Saudi Journal of Biological Sciences 27 (2020) 1428–1434

Contents lists available at ScienceDirect

Saudi Journal of Biological Sciences

journal homepage: www.sciencedirect.com

Original article

Potential effect of *Allium sativum* bulb for the treatment of biofilm forming clinical pathogens recovered from periodontal and dental caries

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ARTICLE INFO

Article history:
Received 18 January 2020
Revised 21 March 2020
Accepted 26 March 2020
Available online 2 April 2020

Keywords:
Periodontal
Dental caries
Extracellular polysaccharide
Biofilm
Allium sativum
Drug resistant

ABSTRACT

Biofilm producing clinical bacterial isolates were isolated from periodontal and dental caries samples and identified as, *Lactobacillus acidophilus, Streptococcus sanguis*, *S. salivarius, S. mutans*and *Staphylococcus aureus*. Among the identified bacterial species, *S. aureus* and *S. mutans*showed strong biofilm producing capacity. The other isolated bacteria, *Streptococcus sanguis*, *S. salivarius* showed moderate biofilm formation. These pathogens were subjected for the production of extracellular polysaccharides (EPS) in nutrient broth medium and the strain *S. aureus* synthesized more amounts of EPS (610 ± 11.2 μg/ml) than *S. sanguis* (480 ± 5.8 μg/ml).EPS production was found to be less in *S. salivarius* (52 ± 3.8 μg/ml).The solvent extract of *A. sativum* bulb showed the phytochemicals such as, carbohydrate, total protein, alkaloids, saponins, flavonoids, tannins and sterioids. The solvent extract of *A. sativum* bulb showed wide ranges of activity against the selected dental pathogens. The difference in antibacterial activity of the solvent extract revealed differences in solubility of phytochemicals in organic solvents. Ethanol extract was highly active againstS. aureus (25 ± 2 mm). The Minimum Inhibitory Concentration (MIC) of crude garlic bulb varied widely and this clearly showed that bacteria exhibits different level of susceptibility to secondary metabolites. MIC value ranged between 20 ± 2 mg/ml and 120 ± 6 mg/ml and Minimum Bactericidal Concentration (MBC) value ranged from 60 ± 5 mg/l to 215 ± 7 mg/ml. To conclude, *A. sativum* bulb can be effectively used to treat periodontal and dental caries infections.

1. Introduction

Dental caries is highly common disease in humans and is very common in Latin and Asian countries. About 70% of younger generations are affected by dental caries (Tandon et al., 2010). The important causative organisms of dental caries are *Streptococcus mitis*; *S. mutans*, *S. salivarius*, *S. sanguis* and *S. sobrinus*. The acidogenic dental pathogens involved in the fermentation of carbohydrates. The fermentation of various sugars by these acidogenic bacterial strains is an important factor for the formation of dental caries. The acid producing bacteria involved in demineralization process and involved in the formation of cavity in tooth (Islam et al., 2007). The bacterial strain *S. mutans* colonizes the surface of the tooth and readily induces the formation of plaque by producing various polysaccharides by extracellular using sucrose. Further, in the plaque margin accumulation of Gram-negative and Gram-positive bacteria was reported (Iwaki et al., 2006). In developed countries, oral diseases cause about 10% of public health expense. However in developing countries, this expense has been very low, and very rare access of oral health, mainly maintenance restricted to pain relief or emergency dental healthcare (Petersen et al., 2005). The link between microbial population that form cavity in teeth and oral health are well observed (Jenkinson and Lamont, 2005). Dental caries involves decalcification of calcium in teeth and decay. In the case of periodontal disease various anerobic Gram-negative bacterium involved. These include, *Fusobacterium*sp., *Prevotella*sp., *Actinobacillus*sp., *Porphyromonasgingivalis*. Generally, in periodontal condition, the surroundings of gringival crevice were infected, causing various inflammatory responses, including cellular of the gingival and connective tissues (Tichy and Novak, 1998).

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Peer review under responsibility of King Saud University.

