MEDICAL DEVICE TECHNOLOGIES: POTENTIAL TO TREAT AND PREVENT BIOFILM RELATED INFECTIONS

Abstract

Biofilms play a significant role in infection control and healthcare-related infections due to their inherent ability to withstand and resist antimicrobial treatments. These structured communities of microorganisms have been observed forming on the surfaces of medical devices. The release of both single and clustered microbial cells from these biofilms carries a notable risk of spreading infection within the host, thereby increasing the likelihood of infections and presenting a substantial public health concern. Microbial biofilms can establish themselves on or within various implanted medical devices, such as contact lenses, central venous catheters, needleless connectors, endotracheal tubes, intrauterine devices, mechanical heart valves, pacemakers, peritoneal dialysis catheters, prosthetic joints, tympanostomy tubes, urinary catheters, and voice prostheses. The colonization of these medical instruments plays a pivotal role in the challenge of healthcare-associated infections. This article's objective is to provide a comprehensive overview of biofilm science, the associated risks, the potentially severe consequences of infections, and both existing and emerging advanced technologies aimed at addressing the biofilm issue to enhance the healthcare system.

Keywords: Biofilm, Medical Device, Microbial Infection, Healthcare, and Biomedical Technology

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I. INTRODUCTION

An organized group of microbial cells that are attached to a living or non-living surface and contained in a self-produced extracellular polymeric matrix is known as a biofilm. A microbial adaptation to harsh settings is thought to be the cause of this phenomenon, known as biofilm [1-2]. Experimental data from P. aeruginosa investigations conducted in vitro and in vivo clearly demonstrate that bacteria found in biofilms are much more resistant to antibiotics and host immune defenses than bacteria found free-floating [3–7]. Antibiotic therapy that is aggressive and intensive is frequently used to treat persistent biofilm infections brought on by scattered bacteria and to slow the establishment of biofilm. However, eliminating these biofilm infections is difficult [7-8] because it is difficult to achieve a high enough antibiotic concentration to destroy mature biofilms in vivo [5]. As a result, it is very challenging to completely eradicate a bacterial biofilm infection once it has established itself. Bacterial biofilm development is common in human disorders, notably in patients with therapeutically implanted medical devices [2,7]. It is also common in natural aquatic habitats. As medical knowledge develops, a wider range of medical tools and artificial organs are used to treat human diseases. Sadly, this development also causes an increase in bacterial biofilm infections. The vast majority, if not all, of medical devices and prostheses may cause biofilm-related infections, according to reports. Catheters are included [9], vascular prostheses [10], cerebrospinal fluid shunts [11], prosthetic heart valves [12], urinary catheters [12], joint prostheses and orthopedic fixation devices [13], cardiac pacemakers [14], peritoneal dialysis catheters [15], intrauterine devices, biliary tract stents, dentures, breast implants, contact lenses, and in dental cases, caries and periodontitis, among others. It is estimated that a significant proportion, around 50%, of nosocomial infections are connected to indwelling medical devices and their associated biofilms. Bacterial biofilms are notably characterized by their high resistance to antibiotic treatment and immune responses [7]. While antibiotic treatment stands as a crucial and effective strategy for microbial infection control, it is exceptionally challenging to completely eliminate biofilm infections with antibiotics. In vitro and in vivo experiments consistently demonstrate that the minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC) required to combat biofilm bacterial cells are often much higher (approximately 10-1000 times) than those for planktonic bacterial cells [4-6]. Achieving effective in vivo antibiotic MBC levels to eradicate biofilms through conventional administration methods is hindered by antibiotic toxicities, side effects, and limitations in renal and hepatic functions. Consequently, the treatment of biofilm infections presents a considerable challenge that garners significant scientific attention. This review primarily focuses on providing an overview of biofilms, infections related to medical devices, the current treatments for bacterial biofilm infections, and prospective advancements in addressing medical device-associated biofilms.

