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FORMULATION AND EVALUATION OF NATURAL TOPICAL GEL FOR THE TREATMENT OF ACNE VULGARIS

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ABSTRACT

Acne is a disease characterized by inflammatory and non-inflammatory lesions. The pathogenesis includes various factors like hormonal, bacterial and immunological, which causes acne lesions. Propionibacterium acnes and Staphylococcus epidermidis have been recognized as pus-forming bacteria triggering an inflammation in acne. The Present research work deals with the Formulation and evaluation of natural topical gel for the treatment of acne vulgaris containing hydro alcoholic extract of Lantana camara. They contain several medicinal properties that include it benignant-diabetic, antioxidant, antimicrobial, anti- inflammatory, anti-carcinogenic and with hepatic-protective properties. The aim of this study was to overcome the antibiotic resistance and side effects of synthetic drugs by avoiding use of synthetic agents. In vitro antibacterial activity was performed against S. aureus, using agar well diffusion method. The prepared gel were evaluated for pH, viscosity, spreadability, washability, drug content, chemical tests, skin irritancy and in vitro antimicrobial test. The results from the agar well diffusion showed that formulation containing herbal extract of lantana camara would inhibit the growth of S. aureus and gel showed significance antimicrobial activity against bacteria This research work is essential for understanding of all aspects of herbal anti-acne gel.

KEYWORDS: Acne vulgaris, Inflammatory lesions, Lantana camara, Propionibacterium acnes, extract, Antimicrobial assay.

INTRODUCTION

Acne is a common skin condition that happens when hair follicles under the skin become clogged. Sebum oil that helps keep skin from drying out and dead skin cells plug the pores, which leads to outbreaks of lesions, commonly called pimples or zits. Most often, the outbreaks occur on the face but can also appear on the back, chest, and shoulders. Acne is an inflammatory disorder of the skin, which has sebaceous (oil) glands that connects to the hair follicle, which contains a fine hair.[1] In healthy skin, the sebaceous glands make sebum that empties onto the skin surface through the pore, which is an opening in the follicle. Keratinocytes, a type of skin cell, line the follicle. Normally as the body sheds skin cells, the keratinocytes rise to the surface of the skin. When someone has acne, the hair, sebum, and keratinocytes stick together inside the pore. This prevents the keratinocytes from shedding and keeps the sebum from reaching the surface of the skin. The mixture of oil and cells allows bacteria that normally live on the skin to grow in the plugged follicles and cause inflammation swelling, redness, heat, and pain. When the wall of the plugged follicle breaks down, it spills the bacteria, skin cells, and sebum into nearby skin, creating lesions or pimples. For most people, acne tends to go away by the time they reach their thirties, but some people in their forties and fifties continue to have this skin problem. Although it is considered not as a dangerous disease, but infect, almost all Acne sufferers feel disturbed appearance that often leads to lower levels of Confidence and interfere with the daily activities. Acne usually affects everyone at some point in their lifetime.[2] It's most common among teenagers and young adults undergoing hormonal changes, but acne can also occur during adulthood. Adult acne is more common among women and people assigned female at birth (AFAB). You may be more at risk of developing acne if you have a family history of acne (Genetics).

Figure no. 1.1: Leaves and Flowers of lantana camara.

Causes of acne, Clogged hair follicles or pores cause acne. Hair follicles are small tubes that hold a strand of hair. There are several glands that empty into hair follicles.[3] When too much material is inside the hair follicle, a clog occurs. The pores can clog with:

- **Sebum:** An oily substance that provides a protective barrier for your skin.
- **Bacteria:** Small amounts of bacteria naturally live on your skin. If you have too much bacteria, it can clog your pores.
- **Dead skin cells:** Your skin cells shed often to make room for more cells to grow. When your skin releases dead skin cells, they can get stuck in your hair follicles.[4]

For many years, antibiotics have been used to treat acne vulgaris. However, antibiotic resistance has been increasing in prevalence within the dermatologic setting. The development of antibiotic resistance including the specific nature of the relationship of bacteria to antibiotics, how the antibacterial is used, host characteristics, and environmental factors. To overcome the problem of antibiotic resistance, medicinal plants have been extensively studied as alternative treatments for diseases. Many medicines are currently available for the treatment of microbial infection, but most of them are becoming abortive because of the microorganism antimicrobial resistance. To address antimicrobial resistance and side effects, there is an enormous need for the discovery of novel antimicrobial agents.[5] So our aim and objective to develop safe and effective herbal formulation for effective management of acne.

