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Research Article

Formulation, Evaluation and Antibacterial Properties of Novel Polyherbal Toothpaste for Oral Care

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ABSTRACT

The demand for herbal based product such as toothpaste is high these days. Consumers believed by using herbal based toothpaste are safe, effective and less toxicity because less and only safe chemical used as compared to the synthetically produced toothpaste. Therefore, this study was aimed to formulate and evaluate new polyherbal toothpaste which containing herbal extracts that available in Malaysia to treat periodontal problem. The polyherbal toothpaste was formulated using three herbal extracts namely banana peel (*Musa acuminate)*, guava leaves (*Psidium guajava)* and orange peel (*Citrus arranticum)* and tested against *Staphylococcus aureus, Bacillus cereus, Escherichia coli* and *Pseudomonas aeruginosa* with different concentrations varying from 100 mg/mL, 250 mg/mL and 500 mg/mL. The significant inhibition has seen against *Staphylococcus aureus* (12-18 mm) followed by *Bacillus cereus* (11-15 mm) and there is no inhibition for both *Escherichia coli* and *Pseudomonas aeruginosa*. The formulated toothpaste was also evaluated with the standard physiochemical parameters along with the antimicrobial activity. The formulated toothpaste showed potent inhibition against gram positive bacteria but not against gram negative bacteria. Thereby, it opens a window for future study to enhance the ability of the toothpaste and to prove the efficacy and safety of the formulated toothpaste.

Keywords: Polyherbal toothpaste, antibacterial activity, *Musa acuminate, Psidium guajava Citrus arranticum*

INTRODUCTION

Toothpaste is a gel or paste formulation product and is used to clean and maintain oral hygiene with the aid of toothbrush. It is a common product used by the community for dental care. Although it is recommended by most dentist to brush the teeth twice daily and it is highly effective for plaque removal but it is not possible for bacterial infection. By removing the plaque it will helps in decrease the chances of periodontal inflammation which mainly caused by bacteria or oral flora. To overcome this problem, it is recommended for the patient to use toothpaste with better antibacterial activity[1]. Most of the toothpaste available in the market nowadays contains two types of ingredient which are the active and non-active toothpaste ingredient or the excipient. One of the active toothpaste ingredients is abrasive which helps in removing the plaque. It constitutes at least 50% from the total preparation of the toothpaste. It really helps in minimized periodontal disease. The commonly used abrasives are sodium bicarbonate, calcium carbonate and aluminium hydroxide. Whitening agents helps to remove stain on the teeth but the effect is temporary. The commonly used whitening agents for toothpastes in the market are peroxide and bleach. Fluoride and its derivatives are used to strengthen the enamel and prevent cavities. The most common used fluorides are sodium fluoride, sodium monofluorophosphate, olaflur and stannous fluoride. Between them, stannous fluoride shows effective controls of gingivitis[2]. The problem now is most people do not know the long term consequence of using the commercial toothpastes. This is because the marketed toothpastes contain substance which consider as unhealthy and could harm the body in the future. Recently there is some issues arise on the harmful effect of fluoride when being used for a longer period of time[3]. The bleach and peroxide used as whitening agent in the toothpaste is consider as hazardous as they may cause irritation to the mouth and skin in small amount and chemical burn in large amount. The flavouring agents used are synthetically and chemically produced and commonly derived from petrochemicals[4]. After studying the drawbacks of commercial toothpastes, people are now more inclined toward the use of herbal formulations. Herbal toothpaste does not contain the artificial colors, flavors or fluoride that many, when compare the contents of artificial products. The antimicrobial activity of *Musa acuminata* as known as banana, *Malus domestica* known as apple and *Citrus arranticum* also known as orange against *Streptococcus mutans, Staphylococcus aureus, Enterococcus faecalis* and *Porphyromonas gingivalis* have been proven recently[5]. *Psidium guajava* or known as guava and its leaf exhibits antimicrobial activity against dental pathogens[6]. This study is selected to overcome the problems associated with the usage of unhealthy ingredient used in commercial toothpastes which widely used around the world. In addition, to reduce the cases of periodontal cases which mainly caused by dental pathogen. Hence, in

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Table 1: Composition of polyherbal toothpaste.

Components	Amount (g)
Guava leaves extract	5
Banana peel extract	5
Orange peel extract	5
Hydroxypropylmethylcellulose (HPMC)	3
Sodium lauryl sulphate	10
Calcium carbonate	20
Glycerine	5
Methyl paraben	0.5
Propyl paraben	0.25
Peppermint oil	2-3 drops
Demineralised water	80 mL

Table 2: Percentage yield of methanolic extract of *Musa acuminate*, *Psidium guajava* and *Citrus arranticum.*

Name of the plant	Quantity of plant powder	Methanol	Yield
Musa acuminate	100 g	500 mL	24.36%
Psidium guajava	100 g	500 mL	23.7%
Citrus arranticum	100 g	500 mL	32.6%

Table 3: Evaluation of formulated polyherbal toothpaste.

