FORMULATION AND EVALUATION OF HERBAL ANTIFUNGAL LOTION CONTAINING CURCUMA LONGA AND AZADIRCHTA INDICA

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Abstract
The current work focuses on creating and assessing a natural antifungal lotion with neem oil from *Azadirachta indica* and curcumin from *Curcuma longa*. The study's objective was to create a topical herbal formulation that is stable, safe, and efficacious for treating Candida albicans-caused fungal skin infections. Neem oil, curcumin powder, stearic acid, cetyl alcohol, liquid paraffin, glycerin, sodium lauryl sulfate, methyl paraben, and rose water were among the materials used in the oil-in-water emulsion process to formulate the herbal lotion.

A number of physicochemical properties were assessed for the produced formulation, including color, odor, texture, pH, viscosity, spreadability, homogeneity, washability, skin irritation, absorption, and stability. The lotion showed satisfactory results with good homogeneity, smooth texture, pleasant odor, pH of 6.2, and no signs of skin irritation or phase separation. The agar well diffusion method was used to assess antifungal efficacy against Candida albicans. In contrast to the conventional antifungal medication miconazole, which demonstrated a zone of inhibition of 28 mm, the prepared herbal lotion demonstrated a notable zone of inhibition of 19 mm. The findings show that curcumin and neem oil together have potent antifungal activity and can be a viable natural substitute for synthetic antifungal formulations with fewer adverse effects.

Keywords: Herbal lotion, Curcumin, Neem oil, *Curcuma longa*, *Azadirachta indica*, Antifungal activity, *Candida albicans*, Herbal formulation, Topical preparation, Agar well diffusion method.

INTRODUCTION
In dermatology, superficial fungal infections especially those brought on by the opportunistic pathogen Candida albicans represent a major clinical issue. Millions of people worldwide suffer from candidiasis, which is primarily caused by Candida albicans and affects their mucosal and cutaneous surfaces [1]. An estimated 1.7 billion people worldwide suffer from superficial fungal diseases, mostly dermatophytosis, which pose a serious threat to public health [2].Although synthetic azoles and polyenes play a major role in conventional therapy, these therapies are increasingly linked to serious clinical limits. Long-term usage of synthetic antifungals has been linked to localized allergic dermatitis and the concerning rise of multidrug-resistant (MDR) fungal strains, according to studies. As a result, there is renewed interest in investigating phytochemicals as potential substitutes [3]. The creation of biocompatible substitutes derived from plants has become necessary because to the high frequency of localized skin toxicity [4]. The complex chemical profile of neem (Azadirachta indica) is well known around the world [5]. A powerful bio-control agent against Candida species is neem (Azadirachta indica) seed oil. Bioactive triterpenoids like nimbin and nimbidin, which prevent Candida albicans from changing from yeast to its invasive hyphal form, are largely responsible for its antifungal effectiveness [6]. Because it prevents spore germination and disrupts the permeability of fungal cell membranes, it is highly effective against a variety of dermatophytes.[7]. Neem oil effectively restricts the fungus's capacity to enter deeper skin layers by blocking this transition. Additionally, it is known that components of neem disrupt the formation of ergosterol, which results in the integrity of cell membranes being compromised [5]. Curcumin, the active polyphenol found in turmeric (Curcuma longa), simultaneously attacks Candida cells in multiple ways. According to research, curcumin causes Reactive Oxygen Species (ROS) to build up, which causes C. albicans to undergo programmed cell death, or apoptosis [4]. Curcumin also causes intracellular acidification and subsequent metabolic collapse by interfering with the fungal cell's

H+-ATPase proton pump [8]. These hydrophobic chemicals have strong action, but their limited solubility frequently makes topical distribution difficult. Curcumin powder and neem oil can be effectively solubilized by formulating them into an constant Oil-in-Water (O/W) herbal lotion, guaranteeing prolonged release and enhanced skin permeability [9]. Combining the metabolites of A. indica and C. longa may have a synergistic impact that lowers the necessary concentration of each active, lowers the risk of resistance, inhibits the growth of C. albicans, and has an anti-inflammatory effect to calm diseased tissues [10] . This study's objective is to develop, describe, and assess the in vitro antifungal effectiveness of a polyherbal lotion including curcumin and neem, offering a secure and efficient natural remedy for cutaneous candidiasis.

