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REVIEW ARTICLE

Bacteriolytic Clearance for Zinc Sulfate Induced Peptidoglycan Cell Wall Destruction on Autolytic Cleavage, Hydrolysis and Degradation against *S. Aureus*

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Abstract

Bacterial peptidoglycan (PGN) cell wall clearance on autolysin cleavage, hydrolysis, and degradation by zinc sulfide solution has been investigated on the ground of the results obtained from halo antibacterial susceptibility tests in metal sulfate solutions against *Staphylococcus Epidermidis*, in which the order of antibacterial effect in the metal sulfate solutions is $Zn^{2+} > Cu^{2+} > Ag^{+} > Al^{3+}$ and the highest antibacterial activity is found to be the zinc sulfate ($ZnSO_4$) solution with a strong acid or strong electrolyte.

PGN autolytic cleavage by zinc-containing autolysin amidase; AmiE, Rv3717, AmiA: Zn^{2+} ions in $ZnSO_4$ solution can cleave bacterial PGN murein chains that as PGN cleavage by *Staphylococcal* amidase autolysin, zinc dependent metalloenzyme AmiE is efficient as prevention of the pathogen growth**.** The other, Zn^{2+} binding AMIDASE Rv3717 showed no activity on polymerized PGN and however, it is induced to a potential role of N-Acetylmuramyl L-alanine Amidase. PGN cleavage by zinc-binding autolysin *Staphylococcus aureus* Amidase is involved that AmiA shed light on PGN binding and cleavage**.**

PGN hydrolysis by zinc-containing enzymes: PGN murein hydrolase activity is involved that PGN hydrolysis by Zn^{2+}-containing catalysis is consistent in peptidoglycan recognition proteins (PGRPs), PGN murein hydrolase activity and generalized autolysis. Amidase MurA, LytA amidase, the major murein hydrolase of *S. pneumoniae* are considered an important virulence facto that A glycylglycine endopeptidase LytM and LytB of enzymatically active domain of autolysin are necessary for its optimal activity toward peptidoglycan hydrolysis and for pneumococcal adhesion to respiratory epithelial cells.

PGN degradation by inhibitive PGN elongation: Zinc ions-induced PGN inhibitive elongation occurs due to inhibitory TG enzyme and activated autolysin against *S aureus.* Inhibition of PGN elongation due to the activated Zn^{2+} binding Rv3717 autolysins showed no activity on polymerized PGN and but, it is induced to a potential role of N-Acetylmuramyl L-alanine Amidase. Thus, zinc(II) inhibits PGN biosynthesis, activates PGN autolysin, and inhibits PGN elongation against *S. aureus* cell wall, in which zinc induced PGN autolytic cleavage and hydrolysis by activated autolysins, respectively, of AmiE, Rv3717 and PGRPs, MurA, LytA, LytM-endopeptidase, hydrolase LytB can be promoted, and then causing PGN autolysin cleavage, hydrolysis, degrading, and leading PGN bactericidal dissolution.

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Keywords: Zinc(II), Halo-test, Zinc sulfate, PGN autolysin and elongation, *S. aureus* PGN cleavage, hydrolysis, and degradation.

Abbreviation:

CV= Carvacrol, **CMC**=chitosan metal complex, **COVID**-19=coronavirus disease of 2019, Eps=endopeptidase, **ICU**=intensive care unit, **PGN**=peptidoglycan, PGRPs=peptidoglycan recognition proteins, **PTEN**=403 amino acids, **LYS1**=Lysozyme1, MIC=minimum inhibitory concentration, **MBC**=minimum bactericidal concentration, **PRRs**=pattern recognition receptor, **TG**=Transglycosylase, **TP**=Transpeptidase.

