academicJournals

Vol. 11(26), pp. 1051-1060, 14 July, 2017
DOI: 10.5897/AJMR2016.8180
Article Number: 93FC33965161
ISSN 1996-0808

http://www.academicjournals.org/AJMR

African Journal of Microbiology Research

Review

Antibiofilm activity of natural substances derived from plants

José Walter Araújo Nogueira*, Renata Albuquerque Costa, Magda Turini da Cunha and Theodora Thays Arruda Cavalcante

Núcleo de Bioprospecção e Experimentação Molecular Aplicada - NUBEM. Faculdades INTA, CE, Brasil.

Received 23 June, 2016; Accepted 9 June, 2017

The main objective of this study was to conduct a review of bioactive substances derived from plants which show antimicrobial/antibiofilm properties. Vegetable species were widely employed as ingredients in medicine based on traditional knowledge. Various secondary metabolites have been proven to inhibit bacterial growth. Bacterial resistance mechanisms have increased over the years. Biofilms are types of bacterial association which gives the communities a higher resistance to drugs. The formation process of biofilms, the problems caused by them and the natural substances, as well as their main chemical components and action mechanisms, have been described according to existing literature.

Key words: Natural product, antimicrobial activity, biofilm.

INTRODUCTION

Pathogenic bacteria present a number of defense mechanism against antimicrobial agents, moreover, their resistance to drugs presently available in the market is increasing. Among the bacterial defense mechanisms, formation of bio-films is one that is responsible for a fair amount of chronic diseases and show extreme resistance to antibiotics and to the host's defense system (Lewis, 2001; Fux et al., 2003; Rozalski et al., 2013).

Biofilms are a more resistant form of bacterial life when compared with free-living planktonic form. Its resistance is directly related to the natural survival characteristics of microbial cells living in such communities, among which we highlight the slower growth of cells associated with biofilm, as opposed to the free-living microbial cells, and low regulation of cellular process, mainly caused by the more restricted contact of the cells in the interior of the biofilm with external nutrients. Besides that, these bacteria produce an extracellular polysaccharidic matrix that hinders the action of antimicrobial agents, collaborating even further to its resistance, since this matrix acts like a diffusion barrier against small molecules (Anderson and O'Toole, 2008; Hall-Stoodley and Stoodley, 2009).

Currently, natural compounds have emerged as potential candidates given the biotechnological focus in the search for antimicrobial and antibiofilm drugs

*Corresponding author. E-mail: nogueira.jwa@gmail.com.

(Schachter, 2003). The relevance of these researches is due to the large role played by biofilms in the etiology of a vast amount of persistent and chronic human diseases (Simões et al., 2010).

The observation of microbial worlds by different techniques of microscopy has made possible to researchers, throughout the years, to observe micro-organisms arranged in communities, sharing nutrients, metabolites, genetic elements and, thus, becoming able to resist environmental turbulence, causing diseases difficult to eradicate. Biofilms impacts men in a variety of ways, being formed in natural environments, medical instruments and industrial machines (Al-Bakri et al., 2010; Lopez et al., 2010).

In 1847, Leuwenhoek used a primitive microscope and described animalcules on a sample scrapped from human teeth. Almost 100 years later, in 1934, Claude Zobell, examining marine populations directly under the microscope, concluded that these bacteria were adhered to surfaces, forming sessile populations (Zobell, 1943). Between 1935 and 1978, microbiologists Ron Gibbons and Van Houte of the Forsyth Dental Center examined microbial biofilms that constitute dental plaque. The first stage of a biofilm formation in pure culture was observed in 1964, when it was established that the state of irreversible adhesion of microorganisms to a surface is the first in the formation of these microbial communities (Costerton, 1999).

More than seventy years after the first account of biofilm (Zobell, 1943), they continue to be a source of preoccupation to a wide array of activities, specifically food industry, environment and biomedics (Flint et al., 1997; Maukonen et al., 2003; Sihorkar and Vyas, 2001; Veran, 2002). Based on observations of dental plaque and sessile communities in mountain streams, Costerton and collaborators presented in 1987 a theory of biofilms that explained the mechanisms through which the microorganisms adhere to living or inert surfaces, and what benefits they receive from living in these communities.

According to Hoiby et al. (2010), a biofilm is a structured consortium of bacteria capable of producing a polymeric matrix which consists of polysaccharides, proteins and DNA. These communities can be established on a wide variety of surfaces (Abee et al., 2011). Aside from the ability to produce extracellular biopolymers, cells in community show, in a high or lower degree, a decreased growth pattern and specifically regulated genes. The organization of microorganisms in biofilms occurs naturally, for living in these communities considerably raises the chances of survival for these microscopic beings. The production of extracellular polymeric substances by microorganisms is accepted as a key mechanism in making irreversible the cellular adhesion to inanimate surfaces in watery environments, resulting in the development of a biofilm (Beech et al., 2005).

Bacterial biofilms are closely related to human health problems; they are responsible for many infectious diseases acquired from inert surfaces, including medical instruments for internal and external use. They might also be present in the water tubing of hospitals, leading to infections after internment (Bordi and de Bentzmann, 2011). It is important to highlight that the formation of biofilm in medical material, such as catheters or implants, result in chronic infections difficult to treat (Donlan, 2008; Hall-Stoodley et al., 2004; Hatt and Rather, 2008).

Since the first observations using confocal microscopy, it became evident that living mature biofilms are not single structured layers of microbial cells on a surface. On the contrary, they appear as heterogeneous entities in time and space, constantly changing as a result of external and internal processes (Dolan and Costerton, 2002). A biofilm might be formed of a single species of bacteria or fungus; however, it might consist of various bacterial, fungal species and even algae and protozoans (Batoni et al., 2003). An example of monospecific biofilm is those formed in cardiac valves of patients with infectious endocarditis, composed of *Staphylococcus epidermidis* (Butany et al., 2002). Besides that, infections have been associated with the formation of biofilms in human organic surfaces such as teeth, skin and urinary tract (Hatt and Rather, 2008). This organization in communities offers the microorganisms resistance to many antimicrobials, protection from protozoan attacks and hosts defenses (Anderson and O'Toole, 2008; Matz and Kjelleberg, 2005).

Currently, it is known that in natural environments, 95-99% of microorganisms exist in biofilm form (Nikolaev and Plakunov, 2007). These communities protect their microbial inhabitants not only from oxygen, but also from consequences of other damaging environmental factors (Paerl and Pinckney, 1996). Bacteria in biofilm may cause chronic infections (Costerton et al., 2003) that are characterized by persistent inflammation and tissue damage (Bjarnsholt et al., 2009). Chronic infections, including foreign body infections, are persistent despite antibiotic therapy, innate and adaptive immune system and inflammatory response of the host. In contrast to colonies, they show an immune response and pathological persistence (Hoiby et al., 2010).

