

International Journal of Advanced Biochemistry Research



ISSN Print: 2617-4693
ISSN Online: 2617-4707
NAAS Rating (2026): 5.29
IJABR 2026; SP-10(4): 1057-1062
www.biochemjournal.com
Received: 22-01-2026
Accepted: 25-02-2026

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Bioactive metabolite profiling and antifungal potential of *Curcuma longa* L.

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DOI: <https://www.doi.org/10.33545/26174693.2026.v10.i4Sm.8287>

Abstract

Curcuma longa L. (turmeric) is a medicinal plant renowned for its diverse therapeutic properties, including antioxidant, antimicrobial, anti-inflammatory, and antifungal activities. This study evaluated and compared the phytochemical composition of aqueous and methanolic extracts of *Curcuma longa* rhizomes and assessed their antifungal potential against plant pathogenic fungi. Fresh rhizomes were collected, washed, air-dried, and powdered, extracted using distilled water and methanol. Organoleptic evaluation revealed a characteristic yellow-orange color, aromatic odor, slightly bitter taste, and rough crustaceous texture. Qualitative phytochemical screening showed the presence of alkaloids, saponins, glycosides, carbohydrates, proteins, amino acids, flavonoids, tannins, and phenols in both extracts, whereas steroids, terpenoids, and quinones were detected only in the methanolic extract, indicating the higher efficiency of methanol in extracting semi-polar and non-polar compounds. The methanolic extract exhibited notable antifungal activity, significantly inhibiting *Magnaporthe oryzae*, *Rhizoctonia solani*, *Pythium aphanidermatum*, and *Fusarium oxysporum*. While both aqueous and methanolic extracts were screened for phytochemicals, antifungal activity was assessed only for the methanolic extract because of its richer spectrum of bioactive compounds. These results highlight the rich phytochemical composition of *Curcuma longa* and its potential as a natural antifungal agent, providing a scientific basis for its traditional medicinal use and future pharmacological applications.

Keywords: *Curcuma longa* L., turmeric, phytochemical screening, Methanolic extract, aqueous extract, bioactive compounds, secondary metabolites, traditional medicine

1. Introduction

Curcuma longa L., commonly known as turmeric, is a rhizomatous herbaceous perennial plant belonging to the Zingiberaceae family (Aggarwal *et al.*, 2007) [1]. Native to Southeast Asia, *Curcuma longa* has been used for centuries in Ayurvedic and traditional Chinese medicine. The present study aimed to analyze and compare the phytochemical profiles of aqueous and methanolic extracts of *Curcuma longa* L. rhizomes and to evaluate the antifungal potential of the methanolic extract, in order to determine the efficiency of each solvent in extracting different classes of bioactive compounds. The present study aimed to analyze and compare the phytochemical profiles of aqueous and methanolic extracts of *Curcuma longa* L. rhizomes and to evaluate the antifungal potential of the methanolic extract, in order to determine the efficiency of each solvent in extracting different classes of bioactive compounds. due to its diverse pharmacological properties (Pawar and Dingra, 2024) [9]. The plant is characterized by bright yellow-orange rhizomes rich in curcuminoids, the primary bioactive compounds responsible for its medicinal effects (Patel *et al.*, 2023) [8]. *Curcuma longa* L. has been traditionally used for its anti-inflammatory, antioxidant, and antimicrobial properties, and has shown promise in the treatment of various ailments, including arthritis, digestive disorders, and skin conditions. This study aimed to analyse and compare the phytochemical profiles of aqueous and methanolic extracts of *Curcuma longa* L. rhizomes and to evaluate the antifungal potential of the methanolic extract, in order to determine the efficiency of each solvent in extracting different classes of bioactive compounds.

