The Bioscan
AN INTERNATIONAL QUARTERLY JOURNAL OF LIFE SCIENCES
21(1): 57-67, 2026 www.thebioscan.com

PHYTOCHEMICAL SCREENING AND ANTIPYRETIC ACTIVITY OF *DILLENIA INDICA* LEAVE EXTRACT ON RATS

Pooja Sharma*, Abhishek Sharma, Ritesh Malvi, Surendra Kumar Jain

Lakshmi Narain College of Pharmacy, Kalchuri Nagar, Raisen Road, Bhopal, Madhya Pradesh-462021, India

Corresponding Author: Pooja Sharma, **Email:** sharmabittu.ps@gmail.com

DOI: 10.63001/tbs.2026.v21.i01.pp57-67

Keywords
Dillenia indica, phytochemical screening, antioxidant activity, DPPH, antipyretic activity, yeast-induced pyrexia, flavonoids, phenols, herbal medicine

Received on:
11-11-2025

Accepted on:
12-12-2025

Published on:
09-01-2026

ABSTRACT

The present study evaluated the phytochemical composition, antioxidant activity, and antipyretic potential of the ethanolic extract of *Dillenia indica* leaves. The extract showed a yield of 12.5% and tested positive for major phytoconstituents, including flavonoids, phenols, tannins, proteins, and alkaloids. Quantitative analysis revealed measurable levels of total flavonoids (0.63 mg/100 mg) and phenolic content (0.47 mg/100 mg). Antioxidant activity assessed by the DPPH method demonstrated moderate free radical scavenging potential with an IC_{50} value of 96.17 µg/mL. In the yeast-induced pyrexia model, the extract produced a dose-dependent reduction in rectal temperature, with the 350 mg/kg dose showing significant antipyretic activity when compared to the standard drug paracetamol. The findings support the traditional therapeutic use of *Dillenia indica* and indicate its potential as a natural antipyretic agent, warranting further investigation for bioactive compound isolation and detailed pharmacological evaluation.

Introduction

Dillenia indica (Family: Dilleniaceae), commonly known as elephant apple, is a widely used ethnomedicinal plant in Southeast Asia and the Indian subcontinent. Traditionally, different parts of the plant—particularly the leaves and fruits have been used for treating fever, inflammation, digestive disorders, and microbial infections (1,2). The leaves of *Dillenia indica* are rich in diverse phytochemicals such as flavonoids, phenolic acids, saponins, tannins, and triterpenoids, which are known for multiple pharmacological activities including antioxidant, anti-inflammatory, and antipyretic effects (3–5).

Fever is a systemic response triggered by infection, tissue damage, or inflammatory mediators such as prostaglandins. Current antipyretic drugs such as aspirin and paracetamol act primarily

by inhibiting cyclooxygenase (COX) and reducing prostaglandin E_2 synthesis, but they often produce side effects like gastric irritation, hepatotoxicity, and renal complications (6). This has created a growing interest in plant-based antipyretic agents that are safer, economical, and effective.

Several studies have highlighted the biological potential of *Dillenia indica*. Extracts of the fruit and leaves have demonstrated antioxidant, anti-inflammatory, antimicrobial, and analgesic properties (7–9). These activities are closely associated with its phytochemical constituents, particularly flavonoids and phenolics, which can modulate inflammatory mediators and oxidative stress pathways. However, scientific evidence supporting the antipyretic activity of *Dillenia indica* leaf extract remains limited, despite its strong traditional use in fever management.

Therefore, conducting phytochemical screening helps establish the presence of bioactive constituents, while evaluating antipyretic activity in rat models provides experimental validation of its traditional medicinal claims. This study aims to investigate the phytochemical profile and assess the antipyretic potential of the hydroalcoholic extract of *Dillenia indica* leaves using Brewer's yeast-induced pyrexia in rats.