Evaluation parameter	Result
Colour	Mud green
Taste	Bitter
Odour	Pleasant (mint)
Texture	Smooth
Moisture content	24.57%
Foaming character	27 mL
Storage stability	After 2 months, separation of liquid and solid phase can be seen clearly
pH	5.7

the present study we are interested to formulate and evaluate new polyherbal toothpaste and to study its antimicrobial potency.

MATERIALS AND METHODS

Plant collection and indentification

The fresh banana fruits (*Musa acuminate)* and orange fruits (*Citrus arranticum)* were purchased from local market, Ipoh and identified. The fresh guava fruits leaves (*Psidium guajava)* were collected in Ipoh and Identified.

Extraction

The peels of banana and orange fruits were removed off using a peeler and shade dried at room temperature for a week. The dried peels and guava fruit leaves were coarsely powdered and then defatted with petroleum ether. The defatted materials were separately subjected to extraction by cold maceration separately by agitation using 70% methanol for 7 days. The marc was squeezed out and filtered off. The combined filtrates were concentrated separately by allowing it to be evaporated from the petri dish. The concentrated extracts were stored in air-tight containers in a refrigerator at 4 °C until further use.

Formulation of polyherbal toothpaste

The extracts were used to formulate toothpaste. The composition of the polyherbal toothpaste were stated in Table 1. Hydroxypropylmethylcellulose (HPMC) was triturated together with methyl paraben and propyl paraben. Glycerin was added and mixed well by triturating them all together. Calcium carbonate was added and mixed well. Then, all the extracts were added and again triturate to mix the ingredients. Water was added until the desired consistency achieved.

Evaluation of the formulated toothpaste

Organoleptic evaluation

Organoleptic evaluation (colour, taste, odour) were done by sensory and visual inspection.

Moisture content

10 g of toothpaste was weighted and dried it in the oven at 105 ^{o}C then it was cooled. The loss of weight was recorded as percentage moisture content and calculated by the given formula.

$$\% \text{ moisture} = \frac{\text{Original sample weight} - \text{dry sample weight}}{\text{Original sample weight}} \times 100$$

Foaming character

For the evaluation of foaming character 5 g of sample was soaked with 10 mL of distilled water in 100 mL of beaker and closed with parafilm for 30 min to ensure the toothpaste disperse in the water. After 30 min, the mixture was stirred and transfer the slurry into a 250 mL measuring cylinder a make sure there is no foam formed. Rinsed the excess slurry in the beaker with 5-6 mL of distilled water and then again transferred into the measuring cylinder slowly. To the measuring cylinder, the volume was marked upto 50 mL volume. The measuring cylinder was heated in water bath and maintain the temperature at 30 °C. The slurry was stirred to make a uniform suspension. Then, the measuring cylinder was closed with parafilm and the measuring cylinder was shaken with 12 complete cycles and allowed it to stand for 5 min. After 5 min, the volume of foam with water (V_1) and volume of water only (V_2) were measured.

Foaming power = $V_1 - V_2$

V_1 - volume in mL of foam with water

V_2 - volume in mL of water only

Determination of pH

5 g of sample was diluted with 45 mL of distilled water in a 150 mL beaker. This was stirred well to make a thorough suspension. The pH of the sample with pH meter within 5 min was measured.

Storage stability

For the evaluation of the storage stability, the toothpaste was filled in a toothpaste tube for storage and stored for 45 days at each of 5 ^{o}C, room temperature and 40 ^{o}C. The tube was then cut through and checked whether the liquid component was separated from the toothpaste.

Table 4: Zone of inhibition for antimicrobial activity testing.

	Gram Positive		Gram Negative	
Concentration (mg/mL)	*Staphylococcus aureus*	*Bacillus cereus*	*Escherichia coli*	*Pseudomonas aeruginosa*
	Zone of inhibition (mm)			
100	12	11	-	-
250	15	13	-	-
500	18	15	-	-
Positive control (Ciprofloxacin 5 µg)	31	26	32	29

Antimicrobial screening

Test microorganisms

Panels of common pathogenic microorganisms were used in the study, which includes two gram-positive bacteria (*Staphylococcus aureus and Bacillus cereus*) and two gram-negative bacteria (*Escherichia coli* and *Pseudomonas aeruginosa*).