Introduction:

$ZnSO_4 \cdot 7H_2O$ is an important inorganic chemical material with wide application in various industrial fields, such as manufacturing lithopone, preservation of wood and leather, bone glue clarification and artificial fiber precipitation agent. In addition, it is widely used in medicine, electroplating, agriculture, etc. The $ZnSO_4 \cdot 7H_2O$ was traditionally produced through the procedure of sphalerite roasting and leaching with sulfuric acid, in which the role of Zn(II) ions in $ZnSO_4$ solution is very important for antibacterial and antiviral drug agents [1]. A minimum inhibitory concentration of active Zn in $ZnSO_4.7H_2O$ ranging from 0.03 mg/ml to 1 mg/ml was inhibited at a concentration of 0.06 mg/ml to 0.5 mg/ml of Zn, in which zinc has an excellent antibacterial activity against enteric bacterial pathogens [2]. In the metallic sulfate solutions, Al^{3+}, Zn^{2+}, Cu^{2+}, Ag^{+} have higher antibacterial activities. From these observations, Zn^{2+} ions in $ZnSO_4$ solution have indicated the highest antibacterial effect [3]. On the other hand, zinc sulfate to act as an inhibitor of mycelial growth of fungus such as the peach gummosis that was caused by Lasiodiplodia theobromae and significant inhibition of mycelial growth of L. theobromae by zinc sulfate [4]. All concentrations of zinc sulfate showed antibacterial effect that the maximum antibacterial effect of zinc sulfate had shown with (10mg/ml) concentration for *Staph. aureus*, *Staph. epidermidis* and *klebsiella* whereas the maximum antibacterial effect of zinc sulfate had shown with (14mg/ml) concentration for *E. coli*, *Enterobacter*, *proteus* and *pseudomonas* [5]. Thus, these metallic zinc salts possess with the higher antibacterial effects.

Zinc(II) (Zn^{2+}) ions exhibit antimicrobial activity against various bacteria that released Zn^{2+} contributes inhibition of bacterial cell growth and DNA damage which the effectiveness of Zn inhibition of bacterial growth results from changing the active transport system and impeding the initial phase of bacterial mating [6].

The other, bacterial PGN cleavage and hydrolysis plays important roles for anti-bacterial functions that zinc induced bacterial PGN cleavage is composed of decomposition and hydrolysis, in which bacterial killing occurs by PGN cell wall destruction through balanced reaction between PGN suppressive biosynthesis and activated autolysin. PGN cleavage is involved that AmiA distinguishes PGN mostly by the peptide, and cleavage is facilitated by a zinc-activated water molecule [7].

Peptidoglycan (PGN) recognition proteins (PGRPs) are pattern recognition receptors of the innate immune system that bind and, in some cases, hydrolyze bacterial PGN hydrolysis by Zn^{2+}-containing PGRPs [8]. Plants employ hydrolytic activities for the decomposition of complex bacterial structures, in which a plant enzyme activity Lysozyme1(LYS1) that hydrolyzes β (1,4) linkages between N-acetylmuramic acid and N-acetylglucosamine residues in PGN and between N-acetylglucosamine residues in chitooligosaccharides, thus PGN breakdown products produced by LYS1 are immunogenic in plants, and LYS1 mutant genotypes were immunocompromised upon bacterial infection, in which Eukaryotic hosts more generally make concerted use of PGN hydrolytic activities and of pattern recognition receptor (PRRs) in order to cope with bacterial infections [9]. Thus, bacterial PGN cleavage may be consisted of decomposition, hydrolysis, and PGN inhibitive elongation.

In this short-review article, antimicrobial effective activity of metal salts and bacterial PGN cell wall clearance on PGN autolysins, hydrolysis, and degradation by Zn^{2+} ions in zinc sulfate solution are elucidated through zinc(II)-induced PGN suppressive biosynthesis, activated PGN autolysin, and PGN elongation inhibition.

Experimental results for antibacterial halo-test susceptibility against *Staphylococcus Epidermidis*.