These communities present an unique profile also because they shelter different species in a structure in which they are able to cooperate preferentially rather than compete (Bordi and de Bentzmann, 2011). They constitute microbial societies with their own set of social rules and behavior patterns, including altruism and cooperation, favoring group success (Parsek and Greenberg, 2005; Shapiro, 1998) by replicating behaviors on one side and competition (Velicer, 2003) on another. Certain subpopulations may show specialization; these behavioral patterns are orchestrated by chemical

communications (Weigel et al., 2007). Thus, they constitute a unique form of interaction between species, inducing drastic changes in symbiotic relations between their components (Hansen et al., 2007).

MECHANISMS OF BIOFILM FORMATION

The understanding of molecular bases in the creation of biofilms has been favored by improvements in the genetic and genomic methods and the development of visualization techniques that reveal the processes involved in the growth, physiology and adaptation of microorganisms to this life condition. A plethora of systems allows bacteria to identify and anchor to proper surfaces, and adhere one to another in order to form multicellular communities (Bordi and de Bentzmann, 2011).

Bacterial growth in pure cultures has been the main approach to microbiological culture, from pasteur to the present day. This kind of experiments are efficient in furnishing knowledge and understanding of the prokaryotic genetics and metabolism, and have simplified the isolation and identification of pathogens of a number of diseases (Costerton et al., 1987).

When it became evident that the behavior of bacteria associated on surfaces could not be predicted from microorganisms cultured in suspension, in their planktonic form, a new term to describe sessile microbial populations was introduced in the researches with biofilm (Jakubovics and Kolenbrander, 2010).

The formation of biofilm can be considered a protection mechanism for the bacterial community against external injury, thus, it seems reasonable that specific extracellular signals regulate the activation of metabolic patterns which set off the establishment of biofilms. This external signalization may come from diverse sources: they can be reproduced and secreted by the community itself, where there are molecules designed as autoinducers, which accumulate in the extracellular medium, their concentration correlated to the population density (Lopez et al., 2010), and that in high concentration might set off chains of signalization that lead to multicellular responses in bacterial population. This mechanism of cell-cell communication (called quorum sensing) controls a large number of processes, including those related to the formation of biofilm (Camilli and Basssler, 2006).

Each bacterial species has its own set of tools to perform adhesion, and there is a large number of different molecules for each species that can be used antagonistically or synergically, depending on the situation (Hagan et al., 2010).

The formation process of biofilms has been extensively described (Dolan and Costerton, 2002; Costerton et al., 1995; Habash and Reid, 1999). It is a process that follows many stages: the initial reversible adhesion of planktonic cells to a surface, followed by a maturation phase. The initial adhesion is based on attraction and repulsion forces between the cells and the surface. These forces include electrostatic and hydrophobic interactions, Van der Waals forces and hydrodynamic forces, in adequate temperatures (Agarwal et al., 2010). After the adhesion to the surface, the bacteria grow and divide, forming dense agglomerates which are characteristic of biofilm. This phase is associated with the production of polysaccharides by the bacterial cells, that become irreversibly adhered to the substrate. After a while, microcolonies develop in a mature biofilm, acquiring a typical architecture, with mushroom-shaped projections, separated by channels filled with fluids. The final stage (dispersion) consists of the detachment of unitary cells or cellular groups from the mature biofilm, it is considered an essential stage in bacterial dissemination (Batoni et al., 2003).

Considering cellular hydrophobicity and the presence of frimbriae and flagella, the production of EPS (extracellular polymeric substance) is one of the main factors that influences the rate and degree of adhesion of microbial cells to different surfaces, aside from protecting against environmental stress and dehydration (Vu et al., 2009). Thus, EPS production has been the topic of many researches to prevent the process of formation and maturation of these microbial communities (Murray et al., 2009; Nagorska et al., 2010).

The initial colonizers interact with the surface through weak interactions, mostly Van de Waals-type forces. If these microorganisms are not removed from the surface by mechanical or chemical action, they might anchor permanently through cellular adhesion molecules, such as pili and flagella (Boks et al., 2008; Hermansson, 1999). The adhesion of microorganisms to adjacent soft tissues is determined by the existence of adhesion molecules (adhesins), fixed by specific receptors, commonly simple sugars (Pereira et al., 2010).

The first microcolonies create a substratum and propitious environment for the arrival of other cells through adhesion sites and start to build the matrix which will form the biofilm. Only some species are able to adhere to a surface itself, others may anchor to the matrix or to preexisting colonies. Once the colonization has started, the biofilm develops in a combination of cellular division and recruitment of other cells (Carneiro et al., 2010).

BIOFILM RESISTANCE AND ANTIMICROBIAL MEASURES

Many human diseases are related to bacterial biofilms, including cavities, periodontitis, endocarditis and prostatitis. Biofilms have been described as bacterial mechanisms of persistence and resistance to anti-

microbial agents, unlike those found in free-living cells (Namasivayam and Roy, 2013). Various streptococcus infections, specially chronic infection, might be related to the formation of bacteria in biofilms (Al-Dhabi et al., 2012). The development of biofilms grants high resistance to the microorganisms, due to their involution in a matrix of extracellular polymeric substances, forming a barrier which stops or hampers the diffusion of antimicrobial substances through the colony (Trentin et al., 2011a, b). The ideal form of avoiding their action in pathogenic processes is to stop their development (Rozalski et al., 2013) in order to do that, the investment in the search of products and methods that interfere with the bacterial accumulation through adhesion control, inhibition of interbacterial communication or the establishment of the polysaccharidic matrix is needed (Jakubovics and Kolenbrander, 2010).

Among the mechanisms that grant resistance to antimicrobial measures are the bacterial quorum sensing, a form of communication and cooperation for the execution of different behaviors, including the production of toxins and the biofilm formation or the efflux pumps, a mechanism of active pumping of biomolecules to the extracellular medium, responsible for the resistance of Gram-negative bacteria to the majority of natural products (Savoia, 2012).

The oral cavity is colonized by a rich collection of beneficial microorganisms that live in harmony with the host, an advantageous situation to both parties. In this context, products designed for oral heath should preferentially aim to control the level of plaque instead of eliminating it, so that the beneficial properties of the resident microflora are maintained (Marsh, 1992, 2010).

Some measures taken to prevent the adherence of bacteria to biotic and abiotic surfaces and, consequentially, the formation of biofilm have been studied, these antimicrobial measures comprise experiments on the inhibition or death of the aim-bacteria driven by tests for the determination of the minimal inhibitory concentration (MIC) and minimal bactericidal concentration (MBC). These techniques are applied in tests with bacteria in their planktonic form. Nevertheless, for clinical use, the realization of tests using biofilm formation techniques or even evaluation of mature biofilm is more predictive.