2. Materials and Methods

2.1 Collection of sample

Fresh rhizomes of *Curcuma longa* L. were collected from the Banusultan, Chatha region of Jammu district, India, and transported to the laboratory of Sher-e-Kashmir University of Agricultural Sciences and Technology (SKUAST-Jammu). The plant material was washed thoroughly with distilled water to remove adhering dust and impurities, cut into small pieces, and air-dried at room temperature until completely dried. The dried rhizomes were then ground into a fine powder using an electric grinder for further analysis.

2.2 Extraction

Approximately 10 g portion of the powdered rhizome sample was accurately weighed and extracted with 100 ml of methanol and distilled water separately. The mixture was allowed to stand with occasional shaking to ensure complete extraction of phytochemicals. The solution was then filtered through Whatman No. 1 filter paper, and the filtrate was collected for further qualitative phytochemical analysis (Fig.1).

2.3 Macroscopic (Organoleptic) Evaluation

The macroscopic characteristics of both the rhizome powder and methanolic extract were evaluated based on parameters such as color, odor, texture, appearance, and solubility using visual and sensory observations.

2.4 Qualitative Phytochemical Analysis

Both aqueous and methanolic extract were subjected to qualitative phytochemical screening to identify the presence of various bioactive compounds. The following tests were performed (Harborne, 1998) [3].

2.4.1 Alkaloid Test (Mayer's test)

Approximately 2ml of extract was acidified with a few

drops of dilute hydrochloric acid and treated with Mayer's reagent. The presence of alkaloids was indicated by the formation of a yellow or cream coloured precipitate (Shaikh and Patil, 2020) [13].

2.4.2 Saponins Test (Foam Test)

1 ml of the methanolic extract was mixed with 5 ml of distilled water and shaken vigorously for 5 minutes. The formation of stable foam indicated the presence of saponins (Mujeeb *et al.*, 2014) [3].

2.4.3 Glycoside Test (Alkaline reagent test)

A small quantity of the filtrate was mixed with 1 ml of distilled water followed by the addition of sodium hydroxide solution. The development of a yellow coloration confirmed the presence of glycosides (Roghini and Vijaylakshmi, 2018) [12].

2.4.4 Phenols Test (Ferric Chloride Test)

Two ml of distilled water was added to 1ml of extract followed by a few drops of 10% ferric chloride. The presence of phenols can be detected by the formation of blue or green colour (Lydia *et al.*, 2016) [4].

2.4.5 Flavonoids Test (Alkaline Reagent Test)

1 ml of sodium hydroxide was added to 2ml of extract. The presence of flavonoids can be detected by their changing colour to yellow (Lydia *et al.*, 2016) [4].

2.4.6 Test for steroids and Phytosteroids (Salkowski's test)

One milliliter of chloroform followed by few drops of concentrated sulphuric acid was added to one milliliter of extracts. The presence of steroids and phytosteroids can be detected by the appearance of brown ring and bluish ring respectively (Ushie *et al.*, 2022) [14].

