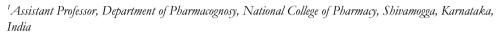
RESEARCH ARTICLE

# Phytochemical Profiling and Evaluation of Anthelmintic Activity of *Cyclea peltata* Leaf Extract

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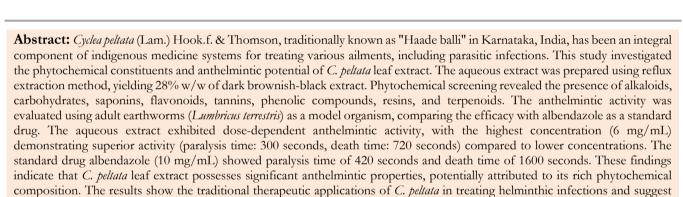


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that it can be a potential candidate as a natural anthelmintic agent.

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# 1. Introduction

Cyclea peltata (Lam.) Hook.f. & Thomson, a climbing shrub belonging to the Menispermaceae family, holds significant ethnomedicinal importance in the Indian traditional healthcare system. Locally known as "Haade balli" or "Pathala beru" in Karnataka, this medicinal plant has been documented extensively in classical Ayurvedic texts including Charakasamhita, Ashtangahridya, and Sushruta Samhita [1]. The plant displays a wide geographical distribution across South and East India, including the Andaman and Nicobar Islands, particularly thriving in evergreen and semi-evergreen forest ecosystems [2]. The plant has garnered substantial attention in traditional medicine for its diverse therapeutic applications. The root system, characterized by its tuberous nature with a starchy cortex and greyish-earthy surface, serves as a primary source of medicinal preparations [3]. Traditional healers have utilized various parts of *C. peltata* for treating conditions ranging from snake bite envenomation to urinary disorders, with particular emphasis on its applications in fever management and diabetic care [4].

C. peltata exhibits distinctive morphological features characteristic of climbing shrubs. The leaves are simple, alternate, and heart-shaped, measuring 2.5-10 cm in length and 2.5-3.75 cm in width, supported by stipules ranging from 5-10 cm. The venation pattern displays 7-11 prominent nerves. The plant produces reniform drupes as fruits, adding to its unique botanical identity [5]. The species are widely distributed across various Indian states. In Maharashtra, it is found in regions including Ahmednagar, Kolhapur, Nasik, Pune, Raigad, Satara, and Sindhudurg. Karnataka hosts populations in Chikmagalur, Coorg, Hassan, Mysore, and Shimoga. The plant maintains a ubiquitous presence across Kerala and is found in specific districts of Tamil Nadu including Coimbatore, Kanniyakumari, Salem, Theni, and Thirunelveli [6]. The ethnomedicinal applications of C. peltata span across various therapeutic domains. The root tuber, consumed in powdered form mixed with diluted curd, has been traditionally prescribed for hemorrhoids treatment over an eight-day regimen [7]. The leaf preparations serve multiple purposes - the paste is utilized for dandruff treatment, while the leaves themselves demonstrate diuretic, antipyretic, and cooling properties [8]. Coastal Karnataka communities specifically employ C. peltata leaves in herpes management [9].

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Figure 1. Leaves of C. peltata

The plant's incorporation into traditional medicine extends to the treatment of various systemic conditions. The root preparations are utilized in managing fever, asthma, gastrointestinal disorders, and jaundice. Additionally, the plant serves as a crucial component in "Shaddharana Choornam," a polyherbal formulation documented in classical Ayurvedic texts [10]. This research focuses on evaluating the anthelmintic potential of *C. peltata* leaf extract, along with comprehensive phytochemical profiling.

#### 2. Materials and Methods

#### 2.1. Collection of Plant Material

Fresh leaves of *Cyclea peltata* were collected from Hirekerur, Karnataka, India, in January 2025. The plant specimens underwent taxonomic authentication at the Department of Botany, S.R.N.M National College of Applied Sciences, Shimoga, Karnataka. A voucher specimen was deposited in the Department of Pharmacognosy, National College of Pharmacy, Shimoga, for future reference.