Biofilm control mechanisms include the use of adhesin analogs, antibody for key epitopes and peptides that block specific sites. The interruption of the quorum sensing mechanisms is another possible goal for biofilm therapy (Njoroge and Sperandio, 2009). Such measures might turn bacteria present in biofilm more susceptible to antimicrobials or lessen its pathogenicity (Bjarnsholt et al., 2005).

A large variety of agents has been formulated so as to increase the potential control of biofilm such as: antibiotics, quaternary ammonium compounds, and chlorhexidine gluconate and acetate (Baehni and Takeuchi, 2003; Oliveira et al., 1998). Chlorhexidine shows good substantivity, with an ample spectrum of activity against Gram-positive, negative bacteria and yeast (Kleerebezem et al., 1997; Mukamolova et al., 1998).

Chlorhexidine found application in periodontal treatments, dermatological infections, skin wounds, eye and throat infections and endodontics (Teixeira and Cortes, 2005). Its frequent and prolonged use presents many collateral effects, such as changes in the taste of food and a burning feeling on the tip of the tongue (Greenberg et al., 2008; More et al., 2008; Porto et al., 2009).

Some factors have to be considered while choosing an antimicrobial substance, such as toxicity, low permeability, retentivity and the capacity of maintaining the balance of the microbiota in the mouth cavity (Cury, 2003). Another important consideration is the administration method of the anti-plaque agents. Their liberation in the oral cavity can be made by mouth washing, sprays, toothpaste, gel or vehicles of prolonged liberation, such as varnishes (Scheie, 2007).

CHEMICAL CONSTITUENTS AND BIOLOGICAL ACTIVITY OF NATURAL SUBSTANCES

Considering the previous observations, the use of natural products to promote health is as old as human civilization and, for a long time, mineral, vegetable and animal products constituted the therapeutic arsenal at hand (Eisenberg et al., 1998). Although, the presence of antimicrobial substances in higher plants is not a recent fact, only from the discovery of penicillin onwards the search has received a greater impulse (Coelho et al., 2004; Tavares, 1996).

Plants have many secondary metabolic pathways that originate various compounds, such as alkaloids, flavonoids, isoflavonoids, tannins, coumarins, glycosides, terpenes and polyacetylenes, that are often specific to certain families, genera or species and whose functions, until not long ago, were unknown (Cowan, 1999; Savoia, 2012; Simões et al., 2007). As researches progressed, these substances were found to be important in the defense mechanism of plants against their predators: fungi, bacteria, viruses, parasites, insects, mollusks or higher animals (de Lima et al, 2006).

Researches concerning the biodiversity of the Brazilian flora appear to be extremely promising sources for the discovery of new substances that might be used in the treatment of diseases. Even if the accounts in literature are few (Aburjal et al., 2001; Aqil et al., 2005; Nascimento et al., 2000), the evaluation of the synergic action between natural products and antibiotics currently used in medical clinic show auspicious signs in the attempt of

minimizing the effect of resistant strains or even the ability of microorganisms organized in biofilms resisting antimicrobial measures.

Natural products derived from medicinal plants have been found to be abundant sources of biologically active compounds, many of which became the base in the development of new chemical products which may lead to a later pharmaceutical output. With respect to diseases caused by microorganisms, the increase in the resistance of many common pathogens to therapeutic agents commonly used, such as antibacterials and antivirals, has lead to a renewed interest in the discovery of new anti-infection compounds. Since there are approximately 500,000 species of plants, distributed all over the planet, and only 1% of which being phytochemically investigated, there is a great potential for finding new bioactive compounds (Palombo, 2011).

In the last years, a great scientific progress has occurred concerning chemical and pharmacological studies of plants aimed at obtaining new compounds with therapeutic properties (Cechinel-Filho and Yunes, 1998). Among the therapeutic agents derived from plants, essential oils - also called volatile or ethereal- occupy a preponderant position.

From a chemical point of view, essential oils are complex mixtures of volatile, lipophilic, generally odorific and liquid substances. They are extracted from various plant parts (flowers, inflorescences, seeds, leafs, sticks, shells, fruits and roots) by specific processes; they possess a frequently pleasant odor and are colorless or lightly yellow when recently extracted, with an oily appearance. Their main characteristic is their volatibility, which distinguish them from the fixed oils, mixtures of lipidic substances, generally extracted from seeds (Simões et al., 2003). These denominations derive from some of their physical-chemical properties, as example, are generally liquid, of an oily aspect at room temperature- hence, oil.

Volatile oils are defined as substances obtained from organs of vegetable species through steam distillation, as well as substances obtained by pressing the pericarps of citric fruits. They might also be called essential oils, ethereal oils or essences.

Another important characteristic is the pleasant and intense odor in most of the volatile oils, thereby named essences. They are also soluble in apolar organic solvents, as aether, hence their name ethereal oils, *aetheroleum* in latin. In water, the volatile oils display limited solubility, but enough to aromatize aquose solutions, named hydrolates.

Essential oils are complex mixtures of mainly terpenes (Edries, 2007) compounds, but they might include terpene hydrocarbons, simple and terpene alcohols, aldehydes, ketones, phenols, esters, ethers, oxides, peroxides, furanes, organic acids, lactones and coumarins (Simões et al., 2007). Strong *in vitro* evidences indicate that essential oils can act as antibacterial agents against a large spectrum of pathogenic bacterial strains (Burt, 2004; Nguefack et al., 2004; Schmidt et al., 2005).

The antimicrobial activity of some essential oils and isolated components was reviewed. It was observed that eugenol showed good antibacterial effectiveness in a 0.5% concentration. In the present study, after 24 h of incubation eugenol showed a statistically significant decrease in bacterial growth in concentrations of 2.5 to 0.078% and there was no significant difference in comparison of all concentrations to tests with chlorhexidine (Burt, 2004).

Carvacrol, a phenolic monoterpene and a potent antimicrobial, was effective against a biofilm formed by *Staphylococcus aureus* and *Salmonella enterica*. Non-biocidal concentrations prevent the accumulation of protein mass and interrupt the normal development of biofilm. This molecule, along with thymol, constitute the phenolic components that enable antimicrobial activities in the oregano oil (Savoia, 2012).

In 2005, Alviano and collaborators studied the antimicrobial and antibiofilm activity of the essential oil of the *Croton cajucara* and its major component linalol, observing that the effect of the oil was more potent than that of the isolated component (Alviano et al., 2005).

Jeon et al. (2011), while testing the capacity of tt-farnesol, a terpenoid that can constitute essential oils, observed its potential to affect biofilms of *S. mutans* through alteration in the proton-motive force, possibly by the interaction of lipophilic domains with the bacterial membrane, as is supposed to happen in the action against bacteria in the planktonic form; damaging the cellular functions of the membrane, and compromising the ability of *S. mutans* to produce and tolerate acids and to synthesize intra/extracellular polysaccharides (Koo et al., 2002, 2003). Data from this study suggest that the treatment with t-farnesol can later make the bacterial accumulation impossible in biofilms subjected to nutritional depletion (Jeon et al., 2011).

