

Antibacterial Activity of Herbal Extracts against Oral Bacteria: An Invitro Study

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Abstract:

Background: Medicinal plants have received a lot of interest for producing agents that could be considered as treatment options against oral bacteria. Dental caries has been strongly associated with *Streptococcus mutans*. Literature suggests that *Enterococcus faecalis* is associated with chronic periodontitis and failed root canal treatments.

Wheat grass, Kokum, Sumac berry and Jambu seed are some of the natural products that have received renewed interest for their antimicrobial effect. The present study was conducted to investigate the effect of Wheat grass, Kokum, Sumac berry and Jambuseed on *Streptococcus Mutans* & *E. Faecalis*. The present study aimed to evaluate and compare the antimicrobial efficacy of four herbal extracts against *E. faecalis* bacteria and *streptococcus mutans*.

Materials and Methods: 25 µl of *E. Faecalis* culture and 25 µl of *Streptococcus mutans* culture were added to the BHI agar and poured into sterile petri plates. Ethanolic extracts of Wheat grass, Kokum, Sumac berry and Jambu seed was obtained and inoculated into the wells of petri plates. 0.2% chlorhexidine was also inoculated into the well which served as control. These plates were incubated aerobically at 37 °C for 24 hours. The zone of inhibition of growth around the well was observed and measured using Vernier caliper.

Results: The zone of inhibition was observed using agar diffusion method. The present study revealed that the herbal plant *Garcinia Indica* (kokum) had higher zone of inhibition when compared to the other products tested.

Conclusion: According to results obtained from the present study the antimicrobial efficacy can be summarized as follows: kokum extract exhibited highest antimicrobial efficacy followed by Jambu seed extract and Sumac Berry extract.

Key Word: Antimicrobial activity, Chlorhexidine, Herbal Extract, *E. faecalis* bacteria and *streptococcus mutans*.

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I. Introduction

Plants provide a natural blueprint for the development of new drugs and hence they have made considerable contributions to human health and well-being.²⁴ In recent years research has focused on herbal medicines due to their wide range of biological and medicinal properties owing to the presence of natural phytochemicals. This could offer an effective alternative to synthetic preparations with higher safety margins and lower costs. The increasing interest in promoting health in natural and simple way has intensified the research in the field of indigenous products to boost oral health.

When compared to synthetically produced antimicrobial agents, natural herbs have been scientifically proven to be safe and effective alternative for bleeding gums, halitosis, mouth ulcers, and preventing tooth decay without side effects.²⁶ Hence it is warranted that further studies need to be undertaken with herbal products and compared with the gold standard in order to show the effectiveness and hence prove its merit.²⁶

Some of the natural products that are receiving renewed interest in research are Wheat grass, Kokum, Sumac berry and Jambu seed.

Wheat grass refers to the young grass of the common wheat plant, *Triticum aestivum* which is widely used as a health food supplement. Wheat grass juice contains nutrients and Vitamins A, C, E and B complex, including B12 and also has high content of bioflavonoids. Wheatgrass has shown to possess good anti-inflammatory, antioxidant, anti-carcinogenic, antibacterial and anti-aging properties.²⁷

Garcinia indica commonly called as 'Kokum', found in Konkan region of Maharashtra, Goa, Karnataka and Kerala has a myriad of health benefits.^{4,5} Kokum fruit has useful antioxidant, anti-cancer, anti-fungal, anti-inflammatory, antibacterial, cardio protective and anti-ulcer properties. It is a traditional home remedy in case of flatulence, heat strokes and infections.⁵ Kokum improves digestion and appetite. It helps to fight cancer, paralysis and bad cholesterol. It is known to improve skin health. Kokum butter is used as skin moisturizer⁶

Sumac is cultivated in a wide area from the Canary Islands to the Mediterranean coast, Iran and Afghanistan. In traditional medicine, this plant has been used for the treatment of anorexia, diarrhea, hemorrhage, and hyperglycemia.⁶ It is used as a spice in Middle Eastern cuisine to add a tart, lemony taste to salads or meat.

Jambul (*Syzygiumcumini*) have several biological activities including anti diabetic, anti-inflammatory, gastro protective and antibacterial activity.⁸

Since the above mentioned natural products have shown to possess beneficial effects on overall general health, along these lines it was proposed to investigate the antimicrobial effect of Wheat grass, Kokum, Sumac berry and Jambu seed against oral pathogens.

