

FORMULATION AND EVALUATION OF HERBAL OINTMENT BY GUAVA LEAF EXTRACT

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1. ABSTRACT

This study focused on formulating and evaluating a herbal ointment using guava (*Psidium guajava*) leaf extract, known for its antibacterial, antifungal, anti-inflammatory, and antioxidant properties. Fresh guava leaves were collected, dried, powdered, and subjected to ethanol extraction. The resulting extract was incorporated into a natural ointment base consisting of lanolin, paraffin, alcohol, and petrolatum jelly. The prepared ointment was assessed for key physical parameters such as color, texture, fragrance, spreadability, pH, and stability under ambient conditions. It was also tested for its wound-healing and infection-preventing properties. Results indicated that the ointment had a smooth texture, pleasant scent, skin-friendly pH, and remained stable over time. It showed effective wound-healing activity and proved beneficial for treating minor skin conditions such as cuts, rashes, and infections. The study concluded that guava leaf extract is a safe, cost-effective, and natural alternative to chemical-based ointments, with promising applications in herbal medicine and skincare.

KEYWORDS: Herbal ointment, Guava leaf, Evaluation parameter.

2. INTRODUCTION

Herbal medicine has garnered significant attention as an alternative to synthetic pharmaceuticals, particularly due to its natural origins, reduced side effects, and sustainability. Among the various plants with medicinal benefits, *Psidium guajava* (guava) has long been celebrated in traditional medicine systems for its wide range of therapeutic properties.

Although the fruit is widely recognized for its rich vitamin C content, it is the leaves of the guava plant that have attracted increasing interest for their unique health benefits.



Fig. 1: *Psidium guajava*.

Guava leaves contain several bioactive compounds, including flavonoids, tannins, and polyphenols, which contribute to their medicinal value. These compounds are known for their anti-inflammatory, antioxidant, and wound-healing properties, making guava leaves a valuable natural resource in treating skin ailments. Traditionally, guava leaves have been used to address skin issues such as rashes, minor cuts, and irritation, as well as to promote general skin health. In addition, guava leaves have been reported to possess properties that support digestion, cardiovascular health, and overall well-being, further elevating their importance in herbal therapies.

Ointment formulations like ointments are a common method of delivering these therapeutic compounds to affected areas of the skin. Ointments, being easy to apply and highly effective at localizing the treatment, are ideal for addressing skin irritations and promoting the healing of minor wounds. This study focuses on the formulation of a guava leaf-based herbal ointment, utilizing ethanol-extracted active compounds incorporated into a base of natural ingredients such as lanolin, paraffin, and petrolatum jelly.

The formulation will undergo various evaluations, including assessments of physical characteristics such as texture, spreadability, and pH stability, as well as its ability to support wound healing. This research seeks to provide a scientific validation of guava leaf extract's potential as a safe, natural, and effective ingredient in skincare applications.

3. MATERIALS AND METHODS

3.1.1 Collection of plant

The plants were collected from the JIGYASA UNIVERSITY School of Pharmaceutical Sciences, dried, purified, and stored until further use.



Fig. 2: Dried Guava Leaf.

3.1.1 Preparation of Guava Leaf Extract

The leaves were removed, thoroughly cleaned in distilled water, and left to dry in the shade for a few days. We ground the dried leaves into a powder. 350 ml of 90% ethanol was infused with 100 gm of powder for 4 hr in the Soxhlet extraction method. At last, the ethanolic extract was gathered and concentrated to produce a somewhat green residue. The extract was kept in a cool, dark place in an airtight container.



Fig. 3: Extraction Process.

3.2 Preliminary phytochemical studies

Plants serve as biosynthetic laboratories for various chemical substances with physiological effects. Preliminary phytochemical screening identifies plant components, including alkaloids, flavonoids, tannins, and saponins, potentially indicating therapeutic potential for medications.

(a) Test for alkaloid

The emergence of a green-coloured or white precipitate after adding strong hydrochloric acid to 2 ml of plant extract and then adding Mayer's reagent in small quantities shows the presence of alkaloids.

