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Biofilm inhibitor and its inhibition mechanism: Plant derived agents

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Abstract:

Bacteria in the natural environment form complexes which are closely associated with abiotic and biotic surfaces of solids and liquids. These bacterial communities are adherent to a surface, an interface or

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to each other and are known as biofilms. Biofilm formation has serious implications in industrial, environmental, public health and medical situations because they express biofilms-specific properties such as increased resistance to antibiotics, UV light and chemical biocides, increased rates of genetic exchange, altered biodegradability and increased secondary metabolite production. In the clinical context, it is estimated that about 60% of all microbial infections involve bacterial biofilms. The occurrence of biofilms in food processing environments can cause post-processing contamination leading to lower shelf life of products and transmission of diseases. The ability to control biofilm growth is an important issue because of health problem and economic losses. Researchers are trying to find green product rather than synthetic one that can be used as generally recognized as safe (GRAS). Plants produce an enormous number of natural products. Many plants extract contain phenol derivatives, terpenes, flavonoids etc. which have ability to suppress the microbial cell attachment and biofilm growth. The aim of this mini review is to combine most of the recently published work on biofilm inhibition (plant based inhibitor) and figure out the inhibition mechanism which can be safely used in food safety and human health issues.

Key words: Biofilm, extract, plant, quorum sensing

1. Introduction

Bacterial growth is characterized by two life forms, one being as single cells (planktonic) and the other being in sessile aggregates. The latter is commonly referred to as the biofilm mode of growth. Many more or less divergent definitions of bacterial biofilm exist; all agreeing that biofilms are multiple bacteria in an aggregate. The different definitions present in the literature differ mainly in whether the cells have to be attached to a surface or whether the bacteria exist in a structured community. As for medical, food microbiology and biofilm applications, the topic of this review, biofilm is defined as: A coherent cluster of bacterial cells imbedded in a matrix –

which are more tolerant to most antimicrobials and the host defense, than planktonic bacterial cells (Bjarnsholt et al. 2011). In most biofilms, the microorganisms account for less than 10% of the dry mass, whereas the matrix can account for over 90%. The matrix is the extracellular material, mostly produced by the organisms themselves, in which the biofilm cells are embedded. It consists of a conglomeration of different types of biopolymers - known as extracellular polymeric substances (EPS) - that forms the scaffold for the three-dimensional architecture of the biofilm and is responsible for adhesion to surfaces and for cohesion in the biofilm. The formation of a biofilm allows a lifestyle that is entirely different from the planktonic state. Although, the precise and molecular interactions of the various secreted biofilm matrix polymers have not been defined, and the contributions of these components to matrix integrity are poorly understood at a molecular level, several functions of EPS have been determined, demonstrating a wide range of advantages for the biofilm mode of life. The biofilm matrix protects organisms against desiccation, oxidizing or charged biocides, some antibiotics and metallic captions, ultraviolet radiation, many (but not all) protozoan grazers and host immune defenses (Flemming et al, 2010). On the other hand, biofilms represent great benefits in biotechnology industries because of their self- immobilization with high concentration of biomass within EPS that provide the high resistance to toxic compounds, long term activity which all facilitate continuous process with the high stability. Biofilm reactor can be set as continuous reactor operated with more cost effectiveness than batch process of free cell because of the reduction in reactor preparation, cell growth and product recovery. Biofilm reactors represent the significant advantage where in the reactor capacity obtained by using free cells is limited by biomass concentration. Therefore, biofilm reactor has been considered to be used in the industry for various economic

reasons. Biofilms of bacteria, fungi and their enzyme products can act as biocatalysts to provide high specificity productions under the mild condition. Various biofilm processes have been implemented commercially with the great success over the last few decades (Qureshi et al., 2005). Biofilms have been used in the food sector in the productions of various value added product organic acid (acetic acid, lactic acid, succinic acid and fumaric acid), polysaccharide, ethanol, butanol. Many microorganisms are capable to develop single species biofilm that are applicable for bioprocess. Mostly the biofilm can be form spontaneously under suitable condition specific for each microbe. Biofilm growth in a multi-stage process involving initial cell attachment to a solid surface and follow by surface adhesion by self produced EPS. Surface properties play an important role in the cell attachment (Goller et al, 2008). The primary method used for cell immobilization is organism κ-entrapment within polymers such as calcium alginate, carrageenan and membrane. Recently, self-immobilized cells and cells that grow as aggregates without the addition of any polymers or cross- linking chemical have been used as biofilm reactors. Biofilm in the fermentation processes can be maintained in the reactor as biofilm reactor in which the cell recycle can be processed without the need for re-inoculation of the culture. The reactors can be manipulated in variety of configurations including batch, repeated batch, continuous stirred tank, fluidized bed, air lift. In order to use biofilm reactor, it is necessary to identify whether the desirable microorganism using in the process can develop biofilm on the supporter otherwise, biofilm reactor cannot be applied for the bioconversion. Mostly single species biofilm has been widely used in the production (Qureshi et al., 2005).Biofilm reactors have been applied for the conversion of agricultural materials such as starch, sugars and glycerol to various alcohol such as ethanol, butanol, 2,3- butanediol) or even organic acids such as