Production and hosting by Elsevier

https://doi.org/10.1016/j.sjbs.2020.03.025
1319-562X/

Drug resistant bacteria pose serious threat in the treatment of many well-known infections and an urgent need to find novel antimicrobial agent to treat these organisms. In developing countries such as, India, plant resource is available in large quantities and is often called as botanical garden of world (Hotwani et al., 2014). In humans, dental caries and periodontal diseases are two important types of pathology frequently affected (Marsh and Martin, 1992). These two conditions are the results of plaque formation by yeast, bacteria which reside in the dental cavity. The microorganisms such as, *Candida* sp., *Streptococcus* sp., *Actinobacillus* and *Actinomyces* are highly associated with periodontal diseases (Van Oosten et al., 1987). *Candida albicans* was reported frequently from immunocompromised people (Samaranayake, 2000). Treatment of dental infections is not easy, and not possible to all sections of people in developing and under developed countries. However, few treatments were recommended to treat dental caries which include phenolic-antiseptics, quaternary ammonium-antiseptics, bisguanide-antiseptics, metal ions and application of oxygenating agents (Addy, 1986). The side effects such as burning sensation at the tip of the tongue, changes in food taste and staining of teeth was reported (Gründermann et al., 2000).

Medicinal plants are ideal sources of various antibacterial and antifungal agents. More than 80% of population in the under developed countries and developing countries use traditional medicine. Plants based medicine are frequently used as herbal medicine for the treatment of various diseases in human (Alviano and Alviano, 2009). Medicinal plants are rich sources of various phytochemicals such as, terpenoids, tannins, flavonoids and alkaloids which have showed potential antimicrobial properties *in vitro* (Dorman and Deans, 2000). The application of medicinal plants has been mainly associated with therapeutic practices and dental hygiene for several decades. In Africa, the tribes, namely, Ethiopians, Vhavenda, Zimbabweans, and Namibians use many plants as chewing sticks (Mabogo, 1990). This chewing stick has potential antibacterial activity against various pathogens. This stick has the ability to control or inhibit the growth of various Gram-negative and Gram-positive bacteria, including, *Fusobactriumnucleatum*, *Porphyromonusgingivalis* and *Bacillus subtilis* (Ndukwe et al., 2004). The chewing stick is commonly called "Muthala", and was known as *Eucleanatalensis* ADC. and *Diospyroslysioides* DESF (Lall and Meyer, 2000). These medicinal plants are from the Anonnaceae, Bombaceae, Ebenaceae and Fabaceae family (Hadissa and Jean-Pierre, 2005).

The decoctions, pastes, juices and other formulations form *Jatropha curcas* are frequently used to maintain oral hygiene and also to treat toothaches, cracked lips, mouth ulcer, carious teeth and bleeding gums. Also the twigs of *J. curcas* are chewed to treat and prevent teeth and gum related problem, pyorrhoea. The twigs are frequently reported by the people of Nigeria to treat and prevent tooth decay. The bacteria such as, *Lactobacillus*, Actinomyces and *Streptococcus* were frequently involved in periodontal and dental caries diseases. Antibiotic resistance against various pathogenic bacteria continues in recent times and the application of antibiotics inhibitors from medicinal plants is used widely. These plants provide many compounds to protect against various types of pathogens. Medicinal plants can target the pathogens in various ways and will be highly active against various drug resistance pathogenic organisms (Ahmad and Beg, 2001). Medicinal plants have been extracted and tested against various dental pathogens. Twigs of plants such as, clove, neem and babul and other medicinal plants have been used for brushing teeth in India (Arora and Kaur, 2007). In this study, *Allium sativum* bulb was used to extract phytochemicals and its efficacy was tested against various bacterial pathogens from patients affected by periodontal and dental caries.

2. Materials and methods

2.1. Culture media

Mueller Hinton agar, Nutrient broth, Antibiotics were purchased from Sigma, USA. Other chemicals were Analytical grade.

2.2. Test bacteria

In this study, dental caries bacteria *Lactobacillus acidophilus, Streptococcus sanguis*, *S. salivarius, S. mutans* and *Staphylococcus aureus* were isolated from the infected patients. Pure culture was performed by serial dilution and plating technique. The isolated strains were identified by biochemical characters and used for antibacterial susceptibility tests.

2.3. Biofilm assay

The isolated periodontal and dental caries bacterial pathogens were subjected for biofilm assay. Quantitative determination of biofilm formation was determined using microtitre plate method as suggested by Elhadidy and Elsayyad (2013). All experiments were performed in duplicate and the results were observed. Based on its biofilm forming ability the bacteria were classified as week-, moderate-, and strong biofilm producer (Stepanović et al., 2007).