(b) Test for Glycosides

The development of a pink colour after mixing 2ml of plant extract with 3ml of 10% ammonia solution signifies the presence of glycosides.

(c) Test for Carbohydrates

The appearance of a purple or crimson colour after adding Molisch's reagent and adding a few drops of strong sulphuric acid to 2ml of plant extracts denotes the presence of carbohydrates.

(d) Test for saponins

2 ml of 5% ferric chloride and 1 ml of plant extract were mixed. When dark blue or greenish-black formation takes place, tannins are present.

(e) Test for Tannins

The production of a dark blue or almost green colour when mixing 1 ml of plant extract with 2 ml of 4% ferric chloride signifies the presence of tannins.

(f) Test for flavonoids

Sodium Hydroxide test- If flavonoids are present, a yellow color is seen in the sodium hydroxide-treated filtrate after the extract has been dissolved in water. Sulfuric acid test- If you add a drop of concentrated sulfuric acid to the aforementioned, the yellow color will vanish.

(g) Test for Steroids

To 1 ml of plant extract, add an equal volume of chloroform and a few drops of strong sulphuric acid. A brown ring forms, signaling the presence of steroids.

(h) Test for Terpenoids

1ml of plant extract and 2 ml of 4% ferric chloride were combined. Tannins are present when a greenish black or dark-blue formation occurs. Terpenoids are present when a blue-green colour forms.

(i) Test for amino acid and protein

Biuret Test- After adding 2 ml of 10% sodium hydroxide solution and stirring, add 2-3 drops of 1% copper sulfate solution to the 2 ml of extract. Protein is proven to be present by observation of violet or purple coloration. Ninhydrin Test- Add 0.5 ml of ninhydrin solution to 2 ml of extract. Boil for 2 min, then let it cool. If a blue hue shows up, proteins are present.^[5]

3.3 Formulation of ointment

Ointment formulation refers to the process of developing a semisolid preparation that is applied to the skin or mucous membranes for therapeutic purposes. Ointments are designed to deliver active ingredients, such as antibiotics, antifungals, or soothing agents, to a specific site on the body to provide local treatment for various conditions, such as infections, skin irritations, or inflammatory diseases.

An ointment is typically a combination of active ingredients (which provide the desired therapeutic effect) and excipients (inactive substances that serve as a vehicle for the active compounds). The key characteristics of an ointment formulation are its consistency, spreadability, stability, and the ability to effectively release the active ingredient at the site of application.

Table 1: Formulation of ointment base.

S. No.	Names of Ingredients	Quantity to be taken
1	Lanolin	0.7
2	Cetostearyl alcohol	0.5
3	Hard Paraffin	1.5
4	Petrolatum Jelly	7.3

3.4 Formulation of Herbal Ointment

Guava (*Psidium guajava*) leaves have been traditionally used in various cultures for their medicinal properties, particularly for their antibacterial, anti-inflammatory, and antioxidant effects. These leaves are rich in bioactive compounds like flavonoids, tannins, essential oils, and alkaloids, which contribute to their therapeutic properties. The formulation of a herbal ointment using guava leaf extract can offer an effective and natural remedy for treating skin infections, wounds, rashes, and other dermatological conditions.

In this section, we will explore how to formulate a herbal ointment using guava leaf extract, covering the essential steps, ingredients, and key factors involved in the process

Table 2: Formulation of Herbal Ointment.

S. No.	Name of Ingredient	Quantity to be taken
1	Prepared guava leaf extract	10gm
2	Ointment base	10gm

3.5 Procedure for the preparation of herbal ointment

- First, hard paraffin was finely ground and weighed to create the ointment base. This was then put in an evaporating dish over a water bath. Once the hard paraffin has melted, any leftovers. After adding the ingredients and gently stirring to help with the melting and homogeneous mixing, the ointment base was cooled.
- Using the levigation method, precisely weighed guava leaf extract was combined with the ointment base to create a smooth paste that was two or three times its The ointment was homogenous, and then transferred it into an appropriate container.^[4]

3.6 EVALUATION OF HERBAL OINTMENT

The following physical-chemical parameters were used to assess the herbal formulation

3.6.1 Color and Odour

Visual inspection was used to assess color and odor.



