

Development and Evaluation of Herbal Cream Incorporating *Cymbopogon citratus* and *Garcinia mangostana* with Antioxidant and Antimicrobial Properties

Ritu¹, Hemangi Gehlot^{*2}

¹Research Intern, Rapture Biotech Pvt. Ltd., Jaipur, India

²Junior Research Scientist, Rapture Biotech Pvt. Ltd., Jaipur, India

^{*}Corresponding author

ABSTRACT: Herbal cosmetics are becoming common in the field of beauty, fashion. Now-a-Days, Herbal Cosmetic is growing rapidly as most people prefer natural products instead of chemicals products. Herbal Cosmetic contains natural nutrients to boost and supply consumer satisfaction because of relatively fewer side effects compared to synthetic cosmetic. The present study is to formulate and evaluate the herbal cream containing extracts of plants such as Lemongrass and Mangosteen, which have highest antioxidant and antimicrobial properties. The herbal formulation was assessed for organoleptic properties, pH, spread ability, homogeneity, irritancy, dilution, sensitivity, and wash ability. The herbal cream was yellow-green, semi-solid, smooth in texture, and had a pleasant fragrance, with pH values ranging from 4.9 to 5.8, within the ideal range for skin compatibility. It exhibited excellent spread ability, homogeneity, and stability, was easily washable, and showed no signs of irritation or sensitivity, confirming dermatological safety. The cream was identified as water-in-oil emulsions, offering enhanced moisturization and prolonged hydration and is safe to use for skin. This study suggests that the composition of extracts and base of cream is stable, safe and natural alternative to synthetic formulations.

KEYWORDS: Herbal Cream, Lemongrass, Mangosteen, Antioxidant, Antimicrobial, Antifungal.

Date of Submission: 12-09-2026

Date of acceptance: 23-09-2026

I. INTRODUCTION

Herbal medicine, also known as botanical medicine or phytomedicine, it refers to the use of any plant's seeds, berries, leaves, roots, bark, fruit peels, or flowers for medicinal purposes [1]. Long practiced outside of conventional medicine, herbalism is becoming more mainstream as up-to-date analysis and research show its value in the treatment and prevention of disease. Recently, the World Health Organization estimated that 80% of people worldwide rely on herbal medicines for some aspect of their primary healthcare [2]. Plant drugs are frequently considered to be less toxic and free from side effects than the synthetic ones [3]. These plants have curative properties due to the presence of various complex chemical substances of different compositions, which are found as secondary plant metabolites in one or more parts of these plants. Along with other dosage forms, herbal drugs are also formulated in the form of ointments and creams. Medicated creams contain a medicament dissolved, suspended, or emulsified in the base. Creams are used topically for several purposes for example, as protectants, antiseptics, emollients, antipruritic and astringents, etc. [5]. Herbal creams offer the added advantage of delivering multiple therapeutic effects through naturally occurring phytochemicals [4]. Medicinal plants used in herbal creams are rich sources of secondary metabolites such as phenolics, flavonoids, tannins, terpenoids, and essential oils, which exhibit antioxidant, antimicrobial, anti-inflammatory, and wound-healing activities. The main aim of our work is to develop a herbal cream which can give multipurpose effect, like moisturizer, reduce acne and skin irritation, reduce skin diseases like eczema, psoriasis, dry skin, wrinkles, rashes etc. and also adding glow to the face.

We used two herbal ingredients in our preparation which are lemongrass and mangosteen. Mangosteen is used as an antifungal and anti-inflammatory, to reduce pimples and acne and also used for maintain skin health, imparting a natural shine as well as for improvement in skin elasticity and skin hydration. Lemongrass is used as a moisturizer, aiding in dead cell removal, slow signs of aging, and it is also used to reduce scar, pigmentation, redness and itching of the skin, skin oil control, and it act as Natural detox for skin and improves skin tone as it has high amount of citral.

II. MATERIALS AND METHODS

Collection of plant material

Lemongrass leaves were collected from a local nursery of Jaipur. Fresh mangosteen fruits were purchased from an exotic fruit shop of Pune market and their peels were taken after washing many times.