acetic acid, fumaric acid and citric acid). *Zymomonasmobilis* biofilm has been efficiently used for ethanol production in polypropylene packed bed reactor in which the production rate was 536 g/Lh while 5 g/Lh was obtained from continuous free cell culture (Kunduru et al, 1996). Acetic acid bacterial biofilm was grown on beech wood shaving that was used to convert ethanol to vinegar to produce final acetic acid concentration of 120 g/L had been obtained through the biofilm packed bed process. Biofilm packed bed reactor of *Lactococcuslactis*on cotton fabric was applied to produce nisin (Liu, 2005). Biofilm processes involving toxic compounds in the substrate have been recently focused, *Z. mobilis* biofilm was found to be tolerated to toxic substrate benzaldehyde than free cells in continuous biofilm reactor (Li et al., 2006). The fluidized bed biofilm reactor (FBBR) have been effectively used more than two decades for treating industrial wastewater in which the biofilm was found to be more resistant to the toxic chemical in the waste than free cells (LaPara et al., 2001; Todhanakasem, 2013). But with other infectious diseases, food safety is a vital public-health concern that connects human health to farming and other areas of food production. Iwamoto et al. (2010) stated that foodborne pathogenicity is a major cause of the worldwide morbidity and hospitalizations that result from consuming various foods, including seafood. Currently, 31 organisms are recognized as foodborne pathogens, and recent statistics released by the United States Centers for Disease Control and Prevention (CDC) indicate that approximately 48 million foodborne illnesses occur annually in the USA alone, resulting in 128,000 hospitalizations and 3000 deaths (CDC, 2012;Mizan et al. 2015). There are many biofilm inhibitor in market. Most of them are synthetic compounds. So, researchers are trying to find less toxic and more specific natural products. The aim of this mini review is to figure out the plant derived (green compounds) biofilm inhibitor and study on their mode of action

based on recent published articles from online (google, pubmed, academia, researchgate, baidu etc.) search.

2. Mechanism of biofilm formation : How biofilms are formed

The transition of microorganisms from the planktonic to the sessile (biofilm) state is often described as occurring in a series of steps or phases. The stages of biofilm formation include: i) the formation of a conditioning film; ii) cellular attachment; iii) the formation of microcolonies which eventually merge to become mature biofilms; and iv) biofilm dispersion and recolonization; duration : sec - days (**Figure 1**).

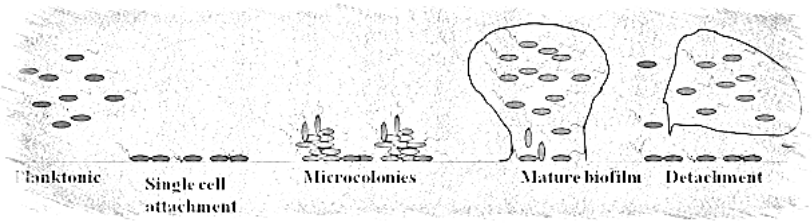


Figure 1 : Stages of bacterial biofilm formation (planktonic-single cell attachment- microcolonies- mature biofilm- detachment) (Todhanakasem, et al, 2013)

This dynamic process depends on the characteristics of the growth medium and the bacterial cell surface, and this process can be affected by various environmental stimuli such as temperature and p^H. Compared to planktonic cells, biofilm cells are more resistant to high temperature and low p^H. Biofilms are also highly resistant to sanitizers (Healy et al., 2009), environmental stresses such as starvation (Spector et al, 2012), desiccation (Gandhi et al, 2007), disinfectants, dry surfaces, and antimicrobial agents. As such, decontamination of biofilms from food products, such as fresh produce, can be challenging. Meanwhile, it is widely accepted that nearly all materials that

are commonly used in food processing, including buna-n rubber (acrylonitrile butadiene) and stainless steel, can support biofilm formation, and Myszka et al. (2011) reported that eliminating bacterial colonization from food-processing equipment is extremely challenging. To prevent biofilm development, food-processing equipment is therefore designed to feature highly polished and smooth surfaces that hamper bacterial adhesion, the first step in biofilm formation. Thus, a comprehensive understanding of this process is required for ensuring that food-industry products are of high quality and are free from microbial contamination. Biofilm adhesion and maturation can be enhanced by treatment with alcohol as a disinfectant, by the presence of NaCl in food substrates or it contains other bacterial species. McDougald et al. (2006) demonstrated that at low and high salinities, *V. vulnificus* exhibited no marked variation in biofilm formation. In contrast, biofilm formation of *V. parahaemolyticus* is highly responsive to environmental factors such as salinity, pH, temperature differences, and the presence of organic matter (Kalburge et al., 2014; Mizan et al. 2015). From Fig. 2, we got a real formed biofilm under CLSM (confocal laser scanning microscopy)observation.