Herbal body lotion's ideal qualities

1. When applied, the The product must to have cooling properties.
2. Look for a possible emollient effect.
3. Get rid of greasy secretions during the application process.
4. Evenly distribute them over the skin's surface.
5. They must not have any negative impact on the skin.
6. Ensuring compatibility with the skin's pH level is crucial [11].

Benefits of Herbal Body Lotion

1. Use water to moisturize dry skin.
2. Rehydrate your dry, rough skin.
3. Make calluses less uncomfortable.
4. Fourth, satisfy the sensations of touch and smell.
5. Lessen your body's roughness where it is most noticeable.
6. Give your skin a more vibrant appearance [12].

Advantages of Herbal Body Lotion

1. There are no negative side effects or allergic responses.
2. They merge seamlessly with skin and hair.
3. These include more effective in small amounts than other cosmetics.
4. The plant extract form provides suitable pharmacological effects while decreasing the amount of features of the cosmetics.
5. They are more potent, pure, and stable because of to their herbal components.
6. Herbal cosmetics are easy to handle and store for longer periods of time [13].

Benefits of *Curcuma longa*:

1) Curcumin is well praised for its antifungal properties, which help to eradicate infections by preventing ergosterol formation and rupturing fungal cell membranes [14].
2) It functions as a strong anti-seditious agent to accelerate the healing of inflammatory tissues, and it is very successful in lowering skin irritation and greenness [15].
3) Curcumin's antioxidant blends support and shield the skin from oxidative stress and fungal pathogen-induced cellular damage [16].

Benefits of *Azadirchta indica*:

1) Strong azadirachtin mixtures found in neem oil have a cooling effect and stop resistant fungal biofilms from growing on the skin's surface [17].
2) The moisture-locking packages of neem oil ensure that the skin stays moisturized and protected throughout therapy by calming and smoothing diseased tissues [18].
3) It facilitates the distribution of herbal actives into deeper skin layers to target fungal roots by acting as a natural penetration enhancer [19].

MATERIALS AND METHOD

Obtaining and Verification of Plant Material

Obtaining and Verification of Plant Material Fresh, healthy Rhizomes of *Curcuma longa* and Seeds of *Azadirachta indica* were gathered early in the day from a clean, pesticide-free area in the area to guarantee the best possible phytochemical content. This authentication certificate verifies the identity of the plant specimen as Curcuma longa, a herbaceous flowering plant species from the ginger belonging to the family Zingiberaceae . The plant was identified based on its distinctive morphological characteristics, including its Perennial herb , yellow-orange rhizomes , aromatic odor , large oblong green leaves , pale yellow or white flowers. The second plant specimen as Azadirachta indica , a tree species and also a flowering plant species from the mahogany belonging to the family Meliaceae. The plant was identified based on its distinctive morphological characteristics, including its Large evergreen tre , bark is rough and brownish-gray , leaves are pinnate and serrated , white fragrant flowers , fruits are yellow drupes , dark green leaflets.

The physical characteristics of the plant were thoroughly examined as part of the verification procedure, 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 Yashwantrao Chavan College of Science, karad], a reutable authority in plant taxonomy and identification. This authentication certificate is an important document for anyone working with curcuma longa and Azadirachta indica, including researchers , botanists , horticulturists , and industry professionals , as it provides a guarantee of the plant`s identity and authenticity.

Chemicals

The source of cetyl alcohol was Research Lab Fine Chem Industries in Mumbai. The source of liquid paraffin was Research Lab Fine Chem Industries in Mumbai. We purchased sodium lauryl sulphate from Loba Chemie Pvt. Ltd. in Boisar. The source of stearic acid was Research Lab Fine Chem Industries in Mumbai. Research Lab Fine Chem Industries in Mumbai provided the glycerin. Methyl paraben was acquired from Mumbai's Research Lab Fine Chem Industries.[20,21].

METHOD OF EXTRACTION

Curcumin Extraction:

Solid-liquid extraction, commonly referred to as the "maceration" or soaking, is a popular and well-known technique for the standard solvent extraction of solid materials. Curcumin has been extracted from plants using a variety of solvents, such as nonpolar organic solvents and mixtures of organic solvents and water. Popuri and Pagala extracted curcumin from C. longa L. using acetone, ethyl acetone, ethanol, methanol, and isopropanol as solvents. According to the data, ethanol extraction at 30 °C for one hour with an 8:1 sample-to-solid ratio generated the highest yield. Consequently, ethanol was the most favored organic solvent for curcumin extraction. The extraction efficiency is also significantly impacted by the solvent combination's ethanol content.[22].