There has been growing interest in the use of natural products. Mouth rinses can be a promising option to help in averting plaque arrangement and dental caries. Dental caries is the most prevalent multifactorial infectious disease. The main causative organism in plaque formation and dental caries is *Streptococcus mutans*. *Enterococcus* species, in particular *enterococcus faecalis*, have been found to be associated with chronic periodontitis and failed root canal treatments involving chronic apical periodontitis.

The therapeutic effect all over the tooth surface including interproximal areas can be delivered better with the use of mouth rinses where even toothpastes are not as effective²⁶

ChlorhexidineGluconate (CHX) a potent antiplaque agent, is regarded as the "gold standard" antimicrobial agent active against many oral pathogens. However prominent side effects like taste alteration, tooth discoloration, mucosal discoloration, supragingival calculus formation, oral ulcerations, unilateral, or bilateral parotid swelling desquamation of oral mucosa and also restricted usage in pediatric patients cannot be ignored.^{25,26,29,30} Hence, there is a need to find alternative to chlorhexidine.

The aim of this study was to evaluate the antibacterial activity of herbal extracts of wheatgrass, kokum, sumac berry and jambu seed against *streptococcus mutans* and *E.faecalis* bacteria.

Hence, the study attempts to explore the possibility of a cost effective herbal oral hygiene product for use in dentistry.

II. Material And Methods

Ethanol extraction of the herbal product.

30 g of the commercially available herbal product was added to 100 ml of 95% ethanol and brought to a paste in a mortar and pestle and ground for 20 minutes, it was then filtered using Whatman filter paper No.2. The filtrate was transferred to a sterile petri plate and alcohol was allowed to evaporate.²⁶

The extracted powder was weighed, i.e. 20mg and dissolved in 1ml of sterile distilled water, and thus 2% concentration of the herbal extract was obtained. This ethanol extract of the herbal product was stored at +4 °C till use.

Preparation of fresh culture of Enterococcus Faecalis

E.Faecalis ATCC29212 was inoculated into 5 ml of BHI broth and incubated at 37⁰ C for 4 hrs. The turbidity of the culture was adjusted to McFarland standard 0.5 (approx.=1.5x10⁸cfu/ml)

BHI agar was freshly prepared in 30 ml quantity and sterilized in autoclave. After sterilization, the agar was cooled to 50⁰C, 25 µl of the adjusted *E.Faecalis* culture was added to the medium, mixed well and poured into sterile petri plates. The agar was allowed to set and wells of 6mm diameter made in the medium using sterile templates.

50 µl of the test materials were inoculated into separate wells i.e. wheatgrass extract, kokum extract, jambu seed extract and sumac berry extract. 0.2% chlorhexidine was also inoculated into the well which served as control.

The plates were incubated aerobically at 37⁰ C for 24 hrs. The zone of inhibition of growth around the well was observed and measured using Vernier caliper.

Preparation of fresh culture of streptococcus mutans

A clinical strain of *Streptococcus mutans* inoculated into 5 ml of BHI broth and incubated at 37⁰C for 4 hrs. The turbidity of the culture was adjusted to McFarland standard 0.5

BHI agar was freshly prepared in 30 ml quantity and sterilized.

After sterilization, the agar was cooled to 50⁰C, 25 µl of the adjusted *Streptococcus mutans* culture was added to the medium, mixed well and poured into sterile petri plates.

The agar was allowed to set and wells of 6mm made in the medium using sterile templates. 50 µl of the test materials were inoculated into separate wells i.e. wheatgrass extract, kokum extract, jambu seed extract and sumac berry extract. The herbal extracts were compared with the 0.2% chlorhexidine inoculated into the well which served as control.

The plates were incubated aerobically at 37°C for 24 hrs. The zone of inhibition of growth around the well was observed and measured using Vernier caliper.

III. Result

Antibacterial Agent/Preparation	Zone of Inhibition against <i>Enterococcus faecalis</i> (Mean ± SD in mm)	Zone of Inhibition against <i>Enterococcus faecalis</i> (Mean rank)	Kruskal Wallis chi square & p value	Zone of Inhibition against <i>Streptococcus mutans</i> (Mean ± SD in mm)	Zone of Inhibition against <i>Streptococcus mutans</i> (Mean rank)	Kruskal Wallis chi square & p value
Chlorhexidine	15.33 ± 1.52	5.00	11.51 & 0.021*	16.00 ± 1.00	5.00	12.83 & 0.012*
Kokum extract	28.33 ± 1.00	12.50		26.33 ± 1.52	13.33	
Jambu Seed extract	27.00 ± 1.73	10.83		23.33 ± 1.15	8.33	
Sumac berry extract	26.00 ± 1.00	9.67		25.00 ± 1.00	11.33	
Wheatgrass Extract	0.90 ± 1.00	2.00		0.83 ± 1.52	2.00	

SD=standard deviation, mm=millimeter

Concentration of Chlorhexidine: 0.2%. Concentrations of all the alcoholic herbal extracts was 2% * p value < 0.05 considered as statistically significant.

