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Research Article

Bio profiling of antibacterial activity of *citrus maxima*: An ethnonutraceutical against cariogenic *streptococci*

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Abstract

Background: Dental caries is a common chronic oral disease that affects people all over the world. It is caused by the etiological agent oral streptococci, which releases virulence factors and causes an imbalance in the mouth biota ecological system. The use of ethno-medicinal plant part crude extracts and herbs as a biocontrol agent for commercially available chemical manufactured medications has recently gained a lot of traction in the fight against the pathogenesis of oral illness and disorders such periodontal disease and dental caries. *Citrus maxima (Rutaceae)* a widely known therapeutic shrub in ayurveda in India is a promising possibility to increase the inhibitory impact endured as antimicrobial agents through synergism. The goal of this study was to see how effective *Citrus maxima* peel extracts were at preventing carcinogenesis in common but important oral pathogenic bacteria: *Streptococcus mutans, S. sanguis, S. salivarius, S. sobrinus,* and *S. mitis.*

Material & Method: The phytochemical study was analysed for fraction of *Citrus maxima* fruits and peels imparted the presence of alkaloids, flavonoids, anthraquinones, carotenoid, coumarin, tannin, phenols, saponins, terpenoids, steroids and amino acids. By determining the Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC), the antibacterial activity was studied utilising the Disc infusion technique (MBC). The extracts were made by serial dilution (dilution by half) with sterile distilled water solvent utilising water, ethanol, and chloroform at various concentrations. On Brain Heart Infusion agar, the crude extracts were tested along with Ampicillin as a positive control and water as a negative control.

Result: Results indicated that *Citrus maxima* crude extracts had an antibacterial activity against oral streptococci and exhibited a wide range of inhibition. The extracts were significantly potent against the bacterial strains in concentrations up to 3.9 and 250 µg/mL respectively. The extracts were shown to be less efficient on *S. mitis* and more efficient on *S. sanguis, S. salivarius* where ethanol extract showed highest efficiency in comparison with Ampicillin.

Conclusions: The findings exhibited in this research study provides the scientific evidence for the primal usage of plant extract as a medicinal ethno-nutraceutical. The antimicrobial activity quantified by appreciable bacterial sensitivity interprets *Citrus maxima* is considered to be effective persuasive therapeutic stand in compared to synthetic medicines.osynthetic adjunct pigments of cyanobacteria.

Keywords: Dental caries, Citrus maxima, Oral streptococci, Antibacterial, Ethno-nutraceutical.

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BACKGROUND

The remarcable discovery of antibiotics from ancient to recent era considered as potential strategy, has beneficially nursed in combating against enormous bacterial infectious diseases in humans. In developed and emergent nations bacterial infection is considered as one of the substantial benefactors to human illness. Although, with each progressing decade, bacteria that could breach multiple antibiotics are setting up to become highly common, steering to a rise in mortality rate, morbidness and healthcare sector cost. The antibiotic resistance catastrophe has been steered to the excessive usage and exploitation of these medicament, even due to reduced economic leverages and demanding regulative necessity there is huge deficiency in development of novel drug by the pharmaceutical production industry (Ahmad & Beg, 2001). Due to multifactorial elements a cognate mechanism occurs in microbial strains causing a raise in drug resistant micro-organisms emergence further leads to antimicrobial resistance that severely jeopardized the usage of antibiotics during therapeutics of biochemical system in humans. Perpetually akin newly developed resistant bacterial mutants reduce the efficiency of present remedial measures giving rise to critical community health hazards (Singh et al., 1980; Arseculeratne et al., 1985). Because the illness produced by pathogenesis of mutant bacteria is difficult to cure, healthcare practitioners are concerned about the development of medication resistance in mutant bacteria.

On that account Balhaddad et al., 2018, it's a challenge presently in controlling microbial infectious illnesses due to the difficulty in validating, profiling, and developing innovative natural antibiotic medicines. Although medicinal research has progressed in recent years as a result of scientific study and the adaptation of new methods, the foundation for its expansion remains firmly entrenched in traditional ayurvedic therapy and nutraceuticals (Dziedzic et al., 2013). In diverse parts worldwide plants as herbs are holistic pharmacological prospective source of antimicrobial agents. About 60 to 90% of populations in under-developed nations primarily uses phyto-derived therapeutics.

Plants have been utilized as natural drug sources exclusively from much before 5000 years, as a constituent of analgesics, antibiotics, antioxidants, cardioprotective. The floral existence i.e. plants are an abundant sources of phytoactive biometabolites such as phenols, alkaloids, tannins, proteins, terpenoids, and flavonoids, and their ability to research and scrutinise *in vitro* antimicrobial qualities enables them to investigate and scrutinise *in vitro* antimicrobial capabilities (Hamilton-Miller et al., 2001; Burapadaja et al., 1995; Baomin et al., 2000). The antimicrobial research study of traditional plant-based medications has been re-evaluated and thoroughly investigated during the previous decade. Secondary metabolites derived from phytomedicines offer a potential and significant function in the prevention, detection and therapeutic ailment of pathogenic illnesses in humans. However, research has shown that crude solvent extracts from flora and fauna are important bioactive roots of anticancer, antibacterial and antiviral agents in the allopathic medication system.

Oral disorders such as dental caries, oral mucosal, and periodontal diseases are among the chronic health conditions that have a global economic impact. However Oral health and the dental healthcare have been overlooked from mainstream establishment in health policy and health-care systems (Park et al., 2014; Huang et al., 2017). In developing countries about 10 % of healthcare expenditure is connected with dental healthcare and hygiene. Oral diseases inordinately affect poor and marginalised species in society, being nearly associated to socioeconomic stature (Benavente-Garcia& Castillo, 2008). Dental caries i.e. tooth decay has been identified being among the ten most common chronic clinical oral infections, and it is a major human illness that affects 9% of the world's population. The chronic dental caries disease is characterised by a number of symptomatic clinical conditions, including the colonisation of pathogenic microbiota, poor nutritional habitual custom, and poor oral hygienic conditions, all of which contribute to the demineralization of teeth hard tissues as consequence of acid production caused by bacterial fermentation of food (Byun et al., 2004). On teeth surfaces oral microbes inclined to form multi microbial communities, known as dental biofilm and that lay out the habitat for cariogenic pathological microbes. Oral streptococci were the first cariogenic species to be identified, and they play a critical role in the blossoming of caries and the creation of dental plaque biofilms De Britto et al., 2012. The aetiology of dental caries has been primarily caused by four reasons: bacteria, time, sensitive surface area of tooth, and fermentable carbohydrates. Because of excessive prevalence of oral caries induced due to biofilm construction leads to increased microbial resistance against antibiotics, hence it necessitates for alternative therapeutic methods. In India there is high incidence and prevalence of dental caries in spite of modernization in diagnostic technologies for treating oral pathological condition compared to other South Asian countries (Badria & Zidan, 2004; Ozkan, 2002). Dental caries is a chronic infectious, epidemic disease occurring from cohesion of specific oral pathogenic bacteria with tooth at distinct sites, primarily *Streptococcus mutans, S. mitis, S. constellatus, S. sanguis, S. salivarius, S. sorbinus, S. Oralis, S. anginosus, S. gordonii, S. intermedius* are a few acid-tolerant groups of cariogenic bacteria are linked to the production of dental biofilms and caries (Zhicen, 1987; Arias& Ramon-Laca, 2005).