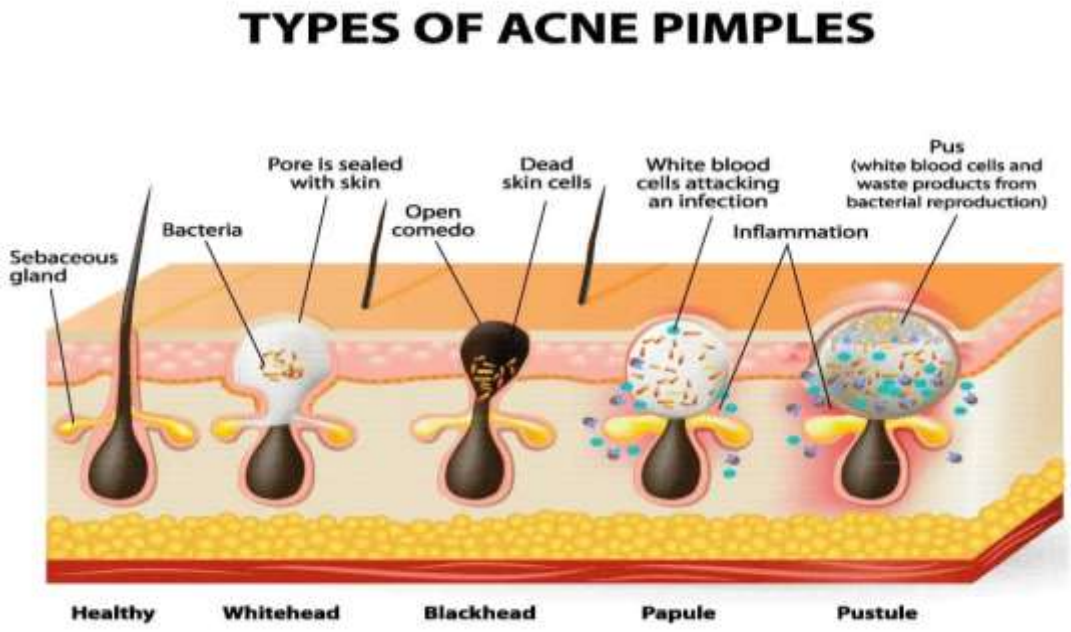

Figure No. 1.2: Type of acne.

Lantana camara, also known as common lantana, shrubverbena, red sage, or yellow sage, is a deciduous shrub native to the West Indies and Mexico to Tropical America. Lantana camara is an adaptable species that can inhabit a wide variety of ecosystems, including between 45ºN and 45ºS and more than 1,400 meters in altitude. It was introduced to Florida in the early 1800s as an ornamental plant, and can be weedy or invasive in areas with more tropical climates. Lantana camara contains 83.91% volatile constituents, including five major constituents in its oil: 28.86% 3,7,11-trimethyl-1,6,10-dodecatriene, 12.28% beta-caryophyllene, 7.63% zingiberene, 7.50% gamma-curcumene, and 3.99% alpha-humulene.[6]

Table No. 1.1: Taxonomic classification.

Category	Botanical description
Domain	Eukarya
Kingdom	Plantae
Phylum	Tracheophyta
Class	Magnoliopsida
Order	Lamiales
Family	Verbenaceae
Genus	Lantanas
Species	Common Lantana

Figure no. 1.3: Pharmacological activities of *lantana camara.*

Uses of *lantana camara*

Lantana camara, also known as wild sage, has many uses in herbal medicine. Here are some uses for lantana camara,

- **Wound healing:** Lantana camara leaves can be used to treat wounds, ulcers and swellings.

- **Fever treatment:** Lantana camara leaves can be used to treat fever, including bilious fevers.
- **Cough treatment:** Lantana camara leaves can be used to treat coughs.
- **Skin itches:** Lantana oil can be used to treat skin itches.
- Antifungal and insecticidal properties: Lantana leaves have antimicrobial, fungicidal, and insecticidal properties.
- **Pectoral:** An infusion of lantana flowers can be used as a pectoral for children.
- **Toothbrush:** Lantana stems can be used as a toothbrush.
- **Wood polishing:** Lantana leaves can be used to polish wood.

Lantana camara has also been used to treat: Influenza, stomach ache, malaria, cancers, chickenpox, measles rheumatism, asthma, high blood pressure, catarrhal infections, and tetanus.[7]

MATERIALS AND METHODS

Selection and Collection of *lantana camara* leaves

When selecting Lantana camara Leaves, look for leaves that are rich in bioactive compounds and have potential therapeutic properties. These leaves should be healthy, Dark green in color, and free from any signs of pests or diseases. It's also important to choose leaves that have been harvested from a clean environment to avoid contamination. *Lantana camara* is a perennial, erect sprawling or scandent, shrub which typically grows to around 2 metres (6+½ feet) tall and form dense thickets in a variety of environments. Under the right conditions, it can scramble up into trees and can grow to 6 m (20 ft) tall.

The leaves are broadly ovate, opposite, and simple and have a strong odour when crushed. *L. camara* has small tubular-shaped flowers, which each have four petals and are arranged in clusters in terminal areas stems. Flowers come in many different colours, including red, yellow, white, pink and orange, which differ depending on location in inflorescences, age, and maturity. The flower has a tutti frutti smell with a peppery undertone. After pollination occurs, the colour of the flowers changes (typically from yellow to orangish, pinkish, or reddish this is believed to be a signal to pollinators that the pre-change colour contains a reward as well as being sexually viable, thus increasing pollination efficiency. In frost-free climates the plant can bloom all year round, especially when the soil is moist.