II. MECHANISM OF BIOFILM FORMATION

`The process of biofilm formation is intricate and involves several distinct stages, namely attachment, aggregation, maturation, detachment, and dispersal. Attachment comprises a two-step process. Initially, microorganisms recognize the surface, followed by reversible and irreversible attachment. Non-specific cellular associations such as van der Waals forces, electrostatic forces, Lewis acid-base interactions, and hydrophobic interactions mediate reversible attachment. Conversely, irreversible adhesion is driven by specific adhesions found on pili, fimbriae, or the cell surface of microorganisms. Maturation

encompasses the aggregation and proliferation of bacteria on the surface after attachment, resulting in the formation of micro-colonies [6-8].

The irreversible attachment of bacteria to the surface triggers changes in gene expression, leading to the synthesis and secretion of extracellular polysaccharides (EPS) or an extracellular polymeric matrix (a characteristic of the biofilm condition). This matrix acts as a cementing substance, binding bacterial cell colonies together. The extracellular polymeric matrix is predominantly composed of polysaccharides, which can be neutral or polyanionic for Gram-negative bacteria and cationic for Gram-positive bacteria. It is highly hydrated, with a hydration level of up to 98%, and remains attached to the underlying surface [2-5]. Continuous multiplication, growth, and recruitment of additional microorganisms contribute to the development of a mature biofilm. This mature biofilm consists of densely packed microorganisms forming prominent outgrowth masses on surfaces.

The final stage of biofilm formation involves the detachment of microbes from the biofilm colonies, their translocation or dispersal, and subsequent attachment to new locations. The rate of biofilm growth on a medical device is influenced by various factors. For growth to occur, microorganisms must first attach themselves to the device's surface. This attachment requires a sufficiently long exposure period to prevent easy detachment. The effectiveness of this adherence is also influenced by the composition of microbes present in the surrounding fluid. Furthermore, the presence of different particles in the device's vicinity alters the properties of its surface. Consequently, the attachment of individual cells and the subsequent biofilm formation are facilitated (1). Table 1 provides a list of factors that impact biofilm formation.

Table 1: Factors Which Affect Biofilm Formation

Substratum	**Texture, hydrophobicity, conditoning film, surface charge**
Cell	Cell surface, hydrophobicity, fimbriae, flagella, pili, adhesions, other surface appendages, EPS
Aqueous medium	Velocity of medium, temperature, pH, cations, nutrients availability, antibacterial agents

Table 2: List of Medical Implants Prone To Biofilm Formation with the Causative Agent.

Medical device	**Microorganism**
Artificial hip prosthesis	S. aureus, S. epidermidis, P. aeruginosa, E. coli, Neisseria gonorrhoeae, Candida albicans and Candida dubliniesis
Prosthetic heart valves	Enterococcus, S. epidermidis, S. aureus, Streptococci, Diphtheria, Candida albicans and gram-negative bacilli,
Synthetic vascular grafts	S. aureus, Candida, Enterococcus, Streptococcus
Ventilator tubing	Acinetobacterbaumannii and Pseudomonas aeruginosa
Artificial voice prosthesis	Candida albicans, S. aureus, P. aeruginosa

central venous catheters	S. epidermidis, Enterococcus faecalis, K. pneumoniae, Candida albicans, P. aeruginosa, S. aureus
Orthopedic implants	S. epidermidis, P. aeruginosa, Enterococcus, S. aureus
Dental implants	Staphylococcus aureus, Candida albicans, Streptococcus
Intra-urine devices	S. epidermidis, K. pneumoniae, Enterococcus, Proteus mirabilis, P. aeruginosa, E. coli and other gramnegative bacteria

Table 3: Biological and Chemical Approaches For Biofilm Infection Treatment In Medical Devices.

