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Research Article

In Vitro Antifungal Evaluation of a Berberine–Ellagic Acid Phytosome-Based Hydrogel Against *Candida albicans*

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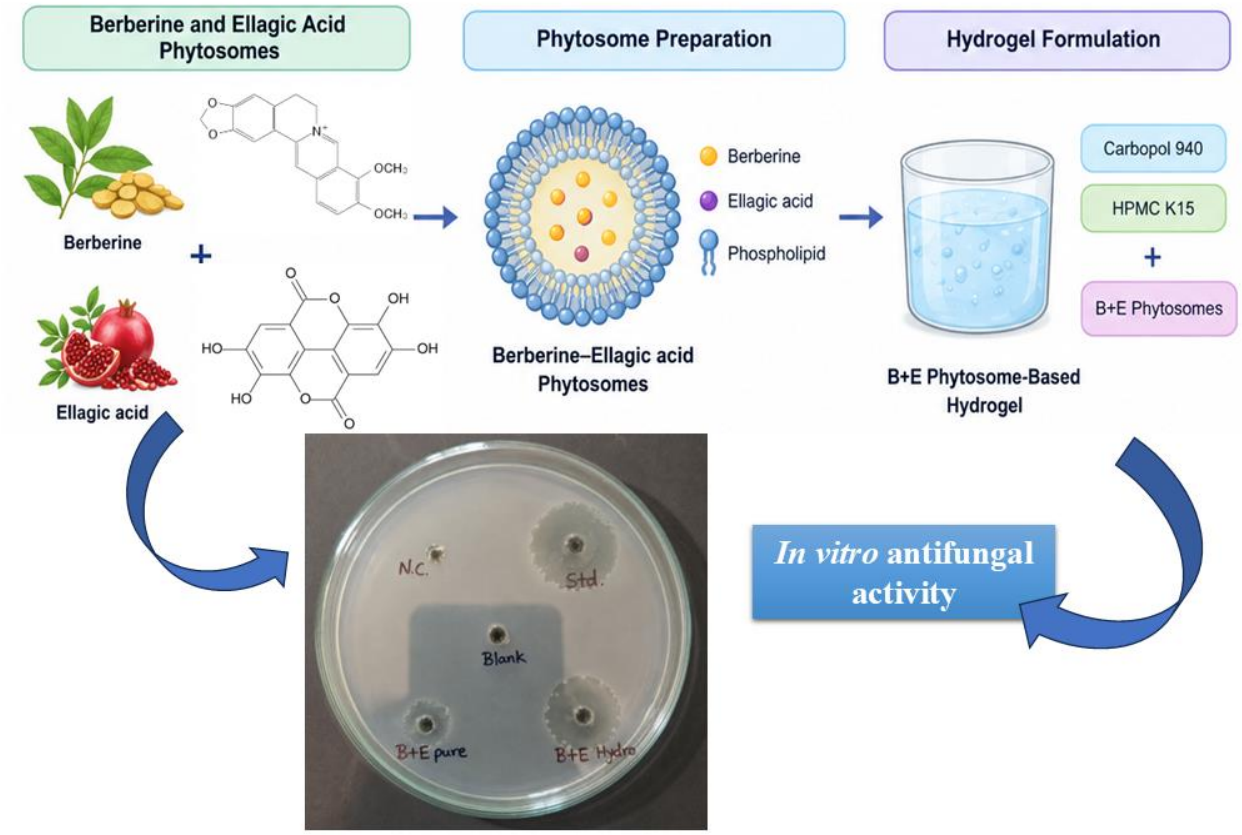
Abstract:
Candida albicans is an important fungal pathogen associated with superficial and opportunistic infections, creating a need for effective topical antifungal delivery systems. The present study aimed to evaluate the *in vitro* antifungal activity of an optimized berberine–ellagic acid (B+E) phytosome-based hydrogel against *C. albicans*. The optimized B+E phytosomes were incorporated into a Carbopol 940 and HPMC K15-based hydrogel matrix to develop a topical formulation. The antifungal activity was evaluated using the agar well diffusion method against *C. albicans*. The study included the pure B+E combination, optimized B+E phytosome hydrogel, ketoconazole as the standard antifungal drug, blank hydrogel, and normal control. Antifungal activity was assessed by measuring the zone of inhibition (ZOI). The optimized B+E phytosome hydrogel exhibited a ZOI of 23.54 mm, compared with 15.29 mm for the pure B+E combination and 26.64 mm for the standard antifungal drug. Thus, the phytosome-based hydrogel demonstrated an approximately 53.89% improvement in inhibition-zone diameter compared with pure B+E and achieved approximately 88.36% of the inhibition produced by the standard treatment. The optimized formulation also showed high drug content, with 95.86 ± 0.52% for berberine and 94.97 ± 0.61% for ellagic acid. The blank hydrogel and normal control showed no detectable inhibition. The enhanced activity may be attributed to improved dispersion and distribution of B+E within the phytosomal system and favorable diffusion characteristics of the hydrogel matrix. Overall, the findings demonstrate the potential of B+E phytosome-based hydrogel as a promising topical antifungal delivery system against *C. albicans*. Further in vivo and mechanistic studies are warranted.

