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Coagulase Negative Staphylococci - Contaminants / Opportunistic Pathogens?

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Abstract:

Coagulase- negative Staphylococci (CoNS) though regarded as normal flora of skin, have recently gained a remarkable importance as nosocomial pathogen causing wide range of infections from skin and soft tissues to systemic blood stream infections. The ability of these bacteria to adhere stubbornly on various devices being biofilm formers, giving the treatment plan is strenuous. These bacteria are also appeared to exhibit several virulent attributes such as haemolysin, protease, DNase, lipase, capsule, haemagglutination, lipoteichoic acid, production of β-lactamase and formation of biofilm considering them as an inevitable clinical pathogen in healthcare facilities. Nevertheless, considering its mere existence on the skin surface of debilitated individuals and patient with implants and other artificial prosthesis, their immunity and virulence of these bacteria cannot be simply obsoleted and clinicians must give more insights and treat them as opportunistic pathogens.

Keywords: Coagulase- negative Staphylococci (CoNS), nosocomial pathogen, virulence, antibiotic resistance.

Introduction

Coagulase- negative Staphylococci (CoNS), formerly disregarded as contaminants, gradually gained importance as a serious healthcare associated pathogen owing to the immune status of the patient as well increased invasiveness in their investigative and diagnostic procedures (**Becker K ., 2014, Bhatt P.,2016).**They still remain as the common distinct group of clinically significant bacteria as they pose a substantial glitch in their identification and differentiation from the pathogenic strains of Coagulase positive Staphylococci (CoPS)(**Kloos WE and Bannerman TL., 1994)**.Increased antibiotic resistance occurring among the serious health care associated pathogens creates an additional major hurdle restricting the choice of treatment of infections caused by CoNS than of CoPS(**Becker K ., 2014).**CoNS resembles *S.aureus* in morphology appearing as Gram positive cocci arranged in irregular clusters. They are negative for coagulase production unlike *S.aureu*(**Rupp ME and Archer GL et al., 1994**).Greater prevalence of these bacterial infections may be accounted for their specific attraction to the foreign substances like prosthetics, catheters and other devices that are commonly employed in the investigative procedures. This may attribute to the emergence of CoNS as a significant nosocomial pathogen associated with substantial fatality, despite being treated as inoffensive organisms(**Huebner J and Goldmann DA ., 1999**).Over than 50 species of CoNS have been described till date including occasional species like *S. pettenkoferi, S.massiliensis and S.petrasii*(Michalik M et al., 2020).Despite not exhibiting specific tropism towards specific niches within the body, they exists as commensals in various *in-vivo* sites such as *S.capitis* on head, *S.auricolaris* in the ear canal, *S. saprophyticus* in the inguinal region , *S. haemolyticus* in the axillary region, *S.epidermidis* and *S. hominis* occurring along all the body sites and readily be transferred among humans through direct contact(**Fontana C andFavaro M., 2018**).Frequently encountered clinically significant species of CoNS includes *Staphylococcus epidermidis* and *Staphylococcus haemolyticus*. Other recently reported clinically potent species of CoNS group includes *Staphylococcus saprophyticus*, *Staphylococcus lugdunensis*with greater virulence potential, analogous to *S. aureus* and *S. caprae*, an occasional member of the CoNSspeciesreported among human infections (**Mazur E et al., 2017, Fontana C andFavaro M., 2018, Heilbronner S and Foster TJ., 2020**).

Clinical significance of CoNS may be due to the following reasons; i) Weakened immune system, ii) Disruption of the skin and mucosa, iii) Ready colonisation of the indwelling catheters and other medical devices**(Heilmann C et al., 2019).**Unlike*S.aureus*, CoNS are always regarded as least pathogenic organisms. Still, they are armed with various unknown armaments that makes them potentially infectious and persistence with in the host**(**Michalik M et al., 2020**).**They are considered clinically significant not only based on their pathogenic ability but even because of their antagonistic property against the pathogenic *S. aureus* and their presence may influence the colonisation of *S. aureus*(**Peng P et al., 2019, Chin D et al.,2021**).Furthermore, the clinical significance of CoNS species is supplemented by the potent ability of forming biofilms especially associated with the indwelling catheters or other medical implants leading to chronic infections and the limitations of the therapeutic options due to the emergence of methicillin resistance and reduced glycopeptide susceptibility among the clinical strains of CoNS species (**Stewart PS and CostertonJW ., 2001, Becker K et al., 2014**).