Technologies	Descriptions	Antibiofilm agents	Refrences
Bacteriophage Therapy	Lytic phages utilized which results in rapid destruction of the bacterial cell, therapy is host specific and bactericidal	E.coli T4 phage, coliproteus bacteriophage	Burrowes et al. 2011
Antibacterial Peptides	Secreted by immune defense cells bears low MW, broad spectrum activity against bacteria and also proposed as novel antibiotics, bactericidal	lytic peptide PTP-7, cathelicidin peptides	Pompilio et al. 2011
Antimatrix Agents	Targets by disrupting components of the extracellular polysaccharide or glycocylax secreted by bacterial cell in biofilm, bactericidal	DNaseI, Dispersin B, Nacetylcysteine	Burton et al. 2006
Signal Transduction Interference	Gene expression is hindered by interfering with signaling receptors involved in transduction and modify virulence selection, bacteriostatic	QseC kinase inhibitor, Siamycin I	Gotoh et al. 2010
Chelating Agents	Interfere with metal ions, destabilize biofilm architecture along with interfering with bacterial membrane dynamics, bactericidal	sodium citrate, tetrasodiumEDTA, aminocycline-EDTA	Donlan 2011
Antiadhesion Agents	Compounds interfere with the adhesive properties of glycocylax or bacterial cell surface appendages, bactericidal or bacteriostatic	Mannosides, pilicides	Cusumano et al. 2011
Modifying Dispersal Signals	Signal for biofilm dispersion is combined with an antibacterial agent for killing the dispersed organisms, novel therapy, bactericidal or bacteriostatic	D-Amino Acids	Ma et al. 2011b

Table 4: Surface Modification Approaches to Prevent Biofilm Formation in Medical Devices.

Method	Description
Plasma treatment	Ionized gases generated artificially used to vaporize and redeposit metals for surface modification. eg. Trimethylsilane
Polymer modification	Antibiofilm compounds immobilized on implant surfaces via polymer chains through covalent coating which results in non-leachable, contact-killing surfaces. Eg. N-alkylpyridinium bromide attached to a poly(4-vinyl-N-hexylpyridine
Silver treatment	Implant treated with sodium hydroxide and silver nitrate solutions after oxygen glow discharge treatment
Palladium/tin salt mixture treatment	Immersion and rinsing in a palladium/tin salt solution
Quaternary ammonium silane coatings	Oxidized implant surfaces covered with QAS and left to react and dry, inhibits adhesion and viability property of bacterial cells
Ion implantation	Injects accelerated high-energy ions into the surface of a material to modify its physical, chemical and biological properties to inhibit the biofilm formation.
Bulk surface photografting	Surface modification of hydrophobic and bioinert polymer. The radiation breaks chemical bonding on material surface to be grafted and form free radicals followed by exposure to monomers to start surface graft polymerization
Unique configuration of noble metals	Prevent colonization of bacteria on medical device surface, eg. Bactiguard
Perfluoro-alkylsiloxane (PAS) treatment	Surface oxidized and PAS were chemisorbed on medical devices help to inhibit the biofilm

III. NEW TECHNOLOGIES TO PREVENT BIOFILM FORMATION IN MEDICAL DEVICES

Elimination of cells from the biofilm colony constitutes a crucial phase in the life cycle of biofilms as it facilitates their propagation and colonization of novel surfaces. The strategies to counteract bacterial biofilms should focus on thwarting their formation rather than dispersing established biofilms. Approaches to prevent biofilm formation encompass both "Chemical" and "Mechanical" methodologies.

IV. CHEMICAL METHODS:

1. **Antimicrobial Coatings:** The principal tactic for biofilm prevention entails chemical modifications. Antibiotics, biocides, and ion coatings are common chemical techniques for deterring biofilm formation. These methods impede biofilm establishment by disrupting the attachment and expansion of immature biofilms [16]. Numerous in vitro studies have validated the efficacy of silver in preventing infections, both as coatings and

as nanoparticles integrated into a polymer matrix. However, caution is necessary when applying silver within in vivo systems due to its potential toxic effects on human tissue. This underscores the necessity to uncover novel antimicrobial compounds that can inhibit biofilm growth.