The native range of *Lantana camara* is Central and South America; however, it has become naturalised in around 60 tropical and sub-tropical countries worldwide. It is found frequently in east and southern Africa, where it occurs at altitudes below 2,000 m (6,600 ft), and often invades previously disturbed areas such as logged forests and areas cleared for agriculture.

Flowers of Lantana camara in West Bengal, India. *L. camara* has also spread across the areas of Africa, Southern Europe, such as Spain and Portugal, and also the Middle East, India, tropical Asia, Australia, New Zealand, and the US, as well as many Atlantic, Pacific and Indian Ocean islands. It has become a significant weed in Sri Lanka after escaping from the Royal Botanical Gardens in 1926. Lantanas were brought to Australia as an ornamental garden plant in 1841, which spread and escaped domestic cultivation and became established in the wild within 20 years. They were brought to India by the British around 200 years ago, which then spread and became invasive there as well.

L. camara leaves were collected from the senior staff quarters of the solankur, Kolhapur, Maharashtra, India in March 2024, and Authenticated by Department of Botany Bhogawati Mahavidyalaya Kurukali, Maharashtra India.

Pre-formulation studies of plant *lantana camara* leaves

Authentication of plant *lantana camara* leaves

This authentication certificate verifies the identity of the plant specimen as Lantana Camara (Red Elder), a flowering plant species belonging to the family Verbenaceae. The plant was identified based on its distinctive morphological characteristics, including its bright red flowers, compound leaves with lance-shaped leaflets, and long, slender fruit pods.

The verification process involved a thorough examination of the plant's physical attributes, comparison with reference materials, and taxonomic research to confirm its identity. This certificate serves as official documentation of the plant's authenticity, providing assurance of its identity for scientific, research, or commercial purposes.

The certificate is issued by [Department of Botany Bhogawati Mahavidyalaya Kurukali], a reputable authority in plant taxonomy and identification, and is signed by [Dr. U.H. Patil Head Department of Botany], a qualified expert in the field. The certificate number and date of issue are also included for reference.

This authentication certificate is an important document for anyone working with Lantana camara, including researchers, botanists, horticulturists, and industry professionals, as it provides a guarantee of the plant's identity and authenticity.

Spectral analysis

UV Spectroscopy

- **Calibration curve**

Stock solution: Take 100mg (0.1gm) Lantana camara leaves extract add into 100ml volumetric flask and makeup volume up to 100 ml with distilled water (Warm if necessary). Shake it properly which makes 1000 ppm solution containing Lantana camara leaves extract.

Then prepare different concentrations solutions (10,20,30,40,50, and 60 ppm) using above prepared stock solution and absorbance of different concentrated solutions detected.

Observation table

Table no. 2.1: Absorbance.

Sr. No.	Sample Name	Absorbance(WL 201.5)
1	10 ppm	0.11
2	20 ppm	0.17
3	30 ppm	0.23
4	40 ppm	0.28
5	50 ppm	0.33
6	60 ppm	0.38

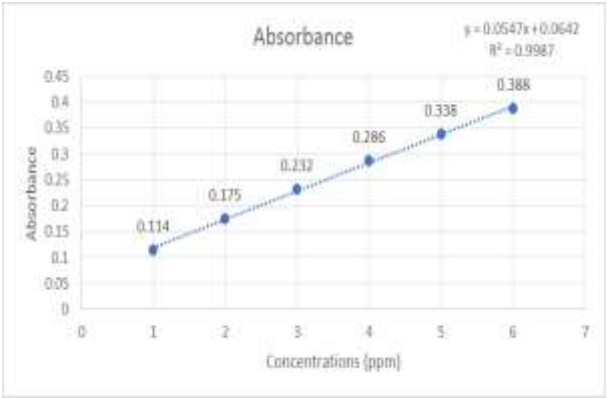

Figure no. 2.1: Graph absorbance vs concentration.

Preparation of Hydro-alcoholic extract of lantana camara leaves

The hydroalcoholic extraction method involves soaking plant material in a solvent, such as ethanol and then evaporating the solvent.

Maceration and sonication technique was used for the preparation of hydro alcoholic extract.

