



PREPARATION AND EVALUATION OF HERBAL MOUTH WASH

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Abstract

The mouth is an extremely vital and sensitive part of our body because we receive all kinds of things through the mouth, so poor oral hygiene condition causes many oral and systemic diseases. Mouthwashes are a very popular additional oral hygiene element and there are plenty of individual products, whose compositions are in a state of flux. The aim of our study was to investigate the compositions of mouthwashes and their functions, as well as to discuss their effectiveness in preventing and curing oral diseases and side effects. Medicinal plants play a predominant role in curing and preventing disease due to their antibacterial and antimicrobial activity against Human microorganism. The aim of present work is to formulate and evaluate antibacterial mouthwash and evaluate its effectiveness against microbes present in oral cavity. They act on mouth pathogens, microbes and reduces the pain and also has no more side effects. The various herbs and their extracts such as Tulsi, Green tea, Nagarmotha were selected for mouthwash and prepared formulation was further screened for antimicrobial activity against culture and further. Evaluated for physical properties. The presence mouthwash possesses a good antibacterial property. This solution can be used to reduce the microbial growth in the oral and may also be given for other reason like for analgesic action, gingivitis, anti-inflammatory activity. Various herbal products and their extracts such as Guava, Pomegranate, Neem, Propolis, Tulsi, Green Tea, Cranberry, Grapefruit etc. Have shown significant advantages over the chemical ones. The present research is aimed to formulate and evaluating herbal mouthwash and evaluating its efficacy against microbial oral cavity load. Due to its physicochemical properties and antimicrobial activity, prepared mouthwash was further evaluated. This research is an attempt to outline such natural substances, which may be used as effective mouthwashes. Evaluate its antimicrobial and anti-inflammatory efficacy against commercially available herbal mouthwash as supplements to daily oral hygiene.

Key words: Polyherbal mouthwash, Oral hygiene, Natural herbs, Anti -bacterial activity.

Introduction

The importance of mouth and teeth cleanliness has been recognized from the earliest days of civilization to the 21st century. Patients and oral health practitioners are faced with a multitude of mouthwash products containing many different active and inactive ingredients. Currently, toothbrushing is the most popular self-performed oral hygiene method to mechanically remove dental plaque. However, this mechanical approach by most individuals is often not sufficiently effective, suggesting that a chemical plaque control by mouthwashes could be beneficial as a supplement to daily oral care. Herbal mouthwashes are in excessive demand, because they act on oral pathogens and relieve the pain instantly and are also less side-effective. Chemical mouthwashes have hydrogen peroxide a chlorine dioxide, and acetyl pyridinium chloride, as an immediate whitener, sterilizer and pain reliever of teeth, but they tend to produce discoloration of teeth and may produce side effect, meanwhile they are cost effective. The mouth washes are concentrated aqueous anti-bacterial solutions that are used against oral microbes to counter oral infection, cleansing, to get rid of bad breath refreshing, anti-septic. The mouthwash plays a prominent role in the oral hygiene of an individual; it helps to relieve symptoms of inflamed gums gingivitis. And also it reliably used to destruct the pathogenic germs.

A variety of different chemical plaque control measures are available in the market, which includes mouthwash, dentifrices, chewing gums, and gel. But they have some undesirable side effects, like vomiting, diarrhea, and tooth staining. Any antimicrobial/antiseptic agents used should be able to modify the oral environment by being specifically effective against the pathogens without altering the normal flora. There are a number of mouthwashes available in the market today worldwide. Many of these mouthwashes have not been tested adequately, and the information is lacking as to when and how to use these agents for maximum benefit. Mouthwashes are widely used solutions due to their ability to reduce the number of microorganisms in the oral cavity. Given the rapid pace of scientific research and clinical data provided by the large number of people who are rapidly infected with SARS-CoV-2, clinicians need reliable evidence of good medical care for this infection, as it is simple to do in-silico analysis in the initial stage with the aid of molecular docking software with help of chemical structure of compound. Herbal mouthwashes are designed and prepared with extracts and essential oils from phyto therapeutic plants, containing a mixture of active agents such as catechins, tannins, and sterols. The mixture of natural compounds inside the herb- or plant-derived substances usually performs gentle remedial effects. Compared with the antimicrobial mechanisms by synthetic chemicals, herbal mouthwashes can have additional anti-inflammatory and antioxidant properties, which could further benefit to health.