Material and Methods

Material

The present study utilized various analytical-grade chemicals and reagents procured from reputable suppliers. Potassium mercuric iodide, picric acid, ferric chloride, and chloroform were obtained from Thomas Baker, Mumbai**.** Iodine, potassium iodide, sodium nitroprusside, lead acetate, ethanol, Folin–Ciocalteu reagent, and other essential reagents were sourced from Loba Chemie Pvt. Ltd., Mumbai**.** Potassium bismuth iodide, pyridine, gelatin, nitric acid, copper acetate, and sodium chloride were supplied by S. D. Fine Chem. Ltd., Mumbai**.** Methanol and ethanol (analytical grade) were procured from Qualigens Fine Chemicals, Mumbai**,** while Fehling's solution was obtained from Central Drug House Ltd., New Delhi**.** All chemicals used were of laboratory or analytical grade, suitable for phytochemical and biochemical analyses.

Methods

Collection of plant material

The plants have been selected on the basis of its availability and folk use of the plant. Leaves of *Dillenia indica* were collected from local area of Bhopal in the month of February, 2025. Drying

of fresh plant parts was carried out in sun but under the shade. Dried leaves of *Dillenia indica* were preserved in plastic bags, closed tightly and powdered as per the requirements.

Extraction by Maceration

50 gram shade dried leaves was coarsely powdered and subjected to extraction with petroleum ether by maceration process. The extraction was continued till the defatting of the material had taken place. Defatted powdered of *Dillenia indica* has been extracted with ethanol solvent using maceration process for 48 hrs, filtered and dried using vacuum evaporator at 40°C (10). Both the obtained pressed out liquid and the strained solvent are mixed together and separated from unwanted materials by filtration. Frequent agitation during maceration facilitates extraction by two processes: (1) promotes diffusion, (2) separates concentrated solution from the sample surface by adding new solvent to the menstruum for increasing the extraction yield.

Determination of percentage yield

The extraction yield is an assessment of the efficiency of the solvent in extracting bioactive components from the selected natural plant samples and was defined as the quantity of plant extracts recovered after solvent extraction compared to the original quantity of plant samples. The yield of the collected plant extracts was measured in grams after extraction, and then converted into percentage. For calculating the percentage yield of selected plant products, formula following was introduced. By using the following formula the percentage yield of extract was calculated:

$$\text{Percentage yield} = \frac{\text{Weight of Extract}}{\text{Weight of powdered drug}} x100$$

Estimation of total flavonoids content

Determination of total flavonoids content was based on aluminium chloride method (11). 10 mg quercetin was dissolved in 10 ml methanol, and various aliquots of 5- 25µg/ml were prepared in methanol. 10mg of dried extracts of were dissolved in 10 ml methanol and filtered. 3 ml (1mg/ml) of this solution was used for the estimation of flavonoid. 1 ml of 2% $AlCl_3$ methanolic solution was added to 3 ml of extract or standard and allowed to stand for 15 min at room temperature; absorbance was measured at 420 nm.

Estimation of total phenolic content

The total phenolic content of the extract was determined by the modified folin-ciocalteu method. 10 mg Gallic acid was dissolved in 10 ml methanol, various aliquots of 10- 50µg/ml was prepared in methanol. 10 mg of dried extract was dissolved in 10 ml methanol and filter. Two ml (1mg/ml) of this extract was for the estimation of phenol. 2 ml of extract and each standard was mixed with 1 ml of folin-ciocalteu reagent (previously diluted with distilled water 1:10 v/v) and 1 ml (7.5g/L) of sodium carbonate. The mixture was vortexed for 15s and allowed to stand for 10min for colour development. The absorbance was measured at 765 nm using a spectrophotometer (11).

***In-vitro* antioxidant activity using DPPH method**

Total free radical scavenging capacity of extract from *Dillenia indica* estimated according to the previously reported method with slight modification (12). Solution of DPPH (6 mg in 100ml methanol) was prepared and stored in dark place. Different concentration of standard and test (10- 100 µg/ml) was prepared. 1.5 ml of DPPH and 1.5 ml of each standard and test was taken in separate test tube; absorbance of this solution was taken immediately at 517nm. 1.5 ml of DPPH and 1.5 ml of the methanol was taken as control absorbance at 517nm. The percentage inhibition of free radical DPPH was calculated from the following equation:

% inhibition = [(absorbance of control - absorbance of sample)/absorbance of control] × 100%.