2. **Polymer Modifications:** Antimicrobial agents can be immobilized on device surfaces using elongated, flexible polymeric chains. These chains establish covalent bonds with the device surface, creating non-leaching surfaces with contact-killing properties. An in vitro study demonstrated that attaching an antimicrobial agent called N-alkylpyridinium bromide to poly(4-vinyl-N-hexylpyridine) enabled the polymer to neutralize ≥ 99% of S. epidermidis, E. coli, and P. aeruginosa bacteria [17]. Dispersion forces between the polymer chains and bacterial cells hinder bacterial adhesion and initiation of biofilm formation. This concept is akin to the steric stabilization of colloids. Polymer chains are either covalently bonded or adsorbed onto a surface.

V. MECHANICAL METHODS

1. **Hydrophobicity, Surface roughness, Surface charge:** The initiation of a biofilm starts with the attachment of suspended cells to a surface. These initial colonizers initially attach with weak and reversible bonds to the surface. If not dislodged promptly, they can establish firmer anchorage using cell adhesion structures like pili. The hydrophobic nature of bacteria also plays a role in their tendency to form biofilms. Some species cannot directly adhere to surfaces and might instead bind to earlier colonizers [17]. On the other hand, certain bacteria face challenges in biofilm development due to their limited mobility. Bacteria that lack motility struggle to recognize surfaces and aggregate as effectively as their motile counterparts. Modifying the surface charge of polymers has proven effective in preventing biofilm formation. By applying electrostatic principles, charged particles repel those carrying similar charges. Adjusting the hydrophobicity and charge of polymeric chains involves various backbone compounds and antimicrobial agents. Positively charged polycationic chains enable molecular extension and provide bactericidal activity [17]. Furthermore, the roughness of a surface impacts biofilm adhesion. Irregular, high-energy surfaces tend to support biofilm growth, whereas smoother surfaces resist biofilm attachment. The surface roughness affects whether contacting substances are hydrophobic or hydrophilic, which in turn affects their ability to adhere [18]. Therefore, it is advisable to maintain smooth surfaces for products that interact with bacteria [18].

VI. STRATEGIES FOR BIOFILM DISPERSAL

It has become essential to increase the effectiveness of biofilm dissolving treatments. Designing new pharmacological therapies depends critically on understanding the role of biofilms in chronic infections and antimicrobial resistance [18]. Traditional antibiotics work by either causing bacterial cell death (bactericidal) or by preventing bacterial cell division (bacteriostatic). While antibiotics have been crucial in the long-term fight against bacterial diseases, evidence suggests that they can seriously damage the host microbiota, fostering the dominance of opportunistic pathogens [3]. Recent developments in techniques try to prevent the formation of biofilms by focusing on bacterial eradication or different stages of biofilm

development [18]. The methods and tactics for preventing the formation of biofilms are described in the discussion that follows.

1. **Bacterial Antibiofilm Polysaccharides:** Polysaccharides, which function as sugar polymers, possess the ability to act as inhibitors of lectins. Lectins are proteins that specifically recognize and bind to sugars without altering their composition. Within bacterial systems, lectins primarily facilitate the attachment of bacteria to host cells. These proteins play a crucial role in the development of biofilms, which are vital for bacterial colonization and subsequent infection. Lectins are typically located on the surfaces of bacterial cells and they engage with glycan substrates on host cells. By competing for the sugar-binding domain of lectins, polysaccharides can hinder the adhesion of pathogens and the subsequent formation of biofilms. Certain types of polysaccharides from plants, microbes, and milk have demonstrated the ability to obstruct various lectins from pathogenic bacteria through a process of competitive inhibition [19]. Polysaccharides also aid in the interactions between cells and surfaces, as well as cell-to-cell interactions, which are crucial for both the formation and stabilization of biofilms. Recent discoveries suggest that specific bacterial exopolysaccharides can inhibit or destabilize the formation of biofilms by other species [19]. The antibiofilm properties of polysaccharides arise from their capacity to: a) alter the physical characteristics of bacterial cells or non-living surfaces, b) function as signaling molecules that influence the gene expression patterns of susceptible bacteria, or c) competitively obstruct multivalent interactions between carbohydrates and proteins, thereby disrupting adhesion.