These cariogenic microbial infections are a crucial etiologic element in the development of dental plaques, leading to colonisation of tooth surfaces and the generation

of plaque via the capacity to synthesise and bind extracellular polysaccharides (glucan) utilising the enzyme glucosyltransferase. By converting sugar to lactic acid, they also aid in the demineralization of inorganic tooth structure. Mouth, throat, heart, and gut are the primary habitation environments for cariogenic streptococci. As a result, dental caries has various chemical components like adherence to enamel surfaces, the formation of acidic metabolites, the ability to generate glycogen reserves, and the ability to synthesise extracellular polysaccharides (Babu et al., 2012). Cariogenic bacteria have been expelled from the oral microflora using a variety of antibiotics and antimicrobial treatments. However, owing to unfavorable adverse effects such as the bacterial susceptibility, tooth discoloration, vomiting, and diarrhea, their clinical use is limited. Such complications highly influence the vitality of advanced research investigations to reinforce a natural substitute antibacterial nutraceutical from plant-based sources with a direct focused emphasis on human health welfare and to improve dental caries treatments (Hayati et al., 2011; Gonçalves et al., 2008; Deng et al., 2012). In current scenario, the cultivars of *Citrus* species and their varieties are outcome of inter species hybridization between different antecedent taxon. Out of them, *Citrus maxima*, familiarly correlated with the pummel botanic groups, were often significant as the prime ancestor of the grapefruit (*Citrus paradisi*) and the tangelo (*Citrus reticulata*). The *Citrus maxima* is more often known as shaddock or pomelo, a herbal traditional ayurvedic aromatic shrub belongs to family *Rutaceae* is indigenous i.e. native to tropical parts of Asia. Because of notable morphological features like it's a medium sized mostly spiny tree; with compound leaves having winged petioles and bisexual prominent fragrant flowers clusters are with sweet smell. The fruit is always oblate, or pear shaped, big size, commercially produced in India and regarded in western world chiefly as the "principal ancestor of the grapefruit"(Zarina& Tan, 2013; Puri et al., 2011). *Citrus maxima* have great ethno-nutraceutical efficiency for healing a variety of pathogenic disorders in humans due to its extraordinary therapeutic impact. As citrus has a very role vital in our diet, like as lemons, oranges, grapefruits and limes and are predominant key sources of major nutrients, have been found to be effective in preventing several infectious diseases. These comprise folic acid, vitamins C, dietary fibres, carotenoids, dietary fibres, potassium, and a broad range of bioactive compounds. *Citrus maxima* can be assessed as "a therapeutic package unto itself" constitutes enormous bioactive metabolites like alkaloids, carbohydrates, carotenoids, coumarins, flavonoids, flavanones, monoterpenes, steroids, Sesquiterpenes, flavones, flavonols and amino acids possessing antimicrobial, antioxidative properties, anti-diabetic, anti-atherosclerotic, anti-tumour, antihypertensive, antiaging, analgesic, and anti-inflammatory properties (Puri et al., 2011).

In systematic traditional research investigations, it has been examined and analysed the existence of various phytochemicals and their originates in pomelo. According to epidemiological studies, consuming a phytochemical enrich diet that includes fruits and vegetables is linked to a lower risk of cancer, hypertension, and diabetes. *Citrus maxima* bioactive components have proven antibacterial activity *in vitro* against microorganisms that cause human illness in recent years (Jannadi et al., 2019; Huang et al., 2018). The peel of Citrus fruit is an abundant element of coumarins, β and α-sitosterol, glycosides, flavonoid, glycosides and volatile oils. Numerous citrus species are well-known for their phytochemicals and antibacterial properties. The hydroalcoholic citrus fruit peel extracts were also shown to have antifungal activity and phytochemical components, according to Pandey et al. The citrus fruit peels include tannins, lowering sugar, and flavonoids, according to certain research investigations. The peels of Citrus fruits which are usually considered as wastes can serve now as a potent and highly economical antimicrobial source as they have no side effects and are available for no cost. A number of research evaluation studies on pomelo extract showed the antimicrobial functionalities *in vitro*. In traditional remedial approach, due to volatility property, the *Citrus maxima* shown how the use of selected fruit peels as remedies for edoema, cough, and epilepsy (Giacaman et al., 2015; KunduSen et al., 2011; Xu et al., 2008).

As a result, the use of perennial *Citrus maxima*, a phyto-nutraceutical ayurvedic agent, as an effective replacement for multi-drug resistance antibiotics is now being explored intensively in India and is thought to play an essential function in human healthcare therapies Weng et al., 2011. Despite the fact that research into the antibacterial efficacy of *Citrus maxima* peels against cariogenic bacteria has been limited so far. Since the primal era, *Citrus maxima* have been known as a nutrient-rich diet with various significant health benefits in healing human ailments. So, in light of the high frequency of oral pathogenic disorders caused by cariogenic microorganisms, as well as the growing public interest in ayurvedic medicinal therapeutics, leads to the foundation of present research study which aims to estimate the bio profile antibacterial activity of crude ethanolic, aqueous, and chloroform extracts of *Citrus maxima* peels *in vitro* against several oral cariogenic microbial pathogen *Streptococcus mutans, S. sanguis, S. Mitis, S. salivarius, S. sorbinus*. However, a number of literature reseach study did not report much research investigation in condusive to the effect of *Citrus maxima* crude peel extract against oral disease causing microbial pathogens. Therefore, the present research study was undertaken to analyse and compare the *in vitro* antibacterial properties of different crude extracts of *Citrus maxima* fruit peels against dental caries causing pathogens and to determine the minimum inhibitory

concentration (MIC) of each crude peel extracts against each pathogen with a perspective of formulating a novel phyto-derived nutraceutical against cariogenic bacteria.

MATERIALS & METHODS

Collection and Identification Plant Material

The fruits of the *Citrus maxima.* "Devanahalli" variety was gathered from a public retail market in the city of Bengaluru, Karnataka, India for experimental investigation. Fruits were rinsed with distilled water thoroughly and surface water was wiped away using tissue paper. Fruits were manually peeled and skins were removed from arils. Additionally, peels were dried for 7 days at 33°C in an incubator before being pulverised using an electric grinder. The Ampicillin used in present study, was obtained from Hi-Media Laboratories pvt ltd.

Preparation of Aqueous, Ethanol and Chloroform extract of *Citrus maxima*

Separating bioactive plant components or secondary bioactive metabolites from inert material by employing a suitable solvent and standard extraction approach is a crucial and vital step in achieving a high-quality result (Kanes et al., 1993). Solvents including water, ethanol, and chloroform were chosen for plant extraction based on previous experimental research done on different regions of *Citrus maxima* (Boudries et al., 2012). *Citrus maxima* peels were properly cleaned in double distilled water. Using a pestle and mortar, 50 g of peels were blended with 100 mL of boiling hot water, then The residue was mixed with 400 mL of hot/cold water and agitated for 30 minutes to yield aqueous extract Brighenti et al., 2012. At 40°C, the pooled extract was centrifuged for 15 minutes at 10,000 rpm. The collected supernatant was concentrated using a lyophilizer and freeze drying. The extracted substance was known as ACM (Aqueous extract of *Citrus maxima* peels). The extract was filtered through a filter of 0.22-micron and kept at 20°C until needed. The weighing, labelling, and storing the dried extract in the refrigerator were all done. 50 gm of peels were weighed individually and then blended with 500 mL of each ethanol and chloroform in a mortar and pestle to preapre the ethanol and chloroform extracts. These were centrifuged for 10 minutes at 10000 rpm. A rotary evaporator was used to concentrate the clear supernatant between 38°C to 40°C. For further usage, the extracts were prepared by disoolving them in ethanol and chloroform, respectively, and stored at -20°C and the extract obtained was called as ECM (Ethanol extract of *Citrus maxima* peels) and CCM (Chloroform extract of *Citrus maxima* peels).