Kokum extract showed maximum zone of inhibition against S.mutans 26.33 ± 1.52 mm followed by sumac berry extract 25.00 ± 1.00 mm, Jambu seed extract 23.33 ± 1.15 mm, chlorhexidine 16.00 ± 1.00 mm and Wheatgrass Extract 0.83 ± 1.52 mm respectively. The results obtained were statistically significant. (p value 0.012)

Similar results were observed against E.Faecalis wherein, kokum extract showed maximum zone of inhibition of 28.33 ± 1.00 mm followed by Jambu Seed extract 27.00 ± 1.73 mm, Sumac berry extract 26.00 ± 1.00 mm, Chlorhexidine 15.33 ± 1.52 mm and Wheatgrass Extract 0.90 ± 1.00 mm respectively. The results obtained were statistically significant. (p value 0.021)



FIGURE A.1: Zone of inhibition of the test products against E.faecalis

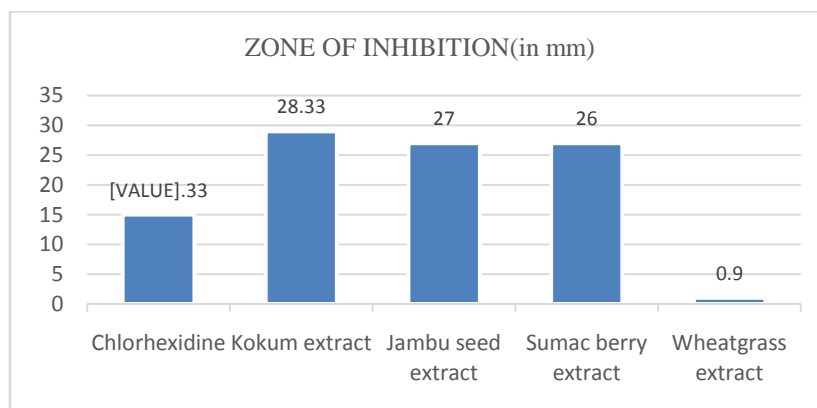


CHART A.1: ZONE OF INHIBITION (mm) *Enterococcus faecalis*

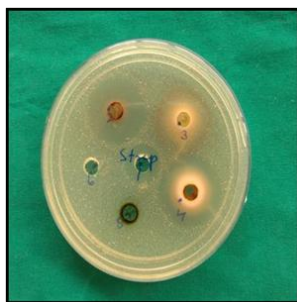


FIGURE A.2:Zone of inhibition of the test products against S.mutans

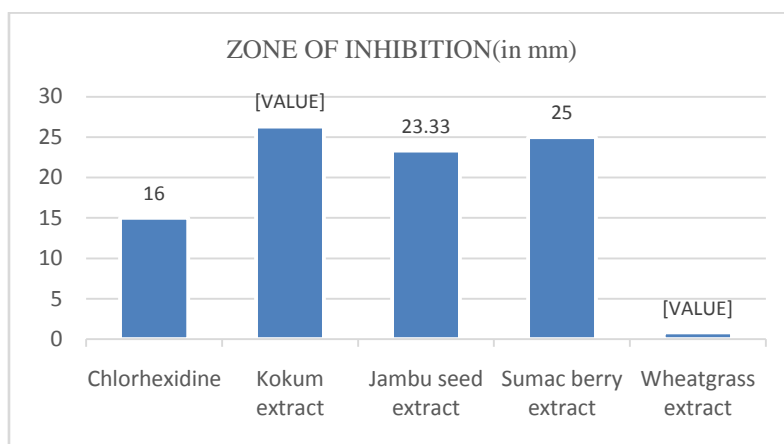


CHART A.2:ZONE OF INHIBITION (mm) streptococcus mutans

TABLE A. 2: COMPARISON OF ANTIBACTERIAL ACTIVITY OF TEST PRODUCTS AGAINST Enterococcus faecalis