Table 1. Environmental Conditions During Plant Collection and Processing

Parameter	Value/Range	
Collection season	Winter	
Average temperature (°C)	$22.4 \pm 2.8$	
Relative humidity (%)	$65 \pm 5$	
Altitude (m)	678	
Soil pH	$6.8 \pm 0.3$	
Drying period (days)	7	
Storage temperature (°C)	4 ± 1	

Values expressed as mean ± SD

#### 2.2. Sample Preparation

The collected leaves underwent thorough cleaning with tap water to remove debris and surface contaminants. The cleaned material was subjected to shade drying for seven days under controlled conditions to prevent microbial contamination and preserve phytoconstituents. The dried leaves were reduced to a fine powder using a domestic grinder and stored in airtight containers until further use. [11, 12]

Table 2. Physical Properties of Fresh and Dried Leaf Material

Parameter	Fresh Leaves	Dried Leaves
Moisture content (%)	82.4 ± 2.1	$7.2 \pm 0.8$
Color	Dark green	Olive green
Texture	Smooth	Brittle
Odor	Characteristic	Mild
Surface area (cm <sup>2</sup> /g)	124.6 ± 8.2	$156.8 \pm 10.4$
Thickness (mm)	$0.28 \pm 0.04$	$0.12 \pm 0.02$

Values expressed as mean  $\pm$  SD (n=3)

### 2.3. Extract Preparation

#### 2.3.1. Aqueous Extraction

Ten grams of dried leaf powder underwent reflux extraction with distilled water. The process involved three successive extractions, followed by two additional two-hour extractions. [13, 14] The combined extracts were concentrated using a rotary evaporator under reduced pressure. The final extract yield was calculated using the following formula:

Percentage yield = (Weight of extract obtained / Initial weight of powder) × 100

The concentrated extract was stored in an airtight container at 4°C until further analysis.

# 2.4. Phytochemical Analysis

#### 2.4.1. Alkaloids

The aqueous extract was subjected to alkaloid screening using three standard tests [15, 16]. The Mayer's test was performed by treating the extract with potassium mercuric iodide solution. For Hager's test, the extract was treated with saturated picric acid solution. Wagner's test was conducted using iodine-potassium iodide solution to assess alkaloid presence.

#### 2.4.2. Carbohydrates

Carbohydrate detection employed three standard procedures. In Molisch's test, the extract was treated with  $\alpha$ -naphthol solution followed by careful addition of concentrated sulfuric acid. Benedict's test involved heating the extract with alkaline copper citrate solution. Fehling's test was performed by treating the extract with Fehling's A (copper sulfate solution) and Fehling's B (alkaline sodium potassium tartrate solution) followed by heating. [17, 18]

#### 2.4.3. Proteins

Protein analysis was conducted using two established methods. The Biuret test involved treating the extract with copper sulfate solution under alkaline conditions. The Ninhydrin test was performed using ninhydrin reagent followed by heat treatment to detect amino acids and peptides. [19]

# 2.4.4. Phenolic Compounds and Tannins

The ferric chloride test was employed for phenolic compound and tannin detection, where the extract was treated with ferric chloride solution and observed for color changes.

# 2.4.5. Flavonoids

Flavonoid test was performed using two methods. The first method involved treating the extract with 10% sodium hydroxide solution. The second method employed ferric chloride test, treating the extract with ferric chloride solution. [19]

#### 2.4.6. Glycosides

Glycoside detection test was carried out using the Keller-Kiliani test, where the extract was treated with glacial acetic acid, ferric chloride, and concentrated sulfuric acid. Legal's test was performed using sodium nitroprusside and sodium hydroxide. [20]

#### 2.4.7. Miscellaneous metabolites

Saponin detection was performed using the foam test, which involved vigorous shaking of the diluted extract. Resin screening utilized both acetic anhydride test and turbidity test in acetone-water mixture. Terpenoid detection was conducted using the Salkowski test, involving treatment with chloroform and concentrated sulfuric acid. [21]