Despite *S. aureus* presumed to be the only pathogenic species exhibiting virulence factors, various clinical CoNS species involved in causing human infections elaborates the presence of various virulence determinants such as haemolysin, protease, capsule, haemagglutination, lipoteichoic acid, β-lactamase and biofilm production(**Türkyilmaz Sand Kaya O ., 2006, Akinkunmi E. O. and Lamikanra A., 2012**). In this review, various virulence factors elaborated by the clinically significant CoNS species are reviewed.

Haemolysin Production:

Among the various clinical CoNS Species, *S. epidermidis* documented to be the predominant member among the CoNS to exhibit haemolysis trailed by other members of the CoNSgroup . Haemolytic property of the CoNS is found to be conferred by the presence of *hla*, *hla_yidD, hld*and *hlb* genes respectively(**Cunha Mde L et al., 2006**).Production of haemolysin by CoNS members exerts a statistically significant substantial association with the patterns of antibiotic resistance. Greater prevalence of levofloxacin and erythromycin resistance was observed among the CoNS strains with *hld*gene while resistance to clindamycin was associated with *hlb*gene and resistance towards rifampicin and novobiocin strongly linked with the presence of *hla*/*yidD* genes(**Nasaj M et al., 2020**).A study conducted by Votava et al, 1990, demonstrated the haemolysin production in *S. haemolyticus*, a leading haemolysin

producer followed by other seven members of CoNS species included in the study (**Votava M et al., 1990**).S. lugdunesis was considered more significant among the coagulase negative staphylococci as they exhibited the presence of a δ-like CoPS haemolysin. This heat stable haemolysin, regulated by *hld* gene, also exhibited synergism with the β toxin secreted by *S. intermedius*(**Vandenesch F, et al., 1991**).

Protease, DNase and Lipase production:

Recent studies have documented the predominant production of protease, DNase and Lipase by Coagulase Negative Staphylococci (CoNS) species, especially among *S. epidermidis* than the species of Coagulase Positive Staphylococci (CoPS) (**Votava M et al., 1990, SaisingJ et al., 2012, Jameel ZJ., 2015**).

Capsule:

Capsular polysaccharide present in CoNS contributes to the major virulence factors deliberates the resistance to the phagocytic process, analogous to capsular polysaccharide present in CoPS(**Hancock IC., 1989**).Expression of capsular polysaccharide among CoNS (i.e., *Staph. chromogenes, Staph. hominis* and *Staph. hyicus.*) serves as a major contributing factor towards their pathogenic nature of causing mastitis among the ovine and bovine animals (**Matthews KR**et al., 1991, BurrielAR and ScottM.A., 1998).

Haemagglutination:

Potential mechanism of microbial pathogenicity or virulence is the attachment of the organism to the mucosal surfaces resulting in the host tissue colonisation (**Beuth J et al., 1988**). Presence of Haemagglutinins (also called as lectins) especially among *S. saprophyticus* facilitates binding to the uroepithelial surfaces, are suggestive of playing a major role inenhancing the mechanism of adhesion among the species of coagulase negative Staphylococci (**Hovelius B and Mårdh PA., 1979,** *WadströmT ., 1987)***.**Several CoNS species such as *S. saprophyticus*, *S. warneri*, *S. haemolyticus* and *S. epidermidis* possess haemagglutinins(**Beuth J et al., 1988**).Haemagglutinin specificity of CoNS species may be of different types which can be majorly influenced by the presence of carbohydrate moiety in the regional area of adhesion. For instance, CoNS species colonised in lungs exhibits N-acetyl Neuraminidase (NANA) specificity while that of CoNS species colonised in uroepithelium exhibits N-acetyl galactosamine (GalNAc) specificity(**Beuth J et al., 1988)**.

