

Formulation and Evaluation of Herbal Gel Containing Powdered Betel Leaf (*Piper betle*) and Tulsi Leaves (*Ocimum sanctum* Linn) Extracts for the Treatment of Fungal Infections

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ABSTRACT

*This study aims to develop a novel herbal gel formulation incorporating powdered betel leaf (*Piper betle*) and Tulsi leaves (*Ocimum sanctum* Linn) extracts for the treatment of fungal infections. The gel formulation was prepared using a standardized procedure and evaluated for various parameters including pH, viscosity, spreadability, drug content, and in vitro antifungal activity against common fungal pathogens. The results demonstrated that the herbal gel exhibited desirable physical characteristics and significant antifungal activity, suggesting its potential as an alternative treatment option for fungal infections. Further clinical studies are warranted to validate its efficacy and safety in human subjects.*

Keywords:-Herbal gel, Betel leaf extract, Tulsi leaf extract, fungal infections, Antifungal activity, Formulation, Evaluation

INTRODUCTION

Gels are primarily semi-solid mixtures containing a liquid phase that has been supplemented with other ingredients to thicken it. For the skin applications, topical gel formulations are utilized. Numerous pathogenic fungi can cause fungal infections, which are a major burden on the world's healthcare systems and can frequently result in chronic illnesses and problems if left untreated. This problem is made more difficult by the advent of drug-resistant fungus strains,

which makes the search for alternate treatments necessary. [1] Fungi can invade many regions of the body and create fungal infections, which can result in a variety of symptoms and health problems. The respiratory system, internal organs, nails, and skin can all be impacted by these illnesses. Athlete's foot, ringworm, candidiasis, and fungal nail infections are common varieties. Additionally, fungi can spread by direct contact with an infected person or animal, contaminated soil, or by inhaling fungal spores. [2]



Fig.1:-Fungal infection

The antibacterial qualities of herbal treatments have long been known, and two in particular, betel leaf (*Piper betle*) and tulsi leaf (*Ocimum sanctum* Linn), have drawn interest due to their potential to treat fungal infections. Many cultures have long utilized betel and tulsi leaves for their

therapeutic benefits, which include their ability to fight against fungus. Bioactive substances found in betel leaf, such as tannins, flavonoids, and phenols, have broad-spectrum antibacterial properties. [3]

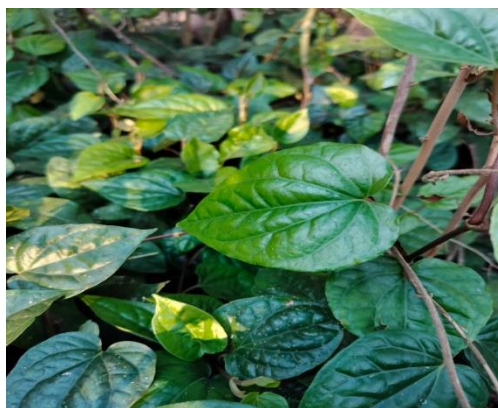


Fig.2:-*Piper betle*

In a similar vein, tulsi leaves are abundant in flavonoids, phenols, and essential oils

that have strong antibacterial and antifungal effects.[4]

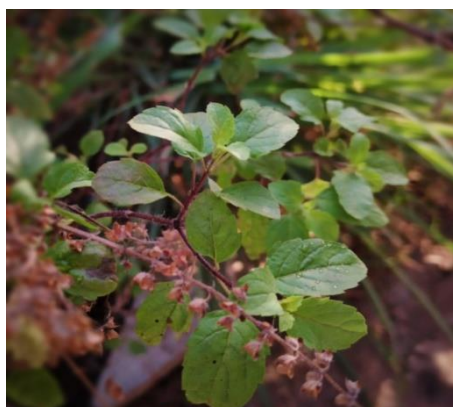


Fig.3:-*Ocimum Sanctum*

The purpose of this study is to create and assess a herbal gel that contains Tulsi and betel leaf extracts as a possible therapy for fungal infections. Synergistic benefits may be obtained by mixing these two herbal constituents, increasing the gel's overall antifungal efficacy while reducing side effects. [5]

MATERIAL AND METHODS

Collection and authentication of plant materials

The leaves of plant *piper betle* and *ocimum sanctum* were collected from the local area

of shahada, Nandurbar, Maharashtra, India in month of January 2024 and The plant specimens are authenticated by Dr. S.K.Tayade Head, department of Botany, P.S.G.V.P. Mandal's Arts, Science and Commerce College, shahada, Dist. Nandurbar respectively.

Chemicals

Carbopol 940, Methyl paraben, propyl paraben propylene glycol 400, Triethanolamine, Distilled water, ethanol, methanol, Glycerin.

Equipment's

Digital balance, PH meter, magnetic stirrer, Digital Water bath, Ultra sonicator, Brookfield viscometer.

The aim of present study is to established the formulation and evaluation of herbal antifungal gel containing extracts of piper betle and ocimum sanctum leaves to treat fungal infection.