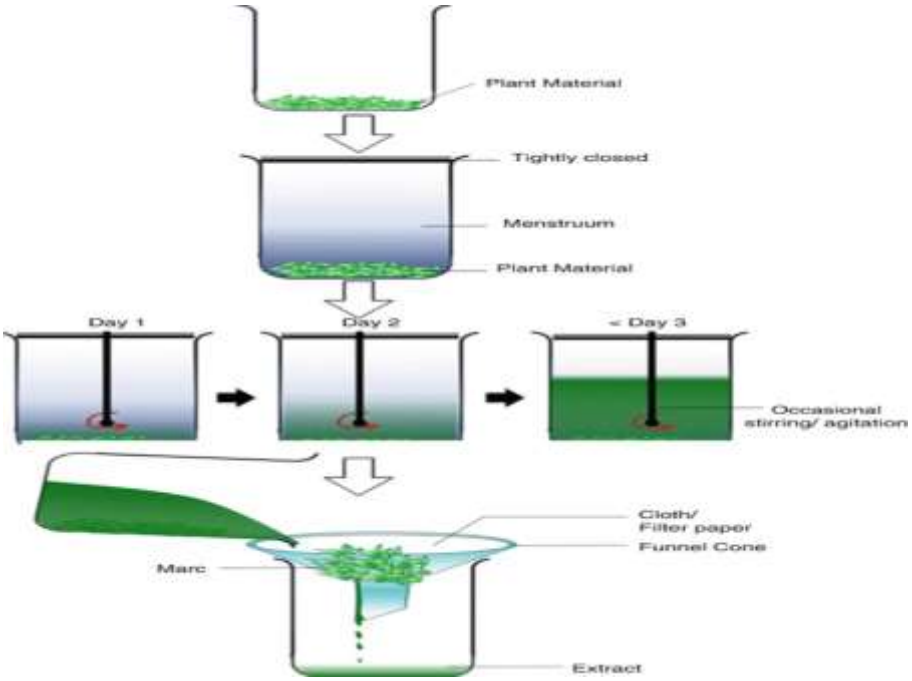

Figure no. 2.2: Extraction steps.

Procedure

1. Before the perform of maceration technique firstly sterilize all the glass wares to avoid cross contamination.
2. Firstly prepared moderately coarse powder from dried leaves of Lantana camara. Here in an extraction process 75% ethanol and remaining will be distilled water was used as a solvent for carried out maceration technique.
3. From moderately coarse powder of Lantana camara took 75 gm powder mixed with 750ml 75% ethanol.
4. After proper mixing of Lantana camara leaves powder and 75% ethanol transported that mixture into closed vessel.
5. That closed vessel containing course powder of Lantana Camara leaves and 75% ethanol that is solvent was allowed to stand for 7 days with shaking occasionally (Magnetic stirrer is used for better mixing).
6. Then liquid strained off. Solid residue (marc) Pressed to recover as much as occluded 75% ethanol.
7. Material included in vessel Clarified by filtration process and keep it for the evaporation process which gave concentrated extract of Lantana camara Leaves.
8. Stored it well with took proper care for avoid cross contamination of an extract of

Lantana camara Leaves.

The percentage of total yield was calculated using the formula below

$$\% \text{ Extraction yield} = \frac{W2-W1}{Wo} \times 100$$

Where

W_0 = The weight of the initial dried sample,

W_1 = The weight of the container alone, and

W_2 = The weight of the extract and the container.[8]

Phytochemical Screening of Hydro-alcoholic extracts of lantana camara leaves

Sample solution: About 100 mg (0.1 gm) of the extract is dissolved in 5 ml of distilled water and filtered. The filtrate is subjected to as an sample solution, used in following tests.

1. Test for alkaloids

a) Mayer's test

Add a few drops of Mayer's reagent to 1 mL of sample solution. A yellowish or white precipitate indicates the presence of alkaloids. For a more specific test, add 2 mL of concentrated HCl to 2 mL of the sample solution, then add a few drops of Mayer's reagent. A white precipitate or green color indicates the presence of alkaloids.

b) Dragendorff's test

Add 1 mL of Dragendorff's reagent to 2 mL of sample solution. An orange red precipitate indicates the presence of alkaloids.

c) Wagner's test

Add a few drops of a solution of 2 g potassium iodide and 1.27 g iodine in 5 mL distilled water, diluted to 100 mL with distilled water, to the sample solution. A brown coloured precipitate indicates the presence of alkaloids.

d) Hager’s test

Treat 2 mL of extract with a few drops of Hager’s reagent. A yellow precipitate indicates the presence of alkaloids.

2. Tests for flavonoids

a) Sodium hydroxide test

Add a few drops of dilute sodium hydroxide to a 1 mL sample solution in a test tube. An intense yellow color will appear, which will turn colorless when a few drops of dilute acid are added.

b) Zinc-HCl reduction test

Add a pinch of zinc dust and a few drops of concentrated hydrochloric acid to 1 mL of sample solution of crude extract in a test tube. A magenta colour indicates the presence of flavonoids.

c) Lead acetate test

Add a few drops of 10% lead acetate solution to 10 mg of extract. A yellow colour precipitate indicates the presence of flavonoids.

d) Shinod's test

Add 10 drops of dilute hydrochloric acid and a piece of magnesium to 1 mL of sample solution. A deep pink colour indicates the presence of flavonoids.

3. Tests for tannins

a) Matchstick test/Catechin test

This test involves warming a matchstick near a flame, which causes the wood to turn pink or red due to the formation of phloroglucinol.

b) Ferric chloride test

A ferric chloride solution to a tannin solution, and a blue, black, violet, or green precipitate or colour will confirm the presence of tannins. Hydrolysable tannins produce a blue–black colour, while a combination of hydrolysable and condensed tannins produce a greenish colour.

c) Gelatin test

This test determines the presence of tannins in a sample solution.

d) Phenazone test

Add 0.5 g of sodium acid phosphate to 5 ml of an aqueous tannin solution, warm, cool, and filter. Then add a 2% phenazone solution to the sample solution, which will precipitate all tannins as a bulky, coloured precipitate.