Preparation of Ethanolic Extract of Lemongrass and Mangosteen

2gm of lemongrass powder and 2gm of mangosteen rind powder were weighed into 250ml Conical flask and 20ml of extracting solvent were added (96% Ethanol) in each flask. It was placed at maceration shaker for 72 hours, after that solution was filtered using Whatman Filter Paper and filtrate was dried.

Preliminary Phytochemical analysis

The extracts were tested in order to find out the presence of active compounds by use of following standard methods:

1. **Flavonoids test:** 1.0ml of each extract were dissolved in sodium hydroxide (NaOH) solution. The appearance of yellow solution, which disappeared on addition of HCL, indicates the presence of flavonoids [6].
2. **Saponins test:** 2 ml of the solvent extract were put into a test tube with 5 ml of distilled water and shaken for 20 min. The formation of a 2 cm layer of persistent foam on the surface indicates the presence of saponins [7].
3. **Alkaloids Test: Wagner's test:** 1 ml extract was treated with Wagner's reagent (100ul), formation of brown reddish precipitate indicates presence of alkaloids.
4. **Phenolic Content Test:** 1ml of 10% Lead Acetate was added in 1ml of plant extract resulting in formation of white/yellow-white precipitate indicates the presence of phenolic content.
5. **Terpenoids Test: Chloroform test:** To 1mL of the extract few drops of chloroform and conc. H₂SO₄ was added carefully along the sides of the test tube to form a layer and observed for the presence of reddish-brown colour.
6. **Quinones test: HCl Test:** 0.1ml of concentrated HCl was added in 1ml of plant extract, formation of green colour indicates the presence of Quinones.
7. **Tannins test: Gelatine Test:** 1ml of plant extract was added in 4ml of distilled water. 1ml of 1% gelatin solution and 1ml NaCl was added to it, formation of white precipitation indicates the presence of tannins.

Quantitative determination of the total phenolic content

The total phenolic content of the lemongrass and mangosteen was determined by Folin-Ciocalteu method. A standard solution of gallic acid (1mg/ml) was prepared in ethanol. Lemongrass and mangosteen extract was prepared from stock solution of lemongrass and mangosteen. Sodium carbonate (7.5%) was also prepared in distilled water.

Solutions of Gallic acid (0.02, 0.04, 0.06, 0.08 and 0.1ml) or Lemongrass and mangosteen extract (0.02 and 0.05ml) was taken in a different test tube and volume was made up to 1ml with distilled water. 2ml Folin-Ciocalteu reagent were added and incubate for 3min at room temperature after thoroughly mixing the solutions. 4ml of 7.5% Na₂CO₃ solution was added and incubate for 30min at 40°C. The solution was mixed well and the absorbance was measured against a prepared reagent blank at 760nm in UV-VIS spectrophotometer.

Quantitative determination of the total flavonoid content

The total flavonoid content was measured with aluminium chloride colorimetric assay. Lemongrass and Mangosteen extracts and standard solution of quercetin (1mg/ml). 5% sodium nitrite (NaNO₂) solution was prepared in distilled water. 10% aluminium chloride (AlCl₃) solution was prepared in methanol. 1M sodium hydroxide (NaOH) was prepared. Standard solution of quercetin (0.20, 0.40, 0.60, 0.80 and 1ml) and Lemongrass and Mangosteen extract (0.2 and 0.5ml) was taken in different tube and volume was made up to 3ml with distilled water. Then 0.3ml 5% NaNO₂ solution was added and incubate for 5 min in dark, 0.3ml 10% AlCl₃ was added. After 6 min, 2ml of 1M NaOH was added. The solution was mixed well and the absorbance was measured against a prepared reagent blank at 510nm in UV-VIS spectrophotometer.