“Smile till you have teeth.”- An ancient and famous saying. Extracts from neem inhibit the growth of *S. mutans* and used in the treatment of periodontitis. It contains anti-microbial, anti-inflammatory, and anti-oxidant property. Tulsi (*Ocimum Sanctum*) as a mouthwash is quite effective for the ulcer and infections in the mouth. Many herbal mouthwashes contain herbs with anti-microbial property such as neem, yavanisatva, nagavali, gandhapurataila, pilu, bibhitaka, ocimum, Echinacea, chameli leaves, etc. some of the herbs that are used in mouthwashes are clove, which is traditionally used for oral health because of their antiseptic, antibacterial and antiviral property, peppermint which gives cooling effect to the mouth.

Natural Herbs such as Tulsi, Green tea, Clove oil, Nagarmotha, Cinnamon oil and many others are used as single or in combination have been Scientifically Proven to be Safe and Effective Medicine against Oral Health Problems such as Bleeding Gums, Gingivitis and Preventing Tooth Decay without side effects. Mouthwashes may serve as a measure in controlling dental plaque and periodontal disease for mentally or physically handicapped patients who are incapable of brushing their teeth themselves or other individuals who are lacking dexterity, skill, or motivation for mechanical plaque removal. Thus, instead of using them solely, mouthwashes should always be used in association with mechanical plaque control measures. Medicinal plants have been used for curing diseases in different traditional systems of medicine such as Ayurveda, Siddha, European, Tibetan, and Unani. Due to its high cultural acceptability, compatibility with the human body, and less side effects, herbal therapy is still the mainstay of therapy in roughly 75–80% of people in many underdeveloped nations for their primary health care. Thus, a potential supporting antimicrobial alternative with minimized side effects would be greatly valued to work on oral infections. There is a need for an alternative medicine enmeshed within precious traditional Indian herbal therapy, which is efficient, safe, and economical.



Materials and Methods

Formulation Table

Sr.no	Ingredients	Botanical name	Role	Quantity
1	Tulsi	Ocimum sacntum	Dental care	10ml
2	Clove	Syzygium Aromaticum	Anesthetic	6ml
3	Cardamom	Elettaria Cardamomum	Antimicrobial	8ml
4	Dry Ginger	Zingiber Officinale Roscoe	Antibacterial	2ml
5	Honey	Apis Mellifera	Sweetning agent	2ml
6	Distilled Water	-	Vehicle	Q.S

1) Tulsi

1. Tulsi (Lamiaceae) mouthwash shown strong anti-microbial activity against various bacterial strains, along with its bacterio statics, anti-oxidant and immune- modulatory properties. It is used also, as therapeutic agent for periodontal diseases.

2. Tulsi has antibacterial properties which helps to relive cold.

3. Tulsi is known for its medicinal values, antimicrobial and anti-viral properties which helps in purifying the air.



Fig no.1 Tulsi



Fig no.2 Extraction Process of Tulsi

2) Clove

1. Cloves (Myrtaceae) contain a strong anesthetic known as eugenol.
2. Protect your teeth and gums from bacterial attacks and ensure strong teeth and gums.
3. Clove is also a popular ingredient in cigarettes.
4. Cloves are flower buds that come from a type of tropical evergreen tree native to Indonesia.



Fig no.3 Clove



Fig. no.4 Extraction Process of Clove

3) Cardamom

1. Cardamom (*Elettaria cardamomum*) is a pungent, aromatic, herbaceous, evergreen perennial of the ginger family.
2. Helps keep your mouth moist and protects you against dental health issues like cavities.
3. Cardamom is widely used as a mouth freshener in India since it is effective in fighting bad breath. It also fights ulcers or other infections of the mouth.
4. Eating Cardamom daily can be beneficial to your overall health. It can help support digestive health, circulation, and the respiratory system.