4. Tests for saponins

a) Ferric chloride test

Two millilitres (2 mL) of the aqueous solution of the extract were added to a few drops of 10% Ferric chloride solution (light yellow). The occurrence of blackish blue colour showed the presence of gallic tannins and a green-blackish colour indicated presence of catechol tannins.

b) Frothing test

Three millilitres (3 mL) of the aqueous solution of the extract were mixed with 10 mL of distilled water in a test-tube. The test-tube was stoppered and shaken vigorously for about 5 min, it was allowed to stand for 30 min and observed for honeycomb froth, which was indicative of the presence of saponins.

c) Foam test

1ml solution of extract was diluted with distilled water to 20 ml and shaken in a graduated cylinder for 15 minutes. Development of stable foam suggests the presence of saponins.

5. Tests for carbohydrates

a) Benedicts test

Concentrated sulfuric acid converts carbohydrates into furfural or its derivatives, which react with α-naphthol to form a purple product. This test is positive for carbohydrates with at least five carbons, as well as proteins and lipids with a linked carbohydrate.

b) Molisch test

A colorimetric test that uses Molisch's reagent to detect carbohydrates Concentrated sulfuric acid in the test converts carbohydrates into furfural or its derivatives, which react with α-naphthol to form a purple product.

c) Fehling's test

Heating a carbohydrate sample with Fehling's reagent turns the reagent from blue to brick red if carbohydrates are present, indicating reducing sugars.

6. Test for anthraquinone glycosides

a) Bromine test

Two ml of the sample solution add an equal volume or an excess of freshly prepared solution of bromine. Record the colour.

b) Modified borntrager's test

Heat an extract with iron chloride and hydrochloric acid, then extract with an organic solvent. A pinkish red colour in the ammonia layer indicates the presence of C-glycosides.[9,13]

OBSERVATIONS

Sr. no.	Phytochemical constituents	Type of test	Result
1	Alkaloids	Mayer's test	+
		Dragandroff's test	+
		Wagner's test	+
		Hager's test	+
2	Flavonoids	Sodium hydroxide test	+
		Zinc-HCl reduction test	+
		Lead acetate test	+
		Shinod's test	+
3	Tannins	Matchstick test/Catechin test	+
		Ferric chloride test	+
		Gelatin test	+
		Phenazone test	+
4	Saponins	Ferric chloride test	+
		Frothing Test	+
		Foam test	+
5	Carbohydrates	Benedicts test	+
		Molisch test	+
		Fehling's test	+
6	Anthraquinone Glycosides	Bromine test	+
		Modified Borntrager's test	+

Excipient compatibility study

a) Carbopol 934

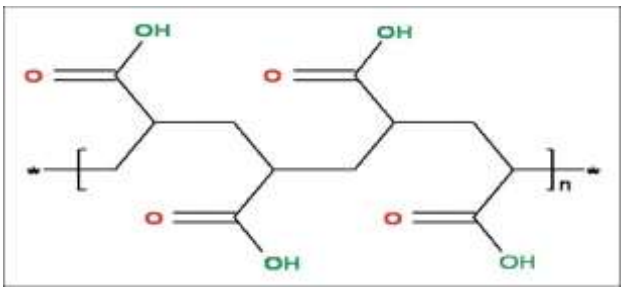

Figure no. 3.1: Structure of carbopol.

Carbopol934 is cross-linked with allyl sucrose and polymerized in benzene. Carbopol 71, 971, and 974 are crosslinked with allyl pentaerythritol and polymerized in ethyl acetate. Carbomer polymers contain 56%–68% of carboxylic acid (– COOH) groups and 0.75%–2% of cross-linking agents.

b) Triethanolamine

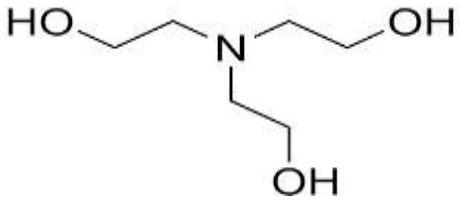

Figure no. 3.2: Structure of triethanolamine.

Triethanolamine (TEA or TEOA) is an oily, viscous organic chemical compound that is a tertiary amine and a triol (a molecule with three alcohol groups). TEA is a bi functional compound that exhibits both properties of alcohols and amines. The compound is used to make surfactants in industrial and cosmetics as a pH adjuster for skin and hair conditioning products as well as sun screen lotions, liquid laundry detergents, polishes, and paints.

c) Methyl parabean

Methylparaben, also methyl paraben, one of the parabens, is a preservative with the chemical formula CH_3. It is the methyle sterof p-hydroxybenzoic acid.

d) Polyethylene glycol

PEG400 is a low-molecular-weight grade of poly ethylene glycol. It is a clear, colorless, viscous liquid Due in part to its low toxicity, PEG 400 is widely used in a variety of pharmaceutical formulations.

Formulation of Anti acne gel containing hydro-alcoholic extract of Lantana camara

Table no. 4.1: Formulation table of Herbal gel containing hydroalcoholic extract of Lantana camara.