***In vivo* anti-pyretic activity using brewer's yeast induced hyperthermia in rats**

Animals

Albino mice (25 -35 g) were used for acute toxicity study and Wistar rats, weighing 150 – 200 g were used for anti-pyretic study. The animals were kept in polypropylene cages in a room maintained under controlled atmospheric conditions. The animals were fed with standard diet (Hindustan liver, Mumbai, India) and had free access to clean drinking water. Pharmacological study was approved by Animal Ethical Committee.

Acute toxicity studies

In the acute toxicity test carried out in mice we take eight doses and 10 mice in each dose of ethanolic extract of *Dillenia indica* i.e. 500, 1000, 1500, 2000 2500 and 3000 mg/kg body weight. All groups of test drug showed neither any toxic effect nor any lethal effect in the dose

range of 500 to 3000 mg/kg body weight. So we had taken a dose 200 mg/kg and 300 mg/kg of body weight for ethanolic extract for further screenings (13-15).

Twenty four male rats were randomly allotted to four groups (6 animals per group). After measuring the rectal temperature of all the rats, hyperthermia was induced by subcutaneous injection of 20% (w/v) aqueous suspension of brewer's yeast. After 18 hours of yeast induction rectal temperatures were measured and only rats those show an increase in temperature by 0.7ºC and more from baseline was used for the study.

Grouping of Animals

Groups I were assigned as vehicle control and administered with Water for Injection (10 ml/kg). Group II were administered with paracetamol (150 mg/kg) and served as positive control. Groups III and IV were administered with ethanolic extract of *Dillenia indica* at the dose of 250 and 350 mg/kg respectively.

Table 1: Study design of the anti-pyretic activity of ethanolic extract of *Dillenia indica* in brewer's yeast induced hyperthermia in rats

Groups	Treatments	Dose (mg/kg)	No: of Animals
Group I	Water for Injection (WFI)	(10 ml/kg)	6
Group II	Paracetamol	150	6
Group III	Ethanolic extract of *Dillenia indica*	250	6
Group IV	Ethanolic extract of *Dillenia indica*	350	6

The temperature was measured at 0 (18 hr after yeast injection), 1, 2, 3 and 4 hrs after administration of doses.

Husbandry Conditions are as follows

Temperature: 20±3°C

Humidity: 30-70%

Light: 12 hours light and 12 hours dark cycle.

Air changes: 12-15 changes per hour

Statistical analysis

Data were analyzed using one way ANOVA followed by Dunnett T method as post-hoc test. All values were reported as mean ± SEM. Statistical significance was set at $p \leq 0.001$.

Results and Discussion

The ethanolic extraction of *Dillenia indica* leaves yielded 12.5% extract**,** as shown in Table 2**,** indicating good extractability of phytoconstituents using ethanol. This high yield reflects the solvent's efficiency in isolating polar and semi-polar compounds.

Phytochemical screening (Table 3) confirmed the presence of several important bioactive groups, including alkaloids, flavonoids, phenols, proteins, carbohydrates, and tannins, while glycosides, diterpenes, and saponins were absent. These constituents particularly flavonoids, phenols, and tannins are widely associated with antioxidant and antipyretic properties.

Quantitative estimation results (Table 4) showed that the ethanolic extract contained 0.63 mg/100 mg of flavonoids and 0.47 mg/100 mg of phenols. These moderate levels support the qualitative findings and suggest that the antioxidant potential of the extract is likely due to these classes of compounds.

The antioxidant activity assessed by the DPPH assay (Table 5) demonstrated that the extract exhibited a concentration-dependent increase in radical scavenging activity, though its IC_{50} value (96.17 µg/ml) was higher than that of ascorbic acid (15.32 µg/ml). While weaker than the standard, the extract still showed a meaningful antioxidant effect attributable to its phenolics and flavonoids.

