A Major Concern in Food Industry, Contamination Reservoir: Bacterial Biofilm

B. Uymaz Tezel[1] and **P. Şanlıbaba**[2]

[1] Food Technology Program, Bayramiç Vocational School, Çanakkale Onsekiz Mart University, Çanakkale, Turkey
[2] Department of Food Engineering, Engineering Faculty, Ankara University, Gölbaşı, Ankara, Turkey

To resistance to inhospitable environmental stresses bacteria have upgraded a variety of mechanisms such as biofilm formation. This self-protection growth pattern of bacteria makes them more resistant to disinfectants and antimicrobial agents since it provides a barrier which prevents or lessens the contact with antimicrobial agents. Formation of biofilms, which acts as a contamination reservoir, is now regarded as a major concern in the food industry since biofilms may contain potential spoilage and pathogenic bacteria including *Listeria monocytogenes*, *Staphylococcus* spp., *Lactobacillus* spp., *Clostridium* spp., *Bacillus* spp., *Salmonella* spp., *Escherichia coli* O157: H7 and *Campylobacter jejuni* etc. Many factors can be sorted for the formation and development of biofilms: specific bacteria strain, material surface properties and environmental parameters such as pH, temperature and nutrient levels. Each biofilm is different due to the diversity of biofilm-forming bacteria in various food industries, as a result, there is no unique system which can remove all biofilms. The first and perhaps the most important thing to do is prevent biofilm formation by regularly cleaning and disinfecting and biofilm problem should be well-analysed to determine an effective cleaning and disinfection operation.

Keywords: Food industry; bacterial biofilm; contamination reservoir

1. Introduction

Foodborne diseases are widespread and crucial public health threat which are the cause of hospitalisation, mortality or significant impediment to socio-economic development. According to global estimates, thirty-one foodborne hazards causing 32 diseases most of which are diarrhoeal disease agents particularly norovirus and *Campylobacter* spp. According to World Health Organization 2015 reports the 31 hazards caused 600 million foodborne illnesses and 420,000 deaths in 2010 however, unfortunately, the real number is likely to be much higher. Foodborne diarrhoeal disease agents caused 230,000 deaths most of whom are children [1]. The burden of foodborne diseases in industrialised countries is still an imported challenge. In a recent report (2105) by the European Food Safety Authority (EFSA) and the European Centre for Disease Prevention and Control (EDC) a total of 5196 foodborne outbreaks were reported in the European Union (EU) in 2013 [2]. According to another statistics released by the United States Centre for Disease Control and Prevention (CDC) emphasise that conjecturally, 48 million foodborne diseases reported resulting in 128,000 hospitalisations and 3000 deaths annually in the USA, with estimated money loss of billions dollar in a single year [3]. Microbial contamination, both from microbiological or chemical, may occur at different steps of the food chain: (1) cultivation environment such as farm, orchard, fishery or livestock farms (2) processing environment such as slaughterhouse, cannery or packing plants (3) preparation, storage and even retail environments [4]. Members of EU have adopted an approach to food safety from the farm to the fork for the protection of consumers against microbial contamination throughout the food chain [5, 6]. Knowing main risks and contaminants in each food processing steps is crucial for their control, prevention or elimination. In order to manage the production of safe and quality foods adoption of risk assessment models such as Hazard Analysis Critical Control Points (HACCP) and Good Manufacturing Practices (GMP) are essential, even though these systems have proven very effective for the control of food safety, it must never be realized that they are designed on the basis of known hazards. Both human and microorganisms related factors lead to evaluation of food safety which occurs on a molecular scale (point mutations or the acquisition of a plasmid by microorganisms) and on short timescales. Innovations according to new validations and verifications are necessary [6]. Furthermore, many bacteria have upgraded a variety of mechanisms such as biofilm formation to be resistant to inhospitable environmental stresses. Formation of biofilms, which acts as a contamination reservoir, is now regarded as a major concern in the food industry since biofilms may contain potential spoilage and pathogenic bacteria including *Listeria monocytogenes*, *Salmonella* spp., *Escherichia coli* O157: H7, *Campylobacter jejuni*, *Bacillus* spp., *Pseudomonas* spp., even *Lactobacillus curvatus*, *Lactobacillus fermentum* etc. [7]. The biofilms increase the risk for post-processing contamination and leading to lowered shelf life of the food product and/or transmission of diseases [8, 9, 10, 11]. Moreover, the formation of biofilms on industry surfaces leads to economic losses due to mechanical blockage, impairment of heat transfer, increase in fluid frictional resistance and corrosion of equipment [12, 13].

In order to gain deeper information about biofilms, a large of number research has been performed in recent years which are focused on both understanding of biofilm formation mechanisms and their effective and control strategies. The main objective of this book chapter is to emphasise the challenges caused by biofilms in food industries and to summarise the knowledge of biofilm formation of foodborne bacteria.

2. Bacterial Biofilm

Bacterial biofilms are defined by Costerton and Stewart [14] as an aggregation of microorganisms attached to and growing on surfaces including plastic, glass, metal and wood which are a prevalent mode of growth for microorganisms in nature [15]. The current description of bacterial biofilms is performed as an assemblage of surface-associated microbial cells that are enclosed in hydrated (from 85% to 95%) extracellular polymeric substances (EPS) mainly constituted of polysaccharides, proteins, phospholipids, teichoic and even nucleic acids [16, 17].

The first observation of bacterial biofilm dates back to 1684. Antonie van Leeuwenhoek, using his primitive light microscope, found that microorganism attached to tooth surfaces, form a sessile community and called them as 'animakuli' [15, 17, 18]. According to the findings of light microscopy bacterial biofilm was observed first as marine bacteria attached to the ships' hulls in the 1920s [15]; Claude E. ZoBell also determined that some marine bacteria could cling to surface of glass slides, form microcolonies and also these structures on the glass surfaces was not distributed by washing or shaking [19]. In the 1980s, by the development of the microscopy techniques, bacteria were observed on the solid surfaces of many ecological environments including wastewater, treatment systems, equipment used to manufacture vinegar, industrial water systems, tooth decay, urinary tract and other implanted medical devices [20, 15]. The first correct question to ask is why bacteria create a structure as a biofilm. This proper colonisation of microorganisms on surfaces, embedded in the EPS, provides survival of bacteria under environmental stress conditions such as ultraviolet radiation, physicochemical stresses or insufficient supply of nutritive resources. This self-protection growth pattern also provides bacteria up 10 to 1000 times more resistance to disinfectants and antimicrobial agents than planktonic bacterial cells [21, 22, 23]. The biofilm structure acts as a barrier and prevents or decreases contact of the bacteria with antimicrobial agents. In order to make pathogens inactive and maintain safe and quality products, chemical disinfectants such as peroxides, chloramines or hypochlorites are widely used in food industry. However, it has been reported in the United States that around 80% of persistent bacterial infections are associated with biofilms [24].

2.1 Biofilm formation and development steps

To explain this threatening issue in a broad range of food industries including brewing, seafood processing, dairy processing, poultry processing and meat processing the second question to ask is which molecular mechanisms, factors and bacteria are involved in biofilm formation.

