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Original Research Article

Physicochemical Parameters, Preliminary Chemical Investigation and Antifungal Activity of Whole Plant of *Euphorbia hirta* Linn

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Abstract: The present study deals with the pharmacognostic characterisation, physicochemical evaluation, preliminary phytochemical screening and antifungal activity of the whole plant of *Euphorbia hirta* Linn (family Euphorbiaceae). The plant material was extracted with methanol by continuous Soxhlet extraction and with water by maceration, giving yields of 31.43% and 15.20% respectively. Physicochemical parameters, namely moisture content (4.5%), total ash (10.5%), acid insoluble ash (2.5%), acid soluble ash (1.8%) and extractive values, were determined. Preliminary phytochemical screening of both extracts showed the presence of carbohydrates, proteins and amino acids, phenolic compounds and tannins, alkaloids, flavonoids and volatile oil. Antifungal activity was evaluated against *Candida albicans* and *Aspergillus niger* by the agar cup plate diffusion method, and the minimum inhibitory concentration (MIC) was determined. The methanolic extract at 50 and 100 mg/ml showed significant antifungal activity, whereas the aqueous extract showed moderate to mild activity. The MIC of both extracts against both fungi was 1000 µg/ml. The findings indicate that the whole plant of *E. hirta* is a promising source of antifungal agents.

Keywords: *Euphorbia hirta* Linn, Pharmacognostic Characterisation, Aqueous Extract, Methanol Extract, Preliminary Chemical Tests, Antifungal Activity, *Candida Albicans*, *Aspergillus Niger*.

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INTRODUCTION

Garden spurge is an herbaceous annual plant considered to be a warm-season weed in the spurge family (Euphorbiaceae). It originates from tropical and subtropical Americas but has spread to Africa, Southeast Asia, the Pacific Islands and Australia. It is found in lawns, agricultural areas, roadsides and in the nursery trade. It can also be found in planting beds, sidewalk cracks or between patio pavers. The specific epithet, *hirta*, is Latin for hairy and refers to the plant's stems. *Euphorbia hirta* L. is a medicinal, rhizomatous herb distributed in the Southern Western Ghats of India and Northern East Coast of Tamil Nadu (Abdul Rahuman A *et al*., 2007). In Eastland West Africa, extracts of the plant are used in the treatment of asthma and respiratory tract inflammations. It is also used for coughs, chronic bronchitis and other pulmonary disorders in Malagasy. The plant is also widely used in Angola against diarrhoea and dysentery, especially amoebic dysentery. In Nigeria, extracts or exudates of the plant are used as ear drops and in the treatment of boils and sores and for promoting wound healing (Ogueke CC *et al*., 2007).

The plant is native to India but is a pantropical weed, found especially on roadsides and waste land. It is a small, erect or ascending annual herb reaching up to 50 cm, with hairy stems. The leaves are opposite, elliptical, oblong or oblong-lanceolate, with a faintly toothed margin and darker on the upper surface. The flowers are small, numerous and crowded together in dense cymes about 1 cm in diameter. The fruits are yellow, three-celled, hairy, keeled capsules, 1–2 mm in diameter, containing three brown, four-sided, angular, wrinkled seeds (Kirtikar KR *et al*., 1991). The parts used are the leaves, stem and flowers (Figure 1). In India, *E. hirta* is commonly known as "Dudhi" (Hindi), "Dugdhika" (Kannada) and "Ammanpacharisi" (Tamil), among other regional names. The present study was undertaken to evaluate the pharmacognostic characters, physicochemical parameters, preliminary phytochemical

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profile and antifungal activity of the whole plant extracts of *E. hirta* Linn (Abdul Rahuman *et al*., 2007); (Agrawal *et al*., 1995); (Al-Jabri, 2005); (Bhattacharya *et al*., 2023); (Deep *et al.,* 2004); (Dhane *et al*., 2023); (Difuntorum *et al*., 2002); (Diwate *et al*., 2025); (Eloff, 2000); (Ghatage *et al*., 2026); (Goyal *et al.,* 2021); (Khadabadi, 2022); (Kirtikar and Basu, 1991); (Konduskar *et al*., 2025); (Lokapure and Patil, 2026); (Malla *et al*., 2023); (McAllister *et al*., 2019); (Memon *et al*., 2026); (Murdoch *et al*., 2012); (Nair and Chanda, 2005); (Neogi *et al*., 2008); (Norrby *et al*., 2005); (Ogueke *et al*., 2007); (Paniker and Ananthanarayana, 2005); (Paniker and Ananthanarayana, 2009); (Patil, 2026); (Patil, 2026); (Patil and Mali, 2026); (Patil, 2026); (Patil and Girija, 2026); (Patil *et al*., 2025).