Determination of Antioxidant activity

Lemongrass and Mangosteen extract were tested by 1,1-diphenyl 2-picrylhydrazyl (DPPH) technique. A total of 24 milligrams of DPPH were dissolved in 100 mL of methanol for making the stock solution. Filtration of DPPH stock solution using methanol yielded a usable mixture with an absorbance of around 0.973 at 517 nm. In a test tube, 3 mL DPPH workable solutions were combined with 100ul of lemongrass and mangosteen extract. In second test tube we made control, 3ml of solution containing DPPH. In third test tube we

make blank ,3ml of methanol. After that, the tubes were kept in complete darkness for 30 min. The absorbance was therefore determined at 517 nm. The following formula was used to compute the percentage of antioxidants.

$$\% \text{ Antioxidant Activity} = [(Ac - As) \div Ac] \times 100$$

Therefore:

Ac = Control reaction absorbance

As = Sample absorbance.[17]

Isolation of skin pathogens

A sterile cotton swab was moistened with sterile saline (NaCl). The Swab was gently and repeatedly rubbed over the selected area of skin. Immediately place the swab into test tube containing 2ml of autoclaved distilled water. Mix it by using vortex mixture. 0.1mL of sample was spread evenly onto agar plate and incubated for 24hours at 37°C temperature. Two distinct colonies from plate were peaked and streaked onto an agar plate with the label skin pathogen1 & 2 (SP1 & SP2) and again incubated for 24hours at 37°C.

Identification of Isolated Bacteria

Identification of the bacteria was done using preliminary biochemical tests such as Gram character, motility, oxidase, catalase and MRVP, Indole and carbohydrate fermentation tests were done.

Biochemical Characterization of Bacterial Isolates

1. Gram Staining: Bacterial smear was prepared on a clean and dry slide then heat fixed. 2-3 drops of gram's iodine were added and incubated for 60 seconds. Then the slide was washed with running tap water and again the slide was washed with 5% ethanol until the crystal violet colour disappeared. The slide was washed gently with running tap water. 2-3 drops of counter stain safranin were added on the slide and incubated for 60 seconds. The slide was washed gently and air dried then observed under a microscope. The gram-positive bacteria appeared violet while gram-negative appeared pink when observed under microscope [15].
2. Motility Test: Hanging Drop Method: A small drop of 24hour old culture was placed on a slide. Petroleum jelly was applied to the four corners of a cover slip to create a seal, prevent drying. Carefully the depression slide was inverted over the coverslip, then flip it over, so the drop hanged in the concavity. Examined under the microscope, actively change in the position of organism indicate motile bacteria[16].
3. Oxidase Test: A small piece of filter paper was dipped in 1% Oxidase reagent and dried. A bacterial colony was added onto it and observation of dark blue/ purple colour indicate the positive test.
4. Citrate Test: Simmons Citrate Agar Slants were prepared in test tubes and sample gently streaked onto it and incubated for 24hours at 37°C. Colour Change from green to blue, indicated the positive test.
5. Carbohydrate Fermentation Broth: That test was helpful in the identification of bacteria that can ferment carbohydrate or those cannot ferment the carbohydrate. Colour change to yellow/purple and potentially gas visible in inverted Durham tube indicated the positive results [8].
6. Methyl Red Test: Bacteria were grown in special MR-VP broth containing glucose and incubated at 37°C for 24-48 hours. After incubation, a few drops of methyl red reagent were added and the colour change was observed. Acid production decreases the medium pH that changes the colour of the methyl red from yellow to red indicate the potential of bacteria to use glucose present in the culture medium [8]. (write procedure)
7. Voges-Proskauer Test: Bacteria were grown in special MR-VP broth containing glucose and incubated at 37°C for 24-48 hours. After incubation, 6-7 drops of alpha-naphthol and 2 drops of 40% KOH were added and incubated for 30 minutes then the colour change was observed. The production of red colour indicates positive test. This was done with the MR Test [8]. (Write procedure not principle)
8. Indole Test: 5ml tryptophan rich broth was prepared and a small amount of pure bacterial culture was inoculated aseptically and incubated for 24 – 48 hours at 37°C. After incubation, Kovac's Reagent was added down the to form a separate layer. The development of red/pink colour within a few minutes was observed[14].
9. Catalase Test: A small bacterial colony was taken on a clean slide; a few drops of 3% H₂O₂ were added. Formation of bubbles on the surface; indicates the positive test.

Antimicrobial Activity of Lemongrass and Mangosteen Extracts against SP1 and SP2:

Nutrient agar media was prepared and autoclaved for the sterilization, then the media was poured on two petri dishes equally in laminar air flow and solidify the media. After solidification, the inoculating sample (100ul) were spreaded onto the plate by using spreader. Then 4wells were formed in plate and mangosteen and lemongrass extract were loaded into the wells respectively (40ul and 80ul) and incubated for 24 hours in an

incubator at 37 °C. The antimicrobial activity was measured by measuring the diameter of the inhibition zone by vernier scale.