2.4. Extracellular polysaccharide (EPS) production and assay

The isolated bacterial strains were cultured in 250-ml Erlenmeyer flask containing 100 ml culture medium. All Erlenmeyer flasks were incubated at 37 °C for 72 h. After 72 h, the culture was centrifuged and subjected to EPS assay (Lima et al., 2008).

2.5. Inoculum preparation

The test bacteria (*Lactobacillus acidophilus, Streptococcus sanguis*, *S. salivarius, S. mutans* and *Staphylococcus aureus*) were inoculated into nutrient broth medium and incubated for 24 at 37 °C. The growth was measured and the bacterial colonies were counted. The bacterial suspension with 10^5CFU/ml was used for analysis.

2.6. Medicinal plant

In this study, *Allium sativum* bulb was used as the sample for antimicrobial activity analysis. Fresh *Allium sativum* bulb was collected from the market and directly used for the extraction of phytochemicals.

2.7. Extraction of plant phytochemicals

About 100 g of *Allium sativum* bulb was added in Soxlet apparatus and extracted with solvents such as, ethanol, hexane, acetone, water and diethyl ether for 3 days. After complete extraction, the extract was passed through Whatman no-40 filter paper and the obtained filtrate was concentrated using vacuum evaporator at room temperature (30 ± 2 °C).

2.8. Phytochemical screening

The dried bulb of *A. sativum* was extracted with solvent such as, ethanol, diethyl ether, acetone, hexane and water. Phytochemcials were screened by the method of Parekh and Chanda (2007) and Evans (1996).

2.9. Minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC)

A. sativum bulb was subjected to MIC and MBC analysis. The microdilution technique was used for the determination of MBC in 96 well microtitre plate and the crude bulb extract was used for analysis. The plant extract was serially diluted and the concentration ranged between 50 mg/ml to 0.25 mg/ml. The MIC value was determined as the extreme lowest phytochemical extract which effectively inhibited 100% bacterial growth in the standard condition (Cohen et al., 1998). The growth of microorganisms was monitored by adding *p*-iodonitrotetrazolium violet to 96-well microtitre plate and incubated for 48 h at 37 °C. The MBC value of the crude plant extract was evaluated by including plant extract diluted appropriately and the well which did not show any bacterial growth after inoculate-d with bacterial suspension. MBC is defined as the extreme lowest concentration of plant extract required to inhibit the colour change of *p*-iodonitrotetrazolium.

2.10. Statistical analysis

All experiments were repeated thrice and an average value of zone of inhibition (mm) was calculated. One way analysis of variance was used to determine the significance ($P < 0.05$).

3. Results and discussion

3.1. Screening of organism for biofilm production

The present study revealed that all the isolated strains showed visible biofilm than control, however, the degree of biofilm formation varied widely. The results revealed that of *Lactobacillus* sp. from the periodontal sample showed moderate biofilm producing ability, while all isolated *Streptococcus* strains produced biofilm showed strong biofilm formation. Among the identified bacterial species, *Staphylococcus aureus* and *Streptococcus mutans* showed strong biofilm producing capacity. The other isolated bacteria, *Streptococcus sanguis*, *S. salivarius* showed moderate biofilm formation. Periodontal infections are caused by various Enterococci species (Peciuliene et al., 2000). Previously various biofilm producing Enterococci species have been reported by Joyanes et al. (2000). Bacteria involving periodontal infections show potential risk to humans. Periodontal pockets may serve as effective reservoirs of various bacterial pathogens, and their synthesized products and transmits to the other parts of the body (Scannapieco, 1998; Han and Wang, 2013). According to these studies, various Gram-positive bacteria and Gram-negative bacteria have been previously reported from oral cavity (Sedgley and Samaranayake, 1994). The high frequency of *Staphylococcus aureus* and *Lactobacillus* sp. would be highly expected since they are predominant in the dental surfaces and commensal inhabitants of the mucosa of humans.