Fig no.5 Cardamom

4) Dry Ginger

1. Dry ginger (*Zingiberaceae*) can even reduce your risk of gingivitis, gum disease, and oral cancers.
2. Ginger can help whiten and strengthen your teeth and gum line.

3. The antibacterial properties of ginger can slow down the bacteria growth. These properties are the result of the oil, resin, starch and spices found in ginger.

4. Dried Ginger, when powdered, is an effective cure for indigestion, sore throat, cold and cough.



Fig no.6 Dry Ginger

5) Honey

1. Honey (Apidae) can help treat gum diseases, including gingivitis, bleeding, and receding gums. 95% of adults have periodontal disease at least once in their lives.

2. Honey-based mouthwash showed a promising antimicrobial effect on dental caries and plaque and gingival scores.

3. Organic honey is powerful antibacterial and antiseptic properties can help treat gum disease and prevent tooth decay.

4. Honey possesses antibacterial effects that work to neutralize nearly 60 species of bacteria while also working to prevent the development of resistant bacterial strains.



Fig no.7 Honey

Procedure of Mouthwash

1. Take 6 ml of clove extract + 8 ml of Cardamom extract + 2ml of Dry ginger in 10 ml of tulsi extract.

2. Also add 2ml of Honey as a sweetener.

3. Followed by methyl paraben as a preservative, and make up with distilled water until the 50 ml.

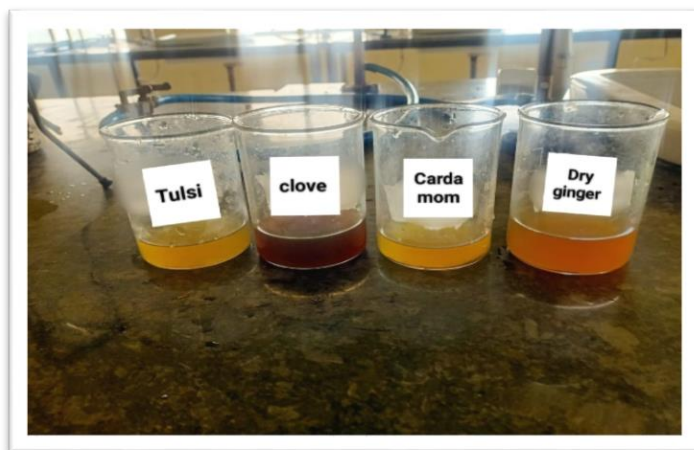


Fig no.8 Extraction of Ingredients

Evaluation Tests

Colour and Odour : Physical parameters like odour and colour were examined by visual examinations.

2. PH:

PH of prepared herbal mouthwash was measured by using digital pH meter. The pH meter was calibrated using standard buffer solution about 1 ml of mouthwash was weighed and dissolved in 50ml of distilled water and its pH was measured.

3. Density:

The density is given to us as $D=0.789\text{g/ml}$

Result of stability of stability studies of Herbal Mouthwash

Temperature	Evaluation parameters	Observation(months)			
		1	2	3	4
3-5°C	Visual Appearance	Light green	Light green	Light green	Light green
	Phase Separation	Nil	Nil	Nil	Nil
	Homogeneity	Good	Good	Good	Good
Room Temperature(25°C RH =60%)	Visual Appearance	Light green	Light green	Light green	Light green
	Phase Separation	Nil	Nil	Nil	Nil
	Homogeneity	Good	Good	Good	Good
40°C±2°C RH=75%	Visual Appearance	Light green	Light green	Light green	Light green
	Phase Separation	Nil	Nil	Nil	Nil
	Homogeneity	Good	Good	Good	Good



Test for Microbial growth in Formulated Mouthwash

The formulated mouthwash was inoculated in the plates of agar media by streak plate method and a control was prepared. The plates were placed in the incubator and are incubated at 37°C for 24 hours. After the incubation period plates were taken out and checked for microbial growth by comparing it with the control.