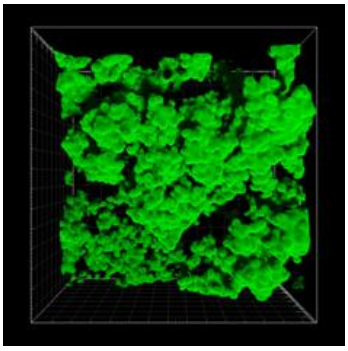


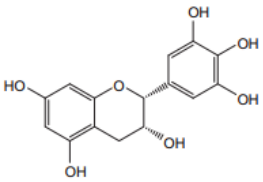
Figure 2: **CLSM of *S. cerevisiae* biofilm (http://www1.bio.ku.dk/english/research/fys/regenberg/research/)**

3. Plant derived agents

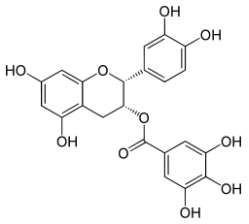
Plants produce an enormous number of natural products. Many of them have been used by people in the form of whole plants, their parts or extracts since ancient times. Despite the long-term research, plants seem to be an inexhaustible treasure of new compounds in the present. Extracts of many plants contain components (phenol derivatives, terpenes, etc.) that have an ability to suppress microbial cell attachment and a biofilm growth. Anti-biofilm activity of green tea polyphenols was also demonstrated on attached pathogenic yeast *C. albicans* , EGCG being more effective than epigallocatechin (Structure 1) or epicatechin-3-gallate (structure 2). This study suggests that the metabolic instability produced by the catechin-induced proteasome nactivation was a contributor to the decrease in the growth rate constant as well as biofilm formation and maintenance (Rˇezanka et al. 2012).

Zeng et al. carried out an analysis of 51 active compounds used in traditional Chinese medicine. Five of them had a proven ability to inhibit biofilm formation, the flavonoid baicalein (structure 3) being the most effective. This substance is contained, for example, in Oroxylum indicum or in the roots of Scutellaria baicalensis. Baicalin, the glucuronide of baicalein, has significant antibiofilm activity against *Burkholderia cenocepacia* or *B. multivorans* (Brackman, 2009). Many flavonoids are prominent secondary metabolites present in citrus species. So naringenin (structure 4), kaempferol , quercetin (structure 5), and apigenin (structure 6) have shown the ability to inhibit biofilm formation in *Vibrio harvey* and *E. coli* O157:H7.

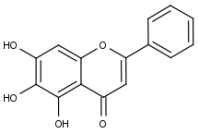
structure 1



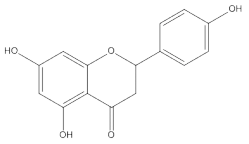
structure 2



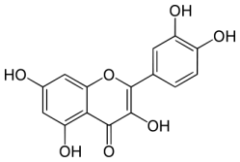
structure 3



structure 4



structure 5



structure 6

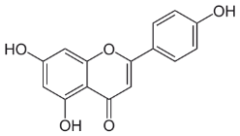


Table 1 is giving an over view of bacterial biofilm inhibition mechanism. From this table it is clear that biolim is mostly controlled by QS (quorum sensing). In recent years, drug resistance of human pathogenic bacteria has been extensively reported. Moreover, persistent infections were also observed due to improved resistance of bacteria in biofilm. This creates a tremendous economic loss and pressure on the medical community to find alternative approaches for the treatment of diseases related with biofilms. Therefore, efforts are been applied to discover efficient antimicrobial molecules not so vulnerable as current drugs to bacterial resistance mechanisms, including those in biofilms. Some natural products have distinctive properties that make them perfect candidates for these much needed therapeutics. Plants produce an enormous array of secondary metabolites (phytochemicals) that are not essential for their normal physiological functions. However, these bioactive compounds are used to protect plants

against attacks of microorganisms, herbivores, insects, nematodes and even other plants. The importance of diverse natural product has been recognized by humans due to their beneficial properties for health. Inclusively, many classes of plant secondary metabolites have demonstrated their potential as antimicrobials or synergists of other products. So, nowadays phytochemicals are a fundamental source of chemical diversity and important components of the current pharmaceutical products(Dixon, 2001; Prior et al. 2000; Kubov et al.2006; Newman et al. 2007; Saavedra et al. 2010; Borges et al. 2013). Crude extract, essential oils, phenolics, isothiocyanates etc. are mostly used.