Activation of Cariogenic Microorganisms

Standard strains of cariogenic microorganisms *S. mutans* (ATCC 25175), *S. salivarius* (ATCC 13419), *S. sobrinus* (ATCC 33402), *S. sanguinis* (ATCC 10556) and *S. mitis* (MTCC 497) were obtained from the Hi-Media Laboratories Pvt Ltd and Micro Type Culture Collection and gene bank (MTCC) in lyophilized form. Cariogenic Bacterial strains were activated by inoculation in the brain heart infusion agar (BHIA) culture medium followed by 24 hours of incubation at 37° C. A 24-hour culture was used for preparation of microbial suspension. The concentration of microorganisms in the microbial suspension was adjusted to 0.5 McFarland standard using a spectrophotometer at a wavelength of 625 nm (a McFarland standard is a chemical solution with a turbidity comparable to that of microbial suspension. Using this suspension, number of bacteria per each milliliter of the suspension can be estimated, which is equal to 1.5 × 108 CFU/mL).

Preliminary Phytochemical Screening of *Citrus maxima*

Phytochemical examinations for *Citrus maxima* peels were carried out for all the extracts as per the standard methods as shown in Table 1. Chemicals and Reagents Solvents Millons reagent, Fehling's solution A and B, Benedict's reagent, gelatin powder, bromine water, sulphuric acid, ferric chloride and glacial acetic acid were purchased from Hi-Media Laboratories pvt. ltd.

Establishment of Antibacterial Activity Assay

The well diffusion method was used for the primary screening of extracts for antibacterial activity. BHIA was used to culture 500 μL of each microbial suspension at a standard concentration of 0.5 McFarland (swabbed on the plate. Test pure cultures of human cariogenic bacteria i.e. *S. sanguinis, S. mutans, S. salivarius, S. sobrinus and S. mitis* were evenly smeared to the sterile BHI agar medium (20 ml) Petri plates using sterile cotton swabs. The agar surface was then drilled with 8mm-diameter wells. Using sterile distilled water as the solvent, the extracts were successively diluted (by half) to create different concentrations; 100 μL of each concentration of extract was poured into each well. The plates were incubated at 37^0 C for 24 hours. The diameter of the growth inhibition zone was measured in millimeters. This process was repeated in triplicate and the mean diameter of the growth inhibition zone was calculated for different concentrations of the extract Petersen et al., 2005.

Minimum Inhibitory Concentration

The MIC is defined as the lowest concentration of an antimicrobial agent that inhibits the growth of a microorganism (0.5 McFarland standard in this study). It is the minimum concentration of the extract that completely prevents visible growth and multiplication of bacteria compared to the negative control group. To determine MIC, macro dilution method according to the standard technique

Table 1: Qualitative phytochemical analysis of *Citrus maxima* peel extracts.

		Peel extract			Fruit extract		
S No.	Phytochemical constituent	Aqueous	Ethanol	Chloroform	Aqueous	Ethanol	Chloroform
1	Alkaloid	+	+	+	+	+	+
2	Amino acid	+	+	+	+	+	+
3	Anthraquinones	+	+	+	+	+	+
4	Carotenoid	_	+	+	_	+	+
5	Carbohydrate	+	+	+	+	+	+
6	Coumarin	+	+	+	+	+	+
7	Flavonoids	+	+	+	+	+	+
8	Phenol	-	+	+	-	+	+
9	Saponin	+	+	+	-	-	-
10	Steroids	+	+	+	+	+	+
11	Tannin	-	+	+	-	+	+
12	Terpenoids	+	+	+	-	+	-
13	Reducing Sugar	-	+	+	-	+	+
14	Quinones	+	+	+	+	+	+
15	Protein	-	+	+	-	-	-

described by the clinical and laboratory standards institute (CLSI) was used. Different concentrations of the extract were prepared by serial dilution (dilution by one-half) in BHI broth medium. Using this medium, the 0.5 McFarland standard suspension was diluted 1 to 150 to obtain a bacterial count of 106 CFU/mL. Microbial suspension was then diluted by one-half using the culture medium and 1mL of it was added to the tubes containing serially diluted extract Dastjerdi et al., 2014. The negative control tube only contained the culture medium and extract with no microbial suspension. The positive control tube contained culture medium and microbial suspension with no extract. After 24 hours of incubation at 37^0 C, growth and proliferation of microorganisms were evaluated and the MIC value of the extract for each bacterial strain was determined. This test was repeated in triplicate for each microorganism.

Minimum Bactericidal Concentration

After determination of MIC, 20 μL of the suspension in the tube containing MIC of the extract and tubes showing no bacterial growth were cultured on plates containing BHIA Vasconcelos et al., 2006. After 24 hours of incubation at 37^0 C, the plates were evaluated for growth of microorganisms. The concentration with no bacterial growth was determined as MBC. This test was repeated in triplicate for each microorganism. Using the disc diffusion technique, the impact of 0.2 percent ampicillin on the microbes was assessed, and the MIC and MBC values of ampicillin for the microorganisms were also calculated Philip et al., 2010.

Statistical Analysis

All values were analysed that obtained in zone of inhibition measuring experiment for statistical significance by one-way analysis of variance (ANOVA) using statistical software GraphPad prism version 8.0. The graphs were also derived using the above-mentioned software. The data were demonstrated as Mean ± SEM of three trials. A P value <0.05 was considered significant. The present study results obtained in MIC and MBC were interpreted for statistical significance using one-way Analysis of Variance (ANOVA) and variations among means were compared using Post Hoc Duncan's Multiple Range test at $P \leq 0.05$ was considered significant [SPSS version 11.5.0 (SPSS Inc., Chicago, IL)].

RESULTS

The present research study of preliminary phytochemical screening of *Citrus maxima* peels is very efficient in examining chemical bio-metabolites that may further lead to their quantitative estimation and also in locating the derived source of pharmacologically bioactive metabolic constituents. Peels of the *Citrus maxima* plant are expected to offer substantial ethno-nutritive benefits since they contain a wide range of phytochemicals. Based on primary biochemical screening experimental research investigation exhibited in Table 1, pomelo peels contained certain important derivatives like Alkaloids, phenols, tannins, flavonoids, quinones, carotenoids, steroids, terpenoids, proteins, coumarin, carbohydrate, anthraquinones and saponin is evaluated in the fruits and peels crude extract mostly leading up to the fact that *Citrus maxima* is significantly rich in bioactive metabolites. As per systematic research evaluation its investigated that bioactive metabolites consist essential medicinal therapeutic properties for human health and diseases.

Due to presence of potential phyto-metabolites, it extends further perspective for antibacterial investigation in different crude extracts of *Citrus maxima* peels to proclaim therapeutical efficacy for treatment of various human oral

pathological diseases. Additionally water, ethanol, and chloroform were used to deracinate crude extracts of *Citrus maxima* peels. The positive control for all microbes revealed exceptionally broad inhibition zones, whereas the negative control showed no significant inhibitory impact while examining *Citrus maxima* peels' antimicrobial properties.

The mean (and standard deviation) diameter of the growth inhibition zone caused by the activity of crude aqueous extract, crude ethanolic extract, and crude chloroform extract of *Citrus maxima* peels against *S. sanguinis, S. salivarius, S. mutans, S. sorbinus, and S. mitis* is depicted in Figure 1, 2,3 ,4, 5.

