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FORMULATION AND DEVELOPMENT OF MOUTH ULCER GEL

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ABSTRACT

The objectives of present investigation were to formulate and evaluate herbal gel for mouth ulcer treatment of dried powdered guava leaves. Herbal gel was prepared by using different concentration of powdered guava leaves and Carbopol 934, Propylene glycol as a gel base. Formulations were evaluated for various parameters Infrared spectroscopy revealed that there was no interaction between powdered Guava leaves and Polymer. The formulated gel was transparent, homogeneous and pH ranges from 7 to 7.5. Formulation showed acceptable rheological behavior with applicable Spread ability and Extra debility properties. Anti-fungal studies of formulations showed excellent efficacy against Aspargiliousaureus, Candida albinos. From the experimental evidence of investor studies it was observed that powdered guava leaves contain flavonoids so it showed significant antioxidant effect. Developed herbal formulation was stable, safe and effective over to synthetic formulations for the treatment of mouth ulcer.

KEYWORDS: *Guava Leaves (Psidium Guajava), Aphthous Ulcers, Mouthwash .*

INTRODUCTION

Gels are typically semi-solid formulations having a liquid phase that has been Thickened with other components. Uses of topical gel preparations are for skin application or percutaneous penetration of medicament or local action to certain mucosal surfaces (Singh, 2014).

A mouth ulcer is a break or breach in the mucous membrane, which is lines the inside of the mouth. It usually has yellow or white color and usually looks like a depression in mouth that is the mucous membrane (Dosani, 2011).

The Commercially available gels containing synthetic and semi synthetic active agents which have several disadvantages like staining on the teeth, irritation, and burning sensation only because presence of high degree of alcohol content and some organic compounds.

The present investigation deals with use of herbal powdered Guava Leaves in the treatment of mouth ulcer in pharmaceutical gel. Commonly known as guava, Peru, Ambrud. A biological source is Psidiumguajava belongings to family Myrtaceae. Chemical composition contains Flavonoids, Triterpinoids,

Steroids, Carbohydrates, Oils, Lipids, Glycosides, Alkaloids, Tannins and Saponin. Used as Antioxidant, Antibacterial activity, Anti-inflammatory activity, anticancer activity (Wang, 2014). Importance of herbal medicine has both medicinal and economical.

Although herbal medicines has benefits to increased, their safety, efficiency, quality and importance of industrialized and developing countries. Herbal medicines are getting increasing patient compliance as they are avoiding typical side effects of allopathic medicines.

It is no wonder that the world's one-fourth population i.e. 1.42 billion people, are dependent on traditional medicines for the treatment of various diseases. Medicinal plants have been a major source of cure for human diseases since time immemorial. Recently considerable attention has been paid to utilize bio-friendly and eco-friendly plant based products for the cure and prevention of different diseases, so it is documented that most of the World's population has taken in traditional medicine.

The India offers a variety of plants having medicinal properties. Medicinal plants can be use to find out effective alternative to synthetic drugs (Jadhav, 2015). The use of the medicinal plant based medication is gradually becoming popular throughout the world.

Near about half of the worlds, twenty five bestselling pharmaceutical innovator agents are derived from natural products (Das, 2011). The use of medicinal plants as raw materials in the preparation of new drugs is ever increasing because of their potentials and the problem of drug resistance in micro-organisms.

Demand for medicinal plants is increasing in both developed and developing countries. Research on herbal medicinal plants is one

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of the leading areas of research globally (Dwivedi, 2012)

Herbal formulations have now a day's undergone more thorough investigation for their potential in preventing and cure oral disease (Silva, 2012). Herbs have long been used traditionally for routine cleaning of teeth and dental disease and to treat various oral diseases(Deepa, 2011)

Oral diseases like oral cancer, dental caries and periodontal diseases among the most important oral health problems. There is a well-established link between the activities of microbial species that form part of the micro biota of the oral cavity and oral diseases.

The big need for alternative treatment, products and prevention options for oral diseases that are safe, economical and effective comes from the rise in disease incidence particularly in developing countries, increased resistance by pathogenic bacteria to currently used chemotherapeutics and antibiotics opportunistic infections in immunocompromized individuals and financial that is economical considerations in developing countries.

