

EVALUATIONS OF SECONDARY METABOLITES ANTIOXIDANT AND ANTIMICROBIAL POTENTIAL IN LEAF EXTRACTS OF *CORBICHONIA DECUMBENS* (FORSSK.) EXELL

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ABSTRACT

Objectives: The present work is to evaluate the presence of phenol and flavonoid contents, 2, 2-Diphenyl-1-picrylhydrazyl (DPPH) activity and antimicrobial activity of *Corbichonia decumbens* leaf extracts.

Methods: *C. decumbens* leaves were collected, shaded, dried, powdered, and were subjected to Soxhlet apparatus for extraction using ethanol and petroleum ether. The ethanol and petroleum ether extract of the plant was tested for phenol, flavonoid, H₂O₂ contents, and DPPH scavenging activity using standard procedures. It is also subjected to antimicrobial activity against pathogenic bacteria and fungus using the well diffusion method.

Results: The result revealed that the leaf extracts contain phenol, flavonoid, and H₂O₂ contents. The values of DPPH radical scavenging activity recorded for ethanol and petroleum ether extract were 55.90±0.10 and 41.23±0.20, respectively. Antimicrobial activities of ethanolic leaf extracts showed lesser values compared to standard values.

Conclusion: The result of this study showed the presence of phenol and flavonoid content, evidenced the potential antioxidant and antimicrobial activity against all tested micro-organisms.

Keywords: *Corbichonia*, Phenol, 2-Diphenyl-1-picrylhydrazyl, Ethanol and Antimicrobial activity.

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INTRODUCTION

Plant can serve as a source of innovative therapeutic agents against infectious diseases [1]. Plant leaves have been used as herbal medicine for their healing properties since ancient times. Some bioactive compounds within these plants are responsible for their medicinal value. The most prominent of these bioactive compounds are alkaloids, tannin, flavonoid, and phenolic compounds [2]. Plants have been endowed with the innate ability to synthesize aromatic substances such as phenols and their derivatives [3], these secondary metabolites have therapeutic tendencies. Pharmacists are interested in these compounds due to their therapeutic performance and low toxicity [4]. Phenolic compounds are common plant secondary metabolites which have not only physiological functions in plants but also positive effects for human health because they can act as antioxidants [5]. Natural antioxidants belonging to the higher plants, especially the typical compounds, such as vitamins, carotenoids, and phenolics exhibit antioxidant activity, and they reduce disease-associated chronic health problems [6]. Medicinal plants represent a rich source of antimicrobial agents. Plants are used medicinally in different countries and are a source of many potent and powerful drugs [7].

METHODS

The fresh leaves of *Corbichonia decumbens* (*Lophiocarpaceae*) were that plant was collected and authenticated by the Botanical Survey of India, Coimbatore, Tamil Nadu. The 25 g of powdered *C. decumbens* leaves were successively extracted using 250 mL of ethanol and petroleum ether using the Soxhlet extractor for 8–10 h.

Determination of secondary metabolites

The total phenolic content of *C. decumbens* in ethanol and petroleum ether extract was performed by referring to the method developed by [8]. The total flavonoid content of the crude extract was determined by the method given [9,10]. The total hydrogen peroxide (H₂O₂) content of the extracts was determined by the method [11].

Antioxidant assay**2, 2-Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity**

The antioxidant activity of the ethanolic and petroleum ether extraction of *C. decumbens* was measured on the basis of the scavenging activity of the stable DPPH free radical according to the standard methods with slight modifications. Inhibition % = $(A_c - A_s) / A_c \times 100$.

Where A_c is the absorbance of the control A_s is the absorbance of the sample.

Antimicrobial assay**Antibacterial and antifungal assay**

Antibacterial activity of the extracts was determined against three bacterial strains, that is, *Bacillus subtilis*, *Klebsiella pneumonia*, and *Salmonella paratyphi* using the well diffusion method. Different concentration of the extracts (50 and 100 µg/ml) was prepared by reconstituting with ethanol. The antifungal activity against two fungal strains (*Candida albicans* and *Aspergillus fumigatus*) was determined by the well diffusion method. The potato dextrose agar plates were inoculated with each fungal culture (10 days old) by point inoculation. Different concentration of the extracts (50 and 100 µg/ml) was prepared by reconstituting with ethanol [11].

RESULTS**Total phenol content**

The Folin-Ciocalteu test was chosen to measure total phenolic content of *C. decumbens* ethanol and petroleum ether extract of the leaves of *C. decumbens*. The ethanolic extract revealed the presence of phenol in a higher amount when compared to petroleum ether extract. The values of phenolic content are recorded in Table 1.