Using the well diffusion method, the crude ethanolic extract of *Citrus maxima* peels demonstrated significant inhibitory activity on the growth and multiplication of all five bacteria *Streptococcus mutans, S. salivarius, S. sanguis, S. sorbinus, S. mitis* using the well diffusion technique. The widest diameter of growth inhibition zone belonged to *S. sanguinis, S. sobrinus* and the smallest zone shown by *S. mutans* respectively. The crude aqueous extract of *Citrus maxima* peels showed intermedial inhibitory effects on the growth and proliferation of all five bacteria *Streptococcus mutans, S. salivarius, S. sanguis, S. sorbinus, S. mitis* using the well diffusion technique. The largest diameter of growth inhibition zone belonged to *S. sangunis*, *S. sorbinus and* the smallest shown by *S. mitis* respectively Jalasvuori et al., 2012.

Using the well diffusion method, the crude chloroform extract of *Citrus maxima* peels had extremely substantial inhibitory impact on the growth and multiplication of all five bacteria *Streptococcus mutans, S. sanguis, S. salivarius, S. sorbinus, S. mitis* using the well diffusion technique. The largest diameter of growth inhibition zone belonged to *S. salivarius*

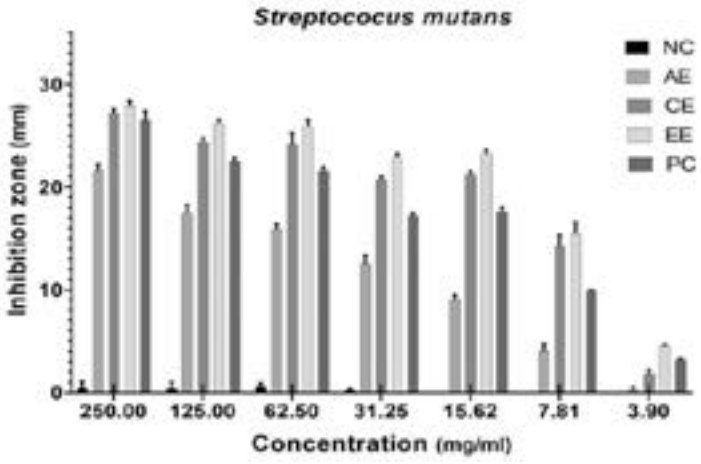

Figure 1. Efficacy of various crude extracts of *C.maxima* peel on *S.mutans*. NC-Negetive control, AE-Aqueous Extract, CE-Chloroform Extract, EE-Ethanol.

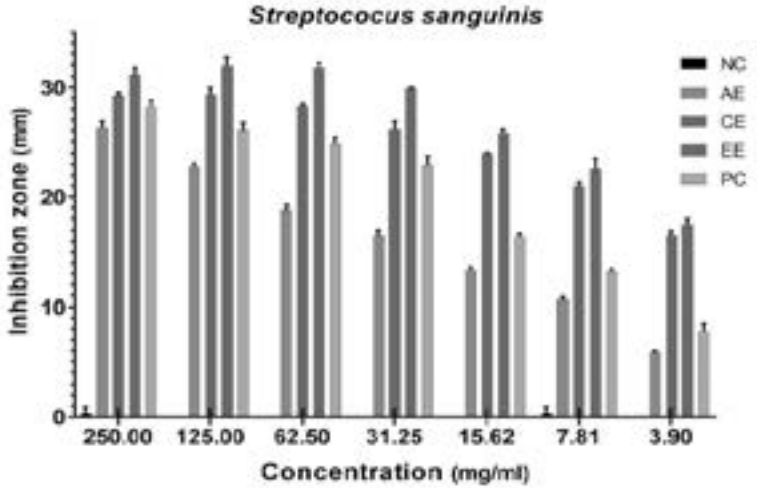

Figure 2. Efficacy of various crude extracts of *C.maxima* peel on *S.sanguinis*. NC-Negetive control, AE-Aqueous Extract, CE-Chloroform Extract, EE-Ethanol Extract, PC-Positive Control.

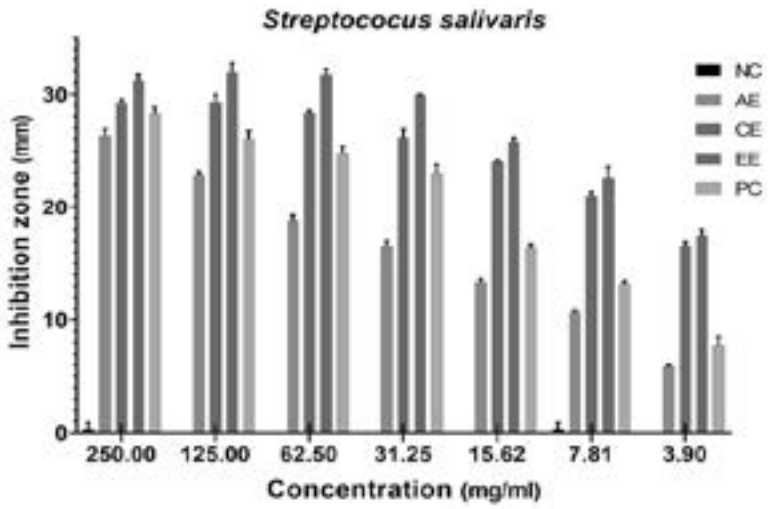

Figure 3. Efficacy of various crude extracts of *C.maxima* peel on *S.salivaris*. NC-Negetive control, AE-Aqueous Extract, CE-Chloroform Extract, EE-Ethanol Extract, PC-Positive Control.

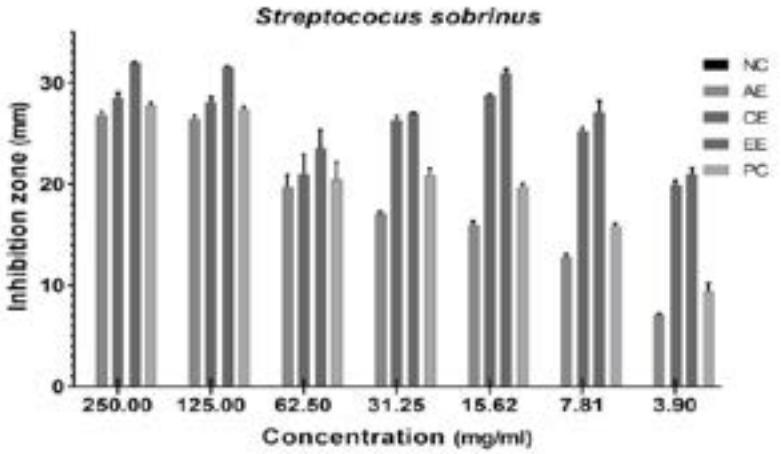

Figure 4. Efficacy of various crude extracts of *C.maxima* peel on *S.sobrinus*. NC-Negetive control, AE-Aqueous Extract, CE-Chloroform Extract, EE-Ethanol Extract, PC-Positive Control.