Recent studies reported complementary and alternative treatment options to combat *P. aeruginosa* infections. Quorum sensing inhibitors, phages, probiotics, anti-microbial peptides, vaccine antigens and antimicrobial nanoparticles have the potential to act against drug resistant strains. Unfortunately, most studies considering alternative treatment options are still confined in the pre-clinical stages, although some of these findings have tremendous potential to be turned into valuable therapeutics (Biswas et al., 2015).

It is relevant to observe that compounds derived from plants have received larger attention in the research of alternatives in the control of infections, especially those related to the formation of biofilm, which present a recalcitrant character. It is well known that there are two main reasons for the essential oils to restrain the development of resistant bacterial strains: they are

complex and constituted by a good number of compounds in varied proportions, depending on the chemotype of the plant. Therefore, even if the bacteria are able to resist the effect of one component, there are others, possibly, with different action goals to complete the global antimicrobial activity of the essential oil (Bakkali et al., 2008; Reichling et al., 2009).

ANTIBIOFILM ACTIVITY OF NATURAL SUBSTANCES

In the last years, medicinal plants have raised the attention of researchers because of their promising potential as a source of antimicrobials. In many communities around the world, the traditional medicine systems are the only means available for the treatment of infections (Savoia, 2012). Many of these agents have their activity tested in planktonic cultures. However, the activity on sessile forms of life, responsible for the gravest problems, is still understudied. Studies that evaluate antibiofilm activity should deal with the activity against consolidated biofilms and anti-adherent properties as a prophylactic measure against the formation of biofilm (Al-Bakri et al., 2010).

Efforts in the discovery of new medicine with antimicrobial properties are the basis to overcome the worldwide problem of microbial resistance. Extracts and oils obtained from plants have been used for a great variety of objectives for centuries (Jones, 1996). These objectives vary from the use of rosewood and ceder in perfume making, flavoring of beverages with lemon oil (Lawless, 1995) and the preservation of stored food (Mishra and Dubey, 1994). Particularly, the antimicrobial activity of plant oils and extracts have been in the roots of several applications, including conservation of raw and processed food, pharmaceutical products, alternative medicine and natural therapies (Lis-Balchin and Deans, 1997; Reynolds, 1996).

The search for natural products with anti-incrustation properties has been strongly encouraged for the reason that these components are not toxic to the environment. The larger part of these components was identified as terpenes, steroids, carotenoids, phenols, furanones, alkaloids, peptides and lactones. Isolates from a vast array of organisms, including sponges, coral, algae and microbes, have been studied worldwide (Viju et al., 2013).

A review of literature made by Agra et al. (2007) listed the popular use of 483 plants with bioactive potential in the Brazilian Northeastern region. Many are yet to be studied in relation to their chemical constituents and/or biological activity, but the number demonstrates the potential of the region as a source for future studies.

A study made with 24 plants empirically used by the Brazilian semiarid community for the treatment of a variety of diseases, such as skin infections, gastrointestinal disturbs, tuberculosis and urinary tract infections, showed that the trunk bark extract of *Commiphora leptophloeos*, known as Imburana in the region, showed inhibitory effect over *Staphylococcus epidermidis* in a minimal concentration of 1.0 mg/mL and decreased the formation of biofilm in 80%. Extracts of *Bauhinia acuruana* and *Pityrocarpa moniliformis* presented biofilm inhibitory activity for the same microorganism, without causing bacterial death (Trentin et al., 2011a, b).

The elimination of mature biofilm is still a very difficult task. Thus, the inhibition of the adherence of microorganisms in a way that does not involve bacterial death constitutes a new concept of antivirulence therapy. Substances that hamper the fixation of organisms, without affecting their growth, maintaining the cell in planktonic stage, turn them more susceptible to other antimicrobial agents and to the hosts immune system.

Rubus ulmifolius extracts studies using confocal microscopy revealed ellagic acid derivatives with biofilm formation inhibitory properties without bacterial growth inhibition in methicillin-sensitive *S. aureus* isolates (Fontaine et al., 2017). In 2008, Silva and collaborators investigated the *in vitro* antimicrobial action and adherence inhibition of the hydroalcoholic extract of *Rosmarinus officinalis* Linn. (rosemary) on standard strains of *Streptococcus mitis* ATCC 98811, *Streptococcus sanguinis* ATCC 10556, *Streptococcus mutans* ATCC 25175, *Streptococcus sobrinus* ATCC 27609 and *Lactobacillus casei* ATCC 7469. In this study, the extract of *Rosmarinus officinalis* Linn. was effective in the adherence inhibition of *S. mitis* ATCC 98811, *S. mutans* ATCC 25175 and *S. sobrinus* ATCC 27609 and might be used in future tests on the inhibition of biofilm formation (Silva et al., 2008).

A research carried out by the University of Barcelona, Spain Department of Genetics, in partnership with the MEDINA Foundation examined a set of 1120 natural product extracts identifying different activities, among them: inhibition of biofilm formation, detachment of mature biofilms, antimicrobial activity against planktonic cells and on biofilm component cells, using high-throughput screening (HTS), automated technology involving robotic and bioinformatics instruments to perform the pharmacologically important substances scanning. Of the 1120 extracts tested, 40 presented metabolites with potential antimicrobial action and antibiofilm against *Salmonella enteritidis* (Paytubi et al., 2017).

The casbane diterpene, isolated from the ethanolic extract of *Croton nepetaefolius,* can interact in a non-specific form with the bacterial membrane, destabilizing interactions and interfering in the cellular development. Experiments with microdilution in polystyrene plates have shown a promising activity of this compound over streptococcus species, achieving efficiency similar to that of chlorhexidine (Sá et al., 2012).

The essential oil and extracts of *Cupressus*

sempervirens were tested in their capacity of inhibiting biofilm formation. Among the studied microorganisms, *K. pneumoniae* was the most sensible strain. The methanolic extract and the essential oil showed a significant decrease in the fixation of *K. pneumoniae* in polyvinyl chloride (PVC), material used in the confection of medical catheters. The antimicrobial activity of these compounds can be mostly attributed to their phenolic constituents, α-pinene and cedrol (Selim et al., 2014).

Extracts of coconut shell fibers (*Cocos nucifera*) increased the hydrophobicity of *Pseudomonas* sp. and *Alteromonas* sp. isolated from marine biofilms, which, according to the physical-chemical theory of bacterial adhesion, decreases the adherence rate of micro-organisms (Viju et al., 2013). Silva et al. (2012) made a study of bioprospection of medicial plants in the Brazilian semi-arid region. The most promising results were obtained from extracts of *Schinopsis brasiliensis* which inhibited the formation of biofilm both in Gram-negative (*Pseudomonas aeruginosa*) as well as in Gram-positive (*Sthaphylococcus aureus*) bacteria, however, it was toxic to *Artemia salina*.

