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Review Article on Bacterial Biofilm Production and Ant Biofilm Approaches

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Abstract

Matrix-enclosed populations of bacteria known as Biofilms, stick to one another or to the different surfaces or interface. This definition comprises floccules, adherent populations and microbial clumps within the porous media having pore spaces. This ability of biofilm formation is a special property of bacteria. Multicellular communities existing in the natural environment known as biofilms are unique and have architectural features by interstitial voids such as micro and macro colonies. Biofilms are basically an ordered aggregate of microorganisms living within their self-produced extracellular matrix and attach to the surfaces irreversibly but these aggregates are not easy to remove unless rinse quickly. In the attachment stage of biofilm to the surface, formation of (EPS) extracellular polymeric substances occurs. Phylogenetic history of different related biofilms can be find by using different computational techniques like tree viewer and quorum sensing. Biofilm formation is not a good thing in many ways so there must be some ways to stop the formation of biofilm so there are some ant biofilm approaches and by using them we can stop the growth of biofilms. Some of these techniques include a tamers, enzyme treatment, nanoparticles, photo-dynamic therapy anti adhesion approaches etc. The development of ant biofilm agents against different microbial targets and their subsequent application as adjuvants with antimicrobial agents seems to be more efficient.

Keywords: Biofilm; Extracellular polymeric substances; Ant biofilm Approaches; Adhesion; Anti-adhesion; Phylogenetic history; Quorum sensing; Photo dynamic therapy; Adjuvants

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Introduction

Biofilms can be defined as aggregates of microbes held together in their self-made extracellular matrix. Biofilms are present in the natural environments cling to each other or to the surfaces [1].

Since the first description about bacterial biofilms, their real importance has gradually emerged and the first recognition of the ubiquitous nature of biofilms. During the fifteen decades, ensuing the discoveries of Louis Pasteur. It has come to be interestingly clear that the biofilms possessed a noticeably different growth phase of bacteria that is extremely diverse from the planktonic growth phase being studied so diligently.

Bacterial cells change their phenotypes during the complex process of adherence in response to the proximity of a surface. Sessile bacteria throughout the initial stages in formation of biofilm find themselves in unchanging vicinity with cells of the same species and of other species as single and assorted species micro colonies are then formed. The vivacious extra polysaccharide matrix production in the emerging biofilm and the cellular juxtapositions are the conditions for a micro-environment of every biofilm bacterium. Many biofilm bacteria react to their unique specific micro environmental conditions showing different patterns of growth and a structurally complex mature biofilm gradually develops accordingly. A major factor responsible

in shaping the structure of a biofilm and in forming the ultimate associations which will form the mature biofilms well organize enough to attach to the surface is physiological cooperatively [2].

Biofilm Formation; Characteristic of Bacteria

Biofilm formation is a characteristic property of bacteria. Biofilms constitute multicellular colonies of microorganisms that are joined together through a matrix. The mechanisms underlying in the formation of biofilms vary for different bacteria that solely depends on different environmental circumstances and strain specificity. In my review, I have emphasized on 4 well-known model organisms so that an overview about how different organisms are involved in biofilm formation can be given: Pseudomonas aeruginosa, Staphylococcus aureus, Escherichia coli and Bacillus subtilis. These bacteria are used as an example to argue the salient charactristics of biofilm formation and the process involved in these when extracellular signaling activates them. Formation of biofilm can influence humans in different ways as they form in natural, industrial and medical settings. For example, biofilm development of medical devices such as catheters or implants that often results in difficulty in order to treat chronic infections [3]. Besides this, infections have been related with biofilm formation on human surfaces such as skin, urinary track and teeth. In spite of that biofilms on human surfaces are not always harmful. For instance, dental plaque biofilms consists of dozens of species and their composition immediately inform either the disease is present or absent.

There is a progression of colonization in dental plaque but the presence of valuable species opposes settlement by harmful organisms. But biofilms are ubiquitously found universally. Like, biofilms are formed on the hull of ships and inside pipes also where they cause severe damage. Biofilms are also formed in many natural settings but there they allow mutualistic symbioses. For example in order to allow ants to maintain pathogen free fungal gardens, the Actinobacteria often grow on ants. There a large number of welfares and damages that a biofilm can converse, so it is important for us to know that how bacteria develop in these communities. From biofilm formation, a bacterial community may get a number of benefits. Biofilms provide confrontation to many anti-microbial and protection against host defenses [4].

Within the biofilm arise in the percentage of persisted cells appears to be one of the possible reasons behind the increased resistance against environmental stresses. Persisted cells are non-dividing and are resistant to many antibiotics even though of the fact that being hereditarily alike to the rest of the population. Persisted cells are supposed to be protected from the antibiotic actions because they express such toxin antitoxin systems which through their toxin modules block the target of antibiotics. In addition to persisters, presences of extracellular matrix provide protection to constituent cells from external harms. These extracellular matrices also provide diffusion barrier to small molecules. In relation to this, some of the cells in bacterial community are metabolically inactive due to slower diffusion of vitamins, nutrients and co factors in biofilms.