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Graphical Abstract



1. Introduction

Fungal infections represent a significant public health concern, with superficial and mucocutaneous mycoses being particularly prevalent worldwide. Among pathogenic fungi, *Candida albicans* is one of the most clinically important opportunistic microorganisms and is responsible for a broad spectrum of infections ranging from superficial mucosal infections to invasive candidiasis [1]. The organism is capable of adhering to host tissues, forming biofilms, and adapting to diverse environmental conditions, which contribute to persistent infections and therapeutic challenges. Although several conventional antifungal agents, including azoles, polyenes, and echinocandins, are available for the management of candidiasis, their clinical effectiveness may be limited by poor drug penetration, toxicity, recurrence, and the emergence of antifungal resistance [2]. These challenges have stimulated interest in alternative therapeutic approaches based on bioactive phytoconstituents and advanced drug-delivery systems.

Berberine is a naturally occurring isoquinoline alkaloid obtained from several medicinal plants and has attracted considerable attention because of its broad pharmacological activities [3]. In particular, berberine exhibits antimicrobial and antifungal effects against various pathogenic microorganisms, including *C. albicans*. Its antifungal activity has been associated with multiple mechanisms, including disruption of fungal cell integrity, interference with cellular metabolism, and inhibition of processes involved in fungal growth and survival [4]. However, the practical pharmaceutical application of berberine may be constrained by its physicochemical characteristics, limited aqueous solubility, and restricted bioavailability. Ellagic acid, a naturally occurring polyphenolic compound found in several fruits and medicinal plants, also possesses antimicrobial, antioxidant, and anti-inflammatory properties [5]. Its ability to interact with microbial cellular components and interfere with microbial growth makes it a promising complementary phytoconstituent for

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antifungal formulations. Combining berberine and ellagic acid may therefore provide an opportunity to exploit their complementary biological activities against fungal pathogens [6].

Despite their therapeutic potential, the formulation of plant-derived compounds can be challenging because of inadequate solubility, stability, permeability, and effective delivery to the site of infection [7]. Phytosomes represent an advanced phytoconstituent delivery system designed to improve the incorporation and delivery of poorly bioavailable plant-derived compounds [8]. In phytosomes, active phytoconstituents are generally complexed with phospholipids, Thereby potentially improving interactions with biological membranes and enhancing drug delivery [9]. A berberine–ellagic acid phytosomal system may therefore improve the pharmaceutical performance of these phytoconstituents while providing a suitable platform for topical antifungal application [10].

Hydrogels are three-dimensional polymeric networks capable of retaining substantial quantities of water and are widely investigated for topical and localized drug delivery [11]. Their semisolid nature facilitates convenient application, prolonged contact with the skin or mucosal surface, and controlled release of incorporated therapeutic agents. Incorporation of a phytosome system into a hydrogel matrix may combine the advantages of nanostructured phytoconstituent delivery with those of a topical hydrogel, potentially enhancing localization and antifungal activity while improving formulation acceptability [12].