Table 1 : Different plants as biofilm inhibitor and the mode of action

Plant name	Biofilm of	Type of Solvent	Mode of action	Ref.
Equisetum arvense, Herniariaglabra, Galiumodoratum, Urticadioica, Vacciniumvitisidaea Betulapendula,	Uropathogenic *Escherichia coli* rods	Water	anti-biofilm effect of plant extracts can be caused by modifications in the bacterial surface structures responsible for binding to the occupied surface.	Wojnicz et al. 2012
Euphorbia hirta L.	*Klebsiella pneumonia, Pseudomonas aeruginosa, Salmonella typhi, Shigelladysenteriae, Enterobacteraerogens, Escherichia coli, Enterococcus faecalis, Proteus mirabilis, Proteus vulgaris, Bacillus subtilis and Bacillus cereus*	Methanol	The most abundant phyto compound found was terpenoids. Probably the terpenoids influenced the membrane integrity in all organisms and helped to eradicate most biofilm cells. The significant reduction in cell attachment made terpenoids an ideal anti-adhesive compound. Terpenes have also been frequently reported to be active against bacteria. Additionally, the expression of synergy, antagonism or additive effects among the major phytocompounds found in the crude extract may also be the rationale to the apparent anti-biofilm activity.	Perumal et. al. 2013
Moringaoleifera, Murrayakoenigii, Psidiumguajava, Eclipta prostrate, Phyllanthusfraternus	*Streptococcus mutans,Streptococcusmitis*	Hot and cold extractions using ethanol, water and aqueous-ethanol (1:1) as solvents	*E. prostrata, M. koenigii and P. guajava* are rich in flavonoids, tannins and terpenes, which may be responsible for observed antibacterial activity against oral bacteria.	John et. al 2013

*Azadirachtaindica, Vitexnegundu ,Tridaxprocumbens,Ocimumtenuiflorum*i	Clinical isolate of *E.coli*	Methanol (chitosan coated extracts)	The increased antibiofilm effect of chitosan coated plant extracts may be due to the inhibition of exopolysaccharide synthesis limiting the formation of biofilm or due to diffusion of CS-plant extracts through the channels present in the biofilms followed by the sustained release of phytochemicals in the respective plant extracts which may then impart antimicrobial function.	Smitinont et al. 1999, Namasivayam et al., 2013
*Hibiscus sabdariffa*calyx	*Streptococcus pneumonia, Pseudomonas aeruginosa*, *Escherichia coli*, *Staphylococcus aureus*	petroleum ether, ethanol (70 %)	These observations revealed that phenolic compounds at concentration that did not or weakly inhibit bacterial growth increased biofilm formation.	Plyuta V et al. 2013, Mutalib et. al., 2015
Fifty four (54) Indonesian medicinal Plants	*Pseudomonas aeruginosa* PAO1, *Staphylococcus aureus* Cowan I	Ethanol	Biofilm formation can be controlled by quorum sensing(QS), a bacterial communication system which causes a rapidly and coordinately change of expression pattern in the bacterial population in response to population density.	Pratiwi et al. 2015
Duabangagrandiflora	methicillin-resistance *Staphylococcus aureus*(MRSA)	Hexane (He), ethyl acetate (EA), and ethanol (EtOH)	The major classes of phytochemicals detected, which include alkaloids, tannins, saponins, steroids, glycosides and flavonoids, are known to be associated with antibacterial effects. For instance, flavonoids are known to affect the sortase activity, which is known to influence the adhesive property of bacterial cell wall, leading to interruption of biofilm development.	Saleem et. al. 2010, Ozçelik, et. al. , 2006,. Cushnie, et. al 2011, Santiago et al. 2015,
S-substituted cysteine sulfoxides and their corresponding disulfide derivatives	*Pseudomonas aeruginosa* PAO1		The inhibition of planktonic growth and biofilm formation are decoupled for those compounds, and this inhibition does not rely upon strict biocidal activity.	Cady et al. 2012
cinnamaldehyde, carvacrol, thymol, eugenol, ß resorcylic acid and caprylic acid	*Listeria monocytogenes*		They prevent biofilm production from planktonic cells by down-regulating critical biofilm-associated genes, whereas bactericidal concentrations eradicate pre-formed biofilms by directly killing *L. monocytogenes*in biofilms.	Upadhyay, 2014
Kramerialappacea, Aesculushippocastanum, Chelidoniummajus(Cheleryth rine, Sanguinarine, DiHydroxyBenzoFuran, proAnthocyanidin A2-phosphatidylCholine)	*Staphylococcus aureus , Staphylococcus epidermidis*		These compounds are able to inhibit the biofilm formation through a mechanism different from mere killing bacterial cells in the planktonic form. Proteomic experiments showed the treatment downregulates some important proteins belonging to different pathways. Of note is that even if there are several cell surface proteins affected, the vast majority are cytoplasmic proteins. Thus hinting that these compounds are cell penetrating and that they are likely to affect intracellular processes.	Artini et al. 2012
Ellagic acid, Tannic acid	*Escherichia coli.*		The very different effects of the plant compounds on the knockout mutants	Correspondence , 2010