3.2. Exopolysaccharides and its role in pathogenic organisms

EPS production was found to be high in the culture of *S. aureus* (610 ± 11.2 μg/ml), followed by *S. sanguis* (480 ± 5.8 μg/ml). EPS production was found to be less in *S. salivarius* (52 ± 3.8 μg/ml) (Fig. 1). These extracellular polysaccharides protect these organisms from harsh environment. It was reported that EPS in the matrix pose a diffusion barrier, which effectively protects biofilm-producing bacterial cells from the penetration of antimicrobial agents. So, EPS production and biofilm formation linked with antibiotic resistance. Moreover, it was reported that most of the antibiotics can easily penetrate the synthesized biofilm

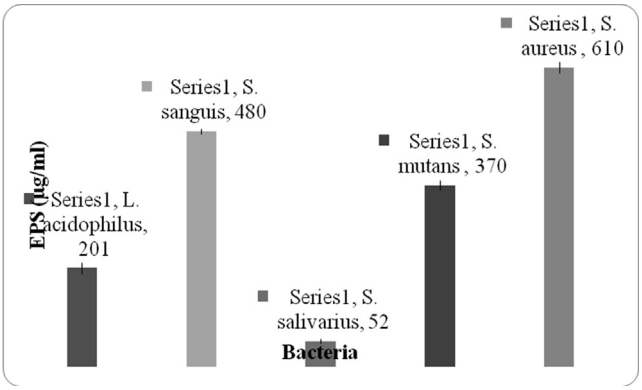

Fig. 1. Production of EPS by dental isolates in nutrient broth medium. The selected strains were inoculated and incubated for 72 h at 37 °C and the yield was expressed as μg/ml.

(Walters et al., 2003). In some cases penetration of drug is neutralized by enzymes within microbial matrix before it reaching the bacterial cell (Hoiby et al., 2010). The important and prevalent role of EPS is protection from nonopsonic and opsonic phagocytosis. EPS provided highly hydrated biofilm environment, which provide heavy protection from desiccation and generally promote biofilm fluidity (Sutherland, 2001). Hence, analysis of EPS production is also very important to analyze the virulence of any particular pathogenic strain.

3.3. Phytochemical properties of A. sativum

The solvent extract of *A. sativum* bulb showed the phytochemicals such as, carbohydrate, total protein, alkaloids, saponins, flavonoids, tannins and sterioids. However, the ethanol extract devoid of flavonoids, however acetone extract devoid of alkaloids, saponins and tannins. Water extract showed the presence of phytochemicals such as, carbohydrates, total protein, saponins and tannins (Table 1). These bioactive compounds were highly responsible for the biological activity. Many researchers have attributed the antibacterial activity of plant extract to the availability of these phytochemicals (Gandhiraja et al., 2009; Nweze et al., 2004). The presence of various phytochemicals in the bulb of garlic could be effectively used to formulate herbal medicine to treat various ailments.

3.4. Antimicrobial activity of A. sativum extracted with various solvent

The solvent extract of *A. sativum* bulb showed wide ranges of activity against the selected dental pathogens. The difference in antibacterial activity of the solvent extract revealed differences in solubility of phytochemicals in organic solvents. Ethanol has been frequently used as the solvent for the extraction of phytochemicals. Ethanol fraction was highly active against *S. aureus* (25 ± 2 mm). The other solvent extracts did not show much antibacterial activity and the results were presented in Fig. 2 and Table 2. Ethanol solubilised number of phytochemicals than ethyl acetate, methanol and chloroform (De Boer et al., 2005). It has been reported that Gram-positive bacteria were highly sensitive to antibacterial substances, these organisms have only peptidoglycan layer, in the case of Gram-negative bacteria, phospholipid membrane which affected permeability of antimicrobials. In a study, Kallel et al. (2014) reported moderate antimicrobial activity of ethanol extract of *A. sativum* against *S. aureus* and *B. subtilis* and the zone of inhibition ranged between 10 and 15 mm and activity was found to be less against *P. aeruginosa* and *B. thuringiensis* (<10 mm zone of inhibition), however, no antibacterial activity

Table 1
Phytochemical screening of bulb of *A. sativum*.

	Solvent				
Phytochemicals	Ethanol	Acetone	Diethyl ether	Hexane	Water
Carbohydrates	+	+	+	+	+
Total protein	+	+	+	+	+
Alkaloids	+	–	–	+	–
Saponins	+	–	–	–	+
Flavonoids	–	+	–	+	–
Tannins	+	–	+	–	+
Sterioids	+	+	+	–	–