Halo antibacterial susceptibility test procedure had been performed that This method is characteristics of finding of inhibitory halo zone measurements as less qualitative antibacterial activity assay. Halo antibacterial tests have been carried out for the zinc sulfate aqueous solutions against *Staphylococcus epidermidis*. The other, the antibacterial reagents were prepared metallic ions 100 mM/L aqueous solutions from metallic salt reagents, wherein the crystalline powders of metallic salts of 0.01mol are dissolved in distilled water of 100 cc, preparing metallic ion concentration of 100 mM/L as antibacterial reagents (crystalline powders of 0.005 mol for zinc, copper, silver, and aluminum sulfates were used) [10].

Figure 1 shows sample appearance of halo inhibitory zone tests, in which from these observations, the antibacterial

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active effect order is Zn^{2+}>Cu^{2+}>Ag^{+} >Al^{3+} for metal sulfate solutions and the highest antibacterial effect has been found to be the zinc sulfate solution, namely, Zn^{2+} ions in sulfate solutions have the highest antibiotics characterization.

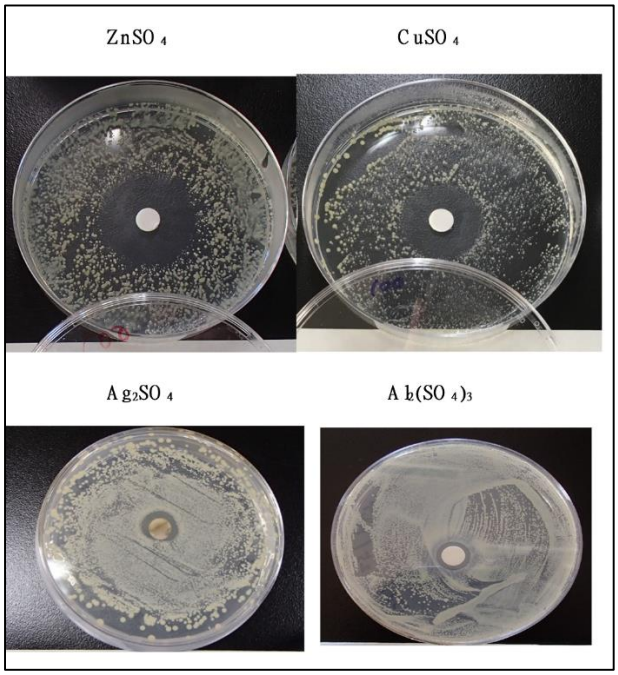

Figure 1: Shows sample appearance of halo inhibitory zone tests.

Figure 2 shows relationship of halo inhibitory zone (in mm) in Aluminum, Zinc, Lead, Copper and Silber Nitrates and Sulfates and Nitrates against *Staphylococcus epidermidis*, in which antibacterial effect order for the metal nitrate solutions is $Cu^{2+} > Zn^{2+} > Ag^{+} > Al^{3+}$ and the highest antibacterial activity is found to be the Copper Nitrate ($Cu(NO_3)_2$) solution.

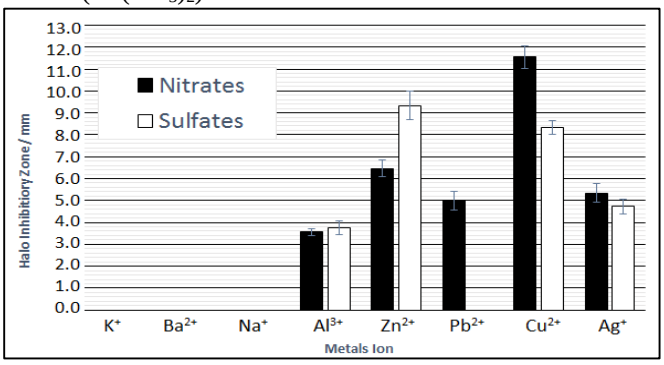

Figure 2: Relationship of halo inhibitory zone (in mm) and some metallic ions of Aluminium, Zinc, Lead, Copper and Silber Sulfates and Nitrates against *Staphylococcus epidermidis*.