2. **Anti-biofilm Enzymes:** Enzymes capable of degrading biofilm extracellular matrices could contribute to biofilm dispersion and serve as anti-biofilm agents. An enzyme like N-acetyl-D-glucosamine-1-phosphate acetyltransferase, pivotal in the peptidoglycan and lipopolysaccharide synthesis of Gram-positive and Gram-negative pathogens respectively, is a target for matrix disruption [18]. Employing such enzymes prevented biofilm formation by Staphylococcus and Enterococcus and dispersed preformed biofilms in vitro [18]. Dispersin-B, a glycoside hydrolase, is another example that cleaves β 1–6 N-acetylglucosamine polymers in the bacterial peptidoglycan layer. Dispersin-B treatment proved effective against S. aureus and S. epidermidis biofilms and bacteria [19].

3. **Chelating Agents:** Metal cations like calcium, magnesium, and iron are implicated in maintaining matrix integrity. Consequently, chelating agents have been shown to disrupt biofilm architecture and interfere with bacterial membrane stability. For instance, sodium citrate inhibited biofilm formation by multiple Staphylococci species in vitro [21]. Additionally, tetrasodium-EDTA eradicated biofilms in in vitro models and on explanted hemodialysis catheters, while disodium-EDTA, in tandem with tigecycline or gentamicin, reduced biofilm formation by Staphylococcus species and P. aeruginosa.

4. **Antimicrobial Peptides:** Innate immune responses generate antimicrobial peptides, potential candidates for novel antibiotic development. However, their range of activity and mechanism need further definition before considering them as therapeutic strategies [22]. Recent research focusing on reducing biofilm formation by multidrug-resistant P. aeruginosa strains from cystic fibrosis patients revealed bacterial eradication within

preformed biofilms. Lytic peptides, another group of antimicrobial peptides, have shown inhibitory effects on biofilm formation by binding to lipopolysaccharides, disrupting membrane stability [22].

5. **Anti-adhesion Agents:** Attachment initiates virtually all biofilm formation, motivating several studies on hindering bacterial adhesion. Efforts have centered on preventing assembly of various pili using pilicides, compounds designed to disrupt pilin subunit export. Pilicides reduced in vitro biofilm formation by 50% at concentrations as low as 3 μM [23]. Similar compounds demonstrated effectiveness against curli (curlicides), inhibiting in vitro curli biogenesis and biofilm formation [24].

6. **Nanotechnology:** Nanotechnology techniques encompass altering nanoscale surface topography (nanotopography) and functionalizing surfaces with antibacterial agents, anti-adhesive polymers, or immobilized bactericidal substances. These modifications resist bacterial adhesion, thwart biofilm growth on medical devices and implants, or kill bacteria upon initial surface attachment. It involves electrostatic attraction between charged surfaces and oppositely charged polyelectrolytes, creating multilayered films with thickness ranging from tens to hundreds of nanometers [24].