			suggest that they act by different mechanisms on biofilm formation. Plant secondary metabolites, to a large extent polyphenols, show bactericidal effects as a result of damage to the cell membrane	
498 plants (331 genera, 466 species, and 20 different parts of plants)	*Escherichia coli* O157:H7	Methanol	Carbon flux, cyclic di-GMP, intercellular signal molecules (autoinducer-2, indole), and other global regulators are known to be involved in the formation of biofilm of *E. coli*.	Lee et. al., 2013
Terminaliacatappa	*Staphylococcus aureus, Pseudomonas aeruginosa*	Methanol	By controlling Quorum Sensing systems that control a number of genes involved in biofilm formation and production of virulence factors.	Tagannanet al. 2011
Rosa rugosa tea polyphenol (rich in polyphenols (87.52%) and flavonoids (61.03%))	*Escherichia coli* K-12, *Pseudomonas aeruginosa* PAO1		Biofilm inhibition activity of *Rosa rugosa* was not due to the antibacterial effect but also for by inhibiting quorum sensing	Zhang, J. et al. 2014
curcumin from *Curcuma longa* (turmeric)	*Escherichia coli*, *Pseudomonas aeruginosa* PAO1	Methanol	Development of these biofilms is based on the signal-mediated QS system. An interference with QS may prevent the development of uropathogenic bacterial biofilms	Packiavathy et al. 2014
wheat-bran	*Staphylococcus aureus*	Demineralized water	Proteases can be an endogenous tool of *S. aureus* to remodel its own biofilm structure, they can also become a way to destroy its biofilm. Recent strategies for controlling biofilm growth are based on the degradation of the matrix using enzymatic treatments. It would therefore be plausible that endogenous proteases in wheat bran could be related to the inhibition and destruction of biofilm.	González-Ortiz. et al. 2014
522 Asian medicinal plant extract (*Carexdimorpholepis), C. pumila extract, P. lactiflora*	*Pseudomonas aeruginosa, Escherichia coli* O157:H7	Methanol	It enhanced the biofilm, from the ecological perspective, it is likely that *P. aeruginosa* has developed a defense system against plant source agents that allows it to form more biofilms, which is similar to *P. aeruginosa* inducing its biofilm formation in the presence of sub inhibitory concentrations of aminoglycoside antibiotics	Cho, et al, 2013
Melastomataceae, Meliaceae, Piperaceae, Lepidobotryaceae, Sapindaceae, Simaroubaceae.	*Pseudomonas aeruginosa*PA14	Ethanol	Tropical plants contain phytochemicals capable of interfering with bacterial quorum sensing and biofilm formation.	Ta , 2015
Burdock (*Arctium lappa* L *Asteraceae*)	*S. aureus*	Ethanol	Ethanol fraction of burdock leaf was not all due to its inhibition effect on bacterial growth, and there are other reasons. Quercetin , P-coumaric acid were found to inhibit biofilm formation.	Tang et al. 2014
Burdock leaf	*Staphylococcus aureus*, *Listeria monocytogenes*.	Ethanol	The burdock leaf fraction based on its interfering with quorum sensing systems and the significant inhibition on the formation of biofilm	Lou et al. 2014

4. Chemistry behind biofilm formation and control

Bacteria communicate via a phenomenon termed quorum-sensing (QS) in which they secrete and detect chemical signal molecules in their surrounding environment. QS allows a bacterial community to activate a coordinated cellular response such as virulence factor production, biofilm development and shielding . The basic mechanism of quorum sensing involves the interaction of a diffusible signal with a transcriptional regulator, either directly or through activation of a sensor kinase. QS signals are also called "autoinducers" because their recognition often increases their own transcription. Although QS phenomena are typically associated with large-bacterial populations, it is important to note that when a few bacterial cells are confined to a small volume, the accumulation of QS signal is sufficient to activate QS.

There are broad differences between the intra-species QS systems employed by Gram-negative and Gram-positive bacteria (Fig. 3). Most of the Gram-negative quorum sensing bacteria use autoinducer syntheses of the LuxI family to produce QS signals containing a core homoserine lactone moiety connected to a variable side chain. These autoinducers diffuse into the bacterial cells and bind their cognate intracellular LuxR receptors. The activated AHL/LuxR complex interacts with responsive promoters and regulates transcription of downstream genes. There is a fair amount of promiscuity in the production and recognition of AHL signals; a single species may produce more than one type of AHL, and a single type of AHL may be recognized by multiple species. For example, the opportunistic human pathogen Pseudomonas aeruginosa produces the QS signals N-(3-oxododecanoyl)-homoserine lactone (3-oxoC12-HSL) through the autoinducer synthase LasI, as well as N-(butanoyl)-homoserine lactone (C4-HSL) through another autoinducer synthase RhlI. On the other hand, the QS

signal N-(heptanoyl)- homoserine lactone (C7-HSL) is produced by a variety of bacteria such as Edwardsiellatarda, Erwiniapsidii, Pantoeaananatis, Rhizobium leguminosarum and Serratiamarescens (Tan S.Y.E, et al. 2014; Rasmussen et al. 2005, Rabin N, et al. 2015). bis-(3′-5′)-cyclic dimeric guanosine monophosphate (c-di-GMP) is a second messenger used for signal transduction by various bacteria and reportedly modulates lifestyles associated with biofilm formation. Some enzymes (e.g., acylase and lactonase) also interfere with bacterial QS and biofilm formation (Kim et al. 2013)

