



In Vitro* Anthelmintic Efficacy of *Spathodea campanulata* P. Beauv. (Bignoniaceae) Against *Pheretima posthuma

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Abstract: To investigate the anthelmintic activity of leaves of *Spathodea campanulata* P. Beauv. (Bignoniaceae) against *Pheretima posthuma* (Indian earthworms). The methanolic extract of the leaves of *Spathodea campanulata* P. Beauv. (Bignoniaceae) was investigated for anthelmintic efficacy against Indian adult earthworms (*Pheretima posthuma*). Earthworms were grouped and treated with extracts at 5, 15 and 20 mg/ml concentration, albendazole as a standard and normal saline as control. The paralysis time and death time of earthworms is considered as a indicator for anthelmintic activity. Methanolic extract of leaves of *Spathodea campanulata* showed dose dependent anthelmintic activity but significant activity was observed at 20 mg/ml concentration. At 20 mg/ml concentration extract, showed better activity with paralysis time (4.23 min) and death time (10.32 min) when compared to standard drug albendazole. The study reveal that the methanolic extract of *Spathodea campanulata* P. Beauv. leaves has significant anthelmintic activity against Indian adult earthworms.

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Keywords: *Spathodea campanulata*; Anthelmintic activity; Albendazole; Methanolic extracts

INTRODUCTION

Helminthiasis is one of the major cause of morbidity and mortality worldwide. It is most common infection in tropical and subtropical countries. Globally, intestinal parasitic infections including helminths and protozoa, remain endemic. About one-third of the world's populations, more than 2 billion people, are infected with the largest majority being children. Helminths parasites are mostly *Ascaris lumbricoides*, *Trichuris trichiura* and hookworms. *A. lumbricoides* is the largest and most common helminths, infecting about 819 million people globally; this is followed by *T. trichiura*, which infects about 464.6 million, and hookworm infecting about 438.9 million people globally (Vincent P



et al., 2019). In developing countries, they pose a large threat to public health and contribute to the prevalence of malnutrition, anemia, eosinophilia and pneumonia. Hence, the increasing prevalence of helminth parasites those are resistant to conventional anthelmintic has been the spur for different research programs exploiting alternative approaches to parasite control (AnujKumar A et al., 2011). Medicinal plants serve as an important therapeutic agent as well as valuable raw material for manufacturing numerous traditional medicines and also acts as the lead for modern medicines. The history of medicinal plants uses for treating disease and ailments is probably dated back to human civilization. *Spathodea campanulata* P. Beauv. tree is native of Africa and it is widely grown in tropical and subtropical regions outside Africa. This plant is also commonly found in India as an ornamental plant. *Spathodea campanulata*, a perennial tree belonging to family Bignoniaceae commonly known as African Tulip tree, fire ball, fountain tree, flame of the forest and Pichakari. It is a large upright tree with a spreading crown and a slightly buttressed trunk. The branches are thick and marked with small whitish-coloured corky spots. Younger branches vary from being almost hairless to having a sparse covering of small hairs. The large leaves are up to 50 cm long, compound with 7-17 leaflets. These leaves are usually oppositely arranged along the stems. The large, orange-red flowers are arranged in dense clusters (8-10 cm long) at the tips of the branches. The reddish-orange coloured petals are also fused together and are shaped somewhat like a tulip flower i.e. tubular. Flowering occurs throughout the year, but usually peaks during spring. The fruits are long, elongated capsules resemble pods. When mature they split open and release about 500 papery seeds. These seeds are very light and surrounded by membranous wing (Orwa C & Mutua A, 2009). Plant parts such as flowers, leaves stem, bark and roots have been reported for possessing anti-inflammatory, analgesic, cytotoxic, anti-solar anti anticonvulsant activity (Wagh A & Butle S, 2017). Phytochemical screening shows the presence of alkaloids, tannins, flavonoids, glycosides and sterols (Ngouela S, 1988). Almost all parts of the plants are used traditionally. Literature survey reveals that till date no reports were found on the anthelmintic activity of the leaves of *Spathodea campanulata* P. Beauv. Hence the present study was carried out to investigate the efficacy of methanolic extract of the leaves of *Spathodea campanulata* P. Beauv.