Neem Oil Extraction:

To obtain neem extracts during the maceration process, 100 g of dried the neem powder containing seeds was weighed and put in many containers with different solvents (methanol, ethanol, acetone, and chloroform).[1:8]. The sample was submerged in the solvent for at least 72 hours. Cold extraction was used because we are aware that heating degrades the nutraceuticals. The solution was filtered after 72 hours, and Nutraceuticals were present in the filtrate. The solvent can be eliminated by vacuum drying or by leaving the container open at room temperature. Any remaining moisture can be removed using a hot air oven set to 50°C. The semi-solid components that are left at the end contain nutraceuticals. [23] .

FORMULATION TABLE

Table No 1: formulation table

Phase	Ingredients	Quantity	Uses
Oil phase (A)	Neem Oil	0.5ml	Active Antifungal
	Curcumin Powder	0.1g	Active Antifungal
	Stearic Acid	0.5g	Emulsifier
	Cetyl Alcohol	0.2g	Texture Enhancer
	Liquid Paraffin	1ml	Base / Emollient
Aqueous phase (B)	Distilled Water	7ml	Vehicle / Base
	Glycerin	0.5ml	Humectant
	Sodium Lauryl Sulfate	0.1g	Surfactant
Cooling Phase (C)	Methyl Paraben	0.02g	Preservative
	Rose water	0.08ml	Masking agent

Uses of ingredients in lotion

1. Cetyl Alcohol :

Cetyl alcohol is widely praised for its remarkable emollient and thickening properties, which are beneficial for enhancing the skin feel and consistency of herbal lotions. It works especially well to stabilize the oil-in-water emulsion, make the formulation easier to spread, and serve as a natural moisture-locking agent.Similar to this, cetyl alcohol contains fatty alcohols that support and smooth the skin barrier, keeping the formulation from feeling oily while guaranteeing a polished, matte finish

- One great source of stabilizing mixes that help to avoid phase separation and improve the lotion's overall texture is cetyl alcohol.
- Because of its superior occlusive effect, it effectively reduces moisture loss from infected skin irritations and encourages a higher level of hydration.
- The mild doses of this fatty alcohol ensure that the antifungal agents are administered smoothly and expertly by calming and smoothing skin surfaces [24].

Fig No 1: cetyl alcohol

2. Sodium Lauryl Sulphate

Sodium lauryl sulphate is well praised for its remarkable surfactant and emulsifying properties, which are beneficial for preserving the consistency of herbal mixtures. It works especially well as a natural cleanser, simplifying the blending of water and oil phases, and lowering surface tension. In a similar vein, it guarantees that the antifungal agents are evenly dispersed to aid in covering and nourishing the skin.

- SLS is a great place to find surface-active mixtures that help create a stable emulsion and ease the application process.
- It ensures that the lotion penetrates deeply into skin irritations by providing an exceptional foaming and spreading effect.

- The formulation is stabilized by the mild doses of this surfactant, which enhance its smoothness and professional appearance [25].

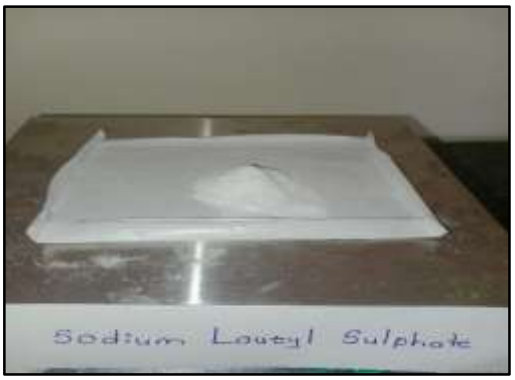

Fig No 2: sodium lauryl sulphate

3. Liquid Paraffin

Liquid paraffin is well praised for its hydrating, calming, and smoothing properties, which are beneficial for dry and sick skin. It works especially well as a natural occlusive barrier, minimizing moisture loss, and relieving skin greenness. Similarly, emollient blends found in liquid paraffin help protect and nourish the skin against environmental irritations.

- Liquid paraffin is a great source of moisturizing mixtures that help reduce skin irritation and inflammation.
- It effectively relieves irritation in sensitive and cracked skin by providing a cooling and protective film effect.
- The mild doses of this mineral oil provide a more hydrated complexion by calming and smoothing skin irritations [24].