Disc-diffusion method

The antibacterial activity of the toothpaste was evaluated using disc-diffusion method. A suspension of the tested microorganisms were uniformly swabbed on agar plates using sterile cotton swabs. Sterile blank discs were individually impregnated to the different concentrations of formulated paste (100, 250 and 500 mg/ml) were placed onto the inoculated agar plates[8]. The plates was inverted and incubated at 37 ºC for 24 h. The antibacterial activity was measured by measuring diameter of the resulting zone of inhibition against the tested organisms. The test for positive control and negative control was performed in duplicate.

RESULTS AND DISCUSSION

The percentage yield of methanolic extract of *Musa acuminate*, *Psidium guajava* and *Citrus arranticum* were shown in Table 2.

Evaluation of the formulated toothpaste

The formulated toothpaste was evaluated with the standard evaluation parameters for toothpaste. The results of the evaluation test were shown in Table 3 below:

Antimicrobial activity

The main objective of this study is to formulate and evaluate the antimicrobial activity of the prepared herbal toothpaste. Various extracts have been used in formulating the toothpaste such as orange peel extract, banana peel extract and guava leaves extract which are believed to have antibacterial properties. The toothpaste was evaluated for antimicrobial activity as given in the table above. The positive result was shown for Gram-positive bacteria, *Bacillus cereus* and *Staphylococcus aureus* at all level of dilution from 100 mg/mL up to 500 mg/mL and the zone of inhibition increased with increasing of concentration, whereas there is no zone of inhibition for Gram-negative bacteria, *Pseudomonas aeruginosa and Escherichia coli*. The most significant result can be seen against *Staphylococcus aureus* (12-18 mm) followed by *Bacillus cereus* (11-15 mm) and there is no inhibition against *Escherichia coli* and *Pseudomonas aeruginosa*. The zone of inhibition for antimicrobial activity testing have been shown in Table 4. A study has been carried out on antimicrobial property of orange peel extract. The result shown for methanolic extract was positive against *S. aureus* and *E. coli*[8]. For banana peel extract, the antimicrobial activity has been established. The methanolic extract shows positive result against *S. aureus, Bacillus sp, P. aeruginosa* and *E. coli* and the zone of inhibition increased with increasing of the extracts concentration[9]. The same goes to methanolic extract of guava leaves where the antimicrobial property has been known against *S. aureus* and *E.coli.* It is said to have antimicrobial activity due to flavonoid present in the leaves[10]. From the studies as mentioned above, the results for Gram-positive bacteria has been well correlated with our present study results. The activity may be due to the presence of various phytoconstituents.

CONCLUSION

Herbal-based products have growing demand in the market. People believed using product made up of natural sources ingredient are safer with fewer side effects than the synthetic one. It is a good attempt to establish such herbal toothpaste containing orange peel extract, banana peel extract and guava leaves extract which helps in reducing bacterial growth in the mouth. Further study are warranted to improve the stability of the formulation, prove the efficacy and safety of the formulated toothpaste and to improve the antimicrobial activity of the toothpaste.

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CONFLICT OF INTEREST

All the authors declared that there is no conflict of interest.

REFERENCES

1. Abdul Wahad I. A. K (2011). Comparison between the efficacy of herbal and conventional dentrifices on established gingivitis. Dent Res J, 8: 57-63.
2. Strassler, H. E (2013). Toothpaste ingredients make a difference. Benco Dental, 1: 101-107.
3. McCaffery K (2003). Fluoride and dermatitis. J American Dental Association, 134: 1166.
4. Abhay S, Dinnanth B. M, Hullatti K. K (2014). Formulation and spectral analysis of new poly herbal toothpaste. J Drug Delivery & Therapeutics, 4: 68-74.

5. Abhay S, Dinnanth B. M (2015). Formulation and evaluation of new polyherbal toothpaste for oral care. Indian J Health Sci, 8: 24-27.
6. Deepa G, Nikhil M (2015). Phytochemical, antioxidant, and antimicrobial activity of *psidium guajava* leaves against oral dental pathogens. Indian J Applied Res, 5: 52-54.
7. Kamarezi T. S, Samah O. A, Taher M, Susanti D, Qaralleh H (2012). Antimicrobial activity and essential oils of *Curcuma aeruginosa*, *Curcuma manga* and *Zingiber cassumunar* from Malaysia. Asian Pacific J Tropical Med, 5: 202-209.
8. Mamta A, Parminder K (2013). Antimicrobial and antioxidant activity of orange pulp and peel. Int J Sci Res, 2: 412-415.
9. Ehiowemwenguan G, Emoghene A. O, Inetianbor J. E. (2014). Antibacterial and phytochemical analysis of banana fruit peel. J Pharmacy, 4: 18-25.
10. Ismail M, Minhas P. S, Fathima K, Sahana V. M, Sowmya C**.** (2012). Antibacterial activity of leaves extract of guava (*Psidium guajava*). Int J Res Pharma Biomed Sci, 3: 1-2