The biofilm formation process that involves both physicochemical and biological factors starts with the initial attachment of planktonic bacteria to solid surfaces and takes in a short time and it can be active (depend on bacterial motility) or passive (gravitational transportation of planktonic cells) [17, 24, 25, 26]. The attached microorganisms may detach from the surface and return to planktonic forms at any moment, it is considered as a reversible adhesion of the bacteria and surface through van der Walls, electrostatic and Lewis acid-base interactions. The main determinant factor is physiochemical properties of the bacterial surface – mostly negative charged- due to the presence of the proteins anchored to the cell wall [25]. For example, the electrostatic charge of bacterial cell walls and cell surface hydrophobicity of *L. monocytogenes* have been the determinate that govern its attachment to stainless steel [27], also BapA surface protein has been found to be directly involved in biofilm formation at the liquid-air interface in *S. enterica* [28]. The cell surface appendages such as flagella (present in both Gram-positive and Gram-negative bacteria), pili (thinner and shorter than flagella, present in both Gram-positive and Gram-negative bacteria) or curli fibres (amyloid like protein, present in only Gram-negative bacteria) not only the principal components for bacterial motility, but also may play a crucial role (involved in cell-cell and cell-surface contacts) in the initial stages of biofilm formation [29, 25].

The other important factors that influence the initial cell attachment are the physical characteristics of solid surfaces such as texture (rough or smooth), surface charge, hydrophobicity and weather be covered by a film of organic molecules (proteins or pre-existing polymeric substances). For instance, high free energy, wet surfaces promote bacterial adhesion: Although hydrophilic surfaces (stainless steel, glass etc.) are more adherent than hydrophobic surfaces (plastics) [17, 24, 30], rougher and hydrophobic surfaces are preferred attachment. According to findings of research by Sinde and Carballo which shown that *Salmonella* and *Listeria* can attach in high numbers to hydrophobic surfaces than the hydrophilic ones [31].

The production of EPS induces to turn weak interaction between bacteria and surface to a permanent bonding (by dipole-dipole, hydrophobic, ion-ion, ion-dipole, covalent and hydrogen interactions); so this stage of biofilm formation is called as an irreversible attachment. In the irreversible stage the biofilm grows through cell division and bacterial cells upregulate the expressions of specific attachment-related genes [17, 23, 24, 26]. The microcolony formation in the early development of biofilm architecture stage helps strengthen the bond between the bacteria and the substratum and stabilises the colony from any environmental stress and also allows for interspecies substrate exchange [32].

The maturation stage usually has been characterised by developing of flat or mushroom shape which requires periods of 10 days or more [17, 24, 26]. A mature biofilm is a highly organised ecosystem including heterogeneous complex-enclosed microcolonies with water channels are dispersed and able to provide passages for the exchange of nutrients, metabolites and waste products [33].

The third question to ask is how different bacteria microcolonies have attended to biofilm architecture. Although its role has not been completely explained, Quorum Sensing (QS) which is a kind of communication between bacteria responsible for heterogeneous biofilm architecture and provides information about cell density for adjusting their gene expression [23]. QS includes production and sensing of signal molecules which regulate the behaviour and physiological processes of bacteria such as growth, sporulation, pathogenicity or starting to biofilm formation [34, 35, 36]. There have been several studies emphasise effect of the QS molecules in different bacteria [25]: the second messenger cyclic-di-GMP responsible from cellular transition between motility and sessility in *S. enterica* and many other pathogens [37, 38]; the accessory gene regulator (*agr*) system plays a central role to mature biofilm phenotypes and 3D architectures in *S. aureus* [39, 40]; the CodY regulator, induces both biofilm formation (under nutrient starvation conditions) and expression of motility and virulence factors (under abundant nutrient conditions), controls the multicellular behaviour in *B. cereus* [41].

The detachment of bacterial cells from biofilm architecture and revert to their planktonic form is the last stage in the biofilm formation cycle, called as Dispersion. Both external and internal biofilm processes may lead to biofilm detachment [24]. The dispersion stage is an active process which has been controlled by QS signals and allows for the colonisation of new niches. Generally, the acly-homoserine lactones (AHLs) and oligopeptides are the extracellular signalling molecules for Gram-negative and Gram-positive bacteria, respectively [42]. The *agr* system also controls dispersion in *S. aureus* biofilm by the activation of protease production [43]. Sometimes some molecules produced by biofilm cells disrupts other species' biofilm architecture. For example, the polyamines as norspermidine and norspermine produced by *B. subtilis*, disrupt the *S. aureus* and *E. coli* biofilm architecture [44].

Many effective factors can be sorted on the formation and development of biofilms: specific factors of bacterial strain (Gram-positive/negative, microbial shape, structure, presence of flagella, pili, capsules or exopolymeric substances molecular composition, species, growth phase and age), material surface properties (chemistry, topography, physiochemistry) and environmental parameters such as pH, temperature, nutrient levels and fluid dynamics [15, 17, 23]. The factors grouped as environmental parameters may be considered to play a crucial role in the turning of planktonic cells to the sessile form. There are several evidence demonstrate that the different adhesion levels of various species at different pH, temperature and concentrations of phosphates or pH. Main effects of environmental parameters on biofilm formation in the food industry can be summarized as follows: rough, hydrophobic and opposite charges of surface favours biofilm initiation; lower temperatures either stimulate biofilm formation which leads to more uniform properties of polysaccharides- or lower biofilm formation -decrease cell surface hydrophobicity levels; decrease of oxygen inhibits biofilm initiation; higher shear rates decrease bacterial attachment but increase density and thinness of biofilms; high osmolarity of food matrix inhibits biofilm formation; influence of pH and ionic strength on biofilm formation through changes in surface hydrophobicity and charge [45]. In addition to abiotic environmental factors listed above, one should consider that the effect of biotic factors is also decisive. Interactions between different microbial populations, especially in multi species biofilms, could greatly impact the development of biofilm as far as the species specific factors. Whereas these interactions mostly are beneficial for the partners, in some cases presence of a bacterial species could inhibit biofilm formation. For example, while *L. monocytogenes* in a mixed biofilm with *Lb. plantarum* exhibited higher resistance to benzalkonium chloride and peracetic acid than single species biofilm [46]; biofilm formation of *L. monocytogenes* can be reduced by the presence of *S. xylosus* and *P. fragi* or bacteriocin-producing *Lactococcus lactis* [29, 45, 47].

3. Biofilm formation in Gram-positive and negative bacteria

According to the nature of microorganisms and depending on the species-specific factors biofilms have displayed diversity. Each biofilm is different due to the diversity of biofilm-forming bacteria in different food industries and also same species can form different biofilm architecture under different grown conditions. According to evidence of SEM, epifluorescence microscopy and confocal laser scanning microscopy (CLSM), biofilms of *L. monocytogenes* under static conditions consist of a homogeneous layer of cells with a similar morphology to planktonic cells but, under continuous flow conditions consist of spherically shaped microcolonies [7, 48]. The structural and developmental features of bacteria such as motility and sporulation are eminent importance for the biofilm formation and are strictly connected with each other. Formation of biofilms provides an optimal environment for sporulation of *Bacilli* spp. Although, cells in biofilm architecture are not motile, in several bacteria such as *L. monocytogenes* and *B. subtilis* motility has been found to be substantial for initial attachment of cells to a surface and formation of biofilm [7]. The flagellar based motility appeared to be essential to propel cells towards the surface before attachment for static biofilm formation but not for biofilm formation under continuous flow conditions in both *L. monocytogenes* and *B. subtilis* [49, 50]. In this situation evaluating biofilms according to the structural and developmental features of microorganisms, growing conditions and food processing environments is a rational approach.