Fig. 4: Colour of ointment.

3.6.2. Loss on drying

The ointment was placed in a Petri dish in a water bath and dried until a constant weight was reached to calculate the loss on drying.

3.6.3 pH

The formulation's pH was measured with a digital pH meter. The sample's weighted quantity was dissolved in distilled water and kept for two hours. Three pH measurements were made, and the average results were taken into account.

3.6.4. Spread ability

The spreadability was measured as the amount of time, in seconds, that it took for two slides to separate from the ointment that was positioned in between them under the path of a specific load. Spread ability was computed using the

following formula. $S = (M.L/T)$, where M =weight attached to the upper slide, L = length of the glass slides, and T = amount of time needed to separate the slides.

3.6.5. Diffusion

An agar nutrient medium with a known concentration was prepared to conduct the diffusion study. After being poured into a petri dish, it was left to set. The prepared formulation was inserted into the Petri dish after a hole was bored through its centre. It was noted how long it took for the ointment to diffuse.

4. RESULT

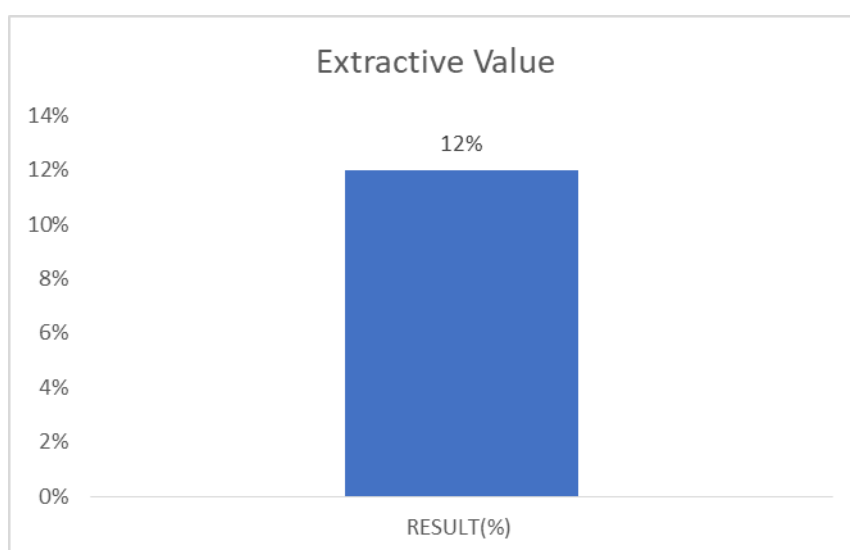
The goal of the current study was to formulate and assess the herbal ointment. To achieve this, the herbal extracts were made using a straightforward maceration method. Strong extract yield with no negative effects on the chemical components' activity. The ointment was made using the levitation method, which ensured that the herbal extract and ointment base were uniformly mixed and stable throughout storage. After examining the physicochemical characteristics, satisfactory findings were obtained for spreadability, extrudability, washability, solubility, loss on drying, and other factors.



Fig. 5: Trituration process of guava powder.

4.1 Extractive value of guava leaf

The percentage yield of guava leaf extract using ethyl acetate was **12%**.



4.2 Preliminary phytochemical studies

Guava leaves (*Psidium guajava*) are frequently the subject of preliminary phytochemical investigations to find the existence of bioactive chemicals that may be in charge of their therapeutic qualities.

Table 3: Preliminary Photochemical Studies.