7. **Disruption of Bacterial Amyloids for Controlling Biofilms:** Numerous bacteria can produce functional amyloid fibers on their cell surfaces. Several bacterial amyloids contribute to biofilm development and community behaviors. For instance, curli are extracellular amyloid fibers generated by Escherichia coli and other Enterobacteriaceae. Certain peptidomimetics inhibit curli biogenesis, with unique anti-biofilm and anti-virulence activities [25]. (Citation in bracket)

8. **Manipulating c-di-GMP Signaling as a Strategy for Dispersing Biofilm Infections:** C-di-GMP, a bacterial second messenger discovered 25 years ago, has emerged as a pivotal player in bacterial communication. It holds significant importance due to its involvement in diverse bacterial lifestyle shifts. For instance, it facilitates the transition from motile to sessile states, enabling the establishment of multicellular biofilm communities. Moreover, it drives the shift from virulent acute infections to less virulent yet chronic infections. Consequently, modulating c-di-GMP signaling pathways within bacteria presents a novel avenue for managing biofilm formation and dispersal in clinical contexts [26-30].

VII. CONCLUSION

The Modern Surge in Medical Devices: Balancing Benefits and Risks in the Battle Against Infections. The modern surge in medical devices brings both progress and challenges, notably in the realm of infections associated with these devices. Despite their contributions to medical advancements, these devices can also significantly contribute to morbidity and mortality rates due to the susceptibility of clinically associated infections. Startlingly, recent statistics reveal a concerning pattern: 95% of urinary tract infections stem from urinary catheters, 65% of pneumonia cases are linked to mechanical ventilation, and intravascular devices are responsible for 87% of bloodstream infections. Among these infections, catheter-related bloodstream infections (CRBSIs) emerge as particularly grave

threats. The complexity of factors involved, including the specter of antibiotic resistance, underscores the ongoing challenge of finding effective methods to eliminate biofilms formed on medical devices. Although current antibiotics target free-floating planktonic cells, they often prove ineffective against biofilms. Nevertheless, the potential exists to both manage and prevent infections associated with medical devices by impeding the formation of biofilms through a variety of anti-biofilm technologies explored above. As we look toward the future, further research should focus on unraveling the intricate interplay between medical devices designed to resist biofilm formation and the bacteria that produce these biofilms. It's essential to assess the stability, specificity, and sensitivity of these innovative medical devices within the human body. Moreover, a comprehensive evaluation of the advantages and disadvantages of emerging anti-biofilm technologies will be pivotal in guiding the path forward.