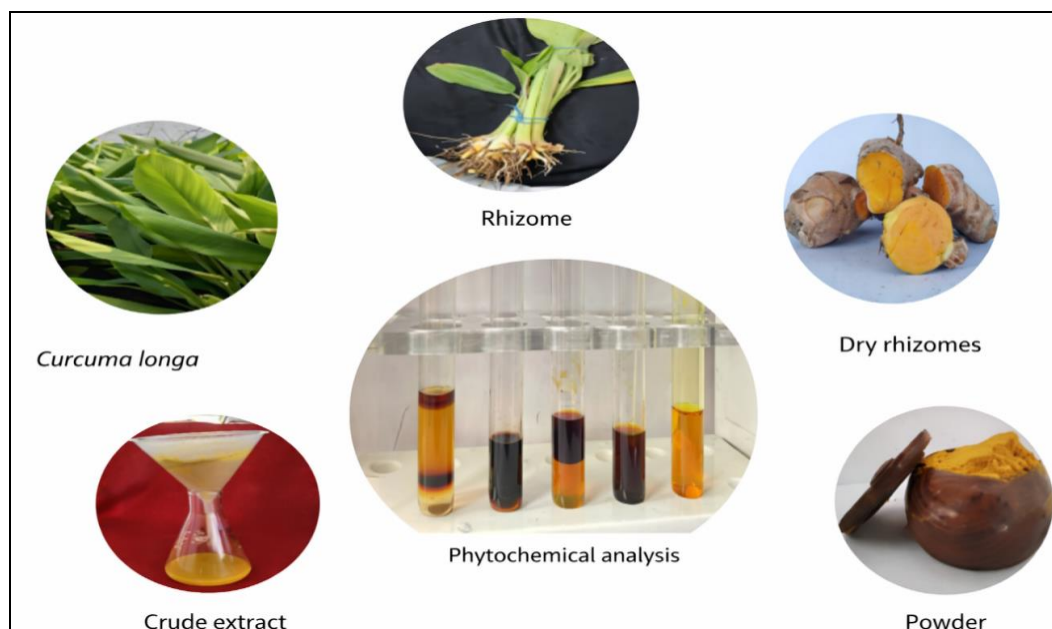


Fig 1: Sequential stages of *Curcuma longa* L. processing from rhizome collection to phytochemical analysis.

2.4.7 Tannins Test (Braymer's Test)

Two ml of 5% ferric chloride was added to 1ml of rhizome extract. The presence of tannins can be detected by their changing colour to greenish black or dark blue (Lydia *et al.*, 2016) [4].

2.4.8 Test for carbohydrates (Molisch's reagent)

Approximately, 1ml of the filtrate was treated with two drops of Molisch's reagent, followed by the careful addition of concentrated sulfuric acid along the sides of the test tube.

The formation of a violet or reddish ring indicated the presence of carbohydrates (Lydia *et al.*, 2016) ^[4].

2.4.9 Test for Proteins (Millon's Test)

Two milliliters of the plant extract were treated with 2 ml of Millon's reagent and allowed to stand for two minutes. The formation of a white precipitate, which turned red upon gentle heating, confirmed the presence of proteins (Mishra *et al.*, 2024) ^[5]

2.4.10 Test for Amino Acids (Ninhydrin Test)

Two millilitres of the plant extract were treated with 2 ml of ninhydrin reagent and heated gently. The appearance of a violet coloration indicated the presence of amino acids (Bhati *et al.*, 2022) ^[2].

2.4.11 Test for Quinones (Sulphuric acid Test)

1 ml of extract was treated with concentrated sulphuric acid. The formation of red coloration indicated the presence of quinones (Lydia *et al.*, 2016) ^[4].

2.5 Evaluation of the antifungal activity of *Curcuma longa* extracts against fungal pathogens

2.5.1 Agar Well Diffusion Method

The antifungal activity of the plant extract was evaluated using the agar well diffusion method. The fungal pathogen was first cultured on Potato Dextrose Agar (PDA) plates and incubated at 28-30 °C for 7 days to obtain fresh growth. A small portion of the fungal colony from the PDA plate was then inoculated into Potato Dextrose Broth (PDB) and incubated at 28-30 °C for 3-5 days to obtain a uniform fungal suspension (Navoda and Dinushani Anupama 2022) ^[7].

Sterile PDA plates were inoculated evenly with the prepared fungal suspension using a sterile cotton swab. Wells of 6 mm diameter were made in the agar using a sterile cork borer. Each well was filled with 0.1 ml of plant extract. While Propiconazole was used as the positive control.

The plates were allowed to stand at room temperature for 1 hour to permit diffusion of the extract and then incubated at 28-30 °C. After incubation, the zone of inhibition around each well was measured in millimeters (mm).

All experiments were carried out in triplicate, and the results were expressed as mean±standard deviation (SD).

3. Result

3.1 Organoleptic Evaluation

The organoleptic evaluation of *Curcuma longa* rhizomes revealed distinct physical and sensory characteristics. The rhizomes exhibited a brown outer surface, while the inner surface was bright yellow, a characteristic feature often associated with its high curcuminoid content. In terms of taste, the rhizome was slightly bitter, aligning with its traditional use in Ayurvedic medicine for its digestive and anti-inflammatory properties. The detailed organoleptic characteristics are summarized in Table 1.

