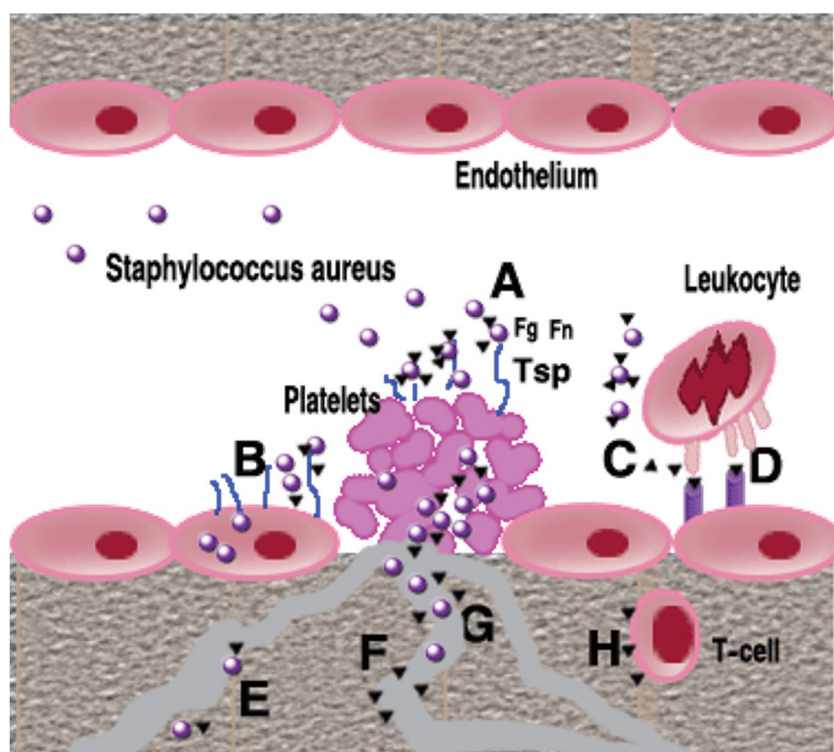


Fig. 7. Role of SERAM in endovascular *Staphylococcus aureus* infection: During wound infection, *S. aureus* organisms secreting and/or decorated by secretable extended repertoire adhesion molecules SERAM) (black triangles) gain access not only to the intact vessel wall, but also to subendothelial matrix and activated platelets and will form a bacteria-entrapped platelet clot (A). SERAM bind to various extracellular matrix components including fibrin (ogen) (Fg), fibronectin (Fn) or thrombospondin (Tsp) that are present in this vegetation and thereby augment bacterial attachment through molecular bridging mechanisms (A). In addition, SERAM may facilitate bacterial binding to Fn expressed on endothelial cells resulting in enhanced uptake of microorganisms (B). Through interaction with complement factor C3, the extracellular fibrinogen binding molecule (Efb) may interfere with opsono-phagocytosis (C). Among other interactions with host factors, the extracellular adhesion protein (Eap) binds to ICAM-1 and other ligands on endothelial cells thereby inhibiting leukocyte adhesion and preventing their extravasation (D). After passing through the endothelial cell layer and/ or the extracellular matrix stroma, *S. aureus* may gain access to other capillaries, arterioles and venules, attach to the vessel wall and start disseminating from focal sites of infection (E) Eap may inhibit vascular cell proliferation and angiogenesis possibly via direct interference with agonist-stimulated endothelial functions (F), while Efb interacting both, with platelets and with fibrinogen, may interfere with fibrin formation resulting in altered wound healing (G). Finally, Eap interferes with T-cells function shifting towards a Th2-cell response and reducing delayed- type hypersensitivity reactions^[42, 43].

The γ -toxin lyses both erythrocytes and leukocytes, whereas PVL is toxic only for leukocytes^{36,38}.

Other evasion mechanism of *S. aureus* Immunomodulatory molecules

Protein A

In addition to its immunoglobulin binding ability, Protein A is an important immunomodulatory molecule owing to its capacity to combine with the VH3 region that is next to the antigen-binding domain of IgM molecules located on the B lymphocyte surface (Fig. 6). Those cells carrying VH3 IgM are induced to proliferate and undergo apoptosis, resulting in depletion of a large proportion of potential antibody-secreting B cells in the spleen and bone marrow^{39,40}.

