ISSN NO: 2770-0003
Date of Publication: 24-08-2022

Immunoregulatory activity of Staphylokinase produced by Staphylococcus aureus

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Abstract*: Staphylococcus aureus* is one of the world's most frequent pathogens that responsible for a wide range of infectious diseases. As plasminogen is converted into the active fibrinolytic enzyme, plasmin by staphylokinase, a staphylococcal protein Staphylokinase is also produced by S. aureus bacteria that infect people. Antibiotic-resistant bacteria are blocked from invading tissues by Sak achieve's capacity to form complexes of plasminogen activating complexes with plasmin as well as with plasmin. As a result of staphylokinase's tiny size, it also acts as an immunomodulator that is represented by an increase in IL-10 blood level (an anti-inflammatory cytokine) and a decrease in IL-4 serum level, which is associated with a reduction in the development of hypersensitivity. Both beneficial and detrimental effects can be achieved by immunomodulation. Immunomodulators abruptly modify informational molecules, like as cytokines, to affect the immune system's activity. If streptokinase is administered regularly, it can cause allergic reactions and the formation of anti-streptokinase antibodies.

Keywords: *Staphylococcus aureus*, staphylokinase, immunomodulation

Staphylococci

Generic name Staphylococcaceae is Staphylococci are gram-positive, spherical organisms of the family. It is possible that Staphylococcus forms clusters that look like grapes and may even form pairs, tetrads, or short chains of various lengths. Their oxidase and catalase enzymes are both negative, and they do not generate spores or form capsules (Khan, 2017).

Bacteria belonging to the staphylococci family can cause a wide range of illnesses, from mild skin infections to bacterial meningitis. In spite of extensive attempts to prevent their distribution, Worldwide, they continues to be a major cause of both hospital and community infections.' *Staphylococcus aureus* and *Staphylococcus epidermidis* are the two most common opportunistic infections in this genus. These bacteria can release free plasma coagulase, unlike Staphylococci that are coagulase-negative (Hadi, 2017).

More than thirty-two varieties of *Staphylococcus* may be found throughout the world, including eight subspecies. These bacteria can thrive in a wide range of conditions, breaking down carbohydrates into their component parts and producing a wide range of colors. Temperatures between 30 and 37 degrees Fahrenheit were optimum for the experiment. An raised, spherical solid media colony may be seen here. hemolysin is produced by certain staphylococci, which is a hallmark of *staphylococci*, and they create a zone of haemolysis on blood agar (Frank, 2017).

Staphylococcus aureus

There are clusters of "grape-like" *Staphylococcus aureus* bacteria, which are gram-positive (stain purple when stained with the Gram stain). Salt-tolerant bacteria may thrive in 10% salt of medium, and golden or yellow colonies frequently appear. It is possible for these organisms to develop either aerobically or anaerobically (facultative) at 18ºC and 40ºC (Taylor and Unakal, 2019).

Toxin-related illnesses and severe life-threatening infections are all possible outcomes of *Staphylococcus aureus* infections (Reddy *et al.*, 2017). According to CDC data, 14.9 and 18.8 percent of all bacterial infections seen in the clinical environment as either an outpatient or inpatient case, respectively, are caused by *Staphylococcus aureus* (Baum et al.,2009; David,2014).

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Among a widespread pathogens was *Staphylococcus aureus* that the most in the globe, responsible for a wide variety of infectious illnesses. Even with effective care, some illnesses may have a death rate of up to 50%. There are three types of infections that can be caused by it: Toxins like food poisoning, scalded skin syndrome, and toxic shock syndrome, as well as more serious illnesses like endocarditis, osteomyelitis, pneumonia, brain abscesses, meningitis, and bacteremia are all examples of systemic infections that can be life-threatening and which are all potentially fatal (Bien *et al.*, 2011; Gnanamani *et al*., 2017).

Staphylokinase

plasminogen is converted into the active fibrinolytic enzyme, plasmin, by an extracellular protein known as Staphylokinase (staphylococcal fibrinolysin or Miiller's factor). Enzymological and clinical research used to name it staphylokinase, but books on staphylococcal classification, biotyping, and epidemiology termed it fibrinolysin. Previously, the name Miiller's factor was used to describe an unknown component that generates several zones of clearing surrounding staphylococci colonies on heated plasma agar plates (Devriese and Kerckhove, 1980).

Staphylokinase, a Staphylococcal protein, was a tiny protein molecule. It is at the late exponential phase of *S. aureus*'s development, By breaking blood clots created at the site of injury, it can penetrate host tissues , that SaK is produced (Nguyen and Vogel, 2016).