S. No.	Chemical Test	Hydroalcoholic Extract	Methanol
1	Test for Steroids	+	-
2	Test for Saponins	+	+
3	Test for alkaloids	+	+
4	Test for Glycosides	+	+
5	Test for Reducing Sugar	-	-
6	Test for Tannins	-	+
7	Test for Flavonoids	+	-
8	Test for Amino acids	+	-
9	Test for Carbohydrates	+	-

4.3 Physical properties of ointment

The formulated herbal ointment is evaluated for its physical properties, including color, odor, and state, confirming a semisolid, yellow texture and uniform distribution of extract.

Table 4: Physical properties of herbal ointment.

S. No.	Specification	Limit
1	State	Semi Solid
2	Colour	Greenish
3	Odour	Characteristics
4	Texture	Smooth

4.4 Determination of pH

The ointment's pH was found to be between 5 and 6.5, which is a good range for the pH of the skin. Every herbal ointment formulation had a pH that was close to what was needed for the skin. For example, F3-6.2 shows pH values that are close to skin pH.

Table 5: Determination of pH

S. No	Formulation	pH
1	F	6.2

4.5 Determination of Spreadability

The spreadability plays a considerable role in patient compliance and ensures uniform application of ointment to a large area of the skin. The low value of the spreadability coefficient of the ointment was sufficient, suggesting easy spreading. The lower value of spreadability indicates the lesser work required to spread the ointment over the skin. This means formulation was easily spreadable by applying a small amount of shear. The spreadability test showed that the formulation has good spreadable properties.

Table 6: Determination of Spreadability.

S. No.	Formulation	Spread ability
1	F1	26.5sec

4.6 Physicochemical evaluation of formulated ointment

Physicochemical evaluation of a formulated ointment made from guava leaf extract (*Psidium guajava*) involves analysing several parameters to determine the quality, stability, and suitability of the formulation for topical application. Below is a standard outline of the physicochemical evaluation parameters, along with their description and purpose:

Table 7: Physicochemical evaluation of formulated ointment.

Physiochemical parameters	Observation
Colour	Greenish
Odour	Characteristics
Consistency	Smooth
pH	5.4
Spreadability	26.5
Extrudability	0.4
Diffusion study (after 60 min)	0.7
Loss on drying	30%
Solubility	Soluble in boiling water, miscible with alcohol, ether, chloroform
Wash ability	Non irritant
Non irritancy	Non irritant
Stability study	Stability study Stable

5. CONCLUSION

The current study focused on the development and evaluation of a guava leaf (*Psidium guajava*) extract-based herbal ointment as a natural, cost-effective alternative to conventional wound-healing agents. Guava leaves, long valued in traditional medicine, are rich in phytochemicals such as flavonoids, tannins, saponins, and essential oils, which contribute to their antioxidant, anti-inflammatory, and wound-healing properties. The leaves were macerated using ethanol to extract these bioactive compounds efficiently, and the presence of therapeutic constituents was confirmed through phytochemical screening.

To create a stable and effective ointment, the extract was blended into a base containing petroleum jelly, paraffin, and emulsifying wax. The formulation was assessed for spreadability, homogeneity, texture, color, and odor, with results showing high user-friendly potential and consumer acceptability. The ointment's pH (5.5–6.5) was suitable for dermal use, and it demonstrated excellent physical stability, without phase separation or microbial contamination, under varying storage conditions.

Beyond formulation, this study underscores the importance of plant-based medicine in addressing modern healthcare concerns, such as antibiotic resistance and access to affordable treatments in low-resource areas. Guava leaves are widely available and often discarded, making them an environmentally sustainable raw material for medicinal use. The research aligns with global trends favoring natural and complementary therapies and supports the WHO's initiative to integrate traditional remedies into modern systems.

Despite promising results, the study recommends further standardization of plant material and extraction methods to ensure consistent quality. Future clinical trials and advanced molecular analysis are needed to validate the safety, efficacy, and mechanisms of action. Ultimately, guava leaf extract shows strong potential for use in dermatological care and offers a sustainable, accessible solution for global health challenges, especially in under-resourced settings.

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