The initial body weight and baseline rectal temperature of animals (Table 6) did not significantly differ across groups, indicating uniformity and tolerance of the extract. After yeast-induced pyrexia, rectal temperatures increased in all groups (Table 7), confirming successful fever induction.

The antipyretic evaluation across different time intervals (Table 8) revealed that both doses of *Dillenia indica* extract (250 and 350 mg/kg) produced a reduction in rectal temperature. The 350 mg/kg dose demonstrated better temperature-lowering activity, especially at the 2–4 hour marks, although paracetamol remained the most effective treatment. The antipyretic effect of the extract may be associated with inhibition of prostaglandin synthesis, reduction of oxidative stress, and modulation of pyrogenic mediators. When examined together with the observed antioxidant

potential and phytochemical composition (Tables 3–5), the antipyretic activity (Tables 7–8) supports the traditional medicinal use of *Dillenia indica* leaves for fever management.

Table 2: Percentage yield of *Dillenia indica* extract

S. No.	Extract	Percentage yield
1.	Ethanolic	12.5%

Table 3: Phytochemical screening of extract of *Dillenia indica*

S. No.	Constituents	Ethanolic extract
1.	**Alkaloids**	
	Mayer's Test	-ve
	Wagner's Test	+ve
	Dragendroff's Test	+ve
2.	**Glycosides**	
	Legal's Test	-ve
3.	**Flavonoids**	
	Lead acetate test	+ve
	Alkaline test	-ve
4.	**Phenol**	
	Ferric chloride test	+ve
5.	**Proteins**	
	Xanthoproteic test	+ve
6.	**Carbohydrates**	
	Molisch's Test	-ve
	Benedict's Test	-ve
	Fehling's Test	+ve
7.	**Saponins**	
	Froth Test	-ve
8.	**Diterpenes**	
	Copper acetate test	-ve
9.	**Tannins**	

	Gelatin Test	+ve

[+ve= positive; -ve= negative]

Table 4: Estimation of total flavonoids and phenol content of extract of *Dillenia indica*

S. No.	Extract	Total flavonoids content (mg/ 100 mg of dried extract)	Total phenol content (mg/ 100 mg of dried extract)
1.	Ethanolic	0.63	0.47

Table 5: % Inhibition of ascorbic acid and ethanolic extract of *Dillenia indica* using DPPH method

S. No.	Concentration (µg/ml)	% Inhibition	
		Ascorbic acid	Ethanolic extract
1	10	41.61	14.96
2	20	57.43	25.74
3	40	64.02	30.55
4	60	70.96	38.15
5	80	78.35	42.43
6	100	85.57	50.86
IC_{50} value		**15.32**	**96.17**

Table 6: Effect of ethanolic extract of *Dillenia indica* on initial rectal temperature and body weight in albino winstar rats

Groups	Treatments	Dose (mg/kg b.wt.)	Body Weight of Rats (g)	Initial Rectal Temperature (°C) before yeast injection
I	Water for Injection (WFI)	0	195.1 ± 2.85	37.48 ± 0.12
II	Paracetamol	150	192.6 ± 3.74	37.19 ± 0.16

III	Ethanolic extract of *Dillenia indica*	250	187.2 ± 3.15	37.25 ± 0.15
IV	Ethanolic extract of *Dillenia indica*	350	190.3 ± 3.82	37.34 ± 0.18

Table 7: Effect of ethanolic extract of *Dillenia indica* on rectal temperature after 18 hrs of yeast injection in albino Winstar Rats

Groups	Treatments	Dose (mg/kg b.wt.)	Temperature (°C) 18 hrs after Yeast Injection (0 h)
I	Water for Injection (WFI)	–	39.08 ± 0.25
II	Paracetamol	150	38.89 ± 0.21
III	Ethanolic extract of *Dillenia indica*	250	38.84 ± 0.25
IV	Ethanolic extract of *Dillenia indica*	350	39.09 ± 0.23

Table 8: Effect of ethanolic extract of *Dillenia indica* on rectal temperature in different time intervals