Sr. no.	Ingredients	Role	Quantity
1	Carbopol 934	Gelling agent	1 gm
2	Lantana camara Extract	Herbal extract	1 gm
3	Methyl paraben	Preservative	0.3 gm
4	Propyl paraben	Preservative	0.2 gm
5	Triethanolamine (TEA)	pH adjustment, Enhance penetration	q.s
6	Propylene glycol	Improve stability	5 ml
7	Distilled water	-	q.s

Formulation procedure

a) 1 gm of Carbopol 934 was suspended in 50 ml of distilled water in beaker no.1 With continuous stirring.
b) The required quantities of methyl and propyl parabens were dissolved into 5 ml of Distilled water in beaker no 2 by heating on a water bath.
c) Propylene glycol were the need to the cooled solution.
d) Further, the required quantity of extract of lantana camara leaves was added in the Beaker no.2.
e) Then the solution present in beaker no 2 was added in beaker no.1 and volume was made upto 100 ml by distilled water.
f) After mixing all the ingredients, drop wise addition Triethanolamine was made to the formulation for obtaining the desired Consistency of the gel and to adjust the desired skin Ph.[14,15]

Evaluation tests for herbal gel

To evaluate the prepared formulation different tests were Performed.

Physical evaluation of gel

Organoleptic characteristics

- **Color:** The color of the formulation was checked out against white Background.
- **Odour:** The odour of the gel was checked by mixing the gel in Water and taking the smell.
- **Greasiness:** The greasiness was assessed by the application on to the skin.
- **Consistency:** The consistency was checked by applying on skin.
- **Homogeneity:** Homogeneity was tested by visual inspection, They were evaluated for their appearance And presence of aggregates.

P^H Determination: Digital pH meter (Systronics digital-DI-707) was used to determine pH of the prepared formulations and control (base). 3 gm of gel was accurately weighed and dispersed in 30 mL of distilled water and stored for two hours, then pH was measured separately. The measurement of pH of each formulation was done in triplicate and the average values have been represented in result.[16,18]

Spreadability: Two sets of glass slides of standard dimensions were taken. The herbal gel formulation was placed over one of the slides. The other slide was placed on the top of the

gel, such that the gel was sandwiched between the two slides in an area occupied by a distance of 7.5 cm along the slides. Hundred g weight of gel was placed on the upper slides so that the gel was between the two slides was pressed uniformly to form a thin layer. The weight was removed and the excess of gel adhering to the slides was scrapped off. The two slides in position were fixed to a stand without slightest disturbance and in such a way that only upper slides to slip off freely by the force of weight tied on it. A 20 g weight was tied to the upper slide carefully. The time taken for the upper slide to travel the distance of 7.5 cm and separated away from the lower slide under the influence of the weight was noted.[19] The experiment was repeated for three times and the mean time was taken for calculation The dispersion time was calculated from the moment the second glass slide was placed until the gel was completely dispersed between the two lines. The experiment was repeated three times, and the meantime was calculated. The following equation was used to calculate the dispersibility

S= m × l/t

Where, S= spreadability, *m*= weight of the upper slide (110 g), l- the distance of two lines (4 cm), t- time is taken in sec.

Viscosity: Viscosity of gel was determined using Brookfield viscometer (S-62, model LVDV-E) at 25 ^{0}C with a spindle speed of the viscometer rotated at 12 rpm.[20]

Washability: Formulations were applied on the skin and then ease and extent of washing with water were checked manually. All the formulations exhibited good washability and left no traces over the skin on washing with water due to non-greasy properties.[21]

Drug content determination: Drug content of gel was determined by dissolving accurately weighed 1gm of gels in 0.1N NaoH. After suitable dilution absorbance was recorded by using UV- visible spectrophotometer (UV – 1900, Shimadzu, Japan) at 410 nm. Drug content was determined using slope of standard curve. The drug content was determined by using following equation.[22]

Drug content = (absorbance/slope) × dilution factor (1/1000)

Chemical tests

Acid value: 0.5 gm of gel was taken and dissolved in 10 times of absolute alcohol. It was heated on a hot plate for 5 min and 2 to 3 drops of phenolphthalein indicator was added to it and titrated with 0.1 N KOH until faint pink colour appeared.[23]

Acid Value = 56.1 × Titre value × N of KOH/Weight of sample

Peroxide value: In a separate 200 mL flask, 5 gm of gel sample of control (base) and formulations, 30 mL of acetic acid and chloroform solution were added and swirled gently. Then 0.5 mL of potassium iodide solution was added with continuous shaking and 30 mL of water was added thereafter. Finally the solution was then titrated with 0.1 M sodium thiosulfate solution with vigorous shaking. End point of titration was noted when yellow color almost disappears. Then 0.5 mL of 1% starch was added and titration was continued with vigorous shaking to release all iodine from chloroform layer, until blue color disappeared.

Peroxide value = S × M × 1000/gm sample Where S = mL of sodium thiosulfate and M = Molarity of sodium thiosulfate solution.