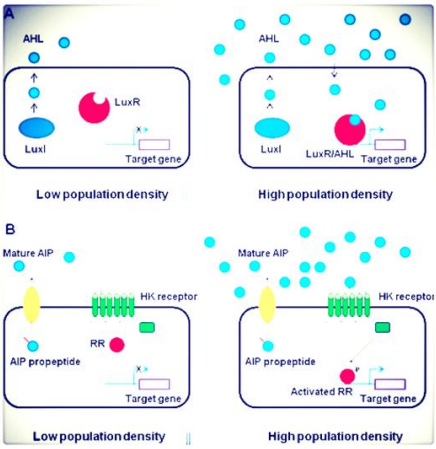


Fig.3 Bacterial QS systems. AAcyl homoserine lactone (AHL) mediated QS in Gram-negative bacteria. AHL signals are produced by LuxI family of autoinducer synthases. At lowpopulation density, the AHL concentration is low and the unstable LuxR receptor is rapidly degraded. When the AHL concentration reaches a threshold, the AHL binds LuxR, thus activating it. The activated AHL/LuxR complex modulates transcription of target genes. B Auto-inducing peptide (AIP) mediated QS in Gram-positive bacteria. AIPs are produced as precursor peptides (propeptides) and are usually modified posttranslationally before secretion via specialized transporters. At high concentration, the mature AIP binds and

activates the cognate trans membrane histidine kinase (HK) receptor, which in turn activates the downstream response regulator (RR). The activated RR directs transcription of target genes.

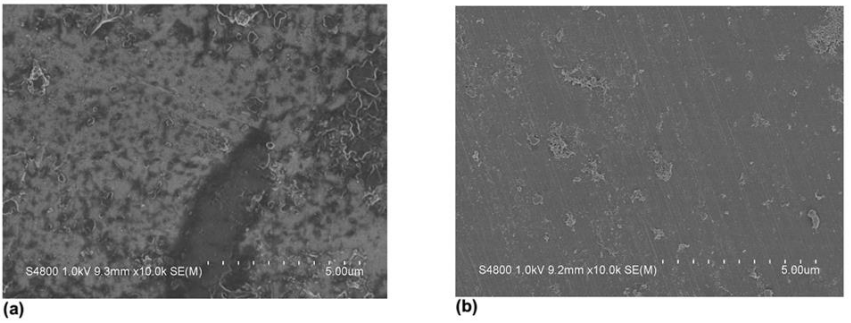


Figure 4: SEM of bacterial biofilms of 70 % ethanol elution fraction (0.5 mg/ml) of (a) absence of burdock leaf fraction (b) treated with burdock leaf fraction (Tang et al. 2014)

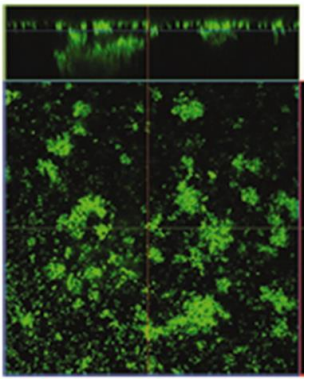


Figure 5: Disturbed biofilm under CLSM observation **(**http://www.pnas.org/content/104/19/8113/F3.expansion.html

5. Concluding remarks and future perspectives

Most of mechanisms are still not fully understood about biofilm formation and inhibition. But in vitro and vivo we have found many green biofilm inhibitor to fight against diseases and

spoilage. More research needs to do for screening out the major components from extract. And we should be more careful on application of natural biofilm inhibitor considering gut microbial challenges.

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Conflict of interest statement

The authors declare that they have no conflict of interest.

REFERENCES

Artini M. et al. 2012, Bacterial biofilm formation inhibitory activity revealed for plant derived natural compounds, Bioorg. Med. Chem. 20 : 920–926

Borges, A. C. Abreu1, J. Malheiro1, M. J. Saavedra, M. Simões 2013, Biofilm prevention and control by dietary phytochemicals, Microbial pathogens and strategies for combating them: science, technology and education, Formatex

Brackman, G. U. Hillaert, S. Van Calenbergh, H.J. Nelis, T. 2009, Coenye, Res. Microbiol. 160, 144–151.

Cady NC, McKean KA, Behnke J, Kubec R, Mosier AP, et al., 2012, Inhibition of Biofilm Formation, Quorum Sensing and Infection in Pseudomonas aeruginosa by Natural Products-Inspired Organosulfur Compounds. PLoS ONE 7(6): e38492

Centers for Disease Control and Prevention (CDC), 2012. Multistate Outbreak of Salmonella Bareilly and Salmonella Nchanga Infections Associated with a Raw Scraped Ground Tuna Product (accessed, 02.10.12.). http://www.cdc.gov/salmonella/bareilly-04-12/index.html.

Carvalho D. C., Fonseca D. M.M., 20007, Preventing biofilm formation: promoting cell separation with terpenes. FEMS MicrobiolEcol,61:406–413.

Cho H.S., et al. 2013, Inhibition of *Pseudomonas aeruginosa* and *Escherichia coli* O157:H7 Biofilm Formation by Plant Metabolite ε-Viniferin, J. Agric. Food Chem., 61, 7120−7126

Correspondence, JMM, 2010, Dietary plant components ellagic acid and tannic acid inhibit *Escherichia coli* biofilm formation, Journal of Medical Microbiology, 496-498

Cowan MM, 1999, Plant products as antimicrobial agents. ClinMicrobiol Rev, 12:564–582.