Lipoteichoic acid:

Presence of lipoteichoic acid in the cell wall of Gram- positive cocci (particularly among the Staphylococcus species) are found to intercede the attachment of the organism to the host tissues (**Beuth J et al., 1988, MG andGründlingA., 2014, Ahn KB et al., 2018).**Teichoic acid serves as a persistent indicator to identify the chemotaxonomic character of a bacteria within the given species. Nevertheless, the different species of Staphylococci exhibit differences in their aminoacid residues. For instance, poly ribitol phosphate in CoPS is replaced by poly-glycerol phosphate in CoNS(**Endl J et al., 1984**).Also, theWTA structure of CoNS is a poly-glycerol phosphate identical with the newly described ST395 lineage of *Staphylococcusaureus*(Winstel V et al., 2014).

β-lactamase:

Studies on expression of β – lactamase among penicillin resistant CoNS species conducted by Rosdahl*et al*, 1986 documented the potent β – lactamase production with majority being reported with *S. epidermidis* among the clinically significant members of CoNS species, parallel to β – lactamase produced by *S. aureus*(Rosdahl VT et al., 1986).β-lactamase production by CoNS species is conferred by the presence of PBP 2a, a homologous regulatory protein with low affinity towards β-lactam antibiotics encoding *mecA* gene expressed among the Methicillin Resistant Staphylococcus Aureus (MRSA) strains. Presence of Sccmec type III and type IV among *S. epidermidis* exhibits multidrug resistance (Xu Z et al., 2020).

Biofilm production:

Biofilms produced by bacteria constitutes a organised framework of bacteria bounded within its polymeric matrix or adherence to biotic or abiotic surfaces, which may contribute to a major virulence factor associated with the persistence of chronic infections(Costerton JW et al., 1999).Ability of biofilm production among CoNS is entrusted with the polysaccharide intercellular adhesin (PIA) encoded by *icaADBC* locus and *ica* independent locus.Biofilm formation is regarded as the significant factor contributing to the virulent nature of CoNS(Soumya KR et al., 2017).Biofilm formation among CoNS takes place in four steps;

attachment, multiplication, biofilm formation, detachment and dissemination of the multiplied bacterial cells either singly or in agglomerates to the blood stream to initiate biofilm formation in the various other sites of colonisation(**Becker K ., 2014**).Initial adherence of CoNS to various biotic or abiotic surfaces, considered to be a predominant necessitating factor influencing biofilm formation, are found to be mediated by an enzyme AtlE (especially in *S. epidermidis*) identical to the autolysin *Atl* of the Coagulase positive *Staphylococcus aureus*(**Heilmann C et al., 1997**).Presence of *Atl* identical proteins designated as AtlC, Aas, AtlL and AtlWM are documented among other CoNS species such as *S. caprae*, S. *saprophyticus*, *S. lugdunensis* and *S. warneri M* respectively (Hell W **et al., 1998, Allignet J et al., 2001,** *Yokoi KJ et al., 2008,* Bourgeois I et al., 2009).Additionally, other cell wall proteins such as *Bhp*, *Fbe* and intercellular adhesins mediates the process of initial adherence(Soumya KR et al., 2017).

Furthermore, ClpP protease, an ATP dependent bicomponent protease enzyme, plays a major role in the development of biofilm and virulence among S. epidermidis. This bicomponent protease bears a proteolytic component with two distinct subunits (ClpP and ClpQ) and a regulatory component (*Wang C et al., 2007*).