3.1 Biofilm forming Gram-positive bacteria

L. monocytogenes is a psychotropic bacterium which can lead to mild gastroenteritis or severe infections and mostly transmitted to man through food. *L. monocytogenes* strains can grow between pH 4.6 and 9.5 and at 0.92 aw and are recurrently found on floors, drains and equipment of the food industry, even in the cold and wet atmosphere of refrigerated rooms where non-psychrotrophic can only survive. The studies focused on to understand why *L. monocytogenes* persists in the food industry have found several reasons including adhesion potential, biofilm forming ability, resistance to desiccation, acid and heat, tolerance or resistance to disinfectants[51]. It has been demonstrated that *L. monocytogenes* can form a biofilm which protects cells from the action of antimicrobials and sanitizers and allows long term persistence them on surfaces such as rubber, plastics, glass and stainless steel [52]. Having flagella, QS and extracellular DNA (eDNA) may play a major role in the formation of listerial biofilm. The attachment of *L. monocytogenes* has been governed by electrostatic charge of bacterial cell walls (conferred by peptidoglycan anionic teichoic acids) and cell surface hydrophobicity (enhanced by the presence of lactic acid) [17]. *PrfA* flagellar biosynthesis regulator, *LuxS* system, *agr* system have been shown to be involved in the QS systems of *L. monocytogenes* biofilm. Alonso et al., [53] found that both disruptions in several purine biosynthesis genes and mutation in flagella related genes at 30-35 ^{o}C caused reduced biofilm formation in *L. monocytogenes*. Piercey et al., [54] carried out that at a temperature common to food environments identified the involvement of nine genetic loci that had not previously linked to biofilm formation in *L. monocytogenes* as well as ten genes also known to be associated with *L. monocytogenes* biofilm temperatures. The role of eDNA has been described by Harmsen et al., [55] in the early stages of cell attachment to surfaces, and also reported that N-acetylglucosamine acted as a co-factor and/or scaffolding in eDNA formation.

As discussed above, the biofilm formation is strongly affected by the hydrophobic or hydrophilic interactions between microbial cell charge and contact surfaces. For the risk assessment and reduction of biofilm production which are a great expense for the enterprises, several measures should be taken as the choice of correct materials for installations. Both the hydrophilic (stainless steel and glass) and the hydrophobic (polymeric) materials are the most common materials used in the food industry [52]. According to several researchers who are investigating relationships between biofilms and contact surfaces suggest that *Listeria* biofilms may adhere more tightly to hydrophobic surfaces than hydrophilic surfaces at cellular level [56]; but some found that biofilm levels of *L. monocytogenes* were significantly higher on glass at different temperatures compared to polystyrene and stainless steel [57]. By the same researchers, a positive correlation between hydrophobicity and heat was suggested that the biofilm formation was influenced by temperature due to modification of cell surface hydrophobicity. The presence of glucose or high concentrations of NaCl (4% to 10%) may lead to an increased ability to self-aggregation and biofilm development in *L. monocytogenes* [58, 59]. With regard to a study conducted on the influence of growth medium, surface and pH on the initial 2D structure of static listerial biofilms, three of the tested strains were able to reach a honeycomb like structure and could develop complex biofilms but only one could not enter to the second step. Time-shift differences according to growth medium and surface have been observed on the plastic surface, except one strain. However, the first step of the primary structure of *L. monocytogenes* which is a critical stage for the development of listerial biofilms, is impaired for all strains in acidic conditions [60]. In accordance with this study, both Tresse et al., [61], and Smooth and Pirson [62] observed a lower cell attachment at pH five than pH 7 or 8. The down-regulation of flagellin synthesis in acidic conditions was also reported by Tresse et al., [63]. The dependency of *L. monocytogenes* biofilms on nutritional conditions is one of the most studied subjects. Although most of the previous studies show that growth in rich media (BHI or TSB) was not lead to switch cells from planktonic to sessile state [64], but growing in synthetic media enhanced attachment to surfaces and biofilm formation [65].

Due to the contamination of food products by both pathogens and spoilage microorganisms is a crucial issue the management of the food processing plant should consider the prevention strategies. The proper design of food contact equipment as the primary strategy should be performed. This should never be disregarded that the most common sources involved in biofilm accumulation are the floors, waste water pipes, bends in pipes, rubber seals, conveyor belts, etc. Materials for industrial installations should be preferred with smooth, resistant to corrosion and damage, be free of sharp edges, easily cleanable and allowing the 'clean in place' [17, 52].

Because of its rich nutrient content and high aw, milk is tremendously vulnerable to contamination against microorganisms. Outbreaks of pathogens associated with biofilms in the dairy industry are including *L. monocytogenes*, *Bacillus* spp., *Pseudomonas* spp., even *Lactobacillus curvatus*, *Lactobacillus fermentum* [17, 66, 67]. According to results of a study aimed to evaluate the biofilm production of *L. monocytogenes* strains isolated from the environment of cheese processing plants in Brazil, all isolates showed ability to produce biofilms on polystyrene microplates, also on stainless steel (24.7%) and 4.7% of the strains were classified as strong biofilm producer [68]. *L. monocytogenes* is able to form multi-species biofilms with both Gram-positive and negative bacteria depending on the genera implicated and the environmental conditions [69]. In a recent study which aimed to find out possible environmental relationships among *L. monocytogenes* and other species, environmental samples belonging to work surfaces from fish, meat and dairy industries were analysed by serotyping and PFGE. 12 samples were found positive for *L. monocytogenes* and *E.coli* and *Carnobacterium* spp. were founded accompanying microbiota in fish and meat industry, respectively [70]. Although the quality of both equipment and water is the main cause of the risks in the fish processing industry, many

types of fish-contaminated-bacteria are found to be biofilm-forming, including *Vibrio* spp., also many other genera such as *L. monocytogenes*, *Salmonella* spp., *Bacillus* spp., *Aeromonas*, and *Pseudomonas* spp., [17].

Mainly three conventional methods have been applied including physical, chemical and biological methods in order to prevent or inhibit the biofilms in food-processing plants. The physical methods commonly used are mechanical scrubbing, jet cleaning, ultrasonic cleaning and other ways [71]. The strong oxidising agents with a broad antimicrobial spectrum such as hypochlorous acid, chlorine, iodine, ozone, hydrogen peroxide, peroxyacetic acid and quaternary ammonium chloride and anionic acids are the most used chemical agents for sanitation procedures in food producing plants [52]. Several comparative experimental studies have been performed in order to establish the most effective treatment for eradication *L. monocytogenes* biofilm. The gaseous ClO_2 [72]; aerosolised sanitizers [73] and nanostructured titanium dioxide combined [74] have been effective on both *L. monocytogenes* planktonic cells and biofilms. With the increasing consumer demand rather than chemicals preferring environment-friendly treatments launch to researching new emerging strategies. These innovative strategies can be sorted as affecting cell adhesion by modifying contact surface hydrophobicity [75], using different essential oils as anti-biofilms agents [76], using bio-solutions such as probiotic [77], bacteriocin-producing strains [78] or phages [79] and using enzymes, antimicrobial molecules from microbial origin [80]. In a study was conducted with a innovate approach, the methanol extracts of several plants have been found out to be effective on *L. monocytogenes* and *S. aureus* biofilm formation [80].