Moreover, allopathic medicine is too expensive and capital intensive for a developing country like India and has only limited success in the prevention and treatment of oral diseases and periodontal disease.

Hence, the plant extracts used in traditional medicine and alternative products continues are considered as good alternatives to synthetic and organic medicine (Nagi, 2015)&(Jose, 2011).

The present investigation deals with use of herbal Guava Leaves in treatment of mouth ulcer in pharmaceutical gel.



GUAVA LEAVES

Description/Taste

Guava leaves are oblong to oval in shape and average 7-15 centimeters long and 3-5 centimeters wide. The leaves grow in an opposite arrangement, which means two leaves grow at the same point on either side of the stem, and have short petioles, or stalks that join the leaf to the stem. The surface of the deep green Guava leaf is wide and leathery with faint white veins and some light brown patches. Guava leaves are aromatic when crushed and have a scent similar to that of the guava fruit. Guava leaves grow on a small tree with widespreading branches and copper-colored flaking bark that revealsagreenbase.

Seasons/Availability

Guava leaves are available year-round. Current Facts

Guava leaves, botanically classified as Psidium guajava, are members of the Myrtaceae, or myrtle family along with eucalyptus, allspice, and clove. Guava leaves have been used in traditional Eastern medicine since ancient times and have recently gained in notoriety as an alternative natural medicine.

Nutritional Value

Guava leaves have many anti-inflammatory properties and also contain vitamin C, vitamin B, antioxidants, and tannins.

Applications

Guava leaves are most popularly consumed in tea, as capsules, ground into pastes, and extracted as essential oils. Young leaves are traditionally preferred for medicinal benefits and can be found in health stores in various forms. They can also be found dried and ready for use in specialty tea stores. When dried, the leaves can be crushed and boiled to make the medicinal tea.

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Ethnic/Cultural Info

Guava leaves have traditionally been used in Eastern medicine as diarrheal remedy and to reduce symptoms of food poisoning. They have also been used in China and India as a method to reduce symptoms of coughs and aid indigestion. In addition to oral remedies, Guava leaves are also being used in Brazil and Mexico externally to reduce symptoms of skin and body wounds.

Geography/History

The guava tree is believed to be native to Mexico, Central America, and the Caribbean and then spread to tropical and sub-tropical regions of the Americas, Australia, and Asia. Today guava trees are being produced in India, Nigeria, Philippines, Southeast Asia, Pakistan, Bangladesh, Brazil, China, and Mexico and the leaves can be found in specialty markets and online stores across the world.



Fig-Mouth Ulcers



Fig-Mouth Ulcer



Fig-Mouth Ulcer

MOUTH ULCERS

A mouth ulcer is a sore that develops in the soft tissue lining of your gums, tongue, inner cheeks, lips or palate. They're usually yellow or red, and they can be quite painful. Mouth ulcers may also be called aphthous ulcers or canker sores.

Mouth ulcers are common and should clear up on their own within a week or 2. They're rarely a sign of anything serious, but may be uncomfortable to live with.

Symptoms of a Mouth Ulcer

Mouth ulcers are easy to spot. They usually appear as sores on your lips, gums, tongue, inner cheeks or roof of the mouth. While red around the edges, mouth ulcers are typically white, yellow or gray in the center. You may only develop one ulcer, or there might be

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more. Other symptoms could include:

- Swelling around the ulcer.
- Increased soreness when brushing your teeth.
- Pain that worsens when eating spicy, salty or sour foods.

What causes mouth ulcers?
The exact cause of mouth ulcers is unknown. But there are several factors that can contribute to the development of these sores:
Minor tissue injury from dental work, such as having a cavity filled. Accidentally biting your cheek or tongue.
Allergic reaction to certain bacteria. Wearing orthodontic braces or retainers. Vitamin deficiencies.
Using harsh or abrasive toothpaste.
Eating a lot of acidic foods, such as oranges, pineapples and strawberries. Hormonal changes during your period.
Stress. Lack of sleep. Viral,bacterial or fungal infections.

Aim
Formulation And Development of Mouth Ulcer Gel

Objectives
To determine whether Psidium guajava leaves mouthwash is effective in the Management of patient with aphthous ulcer.