These enzymes are found to be mediating a major supplemental role in *S. epidermidis* towards adaptation of bacteria and formation of biofilm thereby contributing to its virulence (*Wang C et al., 2007*). Formation of polysaccharide intercellular adhesion requires the genes (*icaA*, *icaD*, *icaB* and *icaC*) encoded by*ica* gene locus indispensable for the biofilm formation (**NamvarAE** et al., 2013[)]**.**

Production of biofilms and multidrug resistance are found to be elevated among the CoNS species especially *S. haemolyticus*evidenced in a study by Allori*et al*, 2006.

Newly identified virulence factors among CoNS species:

i) Phenol soluble modulins (PSM):

CoNS species also elaborates the presence of toxic peptides with appreciable ability to produce cytolysis promoting sepsis designated as Phenol soluble modulins **(**PSM)(Da F et al., 2017, Qin L et al., 2017). Although most characterised PSMs are found to be gene encoded at its core, potential expression of a mobile genetic element *psm-mec* gene is found to be present

in Methicillin resistant CoNS species identical to Methicillin Resistant Staphylococcus aureus (Queck SY et al., 2009).

ii) Cell Wall anchored proteins:

Cell wall anchored proteins (CWA proteins) are regarded as essential virulence factorsamong Gram positive bacteria playing a vital role in theformation of biofilm (Arora S et al., 2016, Pickering AC et al., 2020).CWA proteins present in Gram positive organisms can be broadly classified into two broad types which includes i) Microbial surface Components Recognizing Adhesive Matrix Molecules and ii) Serine Rich Repeat Proteins (SRRPs) (Arora S et al., 2016).Biofilm formation by CoNS is found to be mediated either by binding to these matrix proteins or aggregation of intercellular substance such as Accumulation associated protein (Aap), serine-aspartate dipeptide repeat protein F (SdrF), extracellular matrix binding protein (Embp) and a recently reported SesC protein from *Staphylococcus epidermidis*(Pei L and Flock JI., 2001, Arrecubieta C et al., 2009, Christner M et al., 2010, .Shahrooei M.2010, Schaeffer CR et al., 2015,).

Conclusion:

Hence, dissemination of CoNS originates from the skin and mucous membranes of their common colonising habitat to the external environment(Michalik M et al., 2020).Though being considered as harmless commensal, their nature of dissemination as well as recurrently recovered pathogen among the nosocomial patients with implants and immunocompromised patients have induced the curiosity about the pathogenicity and the virulent armaments of CoNS species(**Fontana C andFavaro M., 2018**).Nevertheless, they have gained significance as their multiplication predominantly occurs only among humans and animals(Michalik M et al., 2020).Contemporary medical approaches generally employ medical devices for either investigation or treatment, it is more probable for the CoNS species to emerge as conducive potential pathogen leading to morbidity and mortality which really deserves attention by the clinicians as well as microbiologists(Fontana C &Favaro M., 2018).

REFERENCES:

Ahn KB, Baik JE, Yun CH, Han SH. Lipoteichoic Acid Inhibits *Staphylococcus aureus* Biofilm Formation. Front Microbiol. 2018 Feb 27;9:327.

Akinkunmi E. O. and Lamikanra A. Phenotypic Determination of Some Virulence Factors in Staphylococci IsolatedFrom Faecal Samples of Children in Ile-Ife, Nigeria. Afr. J. Biomed. Res.2012 May; 15: 123 – 128.

Allignet J, Aubert S, Dyke KG, El Solh N. *Staphylococcus caprae* strains carry determinants known to be involved in pathogenicity: a gene encoding an autolysin-binding fibronectin and the ica operon involved in biofilm formation. Infect Immun. 2001 Feb;69(2):712-8.

Allori MCGD, Jure MA, Romero C and Castillo MECD. Antimicrobial Resistance and Production of Biofilms in Clinical Isolates of Coagulase-Negative Staphylococcus Strains. Biol. Pharm. Bull. 2006: Vol. 29, No. 8

Arora S, Uhlemann A-C, Lowy FD and Hook M (2016) A Novel MSCRAMM Subfamily in Coagulase Negative Staphylococcal Species. Front. Microbiol. 2016: 7:540.