#### 2.5. Evaluation of Anthelmintic Activity

# 2.5.1. Test Organism

Adult earthworms (*Lumbricus terrestris*) were selected as the test organism due to their physiological and anatomical similarities with intestinal roundworms. The earthworms were collected locally and acclimatized to laboratory conditions before experimentation. [22]

# 2.5.2. Experimental Design

The evaluation followed a concentration-dependent protocol with test concentrations of 1, 3, and 6 mg/mL of aqueous extract. Albendazole (10 mg/mL) served as the standard drug, while distilled water functioned as control. Six earthworms were utilized per

treatment group to ensure statistical significance. The evaluation focused on two primary parameters. The time taken for paralysis was recorded when no movement was observed except when vigorously shaken. Time of death was noted when worms showed no movement upon gentle mechanical stimulation [23]

#### 3. Results

#### 3.1. Extract Yield

The aqueous extraction of *Cyclea peltata* leaves produced a dark brownish-black extract with characteristic organoleptic properties. The extraction process yielded 3.06 grams of concentrated extract from 10 grams of dried leaf powder, representing a yield of 28% w/w. This significant yield indicates efficient extraction of water-soluble constituents from the plant material, suggesting the potential economic viability of large-scale extraction processes. [24]

Parameter	Value	
Initial powder weight (g)	10.00	
Final extract weight (g)	3.06	
Percentage yield (% w/w)	28.06	
Extract appearance	Dark brownish-black	
Consistency	Semi-solid	
Solubility in water	Highly soluble	
pH (1% solution)	6.8	

**Table 3.** Extraction Yield Data for Cyclea peltata Leaves

#### 3.2. Phytochemical Screening

Phytochemical screening revealed the presence of significant primary metabolites in the aqueous extract. Carbohydrates were consistently detected through multiple confirmatory tests, indicating the presence of both reducing and non-reducing sugars. However, protein screening yielded negative results, suggesting minimal protein content in the aqueous extract. [25]

The extract demonstrated a rich profile of secondary metabolites. Alkaloids showed strong positive results across all three detection methods (Mayer's, Hager's, and Wagner's tests), indicating substantial alkaloid content. This finding aligns with previous reports of alkaloid presence in various Menispermaceae family members, known for their therapeutic properties. [26]

Phenolic compounds and tannins were detected in significant quantities, as evidenced by positive ferric chloride tests. The presence of these compounds suggests potential antioxidant and antimicrobial properties. Flavonoids, detected through both sodium hydroxide and ferric chloride tests, further contribute to the extract's therapeutic potential. Saponins, resins, and terpenoids were also identified in the extract, while glycoside tests yielded negative results. [27]

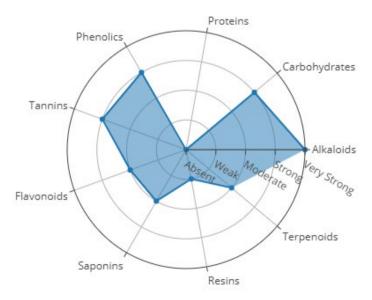


Figure 2. Phytochemical Profile of C.peltata extract

Table 4. Results of Phytochemical screening of Cyclea peltata Aqueous Leaf Extract

Phytochemical Test	Test Test Method Observation		Result
Alkaloids			
- Mayer's test	Cream precipitate	Cream precipitate	++++
- Hager's test	Yellow precipitate	Yellow precipitate	+++
- Wagner's test	Reddish-brown ppt	Reddish-brown ppt	+++
Carbohydrates			
- Molisch's test	Violet ring	Violet ring	+++
- Benedict's test	Orange-red ppt Orange-red ppt		++
- Fehling's test	Brick-red ppt	Brick-red ppt	++
Proteins			
- Biuret test	Violet color	No color change	-
- Ninhydrin test	Purple color No color change		-
Phenolic compounds	Blue-green color	Dark green color	+++
Tannins	Blue-black color	Blue-black color	+++
Flavonoids	Yellow color	Yellow color	++
Glycosides	Brown ring	No ring formation	-
Saponins	Persistent foam	2cm foam layer	++
Resins	Turbidity	Slight turbidity	+
Terpenoids	Reddish-brown color	Reddish-brown color	++

