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# In-Vitro Antimicrobial Activity and Phytochemical Screening of Ethanolic Fruit Extract of Luffa aegyptiaca Against Selected Skin-Associated Pathogens

**Enass El Sharkawy Ali** *[1]

[1] *Cairo Fatemic Hospital, Old Cairo Medical District, Cairo, Egypt*

*Corresponding Author: *Enass El Sharkawy Ali, Email (* enaselsharkawy550@gmail.com )

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## ABSTRACT

**Background:** Skin infections caused by microorganisms are a great clinical and public health problem, especially because antimicrobial resistance is increasing and there is a need for safer natural antimicrobial agents. *Luffa aegyptiaca* It is traditional bathing with a natural sponge and is expected to contain bioactive phytochemical constituents that may act as an antimicrobial agent. **Aim:** This study is looking forward to evaluate the qualitative phytochemical profile and in-vitro antimicrobial effects of the crude ethanolic fruit extract of *Luffa aegyptiaca* against selected skin-associated pathogens, including *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Staphylococcus aureus*, and *Candida albicans*. **Methods:** Dried fruits of *Luffa aegyptiaca* were extracted using hydro-alcohol by maceration. The crude extract was subjected to phytochemical screening to detect secondary metabolites classes. Antimicrobial activity was assessed using the agar well diffusion method at three extract concentrations: $0.001, 0.002$, and $0.003\ mg/mL$. The diameter of the inhibition zone is measured in mm, and a negative control was included to confirm the absence of antimicrobial activity from the diluent. **Results:** Phytochemical screening showed that the presence of terpenoids/steroids, tannins, alkaloids, saponins, cardiac glycosides, and flavonoids. The ethanolic extract exhibited concentration-dependent antimicrobial activity against the tested microorganisms. At 0.003 mg/mL, inhibition zones of 20 mm were recorded against *S. aureus*, *K. pneumoniae*, and *P. aeruginosa*, while *C. albicans* showed the highest inhibition zone of 22 mm. At 0.002 mg/mL, the inhibition zones were 13 mm, 14 mm, and 13 mm against *S. aureus*, *Klebsiella pneumoniae*, and *P. aeruginosa*, respectively, and 20 mm against *C. albicans*. No detectable inhibition was observed against the bacterial isolates at 0.001 mg/mL, whereas *Candida albicans* showed an inhibition zone of 15 mm. The negative control showed no inhibition. **Conclusion:** The crude ethanolic fruit extract of *L. aegyptiaca* demonstrated preliminary antibacterial and antifungal activity against selected skin-associated pathogens, with the greatest activity observed at the highest tested concentration. The antimicrobial effect may be linked with the presence of multiple bioactive phytochemical constituents detected in the extract. Further studies, including MIC, MBC or MFC, compound characterization, and cytotoxicity assessment, are required to confirm the potential use of Luffa *aegyptiaca* extract in topical pharmaceutical and dermatological formulations.

## 1. INTRODUCTION

The skin -the largest organ in the human body- which serves as a barrier against environmental and microbial threats [1]. The skin hosts a large group of microorganisms called skin microbiota, that may be bacteria, fungi, and viruses. Some of these microorganisms are either benign or beneficial, that can be opportunistic pathogens in certain situations like immuno-compromised patients, poor hygiene, or skin damage [2]. The previous studies suggest that it is essential to keep good skin hygiene to preserve microbial balance and prevent infections [3]. Antimicrobial compounds are used always in personal hygiene products like soaps, creams, and sanitizers to kill skin microbes [4]. These compounds act as infections preventive, body odor management, and enhancement of overall skin wellness as these are crucial medical technique for skin health [5] The use of synthetic antimicrobial agents may induce medical health concerns. Many health concerns have been raised including antimicrobial resistance, skin irritation and