Arrecubieta C, Toba FA, von Bayern M, Akashi H, Deng MC,etal.SdrF, a *Staphylococcus epidermidis* Surface Protein, Contributes to the Initiation of Ventricular Assist Device Driveline–Related Infections. PLOS Pathogens; 2009 5(5): e1000411.

Bhatt P, Tandel K, Singh A, Mugunthan M, Grover N, Sahni AK. Species Distribution and Antimicrobial resistance pattern of Coagulase-negative Staphylococci at a tertiary care centre. Med J Armed Forces India. 2016 Jan;72(1):71-74.

Becker K, Heilmann C, Peters G. Coagulase-Negative Staphylococci. Clinical Microbiology Reviews .2014 October; 27(4): 870 –926

Beuth J, H. Ko HL, Schumacher-PerdreauF,Peters G, Heczko P and Pulverer G . Hemagglutination by Staphylococcus saprophyticus and other coagulase-negative staphylococci. Microbial Pathogenesis1988; 4: 379-383

Bourgeois I, Camiade E, Biswas R, Courtin P, Gibert L, Götz F, Chapot-Chartier MP, Pons JL, Pestel-Caron M. Characterization of AtlL, a bifunctional autolysin of *Staphylococcus lugdunensis* with N-acetylglucosaminidase and N-acetylmuramoyl-l-alanine amidase activities. FEMS microbiology letters. 2009 Jan 1;290(1):105-13.

BurrielAR.ScottM.A comparison of methods used in species identification of coagulase-negative staphylococci isolated from the milk of sheep. The Veterinary Journal :1998 March: 155 (2) :183-188

Chin D, Goncheva M I, Flannagan R S, Deecker SR, Guariglia-Oropeza V, Ensminger AW & Heinrichs DE. Coagulase Negative Staphylococci release a Purine analog that inhibits *Staphylococcus aureus* virulence. Nature Communications .2021: 12:1887.

Christner M, Franke GC, Schommer NN, Wendt U, Wegert K, Pehle P, Kroll G, Schulze C, Buck F, Mack D, Aepfelbacher M, Rohde H. The giant extracellular matrix-binding protein of *Staphylococcus epidermidis* mediates biofilm accumulation and attachment to fibronectin. Mol Microbiol. 2010 Jan;75(1):187-207.

Costerton JW, Stewart PS, Greenberg EP. Bacterial biofilms: a common cause of persistent infections. Science. 1999 May 21;284(5418):1318-22.

Cunha Mde L, Rugolo LM, Lopes CA. Study of virulence factors in coagulase-negative staphylococci isolated from newborns. Mem Inst Oswaldo Cruz. 2006 Sep;101(6):661-8.

Da F, Joo HS, Cheung GYC, Villaruz AE, Rohde H, Luo X, Otto M. Phenol-Soluble Modulin Toxins of *Staphylococcus haemolyticus*. Front Cell Infect Microbiol. 2017 May 24;7:206.

Endl J, Seidl PH, Fiedler F, Schleifer KH. Determination of cell wall teichoic acid structure of staphylococci by rapid chemical and serological screening methods. Arch Microbiol. 1984 Mar;137(3):272-80.

Fontana C, Favaro M. Coagulase-Positive and Coagulase-Negative Staphylococci in Human Disease.2018 Elsevier: 25-42

Hancock IC. Encapsulation of Coagulase-Negative Staphylococci. ZentralblBakteriol: 1989: 272 (1): 11-18.

Heilbronner S, Foster TJ. *Staphylococcus lugdunensis*: a Skin Commensal with Invasive Pathogenic Potential. Clin Microbiol Rev. 2020 Dec 23;34(2): e00205-20.

Heilmann C, Hussain M, Peters G and Gotz F. Evidence for autolysin-mediated primary attachment of *Staphylococcus epidermidis* to a polystyrene surface.Molecular Microbiology (1997)24(5), 1013–1024.