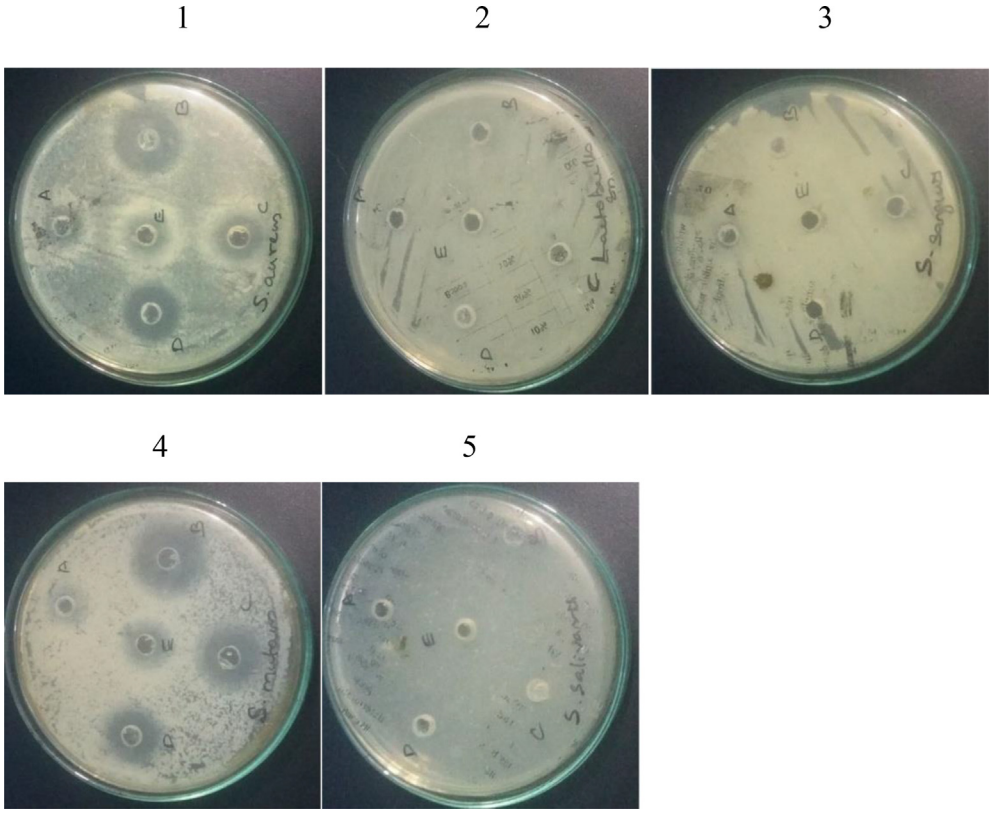

Fig. 2. Antibacterial activity of crude bulb extract against dental pathogens. 25 μg sample was loaded and zone of inhibition (mm) was measured after 24 h (1 = *S. aureus*, 2 = *Lactobacillus* sp., 3 = *S. sanguis*; 4 = *S. mutans*, 5 = *S. salivarius*), (A = ethanol, B = diethyl ether, C = acetone, D = hexane, E = water).

Table 2
Antimicrobial activity of crude extract against bacterial pathogens isolates from periodontal and dental caries.

	Zone of inhibition (mm)				
Test bacteria	Ethanol	Diethyl ether	Acetone	Hexane	Water
S. aureus	25 ± 2	13 ± 3	13 ± 3	19 ± 1	13 ± 3
Lactobacillus sp.	9 ± 2	9 ± 4	11 ± 3	9 ± 2	8 ± 4
S. sanguis	8 ± 1	8 ± 1	12 ± 1	8 ± 0	9 ± 7
S. mutans	17 ± 3	21 ± 2	18 ± 2	11 ± 21	9 ± 6
S. salivarius	10 ± 1	12 ± 1	10 ± 1	10 ± 1	13 ± 3

was reported against, *S. typhimurium*, *E. coli* and *K. pneumonia*. Karuppiah and Rajaram (2012) analyzed the antimicrobial properties of ethanol extract of *A. sativum* and reported antimicrobial activity against various drug resistance bacteria such as, *S. aureus* (13.50 mm), *Enterobacter* sp. (13.50 mm), *Proteus* sp. (13.50 mm), *Bacillus* sp. (16.5 mm), *E. coli* (18.50 mm) and *P. aeruginosa* (19.45 mm). In the present work based on inhibitory zone on Meuller Hinton Agar plates, the ethanol extract showed high antibacterial activity against the tested bacteria. Allicin is an important antimicrobial compounds reported from *A. sativum*. Recent findings revealed antimicrobial property of allicin against pathogenic organisms (Goncagul, 2010). It has been purified by various methods and the purified compound showed antibacterial against various Gram's-negative and Gram's-positive bacteria and also showed activity against the fungus, *Candida albicans*, and antiviral activity and anti-parasitic activity (Ankri and Mirelman, 1999). Protease inhibiting ability of allicin was reported and subsequently acting against parasitic protozoa (Waag et al., 2010).