Bacteria	TEST PRODUCT (a)	Comparator Test Product (b) Mean ±SD	Mean Difference (a-b ± SE)	P value (Mann –Whitney U test)
E. Faecalis	(15.33 ± 1.52) chlorhexidine	(28.33 ± 2.08) Kokum extract	-13.00 ± 1.19	0.05*
		(27.00 ± 1.00) Jambu seed extract	-11.67 ± 1.19	0.05*
		(26.00 ± 1.73) Sumac berry extract	-10.67 ± 1.19	0.04*
		(0.90 ± 0.10) Wheatgrass extract	14.43 ± 1.19	0.05*
	(28.33 ± 2.08) Kokum extract	(27.00 ± 1.00) Jambu seed extract	1.33 ± 1.19	0.37
		(26.00 ± 1.73) Sumac berry extract	2.33 ± 1.19	0.27
		(0.90 ± 0.10) Wheatgrass extract	27.43 ± 1.19	0.05*
	(27.00 ± 1.00) Jambu Seed extract	(26.00 ± 1.73) Sumac berry extract	1.00 ± 1.19	0.48
		(0.90 ± 0.10) Wheatgrass extract	26.10 ± 1.19	0.05*
	(26.00 ± 1.73) Sumac berry extract	(0.90 ± 0.10) Wheatgrass Extract	25.10 ± 1.19	0.04*

SD=standard deviation, mm=millimetre

Following the Kruskal Wallis test as reported in table A.1. A post hoc analysis indicating the mean differences in zones of inhibition between 2 different intervention groups was performed using Mann-Whitney U test. The following observations were ascertained as per the table A.2. Mean Zone of inhibition (mm) was

observed for 5 different test product against *Enterococcus feacalis*. The mean zone of inhibition for group 1 i.e chlorhexidine was 15.33 ± 1.52 mm. This was compared with mean zone of inhibitions of kokum extract (28.33 ± 2.08 mm), Jambu seed extract (27.00 ± 1.00 mm), Sumac berry extract (26.00 ± 1.73 mm) and wheatgrass extract (0.90 ± 0.10 mm) respectively. The mean differences among all these comparisons were found to be statistically significant (p value 0.05).

The mean zone of inhibition for group 2 i.e kokum extract was 28.33 ± 2.08 mm. This was compared with mean zone of inhibitions of Jambu seed extract (27.00 ± 1.00 mm) and Sumac berry extract (26.00 ± 1.73 mm) respectively and the mean differences among these comparisons were found not to be statistically significant. The mean zone of inhibition for group 2 i.e kokum extract (28.33 ± 2.08 mm) when compared with wheatgrass extract (0.90 ± 0.10 mm) was found to be statistically significant (p value 0.05).

The mean zone of inhibition for group 3 i.e Jambu Seed extract was 27.00 ± 1.00 mm. This mean difference when compared with mean zone of inhibitions of Sumac berry extract (26.00 ± 1.73 mm) was found not to be statistically significant. (p value 0.48) whereas when compared with mean zone of inhibitions of wheatgrass extract (0.90 ± 0.10 mm) it was found to be statistically significant (p value 0.05).

The mean zone of inhibition for group 4 i.e Sumac berry extract (26.00 ± 1.73 mm) when compared with mean zone of inhibition of wheatgrass extract (0.90 ± 0.10 mm) it was found to be statistically significant (p value 0.04).

TABLE A.3: COMPARISON OF ANTIBACTERIAL ACTIVITY OF TEST PRODUCTS AGAINST *Streptococcus mutans*

Bacteria	TEST PRODUCT (a)	Comparator Test Product (b) Mean \pm SD	Mean Difference (a-b \pm SE)	P value (Mann –Whitney U test)
S. Mutans	(16.00 \pm 1.00) chlorhexidine	(26.33 \pm 1.52) Kokum extract	-10.33 \pm 0.87	0.05*
		(23.33 \pm 1.15) Jambu Seed extract	-7.33 \pm 0.87	0.04*
		(25.00 \pm 1.00) Sumac berry extract	-9.00 \pm 0.87	0.05*
		(0.83 \pm 0.15) Wheatgrass Extract	15.17 \pm 0.87	0.05*
	(26.33 \pm 1.52) Kokum extract	(23.33 \pm 1.15) Jambu Seed extract	3.00 \pm 0.87	0.10
		(25.00 \pm 1.00) Sumac berry extract	1.33 \pm 0.87	0.26
		(0.83 \pm 0.15) Wheatgrass Extract	25.50 \pm 0.87	0.05 *
	(23.33 \pm 1.15) Jambu Seed extract	(25.00 \pm 1.00) Sumac berry extract	-1.67 \pm 0.87	0.04*
		(0.83 \pm 0.15) Wheatgrass Extract	22.50 \pm 0.87	0.04*
	(25.00 \pm 1.00) Sumac berry extract	(0.83 \pm 0.15) Wheatgrass Extract	24.17 \pm 0.87	0.05*