Staphylococcus spp. is widely distributed in nature including air, water, body surfaces of skin which is bunch shaped cocci without flagella, spore and capsule. The enterotoxin of *S. aureus* causes food poisoning which is a significant concern in the food industry. There for the primary enterotoxigenic pathogenic carriers are humans, food handlers may directly contaminate food. The presence of methicillin-resistance and ability to develop biofilms on different materials also makes a serious risk for safety food production [71]. According to a comparative investigation on biofilm formation capability of many common pathogenic bacteria indicated that *Staphylococcus* tend to develop more biofilm than *E.coli*, *Salmonella* spp., and *Bacillus cereus* [81]. Among the Staphylococcal species *S. aureus* and *S. epidermis* adhere to both biomaterials and food contact surfaces and form a biofilm in 24-48 hours at 37 ^{o}C. The process of heterogeneous multi-layered *S. aureus* biofilm formation is regulated by multiple genes including expression of the PIA by the *ica*ADBC operon; release of eDNA; expression of numerous surface proteins (Bap, SasG, FnBPs or Spa) [82]. eDNA has been reported as a major component of biofilm which has roles in initial attachment and accumulative stages of *Staphylococcus* spp. [83]. The PCR based analyses have shown that eDNA is similar to genomic DNA which supports the hypothesis that eDNA originates from the lysis of a subpopulation of the bacteria [84]. According to results of a study aimed to investigate the silico biofilm production ability of *S. aureus* from dairy environments, 31 *S. aureus* were obtained and detected as producer of biofilm on stainless steel, rubber and silicon surfaces [85]. Lopez et al., [86] reported that the glycone (myricitrin, hesperidin and phloridzin) and aglycone (myricetin, hesperetin and phloretin) flavonoids inhibit biofilm formation by *S. aureus* strains by overexpressing the *msrA* and *norA* efflux protein genes.

Lactic acid bacteria (LAB) are non-motile bacteria which have been widely used for the fermentation of raw milk, meat and vegetables. In spite of their role in the food industry, LAB -especially genus *Lactobacillus*- may lead to food spoilage. Non-starter *Lactobacillus* spp. can produce biofilm on the both raw material and production environment which acts as contamination reservoir [87]. Although many biofilm producer bacteria such as *Listeria* spp., *Bacillus* spp. and *Salmonella* spp. have flagella which are necessary for initial attachment to surfaces, the adhesion of non-motile bacteria to surfaces can be explained by the sedimentation processes, so the initiation of biofilm and maturation of non-motile bacteria is different from the reported for motile bacteria [79, 88]. There is another risk for the fermented foods and beverages that capacity of biogenic amine (BA) producers to form biofilms. The histamine, tyramine and putrescine are low-molecular-weight organic compounds that may found in large quantities. However much BAs play a major role in human physiology, ample consumption of food containing them may lead to toxicological effects. The aminoacyl decarboxylase activity which mainly presents in Gram-positive bacteria of the LAB group is responsible for the formation of BAs [89]. The formation of biofilms by both clinical and food-related *Enterococcus* spp. species have been studied since their pathogenic potential [90, 91].

Pseudomonas spp. includes major proteolytic enzyme producer species including *Pseudomonas aeruginosa* which is pathogenic and spoilage bacterium commonly found in food processing plants and has the tendency to form biofilms [92]. So much so that *Pseudomonas aeruginosa* has demonstrated biofilm formation in the lungs of patients suffering from cystic fibrosis which is noncurable and eventually results in death [93]. In order to inhibit biofilm formation, many studies have been focused on the AHLs QS system which is involved in biofilm formation by knock-out AHLs synthase genes [94]. Another biofilm producer Gram-positive bacteria is *Clostridium perfringes* which are aerotolerant anaerobic, spore-forming and an opportunistic pathogen. *Clostridium perfringes* causes food poisoning both in human and animals as result of its ability to produce many different toxins. The production mono- and dual- species biofilm protects *Clostridium perfringes* cells from the action of potassium monopersulfate, quaternary ammonium chloride, and hydrogen peroxide and glutaraldehyde solutions [95].

3.2 Biofilm forming Gram-negative bacteria

Salmonella spp. is one of the most important foodborne pathogens and cause of Salmonellosis. The dynamics of *Salmonella* infection is variable and are effected mainly by contaminating food and water supplies –especially in

underdeveloped countries-, personal lifestyle, changes in the industry, technology, commerce and travel. [96]. Approximately 2500 Gram-negative *Salmonella* serovars have been reported and can be found in the intestinal tract of both human and animals in nature. *Salmonella* spp. is the first reported biofilm producer foodborne pathogen. This enteric pathogen follows a cyclic lifestyle in which host colonisation is alternated with periods of survival outside the host. The ability of *Salmonella* to form biofilms both host colonisation and non-host conditions contributes to survival and transmission to new hosts [97].

The abiotic surfaces such as plastic, rubber, cement, and stainless steel which are extensively used in farms, food processing industry, kitchens, toilets and bathrooms are the well-documented surfaces on which *Salmonella* is able to form biofilms. Several reports have demonstrated the *Salmonella* biofilms on these abiotic surfaces: According to Stepanovic et al., [98] findings, there was no difference on the ability of biofilm formation on polystyrene microplates of tested 122 *Salmonella* spp. isolates from both humans, animals and food. In accordance with this results, Vestby et al., [99] also found biofilm formation capacity of 111 *Salmonella* isolates feed and fish meal factory environment. It should never be forgotten that toilets and bathrooms in the houses even factories are the most common *Salmonella* contamination source. This fact has been supported by a study [100] which detected survival of *Salmonella* in toilet and bathroom in homes of salmonellosis patients. *Salmonella* spp., *Campylobacter* spp., and *E. coli* O157: H7 are commonly found in meat, poultry and on contact surfaces of meat and ready-to-eat industry due to ample organic residues which could be a niche for microorganism's accumulation and biofilm formation. [17]. The study about biofilm formation by poultry isolates of *Salmonella* on commonly used contact surfaces and their sensitivity to sanitizers was detected [101]. Compatible with this study results Díez-García et al., [102] found the relationships between biofilm formation and growth kinetic parameters of *S. enterica* isolates from poultry. Another study has indicated that there is no significant difference in biofilm-forming ability among the *Salmonella* serotypes (*S.* Typhimurium, *S.* Enteritidis, *S.* Typhi) from different sources or geographical regions [103]. According to a remarkable study's results which detect the effects of different temperature, pH and nutrient availability on *S.* Enteritidis biofilm formation, the environmental stress conditions may alter biofilm resistance to sanitizers [104].

Salmonella is able to adhere to biotic surfaces such as epithelial cells, gallstones and even form biofilms which lead to the development of chronic infections in hosts. Although plants are not considered as a host for enteric pathogens, numerous *Salmonella* outbreaks reported in less developed countries due to contamination of water, soil with sewage. Remarkably, several reports have demonstrated that *S. enterica* can colonise on plants such as sprouted seeds, fresh vegetables and fruits, cilantro; are not killed by surface sterilisation methods [97].

The intrinsic and extrinsic mechanisms involve the biofilm establishment. The extracellular components of *Salmonella* such as flagella, fimbriae (*Pef*), curli fimbria (*Csg*), long polar fimbriae (*Lpf*) and pili are essential equipment for initial attachment, colonisation or cell invasion. The EPS fraction is primarily made by cellulose (encoded by *bcsABZC-bscEFG*), O-antigenic capsule (O-Ag-capsule) and Lipopolysaccharide (LPS) are the sequent fragments. *CsgD* is a major control and integration unit for *Salmonella* biofilm formation which regulates the expression of specific matrix compounds; *RpoS* and *Crl* are the other two main regulators [97].