Extracts of *Humulus lupulus* (Cannabaceae), which contains xanthohumol as a major component, inhibited biofilm formation in *S. aureus* in 99.9%. Synergetic studies have shown that the addition of hop compounds decreased the value of MIC for commercial antibiotics oxacillin and linezolide from 0.125 and 0.5 to 0.094 and 0.38 µg/mL, respectively (Rozalski et al., 2013).

Trentin et al. (2011a, b) conducted a study on the effect of the filtrate of a *Cobetia marina* colony, a Gram-negative marine bacterium, on the formation of biofilm in *S. epidermidis*, an important agent in hospital-acquired infections. The filtrate does not possess the capacity of destructing consolidated biofilm; however, it inhibited its formation in 84.7%. It is possible that the action mechanism of this substance is related to its ability to alter the bacterial quorum sensing, avoiding the production of extracellular polymers and, consequently, biofilm formation.

Antibacterial agents used in the prevention and treatment of oral diseases, including cetylpyridinium chloride, chlorhexidine, fluorinated amines or products containing such agents might show undesired effects such as dental staining or, in the case of ethanol commonly found in mouthwash, in relation to the development of oral cancer (Knoll-Kohler and Stiebel, 2002; Lachenmeier, 2008; Mccullough and Farah, 2008; Neumegen et al., 2005; Rodrigues et al., 2007). Thus, the search for alternative products and phytochemical isolates in plants used in traditional medicine is considered a good alternative over synthetic drugs (Prabu et al., 2006).

The use of essential oils as antimicrobial agents is attested from a long time ago (Abee et al., 2011) and, in the specific case of oral microorganisms, mouthwashes containing essential oils have been shown beneficial and safe for daily use in oral hygiene (Claffey, 2003), depending on further studies to understand the spectrum of its action against these microorganism (Abee et al., 2011).

There are few mentions in literature about the antibiofilm activity of essential oil or their isolates on pathogenic biofilms related to oral diseases. Millezi et al. (2010) reported on the activity of cleaning detergents made of *C. citratus* and *T. vulgaris* over biofilms of *Aeromonas hydrophila*, a microorganism related to food contamination, showing decrease in the number of viable cells present in the communities subject to the substances.

Several screenings have shown that natural products, particularly phytochemicals, are an interesting source of quorum sense inhibitor (QSI) (Castillo-Juárez et al., 2015). They have been recognized as a large and attractive repository of QSI, offering a vast chemical diversity with structural complexity and biological activity (Borges et al., 2015). In fact, they resemble what is considered an “ideal” QSI, which includes being chemically stable, highly effective, low-molecular-mass molecules, and being harmless to health (Qian et al., 2013). Therefore, phytochemicals with QS inhibition activity can be promising tools to help the treatment of bacterial infections, including those that are biofilm-related, in an era where the availability of effective antibiotics is no longer guaranteed.

CONCLUSION

The rescue of the historical use of medicinal plants is inestimable in order to direct the investigations on bioprospection. A large number of plants have already been studied with regards to their antimicrobial properties. Brazil possesses a large diversity of plants mentioned with the popular use in the treatment of numerous infirmities that remain yet to be subject to a deeper scientific investigation, in order to prove their applicability, determine their bioactive substances and action mechanisms.

The focus of the study of antibiofilm properties is necessary, considering the problems these bacterial communities have caused in diverse clinical, environmental and industrial contexts. Brazil has a great potential for developing applied natural products. Thus, the use of medicinal plants reported in traditional knowledge, intertwining technology to scientifically validate it, and might become an efficient option in the treatment of various infectious diseases (Borges et al., 2016).

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Abee T, Kovács AT, Kuipers OP, van der Veen S (2011). Biofilm formation and dispersal in Gram-positive bacteria. Curr. Opin. Biotechnol. 22:172-179.

Aburjal T, Darwish RM, Al-Khalil S, Mahafzah A, Al-Abbadi A (2001). Screening of antibiotic resistant inhibitors from local plant materials against two different strains of *Pseudomonas aeruginosa*. J. Ethnopharmacol. 76:39-44.

Agarwal A, Singh KP, Jain A (2010). Medical significance and management of staphylococcal biofilm. FEMS Immunol Med Microbiol. 58:147-160.

Agra MF, França PF, Barbosa-Filho JM (2007). Synopsis of the plants known as medicinal and poisonous in Northeast of Brazil. Rev. Bras. Farmacogn. 17:114-140.

Al-Bakri AG, Othman G, Afifi FU (2010). Determination of the antibiofilm, antiadhesive, and anti-MRSA activities of seven Salvia species. Pharmacogn. Mag. 6(24):264-270.

Al-Dhabi NA, Balachandran C, Raj MK, Duraipandiyan V, Muthukumar C, Ignacimuthu S, Khan IA, Rajput VS (2012). Antimicrobial, antimycobacterial and antibiofilm properties of Couroupita guianensis Aubl. fruit extract. BMC Complement. Altern. Med. 12:242.

Alviano WS, Mendonça-Filho RR, Alviano DS, Bizzo HR, Souto-Padrón T, Rodrigues ML, Bolognese AM, Alviano CS, Souza MM (2005). Antimicrobial activity of *Croton cajucara* Benth linalool-rich essential oil on artificial biofilms and planktonic microorganisms. Oral Microbiol. Immunol. 20:101-105.

Anderson GG, O'Toole GA (2008). Innate and induced resistance mechanisms of bacterial biofilms. Bacterial Biofilms, Springer, Heidelberg: Ed Romeo T. 322:85-105.

Aqil F, Khan MS, Owais M, Ahmad I (2005). Effect of certain bioactive plant extracts on clinical isolates of ß-lactamase producing methicilin resistant *Staphylococcus aureus*. J. Basic Microbiol. 45:106-114.

Baehni PC, Takeuchi Y (2003). Anti-plaque agents in the prevention of biofilm-associated oral diseases. Oral Dis. 9(1):23-29.

Bakkali F, Averbeck S, Averbeck D, Idaomar M (2008). Biological effects of essential oil: A review. Food Chem. Toxicol. 46(2):446-475.

Batoni G, Maisetta G, Brancatisano FL, Esin S, Campa M (2011). Use of antimicrobial peptides against microbial biofilms: advantages and limits. Curr. Med. Chem. 18(2):256-279.

Beech IB, Sunner JA, Hiraoka K (2005). Microbe-surface interactions in biofouling and biocorrosion processes. Int. Microbiol. 8:157-168.

Bjarnsholt T, Jensen PØ, Burmølle M, Hentzer M, Haagensen JA, Hougen HP, Calum H, Madsen KG, Moser C, Molin S, Høiby N, Givskov M (2005). *Pseudomonas aeruginosa* tolerance to tobramycin, hydrogen peroxide and polymorphonuclear leukocytes is quorum-sensing dependent. Microbiology 151:373-383.