Therefore, the present study focuses on the *in vitro* antifungal evaluation of a berberine–ellagic acid phytosome-based hydrogel against *Candida albicans*. The study is intended to assess the antifungal effectiveness of the developed formulation using appropriate microbiological evaluation methods and to determine its potential as a topical phytoconstituent-based approach for the management of *C. albicans* infection. The findings may provide preliminary evidence to support further development and optimization of phytosome-based hydrogel systems containing synergistic or complementary natural bioactives.

2. Materials and Methods

2.1 Previously Reported Berberine–Ellagic Acid Phytosome-Based Hydrogel

The optimized berberine–ellagic acid (B+E) phytosome-based hydrogel (formulation F6) used in the present study was developed and optimized in our previous study and reported under the title, "Development and Physicochemical Characterization of Berberine–Ellagic Acid Phytosomes Incorporated into Hydrogel for Topical Drug Delivery." The optimized B+E phytosomes were incorporated into a Carbopol 940 and HPMC K15-based hydrogel matrix to obtain the final phytosome-based hydrogel formulation. The optimized F6 formulation was subsequently employed in the present investigation for further *in vitro* antifungal evaluation against *Candida albicans* using the agar well diffusion method.

2.2 *In vitro* antifungal activity by agar well diffusion method

The *in vitro* antifungal activity of the optimized berberine + ellagic acid (B+E) phytosome-based hydrogel (F6) was evaluated against *Candida albicans* using the agar well diffusion method [13]. The optimized F6 phytosomes containing berberine and ellagic acid were incorporated into a Carbopol 940 and HPMC K15-based hydrogel.

A fresh culture of C. albicans was used to prepare a uniform fungal inoculum. The prepared fungal suspension was uniformly spread over sterile Sabouraud dextrose agar (SDA) plates [14]. After inoculation, wells of uniform diameter were aseptically prepared in the agar medium.

The test samples included ketoconazole (standard antifungal drug), the pure B+E combination, the B+E phytosome-based hydrogel, the blank hydrogel, and the normal control. (NC) [15]. Ketoconazole was used at a concentration of 10 µg/mL. The B+E pure sample was prepared at a total concentration of 10 µg/mL, containing 5 µg/mL berberine and 5 µg/mL ellagic acid. The B+E phytosome hydrogel was prepared at an equivalent concentration of 10 µg/mL total B+E, based on its experimentally determined drug content. The optimized F6 hydrogel showed a drug content of 95.86 ± 0.52% for berberine and 94.97 ± 0.61% for ellagic acid.

The normal control (NC) consisted of the C. albicans inoculum without any test antifungal formulation and was used to confirm normal fungal growth in the absence of treatment [16]. The blank hydrogel consisted only of the drug-free hydrogel base without berberine or ellagic acid.

Equal volumes of the test preparations and controls were introduced into the respective wells [17]. The plates were incubated under suitable conditions for C. albicans. Following incubation, the plates were examined for clear inhibition zones around the wells. The zone of inhibition (ZOI) was measured in millimetres using a calibrated scale. Measurements were taken in two perpendicular directions, and the mean diameter was calculated [18].

Table 1. Samples used for *in vitro* antifungal activity

Sample	Recommended test concentration	Amount for 1 mL
Ketoconazole (standard)	10 µg/mL	10 µg
B+E pure	10 µg/mL total	5 µg berberine + 5 µg ellagic acid
B+E phytosome hydrogel	10 µg/mL total B+E equivalent	Equivalent to 10 µg total B+E

3. Results and Discussion

3.1 *In vitro* antifungal activity

The *in vitro* antifungal activity of the developed berberine + ellagic acid (B+E) phytosome-based hydrogel (F6) was evaluated against *Candida albicans* using the agar well diffusion method. The antifungal activity was assessed by measuring the diameter of the zone of inhibition (ZOI) produced by the standard antifungal drug, B+E pure, and the optimized B+E phytosome hydrogel, while normal control (NC) and blank hydrogel were used as controls. The ZOI was measured in millimetres following incubation, with the mean diameter used for evaluation.