allergic reactions, and worries about environmental toxicity. Also, prolonged synthetic antimicrobial compounds can overthrow the skin natural microbiome balance that is essential for healthy skin [6] There is an elevated interest in natural antimicrobial substances derived from plants, animals and fungi because of their safety, environmentally safe, and low adverse effects [7]. Medicinal plants synthesize a vast diversity of bioactive compounds like flavonoids, tannins, alkaloids, and phenolics, steroids that is proven to have antimicrobial actions. Herbal formulations are progressively utilized in beauty products and medicines as substitutes for chemical agents [8] Luffa aegyptiaca is a Cucurbitaceae family member grown in subtropical and tropical areas. The fruit is a dry, fibrous structure when it is ripe hosting the seeds, acting as a good natural bathing sponge. Luffa aegyptiaca has been used for vast medicinal implications, mainly skin cleansing, hygiene, dermatological conditions and infections treatments [9]. Phytochemical studies of Luffa aegyptiaca revealed many compounds, such as flavonoids, alkaloids, saponins, tannins, glycosides, terpenoids, and phenolic compounds [10]. Prior research has reported that Luffa aegyptiaca extracts may show antimicrobial properties against various microorganisms, including S. aureus, E. coli, P. aeruginosa, and C. albicans [11]. The L. aegyptiaca ethanolic extracts have demonstrated promising antimicrobial actions particularly against Gram-positive bacteria [12] The sun-dried fibrous fruit of L. aegyptiaca is widely used as a natural sponge as its fibrous structure removes of dead skin cells, promotes exfoliation, and suggested to enhance local blood circulation. In addition to the mechanical cleansing action, L. aegyptiaca may possess antimicrobial properties [13]. Even though it's very common use, its antimicrobial potential has not been sufficiently investigated, particularly in relation to possible pharmaceutical and dermatological applications.

## 2. MATERIALS AND METHODOS

### Study Design

This study was designed as an in-vitro investigation to assess the compounds classes and the antimicrobial effects of ethanolic extract of L. aegyptiaca dry fruit in relation to four main skin pathogens S. aureus, K. pneumoniae, P. aeruginosa, and C. albicans. The concentration of the extract was $(0.003, 0.002, 0.001\ mg/mL)$ [7] The measurement of the inhibition zone diameter (µm) is the main variable. The inhibition zone diameter that was kept constant in the culture medium, temperature, incubation duration, inoculum volume, and type of disk.

### PLANT COLLECTION AND IDENTIFICATION

Ripe dry fruits of L. aegyptiaca were collected at a market in Najaf on January 5, 2026.

### Extract Preparation

Fruits were shade dried at a temperature of $25 \pm 2°C$ for a duration of 14 days, then after the fruits were communed into powder using an electric grinder [13]. 100 g powder soaked in $500\ mL$ (70% ethanol) and sealed in a glass vessel for 72 hours at room temperature. Continuous stirring occurs every 8 hours. The resulting mixture filtered via filter paper Whatman No. 1. The extract was conc. by a rotary evaporator at $40°C$ under reduced pressure then was air-dried yielding 7% dry extract. A stock solution in in 10% DMSO was prepared at a serial concentration of $0.003, 0.002$, and $0.001\ mg/mL$.

### Phytochemical Screening

Phytochemical screening tests were conducted:
Flavonoids ($NaoH$): 1 mL of the *L. aegyptiaca* extract, a few drops of $NaoH$ solution were added. The yellow color that disappears after addition of Dil HCl indicate the presence of flavonoids[14]. Saponins (Froth test): 0.5 gm of *L. aegyptiaca* extract combined with 10 mls of distilled water and shaken if persistent foam over 1 centimeter for at least 15 minutes then the test is considered positive [15] Tannins (Ferric chloride test): 1 ml of *L. aegyptiaca* extract is mixed with 2 to 3 drops of 5% ferric chloride and the bluish black or the olive green indicated the tannins presence[16]. Cardiac glycosides (Keller-Killiani): The combination of 0.5 gm of *L. aegyptiaca* extract with 2 ml of glacial acetic acid, 1 drop of 5% ferric chloride, and 1 ml of conc. sulfuric acid. The cardiac glycosides is present if a brown layer and a blue-green ring is formed [17]. Alkaloids (Dragendorff): Adding 1 ml of extract along with 2 to 3 drops of reagent creates a precipitate, indicating a positive result [18]. Terpenoids/steroids (Salkowski): 0.5 $gm\ of\ L.$ *aegyptiaca* extract added to 2 $mls$ of chloroform and 1 ml of conc. sulfuric acid results in a red coloration at the interface, confirming a positive test [19]

## Microorganisms

*S. aureus, K. pneumoniae, P. aeruginosa, and C. albicans* were used in this study. *L. aegyptiaca* crude extract was evaluated for its antimicrobial activity. The microbial strains were obtained from Al-Amin Research Center. The bacterial isolates were sub-cultured and activated on Mueller–Hinton agar. *C. albicans*. were activated using Sabouraud Dextrose Agar (SDA). All cultures were incubated at $37°C$ for 24 hours till antimicrobial susceptibility testing.