Preparation of plant extract

The procedure for making plant extract from tulsi and betel leaves begins with us Gathered the leaves from both, rinsed under running water to get rid of any dust, and allowed to air dry for three to four weeks. Using a mechanical grinder, the dried plant components were ground into a coarse powder and then sieved through a

40-mesh screen. After that, the powder was extracted by cold maceration with water, methanol, and ethanol to produce the appropriate extracts. For 48 hours, 500 ml of ethanol, methanol, and water were macerated with 100 g of powdered dried betel leaves and 100 g of powdered tulsi leaves in separate conical flasks. at room temperature, shaking every now and again. following 48 hours. Mixture was removed using straightforward filtration method and filtrates were collected in separate vessels.to obtain the extract the solvent was removed from the filtrate under reduced pressure by using a rotary vacuum evaporator at 45-50°C^[6]

Table 1:-Ingredients used in formulation

Sr.no.	Ingredients	Functional category
1	Liquid Extract	Antifungal
2	Carbopol 940	Gelling agent
3	Propylene Glycol 400	Solubilizing agent
4	Methyl paraben, propyl paraben	Preservatives
5	Triethanolamine	pH Adjuster
6	Glycerin	Moisturizing agent
7	Rose water	Fragrance
8	Distilled water	Vehicle

Methods of preparation of gel

Take a 80 ml of refined water in a measuring glass and scatter 2gm carbopol 940 in it with keeps mixing and kept a container to the side of carbopol for 30 moment. In another measuring utencil take 5ml of refined water add 0.2 gm. Methyl paraben and 0.1gm propyl paraben to it by warming on water shower and cool the arrangement then add propylene glycol 400 (10 ml) and blend carbopol and make up volume 100 ml by adding refined water

and triethanolamine drop wise for change of pH.[7]

Formulation Table

The formulas and the above described procedure were tabulated in Table No. 2 Along with control sample gel were prepared by addition of required quantity of piper betle leaves extract and ocimum sanctum leaves extact to prepared antifungal gel^[8]

Table 2:-Formula for preparation of gel

Ingredients	Quantity in gm or ml			
	F1	F2	F3	F4
Betel leaf extract	0.5	0.8	1	1

Tulsi leaf extract	0.5	0.8	1	1
Carbopol 940	1	1.5	1.8	2
Propylene glycol 400	8	9	10	10
Glycerin	1	1.2	1.4	1.5
Triethanolamine	q.s	q.s	q.s	q.s
Rose water	q.s	q.s	q.s	q.s
Distilled water	Make to 100	Make to 100	Make to 100	Make to 100

After the trial of four different formulation batches, we did the visual inspection of all batches. The formulation trial four was identified the best visual inspection and the gel was tested for homogeneity and formulation showed good gelling property. The herbal gel was then prepared and subjected to evaluation of various parameters.

EVALUATION PARAMETERS

1) Physical Evaluation

Physical parameters such as color, odour and consistency were checked visually

Color:

The color of the formulations was checked by visual inspection.

Consistency:

The consistency of the formulations was checked by applying on skin.

Odour:

The odour of the formulations was checked by mixing the gel in water and observing the smell and the odour of gel was checked.

2) Measurement of pH

The pH of gel formulation were determined by using digital pH meter. After dissolving 1 gram of gel in 100 ml of distilled water, separate the mixture for a period of two hours. After that, the measurement of pH formulations was done by fully submerging the glass electrode into the gel system three times then pH of formulation measured, and the average value are given. [9]

3) Homogeneity

Every prepared gel formulation was visually inspected to ensure homogeneity before the gels were placed inside the container. Their presence and look were examined of any kind of aggregate. It was stated in that gel formulation was homogeneous. [10]

4) Viscosity

The viscosity of the prepared gel was measured using a Brookfield viscometer with spindle number 1. At 25 °C. The rotational speeds of the gels were 0.3, 0.6 and 1.5 rotations per minute and at each speed. The matching dial reading was recorded for each speed and beats per minute. The produced gels viscosity was then measured using the dial readings of Brookfield viscometer, which was based on factor found in Brookfield viscometer catalogs. Then viscosity was gel reported. [11]

5) Spreadability

Spreadability is measured in times in seconds or the amount of time it takes for two slides to slip from gel that is positioned between them when a specific stress is applied. If the time taken for separation of two slides is less than better the Spreadability. Spreadability is calculated by using the formula:

$$S = M \times L \div T$$

Where M = weight tied to upper slide

L = length of glass slide

T = time taken to separate the slides [12]

Spreadability of gel formulation was reported.

6) Extrudability

The planned gel had been filled in famous covered folding aluminum tubes and fixed through creasing as far as possible. The heaviness of filled tubes have been recorded and the cylinders were sandwiched between two glass slides and have been clamped. 250 gram weight was once situated over the slides and afterward the cap was eliminated to expel. The amount of expelled gel was once gathered and gauged. Still up in the air by working out the level of expelled gel. When it is greater than 90% then Extrudability is excellent.

When it is greater than 80% then Extrudability is good.

When it is 70% then Extrudability is fair [13]

Extrudability of gel formulation was reported.