MATERIALS AND METHODS

Plant Material collection and authentication: Leaves of *Spathodea campanulata* P. Beauv. were collected from Nanded District, Maharashtra and authenticated from Botanical Survey of India, Pune.. All the reagents and chemicals were purchased from Merck chemicals Ltd.

Preparation of extract: The leaves were washed with distilled water, shade dried and powdered. Powdered drug material was subjected to soxhlet solvent extraction with methanol. The extracts were filtered, evaporated and stored at 40°.

Phytochemical screening: Preliminary phytochemical screening was carried out for the detection of the type of major phytoconstituents present. The phytochemical study of the extract was carried out using standard procedures (Tiwari P et al., 2011, Kokate C et al., 2007, Khandelwal K, 2006).

Worm Collection and authentication: Indian adult earthworms (*Pheretima posthuma*) were used to study anthelmintic activity of the plant extracts. The adult earthworms were collected from Vishnupuri, Nanded, Maharashtra, India. Worms were authenticated at Department of Life Science, S R T M University, Nanded, India. The earthworms obtained resembled with intestinal roundworm parasites of human beings both anatomically and physiologically and hence were considered for anthelmintic activity (Prasad J, 2014).



Preparation of test drug and reference drug: Extracts for *in vitro* study were prepared at the concentrations of 5, 15 and 20 mg/ml. Samples of methanolic extract were prepared by dissolving 100, 200, and 500 mg crude extract of each in 1 ml dimethyl sulfoxide (DMSO) and made the volume up to 10 mL with normal saline solution and final concentration of samples achieved were 5, 15, and 20 mg/mL, respectively. Normal saline solution was used as control and albendazole was used as the standard drug for this study.

Anthelmintic activity: Anthelmintic activity of methanolic extract of the leaves of *Spathodea campanulata* was evaluated on Indian earthworms. Five groups of Indian earthworms, each containing six earthworms approximately of equal size were used for the study. Three groups were tested with extract of different concentration (5 mg/ml, 15 mg/ml, 20 mg/ml), one group were treated with albendazole, as a reference standard and one group was treated with normal saline solution as control group. The anthelmintic activity on earthworms was observed and time for paralysis and death were recorded. (Hussain A et al., 2012, Hapse S et al., 2012, Rubini D et al., 2012, Anubha S et al., 2017)

RESULTS

Preliminary phytochemical investigation of extract revealed the presence of tannins, phenolic compound, flavonoids, steroidal saponins, alkaloids. The effect of different concentrations of methanolic extract *Spathodea campanulata* leaves and albendazole on *pheretima posthuma* is showed in Table 1. The dose dependent onset of paralysis and mortality were observed in the earthworms treated with the extract which was compared with albendazole as reference drug. The methanolic extract at 5, 15, 20 mg/mL concentrations showed paralysis time as 28.05, 20.34, 4.23 and death time as 32.51, 29.17, 10.32 min respectively (Figure 1). At highest concentration it produces paralysis and death in short time which is comparable with albendazole (Figure 1C). The albendazole treated group at concentration 10 mg/mL showed the paralysis time 20.14 min and death time 29.49 min (Figure 2). The normal saline solution treated earthworms have not showed any change in physical activity and remained active with whole body movements. No paralysis or Mortality was observed in normal saline treated earthworms (Figure 3).

Table-1 In-vitro effect of different concentration of methanolic extract of the leaves of *Spathodea campanulata* and albendazole on survival of Indian earthworms

Control/Albendazole/ Extract	Concentration (mg/ml)	Time taken in minutes	
		Paralysis	Death
Control	Normal saline	-	-
Albendazole	10	20.14±0.2	29.49±0.2
Extract	5	28.05±0.02	32.51±0.008
	15	20.34±0.02	29.17±0.01
	20	4.23±0.1	10.32±0.01