Formulation:

Table1: Formulation Table:

<i>Components</i>	<i>Amount (100% w/w)</i>	<i>Role of Components</i>
Lemongrass Extract	25%	Active Ingredients
Mangosteen Extract	25%	Active Ingredients
Coconut Oil	10%	Humectants
Bees Wax	29.4%	Base Ingredients
Borax	0.6%	Emulsifier
Distilled Water	10%	Solvent

1. The composition of herbal cream was shown in table1. The oil consists of coconut oil and other oil soluble component like borax and bees wax were dissolved within the oil phase.
2. The oil phase was placed in a beaker within the water bath.
3. The temperature of water bath was set to 75°C during the heating period.
4. The Water-soluble components and extracts were dissolved in aqueous phase and heated within the same water bath in different beaker at same temperature.
5. After heating, the aqueous phase was added in portions to the oil phase with continuous stirring until the cooling of emulsifiers occurs.

Evaluation of Herbal Cream:

Physical Characteristics: Physical appearance: The formulated herbal cream was inspected visually for their colour, odour, homogeneity and consistency. The developed cream was tested for homogeneity by visual inspection after cream has been set in the container. It was tested for their appearance and presence of any aggregates and the pH of cream was measured [10].

Washability Test: Removal of the applied cream from the skin was conducted by gentle washing under tap water, ensuring minimal force to effectively cleanse the skin [12].

Determination of the type of Emulsion: A crystal violet dye was mixed with the cream. A small amount of cream was placed on microscopic slide and cover with cover slip and observed under a microscope. If the disperse globules appear purple the continual phase colour less, the cream is oil in water (o/w) type. The reverse condition is water in oil (w/o) type cream. It was found that cream is water in oil (w/o) emulsion.

Spreadability: The spreadability was expressed in terms of time in seconds taken by two slides to slip off from the cream, placed in between the slides, under certain load. Lesser the time taken for separation of the two slides better the spread ability. Two sets of glass slides of standard dimensions were taken. Then one slide of suitable dimension was taken and the cream formulation was placed on that slide. Then other slide was placed on the top of the formulation. Then a weight or certain load (70gm) was placed on the upper slide so that the cream between the two slides were pressed uniformly to form a thin layer. Then the weight was removed and excess of formulation adhering to the slides were scrapped off. The upper slide was allowed to slip off freely by the force of weight tied to it. The time taken by the upper slide to slip off was noted.

Spread ability = $m \times l/t$

m = Standard weight which is tied to or placed over the upper slide (70gm)

l = length of a glass slide (7.5cm)

t = time taken in seconds. [9]

Saponification value: About 2 gm of cream refluxed with 25 ml of 0.5 N alcoholic KOH for 30 minutes, to this 1ml of phenolphthalein added and titrated immediately, with 0.5N HCL.

Saponification value = $(b-a) \times M/w$

The volume in ml of titrant = a

The volume in ml of titrating = b

The weight of the substance in gm = w [11].

Test for microbial growth in formulated creams:

The formulated cream was inoculated on the plate of Nutrient agar media by streak plate method. The plate was placed in to the incubator and was incubated at 37°C for 24 hours. After the incubation period, plates were taken out and check the microbial growth by counting the microbial colonies by using colony counter [13].

III. RESULTS

Phytochemical analysis:

The phytochemical analysis of lemongrass extract revealed the presence of alkaloid, phenols, flavonoids, quinones and terpenoids while saponin and tannins were absent. Whereas phenols, alkaloids, flavonoids, terpenoids, were present while saponins, tannins and quinones were absent in mangosteen extract.