Structure of Staphylokinase

One domain protein of staphylokinase is composed of an alpha-helix centrally located, 5-strand alpha-sheet and two shorter alpha-strands, all of which are negatively charged. The enzyme is molecularly weighted at 15.5 kDa and is composed of 136 amino acids. Sak and plasmin in the serum create Sakplasmin, a 1:1 combination. The first 10 N-terminal residues of mature Sak are eliminated in an active Sak-plasmin complex to reveal the charged residue - Lys11. Sak is inactivated when Lys11 is deleted. It has been shown that Sak binding to plasmin guides the active site toward preferring activation loop cleavage in plasminogen and enhances the conversion of activation loops into activation molecules (Tam and Torres,2019).

Mechanism of Action

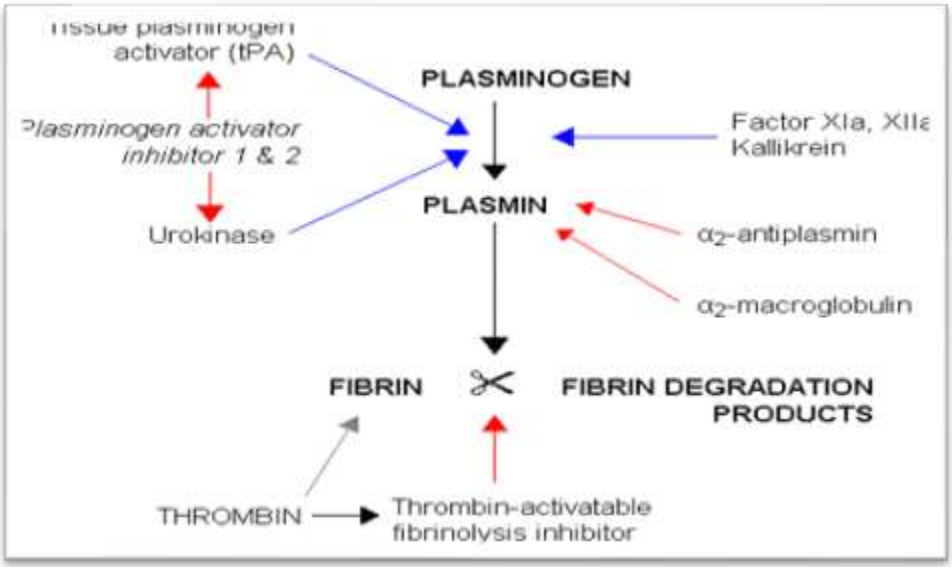
Staphylokinase was one of the thrombolytic drugs, along with tPA and urokinase, which were also used. These substances were used to dissolve clots (Rother et al ., 2013). An enzyme called staphylokinase was used to activate plasminogen. It is possible to summarize these points as illustrated in Figure (1):

1- Inactive enzymatically, the staphylokinase–plasminogen complex is activated by traces of plasmin.

2- Plasminogen is converted into plasmin by activation of the staphylokinases, however the 2-antiplasmin effectively inhibited its activity.

3- SaK is released from the staphylokinase-Plasmin complex when 2-antiplasmin is added to the complex, but its catalytic potential is not affected and it may attach again to any additional plasmin (-ogen) molecules.

4- SaK is largely activated by fibrin-bound plasminogen



Figure(1): fibrinolysis (Banerjee *et al*., 2003)

Staphylokinase and immunity

ISSN NO: 2770-0003
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S. aureus strains that infect humans generate staphylokinase, as well Biofilm development and the fibrinolytic cascade, which helps bacteria invade tissues, are prevented by Sak achieve's ability to form plasminogen activating complexes with plasmin and plasmin itself (Marijke et al., 2014).

Antimicrobial peptide-neutralizing Sak's secondary function (AMPs). All mammals and other animals manufacture AMPs, a diverse group of peptides, as a means of fighting against invading infections (Nguyen and Vogel, 2016).

A peptide called -defensins, which are short -sheet peptides released by neutrophils and prevalent in human, was shown to be able to bind to Staphylokinase. The binding of these peptides to the -defensins appears to raise the plasminogen concentration. Sak's activating characteristics Sak and human neutrophil proteins create a complex, which inhibits each other's bactericidal activity. Consequently (Lehrer and Lu, 2012),

Advantages of Staphylokinase

If plasmin is absent, staphylokinase will not be activated in the plasma milieu. To prevent the development of the SaK–Plasmin (-ogen) complex in the bloodstream, the antiplasmin 2 eliminates plasmin from circulation. If any of the complexes are quickly neutralized by 2 – antiplasmin, the active Staphylokinase molecule is released. As a result of this process, the SaK molecule was reused, which means the dose will be lower. The existence of fibrin clots in the plasma results in activation of plasminogen, which results in plasmin formation (Silence et al ., 1995).