Heilmann C, Ziebuhr W, Becker K. Are coagulase-negative staphylococci virulent? Clin Microbiol Infect. 2019 Sep; 25(9):1071-1080.

Hell W, Meyer HG, Gatermann SG. Cloning of aas, a gene encoding a *Staphylococcus saprophyticus* surface protein with adhesive and autolytic properties. Mol Microbiol. 1998 Aug;29(3):871-81.

Hovelius B, Mårdh PA. Haemagglutination by Staphylococcus saprophyticus and other staphylococcal species. Acta PatholMicrobiolScand B.1979 Feb;87B (1):45-50.

Huebner J, Goldmann DA, Coagulase-negative Staphylococci: Role as Pathogens. Annu Rev Med 1999; 50:223–36.

Jameel ZJ, Nayef SH, Al-Mahdawi MA. Study Some Virulence Factors of Coagulase Positive Staphylococci and Coagulase Negative Staphylococci Isolated from Different Parts of Human Body. Tikrit Journal of Pure Science. 2015 .september: 20 (4).

Kloos WE and Bannerman TL. Update on Clinical Significance of Coagulase-Negative Staphylococci. Clinical Microbiology Reviews. 1994 Jan; 7(1): 117-140

Matthews KR, Harmon RJ, Langlois BE. Effect of naturally occurring coagulase-negative staphylococci infections on new infections by mastitis pathogens in the bovine. J Dairy Sci. 1991 Jun;74(6):1855-9.

Mazur E ,Zychowski P , Juda M, Korona-Glowniak I ,Niedzielska G, Malm A , Koziol-Montewka M. First report of a *Staphylococcus caprae* isolated from middle ear fluid of an infant with recurrent acute otitis media. Annals of Agricultural and Environmental Medicine 2017, 24(3), 357–359.

Michalik M, Samet A, Podbielska-Kubera A,SaviniV,MiędzobrodzkiJ and Kosecka-StrojekM.Coagulase-negative staphylococci (CoNS) as a significant etiological factor of laryngological infections: a review.Ann Clin MicrobiolAntimicrob. 2020; 19: 26.

Namvar AE, Asghari B, Ezzatifar F, Azizi G, Lari AR. Detection of the intercellular adhesion gene cluster (ica) in clinical *Staphylococcus aureus* isolates. GMS Hyg Infect Control. 2013 Apr 29;8(1):Doc03.

Nasaj M, Saeidi Z, Asghari B, RoshanaeiG ,Arabestani MR. Identifcation of Hemolysin encoding genes and their association with Antimicrobial Resistance pattern among clinical isolates of Coagulase-negative Staphylococci. BMC Research Notes. (2020) 13:68

Pei L, Flock JI. Lack of fbe, the gene for a fibrinogen-binding protein from *Staphylococcus epidermidis*, reduces its adherence to fibrinogen coated surfaces. MicrobPathog. 2001 Oct;31(4):185-93.

.Percy MG andGründlingA.Lipoteichoic Acid Synthesis and Function in Gram-Positive Bacteria. Annual Review of Microbiology 2014 68:1, 81-100

Peng P, Baldry M, Gless BH, Bojer MS, Espinosa-Gongora C, Baig SJ, Andersen PS, Olsen CA and Ingmer H. Effect of Co-inhabiting Coagulase Negative Staphylococci on S. aureus agr Quorum Sensing, Host Factor Binding, and Biofilm Formation. Frontiers in Microbiology.2019: 10:2212

47.Pickering AC, Fitzgerald JR. The Role of Gram-Positive Surface Proteins in Bacterial Niche- and Host-Specialization. Front Microbiol. 2020 Oct 29;11:594737.

Qin L, Da F, Fisher EL, Tan DCS, Nguyen TH, et al. (2017) Toxin Mediates Sepsis Caused by Methicillin-Resistant *Staphylococcus epidermidis*. PLOS Pathogens 13(2): e1006153.