Table 3
Minimum Inhibitory Concentration (MIC) of crude bulbs extract from *A. sativum.*

	Minimum Inhibitory Concentration (MIC) (mg/ml)				
Microorganisms	Ethanol	Diethy ether	Acetone	Hexane	Water
S. aureus	35 ± 3	45 ± 4	60 ± 3	80 ± 1	95 ± 3
Lactobacillus sp.	110 ± 4	120 ± 6	100 ± 1	110 ± 5	100 ± 1
S. sanguis	80 ± 3	86 ± 3	120 ± 7	80 ± 3	90 ± 7
S. mutans	20 ± 2	25 ± 3	45 ± 2	75 ± 7	90 ± 12
S. salivarius	98 ± 1	104 ± 1	80 ± 8	70 ± 1	81 ± 2

Table 4
Minimum Bactericidal Concentration (MBC) of crude bulbs extract from *A. sativum.*

	Minimum Bactericidal Concentration (MBC) (mg/ml)				
Microorganisms	Ethanol	Diethyl ether	Acetone	Hexane	Water
S. aureus	60 ± 5	120 ± 2.6	110 ± 1	100 ± 3	120 ± 10
Lactobacillus sp.	150 ± 2.5	190 ± 1.1	120 ± 3	140 ± 6	100 ± 13
S. sanguis	140 ± 3	165 ± 3	215 ± 7	150 ± 10	145 ± 12
S. mutans	70 ± 2.2	65 ± 4	80 ± 8	90 ± 12	125 ± 19
S. salivarius	145 ± 1.6	120 ± 7	110 ± 1	125 ± 7	130 ± 13

The antibacterial activity of *Allium sativum* performed in this study is similarwith the results of various research groups. Garlic extract showed antibacterial activity on wide range of bacteria. The crude extract was highly active against *S. aureus*. Ethanol was the most successful solvent and it showed more antibacterial activity (25 ± 2 mm zone of inhibition), followed by hexane extract (19 ± 1 mm zone of inhibition) on *S. aureus*. The crude extract was also found to be stable against *S. mutans*. Diethyl ether extract showed 21 ± 2 mm zone of inhibition against *S. mutans* and acetone showed 18 ± 2 mm zone against this organism. In *A. sativum*, the antibacterial activity is mainly due to the presence of phytochemicals such as, tannin, flavonoids and alkaloids. The antimicrobial potential of these compounds have been reported by Deresse (2010). Also, the antimicrobial potential of this extract was observed against Gram negative bacteria (*Klebsiella* sp., *Pseudomonas* sp., *Enterobacter* sp., *Citrobacter* sp., *Salmonella* sp., and *E. coli*) Gram's positive bacteria (*Bacillus* sp., *Streptococcus* sp., and *S. aureus*). Tsao and Yin (2001) reported antimicrobial activity of garlic extract against various Gram's-negative and Gram's-positive bacteria such as, *Helicobacter pylori*, *Proteus*, *Salmonella*, *Shigella*, *Pseudomonas*, *Lactobacillus*, *Klebsiella*, *Enterobacter*, *Micrococcus*, *Streptococcus* and *Staphylococcus* sp. Fresh garlic juice has been tested for its antibacterial property against various Gram-positive and Gram-negative pathogens. Also, the antimicrobial property of fresh garlic juice against various drug resistant bacteria was reported by Li et al. (2015). Traditionally, garlic has been effectively utilized by various populations in various countries. In India, it has been used to prevent food spoilage and wound infection. In a study, Arora and Kaur (1999) reported the antimicrobial properties of food spoilage bacteria and also reported the protective role of garlic in wound infections. In Ireland, garlic has been used to treat pulmonary diseases. Garlic extract was found to be effective against multiple drug resistant *Helicobacter* (O'Gara et al., 2000). Skyrme (1997) studied antibacterial activity of *Lactobacillus*; *L. casei* and *E. coli*.