Bacterial Growth Rate

The fact that in a biofilm if cells are confined to a limited space that will influence the bacterial growth rate (Stewart and Franklin). This condition is very much similar to the stationary phase that is produced in laboratory conditions. Hence, biofilm formation basically represents a natural stationary stage of bacterial growth. With the increase making of secondary metabolites such as pigments, antibiotics and other small molecules, bacteria greatly change their physiology during stationary phase. Secondary metabolites function as signaling molecules either to start the biofilm formation procedure or to inhibit it by other organisms that live in the same habitat.

Biofilm constitutes a complex assembly of DNA, protein and polysaccharide in their self-produced EPM (extracellular polysaccharide matrix) and was naturally found on various surfaces including living tissues, potable water system or natural aquatic, medical devices etc. Bacterial biofilms are well studied in avoiding phagocytosis, antibiotics and other disinfectant components. The interstitial voids such as micro and macro colonies found in biofilms allowed the diffusion of gases, antimicrobial agents and nutrients through the biofilms; however, biofilm changes their architecture in response to these changes occurring in the internal and external process. As the cells are in proximity, they exchange their extra chromosomal plasmid, their quorum sensing molecules and show distinct character in each biofilm community [5].

Despite of all the detailed studies on architectural features of biofilms, its composition, mechanisms, benefits and detriments, our review specifically focuses on the steps that are involved in Biofilm formation and Antibiofilm approaches in detail.

Formation of Biofilm

Composition of microbial biofilm

Biofilm is a structured mass of microbes which live in an extracellular matrix and attached with the living surface irreversibly. The development of EPS (Extracellular Polymeric Substances) occurs during the attachment phase of a biofilm to the surface. This EPM provide stability to the biofilm by giving power to the communication of microbes Usually the thickness of EPS matrix is in between 0.2 to 1.0 µm, although size of the biofilm does not out go from 10-30nm. Almost 5 to 35% of the volume of a biofilm is consumed by microbes and the rest of volume if of extracellular matrix. Protein is the main composition of this EPS Through the searching system, some of the useful essential minerals and nutrients are captured from the surrounding environment. Different percentages of biomolecules make up the composition of EPM such as protein is having majority almost greater than 2% , other constituents such as polysaccharides are about 1 to 2% , DNA molecules about less than 1% , RNA less than 1% , ions either free or bound and the rest is water i.e. 97%. Due to the water content in the biofilm, flow of nutrients is possible

within it.

Steps in Formation of a Biofilm

Formation of biofilm occurs in many steps as per genetic studies. Quorum sensing between the cells of microorganisms is a special type of signaling and it is a must requirement for the biofilm formation. In comparison with planktonic forms of the same microbes, transcription of different set of genes are required. The viscous and elastic features of extracellular matrix attribute mechanical stability to a biofilm.

Formation of Biofilms is a difficult procedure but it happens in few common steps according to different researcher's i.e.

1. Attachment to the Surface or Initial Contact

2. Formation of Micro-colony

3. Architecture and Maturation of Biofilms

4. Dispersion or Detachment of the biofilm

Attachment to the surface or initial contact

As this is the initial step in formation of biofilm it requires attachment with a surface, microbial cells get attached to the surface with the help of their adjuncts like pilli and flagella. They may get attached to the other physical forces like electrostatic interactions or vander Waal's forces etc. One cause for growth and attachment of microbes in a biofilm is solid-liquid interface.

Formation of micro-colony

Afterward the stable attachment phase of microbes to a living or a nonliving surface, microbial cells start the procedure of division and multiplication which is introduced through specific chemical signaling occurring within the Extracellular polysaccharide matrix. This will ultimately lead to the micro colonies formation. In a biofilm, bacterial colonies are of many micro communities and these communities interact with each other in multiple ways .In distribution of important metabolic products, exchange of substrate and elimination of the metabolic end-products, this microbial coordination plays a vital role. For example throughout the anaerobic digestion, the complex organic matter is converted to methane and carbon dioxide, 3kinds of bacterial association is required i.e.

I. From organic compounds production of acid and alcohol is start by the fermentative bacteria while depending upon the dissimilation of complex organic compounds.