Fig No 3: liquid paraffin

4. Methyl Paraben

Because of its remarkable antibacterial and antifungal properties, methyl paraben is highly prized and essential to the durability of herbal treatments. It works especially well at lowering the possibility of microbial contamination, allaying worries about product spoiling, and serving as a barrier. Similarly, it has stabilizing mixes that support the lotion's integrity when it is being stored.

- One great source of preservative mixtures that help stop the formation of germs and mold in water-based formulae is methyl paraben.
- It effectively allays concerns about the shelf-life of delicate herbal compounds since it has a broad-spectrum protective effect.
- The formulation is kept sterile by the mild doses of this preservative, which enhances user safety [26].

5. Stearic acid

As an emulsifier, stearic acid is often used in decorative phrasings to cake and stabilize creams and poultices, as well as to improve the thickness and efficacy of icing.

- Stearic acid, an emollient, helps moisturize the skin, leaving it feeling softer and smoother.

- Stearic acid produces a barrier of defense on the skin's face because of its occlusive properties, preventing TransEpidermal Water Loss (TEWL) and aiding in humidity retention.
- It also supports the skin's natural defensive processes and general health by preserving the pH balance of the skin [27].

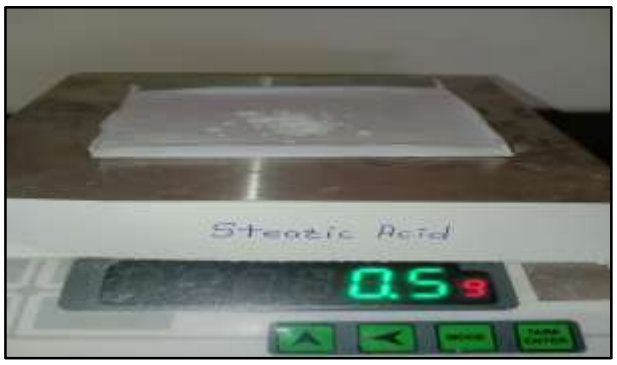

Fig No 4: stearic acid

6. Glycerin

Because of its remarkable moisturizing and hydrating properties, glycerin is highly prized and frequently used in skincare formulas. As a humectant, glycerin keeps the skin's natural moisture barrier intact, ensuring that it stays smooth, supple, and adequately moisturized.

- Glycerin is a powerful moisturizer that draws and holds onto moisture in the skin to prevent dryness.
- It helps replenish comfort and hydration by calming dry and irritated skin. By keeping skin hydrated and encouraging skin healing, glycerin helps treat acne and reduce the appearance of scars.
- Because it improves skin suppleness and moisture retention, it helps minimize the appearance of fine lines and wrinkles [28].

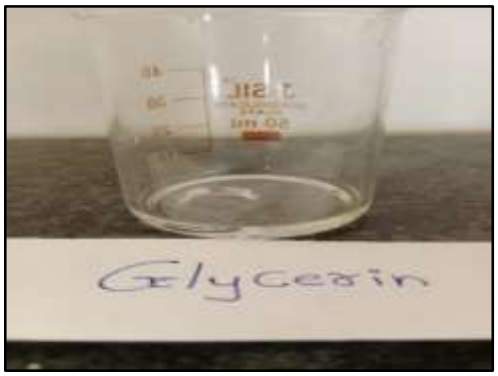

Fig No 5: glycerin

7. Rose water

Rose water is often praised for its energizing, calming, and reassuring properties that are beneficial for a variety of skin conditions. It works very well to ease greenness, lessen irritation, and function as a natural hue. Similarly, rose water has antibacterial and antioxidant combinations that help protect and nourish the skin.

- Rose water is a great source of anti-seditious mixtures that help ease irritation and reduce skin inflammation.
- Because of its cooling action, it works well to reduce greenness and itching, especially on sensitive skin.
- The delicate drops of rose water promote a more radiant complexion by calming and smoothing skin irritations [28]

FORMULATION OF LOTION

1. Prepare the Aqueous Phase:

First, weigh out 7 milliliters of pure water, 0.5 grams of glycerin, and 0.1 grams of sodium lauryl sulphate into a 250 milliliter beaker. Mix everything together and place the beaker into water bath to heat upto 70-75^{0}C to generate the Aqueous phase.

2. Prepare the Oil Phase:

Get ready for the oil phase by measuring out 0.5 grams of stearic acid and putting it in a different plate. Add additional oil-based substances such as 0.5ml Neem Oil, 0.1g Curcumin Powder, 0.2g Cetyl Alcohol, and 1g Liquid Paraffin.Then, while stirring to create the oil phase.