Figure 1: *Euphorbia hirta* Linn

Experimental Section

Plant Material

The whole plant of *Euphorbia hirta* Linn was collected from the local area of Kalaburagi district in August 2016. The plant was authenticated by Dr. Prathiba G. Sangapurkar, Head, Department of Botany, V. G. Women's College, Kalaburagi. A voucher specimen (Ref. No. SVGWCG/05) has been deposited in the college museum.

Preparation of Extracts

The plant was dried under shade, pulverised with a grinder to a coarse powder and stored in air-tight containers for further studies. The powder was extracted with methanol by continuous Soxhlet extraction, and the aqueous extract was obtained by maceration using 5% chloroform (Trease and Evans,2025). The extracts were concentrated under reduced pressure on a water bath and air dried at room temperature. The extracts were weighed, the percentage yield was calculated, and the colour and consistency were noted. The dried extracts were stored in amber-coloured air-tight bottles in a refrigerator at 4 °C.

Physicochemical Parameters

Physicochemical parameters of the whole plant, namely moisture content (loss on drying), total ash, acid insoluble ash, acid soluble ash, water-soluble ash, and chloroform, acetone, alcohol and water-soluble extractive values, were determined. These parameters help in determining adulterants, chemical constituents and impurities such as earthy contaminants and minerals in a drug (Paniker CKJ *et al*., 2005).

Preliminary Phytochemical Screening

The methanolic and aqueous extracts were subjected to the qualitative chemical tests listed in Table 3 (Khadabadi SS, 2022).

Antifungal Activity

The zone of inhibition obtained with different concentrations of *Euphorbia hirta Linn* extracts are shown in antifungal Activity. The zone of inhibition obtained with different concentrations *Euphorbia hirta* Linn whole plant extracts are shown in Table no 4 and 5 and plate no 1 to 4. Methanolic extract at 50 mg/ml and 100 mg/ml of concentration was found to be having significant anti-fungal activity against *C. albicans* and *A. niger*, whereas aqueous extracts were exhibit moderate and mild anti-fungal activity against tested organisms.

Statistical Analysis

Values are expressed as mean ± standard error of the mean (SEM) and were compared by one-way ANOVA followed by Dunnett's test (**p<0.01, ***p<0.001, ****p<0.0001)); (Patil *et al*., 2025); (Patil *et al*., 2025); (Patil *et al.,* 2023); (Pawar *et al*., 2023); (Phalake *et al*., 2026); (PERCH Study Group, 2019); (Rudan *et al*., 2008); (Save, 2026); (Sharma *et al*., 2027); (Shinde *et al*., 2026); (Shingade *et al*., 2026); (Sung and Lee, 2007); (Surwase *et al*., 2026); (Survase *et al*., 2025); (Talsandekar *et al*., 2026); (Trease and Evans, n.d.); (von

Elm *et al*., 2007); (Walvekar *et al*., 2025); (Wang *et al*., 2026); (Woolverton *et al*., 2014); (Zar *et al*., 2017).

RESULTS AND DISCUSSION

Extractive Values

The percentage yields, colour and consistency of the methanolic and aqueous extracts are given in Table 1.

Table 1: Extractive values of whole plant of *Euphorbia hirta* Linn

Sl. No.	Solvent	Colour and consistency	Percentage yield (%)
1	Methanol	Brown	31.43
2	Aqueous	Dark reddish brown	15.20

Physicochemical Parameters

The physicochemical parameters of *E. hirta* Linn were determined, calculated and are reported in Table 2.

Table 2: Physicochemical characters of whole plant of *Euphorbia hirta* Linn

Sl. No.	Physicochemical parameter	Value determined (% w/w)
01	Total ash	10.5
02	Acid insoluble ash	2.5
03	Acid soluble ash	1.8
04	Acetone extractive value	48.9
05	Chloroform extractive value	43.5
06	Alcohol soluble extractive value	59.3
07	Water-soluble extractive value	55.3
08	Moisture content	4.5

Preliminary Phytochemical Screening

Qualitative phytochemical screening of the methanolic and aqueous extracts of *E. hirta* Linn showed the presence of carbohydrates, proteins, amino acids, alkaloids, glycosides, tannins, flavonoids and volatile oils. The results are presented in Table 3.