Another major contaminant in the poultry industry is *Campylobacter* spp. which has similar characteristics with the presence of diarrhoea, cramps [105]. Since *Campylobacter* is present in the industry environment, it can easily form a biofilm on foods, processing areas such as walls, floors, pipes and drains. After the irreversible fixation, *Campylobacter* forms microcolonies which take approximately 4 hours and cells become sessile. The genes *flaA*, *flaB, flaC*, *flhA*, *fliS* encode different flagellar proteins and *cheA*, *cheY* and *CetB* are the responsible for chemotaxis. The QS is activated by the expression of the luxS gene which codes autoinducer-2 synthase. The extrinsic factors such as environmental conditions, the type of available nutrients, and the attachment surface also have effects on the adhesion capacity of *Campylobacter* [106].

E. coli can form biofilms by means of their surface structure as in all members of *Enterobacteriaceae* family. *E. coli* O157: H7 is considered as an enteric pathogen which causes of serious diseases such as haemorrhagic colitis, haemolytic uremic syndrome and thrombotic purpura in humans [107]. The lowest infectious dose (<100 cells) and biofilm forming capacity are the major concerns about *E. coli* O157: H7, this means cross-contamination of foods by food contact surfaces harbouring low numbers of the pathogen can potentially lead to outbreaks [108]. The biofilm formation capacity and degree of *E. coli* O157: H7 on various types of food contact surfaces have been examined in several studies. According to recent survey results, *E. coli* O157: H7 can attach and form a biofilm on wooden surfaces at significantly higher levels compared with steel, glass and plastic and chlorine dioxide show greater lethal activity than NaOCl against *E. coli* O157: H7 in a biofilm on the same surface [108]. Similar findings were demonstrated by Adetunji and Isola [109]. *E. coli* O157: H7 is able to easily adhere and survive on meat contact surfaces even at low temperatures. The presence of other microorganisms on the surfaces is a major factor for the initial attachment. Dourou et al. [110] shown that contact surfaces in the beef fabrication facilities serve as a source of *E. coli* O157: H7 cross-contamination. The type of residual substrate and temperature effect the attachment and attachment occurred not only at a temperature representative of beef fabrication areas during nonproduction hours (15 ^{o}C), but also during cold storage (4 ^{o}C) temperatures.

4. Conclusion

Food preservation is an extensively studied topic in food science. According to these studies results, bacterial elimination can occur either extrinsically or intrinsically. Both extrinsic –influence the food environment to make it non-liveable for microorganisms- and intrinsic- limiting bacterial growth by the direct attack to bacterial- factors contribute to eliminate the microorganisms throughout the food chain. In this respect, the prevalence of microorganisms in biofilm architecture are the major contamination reservoir of both spoilage and pathogenic microflora in the food industry. As discussed above, the ability of pathogenic biofilm formation mainly depends on genetic bases and their regulation, but the properties of the substratum and bacterial cells, environmental conditions such as pH, temperature and nutrient components are the other important factors lead to biofilm formation. As suggested by Meyer [111], there is three significant points to prevent biofilm formation: (1) disinfection before biofilm formation, (2) disinfection biofilm architecture by harsh disinfectants and (3) inhibition of initial attachment by choosing proper surfaces materials which do not allow the bacterial attachment. Because of each biofilm is different due to the diversity of biofilm-forming bacteria in various food industries, the biofilm architecture should be well-analysed to determine an effective cleaning and disinfection operation. The chemical-based control [Sodium hypochlorite (NaClO), Hydrogen peroxide (H_2O_2), Ozone, Peracetic acid], some physical processes such as Hurdle technology, ultrasonication and/or concept of green technology by biological alternatives (natural antimicrobials, phages) are the promising options to inhibiting bacterial quorum sensing and biofilm formation. In this concept, take into account the biofilms must form an essential parameter, while developing the plans such as HACCP, GMP and ISO: 9000 specifications. In order to achieve biofilm-free processing systems, to perform an upgraded HACCP program is necessary for the food industries.

References

[1] WHO Estimates of the Global Burden of Foodborne Diseases 2015; http://apps.who.int/iris/bitstream/10665/199350/1/9789241565165_eng.pdf?ua=1

[2] EFSA (European Food Safety Authority), ECDC (European Centre for Disease Prevention, and Control). Trends and sources of zoonoses, zoonotic agents and food-borne. EFSA J. 2015; 13: 162.

[3] CDC (Center of Disease Control and Prevention). Estimates of Foodborne Illness in the United States. 2014; http://cdc.gov.

[4] Fu, L, Valentino, HR, Wang Y. Bacterial contamination in food production. In: Velazquez JB, editor. Antimicrobial Food Packaging, Elsevier Inc., 125 London Wall, London UK; 2016: 3: 35-43.

[5] Prado, M, Espiña, B, Fernandez-Argüelles, MT, Diéguez, L, Fuciños, P, Vial, S, Oliveira, JM, Reis, RL. 2016. Detection of food pathogens using nanoparticles. Advantages and trends. In: Velazquez JB, editor. Antimicrobial Food Packaging, Elsevier Inc., 125 London Wall, London UK; 14: 183-201.

[6] Havelaar AH, Brul S, Jong AD, Jonge RD, Zwietering MH, Kuile BHT. Future challenges to microbial food safety. International Journal of Microbiology. 2010; 139: 79-94.

[7] Abee T, Kovács AT, Kuipers OP, Veen SVD. Biofilm formation and dispersal in Gram-positive bacteria. Current Opinion in Biotechnology. 2011; 22: 172-179.

[8] Allison DG and Gilbert P. Bacterial biofilms. Science Progress. 1992; 76: 305-321.

[9] Carpentier B and Cerf O. A review: Biofilms and their consequences, with particular reference to hygiene in the food industry. Journal of Applied Bacteriology. 1993; 75: 499-511.

[10] Mattila T, Manninen M, Kylasiurola AL. Effect of cleaning-in-place disinfectants on wild bacterial strains isolated from milk line. Journal of Dairy Research. 1990; 57: 33-39.

[11] Parsek MR and Singh PK. Bacterial biofilms: an emerging link to disease pathogenesis. Annual Review of Microbiology. 2003; 57: 677 - 701.

[12] Russel P. The formation of biofilms. Milk Industry. 1993; 95(9): 10-11.

[13] Sharman M and Anand SK. Biofilms evaluation as an essential component of HACCP for food/dairy processing industry- a case. Food Control. 2002; 13: 469-477.

[14] Costerton JM and Stewart PS. Battling Biofilms. Scientific American. 2001; (285)1: 74-81.

[15] Shi X and Zhu X. Biofilm Formation and Food Safety in Food Industries. Trends in Food Science &Technology. 2009; 20: 407–413.

[16] Sauer K, Rickard AH, Davies DG. Biofilms and biocomplexity. American Society for Microbiology. 2: 2007; 347-353.

[17] Speranza B and Corbo RM. The Impact of Biofilms on Food Spoilage. The Microbiological Quality of Foodborne Spoilers, Woodhead Publishing, ISBN: 978-0-08-100503-3. 2017; 11: 259–282.