3. Combine the Phases:

When the oil and water phases are prepared, Add oil phase into Aqueous phase. gently mix them together by stirring. Continuously stir at 40^0 C until yellow color is appear .The herbal lotion mixture will be produced as a result.

4. Final Product:

Allow the lotion to cool and solidify after thoroughly mixing. You can now apply your herbal body lotion [29].

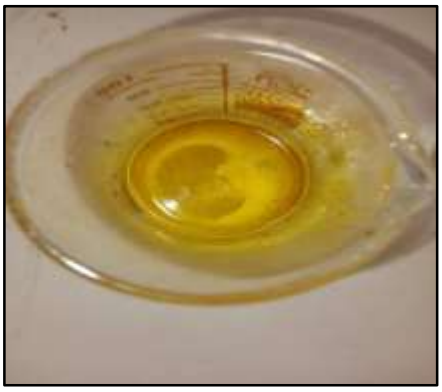

Fig No 6: oil phase

Fig No 7: aqueous phase

Fig No 8: herbal lotion

EVALUATION OF LOTION

1) Organoleptic Properties:

The herbal lotion's organoleptic qualities are assessed using visual examination techniques. This evaluation looked at color, odor, texture, and condition [12].

a) Color:

The lotion's color is examined to ensure that it matches the product's design and appears as intended [29].

b) Odor:

The lotion is tested to make sure it smells well and doesn't contain any overpowering or disagreeable smells [29].

2) Appearance:

To ensure that the lotion appears clear, smooth, and free of flaws like lumps or clumps, it is visually inspected [29].

3) Test for Uniformity:
This test determines whether the lotion has a uniform texture and doesn't separate or lump. It is examined both physically and visually to make sure it is level and smooth [29].

4) Washability:
The hand was subjected to 10 minutes of forceful flowing tap water after a little amount of lotion was applied. When the lotion is completely gone, it will be noted. Look: The lotion's color, aroma, and consistency were evaluated visually [30].

5) Viscosity:
The LV-64 spindle will be used to measure viscosity using a Brookfield viscometer. A rotation speed of 25 revolutions per minute will be chosen. To determine its viscosity, the prepared lotion will be immediately immersed in the spindle [31].

6) Homogeneity test:
Visual inspection and touch were used to evaluate the homogeneity test [32].

7) Test for Absorption:
This test determines how thoroughly the lotion penetrates the skin. To gauge how quickly and well the lotion is absorbed, it is applied to the skin and gently rubbed in [28].

8) Test for Skin Irritation:
A formulation was placed to the back of the hand and left for 15 minutes in order to gauge skin irritation reactions such swelling, itching, and redness. [32].

9) pH Test:
To ensure that the herbal embrocation falls within the skin-friendly range, typically between 4.5 and 7, to alleviate an implicit skin irritation, the pH position was measured using a digital pH cadence [28].

10) Smoothness:
We assessed the lotion's smoothness through touch examination by rubbing it between their fingers and noting its texture. We recorded if the lotion felt smooth, homogeneous, clumpy, or harsh. [33].

11) Spread ability:
This test determines the lotion's spreadability on the skin. To test how well a sample moves and spreads, it is sandwiched between two glass slides and weighted [29].

12) Stability Test:
The prepared lotion was filled in a container and stored at room temperature, Refrigerated condition and accelerated condition. The formulation was stored for 30 days in each condition. Samples were observed at regular intervals at (0,7,14,30) [29] .

Antifungal Activity

The agar well diffusion method, a commonly used in vitro approach for assessing the antifungal efficiency of topical formulations, was used to assess the antifungal activity of the prepared herbal antifungal lotion containing curcumin and neem oil against Candida albicans (ATCC 10231). Since Candida albicans is one of the most prevalent pathogenic fungi that cause a variety of fungal infections of the skin and mucosa, it was chosen as the test organism for this investigation. The fungal strain was cultivated and maintained using Sabouraud Dextrose Agar (SDA) media. Clean, sterile Petri dishes were filled with around 15 mL of sterile molten SDA medium, which was then left to solidify aseptically. To guarantee that the fungal inoculum was distributed evenly, 100 µL of freshly made fungal broth culture was evenly distributed over the agar medium's surface using a sterile spreader after solidification. A sterile cork borer was used to gently create 6 mm-diameter wells in the agar plates. Dimethyl Sulfoxide (DMSO) was used as the solvent to create the test