++++ (very strong), +++ (strong), ++ (moderate), + (weak), - (absent)

# 3.3. Anthelmintic Activity

The aqueous extract exhibited concentration-dependent anthelmintic activity against Lumbricus terrestris. At the highest tested concentration (6 mg/mL), the extract demonstrated superior efficacy with paralysis time of 300 seconds and death time of 720 seconds. The medium concentration (3 mg/mL) showed moderate activity with paralysis and death times of 480 and 1220 seconds, respectively. The lowest concentration (1 mg/mL) exhibited relatively slower action, requiring 700 seconds for paralysis and 2000 seconds for death. The reference drug albendazole (10 mg/mL) showed paralysis time of 420 seconds and death time of 1600 seconds. Notably, the highest concentration of *C. peltata* extract (6 mg/mL) demonstrated more rapid action than albendazole, despite being at a lower concentration. This superior activity suggests potential advantages of the plant extract over conventional anthelmintic drugs. [28]

Table 5. Anthelmintic Activity of Cyclea peltata Aqueous Extract Against Lumbricus terrestris

Treatment Group	Concentration	Time to Paralysis	Time to Death	Mortality
Aqueous Extract	1 mg/mL	$700 \pm 45.2$	$2000 \pm 98.6$	100%
	3 mg/mL	$480 \pm 32.8$	$1220 \pm 76.4$	100%
	6 mg/mL	$300 \pm 28.4$	$720 \pm 54.2$	100%
Albendazole	10 mg/mL	420 ± 35.6	1600 ± 82.8	100%
Control	-	-	-	0%

Values expressed as mean ± SEM (n=6)

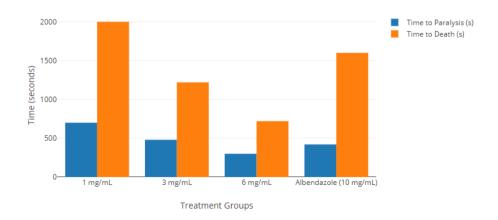


Figure 3. Anthelmintic Activity of C. peltata Extract

#### 3.4. Discussion

The observed anthelmintic activity may be attributed to the synergistic effects of various phytoconstituents identified in the extract. Alkaloids, known for their paralytic effects on nematode nervous systems, likely play a crucial role. Additionally, tannins may interfere with energy generation in helminth parasites by uncoupling oxidative phosphorylation or binding to free proteins in the gastrointestinal tract of the host animal. The exhibited anthelmintic efficacy of *C. peltata* extract, combined with its rich phytochemical profile, supports its traditional use in helminth infections. The rapid action and potentially lower required dosage compared to conventional anthelmintics suggest possible advantages in clinical applications. These findings may contribute to the development of novel anthelmintic formulations based on natural products. [28]

#### 4. Conclusion

The current investigation provides comprehensive scientific validation of *Cyclea peltata*'s traditional therapeutic applications, particularly its anthelmintic properties through systematic phytochemical and pharmacological evaluation. The aqueous extract demonstrated a rich phytochemical profile, containing significant amounts of alkaloids, phenolic compounds, tannins, flavonoids, saponins, and terpenoids, supporting its medicinal value. The extract exhibited potent anthelmintic activity, with the highest concentration (6 mg/mL) showing superior efficacy compared to the standard drug albendazole, suggesting its potential as a natural alternative to conventional treatments. The significant extraction yield (28% w/w) coupled with potent activity at lower concentrations indicates potential economic advantages in therapeutic applications.

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