### SAMPLE

1. Control vehicle for application of the extract: The Negative control was the diluting agent (DMSO)to confirm the method validity[20]
2. Crude extract: Crude Total Extract *L. aegyptiaca* crude extract illustrates the entire phytochemical composition of *L. aegyptiaca* such as Flavonoids, Alkaloids, Saponins, Tannins, Terpenoids, and Cardiac glycosides

### CULTURE MEDIA

MHA was used in this study as a standard medium for the cultivation and antimicrobial testing of bacterial skin pathogens, including S. aureus, K. pneumoniae, and P.

aeruginosa [21] MHA provided good and reproducible results in assessing the inhibitory effects of the plant extract against the selected pathogens [16]. SDA served as a specialized medium in this research for the identification and growth of Candida species linked to skin infections [17] it offers a proper environment to evaluate the antifungal efficacy of L. aegyptiaca ethanolic extracts [17]

### ANTIMICROBIAL WELL DIFFUSION METHOD

The antimicrobial activity of the total extract of L. aegyptiaca was determined by the well diffusion method [10] Sterile agar plates were inoculated with the tested microorganisms under aseptic conditions. Wells containing different concentrations of the extract were then placed on the inoculated agar surface [12] The tested concentrations were:

- 0.001
- 0.002
- 0.003

A control was also included in the experiment. After incubation under suitable conditions, the antimicrobial activity was assessed by measuring the diameter of the inhibition zone around each well in millimeters (mm). A larger inhibition zone indicated greater antimicrobial activity of the extract [12]

### Interpretation of results

The antimicrobial activity was evaluated by comparing the inhibition zones produced by the three tested concentrations against each microorganism. The symbol R was considered to indicate resistance or absence of detectable inhibition.

## 3. RESULTS

### Phytochemical screening of the ethanolic extract of L. aegyptiaca fruit

The qualitative phytochemical screening of the ethanolic extract of L. aegyptiaca fruit revealed the presence of several important secondary metabolites, as presented in Table 1.

***Table 1:*** *Phytochemical screening of ethanolic extract of L. aegyptiaca fruit.*

| Test | Phytochemical constituent | Observation | Result |
|---|---|---|---|
| Alkaline reagent test (NaOH test) | Flavonoids | Formation of an intense yellow color after addition of sodium hydroxide, which becomes colorless upon addition of dilute acid | Positive (+) |
| Froth test | Saponins | Formation of stable persistent froth exceeding 1 cm in height for at least 15 minutes | Positive (+) |
| Ferric chloride test | Tannins | Greenish-black or bluish-black color with ferric chloride solution | Positive (+) |
| Keller–***Killiani*** test | Cardiac glycosides | Brown ring at the interface with bluish-green color in the upper layer | Positive (+) |
| ***Dragendor*** test | Alkaloids | Orange or reddish-brown precipitate on addition of *Dragendorff's* | Positive (+) |
| Salkowski test | Terpenoids / steroids | Reddish-brown coloration at the interface on addition of conc. sulfuric acid | Positive (+) |

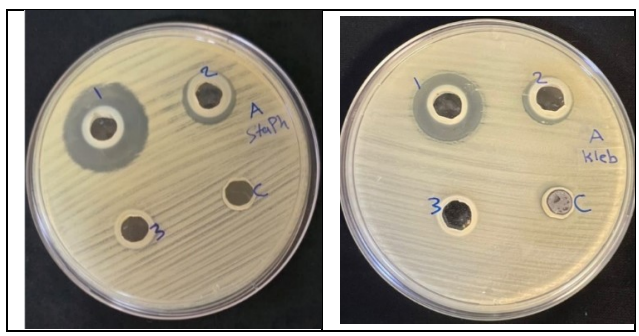
The detection of saponins, flavonoids, tannins, cardiac glycosides, alkaloids, and terpenoids/steroids suggest that the extract contains biologically active compounds that may contribute to its antimicrobial activity.