The errors of Nitrates ■; Al^{3+}=±4.87%, Zn^{2+}=±5.57%, Pb^{2+}=±8.47%, Cu^{2+}=±4.39%, Ag^{+}=±7.96%,

The errors of Sulfates □; Al^{3+}=±8.29%, Zn^{2+}=±7.08%, Cu^{2+}=±3.85%, Ag^{+}=±7.11%

Discussion:

Characteristics of zinc sulfate ($ZnSO_4$) solution is a strong acid or strong electrolyte that zinc is redox-inert and has only one valence state of Zn (II). In proteins, the coordination is limited by His, Cys, Glu, and sulfur donors from the side chains of a few amino acids. In zinc sulfate solution is dissociated into aqua zinc ion $[Zn(H_2O)_6]^{2+}$ and sulfuric ion $(SO_4)^{2-}$. Aqua zinc ions are liable to be bound to ligand L having negative charge. The sulfuric ion has bactericidal inactivity [11]. Zinc sulfate solution may have an excellent antibacterial activity against bacterial drugs.

1. Antimicrobial effective activity for metal salts; Zn^{2+} antibacterial large effect

For *S. sobrinus*, Zinc Sulfate ($ZnSO_4$) recorded MIC 0.125mg/mL value and MBC 4mg/mL value, in which zinc sulfate can inhibit the growth of *S. mutans* and *S. sobrinus* in vitro and alter the normal cell morphology of these bacteria that zinc has a favourable potential antibacterial effect against cariogenic bacteria, primarily *S. sobrinus* and is a promising candidate for the management of dental caries [12]. Pulmonary bacterial clearance by Zinc Sulfate can be achieved to be Carvacrol (CV) + Zn demonstrated strong synergistic effects with antibiotics and effectively managed *P. aeruginosa* lung infections in mice, in which these findings highlight its potential as an innovative therapy for biofilm-associated infections [13].

Zinc sulphate may play a role in therapeutic management for COVID-19, the coronavirus disease of 2019 (COVID-19) caused by the severe acute respiratory syndrome coronavirus, the U.S. Food and Drug Administration authorized the emergency, admission to the ICU and mortality or transfer to hospice for patients who were never admitted to the intensive care unit (ICU), ICU level of care remained significant (OR 0.449, 95 % CI 0.271–0.744) [14].

Zinc sulphate ($ZnSO_4$ $7H_2O$) exhibited relative less antibacterial activity while zinc gluconate was devoid of antifungal activity whereas the other salts showed antimycotic activity but, it was significantly lower than their respective activities against tested bacterial strains [15].

Antibacterial chemically effectiveness by metal salts has been indicated that Zn^{2+}-treatment possessed a great antibacterial activity against *S. aureus*, Cotton fabric treated with Cu^{2+} possessed the most effective antibacterial that treated with Zn^{2+}, Fe^{2+} and Fe^{3+} possessed activity against Klebsiella pneumoniae a slight antibacterial and activity. These findings suggest that antibacterial with metal salt on cotton fabrics [16]. Zinc sulphate for antibacterial drug is used in oral care products,

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typically at concentrations of 0.02–0.5% calculated on Zn^{2+} content. In addition, it has been established that using Zn salts in such products might inhibit tartar formation. Although the antibacterial activity of regular ZnO has been relatively well investigated, in which ZnO nanoparticles have demonstrated significant antibacterial activity with greater activity against *S. aureus* [17]. Antimicrobial tests chitosan and chitosan metal complex (CMC)-Zn against *E. Coli* were shown that chitosan and CMC-Zn show good antimicrobial result on jute fabric and the possible advantages of these enhanced chitosan derivatives for medical textile [18].