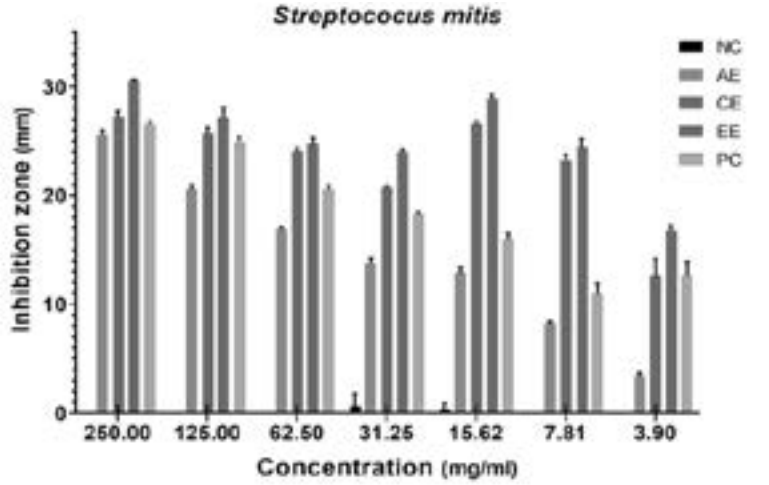

Figure 5. Efficacy of various crude extracts of *C.maxima* peel on *S.mitis*. NC-Negetive control, AE-Aqueous Extract, CE-Chloroform Extract, EE-Ethanol Extract, PC-Positive Control.

S. sorbinus, S. sanguis, and the growth inhibition zone with the lowest dimension belongs to *S. mitis* respectively.

Citrus maxima peels crude aqueous extract, ethanolic extract, and crude chloroform extract were tested for MIC and MBC using the serial dilution technique depicted in Tables 2a, 2b, and 2c and Table 3.

It was found that all the crude extract of *Citrus maxima* peels inhibited oral microbes *Streptococcus mutans, S. sanguis, S. salivarius, S. sorbinus, S. mitis*. In addition, the concentration of 3.9 mg/ml was the most effective crude ethanolic extract against *S. sanguinis* compared with other cariogenic streptococci. Further the concentration of 31.3 mg/ml was the most effective crude aqueous extract against *S. sanguinis* compared with other cariogenic streptococci as well the concentration of 15.6 mg/ml was the most effective crude chloroform extract against *S. sanguinis* compared with pathogens Li et al., 2006.

The ethanolic extract of medicinal plant *Citrus maxima* peels had MBCs of 3.9 – 15.6 mg/mL against *Streptococcus mutans, S. salivarius, S. sanguis, S. sorbinus, S. mitis.* The crude ethanolic extract of *Citrus maxima* peel interpreted the most outstanding in-vitro antibacterial activity with MBC of 3.9 mg/mL against *S. sanguinis* and *S. sorbinus.* In addition, the MBC calculated values indicated that the crude aqueous extract had the greatest impact on *S. sanguinis*, at 31.3 mg/ml, when compared to other cariogenic streptococci. Additionaly, crude chloroform extract shown that highest potency against *S. salivarius, S. sanguis* and *S. sorbinus* at the concentration range of 15.6 mg/ml in comparison to other cariogenic streptococci under research study Ruiz-Linares et al., 2014.

The highest antibacterial effect of *Citrus maxima* peel crude ethanolic extract was on *S. sanguinis* with MIC and MBC values of 3.9 mg/mL and lowest antimicrobial activity on *S. mitis.* As a result, dental caries bacteria were shown to be moderately resistant to *Citrus maxima* peel crude ethanolic, aqueous, and chloroform extracts Karimi et al., 2012.

Table 2a: The MIC (mg/mL) of *Citrus maxima* peel Aqueous extract (ACM) for different cariogenic bacteria.

Pathogens	Aqueous Extract mg/mL						
	250.0	125.0	62.5	31.3	15.6	7.8	3.9
S.salivaris	-	-	+	+	+	+	+
S.mutans	-	-	+	+	+	+	+
S.sanguinis	-	-	-	-	+	+	+
S.sobrinus	-	-	-	+	+	+	+
S.mitis	-	+	+	+	+	+	+

Table 2b: The MIC (mg/mL) of *Citrus maxima* peel Chloroform extract (CCM) for different cariogenic bacteria.

Pathogens	Chloroform Extract mg/mL						
	250.0	125.0	62.5	31.3	15.6	7.8	3.9
S. salivaris	-	-	-	-	-	+	+
S. mutans	-	-	-	-	+	+	+
S. sanguinis	-	-	-	-	-	+	+
S. sobrinus	-	-	-	-	-	+	+
S. mitis	-	-	-	+	+	+	+

Table 2c: The MIC (mg/mL) of *Citrus maxima* peel Ethanol extract (ECM) for different cariogenic bacteria.

Pathogens	Ethanol Extract mg/mL						
	250.0	125.0	62.5	31.3	15.6	7.8	3.9
S. salivaris	-	-	-	-	-	-	+
S. mutans	-	-	-	-	-	+	+
S. sanguinis	-	-	-	-	-	-	-
S. sobrinus	-	-	-	-	-	-	-
S. mitis	-	-	-	-	-	-	+

Table 3: The MBC (mg/mL) of *Citrus maxima* peel extracts for different cariogenic bacteria.

Pathogens	Aqueous Extract (mg/mL)	Chloroform Extract (mg/mL)	Ethanol Extract (mg/mL)
S.salivaris	125	15.6	7.8
S.mutans	125	31.3	15.6
S.sanguinis	31.3	15.6	3.9
S.sobrinus	62.5	15.6	3.9
S.mitis	125	62.5	7.8

DISCUSSION

In paramount, from past 70 years antibiotics have been transformed as a therapeutic keystone of human pathogenic diseases and hence substantially worldwide pitched in development of health sector. The huge public populace is surviving presently from life-threatening diseases due to crucial utilization of antibiotics Perez et al., 1994. But their irrational and improper usage has extensively influenced in rise of resistant microbial inhabitants leading to lessening the efficacy of newly introduced antibiotics (Giacaman et al., 2015). In consequence antibiotic resistance has become a major public health concern for the reason that the antibiotic-resistant microbe allied with the diseases pathogenic to humans transferred via enormous sources like food, animal Lim et al., 2006. As a result of this, a different powerful naturally occurring antimicrobial source has emerged, mostly from plants, with the aim of finding novel bioactive therapies consisting of bioactive chemicals to combat the phenomena of multidrug resistance. In addition, this has led to a growing interest in medications that are produced from traditional ethnomedicinal plants in the modern period in the healthcare sectors of both developed and developing nations (Xu et al., 2008; Jang et al.,2010).

As a source of innovative medication nutraceuticals, herbal medicinal products have been widely employed for ages to treat infectious disorders. In the pharmaceutical business and in a variety of phyto-therapeutic applications, secondary metabolites originating from plants are widely used because of their remarkable structural diversity (Arias& Ramon-Laca, 2005; Xu et al., 2008). Traditional herb extracts from plants and their components, including as roots, stems, leaves, and flowers, have undergone extensive study, trials, and medication evaluations in primary healthcare in order to assess their considerable multilayer therapeutic potential.

In addition to dental disorders, dental caries is one of the most prevelant chronic and multi-factorial oral infective diseases globally, mostly produced by acid diffusion into enamels and dentine bacterial metabolic activity. Cariogenic bacterial biofilms frequently specified as aggregation of microbes (commonly bacteria) anchored in salivary polymer matrix and bacterial origin on tooth surfaces leading to biochemical transformations, a major contributor in the pathogenicity of dental caries and other periodontal diseases (Yamada et al., 2011; Limet al., 2006). As per considerable documentation, Streptococci such as *S. sanguinis*, *S. mutans*, *S. salivarius*, *S. sorbinus*, and *S. mitis* are the first species to cause oral caries. Among all cariogenic microbes *Streptococcus sanguinis* and *Streptococcus mutans* are the primary superintend for origination, accumulation and maturation of dental plaque.