Total fatty matter determination: 2 gm of gel sample and 20-25 mL of 1:1 dilute HCl was taken into the 200 mL flask, then the solution was heated on a water bath till the solution becomes clear. The sample (Aqueous phase) was drawn in a 250 mL separating funnel and then allowed to cool at room temperature.

50 mL of petroleum ether (Organic phase) was then added in the funnel and shaked and left for separation to occur. The organic phase was collected. The above aqueous layer partitioned twice with same quantity of petroleum ether. The organic layers were collectively evaporated to obtain residue which was consequently washed with water. The residue was filtered and sodium sulfate was added to it. The mixture was again filtered, the extract was dried and the content was determined.[24]

Total fatty matter (%) by mass = 100×M1/M2

Where, M1 = mass of residue; M2 = mass of sample in gram.

Antibacterial activity of polyherbal gel

The antibacterial activity of formulated preparation was performed against Gram-positive (S. aureus NCIM 2654,) bacterial strain by agar well diffusion method with slight modifications.[25,26] The suspension of respective test pathogens was prepared in sterile saline

and used for further study. For the antimicrobial activity test pathogens were inoculated on the surface of sterile Muller and Hinton agar and spread on plates by using a sterile spreader. After that agar well was prepared aseptically with the help of a sterilized cork borer having a 0.7 cm diameter. Then 25 to 100 mg/ml weight of the test sample was added to the different wells of the respective test pathogen. Then plates were placed at 4 °C for 20 min for sample diffusion in a culture medium and transferred to an incubator at 37 °C for 24 hrs. Furthermore, the obtained results were compared with the well containing 1000 mg /ml Streptomycin as a positive control.

RESULTS AND DISCUSSION

Physical evaluation of gel

Table no. 5.1: Observations of organoleptic properties.

Sr. no.	Test	Observations
1	Colour	Dark green
2	Odour	Characteristics
3	Greasiness	Non greasy
4	Consistency	Smooth
5	Homogeneity	Homogenous

pH Determination

It was found to be in the range of 6.62 to 7.08, kept at different storage conditions for 5 days. pH of the formulations and base kept at 8°C for 5 days did not show much change and data were significant over control (base) during one month ($p < 0.03$). Interestingly at 40°C, formulation exhibited elevated change in pH (7.08), while the others remained slightly stable during 5 days study. Data of formulation at 40°C were found to be significant in following table.

Table no. 5.2: pH of the formulation at different storage condition.

Duration	Storage condition	pH
1 day	8 ^{0}C	6.63±0.22
	40 ^{0}C	6.91±0.17
3 days	8 ^{0}C	6.87±0.19
	40 ^{0}C	6.93±0.23
5 days	8 ^{0}C	6.80±0.38
	40 ^{0}C	7.08±0.14

Spreadability

Table no. 5.3: Spreadability.

Test	Observation	Result
Spreadability	9.5 gm.cm/ sec	Easily spreadable

Spreadability of the formulation were studied and found to be 9.5 gm.cm/sec. the formulation were found to possess good spreadability.

Viscosity test

Viscosity and Rheological properties of the formulations were found to be 5.09 Cps. The data of viscosity in formulation were significant temperatures.

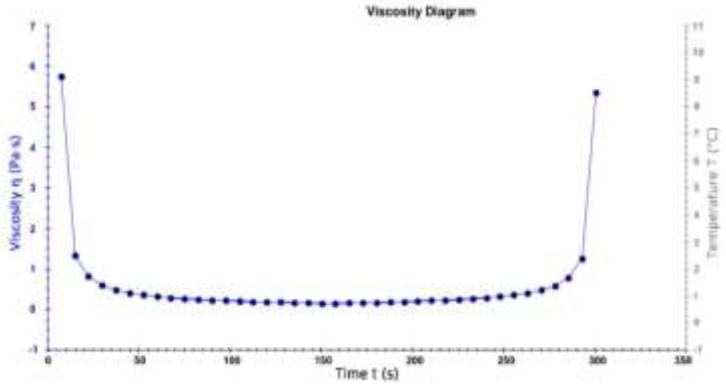

Figure no. 4.1: Graph (Viscosity vs Time).

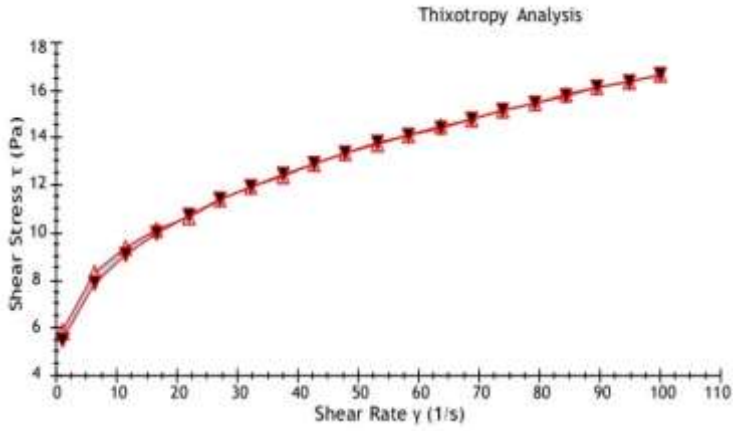

Figure no. 4.2: Graph (Shear stress vs Shear rate).