### ANTIMICROBIAL ACTIVITY RESULTS

The total crude extract of L. aegyptiaca demonstrated antimicrobial activity against the tested microorganisms, and the activity increased with increasing concentration. In general, the highest antimicrobial activity was observed at concentration $0.003\,mg/ml$, followed by $0.002\,mg/ml$, while $0.001\,mg/ml$ showed either weak activity or no activity. The control showed no inhibition zone in all tested organisms. L. aegyptiaca total extract, the inhibition zones against S. aureus, K. pneumoniae, and P. aeruginosa were $20\,mm$, $20\,mm$, *and* $20\,mm$, respectively, at concentration $0.003\,mg/ml$. At concentration 0.002 mg/ml, the inhibition zones were 13 mm, 14 mm, and 13 mm, respectively, whereas concentration 0.001 mg/ml showed resistance. Against C. albicans, L. aegyptiaca total extract showed inhibition zones of 15 mm at $0.001\,mg/ml$, $20\,mm$ at $0.002\,mg/ml$, *and* $22\,mm$ *at* $0.003\,mg/ml$, while the control showed no inhibition.

***Table 2:*** *Antimicrobial activity of L. aegyptiaca total extract against microorganisms of selection expressed as inhibition zone diameter (mm).*

| Sample | Microorganism | Control (mm) | 0.001 mg/ml | 0.002 mg/ml | 0.003 mg/ml |
|---|---|---|---|---|---|
| A | *S. aureus* | 0 | R | 13 | 20 |
| A | *K. pneumoniae* | 0 | R | 14 | 20 |
| A | *P. aeruginosa* | 0 | R | 13 | 20 |
| A | *C. albicans* | 0 | 15 | 20 | 22 |



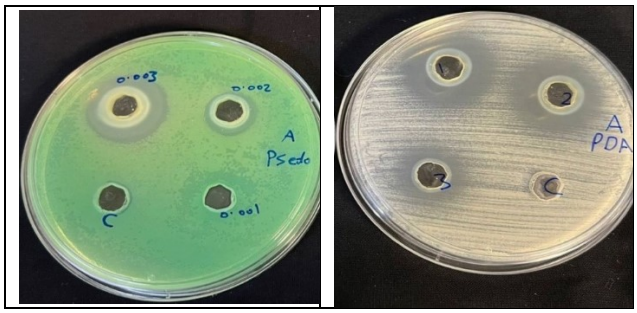



***Figure 1:*** *Antimicrobial effects of L. aegyptiaca extract on S. aureus, P. aeruginosa, K. pneumoniae, C. albicans.*

## 4. DISCUSSION

The present study demonstrated that the ethanolic fruit extract of L. aegyptiaca exhibited antimicrobial activity against the tested microorganisms in a concentration-dependent manner. Overall, the diameter of the inhibition zones increased as the extract concentration increased from $0.001\ to\ 0.003\ mg/mL$. This finding suggests that higher extract concentrations may contain greater amounts of bioactive phytochemical constituents capable of suppressing microbial growth. Such a concentration-dependent response is commonly observed with plant-derived extracts and may reflect the cumulative or synergistic effects of multiple secondary metabolites present in the crude extract. A notable finding of this study was the susceptibility of Candida albicans to the extract, even at the lowest tested concentration. In contrast, the bacterial isolates showed little or no detectable inhibition at $0.001\ mg/mL$. This observation may confirm that C. albicans is sensitive to L. aegyptiaca extract. Antifungal activity may be due to phytochemical disruption for fungal cell wall integrity, membrane permeability or essential metabolic processes that need more extensive investigations. The phytochemical screening of the L. aegyptiaca extract showed flavonoids, alkaloids, tannins, saponins, cardiac glycosides, and terpenoids/steroids that are associated with antimicrobial activity. Flavonoids may have antimicrobial activity through interaction with cell wall components, alteration of membrane permeability and effect on microbial enzyme activity [18]. Tannins are known for their protein's precipitation actions and microbial enzymes and structural proteins disruption [19]. Some saponins affect membrane stability by interacting with sterols [20] while nucleic acid synthesis and cellular metabolism can be intercepted by alkaloids [21]. The L. aegyptiaca antimicrobial activity in the present study may be due to the synergistic effect of these phytochemical constituents. Further investigations are needed. Future studies should include estimation of MIC, MBC and MFC. The isolation and Identification of the active compounds along with the Docking and Insilco Studies are essential to establish the potential use of Luffa aegyptiaca extract in antimicrobial skin care preparations.

## AUTHORS' DECLARATION

We confirm that all the Figures and Tables in the manuscript belong to the current study.

## CONFLICT OF INTEREST

The authors declare that there are no conflicts of interest regarding the publication of this study.

## REFERENCES

[1] Wu M-Y, Yao X. Skin microbiota and the skin barrier. Int J Dermatol Venereol 2024; 7:18–26.