Streptococcus mutans is essential for caries formation, proliferation, and spread *S. salivarius* is a prevailing colonizer of the oral mucosal surfaces especially in the dorsal aspect of the tongue, buccal mucosa, and saliva (Owoseni& Ajayi, 2010; Boudries et al., 2012). The cariogenic phenotypic factors assimilated by the oral streptococci are attributed to the production of indissoluble glucans from sucrose, their acidogenicity further acid tolerance and their ability to adhere to tooth surfaces raises the possibility that they may be an effective competitor in the formation of biofilm. Substantial prophylactic and suppressing therapeutic techniques have been thoroughly studied and experimentally analysed for prevention and degradation of dental caries fully and even presently the analytical research investigations are still going on (Naderi et al., 2011; Khan et al., 2015). Furthermore, most antibacterial sources may promote the emergence and development of resistant microbial strains as a result of this persistent chronic infectivity. Accordingly, it becomes imperative for the present remedial salutiferous research to analyse naturally accessible bioactive candidates which are insusceptible for humans and target specific for dental caries, due to their intangible side effects (Singh, 1986). Worldwide this thrust nobel therapeutic perspective has enormous implications, specifically in India due to the indigenous prevalence and growth of pathogenic oral caries Nishijima et al., 2012. In present era, to diminish the appearance of dental caries various scientific traditional medical research studies exemplified the promising antibacterial properties of diverse number of ayurvedic herbs as an ethno-nutraceutical in oral hygiene. One of the most potent plant in traditional phyto medical system is *Citrus maxima* has been attributed as a holistic medicinal plant to cure a broad range of pathogenic diseases globally (Chopra et al., 1960).

Citrus maxima (shaddock or pomelo), a member of the *Rutaceae* family, is a fragrant perennial plant used in traditional ayurveda medicine, is often regarded as potent nutrient diet and phyto-drug against numerous pathogenic diseases. The plant's varied components extensively used by different folk tribe communities. Ayurvedic, Siddha, and Unani medicine practitioners recommend it highly for the treatment of a broad range of debilitating human pathologies, according to ethnographic study. *Citrus maxima* is a pharmacologically active crop plant primeval traditional plant native of Asia and widely distributed indigenous, commercially grown in Indian subcontinent. Geographically it is also crop plant of Japan, USA, China, Indonesia, Philippine and Thailand. *Citrus maxima* has a range of bioactive metabolites discovered during flora biochemical investigation of the crude extracts of the peel revealed the existence of alkaloid, phenols, tannins, flavonoid, saponins might be an effective nutraceutical in treating a variety of pathogenic disorders (Alviano et al., 2008; Siang, 1983; Saraya et al., 2008). As per Scientific reports, a focused approach is developing in correlating the bioactive constituents of a medicinal herb with its pharmacognistic and pharmacological activity.

The current research analysis was aimed systematically to demonstrate the susceptible antibacterial activity of crude ethanolic, aqueous and chloroform extracts of *Citrus maxima* peels using the well diffusion method *in vitro* on cariogenic bacteria *S. sorbinus, S. mitis*, *Streptococcus mutans, S. sanguinis, S. salivarius*. For a number of pathogenic bacteria, calculations were also performed to figure out the growth inhibition zone diameter, MIC, and MBC values of the crude extracts. The outcomes were compared to ampicillin, which was used as a positive control. Based on the experimental results, it's attributed that the crude extracts of *Citrus maxima* peels showed preclusive effect on the origination, development and proliferation of all five cariogenic microbial strains. In this research study, the MIC of *Citrus maxima* peel is investigated and compared it with ampicillin against adhesion of *S. mutans*, *S. sanguinis, S. salivarius, S. sorbinus* and *S. mitis*. It was discovered that crude extracts of *Citrus maxima* peel had a higher capacity for inhibiting streptococci adhesion. On *S. sanguinis*, *Citrus maxima* peel crude ethanolic extract had the strongest antibacterial activity. As a consequence, dental caries bacteria were shown to be less resistant to *Citrus maxima* peel crude ethanolic, aqueous, and chloroform extracts Kakiuchi et al., 1986.

In demonstration by Sudto et al., 2009, the Pomelo, the citrus fruit belongs to the genus *Rutaceae*, with a diameter of 30 cm and weight of 10 kg, approximately 30% of which is constituted by the peel. Thus, large quantities of pomelo peel are obtainable for use. As per Methacanon *et al.*, 2014 study Pomelo peel does not damage the environment and is easily obtained and its utilisation as an alternative natural source reduces wastage. Huang *et al.,* 2014 evaluated that the pomelo peel contains high amounts of phyto-constituents pectin, phenols, essential oils, soluble polysaccharides, active polysaccharides and flavonoids, which can reduce the filtration of mud, improve its rheological properties and hinder the hydration and expansion of clay.

Barrion *et al.*, 2014 manifested the antibacterial activity of the ethanolic extract (90%) of the phytochemical constituents of the pericarp, mesocarp, and segment membrane of *Citrus grandis* against gram negative bacteria *Escherichia coli* and *Salmonella typhimurium*. The highest antibacterial activity was seen by methanolic extract followed by acetone, water and petroleum ether Hintao et al., 2007.

Citrus flavonoids work as a scavenger of free radicals and exhibits anti-allergenic, analgesic, antimicrobial, anti-inflammatory, antiviral, antiulcer activities. In a comprehensive investigation in correspondence to Makynen *et al*., 2012 the antioxidant activity of *Citrus grandis* cultivars were determined where the main flavonoids in the fruit hesperidin and neohesperidine dihydrochalone showed effectivity against superoxide formation as well as scavenging reactive oxygen species. Zhao *et al.,* 2019 evaluated the anti-inflammatory effect of Citrus grandis by the methanolic (90%) and ethyl acetate extract (coumarin fraction, 32 g) of the peel of the fruit in animal model. C. grandis functions as a good anti-inflammatory agent because of polysaccharides and bioactive coumarins present in the peel which blocks the inflammatory mediators and pro-inflammatory cytokines Ancuceanu et al., 2019.

The predominant factor majorly influencing the MIC is the influence of the geographical setting on the fauna of medicinal plant species causing variance in the amalgam of crude extracts of ayurvedic folk herb, senescence of plant, developing and growing stage, harvesting conditions and season, technique of drying, and extraction procedure. In addition, crude extracts of distinct parts of the ayurvedic medicinal herb have oscillation in the level of antimicrobial effect and cariogenic bacteria have varying susceptibility to different extracts of the herb. Alcoholic extract of *Citrus maxima* peel has significant isolated bioactive elements like tannins, phenols, alkaloids, flavonoids, gamma terpinene, steroids, betacyanins, coumarin, proteins. Therefore, the high antibacterial activity of *Citrus maxima* has been established by all prior study assessments; nonetheless, additional extensive examination is suggested on the therapeutic potency of bioactive metabolites Rhodes et al., 2014.