Cushnie, T.P.T.; Lamb, A.J., 2011, Recent advances in understanding the antibacterial properties of flavonoids. *Int. J. Antimicrob. Agents*, *38*, 99–107.

Dixon RA. 2001, Natural products and plant disease resistance.*Nature*. 2001; 411 (6839):843-847.

Flemming, H.C., Wingender, J. 2010. The biofilm matrix. *Nat Rev Microbiol*, **8**(9), 623-33.

Gandhi, M., Chikindas, M.L., 2007. Listeria: a foodborne pathogen that knows how to survive. Int. J. Food Microbiol. 113, 1-15.

González-Ortiz G. et al. 2014, New properties of wheat bran: anti-biofilm activity andinterference with bacteria quorum-sensing systems, Environmental Microbiology, 16(5), 1346–1353

Goller CC, Romeo T (2008). Environmental influences on biofilm development. Curr. Top Microbiol. Immunol. 322:37-66.

Healy, B., Huynh, S., Mullane, N., O'Brien, S., Iversen, C., Lehner, A., Stephan, R., Parker, C.T., Fanning, S., 2009. Microarray-based comparative genomic indexing of the Cronobacter genus (Enterobactersakazakii).Int. J. Food Microbiol. 136, 159-164.

Iwamoto, M., Ayers, T., Mahon, B.E., Swerdlow, D.L., 2010. Epidemiology of seafood associated infections in the United States. Clin. Microbiol. Rev. 23, 399-411.

John N.R., Gala V.C., Sawant C.S., 2013, Inhibitory effects of plant extracts on multi-species dental biofilm formation in-vitro, Int J Pharm Bio Sci 2013 Apr; 4(2): (B) 487 - 495

Kalburge, S.S., Whitaker, W.B., Boyd, E.F., 2014. High-salt preadaptation of Vibrio parahaemolyticus enhances survival in response to lethal environmental stresses. J. Food Prot. 77, 246-253.

Kim H-S, Park H-D (2013) Ginger Extract Inhibits Biofilm Formation by *Pseudomonas aeruginosa* PA14. PLoS ONE 8(9): e76106. doi:10.1371/ journal.pone.0076106

Kubo I, Masuoka N, Ha TJ, Tsujimoto K. Antioxidant activity of anacardic acids. *Food Chemistry*. 2006;99(3):555-562.

Kunduru MR, Pometto ALIII (1996). Continuous ethanol production by *Zymomonas mobilis* and *Saccharomyces cerevisiae* in biofilm reactors. J. Ind. Microbiol. 16(4):249-256.

Lapara TM, Konopka A, Nakatsu CH, Alleman JE (2001). Thermophilic aerobic treatment of a synthetic wastewater in a membrane-coupled bioreactor. J. Ind. Microbiol. Biotechnol. 26(4):203-209.

Liu XEA (2005). Continuous nisin production in laboratory media and whey permeate by immobilized *Lactococcus lactis*. Process Biochem. 40:13-24.

Li XZ, Jeremy SW, Staffan K, Bettina R (2006). Enhanced benzaldehyde tolerance in *Zymomonas mobilis* biofilms

and the potential of biofilm applications in fine-chemical production. Appl Environ. Microbiol. 72(2):1639-1644.

Lee, J.H., et al. 2013, Diverse plant extracts and *trans*-resveratrol inhibit biofilm formation and swarming of *Escherichia coli* O157:H7Biofouling, 29(10), 1189–1203.

Lou, Z. et al. 2014, Effect of ethanol fraction of burdock leaf on biofilm formation and bacteria growth, European Food Research and Technology, 239 (2) 305-311

McDougald, D., Lin,W.H., Rice, S.A., Kjelleberg, S., 2006. The role of quorum sensing and the effect of environmental conditions on biofilm formation by strains of Vibrio vulnificus.Biofouling 22, 133-144.

Mizan, M. F. Jahid, I. K. Ha, S. D. 2015, Microbial biofilms in seafood: A food-hygiene challenge, Food Microbiol, 49, 41-55

Mutalib L.Y.,, Haji S.H., Hanna S.T., 2015, Estimation of bacterial growth inhibition and antibiofilm activity of Hibiscus *Sabdariffa* plant extracts against standard pathogenic bacteriaWorld J Pharm Sci , ; 3(8): 1525-1530

Myszka, K., Czaczyk, K., 2011. Bacterial biofilms on food contact surfaces - a review. Pol. J. Food Nutr. Sci. 61, 173-180.

Namasivayam S.K.R., Roy E.A., 2013, Anti biofilm effect of medicinal plant extracts against clinical isolate ofbiofilm of *escherichia coli,* International Journal of Pharmacy and Pharmaceutical Sciences, *5(2) 486-489*

Newman DJ, Cragg GM.2007, Natural products as sources of new drugs over the last 25 years.*Journal of Natural Products*.;70(3):461-477.

Ozçelik, B.; Orhan, I.; Toker, G., 2006, Antiviral and antimicrobial assessment of some selected flavonoids. *Z. Naturforsch.* 2006, *61*, 632–638.