SD=standard deviation, mm=millimeter

The following observations were ascertained as per the table A.3.

Mean Zone of inhibition (mm) was observed for 5 different test product against *Streptococcus mutans*. The mean zone of inhibition for group 1 i.e.chlorhexidine was 16.00 ± 1.00 mm. This was compared with mean zone of inhibitions of kokum extract (26.33 ± 1.52 mm), Jambu seed extract (23.33 ± 1.15 mm), Sumac berry extract (25.00 ± 1.00 mm) and wheatgrass extract (0.83 ± 0.15 mm) respectively. The mean differences among all these comparisons were found to be statistically significant (p value 0.05).

The mean zone of inhibition for group 2 i.e. kokum extract was 26.33 ± 1.52 mm. This was compared with mean zone of inhibitions of Jambu seed extract (23.33 ± 1.15 mm) and Sumac berry extract (25.00 ± 1.00 mm) respectively and the mean differences among these comparisons were found not to be statistically significant. The mean zone of inhibition for group 2 i.e. kokum extract (26.33 ± 1.52 mm) when compared with wheatgrass extract (0.83 ± 0.15 mm) was found to be statistically significant (p value 0.05).

The mean zone of inhibition for group 3 i.e.Jambu Seed extract was 23.33 ± 1.15 mm. This mean difference when compared with mean zone of inhibitions of Sumac berry extract (25.00 ± 1.00 mm) and wheatgrass extract (0.83 ± 0.15 mm) was found to be statistically significant (p value 0.04).

The mean zone of inhibition for group 4 i.e. Sumac berry extract (25.00 ± 1.00 mm) when compared with mean zone of inhibition of wheatgrass extract (0.83 ± 0.15 mm) was found to be statistically significant (p value 0.05).

IV. Discussion

There is immense potential for antimicrobial agents from plant origin. Hence the purpose of the study was to evaluate the antimicrobial efficacy of herbal extracts i.e. Wheatgrass extract, Kokum extract, Jambu seed extract and Sumac Berry extract against *E. faecalis* bacteria and streptococcus mutans. In the present study, all the extracts except wheatgrass extract inhibited the growth of Streptococcus mutans and enterococcus faecalis more than chlorhexidine.

In the present study wheatgrass extract did not show antibacterial effect against *S. mutans* and *E. faecalis* but in the study conducted by Sundaresan A et al (2015)⁷ it was found that wheat grass extracts showed antibacterial activity against *Yersinia enterocolitica*, *Staphylococcus aureus* and *Listeria monocytogenes* which are some of the foodborne pathogens.

Murali M. et al (2016)¹⁸ found that chloroform extracts of wheatgrass showed better anti-fungal activity against *E. coli* and *Aspergillus Niger*.

In the study conducted by Deshwal and Deepshikha (2018)²⁶ it was found that wheat grass (*Triticum aestivum* L.) significantly inhibited the growth of *E. coli*. Rucha Diwakar Gore et al. (2017)¹⁴ conducted a study which revealed that aqueous extract of wheatgrass has an inhibitory effect on the oral cancer cell line proliferation.

However, there is very scarce literature on the effect of wheatgrass extract against oral bacteria. The present study revealed that ethanol extraction of wheatgrass did not exhibit a significant inhibitory effect against the tested bacteria.

In the present study, there was a definite reduction in *S. mutans* and *E. Faecalis* with Sumac berry extract. The results are in synchrony with the results obtained by study done by Vahid-Dastjerdi E et al. (2014)⁶ to evaluate the effect of *Rhus coriaria* L. (sumac berry) water extract on five common oral bacteria i.e. *Streptococcus mutans*, *Streptococcus sanguinis*, *Streptococcus sobrinus*, *Streptococcus salivarius* and *Enterococcus faecalis* and bacterial biofilm formation on orthodontic wire and the study revealed that the sumac berry extract was able to inhibit bacterial biofilm formation on orthodontic wire.