2. Zinc(Ⅱ) inhibits PGN biosynthesis, enhances PGN autolysins, and then inhibits PGN elongation

Figure 3 shows the molecular structure of *S. aureus* PGN cell wall that indicates the action sites of PGN biosynthesis enzymes of TG)/TP and PGN forth autolysins and Lysostaphin enzyme. *Table 1* is represented PGN biosyntheses and forth autolysins against *S. Aureus.*

For the sake of growth of *S. aureus* PGN cell wall, there is necessarily required for the adequate balance between PGN biosynthesis and PGN autolysin. When the balance is broken to be become imbalanced, bacteriolysis and destruction of the cell wall should occur. Hence, it became apparent that PGN cleavage and hydrolysis of *S. aureus* PGN cell wall by Zn^{2+} ions are caused by inhibition of PGN elongation due to inactivation of PGN Transglycosylase (TG) or Transpeptidase (TP) and enhancement of PGN activated autolysin of amidases. By the reaction of Zn^{2+} ions with *S. aureus* surface, zinc-protein complexes are formed on the ground that are due to formation of S-atom containing Zn-cysteine complex in bacteria [19]. Zinc may be shown to inhibit PGN biosynthesis TG that the bactericidal activity of Zn^{2+}-dependent peptidoglycan recognition proteins (PGLYRPs) is salt insensitive and requires N-glycosylation of PGLYRPs, namely, zinc may be shown to inhibit PGN biosynthesis TG, but these limited PGLYRPs don't be applicable for Gram-negative bacteria [20].

Zinc ions can inhibit PGN biosynthesis TG against *S. aureus* that zinc regulates PGN biosynthesis, in which Zn^{2+} ion can inhibit PGN synthetic enzymes that Zn^{2+} ions are most commonly coordinated by cysteine, followed by histidine, aspartate, and glutamate that Zn-cysteine complex in bacteria, and the Zn^{2+} chelation represents a potential therapeutic approach for combating biofilm growth in a wide range of bacterial biofilm-related infections [21].

Wall teichoic acids are spatial regulators of PGN cross-linking biosynthesis TP, however, it is not explicit whether zinc ions could inhibit both TG and TP enzymes of the PGN, wherein due to uncertain relation between wall teichoic acids biosynthesis and PGN biosynthesis [22]. Zinc can inhibit PGN biosynthesis that zinc inhibition of phosphoglucomutase results in decreased capsule biosynthesis and Zinc intoxication also is observed to disrupt or inhibit PGN biosynthesis [23].

Metalation of Zn^{2+} enzymes are activated by Zn^{2+} metalation via Zn^{2+} transporters with that Zn(II) disrupts this coordination, resulting in depression of heme synthesis but continued repression of catalase that Zn(II) intoxication leads to intracellular heme accumulation from measurement of heme content of crude extract of cells treated with zinc concentration 50 μM Zn(II) [24].

Zinc ions-induced bacterial cell wall functions PGN inhibitive synthesis enzymes of TG and TP against *S. aureus*, in which zinc ions inhibit PGN biosynthesis and zinc disrupts PGN biosynthesis in bacterial cell wall [25]. The zinc intoxication on *S. pneumoniae*, observing disruptions in central carbon metabolism, lipid biogenesis, and peptidoglycan biosynthesis. Thus, zinc(II) regulates PGN biosynthesis TG/TP that zinc ion can inhibit PGN synthetic enzymes and zinc intoxication could inhibit PGN biosynthesis TG against *S. aureus.*

3. PGN autolytic cleavage by zinc-containing autolysin amidase; AmiE, Rv3717, AmiA

Zn^{2+} ions in $ZnSO_4$ solution cleave PGN cell wall murein by *Staphylococcal* amidase autolysin [26]. Zinc(II) can cleave bacterial PGN murein that As PGN cleavage by *Staphylococcal* autolysin, zinc dependent metalloenzyme AmiE is efficient as prevention of the pathogen growth [27]. The other, Zn^{2+} binding AMIDASE Rv3717 showed no activity on polymerized PGN and however, it is induced to a potential role of N-Acetylmuramyl L-alanine Amidase [28]. PGN cleavage by zinc-binding autolysin *Staphylococcus aureus* Amidase is involved that the AmiA shed light on PGN binding and cleavage [7].