Table2: Results of the phytochemical analysis of Ethanolic Extracts

Tests	Lemongrass	Mangosteen
Flavonoid	Positive	Positive
Phenol Content	Positive	Positive
Alkaloids	Positive	Positive
Saponins	Negative	Negative
Terpenoids	Positive	Positive
Tannis	Negative	Negative
Quinines	Positive	Negative

Total flavonoid Content:

Table3: Total Flavonoid Content in Lemongrass and Mangosteen Ethanolic extract:

Ethanolic Extract	Total Flavonoid Content
Lemongrass	0.31mg/ml
Mangosteen	0.26mg/ml

Total Phenolic content:

Table 4: Total phenolic content in Lemongrass and Mangosteen Extract:

Ethanolic Extract	Total Phenolic Content
Lemongrass	0.069mg/ml
Mangosteen	0.468mg/ml

Antioxidant activity:

The results revealed that lemongrass and mangosteen extracts show highest antioxidant activity.

Table5: Antioxidant activity of Lemongrass and Mangosteen with DPPH:

Ethanolic Extract	% Antioxidant
Lemongrass	69.85%
Mangosteen	60.14%

Antimicrobial Activity of Lemongrass and Mangosteen Ethanolic Extract against SP1 and SP2:

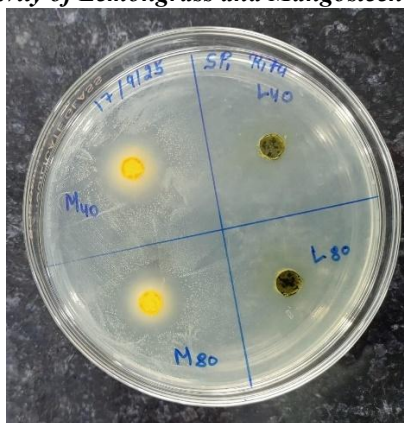


Figure1: Antibacterial activity Against SP1

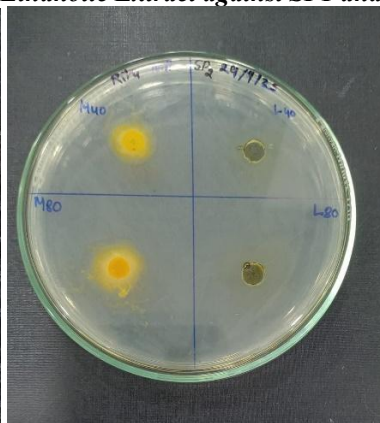


Figure2: Antibacterial activity Against SP2

Table6: Antimicrobial Activity of Lemongrass and Mangosteen Ethanolic Extract against SP1 and SP2:

Bacterial Isolates	SP1		SP2	
	40ul	80ul	40ul	80ul
Lemongrass	17.9	18.38	14.01	15.40
Mangosteen	19.50	22.50	14.74	19.71

Physical Evaluation of Formulated Herbal Cream:

The prepared formulation was evaluated for colour, odour, and consistency. The colour of the cream was observed visually and appeared faint yellow-green. The odour of the cream was pleasant. The cream state was also examined visually and found to be semisolid in nature. The formulation's consistency was assessed by

manually applying and rubbing it on the hand, providing a smooth texture. The results are listed in the below table.



Figure3: Formulated Herbal Cream

Table8: Results of Physical Evaluation of Herbal Cream:

S.no.	Parameter	Cream
1	Colour	Yellow-green
2	Odour	Pleasant
3	State	Semi-solid
4	Consistency	Smooth
5	pH	5.5

Irritability: After the application of the Herbal cream, the irritability test shows no indication of Irritation at all. Results show cream is non-irritant.

Washability: Formulation was applied on the hand and then ease extends of washing with water was checked. Results show, cream is easily washable with water.

Greasiness: The cream was applied to the hand surface as a smear, and its texture was evaluated to determine whether it was oily or greasy. Based on the results, all the formulation was found to be non-greasy.

Determination of pH: The pH of the cream was measured with a pH meter. The pH of the formulation was found slight acidic.

After Feel: The slipperiness and amount of residue left after applying a fixed amount of cream was found to be good.

Microbial Growth Test: The microbial growth test was conducted to determine total viable count (Bacterial and Fungal) in cream formulation. The total Microbial count by standard plate count technique was found to be 315×10^3 cfu/m.

Antimicrobial activity of Herbal Cream against SP1 and SP2

The anti-microbial activity of the formulated cream was screened by well diffusion method and the observations were noted. The antibacterial activity against the skin isolated bacteria was confirmed.