Table 1: Organoleptic characteristics of *Curcuma longa* L. rhizome

S. No.	Parameters	Description of rhizomes
1.	Color	Yellow-orange
2.	Odor	Characteristic
3.	Taste	Slightly bitter
4.	Texture	Crusty
5.	Touch	Rough

3.2 Qualitative phytochemical analysis

The qualitative phytochemical screening of *Curcuma longa* L. revealed the presence of multiple bioactive compounds in both aqueous and methanolic extracts, indicating the plant's rich phytochemical profile. In the aqueous extract, alkaloids were detected using Mayer's test, saponins were confirmed by the foam test, and glycosides were identified through the alkaline reagent test. Carbohydrates were present as indicated by, while proteins were detected using Millon's test. Amino acids were also observed in the aqueous extract with the Ninhydrin test.

However, certain compounds such as steroids, terpenoids, and quinones were not detected in the aqueous extract, suggesting their limited solubility or absence in water. In contrast, the methanolic extract demonstrated a broader spectrum of phytochemicals. All compounds present in the aqueous extract, including alkaloids, saponins, glycosides, carbohydrates, proteins, and amino acids, were also present in the methanol extract. Additionally, steroids, terpenoids, and quinones were successfully detected in the methanolic extract using Salkowski's and sulphuric tests, highlighting the superior extraction efficiency of methanol for non-polar or semi-polar compounds.

Furthermore, flavonoids, tannins, and phenols were detected in both aqueous and methanolic extracts. Flavonoids were confirmed by the alkaline reagent test through the development of a yellow coloration. Tannins were identified using Braymer's test, which produced a greenish-black coloration, while phenolic compounds were confirmed by the ferric chloride test showing a characteristic blue-green color. The presence of these phytochemicals indicates that *Curcuma longa* rhizomes contain significant phenolic constituents that may contribute to their antioxidant and therapeutic properties.

Table 2: Qualitative Phytochemical Screening of Aqueous and Methanolic Extracts of Rhizome

S. No	Phytochemical	Test name	Aqueous	Methanol
1.	Alkaloids	Mayer's test	Present	Present
2.	Saponins	Foam test	Present	Present
3.	Glycosides	Alkaline reagent test	Present	Present
4.	Steroids	Salkowski's test	Absent	Present
5.	Carbohydrates	Molisch's reagent	Present	Present
6.	Protein	Millon's Test	Present	Present
7.	Amino acids	Ninhydrin test	Present	Present
8.	Terpenoids	Salkowski's test	Absent	Present
9.	Quinone	Sulphuric test	Absent	Present
10.	Tannins	Braymer's Test	Present	Present
11.	Flavonoids	Alkaline Reagent Test	Present	Present
12.	Phenols	Ferric Chloride Test	Present	Present

The qualitative phytochemical analysis of *Curcuma longa* L. rhizome extracts revealed the presence of flavonoids, tannins, and phenols in both aqueous and methanolic extracts. Flavonoids were confirmed by the alkaline reagent test, which showed the development of a yellow coloration indicating a positive reaction. Tannins were detected using Braymer's test, where the formation of a greenish-black coloration indicated their presence. Similarly, phenolic compounds were identified by the ferric chloride test, which produced a characteristic blue-green coloration. The presence of these phytochemicals in both extracts suggests that *Curcuma longa* rhizomes contain significant amounts of phenolic compounds that may contribute to their antioxidant and therapeutic properties.

Overall, these findings demonstrate that *Curcuma longa* L. contains a diverse range of phytochemicals, with the methanolic extract showing a higher capacity to extract a wider variety of secondary metabolites compared to the aqueous extract. These results support the traditional

medicinal use of *Curcuma longa* L. and highlight its potential for further pharmacological and therapeutic investigations. These results are presented in Table 2. and Fig. 2.