4. PGN hydrolysis by zinc-containing enzymes; PGRPs, MurA, LytA, LytM=Endopeptidase, Hydrolase LytB

Zinc can hydrolyze bacterial PGN protein that PGN hydrolysis by Zn^{2+}-containing catalytic peptidoglycan recognition proteins (PGRPs) is involved that Zn^{2+}-dependent amidases that hydrolyze PGNs by cleaving the amide bond between MurNAc and L-alanine, in which no zinc ion is present in the PGN-binding site of human PGRP-IαC, which binds but does not hydrolyze PGNs due to substitutions in zinc-coordinating residues [29].

MurA of PGN-hydrolases; PGN murein hydrolase activity and generalized autolysis; Amidase MurA [30], LytA amidase; the major murein hydrolase of *S. pneumoniae*, is considered an important virulence factor [31]. PGN murein

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hydrolase activity and generalized autolysis; Lytic Amidase LytA [32]. LytM; LytM is a glycylglycine endopeptidase Eps) that promotes lethal cell wall degradation after exposure to antibiotics that inhibit PG biosynthases [33]. Lysostaphin-like PGN hydrolase and glycylglycine endopeptidase LytM are efficient, in which PGN murein hydrolase activity and generalized autolysis are suitable to enzymatically active domain of autolysin LytM [34]. PGN Hydrolase LytB; PGN-hydrolases LytB is necessary for its optimal activity toward peptidoglycan hydrolysis and for pneumococcal adhesion to respiratory epithelial cells [35].

5. PGN degradation by zinc-containing inhibitive PGN elongation

Zinc inhibits protein substrates such as that each individual caspase has a variety of interactive surface elements as well as a reactive cysteine nucleophile in the active site, which could facilitate binding zinc or other biologically-relevant metals [36]. Namely, zinc ions-induced PGN inhibitive elongation due to inhibitory TG enzyme and activated autolysin against *S aureus* is involved that inhibition of PGN elongation due to the activations of autolysins Zn^{2+} binding Rv3717 is induced to a potential role of N-Acetylmuramyl L-alanine Amidase [28]. Zn^{2+} ions also causes tumor suppressor PTEN degradation which is mediated by ubiquitin-associated proteolytic process in the airway epithelium that A phosphatidylinositol 3-kinase inhibitor blocked PTEN degradation induced by Zn^{2+}, suggesting that phosphatidylinositol 3-kinase may participate in the regulation of PTEN [37]. Lysis of bacteria PGN cell walls by electrolyte ions results from damage to PGN biosynthesis by TG and TP and PGN inhibition by PGN autolysin activation. The other is caused by lysis of the outer membrane cell wall of bacteria by electrolyte ions and results from disruption of the outer membrane structure and inhibition of PGN elongation through disruption of PGN biosynthetic TP, and activation of PGN autolysin that metal ions also are required for PGN cell wall integrity, and the binding to peptidoglycan provides this stability [38].

However, it is thought that the activations of these PGN autolysins could be enhanced the inhibitions of PGN elongation simultaneously, with bacteriolysis of *S. aureus* PGN cell wall, namely, zinc(II) inhibits PGN biosynthesis and activates PGN autolysin, in which zinc promotes PGN cleavage, hydrolysis, activated autolysin, and inhibitive elongation, and then accordingly causing PGN cleavage and leading PGN elongation inhibition. Zinc can activate PGN autolysins that are associated enzymes, in which are essential for cell proliferation, represent one promising such new minimal peptidoglycan fragment that zinc induced PGN inhibitory synthesis corresponds to disruption of bacterial cell wall, but zinc ions may be possible to inhibit PGN synthesis TG and PGN elongation by PGN activated major autolysin of amidase against *S. aureus.*