[18] Leeuwenhoek AV. Some microscopical observations about animals in the scarf of the teeth. Phil. Trans. 1684; 14: 568-574.

[19] Zobell EC and Eshter CA. The Significance of Marine Bacteria in the Fouling of Submerged Surfaces. Journal of Bacteriology. 1935; 29(2): 239-251.

[20] Zottola EA and Sasahara KC. Microbiol biofilms in the food processing industry- should they be a concern. International Journal of Food Microbiology. 1994; 23(2): 125-148.

[21] Cos P, Tote K, Horemans T, Maes L. Biofilms an extra hurdle for effective antimicrobial therapy. Cur. Parma. Design. 2010; 16: 2279-2295.

[22] Simoes M. Antimicrobial strategies effective against infectious bacterial biofilms. Current Medicinal Chemistry. 2011; 18: 2129-2145.

[23] Akbas MY. Bacterial biofilms and their new control strategies in food industry. In: Méndez-Vilas A, editor. The Battle against Microbial Pathogens: Basic Science, Technological Advances and Educational Programs. Formatex Research Centre; 2013. 114-123.
[24] Sokunrotarak S, Jahid KI, Ha SD. Biofilm Formation in Food Industries: A food safety concern. Food Control. 2013; 31: 572–585.
[25] Bridier A, Sanches-Vizuete P, Guilbaud M, Piard JC, Naïtali M. Biofilm-associated persistence of foodborne pathogens. Food Microbiology. 2015; 45: 167-178.
[26] Stoodly P, Cargo R, Rupp CJ, Wilson S, Klapper I. Biofilm material properties as related to shear-induced deformation and detachment phenomena. Journal of Industrial Microbiology and Biotechnology. 2002a; 29(6): 361-367.
[27] Briandet R, Leriche V, Carpentier B, Bellon-Fortaine MN. Effects of the growth procedure on the surface hydrophobicity of *Listeria monocytogenes* cells and their adhesion to stainless steel. Journal of Food Protection. 1999a; 62: 994-998.
[28] Latasa C, Roux A, Toledo-Arana A, Ghigo JM, Gamazo C, Penades JR, Lasa I. BapA, a large secreted protein required for biofilm formation and host colonisation of *Salmonella enterica* serovar enteritidis. Molecular Microbiology. 2005; 58: 1322-1339.
[29] Van Houdt R and Michiels CW. Biofilm formation and food industry, a focus on the bacterial outer surface. Journal of Applied Microbiology. 2010; 109: 1117-1131.
[30] Donlan RM. Biofilms: microbial life on surfaces. Emerging Infectious Disease. 2002; 8(9): 881-890.
[31] Sinde E and Carbolla J. Attachment of *Salmonella* spp. and *Listeria monocytogenes* to stainless steel, rubber, and polytetrafluorethylene: the influence of free energy and the effect of commercial sanitizers. Food Microbiology. 2000; 17: 439-447.
[32] Costeton JW, Lewandowski Z, Debeer D, Caldwell D, Korber D. Biofilms, the customised microniche. Journal of Bacteriology. 1994; 176(8): 2137-2142.
[33] Stoodley P, Sauer K, Davies DG, Costerton JW. Biofilms as complex differentiated communities. Annual Review of Microbiology. 2002b; 56: 187-209.
[34] Miller MB, Bassler BL. Quorum sensing in bacteria. Annual Review of Microbiology. 2001; 55: 165-199.
[35] Annous BA, Fratamico PM, Smith JL. Quorum sensing in biofilms: why bacteria behave the way they do. Journal of Food Science. 2009; 74: 24-37.
[36] Yang L, Hu Y, Liu Y, Zhang J, Ulstrup J, Molin S. Distinct roles of extracellular polymeric substances in *Pseudomonas aeruginosa* biofilm development. Environmental Microbiology. 2011; 1705-1717.
[37] Cotter PA, Stibitz S. c-di-GMP-mediated regulation of virulence and biofilm formation. Current Opinion in Microbiology. 2007; 10: 17-23.
[38] Ahmad I, Wigren E, Le Guyon S, Vekkeli S, Blanka A, El Mouali Y, Anwar N, Chuah ML, Lunsdorf H, Frank R, Rhen M, Liang ZX, Lindqvist Y, Romling U. The EAL-like protein STM 1697 regulates virulence phenotypes, motility and biofilm in *Salmonella typhimurium*. Molecular Microbiology. 2013; 90: 1216-1232.
[39] Arvidson S and Tegmark K. Regulation of virulence determinants in *Staphylococcus aureus*. International Journal of Medical Microbiology. 2001; 291: 159-170.
[40] Kon KF, Vuong C, Otto M. *Staphylococcus* quorum sensing in biofilm formation and infection. International Journal of Medical Microbiology. 2006; 296: 133-139.
[41] Linback T, Mols M, Basset C, Granum PE, Kuipers OP, Kovacs AT. CodY, a pleiotropic regulator, influences multicellular behaviour and efficient production of virulence factors in *Bacillus cereus*. Environmental Microbiology. 2012; 14: 2233-2246.
[42] Davies DG, Parsek MR, Pearson JP, Iglewski BH, Costerton JW, Greenberg EP. The environmental of cell-to-cell signals in the development of a bacterial biofilm. 1998; Science 280: 295-298.
[43] Boles BR, Thoendel M, Singh PK. Self-generated diversity produces 'insurance effects' in biofilm communities. Proceedings of the National Academy of Sciences of the United States of America. 2004; 101: 16630- 16635.
[44] Kim TJ, Young BM, Young GM. Effect of flagellar mutations on *Yersinia enterolitica* biofilm formation. Applies Environmental Microbiology. 2008; 74: 5466-5474.
[45] Garcia-Gonzalo D and Pagán R. Influence of environmental factors on bacterial biofilm formation in the food industry: a review. Journal of Postdoctoral Research. 2015; 3(6): 3-13.
[46] van der Veen S and Abee T. Mixed species biofilms of *Listeria monocytogenes* and *Lactobacillus plantarum* show enhanced resistance to benzalkonium chloride and peracetic acid. International Journal of Microbiology. 2011; 144: 421-431.
[47] Nilsson RE, Ross T, Bowman JP. Variability in biofilm production by *Listeria monocytogenes* correlated to strain origin and growth conditions. International Journal of Food Microbiology. 2011; 150: 14-24.
[48] Rieu A, Briandet R, Habimana O, Garmyn D, Guzzo J, Piveteau P. *Listeria monocytogenes* EGD-e biofilms: no mushrooms but a network of knitted chains. Applied Environmental Microbiology. 2008; 74: 4491-4497.
[49] Lemon KP, Higgins DE, Kolter R. Flagellar motility is a critical for *Listeria monocytogenes* biofilm formation. Journal of Bacteriology. 2007; 189: 4418-4424.
[50] Houry A, Briandet R, Aymerich S, Gohar M. Involvement of motility and flagella in *Bacillus cereus* biofilm formation. Microbiology. 2010; 156: 1009-1018.
[51] Carpentier B and Cerf O. Persistence of in food industry equipment and premises. International Journal of Food Microbiology. 2011; 145: 1-8.
[52] Botticella G, Russo P, Capozzi V, Amodio ML, Massa S, Spano G, Beneduce L. *Listeria monocytogenes*, biofilm formation and fresh-cut produce. In: Méndez-Vilas A, editor. Microbial pathogens and strategies for combating them: science, technology and education. Formatex Research Center; 2013. 114-123.
[53] Alonso AN, Perrye KJ, Regan PM, Higgins DE. Identification of *Listeria monocytogenes* determinates required for biofilm formation. PLoS One 9. 2014; 12: https://doi.org/10.1371/journal.pone.0113696.
[54] Piercey MJ, Hingston PA, Hansen LT. Genes involved in *Listeria monocytogenes* biofilm formation at a simulated food processing plant temperature of 15 ^{o}C. International Journal of Food Microbiology. 2016; 223: 63-74.