Groups	Treatments	Dose (mg/kg b.wt.)	1 hour (°C)	2 hour (°C)	3 hour (°C)	4 hour (°C)
I	Water for Injection (WFI)	0	39.12 ± 0.14	39.18 ± 0.12	38.95 ± 0.10	38.76 ± 0.17
II	Paracetamol	150	38.42 ± 0.16	38.06 ± 0.18**	36.87 ± 0.15***	35.64 ± 0.12***
III	Ethanolic extract of *Dillenia indica*	250	38.91 ± 0.22	38.48 ± 0.24	38.27 ± 0.22*	38.18 ± 0.22
IV	Ethanolic extract of *Dillenia indica*	350	38.70 ± 0.21	38.38 ± 0.17*	38.13 ± 0.18*	37.81 ± 0.17**

Conclusion

The ethanolic extract of *Dillenia indica* showed a good yield and contained key phytochemicals such as flavonoids, phenols, tannins, and alkaloids. It exhibited moderate antioxidant activity in the DPPH assay. In the yeast-induced pyrexia model, the extract produced a dose-dependent reduction in rectal temperature, with the 350 mg/kg dose showing the best effect. Although less

effective than paracetamol, the extract demonstrated significant antipyretic potential, supporting its traditional medicinal use. Further studies are needed to isolate active compounds and confirm its safety.

References

1. Usha K, Kumar PS, Bhatnagar SP. Traditional uses of *Dillenia indica* in Indian folk medicine. J Ethnopharmacol. 2010;128:22–25.
2. Singh R, Singh S, Kumar S. Ethnomedicinal relevance of *Dillenia indica*. Int J Pharm Sci Rev Res. 2012;16:45–49.
3. Sharma A, Sharma P. Phytochemical screening of *Dillenia indica* leaves. Asian J Pharm Clin Res. 2014;7(3):210–213.
4. Hussain H, Ahmed I, Schulz B, Draeger S, Krohn K. Chemical constituents of *Dillenia* species and their pharmacological activities. Phytochemistry. 2012;85:123–131.
5. Das S, Dash S. GC-MS and phytochemical profile of *Dillenia indica* leaf extract. Int J Pharm Bio Sci. 2013;4:350–357.
6. Rang HP, Dale MM, Ritter JM. *Pharmacology*. 6th ed. Churchill Livingstone; 2008.
7. Gupta M, Mazumder UK, Gomathi P, Selvan VT. Antioxidant and anti-inflammatory activities of *Dillenia indica* fruit extract. J Pharm Res. 2011;4(3):720–722.
8. Devi RK, Indira M. Evaluation of antimicrobial activity of *Dillenia indica* leaf. Int J Pharm Sci. 2013;5:110–113.
9. Ghosh A, Das BK. Analgesic potential of *Dillenia indica* extracts. Pharmacogn Res. 2014;6(2):141–145.
10. Mukherjee PK. *Quality Control of Herbal Drugs*. 2nd ed. Business Horizons; 2007. p. 2–14.
11. Mishra AG, Singh R, Meha P, Parkhe G. Determination of total phenolic, flavonoid content, antioxidant and antimicrobial activity of *Gloriosa superb* seed extract. Asian J Pharm Educ Res. 2017;6(2):12–17.
12. Parkhe G, Jain P. Study of antioxidant potential of hydroalcoholic extract of *Anethum graveolens*. Career Int J Sci Technol. 2018;1(2):39–45.
13. Niazi J, Gupta V, Chakraborty P, Kumar P. Anti-inflammatory and antipyretic activity of *Aleuritis moluccana* leaves. Asian J Pharm Clin Res. 2010;1:35–37.

14. Hajare SW, Chandra S, Tandan SK, Sharma J, Lal J, Telang AG. Analgesic and antipyretic activity of *Dalbergia sissoo* leaves. Indian J Pharmacol. 2000;32:357–360.
15. Cheng L, Ming-liang H, Lars B, et al. Is COX-2 a perpetrator or a protector? Selective COX-2 inhibitors remain controversial. Acta Pharmacol Sin. 2005;26:926–933.