Packiavathy I. A. S. V. et al. 2014, Inhibition of biofilm development of uropathogens by curcumin – An anti-

quorum sensing agent from *Curcuma longa*, Food Chemistry 148 : 453–460

Perumal S., Mahmud R., 2013, Chemical analysis, inhibition of biofilm formation and biofilm eradication potential of *Euphorbia hirta* L. against clinical isolates and standard strains, BMC Complementary and Alternative Medicine, 13:346

Plyuta V et al. 2013, Effect of plant phenolic compounds on biofilm formation by Pseudomonas aeruginosa. APMIS

Pratiwi SUT, Lagendijk EL, Hertiani T, WeertSd, Hondel CAMJJVD, 2015, Antimicrobial effects of Indonesian Medicinal Plants Extracts on Planktonic and Biofilm Growth of *Pseudomonas aeruginosa*and *Staphylococcus aureus*. J Horticulture 2: 119.

Prior RL, Cao G. 2000, Antioxidant phytochemicals in fruits and vegetables: Diet and health implications. *HortScience*.;35(4):588-592.

Qureshi N, Annous BA, Ezeji TC, Karcher P, Maddox IS (2005). Biofilm reactors for industrial bioconversion processes: employing potential of enhanced reaction rates. Microb. Cell Fact. 4:24.

Rezanka et al. 2012, Natural Products: Strategic Tools for Modulation of Biofilm Formation, Chp. 10, Studies in Natural Products Chemistry, 38: 269-303.

Rabin N, et al. 2015, Agents that inhibit bacterial biofilm formation, *Future Med. Chem.,* 7(5), 647–671

Rasmussen TB, Bjarnsholt T, Skindersoe ME, Hentzer M, Kristoffersen P, et al. (2005) Screening for quorum sensing inhibitor (QSI) by use of a novel genetic system, the QSI selector. Journal of Bacteriology 187: 1799-1814.

Saavedra MJ, Borges A, Dias C, Aires A, Bennett RN, Rosa ES, Simões M. 2010, Antimicrobial activity of phenolics and glucosinolate hydrolysis products and their synergy with

streptomycin against pathogenic bacteria. *Medicinal Chemistry*.;6(3):174-183.

Saleem, M.; Nazir, M.; Ali, M.S.; Hussain, H.; Lee, Y.S.; Riaz, N.; Jabbar, A., 2010, Antimicrobial natural products: An update on future antibiotic drug candidates. *Nat. Prod. Rep.* **2010**, *27*, 238–254.

Santiago C, Lim K.h., Hwei-San Loh H.S., Ting K.N., 2015, Inhibitory Effect of Duabangagrandifloraon MRSA BiofilmFormation via Prevention of Cell-Surface Attachment andPBP2a Production, Molecules , 20, 4473-4482

Smitinont T et al. Exopolysaccharide-producing lactic acid bacteria strains from traditional Thai fermented foods:isolation, identification and exopolysaccharide characterization. Int J. Food Microbiol 1999; 51:105-111

Spector, M.P., Kenyon, W.J., 2012. Resistance and survival strategies of *Salmonella enterica* to environmental stresses. Food Res. Int. 45, 455-481.

T. Bjarnsholt et al. (eds.), 2011. *Biofilm Infections*, Springer link, 1-9, DOI 10.1007/978-1-4419-6084-9_1

Ta, C.A.K, 2015, Bacterial biofilm inhibition and antifungal activity ofneotropical plants, PhD thesis, University of Ottawa, 14-34

Taganna J.C., et al. 2011, Tannin-rich fraction from *Terminaliacatappa* inhibits quorum sensing (QS) in Chromobacteriumviolaceum and the QS-controlled biofilm maturation and LasAstaphylolytic activity in *Pseudomonas aeruginosa*

Tan S.Y.E, et al. 2014, Emerging frontiers in detection and control of bacterial biofilms, Current Opinion in Biotechnology , 26:1–6

Tang et al. 2014. Chemical Composition and Anti-Biofilm Activity of Burdock (*Arctium lappa* L *Asteraceae*) Leaf

Fractions against *Staphylococcus aureus.* Tropical Journal of Pharmaceutical Research, 13 (11): 1933-1939

Todhanakasem, T., 2013, Review: Microbial biofilm in the industry, African Journal of Microbiology Research, 7(17), 1625-1634

Upadhyay, A., 2014, "Investigating the Potential of Plant-derived Antimicrobials and Probiotic Bacteria for Controlling *Listeria monocytogenes*" *Doctoral Dissertations.*Paper 326.,82-86

Wojnicz D., Kucharska A. Z., Soko A.L., Kicia M., DorotaTichaczek-Goska, 2012, Medicinal plants extracts affect virulence factors expression and biofilm formation by the uropathogenic Escherichia coli, Urol Res , 40:683–697

Zeng, Z. L. Qian, L. Cao, H. Tan, Y. Huang, X. Xue, Y. Shen, S. Zhou, 2008, Appl. Microbiol. Biotechnol. 79 : 119–126.

Zhang, J. et al. 2014, Polyphenolic extract from *Rosa rugosa* tea inhibits bacterial quorum sensing and biofilm formation, Food Control 42, 125-131.