Total flavonoid content

The total flavonoid content of ethanolic and petroleum ether extract of leaves of *C. decumbens* was determined by aluminium chloride method. The highest yield was obtained from ethanolic extract than

the petroleum ether extract. The result was expressed as mg rutin equivalent per gm dry weight. The amount of flavonoid in ethanolic and petroleum ether extracts is recorded in Table 2.

H₂O₂

Levels of H₂O₂ activity are recorded in Table 3. The petroleum ether extract has more activity when compared to ethanol extract, unlike other estimations. In ethanol extract, the values were found to be 0.468±0.01 and in petroleum ether extract.

DPPH activity

The present antioxidant result of the ethanolic and petroleum ether extracts of the leaves of *C. decumbens* showed that the presence of DPPH radical scavenging activity was used to determine the antioxidant activity index. Compared to the petroleum ether extracts, ethanolic extract exhibited considerably significant activity. The antioxidant activities of the ethanol extract were superior to those of the petroleum ether extract. The result is recorded in Table 4.

Antimicrobial assay

Antibacterial activity

Antibacterial activity of leaf extract of *C. decumbens* was tested by the well diffusion method. This activity was tested against the following microorganisms (*Staphylococcus aureus*, *K. pneumonia*, and *S. paratyphi*). The zones of inhibition values are presented in Table 5. In 100% of ethanolic extract showed highest zone of inhibition against micro-organisms such as *S. aureus*, *K. pneumonia*, and *S. paratyphi*. Results are tabulated in Table 5.

Antifungal activity

Similar to antibacterial activity, antifungal activity was also performed by well diffusion. The diameters of the inhibition zones were measured in mm. This activity was tested against two fungal micro-organisms such as *C. albicans* and *A. fumigatus*. For *C. albicans*, the values reported for 50% and 100% zone of inhibition was 02±0.2 and 04±0.26, respectively. For *A. fumigatus*, the respective values for 50% and 100% zone of inhibition was 02±0.13 and 05±0.20, respectively. The ethanol extract of *C. decumbens* leaf exhibited potent antifungal activity toward the tested microbes. Values are recorded in Table 6.

DISCUSSION

Phytochemicals are non-nutritive plant secondary metabolites that prove either protective or disease preventive properties. The result of the qualitative phytochemical study of *C. decumbens* revealed the presence of phenol and flavonoids. In this study, the methanolic and

Table 1: Total phenol content in leaf extracts of *Corbichonia decumbens*

S. No	Extracts	Total phenol content.(mg/g. dr.wt)
1	Ethanol	1.99±0.20
2	Petroleum ether	1.23±0.03

Table 2: Total flavonoid content in leaf extracts of *Corbichonia decumbens*

S. No	Extracts	Total flavonoid content (mg/g.dr.wt)
1	Ethanol	0.205±0.009
2	Petroleum ether	0.140±0.006

Table 3: Hydrogen peroxide content in leaf extracts of *Corbichonia decumbens*

S. No	Extracts	Hydrogen peroxide (µmol/g.dr. wt)
1	Ethanol	0.468±0.01
2	Petroleum ether	0.527±0.03

aqueous extracts of leaves of *Mollugo nudicaulis* were taken for analysis. The result showed significant quantity of total phenol and flavonoids than the ethanolic and petroleum ether extract of *C. decumbens*. This interprets that *M. nudicaulis* leaves possess promising health beneficial phytochemicals compared to *C. decumbens* [12]. The report showed that the ethanolic extracts of leaves of *Solidago Canadensis* have low phenol content compared to the results of *C. decumbens*. The same applies to the total flavonoid content as the result of *C. decumbens* shows a higher value compared to this report [13]. The phytochemical analysis on *Glinus oppositifolius* presented that the methanolic leaf extract reported more phenol and flavonoid content when compared to the ethanolic and petroleum ether extract of leaves of *C. decumbens* and clearly indicates that *G. oppositifolius* is more valuable for therapeutic applications than *C. decumbens* [14]. The study on phytochemical screening of leaf extracts of *Vernonia amygdalina* determined that the methanolic leaf extract of *V. amygdalina* showed the highest total phenol content than other extracts and ethanolic, petroleum ether leaf extracts of *C. decumbens*. As the above result, the total flavonoid content is also higher than the *C. decumbens* [15]. Drugs which quench H₂O₂ radical serve as better therapeutic for the stress-related disorders. The experiment of H₂O₂ scavenging activity on *Crataegus monogyna* showed that the ethanolic extract of *C. monogyna* exhibited very high value when compared with the *C. decumbens* ethanolic and petroleum ether leaf extract [16]. The study on the H₂O₂ scavenging activity of *Costus* after revealed that the methanolic leaf extract showed lower value compared to ethanolic leaf extract of *C. decumbens*. This suggests that ethanolic leaf extract of *C. decumbens* has great potentials being an antioxidant [17]. The results indicate that the water, infusion, and ethanolic extracts of leaves of *Smilax* excels prove to be higher than the values obtained for ethanolic and petroleum ether leaf extract of *C. decumbens*. These results showed that *S.exelsa* leaf extracts had high H₂O₂ scavenging activities [18]. In this study, the ethanolic leaf extract of *Chrysophyllum albidum* has high H₂O₂ scavenging activity than the ethanolic and petroleum ether leaf extract of *C. decumbens* [19].