In the present study, there was a definite reduction in *S. mutans* and *E. Faecalis* with jambu seed extract. However, there is very scarce literature on jambu seed extract against oral bacteria. Haque et al. (2017)⁸ found that Jambu juice had selective bactericidal effects against several enteric pathogens such as *Salmonella typhimurium*, *Shigella flexneri*, *Staphylococcus aureus*, and *ETEC* (*Enterotoxigenic E. coli*) although, no information about the activity against oral pathogens was available.

The present study revealed that kokum had higher zone of inhibition when compared to the other products tested including chlorhexidine.

The study conducted by Ranveer R C and Sahoo A K (2017)⁴ revealed that kokum has many bioactive compounds such as anthocyanin, Hydroxyl citric acid and Garcinol which has showed to possess antimicrobial activity has a role in treatment of gastric ulcers and garcinol acts as antioxidant and anti-inflammatory agent but there is insufficient literature about the activity against oral pathogens.

In the present study, kokum extract gave the maximum zone of inhibition against streptococcus mutans and enterococcus faecalis bacteria.

The results indicate that the active components present in these herbal extracts have the capability of destroying bacterial cell wall which will inevitably inhibit the growth of bacteria. Hence, this herbal extracts can be used for discovery of bioactive natural products that may serve in the development of new pharmaceutical agent.

After clinical validation of the proof of efficacy in prevention of Dental Caries, further studies on these herbal extracts can be encouraged.

In the present study, there was a definite reduction in *S. mutans* and *E. Faecalis* with kokum, Sumac berry and jambu seed extract whereas wheatgrass extract did not show antibacterial effect.

This in vitro evaluation is an attempt to encourage further studies comparing the antimicrobial effects of different herbal extracts on prevalence of oral diseases.

V. Conclusion

The herbal extracts showed significant activity against the investigated microbial strains, which is promising. The study revealed that kokum had higher zone of inhibition among all the groups compared including chlorhexidine. *Garcinia Indica* (kokum) can be further studied so that it can be considered as a cost effective herbal oral hygiene product in future research. Similar findings with *E. faecalis* bacteria can be a potential for use in endodontics. The study revealed the possibility of alternative and effective antimicrobial agents for use in dentistry as compared to chlorhexidine which is considered as the gold standard, requiring further research.