Table 3: Preliminary phytochemical screening of the whole plant extracts of *Euphorbia hirta* Linn (methanolic and aqueous extracts)

Sl. No.	Constituents	Test	Methanolic extract	Aqueous extract
1	Carbohydrates	Molisch's test	+	+
2	Fixed oils and fats	Spot test	–	–
		Saponification test	–	–
3	Proteins and amino acids	Millon's test	+	+
		Xanthoproteic test	+	+
		Ninhydrin test	+	+
		Biuret test	+	+
4	Saponins	Foam test	–	–
5	Phenolic compounds and tannins	$FeCl_3$ test	+	+
		Gelatin test	+	+
		Lead acetate test	+	+
		Bromine water test	+	+
6	Phytosterols	Salkowski test	+	–
		Liebermann–Burchard test	–	–
7	Alkaloids	Dragendorff's test	+	+
		Mayer's test	+	–
8	Gums and mucilage	Swelling test	–	–
9	Flavonoids	Aqueous NaOH test	+	+
		Conc. H_2SO_4 test	+	+
		Shinoda's test	+	+
		Lead acetate test	+	+
10	Volatile oil	—	+	+

(+) indicates positive result and (–) indicates negative result

Antifungal Activity

The zones of inhibition obtained with different concentrations of the methanolic and aqueous whole plant extracts of *E. hirta* Linn are shown in Tables 4 and 5, Figures 2 and 3, and Plates 1 to 4. The methanolic extract at 50 mg/ml and 100 mg/ml showed significant

antifungal activity against *C. albicans* and *A. niger*, whereas the aqueous extract showed moderate to mild antifungal activity against the tested organisms. For the methanolic extract (50 and 100 mg/ml), the mean ± SEM zones were 11.67 ± 0.8819 and 10.67 ± 0.8819 mm against *C. albicans* (standard 17 ± 1.155 mm) and 9.333 ± 0.8819 and 10.33 ± 0.8819 mm against *A. niger* (standard 13.67 ± 0.8819 mm). For the aqueous extract, the corresponding values were 8.667 ± 0.8819 and 9.667 ± 0.8819 mm against *C. albicans* (standard 11.67 ± 0.8819 mm) and 10.33 ± 0.8819 and 8.667 ± 0.8819 mm against *A. niger* (standard 12 ± 0.5774 mm).

Table 4: Antifungal activity of the methanolic extract of *Euphorbia hirta* Linn against selected fungal strains

Sl. No.	Fungal strain	Zone of growth, mean ± SEM (mm)		
		Extract concentrations		Standard
		50 mg/ml	100 mg/ml	
1	*C. albicans*	11.67±0.8819**	10.67±0.8819**	17±1.155****
2	*A. niger*	9.333±0.8819***	10.33±0.8819***	13.67±0.8819***

Each value is expressed as mean ± standard error of the mean; **p<0.01, ***p<0.001, ****p<0.0001 when compared by one-way ANOVA followed by Dunnett's test.

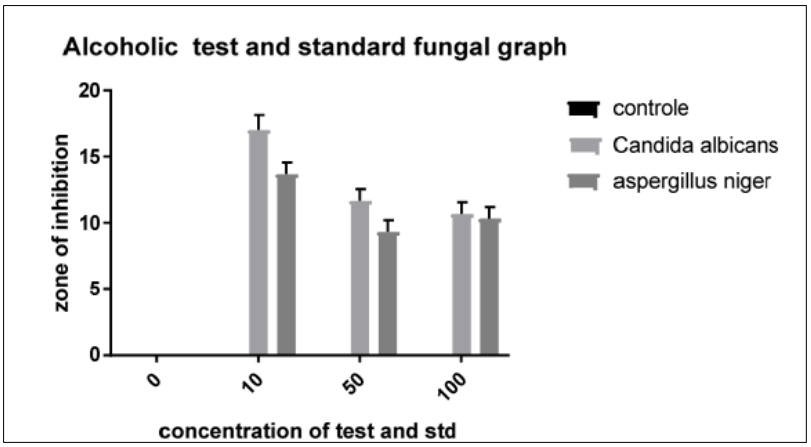


Figure 2: Zone of inhibition of the methanolic extract and standard against *C. albicans* and *A. niger*



Plate 1: *Candida albicans*

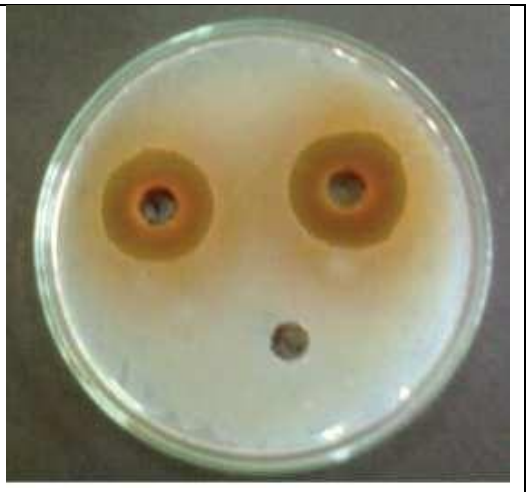


Plate 2: *Aspergillus niger*

(A) Test, (B) Standard, (C) Control

Table 5: Antifungal activity of the aqueous extract of *Euphorbia hirta* Linn against selected fungal strains

Sl. No.	Fungal strain	Zone of growth, mean ± SEM (mm)		
		Extract concentrations		Standard
		50 mg/ml	100 mg/ml	
1	*C. albicans*	8.667±0.8819***	9.667±0.8819***	11.67±0.8819****
2	*A. niger*	10.33±0.8819****	8.667±0.8819***	12±0.5774****

Each value is expressed as mean ± standard error of the mean; ***p<0.001, ****p<0.0001 when compared by one-way ANOVA followed by Dunnett's test.