[55] Harmsen M, Lappann M, Knøchel S, Molin S. Role of extracellular DNA during biofilm formation by *Listeria monocytogenes*. Applied Environmental Microbiology. 2010; 76: 2271-2279.
[56] Rodríguez A, Autio W, Mclandsborough LA. Effects of contact time, pressure, percent relative humidity (%RH) and Material type on Listeria biofilm adhesive strength at a cellular level using atomic force microscopy (AFM). Food Biophysics. 2008; 3: 305-311.
[57] Di Bonaventura G, Piccolomini R, Paludi D, D'Orio V, Vergara A, Conter M, Ianieri A. Influence of temperature on biofilm formation by *Listeria monocytogenes* on various food contact surfaces: relationship with motility and cell surface hydrophobicity. Journal of Applied Microbiology. 2008; 104: 1552-1561.
[58] Choi NY, Kim BR, Bae YM, Lee SY. Biofilm formation, attachment and cell hydrophobicity of foodborne pathogens under varied environmental conditions. Journal of Korean Society of Applied Biological Chemistry. 2013; 56(2): 207-220.
[59] Xu H, Zou Y, Lee HY, Ahn J. Effect of NaCl on the biofilm formation by foodborne pathogens. Journal of Food Science. 2010; 75(9): 580-585.
[60] Pilchová T, Hernould M, Prévost H, Demnerová K, Pazlarová J, Tresse O. Influence of food processing environments on structure initiation of static biofilm on *Listeria monocytogenes*. 2014; 35: 366-372.
[61] Tresse O, Lebret V, Benezech T, Faille C. Comparative evaluation of adhesion, surface properties and surface composition of *Listeria monocytogenes* strains after cultivation at constant pH of 5 and 7. Journal of Applied Microbiology. 2006; 101(1): 53-62.
[62] Smoot LM and Pierson MD. Influence of environmental stress on the kinetics and strength of attachment of *Listeria monocytogenes* Scott A and Buna-N rubber and stainless steel. Journal of Protection. 1998; 61(10): 1286-1292.
[63] Tresse O, Lebret V, Garmyn D, Dussurget O. The impact of growth history and flagellation on the adhesion of *Listeria monocytogenes* various strains to polystyrene. Canadian Journal of Microbiology. 2009; 55(2): 189-196.
[64] Herald PJ and Zottola EA. Attachment of *Listeria monocytogenes* to stainless steel surfaces at various temperatures and pH values. Journal of Food Science. 1988; 53(5): 1549-1562.
[65] Moltz AG and Martin SE. Formation of biofilms by *Listeria monocytogenes* under various growth conditions. Journal of Food Protection. 2005; 68(1): 92-97.
[66] Sharma M, Anand SK. Characterization of constitutive microflora of biofilms in dairy processing lines. Food Microbiology. 2002; 19: 627-636.
[67] Salo S, Ehavald H, Raaska L, Vokk R, Wirtanen G. Microbial surveys in Estonian dairies. LWT - Food Science and Technology. 2006; 39(5): 460-471.
[68] Lee SHI, Barancelli GV, de Camargo TM, Corassin CH, Rosim RE, da Cruz AG, Cappato LP, de Oliveira CAF. Biofilm-producing ability of *Listeria monocytogenes* isolates from Brazilian cheese processing plants. Food Research International. 2017; 91: 88-91.
[69] Daneshvar Alavi HE, T Truelstrup HL. Kinetics of biofilm formation and desiccation survival of *Listeria monocytogenes* in single and dual species biofilms with *Pseudomonas fluorescens*, *Serratia proteamaculans* or *Shewanella baltica* on food-grade stainless steel surfaces. Biofouling. 2013; 29: 1253-1268.
[70] López PR, Ibusquiza PS, Mosquera-Fernández M, López-Cabo M. *Listeria monocytogenes* – carrying consortia in food industry. Composition, subtyping and numerical characterisation of mono-species biofilm dynamics on stainless steel. International Journal of Food Microbiology. 2015; 206: 84-95.
[71] Miao J, Liang Y, Chen L, Wang W, Wang J, Li B, Li L, Chen D, Xu Z. Formation and development of *Staphylococcus* biofilm: With focus on food safety. Journal of Food Safety. 2016; 1-11.
[72] Trinetta V, Vaid R, Xu Q, Linton R, Morgan M. Inactivation of *Listeria monocytogenes* on ready-to-eat food processing equipment by chlorine dioxide gas. Food Control. 2012; 26: 357-362.
[73] Park SH, Cheon HL; Park KH, Chung MS, Choi SH, Ryu S, Kang DH. Inactivation of biofilm cells of foodborne pathogen by aerosolized sanitizers. International Journal of Food Microbiology. 2012; 154: 130-134.
[74] Chorianopoulos NG, Tsoukleris TS, Panagau EZ, Falaras P, Nychas GJE. Use of titanium dioxide (TiO_2) photocatalysts as alternative means for *Listeria monocytogenes* biofilms disinfection in food processing. Food Microbiology. 2011; 28: 164-170.
[75] Simões M, Simões LC, Machado I, Pereira MO, Vieira MJ. Control of flow-generated biofilms with surfactants: evidence of resistance and recovery. Bioproducts. 2006; 84(4): 338-345.
[76] Burt S, Essential oils: their antibacterial properties and potential applications in foods. International Journal of Microbiology. 2004; 94: 223-253.
[77] Woo J and Ahn J. Probiotic-mediated competition, exclusion and displacement in biofilm formation by foodborne pathogens. Letters in Applied Microbiology. 2013; 56(4): 307-313.
[78] Minei CC, Gomes BC, Ratti RP, D'Angelis CEM, De Martinis ECP. Influence of peroxyacetic acid and nisin and coculture with *Enterococcus faecium* on *Listeria monocytogenes* biofilm formation. Journal of Food Protect. 2008; 71: 634-638.
[79] Sillankorva S, Oliveira DR, Vieira MJ, Sutherland IW, Azeredo J. Bacteriophage ϕ S1 infection of *Pseudomonas fluoresecens* planktonic cells versus biofilms. BioFouling. 2004; 20: 133-138.
[80] Nostro A, Guerrini A, Marino A, Tacchini M, Di Giulio M, Grandini A, Akin M, Cellini, Bisignano G, Saraçoğlu HT. In vitro activity of plant extracts against biofilm-producing food-related bacteria. International Journal of Food Microbiology. 2016; 238: 33-39.
[81] Folsom JP and Frank JF. Chlorine resistance of *Listeria monocytogenes* biofilms and relationship to subtype, cell density and planktonic cell chlorine resistance. Journal of Food Protect. 2006; 69(6): 1292-1296.
[82] Archer NK; Mazaitis MJ, Costerton JW, Leid JG, Powers ME, Shirtliff ME. *Staphylococcus aureus* biofilms: Properties, regulation and roles in human disease. Virulence. 2011; 2 (5): 445-459.
[83] Arciola CR, Campoccia D, Speziale P, Montanaro L, Costerton JW. Biofilm formation in *Staphylococcus* implant infections. A review of molecular mechanisms and implications for biofilm resistant materials. Biomaterials. 2012; 33: 5967-5982.
[84] Qin Z, Ou Y, Yang L, Zhu Y, Tolker-Nielsen T, Molin S. Role of autolysin-mediated DNA release in biofilm formation of *Staphylococcus epidermis*. Microbiology. 2007; 153: 2083-2092.