Bjarnsholt T, Jensen PØ, Fiandaca MJ, Pedersen J, Hansen CR, Andersen CB, Pressler T, Givskov M, Høiby N (2009). *Pseudomonas aeruginosa* biofilms in the respiratory tract of cystic fibrosis patients. Pediatr. Pulmonol. 44:547-558.

Boks NP, Norde W, van der Mei HC, Busscher HJ (2008). Forces involved in bacterial adhesion to hydrophilic and hydrophobic surfaces. Microbiology 154(10):3122-3133.

Bordi C, de Bentzmann S (2011). Hacking into bacterial biofilms: a new therapeutic challenge. Ann. Intensive Care, 19:1-8.

Borges A, Abreu AC, Dias C, Saavedra MJ, Borges F, Simões M (2016). New perspectives on the use of phytochemicals as an emergent strategy to control bacterial infections including biofilms. Molecules 21:877.

Borges A, Saavedra MJ, Simões M (2015). Insights on antimicrobial resistance, biofilms and the use of phytochemicals as new antimicrobial agents. Curr. Med. Chem. 22:2590-2614.

Burt S (2004). Essential oils: their antibacterial properties and potential applications in foods - a review. Int. J. Food Microbiol. 94:223-253.

Butany J, Scully HE, VanArsdell G, Leask R (2002). Prosthetic heart valves with silver-coated sewing cuff fabric: early morphological features in two patients. Can. J. Cardiol. 18:733-738.

Camilli A, Bassler BL (2006). Bacterial small-molecule signaling pathways. Science 311:1113-1116.

Carneiro VA, Santos HS, Arruda FV, Bandeira PN, Albuquerque MR, Pereira MO, Henriques M, Cavada BS, Teixeira EH (2010). Casbane diterpene as a promising natural agent against biofilm-associated infections. Molecules 16(1):190-201.

Castillo-juárez, Israel et al. (2015). Role of quorum sensing in bacterial infections. World J. Clin. Cases 3:575-598.

Cechinel-Filho V, Yunes RA (1998). Estratégias para a obtenção de compostos farmacologicamente ativos a partir de plantas medicinais. Conceitos sobre modificação estrutural para otimização da atividade. Quím. Nova 21(1):99-105.

Claffey N (2003). Essential oil mouthwashes: A key component in oral health management. J. Clin. Periodontol. 30:22-24.

Coelho MPG, Reis PA, Gava VB, Marques PR, Gayer CR, Laranja GA, Felzenswalb I, Sabino KC (2004). Antiarthritic effect and subacute toxicological evaluation of Baccharis genistelloides aqueous extract. Toxicol. Lett. 154:69-80.

Costerton JW (1999). Introduction to biofilm. Int. J. Antimicrob. Agents 11(3-4):217-221.

Costerton JW, Cheng KJ, Geesey GG, Ladd TI, Nickel JC, Dasgupta M, Marrie TJ (1987). Bacterial biofihns in nature and disease. Annu. Rev. Microbiol. 41:435-64.

Costerton JW, Lewandowski Z, Caldwell DE, Korber DR, Lappin-Scott HM (1995). Microbial biofilms. Annu. Rev. Microbiol. 49:711-745.

Costerton W, Veeh R, Shirtliff M, Pasmore M, Post C, Ehrlich G (2003). The application of biofilm science to the study and control of chronic bacterial infections. J. Clin. Invest. 112:1466-1477.

Cowan MM (1999). Plants products as antimicrobial agents. Clin. Microbiol. Rev. 12(4):564-582.

Cury JA (2003). Controle químico da placa. In: Promoção de saúde bucal: paradigma, ciência, humanização. 3. ed. São Paulo: Artes Médicas, 141-152.

de Lima MR, de Souza Luna J, dos Santos AF, de Andrade MC, Sant'Ana AE, Genet JP, Marquez B, Neuville L, Moreau N (2006). Anti-bacterial activity of some Brazilian medicinal plants. J. Ethnopharmacol. 105:137-147.

Dolan RM, Costerton JW (2002). Biofilms: survival mechanisms of clinically relevant microorganisms. Clin. Microbiol. Rev. 15:167-193.

Donlan RM (2008). Biofilms on central venous catheters: is eradication possible? Curr. Top. Microbiol. Immunol 322:133-161.

Edries AE (2007). Pharmaceutical and therapeutic potentials of essential oils and their individual volatile constituents: A review. Phytother. Res. 21:308-323.

Eisenberg DM, Davis RB, Ettner SL, Appel S, Wilkey S, Van Rompay M, Kessler RC (1998). Trends in alternative medicine use in United States, 1990-1997: results of a follow-up national survey. J. Am. Med. Assoc. 280:1569-1575.

Flint SH, Bremer PJ, Brooks JD (1997). Biofilms in dairy manufacturing plant-description, current concerns and methods of control. Biofouling 11:81-97.

Fontaine BM, Nelson K, Lyles JT, Jariwala PB, García-Rodriguez JM, Quave CL, Weinert EE (2017). Identification of ellagic acid rhamnoside as a bioactive component of a complex botanical extract with anti-biofilm activity. Front. Microbiol. 8:496.

Fux CA, Stoodley P, Hall-Stoodley L, Costerton JW (2003). Bacterial biofilms: a diagnostic and therapeutic challenge. Expert Rev. Anti-Infect. Ther. 1(4):667- 683.

Greenberg M, Dodds M, Tian M (2008). Naturally occurring phenolic antibacterial compounds show effectiveness against oral bacteria by a quantitative structure-activity relationship study. J. Agric. Food Chem. 56:11151-11156.

Habash M, Reied G (1999). Microbial biofilms: their development and significance for medical device-related infections. J. Clin. Pharmacol. 39:887-898.

Hagan EC, Lloyd AL, Rasko DA, Faerber GJ, Mobley HL (2010). *Escherichia coli* global gene expression in urine from women with urinary tract infection. PLoS Pathog. 6(11):e1001187.

Hall-Stoodley L, Costerton JW, Stoodley P (2004). Bacterial biofilms: from the natural environment to infectious diseases. Nat. Rev. Microbiol. 2(2):95-108.