REFERENCES

[1] De Fuente-Nu´ n ez C, Reffuveille F, Fernandez L et al. Bacterial biofilm development as a multicellular adaptation: antibiotic resistance and new therapeutic strategies. CurrOpinMicrobiol 2013; 16(5): 580–589
[2] Hall-Stoodley L, Costerton JW, Stoodley P. Bacterial biofilms: from the natural environment to infectious diseases. Nat Rev Microbiol 2004;2(2): 95–108.
[3] Yang L, Liu Y, Wu H et al. Combating biofilms. FEMS Immunol Med Microbiol 2012; 65(2): 146–157.
[4] Høiby N, Ciofu O, Johansen HKet al. The clinical impact of bacterial biofilms.Int J Oral Sci 2011; 3(2): 55–65.
[5] Hengzhuang W, Wu H, Ciofu O et al. Pharmacokinetics/pharmacodynamics of colistin and imipenem on mucoid and nonmucoid Pseudomonas aeruginosa biofilms. Antimicrob Agents Chemother 2011; 55(9): 4469–4474.
[6] Hengzhuang W, Wu H, Ciofu O et al. In vivo pharmacokinetics/pharmacodynamics of colistin and imipenem in Pseudomonas aeruginosa biofilm infection. Antimicrob Agents Chemother 2012; 56(5): 2683–2690.
[7] Høiby N, Bjarnsholt T, Givskov M et al. Antibiotic resistance of bacterial biofilms. Int J Antimicrob Agents 2010; 35(4): 322–332. 8 Høiby N. Recent advances in the treatment of Pseudomonas aeruginosa infections in cystic fibrosis. BMC Med 2011; 9: 32
[8] Tomas B, Oana C, Molin S, Michael G, Niels H (2013) Applying insights from biofilm biology to drug development -can a new approach be developed? Nature Reviews Drug Delivery 12: 791-808.
[9] Fletcher M, Loeb GI. Influence of substratum characteristics on the attachment of a marine pseudomonad to solid surfaces. Appl Environ Microbiol 1979; 37:67–72.
[10] Pringle JH, Fletcher M. Influence of substratum wettability on attachment of freshwater bacteria to solid surfaces. Appl Environ Microbiol 1983; 45:811–7.
[11] Characklis WG, McFeters GA, Marshall KC. Physiological ecology in biofilm systems. In: Characklis WG, Marshall KC, eds. Biofilms. New York: John Wiley and Sons, 1990:341–94.
[12] Quirynen M, Brecx M, van Steenberghe D. Biofilms in the oral cavity: impact of surface characteristics. In: Evans LV, ed. Biofilms: recent advances in their study and control. Amsterdam: Harwood Academic Publishers, 2000:167–87.
[13] Korber DR, Lawrence JR, Sutton B, et al. Effect of laminar flow velocity on the kinetics of surface recolonization by Mot and Mot Pseudomonas fluorescens. MicrobEcol 1989; 18:1–19.
[14] Rosenberg M, Bayer EA, Delarea J, et al. Role of thin fimbriae in adherence and growth of Acinetobactercalcoaceticus RAG-1 on hexadecane. Appl Environ Microbiol 1982; 44:929–37.
[15] Christensen GD, Baldassarri L, Simpson WA. Colonization of medical devices by coagulase-negative staphylococci. In: Bisno AL, Waldvogel FA, eds. Infections associated with indwelling medical devices, 2nd ed. Washington, DC: American Society for Microbiology, 1994:45–78.
[16] Murga R, Forster S, Brown E, Pruckler J, Fields B, et al. Role of biofilms in the survival of Legionella pneumophila in a model potable-water system. Microbiology , 2001,147: 3121–6.
[17] Jansen B, Kohnen W, Prevention of biofilm formation by polymer modification. J Ind Microbiol,1995 15: 391-6.
[18] Meiron T, SaguyI , Adhesion Modeling on Rough Low Linear Density Polyethylene. J Food Sci,2007;72: E485–91.

[19] Maria K, Maria H, Scott J , Bacterial Biofilms: Development, Dispersal, and Therapeutic Strategies in the Dawn of the Postantibiotic Era. Cold Spring Harbor Laboratory Press.2014
[20] Kaplan JB Biofilm dispersal: Mechanisms, clinical implications, and potential therapeutic uses. J Dent Res,2010; 89: 205–18.
[21] Shanks R, Sargent J, Martinez R, Graber M, O'Toole G, Catheter lock solutions influence staphylococcal biofilm formation on abiotic surfaces. Nephrol Dial Transpl,2006; 21: 2247–55.
[22] Kharidia R, Liang J ,The activity of a small lytic peptide PTP-7 on Staphylococcus aureus biofilms. J Microbiol,2011 49: 663–8.
[23] Berg V, Das P, Chorell E, Hedenstrom M, Pinkner JS, et al. Carboxylic acid isosteres improve the activity of ring-fused 2-pyridones that inhibit pilus biogenesis in E. coli. Bioorg Med ChemLett, 2008;18: 3536–40.
[24] Cegelski L, Pinkner J, Hammer N, Cusumano C, Hung C, et al.Small-molecule inhibitors target Escherichia coli amyloid biogenesis and biofilm formation. Nat Chem Biol,2009; 5: 913–9.
[25] K.G. Neoh, R. Wang, E.T. KangSurface nanoengineering for combating biomaterials infectionsBiomaterials and Medical Device - Associated Infections, 2015, Pages 133-161
[26] Romling U, Galperin MY, Gomelsky M. Cyclic di-GMP: the first 25 years of a universal bacterial second messenger. MicrobiolMolBiol Rev 2013; 77(1): 1–52.]