MATERIALS AND METHODS

All chemicals used for different experimental studies such as chloroform, conc.H2SO4, ammonium hydroxide, methanol, formic acid, acetonitrile, ampicillin, glucose, phenol red, 5,5dimethylpyrroline-N-oxide (DMPO), FeSO4, H2O2 were ofanalytical grade procured from M/s. SD Fine, M/s.Qualigens and M/s. SRL, India. Pectin was procured from Purix India Pvt. Ltd., 2,2-diphenyl-1picrylhydrazyl (DPPH) and Muller Hinton broth were procured from M/s. Sigma and M/s. HiMedia respectively.

1. Sample Collection and Processing

Fresh leaves of P. guajava L. (guava) were collected from the premises of Vignan's Foundation for Science, Technology and Research, Vadlamudi (16.2334° N, 80.5509° E). The leaves were rinsed gently with double distilled water; shade dried and powdered using a blender. The powder was then passed through aluminium sieve (1 mm) to get uniform particle size. Guava leaf powder was stored in an air tight container for further Studies.

2. Preparation of Guava Leaf Extract

The guava leaf powder (20 g) was boiled at 90 °C in 100 mL of double distilled water in sterile Erlenmeyer flask (150 mL) for 30 min. The mixture was centrifuged at 4000 rpm for 10 min (Biswas et al. 2013). The supernatant was separated and stored at 4 °C for further studies.

3. Phytochemical screening of guava leaf extract:

The guava leaf extract was tested for the presence of bioactive components. A mixture containing glacial acetic acid (2 mL), 2 drops of FeCl3 and 2 mL of conc. H2SO4 was mixed with aqueous extract. A brown ring at the interface confirms the presence of glycosides. For detection of saponin, the extract was taken in a measuring cylinder, diluted with distilled water and was shaken vigorously for the formation of foam (Kokate 1999). The presence of tannins and phenols were confirmed by formation of blue-green/black color upon mixing the extract (1 mL) with 2 mL of FeCl3 (2%, w/v) (Evans 1997). For screening terpenoids, 5 mL of extract was mixed with 2 mL of chloroform and 3 mL of conc. H2SO4 to form a layer. The presence of terpenoids is confirmed by the formation of reddish brown color at the interface. Besides, the presence of flavonoids is confirmed by yellow fluorescence upon treating 0.5 mL of extract with 5 mL of 10% ammonium hydroxide solution (Evans 1997).

4. Characterization of photochemical by mass spectroscopy:

Mass Spectroscopy is an analytical technique used for separation and identification of various components present in a mixture. Agilent 1100 LC/MS System with Chemstation Rev.A.09.01(1206) software was used for sample processing and analysis. The extract was mixed in methanol in 1:10 ratio and 20 µL of the sample was directly injected into the mobile phase [0.1% formic acid in water (50%) and acetonitrile (50%)] at a flow rate of 0.5 mL/min. The electro spray ionization (ESI) was set in negative ionization mode in 60–200 V and capillary voltage at 4000 V. Nitrogen is used as mobilizing gas at 350 °C and 30 psi pressure with flow rate of 8–10 L/min.

5. Preparation of Jelly and its Proximate Analysis:

Two types of jellies, with and without guava leaf extract was prepared by mixing pectin, sugar and lime juice as the major ingredients. In 100 mL of water, pectin (1.5 g) along with 28 g of sugar and guava leaf extract (10%, v/v) was mixed and boiled at 100 °C till the mixture was thickened. To minimize pre-gelling and hydrolysis of pectin, 2 mL of Citrus aurantifolia was added at the end of boiling. The mixture was cooled for the jelly formation and stored in airtight container. Further, the jellies were subjected to proximate composition (AOAC 2008) and texture analysis.

6. Physicochemical Analysis

The total soluble solids (TSS) of samples is determined by using hand refractometer (Model- MCP Metal and PP), having range of

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0–100Brix (Ghosh et al.º 2017). The digital pH meter (Mettler Toledo, USA) was used to measure the pH of the samples. Viscosity of the samples was determined by Brookfield Viscometer (Model DV1 Digital Brookfield, Middleboro, USA) at 30 ± 0.5 °C (Keshani et al. 2012).