Hall-Stoodley L, Stoodley P (2009). Cellular Envolving concepts in

biofilm infections. Cell Microbiol. 11(7):1034-1043.
Hansen SK, Rainey PB, Haagensen JA, Molin S (2007). Evolution of species interactions in a biofilm community. Nature 445:533-536.
Hatt JK, Rather PN (2008). Role of bacterial biofilms in urinary tract infections. In Bactarial Biofilms, Springer, Heidelberg, 163-192.
Hermansson M (1999). The DLVO theory in microbial adhesion. Coloids Surf. B: Biointerfaces. 14(1):105-119.
Hoiby N, Bjarnsholt T, Givskov M, Molin S, Ciofu O (2010). Antibiotic resistance of bacterial biofilms. Int. J. Antimicrob. Agents 35:322-332.
Jakubovics NS, Kolenbrander PE (2010). The road to ruin: the formation of disease-associated oral biofilms. Oral Dis. 16:729-739.
Jeon JG, Pandit S, Xiao J, Gregoire S, Falsetta ML, Klein MI, Koo H (2011). Influences of trans-trans farnesol, a membrane-targeting sesquiterpenoid, on *Streptococcus mutans* physiology and survival within mixed-species oral biofilms. Int. J. Oral Sci. 3:98-106.
Jones FA (1996). Herbs - useful plants. Their role in history and today. Eur. J. Gastroenterol. Hepatol. 8:1227-1231.
Kleerebezem M, Quadri LEN, Kuipers OP, Vos WMD (1997). Quorum - sensing by peptide pheromones and two-component signal-transduction systems in Gram-positive bacteria. Mol. Microbiol. 24(5):895-904.
Knoll-Kohler E, Stiebel J (2002). Amine fluoride gel affects the viability and the generation of superoxide anions in human polymorphonuclear leukocytes: an *in vitro* study. Eur. J. Oral Sci. 110:296-301.
Koo H, Hayacibara MF, Schobel BD, Cury JA, Rosalen PL, Park YK, Vacca-Smith AM, Bowen WH (2003). Inhibition of *Streptococcus* mutans biofilm accumulation and polysaccharide production by apigenin and tt-farnesol. J. Antimicrob. Chemother. 52(5):782-789.
Koo H, Rosalen PL, Cury JA, Park YK, Bowen WH (2002). Effects of compounds found in propolis on *Streptococcus* mutans growth and on glucosyltransferase activity. Antimicrob. Agents Chemother. 46(5):1302-1309.
Lachenmeier DW (2008). Safety evaluation of topical applications of ethanol on the skin and inside the oral cavity. J. Occup. Med. Toxicol. 3:26-33.
Lawless J (1995). The Illustrated Encyclopedia of Essential Oils. Shaftesbury, UK: Element Books Ltd.
Lewis K (2001). Riddle of biofilm resistance. Antimicrob. Agents Chemother. 45(4):999-1007.
Lis-Balchin M, Deans SG (1997). Bioactivity of selected plantessential oils against *Listeria monocytogenes*. J. Appl. Bacteriol. 82:759-762.
Lopez D, Vlamakis H, Kolter R (2010). Biofilms. Cold Spring Harbor Perspect. Biol. 2(7):1-11.
Marsh PD (1992). Microbiological aspects of the chemical control of plaque and gingivitis. J. Dent. Res. 71:1431-1438.
Marsh PD (2010). Controlling the oral biofilm with antimicrobials. J. Dent. 38:S11-S15.
Matz C, Kjelleberg S (2005). Off the hook-how bacteria survive protozoan grazing. Trends Microbiol. 13:302-307.
Maukonen J, Mättö J, Wirtanen G, Raaska L, Mattila-Sandholm T, Saarela M (2003). Methodologies for the characterization of microbes in industrial environments: a review. J. Ind. Microbiol. Biotechnol. 30:327-356.
Mccullough MJ, Farah CS (2008). The role of alcohol in oral carcinogenesis with particular reference to alcohol-containing mouthwashes. Aust. Dent. J. 53(4):302-305.
Millezi AF, Piccoli RH, Cardoso M das Graças, Alves E (2010). Ação bactericida de detergente-sanificante à base de óleos essenciais sobre biofilme de *Aeromonas hydrophila*. XIX CONGRESSO DE PÓS-GRADUAÇÃO DA UFLA.
Mishra AK, Dubey NK (1994). Evaluation of some essential oils for their toxicity against fungi causing deterioration of stored food commodities. Appl. Environ. Microbiol. 60:1101-1105.
More G, Tshikalange TE, Lall N, Botha F, Meyer JJ (2008). Antimicrobial activity of medicinal plants against oral microorganisms. J. Ethnopharmacol. 119:473-477.
Mukamolova GV, Kaprelyants AS, Young DI, Young M, Kell DB (1998). A bacterial cytokine. Proceedings of the National Academy of Sciences of the United States of America 95(15):8916-8921.
Murray EJ, Strauh MA, Stanley-Wall NR (2009). σx Is involved in controlling Bacillus subtilis biofilm architecture through the AbrB homologue Abh. J. Bacteriol. 191:6822-6832.
Nagorska K, Ostrowski A, Hinc K, Holland IB, Obuchowski M (2010). Importance of EPS genes from *Bacillus subtilis* in biofilm formation and swarming. J. Appl. Gene. 51:369-381.
Namasivayam SKR, Roy EA (2013). Anti-biofilm effect of medicinal plant extracts against clinical isolate of Biofilm of *Escherichia coli*. Int. J. Pharm Pharm. Sci. 5:486-489.
Nascimento GG, Locatelli J, Freitas PC, Silva GL (2000). Antibacterial activity of plant extracts and phytochemicals on antibiotic-resistant bacteria. Braz. J. Microbiol. 31(4):247-256.
Neumegen RA, Fernández-Alba AR, Chisti Y (2005). Toxicities of triclosan, phenol, and copper sulfate in activated sludge. Environ. Toxicol. *20*(2):160-164.
Nguefack J, Budde BB, Jakobsen M (2004). Five essential oils from aromatic plants of Cameroon: their antibacterial activity and ability to permeabilize the cytoplasmic membrane of Listeria innocua examined by flow cytometry. Lett. Appl. Microbiol. 39(5):395-400.
Nikolaev YA, Plakunov VK (2007). Biofilm-“City of microbes” or an analogue of multicellular organisms? Microbiology *76*(2):125-138.
Njoroge J, Sperandio V (2009). Jamming bacterial communication: new approaches for the treatment of infectious diseases. EMBO Mol. Med. 1(4):201-210.
Oliveira DC, Oliveira DC, Rosell FL, Sampaio JEC, Rodrigues J, Antonio L (1998). Redução do índice de placa com Listerine: avaliação do índice de placa em relação ao uso de Listerine e/ou escovação. Revista Gaúcha de Odontologia. Porto Alegre 46(2):101-108.
Paerl HW, Pinckney JL (1996). A mini-review of microbial consortia: their roles in aquatic production and biogeochemical cycling. Microbial Ecol. 31(3):225-247.
Palombo EA (2011). Traditional medicinal plant extracts and natural products with activity against oral bacteria: potential application in the prevention and treatment of oral diseases. Evid. Based Complement. Altern. Med. 1-15.
Parsek MR, Greenberg EP (2005). Sociomicrobiology: the connections between quorum sensing and biofilms. Trends in Microbiol. 13(1):27-33.
Paytubi S, de La Cruz M, Tormo JR, Martín J, González I, González-Menendez V, Genilloud O, Reyes F, Vicente F, Madrid C, Balsalobre C (2017). A high-throughput screening platform of microbial natural products for the discovery of molecules with antibiofilm properties against Salmonella. Front. Microbiol. 8:326
Pereira AG, Neves AM, Trindade AC (2010). Imunologia da cárie dentária. Acta Medica Portuguesa 23:663-668.
Porto TS, Furtado NA, Heleno VC, Martins CH, Da Costa FB, Severiano ME, Silva AN, Veneziani RC, Ambrósio SR (2009). Antimicrobial ent-pimarane diterpenes from *Viguiera arenaria* against Gram-positive bactéria. Fitoterapia 80:432-436.
Prabu GR, Gnanamani A, Sadulla S (2006). Guaijaverin - a plant flavonoid as potential antiplaque agent against *Streptococcus mutans*. J. Appl. Microbiol. 101:487-95.
Qian PY, Chen L, Xu Y (2013). Mini-review: Molecular mechanisms of antifouling compounds. Biofouling 29: 381-400.
Reichling J, Schnitzler P, Suschke U, Saller R (2009). Essential oil of aromatic plant with antibacterial, anfungical, antiviral and citotoxic properties - an overview. Forsch Klomplementmed. 16:79-90.
Reynolds JEF (1996). Martindale - the Extra Pharmacopoeia 31st edn. London: Royal Pharmaceutical Society of Great Britain.
Rodrigues F, Lehmann M, do Amaral VS, Reguly ML, de Andrade HHR (2007). Genotoxicity of three mouthwash products, Cepacol®, Periogard®, and Plax®, in the Drosophila wing-spot test. Environ. Mol. Mutagen. 48(8):644-649.
Rozalski M, Micota B, Sadowska B, Stochmal A, Jedrejek D, Wieckowska-Szakiel M, Rozalska B (2013). Antiadherent and antibiofilm activity of *Humulus lupulus* L. derived products: new pharmacological properties. BioMed Res. Int. 2013.
Sá NC, Cavalcante TTA, Araújo AX, dos Santos HS, Albuquerque MRJR, Bandeira PN, Teixeira EH (2012). Antimicrobial and