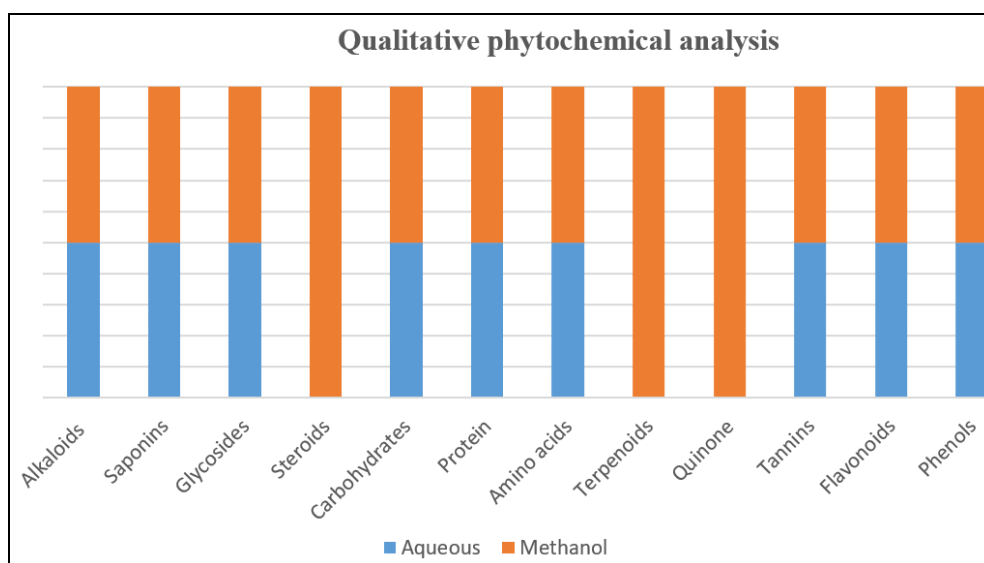


Fig 2: Graphical representation of qualitative phytochemical constituents present in aqueous and methanolic extracts of *Curcuma longa* L.

3.3 Antifungal Activity of *C. longa* Methanolic Extract

The extract of *Curcuma longa* exhibited notable antifungal activity against the tested fungal pathogens. The zone of inhibition (mm) for each pathogen is summarized in Table 1. Among the pathogens tested, the extract showed the highest inhibition against *Magnaporthe oryzae* (20.00 ± 0.20 mm), followed by *Rhizoctonia solani* (18.00 ± 0.60 mm),

Pythium aphanidermatum (14.00 ± 0.02 mm), and *Fusarium oxysporum* (12.00 ± 0.04 mm). The standard fungicide, Propiconazole, showed slightly higher inhibition against *Magnaporthe oryzae* (22.00 ± 0.05 mm) and *Fusarium oxysporum* (19.00 ± 0.20 mm), and comparable activity against *Rhizoctonia solani* (18.00 ± 0.60 mm) and *Pythium aphanidermatum* (18.00 ± 0.50 mm).