[2] Elias PM. The skin barrier as an innate immune element. Semin. Immunopathol., vol. 29, 2007, p. 3–14.

[3] Larson E. Skin hygiene and infection prevention: more of the same or different approaches? Clinical Infectious Diseases 1999; 29:1287–94.

[4] Keswick BH, Berge CA, Bartolo RG, Watson DD. Antimicrobial soaps: their role in personal hygiene. Cutaneous infection and therapy, CRC Press; 1997, p. 69–102.

[5] Caioni G, Benedetti E, Perugini M, Amorena M, Merola C. Personal care products as a contributing factor to antimicrobial resistance: Current state and novel approach to investigation. Antibiotics 2023; 12:724.

[6] Olceroglu B, Balci G, Kati A. The Skin Microbiome and Its Implications for Maintaining Skin Health. Phytochemical Potentials for Dermatological Applications, CRC Press; 2025, p. 76–103.

[7] Abdallah EM, Alhatlani BY, de Paula Menezes R, Martins CHG. Back to nature: Medicinal plants as promising sources for antibacterial drugs in the post-antibiotic era. Plants 2023; 12:3077.

[8] El-Saadony MT, Saad AM, Mohammed DM, Korma SA, Alshahrani MY, Ahmed AE, et al. Medicinal plants: bioactive compounds, biological activities, combating multidrug-resistant microorganisms, and human health benefits-a comprehensive review. Front Immunol 2025; 16:1491777.

[9] Bano S, Ansari JA, Ahsan F, Khan AR. Botanical scenario, phytochemical insights and therapeutic applications of Luffa acutangula in traditional herbal practices. Nat Prod Res 2025:1–19.

[10] Obumseli H, Okoye EL, Okafor UC. Plasmid Profiles of Multidrug Resistant Uropathogens and Antibacterial Efficacy of Leaf Extracts of Luffa cylindrica. International Journal of Research and Innovation in Applied Science 2026; 11:1006–24.

[11] Sadah Al-Halfi AS, Faisal Al-Shammari MZ, Ibrahim SK. Studying the effect of plant extracts on Gram-positive and Gram-negative pathogens. Journal of University of Anbar for Pure Science 2024;18.

[12] Giwa MS, Ibrahim B, Musa F, Abdallah EM. Evaluation of the phytochemical composition and antibacterial efficacy of Momordica balsamina and Luffa aegyptiaca leaf extracts. Journal of Medicinal Natural Products 2025:100002.

[13] Karan H, Özgen \. Inanç, Aydoğmuş E, Arslanoğlu H. Comparative assessment of sun/shade and oven

drying on structural integrity, chemical composition and functional quality of Ziziphus jujuba Mill. fruits. J Sci Food Agric 2026; 106:4087–100.

[14] Nagori M, Rajput D, Choudhary G, Khabiya R. Qualitative and quantitative methods of phytochemical analysis. Pharmacognosy and Phytochemistry: Principles, Techniques, and Clinical Applications 2025:143–66.

[15] Zabad G, Teriaky O, Alsalameh SA, Alzoubi H. Antifungal efficacy of Miswak extracts versus sodium hypochlorite against Candida albicans on CAD-CAM denture base: an in-vitro study. BMC Complement Med Ther 2026.

[16] Odongo EA, Mutai PC, Amugune BK, Mungai NN, Akinyi MO, Kimondo J. Evaluation of the antibacterial activity of selected Kenyan medicinal plant extract combinations against clinically important bacteria. BMC Complement Med Ther 2023; 23:100.

[17] Rahmayanti R, Zildiya Z, Nuraskin CA. Isolation of the fungi candida albicans in teenage girls' urine using sabouraud dextrose agar (SDA) Media. AICH: Aceh International Conference on Health, vol. 1, 2024.

[18] Górniak I, Bartoszewski R, Króliczewski J. Comprehensive review of antimicrobial activities of plant flavonoids. Phytochemistry Reviews 2019; 18:241–72.

[19] Bhat TK, Singh B, Sharma OP. Microbial degradation of tannins–a current perspective. Biodegradation 1998; 9:343–57.

[20] Böttger S, Melzig MF. The influence of saponins on cell membrane cholesterol. Bioorg Med Chem 2013; 21:7118–24.

[21] Facchini PJ. Alkaloid biosynthesis in plants: biochemistry, cell biology, molecular regulation, and metabolic engineering applications. Annu Rev Plant Biol 2001; 52:29–66.