antibiofilm action of casbane diterpene from *Croton nepetaefolius* against oral bacteria. Arch. Oral Biol. 57(5):550-555.
Savoia D (2012). Plant-derived antimicrobial compounds: alternatives to antibiotics. Future Microbiol. 7(8):979-990.
Schachter B (2003). Slimy business-the biotechnology of biofilms. Nat. Biotechnol. 21(4):361-365.
Scheie AA (2007). O papel dos antimicrobianos. In: Cárie dentária: a doença e seu tratamento clínico. São Paulo: Ed. Santos. pp. 179-188.
Schmidt E, Jirovetz L, Buchbauer G, Denkova Z, Stoyanova A, Murgov I, Geissler M (2005). Antimicrobial testingand gas chromatographic analysis of aroma chemicals. J. Essential Oil Bearing Plants 8:99-106.
Selim SA, Adam ME, Hassan SM, Albalawi AR (2014). Chemical composition, antimicrobial and antibiofilm activity of the essential oil and methanol extract of the Mediterranean cypress (*Cupressus sempervirens* L.). BMC Complement. Altern. Med. 14(1):1.
Shapiro JA (1998). Thinking about bacterial populations as multicellular organisms. Annu. Rev. Microbiol. 52(1):81-104.
Sihorkar V, Vyas SP (2001). Biofilm consortia on biomedical and biological surfaces: delivery and targeting strategies. Pharm. Res. 18:1254-1427.
Silva MDSA, Silva MAR, Higino JS, Pereira MSV, Carvalho ADA (2008). Atividade antimicrobiana e antiaderente *in vitro* do extrato de Rosmarinus officinalis Linn. sobre bactérias orais planctônicas. Rev Bras Farmacogn. 18(2):236-240.
Silva MSP, Brandao DO, Chaves TP, Formiga Filho AL, Costa EMMDB, Santos VL, Medeiros ACD (2012). Study bioprospecting of medicinal plant extracts of the semiarid northeast: contribution to the control of oral microorganisms. Evi-Based Complement. Altern. Med. 2012.
Simões CMO, Schenkel EP, Gosmann G, Mello, JCP, Mentz, LA, Petrovick, PR (2007). Farmacognosia: da planta ao medicamento. 6. ed. Porto Alegre/Florianópolis: Editora da Universidade UFRGS/Editora UFSC. 1104 p.
Simões M, Simões LC, Vieira MJ (2010). A review of current and emergent biofilm control strategies. Food Sci. Technol. 43:573-583.
Tavares W (1996). Introdução ao estudo dos antimicrobianos. In: Manual de antibióticos e quimioterápicos antiinfecciosos. 2 ed. São Paulo: Atheneu 3-13.
Teixeira KIR, Cortes ME (2005). Estado actual de la indicacion de antimicrobianos para la medicacion intracanal. Acta Odontol. Venez. 43(2):177-180.
Trentin DS, Gorziza DF, Abraham WR, Antunes ALS, Lerner C, Mothes B, Macedo AJ (2011a). Antibiofilm activity of Cobetia marina filtrate upon Staphylococcus epidermidis catheter-related isolates. Braz. J. Microbiol. 42(4):1329-1333.
Trentin DS, Giordani RB, Zimmer KR, da Silva AG, da Silva MG, Correia MT, Baumlov IJ, Macedo AJ (2011b). Potential of medicinal plants from the Brazilian semi-arid region (Caatinga) against Staphylococcus epidermidis planktonic and biofilm lifestyles. J. Ethnopharmacol. 137:327-335.
Velicer GJ (2003). Social strife in the microbial world. Trends Microbiol. 11:330-337.
Veran J (2002). Biofouling in food processing: biofilm or biotransfer potential? Food Bioprod. Process. 80:292-298.
Viju N, Satheesh S, Vincent SGP (2013). Antibiofilm activity of coconut (*Cocos nucifera* Linn.) husk fibre extract. Saudi J. Biol. Sci. 20:85-91.
Vu B, Chen M, Crawford RJ, Ivanova EP (2009). Bacterial extracellular polysaccharides involved in biofilm formation. Molecules 14(7):2535-2554.
Weigel LM, Donlan RM, Shin DH, Jensen B, Clark NC, McDougal LK, Zhu W, Musser KA, Thompson J, Kohlerschmidt D, Dumas N, Limberger RJ, Patel JB (2007). High-level vancomycin-resistant Staphylococcus aureus isolates associated with a polymicrobial biofilm. Antimicrob. Agents. Chemother. 51(1):231-238.
Zobell CE (1943). The effect of solid surfaces upon bacterial activity. J. Bacteriol. 46:39-56.