7) Gel strength

Gel strength was resolute by the times in seconds required by the weight to penetrate in the gel. A 3-5 ml of formulation was placed in 10 ml graduated cylinder. A weight of 250 gram was placed on the gel surface. The gel strength, which indicates the gel at physiological temperature, was calculated as the times in seconds required by the weight to penetrate 2 cm into the gel. All measurement was done in triplicate. [14]

8) Clarity

The clarity of all four batches was determined by visual inspection. [15]

9) Stability study

The antifungal gel which we are created it was placed into collapsible tubes and stored at room temperature and 40°C with 75% relative humidity. We conducted a stability test over three months, checking its appearance, pH, homogeneity, viscosity and Spread ability each month. [16]

10) Skin irritation test

Applied the herbal antifungal gel formulation on the skin and observed for irritation, redness or Rashes [17]

11) Anti-fungal activity

The antifungal activity of all optimized formulation and blank formulation have been carried out by means of cup-plate method. The antifungal activity test was carried out by using candida albicans. Prepared nutrient brought and poured in to sterile petri plates and stored apart for drying and cooling. After that candida albicans culture were spread through micron wire loop. A sterile stopper drill 6mm measurement used to be utilized to penetrate openings 4 mm profound. Then place plan of gel in this openings. Plates were then brooded at 27°C for 48 hours. Then, at that point, the zone of restraint was estimated. [18]

RESULTS AND DISCUSSIONS**1) Physical evaluation**

The herbal antifungal gel formulation were found to have a yellowish green colour and the odour is characteristics and the consistency is good and feels smooth were applied.

2) Measurement of pH

The determination of pH of antifungal herbal gel formulations ranges from 4-7 and the pH of these antifungal gel is 5.43. Which is appropriate for the skin making the compatibility of the herbal gel formulation with skin. The pH of Antifungal gel is showed in figure no 4

3) Homogeneity

All the gels that were developed were carefully examined for uniformity through visual inspection to check for any lumps, flocculates, or aggregates. It was determined that consistency of all formulation was satisfactory. The homogeneity of herbal gel formulation was good.

4) Viscosity

Viscosity of gel was determined using Brookfield viscometer. Viscosity is an essential requirements for distinguishing the gels as it influences the spreadability, extrudability, and release of the drug. The viscosity of herbal antifungal gel is 4600 cps.

5) Spreadability

The spreadability plays the key part in patient compliances and help in even usage of the gel. A good gel takes less duration to spread and will have excellent spreadability. The spreadability of the herbal antifungal gel formulation is 9.80 gcm/sec.

6) Extrudability

The extrudability of the gel formulation is good extrudability when extruded from the metallic collapsible tube. The extrudability of the herbal gel formulation is 89.2

7) Gel strength

The gel strength is important parameter the gelling strength of the herbal antifungal gel is 25 ± 0.11 .

8) Clarity

The clarity of gel formulation was determined by visual inspection. The clarity of herbal antifungal gel formulation was excellent

9) Stability study

The stability studies were conducted for all the formulations for 3 months. No appreciable changes were found for the proven limits for the proven limits like physical appearance, pH, and odour, Homogeneity and spreadability at room temperature.

10) Skin irritation test

The prepared herbal antifungal gel formulation was applied on skin of hand and exposed to sunlight for 4-5 min. It was found skin compatible and non – irritant.

11) Antifungal studies

The antifungal activity test was performed by using candida albicans. Then the zone of inhibition was measured in diameter in mm that is the zone of inhibition is 20mm.



Fig.3:-Zone of inhibition

Optimization of Formulation

After analysis of batches of formulation for their evaluation parameters like pH, viscosity, spreadability, extrudability, gelling strength it is observed that the formulation containing equal amount of

betel leaf and Tulsi leaves extract showed good result. These formulation optimized with good pH, viscosity, spreadability, extrudability and gelling strength, hence this formulation is further used for antifungal study (figure 3)

Table 3:-Optimization of formulation

Parameters	Result
Color	Yellowish green
Odor	Characteristics
pH	5.43
Viscosity	4600
Spreadability	9.80
Extrudability	89.2
Gelling strength	25±11

CONCLUSION

In conclusion, the formulation of an herbal gel containing extracts of powdered betel leaf (*Piper betle*) and Tulsi leaves (*Ocimum sanctum* Linn) shows promising potential for the treatment of fungal infections. The combination of these two medicinal plants, known for their antifungal properties, demonstrated significant efficacy in inhibiting fungal growth in vitro. Additionally, the gel formulation exhibited good stability and skin compatibility during the evaluation process.

The study highlights the Importance of exploring natural remedies for fungal infections, considering the increasing prevalence of drug-resistant strains and the side effects associated with conventional antifungal medications.

The herbal gel formulation presents a safer and potentially more sustainable alternative for managing fungal infections, offering a holistic approach to healthcare. Further research is warranted to optimize the formulation and dosage, conduct clinical trials to assess efficacy in human subjects, and explore potential synergistic effects with other herbal ingredients. Overall, the development of herbal formulations such as this gel represents a promising avenue for addressing fungal infections with fewer adverse effects and greater accessibility.

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