II. Acetogenic bacteria then consumed these as their substrates.

By converting the acetate, hydrogen and carbon dioxide into methane, methanogens get energy. For the development of a syntrophic association, the need of a complete environment is being fulfilled by biofilm in this phase, with the help of auto-inducer signals microbial cells coordinate with each other. To achieve required microbial density cell-cell coordination is an essential procedure. This coordination ultimately leads to excretion of auto inducers, the signaling molecules. Quorum sensing is facilitated by these signaling molecules At this stage of biofilm maturation, in order to form EPS, expressions of certain gene products are required. Since EPS maintains the 3D structure of a biofilm, then interstitial spaces are being formed within the matrix. To eradicate the leftover from the populations of micro colonies and to distribute essential nutrients among the communities of a biofilm, a circulatory system is required and to accomplish that purpose these channels are occupied with water.

Architecture and maturation of a biofilm

In this phase, with the help of auto-inducer signals microbial cells coordinate with each other. To achieve required microbial density cell-cell coordination is an essential procedure. This coordination ultimately leads to excretion of auto inducers, the signaling molecules. Quorum sensing is facilitated by these signaling molecules. At this stage of biofilm maturation, in order to form EPS, expressions of certain gene products are required. Since EPS maintains the 3D structure of a biofilm, then interstitial spaces are being formed within the matrix. To eradicate the leftover from the populations of micro colonies and to distribute essential nutrients among the communities of a biofilm, a circulatory system is required and to accomplish that purpose these channels are occupied with.

Dispersion or depression stage of a biofilm

In the detachment stage, in the biofilm the microbial cells quickly multiply and disperse in order to change from sessile to motile form. Then dissociation happens in a usual phenomenon. Bacterial cells of some bacteria directly disperse into the environment because these kinds of the bacteria do not form EPS but motorized pressure might too get involved in this procedure. Within the biofilm throughout the dispersion stage the microbial colonies release diverse sacchrolytic enzymes in order to discharge the superficial of microorganisms to a novel extent for the settlement purpose.

For instance *P. flouresence* and *Pseudomonas aeruginosa* yield alginate lyase, Escherichia coliproduce N-acetyl-heparosanlyase and S. equi yield shyaluronidase for the breakdown of Extracellular polysaccharide medium and later dispersion. In this stage up regulation of the manifestation of some proteins is carried out by microbial cells because these proteins are essential to flagella formation which will ultimately allow the bacteria to transfer into a novel place. Detachment of the microbial cells and then move to a new place will be helpful in scattering of the infections.

Anti-Biofilm Approaches

Antibiofilm approaches include the natural and induced process that leads to lessen the bacterial biomass through modifications done in biofilm formation durability and or quality. Antibiofilm approaches can target either the adhesion stage of biofilms or mature biofilms. Biological response to a biomedical device solely depends on the structure and the surface capabilities of the material used in it and most likely device-associated infections are originate from surface material contamination at time of

implantation. Thus, the surface capabilities or compositions of biomaterials are modified in order to achieve appropriate results. To magnify functionality and biocompatibility, surface engineering of materials can be done which ultimately reduce the microbial contamination and put a stop to biofilm infections

Aptamers

Aptamers consists of single stranded RNA or DNA sequences that can specifically bind to their targets and often inhibit them .Very rare studies have examined the aptamers as anti-biofilm agents. In an approach to block the flagella motility as a promising strategy to hinder biofilm formation, developed a single stranded DNA aptamer that specifically targeted S. Choleraesuis flagellin protein. The characterized aptamer inhibited the early attachment by restricting cellular aggregation and production of mature biofilms. Moreover, flagellin aptamer demonstrated synergistic effect with ampicillin antibiotic. Further upgraded the flagella targeting aptamer by linking it with ampicillin. The conjugate had a distinctive antibacterial activity and higher antibiofilm activity when compared to those when either component were applied separately. The aptamer is thought to ensure facilitated entry of ampicillin into the biofilm which decreased its tolerance to the antibiotic [6].

Conclusion

Biofilms are defined as microbial communities stick with one another or to various surfaces also entrenched within a self-produced extracellular matrix. This also includes floccules, adherent populations and microbial aggregates within the porous media. Bacterial sample can be taken from different sources like soil sewage water etc. and can check the aggregation of different bacterial colonies and can check the formation of biofilm. We can check the formation of biofilms through different sources; we can perform different physical and chemical tests to check the formation of different biofilms. Relation of different biofilms can be finding out by using different computational techniques like tree viewer in which we can draw a phylogenetic tree which will assist to find out the relation between different bacterial biofilms. Quorum sensing is a novel technique which can also be used to find the evolutionary history. Biofilm formation is not a good thing in many ways so there must be some ways to stop the formation of biofilm so there are some antibiofilm approaches and by using them we can stop the growth of biofilms. Some of these techniques include aptamers, enzyme treatment, nanoparticles, photo- dynamic therapy anti adhesion approaches etc. The development of antibiofilm agents against different microbial targets and their subsequent application as adjuvants with antimicrobial agents seems to be more efficient.

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