In the case of *Lactobacillus* zone of inhibition was found to be less than Inhibition observed in *E. coli* was more than 10 times greater than that seen in *Lactobacillus casei* for the same garlic dose (Skyrme, 1997). Miron et al. (2000) studied antimicrobial activity of garlic extract against various pathogenic microorganisms and observed varying degree of antimicrobial sensitivity and reported variations in permeability of allicin. In garlic, antimicrobial effect of allicin considerably reduced during storage. It is highly unstable even at room temperature. Gallic acid combined with vancomycin and observed synergistic activity in previous studies. Yusha'u et al. (2008) observed antimicrobial activity of garlic against various Gram-negative bacteria involved in respiratory infections. Among the tested isolates, garlic was found to be effective against *E. coli*. Also the mixture of garlic and ginger extracts was tested and the efficacy was compared with commercial antibiotics. Analysis revealed that the extract from these two sources showed high activity against meat spoiling bacteria such as, *Pseudomonas fluorescence*, *E. coli*, *Staphylococcus* spp., *Enterococcus* sp. and *B. subtilis* (Leita d Souza et al., 2005). Recently Sarhan et al. (2016) used *Allium sativum* extract to prepare honey/chitosan nanofiber wound dress and reported potent activity against *S. aureus*, multidrug-resistant *Pseudomonas aeruginosa*, Methicillin-resistant *S. aureus* (MRSA) and *Escherichia coli*.

The MIC of crude garlic bulb varied widely and this clearly showing that bacteria exhibits different level of susceptibility to plant secondary metabolites. The present finding highly agrees with previous report by Banso and Adeyemo (2007) describing that antibacterial agent with very low activity against bacteria has a high MIC value, while a potent antimicrobial agent gives a low MIC value. The MIC value for *S. aureus* and *S. mutans* were found to be 35 ± 3 mg/ml and 20 ± 2 mg/ml (Table 3). The MBC value for all bacterial isolates was described in Table 4. The antibacterial activity of *A. sativum* essential oil (EO) has studied previously. Extracted essential oil showed potent antibacterial activity against various bacteria including, *Escherichia coli* (inhibition zone 11.0 mm), *Pseudomonas aeruginosa* (inhibition zone 21.1 mm) and *Staphylococcus aureus* (inhibition zone 14.8 mm). The phytochemicals such as, dipropyldisulfide, diallylmonosulfide and diallyldisulfide (DADS), diallyltrisulfide, and diallyltetrasulfide were detected. In garlic, allyl group is highly responsible for antibacterial activity (Casella et al., 2013). O'Gara et al. (2000) reported the influence of *Allium* vegetable intake on chronic Helicobacter pylori infections. The reported MIC level varied between 8 and 32 μg/ml and MIC values were ranged from 16 to 32 μg/ml. Garlic oil act on bacterial pathogens and higher EO concentration showed more activity. These finding revealed the importance of garlic to treat *H. pyroli* infections. Recently, Chen et al. (2018) used *Allium sativum* extract to treat various plant pathogens. The extract showed high activity at pH 4.0, 3-vinyl-1,2-dithiacyclohex-4-ene was detected as the one of the important compounds. Garlic extract showed activity against bacteria such as, *Clavibactermichiganensis* subsp. *michiganensis*, *Xanthomonasvesicatoria*, and these phytopathogens cause bacterial spot disease (Martins et al., 2016). Garlic extract

along with eucalyptus extracts showed wide spread activity against various bacteria involved in pepper and bacterial spot diseases (Mirik and Aysan, 2005). The application of garlic as a food supplement is widely considered as health choice for the treatment of diabetes, hypertension, cardiovascular disease, inflammation, Alzheimer's disease, thrombosis and cancer (Banerjee and Maulik, 2002; Peng et al., 2002; Chauhan, 2006; Fukao et al., 2007).

4. Conclusion

Multiple drug resistant bacterial pathogens pose serious threat to periodontal and dental caries infections. These pathogenic bacteria produce extracellular polysaccharides and biofilm. The biofilm protects the organisms from the penetration of various drugs. *Allium sativum* bulb showed high antibacterial activity against *Staphylococcus aureus* and *Streptococcus mutans*. These two were generally regarded as highly pathogenic strains. Hence, *A. sativum* could be useful to treat these dental infections without any side effects.

Acknowledgment

The authors would like to extend their sincere appreciation to the Deanship of Scientific Research at King Saud University for its funding of this research through the Research Group Project No. RG-1435-071.

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