Table9: Antimicrobial Activity of Herbal Cream Against SP1 and SP2:

Skin pathogen	Zone of inhibition
SP1	11.97
SP2	8.67

REFERENCES

- [1]. Schulz, V., Hänsel, R., & Tyler, V. E. (2001). Rational phytotherapy: a physician's guide to herbal medicine. Psychology Press
- [2]. Bannerman, R. H. O., & Burton, J. (1983). Traditional medicine and health care coverage: a reader for health administrators and practitioners. (No Title).
- [3]. Momin, A. (1987). Role of indigenous medicine in primary health care. In 1st International Seminar on Unani Medicine, New Delhi, India (Vol. 54).
- [4]. MV Vishvanathan, PM Unnikrishnan, Kalsuko Komatsu, Hirotooshi Fushimi. A brief introduction to Ayurvedic system of medicine and some of its problems. Indian J Traditional Knowledge 2003;2:159-69.
- [5]. Navindgikar, N. N., Kamalapurkar, K. A., & Chavan, P. S. (2020). Formulation and evaluation of multipurpose herbal cream. International Journal of Current Pharmaceutical Research, 12(3), 25-30.
- [6]. Ewansiha, J. U., Garba, S. A., Mawak, J. D., & Oyewole, O. A. (2012). Antimicrobial activity of *Cymbopogon citratus* (lemon grass) and its phytochemical properties. Frontiers in Science, 2(6), 214-220.
- [7]. Owusu-Ansah, P., Alhassan, A. R., Ayamgama, A. A., Adzaworlu, E. G., Afoakwah, N. A., Mahunu, G. K., & Amagloh, F. K. (2023). Phytochemical analysis, enumeration, isolation, and antimicrobial activity of lemongrass and moringa leaves extracts. Journal of Agriculture and Food Research, 12, 100579.

- [8]. Shoaib, M., Muzammil, I., Hammad, M., Bhutta, Z. A., & Yaseen, I. (2020). A mini-review on commonly used biochemical tests for identification of bacteria. *International Journal of Research Publications*, 54(1), 1-7.
- [9]. El-Gied, A. A. A., Abdelkareem, A. M., & Hamedelniei, E. I. (2015). Investigation of cream and ointment on antimicrobial activity of *Mangifera indica* extract. *Journal of advanced pharmaceutical technology & research*, 6(2), 53-57.
- [10]. Devi, D., Mishra, A., & Mishra, S. (2025). Formulation, Development and Characterization of Herbal Cream. *Asian Journal of Pharmaceutical Research and Development*, 13(4), 42-46.
- [11]. DUSI, S. (2020). Formulation and Evaluation of Aloe vera and *Daucus Carota* herbal cream. *International Journal of Pharmacy Research & Technology (IJPRT)*, 10(1), 31-36.
- [12]. Rakshitha, C., Shalini, B. V., Yogesha, J. S., & Monisha, M. R. (2025). Formulation and Evaluation of Herbal Anti-Aging Cream Containing Natural Bioactive Ingredients: Formulation of Herbal Anti-Aging Cream. *Journal of Advanced Pharmacy and Drug Development*, 1(01), 62-71.
- [13]. Al-Busaid, M. M., Akhtar, M. S., Alam, T., & Shehata, W. A. (2020). Development and evaluation of herbal cream containing Curcumin from *Curcuma longa*. *Pharm Pharmacol Int J*, 8(5), 285-289.
- [14]. MacWilliams, M. P. (2012). Indole test protocol. American Society for Microbiology, Washington, DC, 1-3.
- [15]. Tripathi, N., Zubair, M., & Sapra, A. (2025). Gram staining. In StatPearls [Internet]. StatPearls Publishing.
- [16]. Jain, A., Jain, R., & Jain, S. (2020). Motility testing—hanging drop method and stab. In *Basic Techniques in Biochemistry, Microbiology and Molecular Biology: Principles and Techniques* (pp. 121-122). New York, NY: Springer US.
- [17]. Bag, G. C., Devi, P. G., & Bhaigayabati, T. H. (2015). Assessment of total flavonoid content and antioxidant activity of methanolic rhizome extract of three *Hedychium* species of Manipur valley. *International Journal of Pharmaceutical Sciences Review and Research*, 30(1), 154-159.