Furthermore, in inference of the present experiential research investigation, its summarised as in dental plaques, there is an existence of a structural complex microbial community. The interpretations showed the presence of various biochemical components like flavonoids, steroids, phenols, alkaloids, carbohydrate glycosides, saponins, and amino acid aids in exhibiting ethno-nutraceutical potent activity of the ethanolic, aqueous and chloroform crude extracts of *Citrus maxima* peels would be crucial bioactive resources for the production of antibacterial agents as a pharmaceutical bioformulation against *S. sanguinis, S. salivarius, S. mutans, S. sorbinus* and *S. mitis*, although, for latter higher concentration of the crude extracts are preferred to attain MIC and MBC. However, for few cariogenic bacteria efficacies of extracts was less than ampicillin, they may have a prospective role for dental caries obstruction. Consequently, this research analysis could potentially exhibit a scientific substratum for the traditional ayurvedic therapeutics of *Citrus maxima* peels on oral and dental pathogenic microorganisms, out laying the improvement of oral health and hygiene as well as suppressing the complications and lessening the treatment cost with synthetic drug. Beside this, additional clinical trial research evaluations seem prerequisite to analyse their significant potency and its beneficiary usage to human infectious illness as well as biological diversity ecosystem. Pomelo peel not only suitable constitutes bioactive metabolites and exhibits anti-bacterial activity, but it is also extensively available, conveniently accessible, low cost and environment friendly and sustainable resource. Accordingly

it's summarised as the *Citrus maxima peels* crude extracts demonstrated a potential ethno-nutraceutical activity against cariogenic streptococci, can further scientific investigation may lead to development of crucial bio-formulation for the therapy of a comprehensive dental caries ailment Ge et al., 2008.

Conclusion

The experimental demonstration of *Citrus maxima* peels establishes the scientific evidence for the usage of *Citrus maxima* peels as a traditional natural alternative against oral streptococci to proclaim therapeutical efficacy for treatment of various human oral pathological diseases, due to presence of ample number of potential phytochemical bioactive compounds Menezes et al., 2006. The findings analysed *in vitro* antimicrobial activity against dental caries pathogens validating perspectives for further *in vivo* clinical research studies to prompt the exact dosages and its efficacy in practical ailing conditions. Though, additional research trials at clinical level seem requisite to evaluate their effectiveness and advantages for human infectious illnesses and the environment. Subsequently *Citrus maxima's* nutraceutical bioformulations for the treatment of dental caries may take a step forward as a consequence of this study's findings. The conclusion is that the crude extracts of *Citrus maxima* peels exhibit high ethno-nutraceutical efficacy against cariogenic *streptococci* and determined as potent bioactive source.

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REFERENCES

Ahmad I, Beg AZ (2001). Antimicrobial and phytochemical studies on 45 Indian medicinal plants against multi-drug and resistant human pathogens. J Ethnopharmacol. 74: 113-123.

Arseculeratne SN, Gunatilaka AAL, Panabokke RG (1985). Studies on medicinal plants of Sri Lanka. J Ethnopharmacol. 13: 323-335.

Ancuceanu R, Anghel AI, Ionescu C, Hovaneț MV (2019). Clinical trials with herbal products for the prevention of dental caries and their quality: A scoping study. Biomolecules. 9:884.

Arias BA, Ramon-Laca L (2005). Pharmacological properties of citrus and their ancient and medieval uses in the Mediterranean region. J Ethnopharmacol. 97: 89–95.

Alviano WS, Alviano DS, Diniz CG, Antoniolli AR (2008). In vitro antioxidant potential of medicinal plant extracts and their activities against oral bacteria based on Brazilian folk medicine. Arch Oral Biol. 53: 545–552.

Benavente-Garcia O, Castillo J (2008). Update on uses and properties of citrus flavonoids: New findings in anticancer, cardiovascular, and anti-inflammatory activity. J Agric Food Chem. 56: 6185–6205.

Badria FA, Zidan OA (2004). Natural products for dental caries prevention. J Med Food. 7: 381-4.

Balhaddad AA, Kansara AA, Hidan D, Weir MD (2018). Toward dental caries: exploring nanoparticle-based platforms and calcium phosphate compounds for dental restorative materials. Bioact Mater. 4: 43–55.

Babu J, Blair C, Jacob S, Itzhak O (2012). Inhibition of Streptococcus gordonii metabolic activity in biofilm by cranberry juice high-molecular-weight component. J Biomed Biotechnol. 590384.

Brighenti FL, Gaetti-Jardim E, Danelon M, Evangelista GV (2012). Effect of Psidium cattleianum leaf extract on enamel demineralisation and dental biofilm composition in situ. Arch Oral Biol. 57: 1034–1040.

Boudries H, Madani K, Touati N, Souagui S (2012). Pulp antioxidant activities, mineral contents and juice nutritional properties of Algerian Clementine Cultivars and Mandarin. Afr J Biotechnol. 11: 4258–4267.

Burapadaja S, Bunchoo A (1995). Antimicrobial activity of tannins from Terminalia citrine. Planta Medica. 61: 365.

Byun R, Nadkarni MA, Chhour KL, Martin FE (2004). Quantitative analysis of diverse Lactobacillus species present in advanced dental caries. J Clin Microbiol. 42: 3128–36

Baomin F, Yuehu P (2000). Chemical constituents of peels of Citrus grandis, Shenyang Yaoke Daxue Xuebao. 17: 332-333.

Chopra CL, Bhatia MC, Chopra IC (1960). In vitro antibacterial activity of oils from Indian medicinal plants. J American Pharma Assos. 49: 780.

De Britto AJ, Sebastian SR, Sujin RM (2012). Antibacterial activity of selected species of Lamiaceae against human pathogens. Indian J Nat Prod Resour. 3: 334-342.

Dziedzic A, Kubina R, Wojtyczka RD, Kabala-Dzik A (2013). The antibacterial effect of ethanol extract of polish propolis on mutans streptococci and lactobacilli isolated from saliva. Evid Based Complement Alternat Med. 681891.

Dastjerdi EV, Abdolazimi Z, Ghazanfarian M, Amdjadi P (2014). Effect of Punica granatum L. flower water extract on five common oral bacteria and bacterial biofilm formation on orthodontic wire. Iran J Public Health. 43: 1688.

Gonçalves FA, Andrade Neto M, Bezerra JN, Macrae A (2008). Antibacterial activity of Guava, Psidium guajava linnaeus, leaf extracts on diarrhea-causing enteric bacteria isolated from Seabob Shrimp, xiphopenaeus kroyeri (Heller). Rev Inst Med Trop Sao Paulo. 50: 11–5.

Giacaman RA, Torres S, Gómez Y, Muñoz-Sandoval C (2015). Correlation of Streptococcus mutans and Streptococcus sanguinis colonization and ex vivo hydrogen peroxide production in carious lesion-free and high caries adults. Arch Oral Biol. 60: 154-9.

G. Xu D, Liu J, Chen X, Ye Y (2008). Juice components and antioxidant capacity of citrus varieties cultivated in China, Food Chem. 106: 545–551.

Ge X, Kitten T, Chen Z, Lee SP (2008). Identification of Streptococcus sanguinis genes required for biofilm formation and examination of their role in endocarditis virulence. Infect Immun. 76: 2551–2559.

G.-F. Deng, C Shen, X.-R. Xu (2012). Potential of fruit wastes as natural resources of bioactive compounds, Int J Mol Sci. 13: 8308–8323.

Gupta, SK, Bhatt P, Joseph GS, Negi PS (2013). Phenolic constituents and biological activities of leaf extracts of traditional medicinal plant Plectranthus amboinicus Benth (Lamiaceae). Tang Humanitas Med. 3: 16

Hamilton-Miller JM (2001). Anti-cariogenic properties of tea (Camellia sinensis). J Med Microbiol. 50: 299–302.

Hillman JD, Cransky SSS, Shivers M (1985). The relationsips between streptoccocal Species and periodontopathic Bacteria IN Human Dental Plaque. Arch Oral Biol. 30: 791–795.

Huang X, Zhang K, Deng M (2017). Effect of arginine on the growth and biofilm formation of oral bacteria. Arch Oral Biol. 82: 256–262.