Stability studies

The formulation and preparation of any pharmaceutical product is incomplete without proper stability studies of the prepared product. This is done in order to determine the physical and chemical stability of the prepared product and thus determine the safety of the product. A general method for predicting the stability of any product is accelerated stability studies, where the product is subjected to elevated temperatures as per the ICH guidelines. A short term accelerated stability study was carried out for the period of 3 months for the prepared formulation. The samples were stored at under the following conditions of temperature as 3- 50C, 250C RH=60%, 400C \pm 2% RH=75%. Finally the samples kept under accelerated study were withdrawn on monthly intervals and were analysed.

In-Vitro Antibacterial Activity

In vitro antibacterial activity was performed on isolated colonies of *Streptococcus mutans*. The Agar well diffusion technique was used for determining the zone of inhibition and minimum inhibitory concentrations (MIC). The strains of *S. mutans* were inoculated in prefabricated blood agar plate. Plates were dried and 4 wells were made with the help of 6 mm agar well cutter. 20 μ l, 40 μ l, 60 μ l, 80 μ l of prepared mouthwash was loaded in all the respective wells. The agar plates were kept undisturbed to allow the passive diffusion of herbal mouth wash into the agar culture medium. Then the plates were incubated at 37°C for 24 hours. The zone of inhibition was calculated in mm. A wide range of mouthwashes containing different active ingredients is available on the market. It is important to know their antimicrobial activity because they are mainly employed to control microorganisms.

Result and Discussion

This mouthwash is a purely herbal prepared without the addition of any kind of alcohol and any other additives as other products found in the market. The formulation was undertaken Stability studies for physical and chemical change. No considerable variations in properties of the formulation were observed.

The pH of the formulation was found to be 6.1. As the skin is having an acidic pH around 5.5 this pH range of the formulation is suitable for oral disorders. The formulation was found to be free from heavy metals. The formulation was free from microbes as they have not produced any microbial growth when they got inoculated in the agar medium. Alcohol consumption as well as alcohol and tobacco use are known risk factors for head and neck cancers. It has always been the question of whether use of alcohol containing mouthwash increases the risk of cancer. When used in mouthwashes antimicrobial ingredient like neem, clove and other essential plant extracts have been found to reduce plaque and gingivitis when combined with daily brushing and flossing. Volatile sulphur compounds are the major contributing factor to bad oral odour. They arise from a variety of sources that is breakdown of food, dental plaque and bacteria associated with oral disease. The antibacterial activity was

evaluated by agar diffusion method for different concentrations of mouthwash. The result of zone of inhibition for *S. mutans* was found to be 18 mm for 80 µl, 15 mm for 60 ml 12 mm for 40 µl and 7 mm for 20 µl respectively and 20 mm for 80 µl, 22 mm for 60 µl, 17 mm for 40 µl and 14 mm for 20 µl respectively for *S. salivarius*. These results showed that the herbal mouthwash has significant antibacterial activity and the present preparation is able to inhibit bacterial growth in oral cavity. The association of oral microbial load on oral microbial load on oral diseases is well established.

Conclusion

The natural herbs used in present formulation have been medicinally proven to prevent the problem of oral hygiene and bad breath. This mouthwash is purely herbal without adding any kind of alcohol and any other additives as other products found in the market. Mouthwash is a liquid accessory to clean and maintain the health of our teeth for oral hygiene. Several herbal mouthwash and herbal extracts have been tested in-vitro and in- vivo in search of suitable adjunct to mechanical therapy for long term use. The pH of the formulation was found to be 6.1. As pH range of the formulation is suitable for oral disorders. When used in mouthwashes antimicrobial ingredients like Clove, Cardamom, Dry ginger and Tulsi plant extracts have been found to reduce plaque and gingivitis when combined with daily brushing and flossing. The results of zone of inhibition also confirmed that this herbal mouth rinses was found to be a potent plaque inhibitor, and were preferred by the patients for its taste, convenience of use and test duration in their mouth after rinsing. Thus, these can be used as an adjunct to mechanical therapy for treating plaque induced gingivitis. Present study has an important impact in order to create an effective and inexpensive herbal oral health intervention for low social economic communities. Since years and decades, these herbs have been known for working wonders as reflected in many research findings. Person can easily rinse his mouth using this herbal mouthwash and stay clear of wide variety of oral health

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