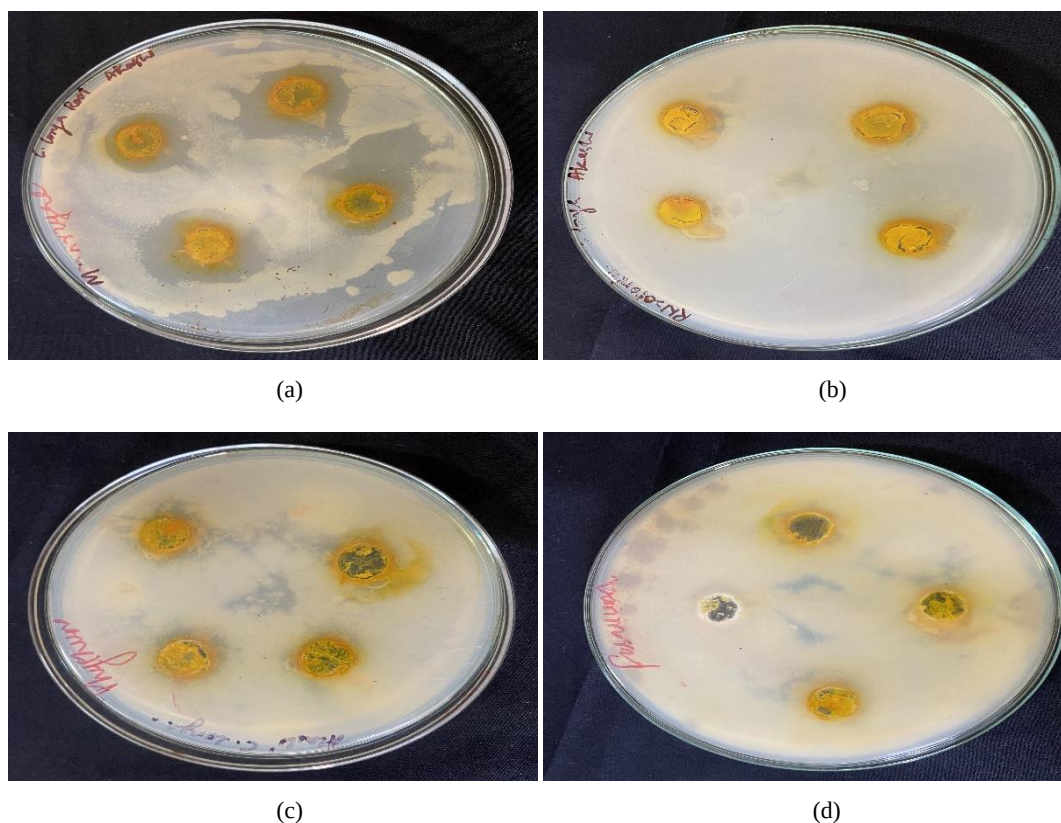


Fig 3: Antifungal activity of *Curcuma longa* extract against plant pathogens: (a) *Magnaporthe oryzae*, (b) *Rhizoctonia solani*, (c) *Fusarium oxysporum*, (d) *Pythium aphanidermatum*

The antifungal activity of *Curcuma longa* extract against different fungal pathogens is visually represented in Fig 3, while the comparative graphical analysis between the plant extract and the standard fungicide is shown in Fig. 4. Based on the results of the phytochemical analysis, the

methanolic extract exhibited a richer phytochemical profile and was therefore selected for antifungal evaluation. Overall, the extract demonstrated moderate to good antifungal potential, suggesting it could be a promising natural alternative to chemical fungicides.

Table 3: Antifungal activity of *Curcuma longa* methanolic extract against plant pathogenic fungi

S. No.	Sample	Zone of inhibition (mm)			
		<i>Magnaporthe oryzae</i>	<i>Rhizoctonia solani</i>	<i>Fusarium oxysporum</i>	<i>Pythium aphanidermatum</i>
1.	<i>C. longa</i> extract (4 mg/ml)	20.00±0.20	18.00±0.60	12.00±0.04	14.00±0.02
2.	Control (Propiconazole)	22.00±0.05	18.00±0.60	19.00±0.20	18.00±0.50