Hintao J, Teanpaisan R, Chongsuvivatwong V, Ratarasan C (2007). The microbiological profiles of saliva, supragingival and subgingival plaque and dental caries in adults with and without Type 2 diabetes mellitus. Oral Microbiol Immunol. 22: 175–181.

Hayati F, Okada A, Kitasako Y, Tagami J (2011). An artificial biofilm induced secondary caries model for in vitro studies. Aust Dent J. 56: 40–47.

Jannadi H, Correa W, Zhang Z, Brandenburg K (2019). Antimicrobial peptides Pep19-2.5 and Pep19-4LF inhibit Streptococcus mutans growth and biofilm formation. Microb Pathog. 133.

Jang HD, Chang KS, Chang TC, Hsu CL (2010). Antioxidant potentials of buntan pumelo (Citrus grandis Osbeck) and its ethanolic and acetified fermentation products. Food Chem. 118: 554–558.

Jalasvuori H, Haukioja A, Tenovuo J (2012). Probiotic Lactobacillus reuteri strains ATCC PTA 5289 and ATCC 55730 differ in their cariogenic properties in vitro. Arch. Oral Biol. 57: 1633–1638.

KunduSen S, Gupta M (2011). Antihyperglycemic effect and antioxidant property of Citrus maxima leaf in Streptozotocin-induced diabetic rats. Diabetologia Croatica. 40-4:113-120.

Khan UA, Rahman H, Qasim M, Hussain A (2015). Alkanna tinctoria leaves extracts: a prospective remedy against multidrug resistant human pathogenic bacteria. BMC Complement Altern Med. 15: 127

Kanes K, Tisserat B, Berhow M, Vandercook C (1993). Phenolic composition of various tissues of Rutaceae species. Phytochemistry. 1993: 967-974

Kakiuchi N, Hattori M, Nishizawa M (1986). Studies on dental caries prevention by traditional medicines. Inhibitory effect of various tannins on glucan synthesis by glycosyltransferase from Streptococcus mutans. Chem Pharm Bull. 34: 720-725.

Karimi E, Oskoueian E, Hendra R, Oskoueian A (2012). Phenolic compounds characterization and biological activities of Citrus aurantium bloom. Molecules 17: 1203–1218.

Lim HK, Yoo ES, Moon JY, Jeon YJ (2006). Antioxidant activity of extracts from Dangyuja (Citrus grandis Osbeck) fruits produced in Jeju Island. Food Sci Biotech. 15: 312–316.

Li B, Smith B, Hossain MM (2006). Extraction of phenolics from citrus peels: II. Enzyme-assisted extraction method. Sep Purif Technol. 48:189–196.

M. Puri A, Kaur WH, Schwarz S, Singh JF (2011). "Molecular characterization and enzymatic hydrolysis of naringin extracted from kinnow peel waste," Int J Biol Macromol. 48: 58–62.

Menezes SM, Cordeiro LN, Viana GS (2006). Punica granatum (pomegranate) extract is active against dental plaque. J Herb Med. 6: 79-92.

Naderi NJ, Niakan M, Kharazi Fard MJ, Zardi S (2011). Antibacterial activity of Iranian green and black tea on streptococcus mutans: An *in vitro* study. J Dent (Tehran). 8: 55–9.

Nishijima T, Teruya K, Yanase M, Tamori Y (2012). Infectious endocarditis caused by Lactobacillus acidophilus in a patient with mistreated dental caries. Internal Medicine. 51(12):1619–1621.

Ozkan M (2002). Degradation of anthocyanins in sour cherry and pomegranate juices by hydrogen peroxide in the presence of added ascorbic acid. Food Chem. 78: 499–504.

Owoseni AA, Ajayi A (2010). Antimicrobial properties of ethanolic and aqueous extracts of Cymbopogon citratus on selected bacteria and fungi. J Med Appl Biosci. 2: 64–73.

Park SJ, Han KH, Park JY, Choi SJ, Lee KH (2014). Influence of bacterial presence on biofilm formation of Candida albicans. Yonsei Medical Journal. 55: 449–458.

Philip N, Leishman S, Walsh L (2019). Potential role for natural products in dental caries control. Oral Health Prev Dent.17: 479–485.

Perez C, Anesini C (1994). In vitro antibacterial activity of Argentine folk medicinal plants against Salmonella typhi. J Ethnopharmacol. 44:41-46.

Petersen PE, Bourgeois D, Ogawa H, Estupinan-Day S (2005). The global burden of oral diseases and risks to oral health. Bulletin of the World Health Organization. 83: 661–669.

Rhodes DV, Crump KE, Makhlynets O (2014). Genetic characterization and role in virulence of the ribonucleotide reductases of Streptococcus sanguinis. J Biol Chem. 289: 6273–6287

Ruiz-Linares M, Ferrer-Luque CM, Arias-Moliz T, de Castro P (2014). Antimicrobial activity of alexidine, chlorhexidine and cetrimide against Streptococcus mutans biofilm. Ann Clin Microbiol Antimicrob. 13: 41.

Singh YN (1986). Traditional medicine in Fiji: Some herbal folk cures used by Fiji Indians. J Ethnopharmacol. 15: 57-88.

Siang ST (1983). Use of combined traditional Chinese and Western medicine in the management of burns. Panminerva Med. 25: 197-202.

Sudto K, Pornpakakul S, Wanichwecharungruang S (2009). An efficient method for the large scale isolation of naringin from pomelo (Citrus grandis) peel. International journal of food science & technology. 44: 1737-1742.

Shakthi Deve A, Sathish Kumar T, Kumaresan K, Rapheal VS (2014). Extraction process optimization of polyphenols from Indian Citrus sinensis - as novel antiglycative agents in the management of diabetes mellitus. J Diabetes Metab Disord.13: 11

Saraya S, Kanta J, Sarisuta N, Temsiririrkkul R (2008). Development of guava extract chewable tablets for anticariogenic activity against Streptococcus mutans. Mah Univer Jou of Pharma Sci. 35: 18–23.

Singh VP, Sharma SK, and Khare VS (1980). Medicinal plants from Ujjain District Madhya Pradesh, Part II. Indian Drgs Pharm Ind. 5: 7-12.

Vasconcelos LC, Sampaio FC, Sampaio MC, Pereira Mdo S (2006). Minimum inhibitory concentration of adherence of Punica granatum Linn (pomegranate) gel against S. mutans, S. mitis and C. albicans. Braz Dent J. 17: 223-227.

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Weng Y, Guo X, Chong VJ, Howard L (2011). Synthesis and evaluation of a novel antibacterial dental resin composite with quaternary ammonium salts. J Biomed Eng. 4: 147–157.

Xiao J, Huang X, Alkhers N (2018). Candida albicans and early childhood caries: a systematic review and meta-analysis. Caries Res. 52:102–112.

Xu G, Liu D, Chen J, Ye X (2008). Juice components and antioxidant capacity of citrus varieties cultivated in China. Food Chem.106: 545–551.

Yamada T, Hayasaka S, Shibata Y, Ojima T (2011). Frequency of citrus fruit intake is associated with the incidence of cardiovascular disease: The Jichi Medical School cohort study. J Epidemiol. 21: 169–175.

Zhicen L (1987). Colour Atlas of Chinese Traditional Drugs. Vol II. Science Press, Beijing, People's Republic of China. 75-76.

Zarina Z, Tan SY (2013). Determination of flavonoids in Citrus grandis (Pomelo) peels and their inhibition activity on lipid peroxidation in fish tissue. International Food Research Journal. 20: 313.