Thus, zinc(II) ions can impair the activity of PGN biosynthesis TG and PGN elongation by bacteriolytic destruction of bacterial cell walls, causing bacterial lysis [39].Accordingly, zinc sulfate solution inhibits PGN biosynthesis, activates PGN autolysins, and inhibits PGN elongation against *S. aureus* cell wall, in which the activations of these PGN autolysins could be enhanced by PGN elongation inhibition, simultaneously, with zinc ions-induced PGN cleavage and hydrolysis by activated autolysins of AmiE, Rv3717 and PGRPs, MurA, LytA, LytM-endopeptidase, hydrolase LytB with bacteriolysis of *S. aureus* PGN cell wall that zinc ions promote PGN cleavage, hydrolysis, activated autolysin, and inhibitive elongation, and then causing further autolytic PGN cleavage, hydrolysis, degrading, and leading bactericidal PGN dissolution.

In summary, such as above-mentioned, zinc(II) in $ZnSO_4$ solution inhibits PGN biosynthesis, activates PGN autolysin, and inhibits PGN elongation against *S. aureus* cell wall, in which zinc(II) induced PGN autolytic cleavage and hydrolysis by activated autolysins, respectively, of AmiE, Rv3717 and PGRPs, MurA, LytA, LytM-endopeptidase, hydrolase LytB can be promoted, and then causing PGN autolysin cleavage, hydrolysis, and degrading, and leading PGN bactericidal dissolution.

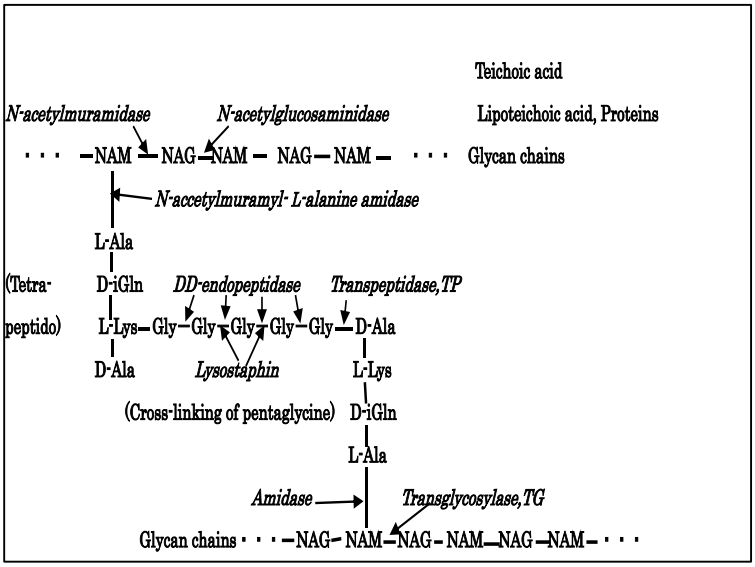

Figure 3: The molecular structure of *S.aureus* PGN cell wall, and the action sites of PGN biosynthesis enzymes of TG/TP and PGN forth autolysins and Lysostaphin enzyme.

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Table 1: PGN biosynthesis and autolysins against *S. Aureus*

PGN biosyntheses	PGN autolysins
• Transglycosylase, TG • Transpeptidase, TP	• N-acetylmuramidase • N-acetylglucosaminidase • N-accetylmuramyl-L-alanine amidase • DD-endopeptidase *(Lysostaphin)*

Conclusions:

Bacterial peptidoglycan (PGN) cell wall clearance on autolytic cleavage, hydrolysis, and degradation by zinc sulfide solution has been elucidated on the ground of the results obtained from halo antibacterial susceptibility tests in metal sulfate solutions against *Staphylococcus Epidermidis*, in which the order of antibacterial effect in the metal sulfate solutions becomes to be $Zn^{2+} > Cu^{2+} > Ag^{+} > Al^{3+}$ and the highest antibacterial activity is found to be the $ZnSO_4$ solution with a strong acid or strong electrolyte.