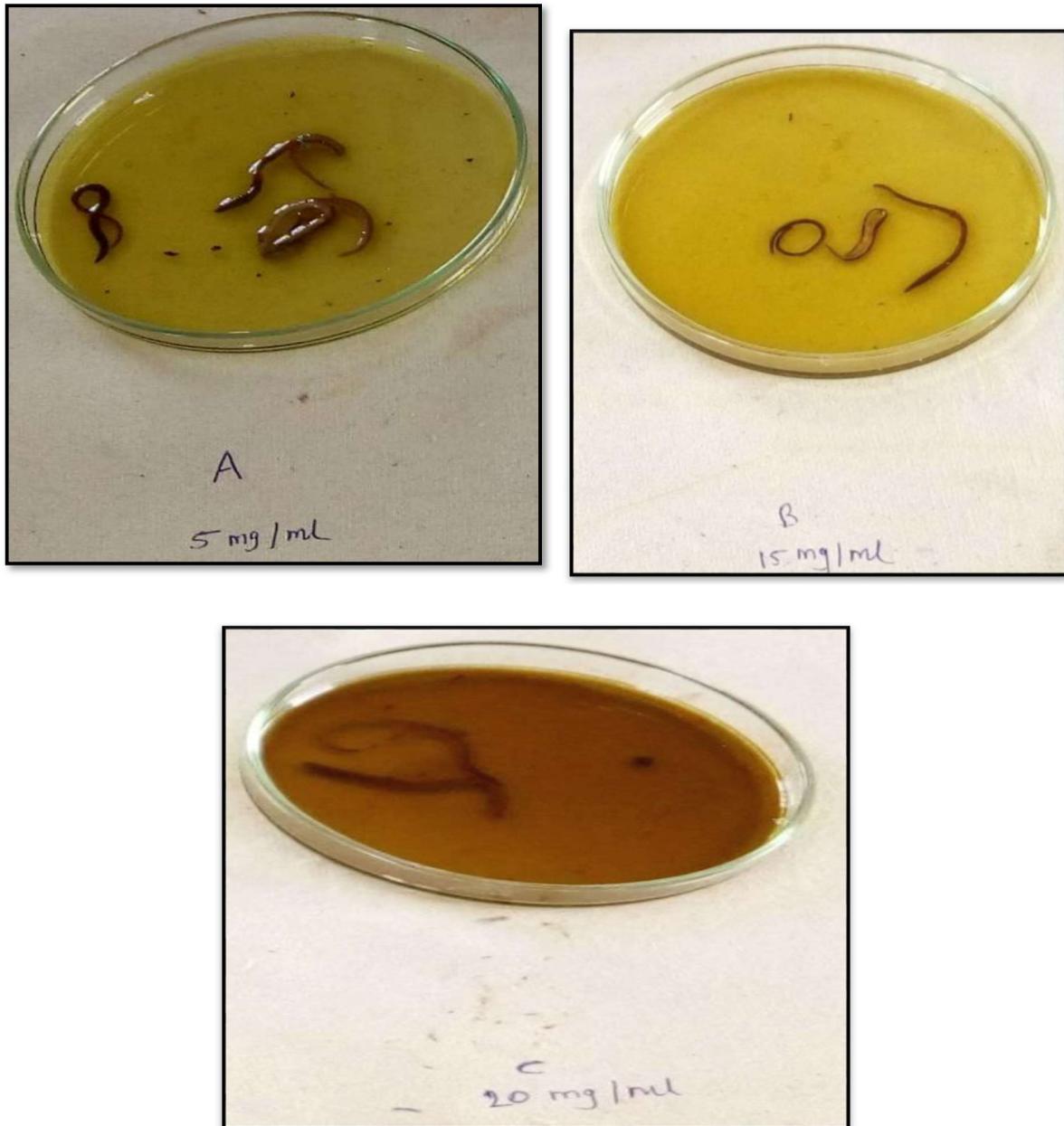


Figure 1. Effect of methanolic extract of *Spathodea campanulata* leaves on paralysis and mortality time for 5 mg/mL (A), 15 mg/mL (B) and 20 mg/mL (C).



Figure 2. Effect of Standard drug Albendazole on paralysis and mortality time for 10 mg/mL



Figure 3. No paralysis or mortality was observed in normal saline treated earthworms

The present study revealed that methanolic extract of leaves of *Spathodea campanulata* exhibits promising in vitro anthelmintic activity. This activity may be due to the presence of various phytoconstituents like tannins, saponins, alkaloids, flavonoids. Further in vivo study is required for evaluation of *Spathodea campanulata* for its effectiveness and pharmacological



rationale as anthelmintic agent. Detailed phytochemicals investigation is needed for the development of new potential drug from leaves of *Spathodea campanulata*.

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CONFLICT OF INTEREST STATEMENT

We declare that we have no conflict of interest.

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