The lysine binding site of these plasmin molecules shielded them against the fast inhibition of 2 – antiplasmin. Thus, Staphylokinase was metabolized in the vicinity of the clot, and its activity continues to be focused in this area. The release of SaK-plasmin complexes from the clot was promptly inhibited. Using this method, staphylokinase is clot-specific and does not generate plasmin in the body (Sumera et al ., 2018).

Streptokinase, on the other hand, was shown to be less effective in breaking up clots that were both platelet-rich and platelet-poor. It was found that Streptokinase was only able to break apart clots that were lacking in platelets. An important clinical consideration for a high platelet content in a coronary thrombus, as well as the retraction and aging of traditional non-fibrin selective treatments, was that this attribute had a substantial clinical impact (Kattula *et al.*, 2018).

Immunomodulation

The immune system is both sophisticated and widespread. There are different cell types that can be found in the bloodstream or confined to a certain organ or tissue. The immune system's involvement-Protecting the host from environmental dangers and infections while also preventing autoimmunity was a marvelously balanced protective strategy for the immune system (Mangino *et al.,* 2017). Immune homeostasis relies on a balance between innate cells (which are pre-programmed to fight infections and malignancies), naïve adaptive B and T lymphocytes (which have antigen receptors that may target any unknown pathogen or neoantigen), and functionally polarized memory B and T lymphocytes(Liang *et al*., 2018).

All processes in which an immune response is manipulated, such as immunopotentiation, immunosuppression, or immunological tolerance, fall under the umbrella term "immunomodulation. Basically, an immunomodulator is a chemical that changes the immune system's responsiveness or activity. Different branches of the immune system are regulated by molecules called immunomodulators, which operate on the pathways that control the immune response by either raising (immunostimulators) or reducing (immunomodulators) (immunosuppressives). Many conditions can benefit from immunostimulators, including infections, malignancies, primary or secondary immunodeficiency and changes in antibody transfer according to Bascones-Martinez and colleagues, 2014).

Additionally, microorganisms were able to alter the immune system's response to their presence, establishing or maintaining an infection. Immunomodulation, on the other hand, may be both useful and harmful. Informational molecules, such as cytokines, are impulsively modulated by immunomodulators to alter the functioning of the immune system (Kapur and Pal, 2018). Streptokinase can produce allergic responses and the production of anti-streptokinase antibodies if it is given repeatedly.

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a- Interleukin-4

Interleukin-4 (IL-4) and interleukin-6 (IL-6) are Inflammatory cytokines that generated by a wide range of cells, including T lymphocytes, basophils, eosinophils and neutrophils. Inflammation and immunological responses are regulated by this protein. I-4 regulates allergic reactions and possesses anticancer and inflammatory properties. B lymphocytes, monocytes, dendritic cells, and fibroblasts are all affected by IL-4's actions, and its expression is upregulated (Reinhart and Kaufmann, 2018).

Th2 cells are formed when naive helper T cells are stimulated to undergo differentiation. It is possible that IL-4 and IL-13 have similar roles since the IL-4 receptor they connect to also binds another cytokine, interleukin 13 (IL-13). Cell culture applications were the planned usage of recombinant human IL-4 (Bao and Reinhardt, 2015). Helper T-cell phenotype and B lymphocyte synthesis of immunoglobulin E (IgE) are only two examples of the many roles of B lymphocytes (Saunders et al., 2019).

b- Interleukin-10

An important cytokine in the host's immunological response to infections, Interleukin-10 (IL-10) has anti-inflammatory capabilities and has an important role in avoiding tissue damage and maintaining normal tissue homeostasis (Brockmann *et al*., 2018). Immune cells such as B lymphocytes produce interleukin 10, a cytokine that can inhibit the functions of T and antigen presenting cells (APC). However, IL-10 promotes B cell functions such as survival, proliferation, differentiation, and antibody production, which is why IL-10 is often used in the treatment of immunodeficiency (Akdis *et al*., 2016).

Anti-inflammatory interleukin-10 (IL-10) has an essential immunoregulatory role and is associated with the development and severity of allergic rhinitis (Wang et al., 2014). While an IL-10 deficiency or abnormal expression might contribute to an increased response to microbial challenge, these conditions could also cause inflammation in the digestive tract as well as the development of autoimmune disorders (Bhurani and Dalai, 2018).

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ISSN NO: 2770-0003
Date of Publication: 24-08-2022

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