[85] Lee SHI, Mangolin BLC, Gonçalves JL, Neff DV, Silva MP, Cruz AG, Oliveira CAF. Biofilm-producing ability of *Staphylococcus aureus* isolates from Brazilian dairy farms. Journal of Dairy Science. 2013; 97:1812-1816.
[86] Lopez LAA, Rodrigues JBDS, Magnani M, de Souza EL, de Siqueira-Júnior JP. Inhibitory effects of flavonoids on biofilm formation by *Staphylococcus aureus* that overexpresses efflux protein genes. Microbial Pathogenesis. 2017; 107: 193-197.
[87] Ramírez MD, Smid EJ, Abee T, Groot MNN. Characterisation of biofilms formed by *Lactobacillus plantarum* WCFS1 and food spoilage isolates. International Journal of Food Microbiology. 2015; 207: 23-29.
[88] Simões M, Simões LC, Vieria M A. Review of Current and Emergent Biofilm Control Strategies. LWT - Food Science and Technology. 2010; (43)4: 573-583.
[89] Ladero MDV, del Rio B, Redruello B, Fernández M, Martin MC, Alvarez MA. Biofilm-forming capacity in biogenic amine-producing bacteria isolated from dairy products. Frontiers in Microbiology. 2016; 7: 1-10.
[90] Langsrud S. Biofilm formation by Gram-positive bacteria including *Staphylococcus aureus*, *Mycobacterium avium*, and *Enterococcus* spp. in food processing environments. In Biofilms in the Food and Beverage Industries, Editors Fratamico P, Annous B, Gunther N. Cambridge: Woodhead Publishing, 2009; 250-269.
[91] da Silva Fernandes M, Kabuki DY, Kuaye AY. Biofilms of *Enterococcus faecalis* and *Enterococcus faecium* isolated from the processing of ricotta and the control of these pathogens through cleaning and sanitization procedures. International Journal of Food Microbiology. 2015; 200: 97-103.
[92] Zang QQ, Ye KP, Wang HH, Xiao HM, Xu XL, Zhou GH. Inhibition of biofilms of *Pseudomonas aeruginosa* by an acylated homoserine lactones-containing culture extract. LWT- Food Science and Technology. 2014; 57: 230-235.
[93] Hassett DJ, Korfhagen TR, Irvin RT, Schurr MJ, Sauer K, Lau GW. *Pseudomonas aeruginosa* biofilm infections in cystic fibrosis: Insights into pathogenic processes and treatment strategies. Expert Opinion on Therapeutic Targets. 2010; 14(2): 117-130.
[94] Venturi V. Regulation of quorum sensing in *Pseudomonas*. FEMS Microbiology Reviews. 30(2): 274-291.
[95] Charlebois A, Jacques M, Boulianne M, Archambault M. Tolerance of *Clostridium perfringens* biofilms to disinfectants commonly used in the food industry. Food Microbiology. 2017; 62: 32-38.
[96] Carrasco E, Morales-Rueda A, García-Gimeno RM. Cross-contamination and recontamination by *Salmonella* in foods: A review. Food Research International. 2012; 45: 545-556.
[97] Steenackers H, Hermans K, Vanderleyden J, De Keersmaecker SJD. *Salmonella* biofilms: An overview on occurrence, structure, regulation and eradication. Food Research International. 2012; 45: 502-531.
[98] Stepanovic S, Cirkovic I, Ranin L, Svabic-Vlahovic M. Biofilm formation by *Salmonella* spp. and *Listeria monocytogenes* on plastic surface. Letters in Applied Microbiology. 2004; 38(5): 428-432.
[99] Vestby LK, Moretro T, Langsrud S, Heir E, Nesse LL, Biofilm forming abilities of *Salmonella* are correlated with persistence in fish meal and food factories. BMC Veterinary Research. 2009; 5: 20.
[100] Barker J and Bloomfield SF. Survival of *Salmonella* in bathrooms and toilets in domestic homes following salmonellosis. Journal of Applied Microbiology. 2000; 89(1): 137-144.
[101] Joseph B, Otta SK, Karunasagar I, Karunasagar I. Biofilm formation by *Salmonella* spp. on contact surfaces and their sensitivity to sanitizer. International Journal of Food Microbiology. 2001; 64: 367-372.
[102] Díez-García M, Capita R, Alonso-Calleja C. Influence of serotype on the growth kinetics and the ability to form biofilms of *Salmonella* isolates from poultry. Food Microbiology. 2012; 31: 173-180.
[103] Nair A, Rawool DB, Daijad S, Poharkar K, Mohan V, Barbuddhe SB, Kolhe R, Kurkure NV, Kumar A, Malik SVS, Balasaravanan T. Biofilm formation and genetic diversity of *Salmonella* isolates recovered from clinical, food, poultry and environmental sources. Infection, Genetics and Evolution. 2015; 36: 424-433.
[104] Yang Y, Mikš-Krajnik M, Zheng Q, Lee SB, Lee SC, Yuk HG. Biofilm formation of *Salmonella* Enteritidis under food-related environmental stress conditions and its subsequent resistance to chlorine treatment. Food Microbiology. 2016; 54: 98-105.
[105] Rossi DA, Melo RT, Mendonça EP, Monteiro GP, Biofilms of *Salmonella* and *Campylobacter* in the poultry industry. In Poultry Science, Edited Malafi M. InTech, 2017; 94-112.
[106] Moe KK, Mimura J, Ohnishi T, Wake T, Yamazaki W, Nakai M, Misawa N. the mode of *Campylobacter jejuni* biofilm formation on smooth surfaces by. Journal of Veterinary Medical Science. 2010; 72: 411-416.
[107] Doyle MP, *Escherichia coli* O157:H7 and its significance in foods. International Journal of Food Microbiology. 1991; 12: 289-301.
[108] Bang J, Hong A, Kim H, Beuchat LR, Rhee MS, Kim Y, Ryu JH. Inactivation of *Escherichia coli* O157:H7 in biofilm on food-contact surfaces by sequential treatments of aqueous chlorine dioxide and drying. International Journal of Food Microbiology. 2014; 191: 129-134.
[109] Adetunji VO, Isola TO, Crystal violet binding assay for assessment of biofilm formation by *Listeria monocytogenes* and *Listeria* spp. on wood, steel and glass surfaces. 2011; 6: 6-10.
[110] Dourou D, Beauchamp CS, Yoon Y, Geornaras I, Belk KE; Smith G, Nychas GJE, Sofos JN. Attachment and biofilm formation by *Escherichia coli* O157:H7 at different temperatures, on various food-contact surfaces encountered in beef processing. International Journal of Food Microbiology. 2011; 149: 262-268.
[111] Meyer B. Approaches to prevention, removal and killing of biofilms. International Biodeterioration & Biodegradation. 2003; 51(4): 249-253.