46.Queck SY, Khan BA, Wang R, Bach THL, Kretschmer D.Mobile Genetic Element-Encoded Cytolysin Connects Virulence to Methicillin Resistance in MRSA. PLOS Pathogens:2009 5(7): e1000533.

Rosdahl VT, Jarløv JO, Knudsen AM. beta-lactamase production in coagulase-negative micrococcaceae. Acta PatholMicrobiol Immunol Scand B. 1986 Dec;94(6):423-7.

Rupp ME, ArcherGL.Coagulase-Negative Staphylococci: Pathogens Associated with Medical Progress**.***Clinical Infectious Diseases,* 1994, August, 19(2): 231–245

SaisingJ , Dube L, Ziebandt AK, Voravuthikunchai SP, Nega M and Götza F. Activity of Gallidermin on *Staphylococcus aureus* and *Staphylococcus epidermidis* Biofilms. Antimicrobial Agents and Chemotherapy .2012 November 56 (11) ;5804 –5810.

Schaeffer CR, Woods KM, Longo GM, KiedrowskiMR,Paharik AE, Büttner H, Christner M, Boissy RJ, Horswill AR, Rohde H, Fey PD. Accumulation-associated protein enhances *Staphylococcus epidermidis* biofilm formation under dynamic

conditions and is required for infection in a rat catheter model. Infect Immun . 2015.83:214–226.

Shahrooei M. Identification of Potential Targets for Vaccination against *Staphylococcus epidermidis* Biofilms. Leuven, Doctoral thesis in Medical Sciences.2010

Soumya KR, Philip S, Sugathan S, Mathew J, Radhakrishnan EK. Virulence factors associated with Coagulase Negative Staphylococci isolated from human infections. 3 Biotech. 2017 Jun;7(2):140.

Stewart PS, Costerton JW. Antibiotic resistance of bacteria in biofilms. Lancet 2001; 358: 135–38

Türkyilmaz S, Kaya O. Determination of some Virulence Factors in Staphylococcus Spp. Isolated from Various Clinical Samples. Turk J Vet Anim Sci 30 (2006) 127-132

18. Vandenesch F, Storrs MJ, Poitevin-Later F, Etienne J,Courvalin P and Fleurette J. 8-Like haemolysin produced by *Staphylococcus lugdunensis*.FEMS Microbiology Letters.1991: 78: 65-68

Votava M,Skalka B, SediváJ. Slime production, hemolysis and proteolysis in Coagulase-negative Staphylococci isolated in Hemoculture from humans.CeskEpidemiolMikrobiol Imunology.1990 Sep;39(5):288-93.

27.WadströmT. Molecular aspects on pathogenesis of wound and foreign body infections due to staphylococci**.** Medical Microbiology, Infectious Diseases, Virology, Parasitolog**y.** 1987 August :266, (1–2),191-211.

Wang C, Li M, Dong D, Wang J, Ren J, Otto M, Gao Q. Role of ClpP in biofilm formation and virulence of Staphylococcus epidermidis. Microbes Infect. 2007 Sep;9(11):1376-83.

31.Winstel V, Sanchez-Carballo P, Holst O, Xia G, Peschel A. Biosynthesis of the unique wall teichoic acid of *Staphylococcus aureus* lineage ST395. mBio. 2014 Apr 8;5(2):e00869.

*33.*Xu Z, Cave R,Chen L, Yangkyi T, KeLi Y, Meng G,Niu K, Zhang W,Tang N, Shen J, MkrtchyanH.Antibiotic resistance and molecular characteristics of methicillin-resistant Staphylococcus epidermidis recovered from hospital personnel in China.Journal of Global Antimicrobial Resistance :2020 September: 22,(195-201).

39.Yokoi KJ, Sugahara K, Iguchi A, et al. Molecular properties of the putative autolysin Atl(WM) encoded by Staphylococcus warneri M: mutational and biochemical analyses of the amidase and glucosaminidase domains. Gene. 2008 Jun;416(1-2):66-76.