sample solution of the produced herbal body lotion at a concentration of 100 µL/mL. Each well received around 100 µL of the prepared test solution. DMSO was utilized as the negative control to ensure that the solvent itself had no antifungal effect, and miconazole (1 mg/mL), a common antifungal medication, was employed as the positive control for comparison. After that, the infected plates were incubated for a full day at 37°C. Following incubation, the diameter of the zone of inhibition—a clear area that formed around each well—was measured in millimetres (mm) to determine the antifungal activity. Stronger antifungal activity of the formulation is indicated by a greater zone of inhibition. To guarantee precision, repeatability, and dependability of the findings, every experiment was conducted in triplicate. The efficacy of the herbal lotion was assessed by comparing its observed antifungal activity with that of the conventional medication. The combined therapeutic benefits of curcumin and neem oil, which are both well-known for their robust antibacterial and antifungal qualities, showed that the formulation has substantial antifungal potential.[34,35].

RESULTS

A] Evaluation of lotion:

SR.NO	TEST	RESULT
1	Color	Bright yellow
2	Odor	Pleasant characteristic odor
3	Texture	Smooth and Creamy
4	State	Semi-solid
5	pH	6.2
6	Skin Irritancy test	No irritation observed
7	Absorption test	Easily absorbed into skin
8	Homogeneity	Good homogeneity with no phase separation
9	Washability	Easily washable with water
10	Viscosity	5800 cP
11	Smoothness	Good smoothness
12	Spread-ability Test	Smooth and light to spread
13	Uniformity	Uniform distribution of ingredients
14	Stability	Formulation remained stable with no phase separation , color change, or odor change during the study period

Table No 2: evaluation parameter of lotion

B] Result of Antifungal Activity

The herbal body lotion shown strong inhibitory effect against Candida albicans, according to the antifungal activity results. The lack of antifungal activity in the negative control was confirmed by the control sample, which displayed no zone of inhibition (0 mm). Strong antifungal efficacy was demonstrated by the conventional medication miconazole, which had the largest zone of inhibition of 28 mm. With a zone of inhibition of 19 mm, the herbal body lotion showed good antifungal activity against Candida albicans.

Table No 3: antifungal activity of test samples against *c. albicans*

SR.NO	SAMPLE CODE	ZONE IN DIAMETER (mm)
1	Control	00
2	Standard (Miconazole)	28
3	**Body Lotion**	19

Fig No 9: zone of inhibition against *candida albicans*

DISCUSSION

The current study demonstrates the significant antifungal effectiveness of the herbal antifungal lotion including neem oil and curcumin against Candida albicans. Fungal growth is significantly inhibited by the formulation, as evidenced by the observed zone of inhibition of 19 mm. The herbal lotion showed encouraging results even if its effectiveness was somewhat lower than that of the common antifungal medication miconazole (28 mm).While neem oil has potent antibacterial and antifungal action because it contains bioactive components such azadirachtin, nimbin, and gedunin, curcumin is well known for its antifungal, anti-inflammatory, and antioxidant qualities. The formulation's increased antifungal efficacy could be attributed to the curcumin and neem oil's synergistic action.The control sample's lack of inhibition indicates that the active herbal constituents, not the solvent system, were responsible for the activity. As a result, the composition might be a more natural and safe substitute for synthetic antifungal medicines with fewer adverse effects.

CONCLUSION

The current study created and assessed a herbal antifungal lotion with neem oil and curcumin.The prepared formulation showed satisfactory physicochemical characteristics such as smooth texture, good homogeneity, pleasant odor, appropriate pH, good spreadability, easy washability, and stability without any phase separation or skin irritation. The antifungal activity study demonstrated that the herbal lotion possessed significant inhibitory activity against *Candida albicans*, showing a considerable zone of inhibition when compared with the standard antifungal drug miconazole. Curcumin and neem oil, which are both rich in bioactive chemicals with antifungal, anti-inflammatory, antioxidant, and antibacterial qualities, may work in concert to produce the antifungal effect.. The formulation not only exhibited effective antifungal activity but also provided good skin compatibility and moisturizing properties, making it suitable for topical application. Therefore, for the treatment of superficial fungal infections, the proposed herbal antifungal lotion might be regarded as a promising natural substitute for synthetic antifungal medicines. To determine its medicinal efficacy and commercial applicability, additional research, such as advanced stability tests, toxicity evaluation, and clinical trials, may be conducted.

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