References

- [1]. Hegde RJ, Kamath S. Comparison of the Streptococcus mutans and Lactobacillus colony count changes in saliva following chlorhexidine (0.12%) mouth rinse, combination mouth rinse, and green tea extract (0.5%) mouth rinse in children. *J Indian Soc Pedod Prev Dent* 2017; 35:150-5.
- [2]. Loesche W.J, Rowan J, Straffon L.H. And Loos P.J. Association of Streptococcus mutans with Human Dental Decay. *Infection and Immunity* 1975; 11(6):1252-1260
- [3]. Wang QQ, Zhang CF, Chu CH and Zhu XF. Prevalence of Enterococcus faecalis in saliva and filled root canals of teeth associated with apical periodontitis. *International Journal of Oral Science* 2012; 4:19–23.
- [4]. Ranveer R C and Sahoo A K. Bioactive Constituents of Kokum and its Potential Health Benefits. *Nutrition and Food Toxicology*. 2017; 1(6): 236-244.
- [5]. Jagtap P, Bhise K, Prakya V. A Phytopharmacological Review on Garcinia indica. *International Journal of Herbal Medicine* 2015; 3(4): 02-07
- [6]. Vahid-Dastjerdi E et al. Effect of Rhus coriaria L. water extract on five common oral bacteria and bacterial biofilm formation on orthodontic wire. *IRAN. J. MICROBIOL.* 2014; 6:269-275.
- [7]. Sundaresan A, Selvi A and Manonmani H.K. The Anti-Microbial Properties of Triticum aestivum (Wheat Grass) Extract. *International Journal of Biotechnology for Wellness Industries*. 2015; 4: 84-91
- [8]. Haque et al. Antimicrobial Activity of Jambul (Syzygium cumini) Fruit Extract on Enteric Pathogenic Bacteria. *Advances in Microbiology*, 2017; 7:195-204.
- [9]. Prusti, A, Mishra S.R., Sahoo S and Mishra, S.K. Antibacterial Activity of Some Indian Medicinal Plants. *Ethno botanical Leaflets*. 2008; 12: 227-230.
- [10]. Veiga N et al. Dental Caries: A Review. *Journal of Dental and Oral Health*. 2016; 2(5):1-3.
- [11]. Haerian-ardakani A et al. Comparison of Antimicrobial Effects of Three Different Mouthwashes. *Iran J Public Health*, 2015; 44(7):997-1003.
- [12]. Swami B S, Thakor N.J and Patil S.C. Kokum (Garcinia indica) and its Many Functional Components as Related to the Human Health: A Review. *Journal of Food Research and Technology*. 2014; 2(4):130-142
- [13]. Sutar, R. L., Mane S. P. and Ghosh, J.S. Antimicrobial activity of extracts of dried kokum (Garcinia indica C) *International Food Research Journal*. 2012; 19 (3): 1207-1210
- [14]. Rucha Diwakar Gore et al. Wheatgrass: Green Blood can help to Fight Cancer. *Journal of Clinical and Diagnostic Research*. 2017 Jun, Vol-11(6): ZC40-ZC42
- [15]. Mishra A, Bapat M, Tilak J.C and Thomas P. A. Devasagayam T.P.A. Antioxidant activity of Garcinia indica (kokum) and its syrup. *current science*. 2006; 91(1):90-93
- [16]. K. Nagendra, R. Kusum and H.D. Ramachandran. Anti-Microbial, Antifungal and Anti-Oxidant Activity of Ethanolic Extracts of Garcinia indica Fruit. *Int. J. Curr. Microbiol. App. Sci.* 2014; 3(9): 800-804
- [17]. Elumalai A and Eswaraiah M.C. A pharmacological review on Garcinia indica choisy. *International Journal of Universal Pharmacy and Life Sciences*. 2011; 1(3):57-60
- [18]. Murali M, Nair A S, Kumar N S. In Vitro Anti-microbial Activities of Wheat Grass. *J. Pharm. Sci. Innov.* 2016; 5(6):201-204.
- [19]. Abdullah T, Albeshari K, Ahmed IA, Baharuddin A. Effect of extraction methods on the antibacterial activity of spices (garlic, cinnamon, turmeric, clove) against Escherichia coli K12 (JM109) *Asian J. Biol. Life Sci.* 2017; 6(2):313-320.
- [20]. Goyal P and Kaushik P. In vitro Evaluation of Antibacterial Activity of Various Crude Leaf Extracts of Indian Sacred Plant, Ocimum sanctum L. *British Microbiology Research Journal*. 2011; 1(3): 70-78
- [21]. Chakravarty A.K, Chatterjee S.N, Yasmin H and Mazumder T. Comparison of efficacy of turmeric and commercial curcumin in immunological functions and gene regulation. *International Journal of Pharmacology*. 2009; 5(6):333-345.
- [22]. Nayak C.A, Rastogi N.K & Raghava Rao K.S. Bioactive Constituents Present in Garcinia indica Choisy and its Potential Food Applications: A Review. *International Journal of Food Properties*. 2010; 13: 441–453
- [23]. Saadat N and Gupta S.V. Potential Role of Garcinol as an Anticancer Agent. *Journal of Oncology*. 2012; 1-8
- [24]. S. S. Kadam, S. M. Mule, P. V. Jadhav, S. V. Surve, V. S. Dandekar and S. C. Warawadekar. Studies on Microbial Profile of Probiotic Low Fat Kokum Lassi by Using Honey as Sweetener. *Int. J. Curr. Microbiol. App. Sci.* 2018; 7: 4767-4777

- [25]. Goyal P and Kaushik P. In vitro Evaluation of Antibacterial Activity of Various Crude Leaf Extracts of Indian Sacred Plant, *Ocimum sanctum* L. *British Microbiology Research Journal* 2011; 1(3): 70-78.
- [26]. Deshwal VK and Deepshikha. Antimicrobial Investigation of Wheat Grass (*Triticum Aestivum* L.) Against *Escherichia Coli*. *European Journal of Molecular Biology and Biochemistry*. 2018; 5(1): 9-12.
- [27]. Arul Selvan K, Madhu P, Prabhu T. A study on the phytochemistry and anti-bacterial effect of ethanol extract of Indian propolis on clinical strains of mutans streptococci isolated from patients with dental caries. *International J of Microb Environ Managemt* 2010 vol I 20-36

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