7. Texture Analysis

Texture profile of jellies was determined by using CT3 texture analyzer connected to a cylindrical probe (TA4/1000, 20 mm L) at pre-test speed: 2.00 mm/s, test speed: 1.00 mm/s, post-test speed: 1.00 mm/s and load cell: 10,000 g. The total profile analysis of jellies was performed for two cycles for 5 replications. Various properties such as firmness, cohesiveness, chewiness, springiness and gumminess were determined by calibrated load cell through measuring the resistance of material against force applied by the spindle, and analyzed results were taken from the installed Texture Pro CT Software in their respective units (Ghosh et al.2017).

PREPARATION OF HERBAL GEL

Specified amount of Carbopol 934 was dispersed in required amount of distilled water with continuous stirring.5 ml of distilled water was taken and required quantity of methyl paraben and propyl paraben were dissolved by heating on water bath after cooling propylene glycol was added. Further varying concentration of Psidiumguajava powder was mixed to the above mixture and volume was made up to 20 ml with distilled water.

Finally full mixed ingredients were mixed properly to the Carbopol 934 gel with continuous stirring and triethanolamine was added drop wise to the formulation for adjustment of required pH (6.8-7) (Das, 2010).





Table 1: Composition of Various Gel Formulations Containing Powdered Guava Leaves.

Ingredients	G1	G2	G3
Guava leaves powder	2%	1%	0.5%
Carbopol 934	2%	2%	2%
Methyl Paraben	0.0015%	0.0015%	0.0015%
Propyl Paraben	0.01%	0.01%	0.01%
Triethanolamine	q.s + pH 6.5-7	q.s + pH 6.5-7	q.s + pH 6.5-7
Distilled water	Up to 20 ml	Up to 20 ml	Up to 20 ml

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RESULTS AND DISCUSSION

From the result it is clearly shown that all the prepared gel formulations having good homogeneity and gelling properties. The pH of all gel formulations was in the range of compatible with normal pH range of the skin. The rheological behavior also indicates that the gels were neither too thick nor too thin. The Spreadability shows that with increasing viscosity of formulation, Spreadability decreases and vice versa. Extrudability study was done by pressing thumb and it's easily extendable. The gelling and bioadhesive strength of all the batches was found in the suitable range. one Month stability study was done with open and close container and it's showed that open container containing gel was not stable and close container gel was stable. Formulated gel containing open container when expose to ambient room temperature then syneresis was observed it means liquid exudates separating. Syneresis occurs when the interaction between particles of the dispersed phase becomes so great that on standing. In that dispersing medium is squeezed out in droplets forms and the gel shrinks. Syneresis it means the form of instability in aqueous gels. In syneresis system separation of a solvent phase is occur only because of the elastic contraction of the polymer means polymeric molecules. All the three batches of developed formulation showed antifungal activity against Aspargilious aureus and Candida Albicans this are main microorganism responsible for mouth ulcer and formulation it can also use to treat mouth ulcer infection.

Comparison of VAS scores of guava treatment and NSS control groups showed that there were no differences in pain experienced on Days 1 and 2. However, the VAS scores from Day3 to 7 had values ranging from 0.02 - 0.0001 which showed significant differences in resolution of pain. There was statistically significant marked improvement of pain symptoms as early asthree days post-treatment among patients who were administered guava leaves mouthwash.Complete resolution of aphthous ulcers in 75% of the study group was observed on Day 7. Meanulcer size post-treatment with guava gargle was 0.25mm compared to 0.75mm for NSS gargle.The mean size difference at day 7 was 1.44mm for the guava treatment group and 0.88mm for theNSS control group. There was a statistically significant faster resolution of ulcer size on day 7 in16/16 or 100 % of patients in the treatment group compared with only 10/16 or 62.5% of patientsin the control group. Patients who were administered guava leaves mouthwash generally faredbetter than those administered isotonic sodium chloride solution.

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CONCLUSION

The data presented in the study, it was demonstrated that the developed herbal gel formulation possess significant, therapeutically efficacious, suitable vehicle for drug delivery in low cost but definitely with high potential. Developed new herbal gel formation is suitable for mouth ulcers treatment. Guava leaves mouthwash was effective for aphthous ulcers in terms of reduction of symptoms of pain and faster reduction of ulcer size. Further clinical trials comparing this mouthwash against other treatment options are recommended

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