C. decumbens leaves may constitute an excellent antioxidant property – DPPH scavenging activity. DPPH activity in ethanolic leaf extract has been higher than the petroleum ether extract. Similarly, in the study of leaf extracts of *Chromolaena odorata*, the ethanolic extract revealed

Table 4: 2, 2-Diphenyl-1-picrylhydrazyl scavenging activity in leaf extracts of *Corbichonia decumbens*

S. No	Extracts	% of inhibition
1	Ethanol	55.90±0.10
2	Petroleum ether	41.23±0.20
3	Ascorbic acid (standard)	83.00±0.55

Table 5: Antibacterial activity in leaf ethanolic extract of *Corbichonia decumbens*

S. No	Pathogenic Bacteria	Zone of inhibition (mm)		Standard (ciprofloxacin)
		50%	100%	
1	<i>Staphylococcus aureus</i>	04±0.01	08±0.017	12±0.02
2	<i>Klebsiella pneumoniae</i>	03±0.02	06±0.02	08±0.01
3	<i>Salmonella paratyphi</i>	04±0.01	07±0.01	10±0.02

Table 6: Antifungal activity in leaf ethanolic extract of *Corbichonia decumbens*

S. No	Pathogenic fungus	Zone of inhibition (mm)		Standard (Ciprofloxacin)
		50%	100%	
1	<i>Candida albicans</i>	02±0.02	04±0.02	06±0.01
2	<i>Aspergillus fumigatus</i>	02±0.01	05±0.02	06±0.01

high antioxidant activity than any other extracts [20]. The ethanolic leaf extract of *Leea indica* showed high DPPH activity than the ethanolic leaf extract of *C. decumbens* [21]. The report shows that the petroleum ether extract of *V. amygdalina* is lower than the other extracts and in *C. decumbens*; the ethanolic extract shows high antioxidant activity than the petroleum ether extract [15]. In the study of DPPH activity of plant extract *M. nudicaulis*, the methanolic leaf extract showed values lower than the ethanolic extract of *C. decumbens*. Our result indicated that the ethanolic extract of *C. decumbens* possesses potent antioxidant capacity than these extracts compared [12]. Results on assessment of antimicrobial potency of leaf extracts from *Vitex negundo* and *Gloriosa superba* showed that the ethanolic extract against *Escherichia coli*, *Salmonella typhi*, *S. aureus*, and *C. albicans* showed more effect than the ethanolic extract of *C. decumbens* tested against *S. aureus*, *Klebsiella pneumoniae*, *S. paratyphi*, *C. albicans*, and *A. fumigatus* [22]. The present study on the antimicrobial activity on the ethanolic leaf extract of *C. decumbens* showed lower inhibition zones against the organisms such as *S. paratyphi*, *S. aureus*, *K. pneumoniae*, *C. albicans*, and *A. fumigatus* than the ethanolic leaf extracts of *Garcinia brasiliensis* against *S. aureus*, *Staphylococcus epidermis*, *Streptococcus pyrogenes*, *E. coli*, *Pseudomonas aeruginosa*, *C. albicans*, and *Candida krusei*. Considering antifungal activity in *G. brasiliensis* was observed that none of the extracts induced activity against *C. albicans* in the tested concentrations, but ethanolic leaf extracts of *C. decumbens* leaves have low antibacterial activity than *G. brasiliensis* leaves and possess a little antifungal activity [23]. The demonstration of antimicrobial activity of methanolic leaf extracts of *Justicia adhatoda* possesses potential antibacterial activity against *C. albicans*, *A. flavus*, and *C. neoformans*. These result when compared with the present study; it revealed low antimicrobial activity in *C. decumbens*.

CONCLUSION

The result showed the presence of total phenol, H₂O₂, and flavonoid content. Hence, this plant can be used as a potential source of useful drugs. Antioxidant activity was also done using DPPH scavenging activity and the plant found to have potential antioxidant capacity. The result suggests the presence of good antimicrobial potency in ethanol extract. The plant has been used in traditional medicines to treat skin diseases, kidney stone, tonic, gonorrhoea, and antinociception activity.

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AUTHORS' CONTRIBUTIONS

All the authors contributed equally for the manuscript preparation, and especially edited and final revised by Dr. A. Arunprasath.

CONFLICTS OF INTEREST

The authors declare that no conflicts of interest were involved in this study.

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