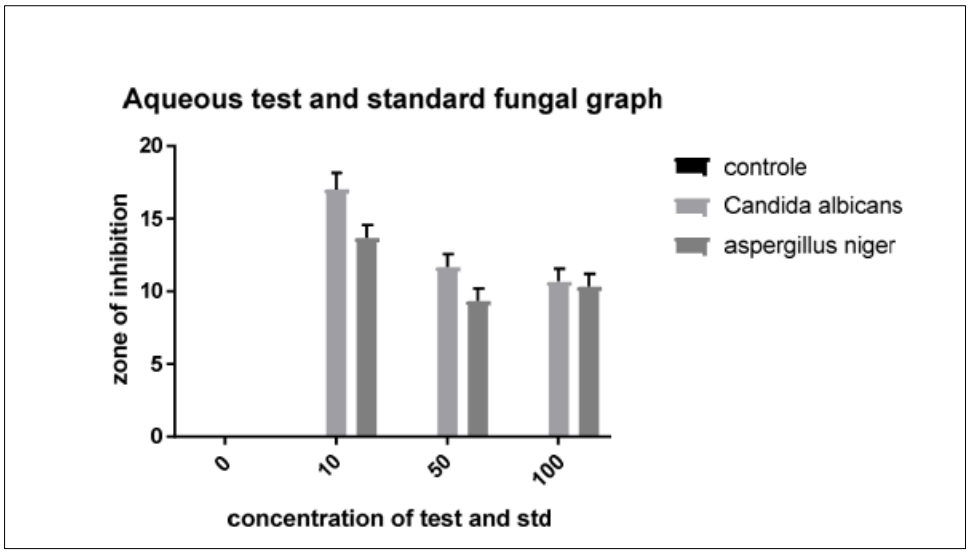


Figure 3: Zone of inhibition of the aqueous extract and standard against *C. albicans* and *A. niger*



Plate 3: *Candida albicans*



Plate 4: *Aspergillus niger*

(A) Test, (B) Standard, (C) Control

Minimum Inhibitory Concentration (MIC)

The MIC of the extracts of *E. hirta* Linn whole plant for fungi was found to be 1000 µg/ml (Table 6). MIC for *E. hirta* Linn extract against bacteria was found to be at 500 µg/ml and for fungi at 1000 µg/ml concentration, but there was no inhibition of bacteria at the tested concentrations.

Table 6: MIC values of *Euphorbia hirta* Linn whole plant methanolic and aqueous extracts for fungi

Fungal strain	Minimum inhibitory concentration (MIC) in µg/ml	
	Methanolic extract	Aqueous extract
Candida albicans	1000	1000
Aspergillus niger	1000	1000

DISCUSSION

The search for antifungals from natural sources has received much attention, and efforts have been made to identify compounds that can act as suitable antimicrobial agents to replace synthetic ones. Phytochemicals derived from plant products serve as prototypes to develop less toxic and more effective medicines for controlling the growth of microorganisms. These compounds have significant therapeutic application against human pathogens, including bacteria, fungi and viruses. Numerous studies have been conducted with extracts of various plants, screening for antimicrobial activity as well as for the discovery of new antimicrobial compounds. In the present investigation, different extracts of the whole plant of *E. hirta* Linn were evaluated for their antimicrobial activity against certain Gram negative and Gram positive bacteria, fungus which are regarded as human pathogenic microorganisms. Susceptibility to the plant extracts was determined by the

serial micro dilution method (MIC) and the agar well diffusion method.

Our preliminary investigation showed that the methanolic and aqueous extracts of *E. hirta* Linn were active against the locally isolated human pathogens *Candida albicans* and *Aspergillus niger*. The present results show that the whole plant extracts are effective against the tested fungi. This may be due to the larger number of secondary metabolites in the whole plant, which may interfere with fungal growth.

CONCLUSION

The results of this study show that the whole plant extracts of *E. hirta* Linn have great potential as antifungal agents in the treatment of fungal infections. Further detailed investigation of the active compounds (carbohydrates, alkaloids, total phenolic compounds, flavonoids and amino acids) of the plant, and of the mechanism of action of the extracts, will contribute greatly to the development of new pharmaceuticals.

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