As shown in Table 2 and Figure 1, the standard antifungal drug exhibited the highest antifungal activity against *C. albicans*, producing a ZOI of 26.64 mm. The B+E pure combination produced a comparatively smaller inhibition zone of 15.29 mm, indicating appreciable antifungal activity of the berberine–ellagic acid combination. In contrast, the optimized B+E phytosome hydrogel (F6) produced a markedly larger ZOI of 23.54 mm, demonstrating enhanced antifungal activity following incorporation of B+E into the phytosome-based hydrogel system.

Importantly, the ZOI produced by F6 was approximately 53.89% greater than that of B+E pure (23.54 versus 15.29 mm). This substantial increase indicates that incorporation of B+E into the phytosomal hydrogel system improved the apparent antifungal performance of the active combination under the agar diffusion conditions. The F6 formulation also produced a ZOI approximately 88.36% of that produced by the standard antifungal drug, indicating that its inhibition zone approached that of the reference treatment, although the standard remained more active under the conditions of the assay.

The enhanced inhibition observed with the F6 formulation may be associated with improved dispersion of berberine and ellagic acid within the phytosomal system, potentially facilitating their effective diffusion from the formulation into the surrounding agar medium. Phytosome formation may also improve the presentation and distribution of the active constituents within the formulation. In addition, incorporation into the Carbopol 940/HPMC K15 hydrogel matrix may provide prolonged contact of the formulation with the test medium and influence the release and diffusion characteristics of the incorporated phytosomal constituents. The optimized F6 formulation showed high drug content for both berberine (95.86 ± 0.52%) and ellagic acid (94.97 ± 0.61%), supporting the presence of substantial amounts of both active constituents in the formulation.

The normal control and blank hydrogel showed no detectable inhibition zone (0.0 mm). This confirms that normal fungal growth occurred in the absence of treatment and that the drug-free hydrogel vehicle did not exhibit appreciable intrinsic antifungal activity. Consequently, the inhibition observed with F6 can primarily be associated with the incorporated B+E phytosomal system rather than the hydrogel base itself.

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The findings demonstrate that the B+E phytosome-based hydrogel (F6) substantially enhanced the *in vitro* antifungal activity of the B+E combination against *C. albicans* compared with the corresponding pure B+E preparation. Although the ZOI of F6 remained lower than that of the standard antifungal drug, the considerable increase in inhibition-zone diameter compared with B+E pure supports the potential of phytosome-based hydrogel incorporation as a strategy for improving the antifungal performance of the B+E combination.

Table 2. *In vitro* antifungal activity of B+E formulations against *Candida albicans*

S. No.	Treatment	Zone of inhibition (mm)
1	Normal control (NC)	0.0
2	Blank hydrogel	0.0
3	Standard antifungal drug	26.64
4	B+E pure	15.29
5	B+E phytosome hydrogel	23.54