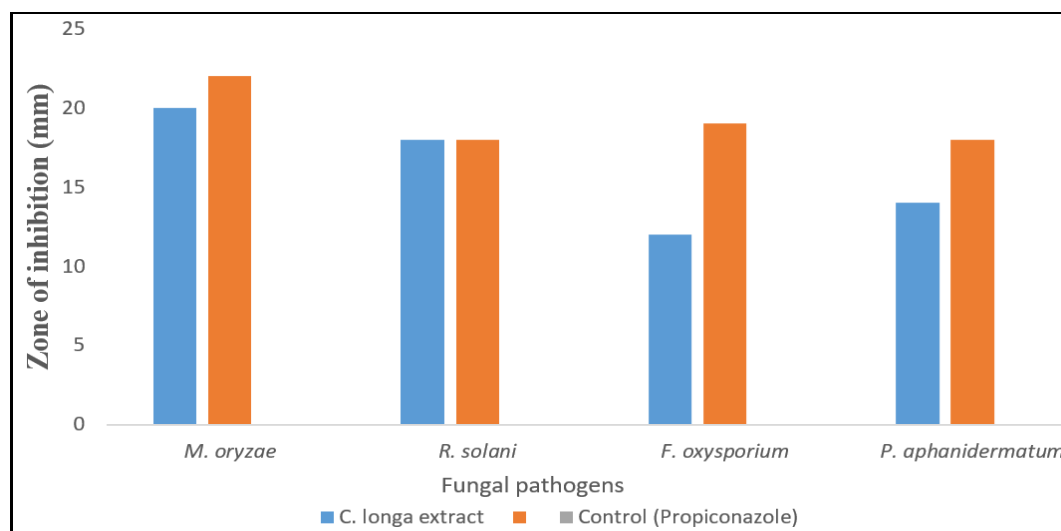


Fig 4: Graphical representation of antifungal activity (zone of inhibition (mm)) of *Curcuma longa* extract compared with Propiconazole against plant pathogenic fungi

4. Summary

This study investigated the phytochemical composition of aqueous and methanolic extracts and the antifungal potential of methanolic extract of *Curcuma longa* L. rhizomes. Fresh rhizomes were collected, cleaned, air-dried, powdered, and extracted using distilled water and methanol. Organoleptic evaluation revealed characteristic features of turmeric rhizomes, including a yellow-orange color, aromatic odor, slightly bitter taste, and rough crustaceous texture.

Qualitative phytochemical screening demonstrated the presence of several bioactive compounds such as alkaloids, saponins, glycosides, carbohydrates, proteins, amino acids, flavonoids, tannins, and phenols in both aqueous and methanolic extracts. However, steroids, terpenoids, and quinones were detected only in the methanolic extract, indicating the higher efficiency of methanol in extracting semi-polar phytochemicals.

Based on the phytochemical screening results, the methanolic extract was selected for antifungal evaluation. The antifungal activity was assessed using the agar well diffusion method against plant pathogenic fungi including *Magnaporthe oryzae*, *Rhizoctonia solani*, *Pythium aphanidermatum*, and *Fusarium oxysporum*. The methanolic extract showed notable antifungal activity, with the highest inhibition observed against *Magnaporthe oryzae*, followed by *Rhizoctonia solani*, *Pythium aphanidermatum*, and *Fusarium oxysporum*. The activity was comparable to the standard fungicide propiconazole in some cases.

These findings indicate that *Curcuma longa* rhizomes possess a rich phytochemical profile and significant antifungal potential, supporting their traditional medicinal and agricultural applications.

5. Conclusion

The present study demonstrated that *Curcuma longa* L. rhizomes contain a diverse range of bioactive phytochemicals with potential biological activity. Qualitative phytochemical analysis revealed that both aqueous and methanolic extracts contain important secondary metabolites such as alkaloids, flavonoids, tannins, phenols, and glycosides. However, the methanolic extract exhibited a broader phytochemical profile, including steroids, terpenoids, and quinones, highlighting methanol as a more efficient solvent for extracting semi-polar compounds.

The methanolic extract showed moderate to strong antifungal activity against several plant pathogenic fungi, particularly *Magnaporthe oryzae* and *Rhizoctonia solani*. These results suggest that the antifungal activity may be attributed to the presence of multiple bioactive phytochemicals present in the rhizomes.

Overall, the study confirms that *Curcuma longa* is a promising natural source of antifungal compounds. The findings support its traditional medicinal use and highlight its potential application in the development of eco-friendly plant-based antifungal agents.

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