Washability

Formulation were applied on the skin and then ease and extent of washing with water were checked manually. The formulation exhibited good washability and left no traces over the skin on washing with water due to non-greasy properties.

Drug content determination

The observations of Calibration curve and absorbance of herbal extract of lantana camara leaves. After the analysis of the extract release using UV spectrophotometer (UV – 1900, Shimadzu, Japan) at 410 nm, at dilutions of 10,20,30,40,and 50 it was 60.07%

Chemical tests

Acid value, peroxide value and total fatty matter for the formulations kept at different storage conditions were observed for 15 days and the values for formulation were found within the range. Acid value was found to be in the range of 2.30 to 2.91, peroxide value was found to be in the range of 1.64 to 1.88 and total fatty matters were found to be in the range of 15.01 to 15.5. Peroxide value data in formulations control at 8^0C and 40°C were found to be significant ($p < 0.05$) Total fatty matter data were found to be significant in all except formulation A, B, C and control at 8^0C and 40°C results will be following table.

Table no. 5.4: Calculated values of chemical tests.

Duration	Storage condition	Acid value	Peroxide value	Total fatty matters
5 Days	8^0 C	2.65	1.49	15.55
	40^0 C	2.59	1.53	15.4
10 Days	8^0 C	2.63	1.55	15.34
	40^0 C	2.74	1.62	15.30
15 Days	8^0 C	2.85	1.64	15.2
	40^0 C	2.91	1.67	15.18

Antibacterial activity of gel

Zone of inhibition of control and herbal formulation and control against S. aureus with different dilutions shown in following table.

Table no. 5.5: Zone of inhibition.

Bacteria used	Concentration of sample (mg/ml)	Zone of inhibition (mm)
S. Aureus	25	18.12
	50	20.08
	75	21.85
	100	25.36

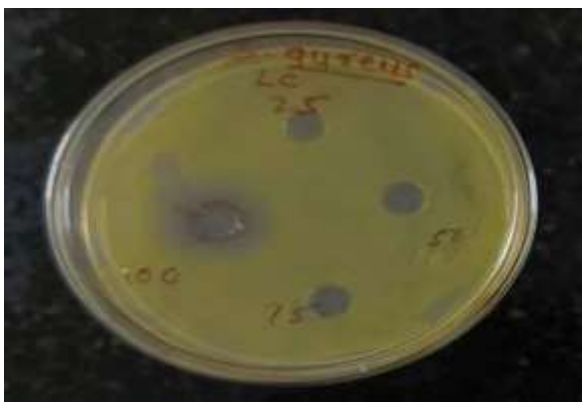

Figure no. 4.3: Zone of inhibition of Control and Herbal formulation against S. Aureus.

Figure no. 4.4: Standard (Streptomycin).

DISCUSSION

Plants are considered to be a vital source of useful constituents for the development of new therapeutic agents, as most of them are safe with less or no side effects. Topical application of gels at pathological sites offer great advantages in a faster release of a drug directly to site of action as compared to cream and ointment. Nowadays, gels have been widely used as a vehicle for topical delivery of drugs. Extracts of plants and herbs with specific medicinal properties can be incorporated in this dosage form as active ingredients in order to additional benefits S. aureus is a common pathogens that can cause skin infections. The antimicrobial properties of Lantana camara plant have been previously investigated on some plant and human pathogens. However, their application and use in the raw form on to the skin surface is difficult therefore the extracts of these plants were developed in the form of gel formulation.

Antimicrobial and antifungal properties of L. camara referred to aurones absorb wavelength between 240–270 and 410–430 nm, respectively, according to the study of Cerovic *et al*.

Carbopol is one of the most ingredients used as the gel bases for the production of antiacne gels. In one study, this compound was used for the preparation a polyherbal antiacne gel, and the results showed that this gel had good consistency and spreadability. Another study is the Mayank *et al*.'s project successfully used this polymer for the formulation a herbal antiacne gel-containing *Aloe vera* extract with suitable appearance, color, consistency, pH, and spreadability.

CONCLUSION

In the present study, we prepared a topical gel formulation containing hydroalcoholic extracts of *Lantana camara*. It showed significant antibacterial effects against *P. acnes* and S. aureus. It is concluded, on the basis of the results obtained in the present analysis, that the herbal formulation of Lantana Camara extracts gel shows satisfactory physicochemical parameters. Herbal cosmetic products are assumed to be safe for longer periods of time. However, quality control for efficacy and safety of herbal cosmetic products is of paramount importance; and quality control tests must therefore be carried out for these preparations. Topical application of gels at pathological sites offer great advantages in a faster release of a drug directly to site of action as compared to cream and ointment. Nowadays, gels have been widely used as a vehicle for topical delivery of drugs.

Thus our research is conclude that *Lantana Camara* shows good antibacterial activity against S. aureus and P. acne hence L. Camara is an best choice for treatment of Acne.

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