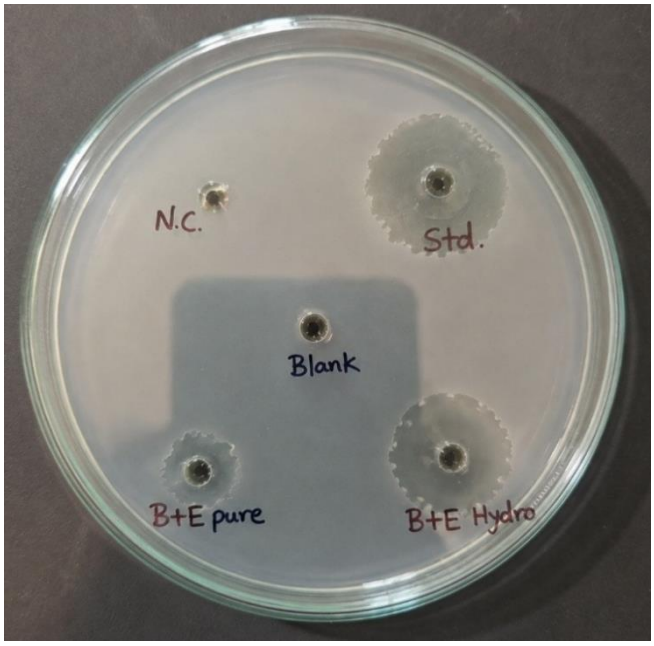


Figure 1. *In vitro* antifungal activity

4. Conclusion

In conclusion, the present study demonstrated the promising *in vitro* antifungal potential of the optimized berberine–ellagic acid (B+E) phytosome-based hydrogel against *Candida albicans*. The optimized phytosomal formulation was successfully incorporated into a Carbopol 940 and HPMC K15-based hydrogel matrix, providing a suitable topical delivery system for the combined phytoconstituents. The developed formulation exhibited enhanced antifungal activity compared with the corresponding pure B+E combination, indicating that phytosome-based incorporation may improve the antifungal performance of the active constituents. The absence of antifungal activity from the blank hydrogel further supports that the observed inhibitory effect was primarily associated with the incorporated B+E phytosomal system rather than the hydrogel vehicle. The improved activity may be attributed to enhanced dispersion and distribution of berberine and ellagic acid within the phytosomal system, along with the favorable contact and diffusion characteristics provided by the hydrogel matrix. Overall, the findings support the potential of B+E phytosome-based hydrogel as a promising topical antifungal delivery system for managing *C. albicans* infections. However, further studies, including comprehensive formulation characterization, mechanistic investigations, stability evaluation, and *in vivo* assessment, are warranted to establish its therapeutic potential.

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None

Conflict of Interest

The authors declare no conflict of interest.

Authors' Contribution

Conceptualization; methodology; software; validation; formal analysis; investigation; resources; data curation; writing—original draft preparation; writing—review and editing; visualization; supervision; project administration by N.G.N, S.P, and V.A.J. All authors have read and agreed to the published version of the manuscript.