Zinc(II) can inhibit PGN biosynthesis TG against *S. aureus* and zinc ion regulates PGN synthetic enzymes that Zn^{2+} ions are most commonly coordinated by cysteine, followed by histidine, aspartate, and glutamate that Zn-cysteine complex in bacteria. Zinc intoxication also is observed to disrupt or inhibit PGN biosynthesis**.**

PGN autolytic cleavage by zinc-containing autolysin amidase, AmiE, Rv3717, AmiA: PGN cell wall cleavage by a *staphylococcal* autolysin is involved in zinc dependent metalloenzyme AmiE and autolysin amidase Rv3717. PGN cell wall cleavage by a *staphylococcal* autolysin is involved in zinc dependent metalloenzyme AmiE and autolysin amidase Rv3717. Zn^{2+} ions in $ZnSO_4$ solution cleave PGN cell wall murein by *Staphylococcal* amidase autolysin. Zinc(II) can cleave bacterial PGN murein chains that As PGN cleavage by *Staphylococcal* autolysin, zinc dependent metalloenzyme AmiE is efficient as prevention of the pathogen growth.

The other, Zn^{2+} binding AMIDASE Rv3717 showed no activity on polymerized PGN and however, it is induced to a potential role of N-Acetylmuramyl L-alanine Amidase. Inhibition of PGN elongation due to the activations of autolysins Zn^{2+} binding Rv3717 is induced to a potential role of N-Acetylmuramyl L-alanine Amidase. PGN cleavage by zinc-binding autolysin *Staphylococcus aureus* Amidase is involved that the AmiA shed light on PGN binding and cleavage**.**

PGN hydrolysis by zinc-containing enzymes; PGN murein hydrolase activity is involved that PGN hydrolysis by Zn^{2+}-containing catalysis is consistent in PGRPs, PGN murein hydrolase activity and generalized autolysis; Amidase MurA, LytA amidase, the major murein hydrolase of *S.pneumoniae*. Zinc can hydrolyze bacterial PGN protein that LytM is a glycylglycine endopeptidase, enzymatically active domain of autolysin LytM, and LytB is necessary for its optimal activity toward peptidoglycan hydrolysis and for pneumococcal adhesion to respiratory epithelial cells.

PGN degradation by inhibitive PGN elongation: *S. aureus* PGN cell wall degradation by Zn^{2+} ions is contributed to the inhibition of PGN elongation due to the activation of PGN autolysins of amidases and endopeptidase. Zinc ions-induced PGN inhibitive elongation due to inhibitory TG enzyme and activated Zn^{2+} binding Rv3717 autolysin against *S aureus* is shown to be induced to a potential role of N-Acetylmuramyl L-alanine Amidase. Thus, zinc(II) promotes PGN cleavage, hydrolysis, activated autolysin, inhibitive elongation, in which zinc inhibits PGN biosynthesis and activates PGN autolysin, and then causing PGN cleavage and hydrolysis, leading PGN elongation inhibition.

Accordingly, zinc sulfate solution inhibits PGN biosynthesis, activates PGN autolysins, and inhibits PGN elongation against *S. aureus* cell wall that zinc ions promote PGN cleavage, hydrolysis, activate PGN autolysins, and inhibit PGN elongation, and then causing further PGN autolytic cleavage, hydrolysis, degrading, and leading bactericidal PGN dissolution. The activations of these PGN autolysins could be enhanced by PGN elongation inhibition, simultaneously, with zinc ions-induced PGN autolytic cleavage and hydrolysis by activated autolysins of AmiE, Rv3717 and PGRPs, MurA, LytA, LytM-endopeptidase, hydrolase LytB with bacteriolysis of *S. aureus* PGN cell wall

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