Enterotoxins and TSST-1

S. aureus produces toxins which work as superantigens such as toxic shock syndrome toxin-1 (TSST-1), the expression of these antigens in the host cell prevents the development of the immune response such as activation of B and T cells or formation of antibodies (Fig. 6)^{36,38}. Furthermore, Antigen-specific T cells are unable to reproduce in response to antigens that are normally presented by MHC class II due to a phenomenon called anergy⁴¹. Therefore, immune-suppression occurred due to the failure of the antibody response. This seems to be important to prevent the production of antibodies to superantigenic toxins themselves^{36,38,41}.

Secretable expanded repertoire adhesive molecules (SERAM)

S. aureus generates some bacterial proteins that anchored in the cell wall which has a significant role in mediating bacterial adherence to host cells and to the components of the extracellular matrix (ECM). The general roles of SERAM (Fig. 7) can be described as: (1) to facilitate bacterial adhesion to host molecules, cells, or tissues, (2) to interact with a broad array of host ligands, thereby sharing diverse activities in that they typically interfere with host defense mechanisms^{42,43}.

Other proteins involved in immune-evasions mechanisms

Furthermore, some studies have shown new functions of some bacterial proteins such as extracellular matrix binding protein (Emp) and extracellular adherence protein (Eap) which play significant roles in *S. aureus* pathogenesis due to its

ability to bind the host components or block some steps in wound healing process. Eap also known as a MHC class II analogous protein. It has a very wide spectrum of binding interactions to components of the host. However, some strains of *S. aureus* lose the ability to express Eap therefore these strains cannot colonize or invade host tissues^{44,45}. This protein can also combine with endothelial ICAM-1 in order to inhibit the interaction between the ICAM-1 and its integrins Mac-1 and LFA-1. Thus, Eap completely destroyed the adhesion systems of respective functional leukocyte, i.e. strong adhesion and endothelial transmigration⁴². In addition to Eap anti-adhesive and anti-migratory function, it has been considered that Eap has an anti-inflammatory role by inhibiting neutrophils and T cells to get to the site of infections to prevent their effects against the bacterium. It also has been described that Eap has an immunomodulatory activity by increasing the synthesis of interleukin (IL)-4 syntheses which is vital for the differentiation of T-cells into Th2-cells in the response of the immune cells in order to down regulate and reduce the response of T-cells to facilitate the intracellular survival of *S. aureus*⁴⁶⁻⁴⁹.

CONCLUSION

It seems clear that *S. aureus* is a widespread organism that lives as a normal flora in the nose and on the skin of humans and animals. It is a very successful pathogen because of its ability to express and produce virulence factors to evade the recognition and the killing of the host immune cells. The bacterium infection starts through its entry to the host tissue which leads to stimulate and induce the innate immune response. The activation of this system occurred via specific pathways which lead to activate other important cells such as neutrophils and macrophages in order to eliminate and remove the pathogen from the host. The function of these cells initiated by the complement system which plays a significant role by producing chemoattractant (chemotactic factors, chemokines, and complement factors etc.) in order to label and opsonise the pathogen for phagocytosis.

However, it has been shown that *S. aureus* improved many ways to avoid the effects of phagocyte cells by producing some proteins such as SpA and Sbi to inhibit and block the recognition

of complement and their receptors interaction on phagocytes. Also, *S. aureus* can inhibit neutrophil chemotaxis by producing CHIPS that can interfere with some important receptors such as C5a receptor protein to prevent the binding of any related agonist. This pathogen can also produce super antigens toxins to make the response of the immune system difficult to achieve and hard to occur. Furthermore, it has been described that *S. aureus* can kill the leukocytes directly by secreting important cytolytic toxins such as PVL and leukocidin E/D. Moreover, SERAM, Eap and Emp are ubiquitous proteins that can help and assist the bacteria to survive and cause infections.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

AUTHORS' CONTRIBUTION

All authors have made substantial, direct, and intellectual contribution to the work and approve it for publication.

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DATA AVAILABILITY

All datasets generated or analyzed during this study are included in this manuscript

ETHICS STATEMENT

This article does not contain any studies with human participants, or animals performed by any of the authors.

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