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References

1. Talapko J, Juzbašić M, Matijević T, Pustijanac E, Bekić S, Kotris I, Škrlec I. Candida albicans-the virulence factors and clinical manifestations of infection. J. Fungi, 7, 1–19 (2021). doi: 10.3390/jof7020079.
2. Zubovskaia A, Vazquez JA. The Upcoming Antifungal Drugs in Clinical Development for the Treatment of Invasive Candidiasis. Infect. Drug Resist., 19, (2026). doi: 10.2147/IDR.S547511.
3. Purwaningsih I, Maksum IP, Sumiarsa D, Sriwidodo S. A Review of Fibraurea tinctoria and Its Component, Berberine, as an Antidiabetic and Antioxidant. Molecules, 28, (2023). doi: 10.3390/molecules28031294.
4. Susilawati S, Anwar C, Saleh I, Salni S. Flavonoid as Anti-Candida Agents. Indones. J. Fundam. Appl. Chem., 8, 88–97 (2023). doi: 10.24845/ijfac.v8.i2.88.
5. Sharifi-Rad J, Quispe C, Castillo CMS, Caroca R, Lazo-Vélez MA, Antonyak H, Polishchuk A, Lysiuk R, Oliinyk P, De Masi L, Bontempo P, Martorell M, Daştan SD, Rigano D, Wink M, Cho WC. Ellagic Acid: A Review on Its Natural Sources, Chemical Stability, and Therapeutic Potential. Oxid. Med. Cell. Longev., 2022, (2022). doi: 10.1155/2022/3848084.
6. Othman B, Beigh S, Albanghali MA, Sindi AAA, Shanawaz MA, Ibahim MAEM, Marghani D, Kofiah Y, Iqbal N, Rashid H. Comprehensive pharmacokinetic profiling and molecular docking analysis of natural bioactive compounds targeting oncogenic biomarkers in breast cancer. Sci. Rep., 15, (2025). doi: 10.1038/s41598-024-84401-4.
7. Ghosh S, Solanki R, Bhatia D, Sankaranarayanan S. Nanomaterials for delivery of medicinal plant extracts and phytochemicals: Potential applications and future perspectives. Plant Nano Biol., 12, (2025). doi: 10.1016/j.plana.2025.100161.
8. Komal Maliwal*, Pankaj Chasta, Md. Zulphakar Ali. Review on Phytosomes: A Novel Drug Delivery System. Int. J. Pharm. Life Sci., 16, 1–5 (2025). doi: 10.64149/ijpls.16.5.1-5.
9. Sakure K, Patel A, Pradhan M, Badwaik HR. Recent Trends and Future Prospects of Phytosomes: A Concise Review. Indian J. Pharm. Sci., 86, (2024). doi: 10.36468/pharmaceutical-sciences.1334.
10. Talebi M, Shahbazi K, Dakkali MS, Akbari M, Almasi Ghale R, Hashemi S, Sashourpour M, Mojab F, Aminzadeh S. Phytosomes: A promising nanocarrier system for enhanced bioavailability and therapeutic efficacy of herbal products. Phytomedicine Plus, 5, (2025). doi: 10.1016/j.phyplu.2025.100779.
11. Pan P, Svirskis D, Waterhouse GIN, Wu Z. Hydroxypropyl Methylcellulose Bioadhesive Hydrogels for Topical Application and Sustained Drug Release: The Effect of Polyvinylpyrrolidone on the Physicomechanical Properties of Hydrogel. Pharmaceutics, 15, (2023). doi: 10.3390/pharmaceutics15092360.
12. Dahma Z, Álvarez-Álvarez C, de la Torre-Iglesias PM. A Mini-Review on Enhancing Solubility in Topical Hydrogel Formulations Using Solid Dispersion Technology for Poorly Water-Soluble Drugs. Colloids and Interfaces, 9, (2025). doi: 10.3390/colloids9020017.
13. Narvekar NG, Panda S, Jagtap VA. Development and physicochemical characterization of berberine–ellagic acid phytosomes incorporated into hydrogel for topical drug delivery. J. Chem. Lett., 7, 569–89

(2026). doi: 10.22034/jchemlett.2026.584392.1434.

14. Rafika R, Rahman R, Pratama R, Artati A, Mursalim M, Nasir M, Widarti W, Nuradi N, Nurdin N, Asyikin A, Nurisyah N, Abdullah T, Asmawati A. Rice Bran Dextrose Agar as a Cost-Effective Alternative to Standard Fungal Culture Media. Trop. J. Nat. Prod. Res., 9, 6209–13 (2025). doi: 10.26538/tjnpr/v9i12.39.
15. Khudhair AS, Abdalkarim Sabri L. Preparation and evaluation of topical hydrogel containing ketoconazole-loaded bilosomes. Acta Pharm. Sci., 62, 762–77 (2024). doi: 10.23893/1307-2080.aps6250.
16. Bicer M, Öztürk E, Sener F, Hakki SS, Fidan Ö. Antifungal Efficacy of 3D-Cultured Palatal Mesenchymal Stem Cells and Their Secreted Factors against Candida albicans. ACS Infect. Dis., 11, 2894–906 (2025). doi: 10.1021/acsinfecdis.5c00657.
17. Safanda Tiara Fitri, Maulidiyah Cholifah, Isna Wulan Mukarromah, Renny Angreyni, Ferdiana Putri Gita Veronika, Dewi Rahmawati, Dzakiya Zhihrotulwida, M. Fithrul Mubarak. Artikel Review : Evaluasi pada Sediaan Steril. An-Najat, 3, 70–81 (2025). doi: 10.59841/an-najat.v3i1.2279.
18. Shaaban S, Hamad GM, Genena S, Meheissen MA, Moussa S. Evaluation of the antibacterial activity of Lactobacilli probiotics supernatants against Enterococcus faecalis (in-vitro study). BMC Oral Health, 22, (2022). doi: 10.1186/s12903-022-02434-5.