



## International Journal of Preclinical & Pharmaceutical Research

Journal homepage: [www.preclinicaljournal.com](http://www.preclinicaljournal.com)

### IN-VITRO ANTHELMINTIC ACTIVITY OF *COUROUPITA GUIANENSIS* LEAVES IN INDIAN ADULT EARTHWORM

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#### ABSTRACT

Different extracts of *Couroupita guianensis* were taken for anthelmintic activity against Indian earthworm *Pheretima posthuma*. Two concentrations (50 and 100 mg/ml) of various extracts were tested and results were expressed in terms of time for paralysis and time for death of worms. Albendazole (20 mg/ml) was used as reference standard and carboxy methyl cellulose (0.5%) as a control group. Dose dependent activity was observed in the plant extracts but methanolic extract exhibited more activity as compared to others. The anthelmintic activity of *Couroupita guianensis* leaves extract has therefore been demonstrated for the first time.

**Key Words:** Anthelmintic, *Couroupita guianensis*, *Pheretima posthuma*, Methanolic extract.

#### INTRODUCTION

Helminthes infections, repeatedly entitled helminthiasis are among the most pervasive infection and a foremost degenerative disease distressing a large proportion of world's population. In developing countries, they pose a large threat to public health and contribute to the prevalence of malnutrition, anemia, eosinophilia and pneumonia [1]. The helminths parasites mainly subsist in human body in intestinal tract, but they are also found in tissue, as their larvae migrate towards them. Most diseases caused by helminths are of a chronic, debilitating nature; they probably cause more morbidity and greater economic and social deprivation among humans and animals than any single group of parasites [2]. Chemical control of helminthes coupled with improved management has been the important worm control strategy throughout the world. However, development of resistance in helminthes against conventional anthelmintics is a foremost problem in treatment of helminthes diseases. Henceforth it is important to look for alternative strategies against gastrointestinal nematodes, which have led to the proposal of screening medicinal plants for their anthelmintic activity [3].

*Couroupita guianensis* Aubl belonging to the family Lecythidaceae. It is widely cultivated for its large showy flowers and reddish - brown woody capsular fruits upto 20 cm in diameter. It is grown in Indian gardens as an ornamental tree [4]. It is native to South India and Malaysia and is commonly known as Nagalinga pushpam in Tamil. Previous work on *Couroupita guianensis* has shown that plant contains several chemical constituents with novel structures and possesses bio-active moieties. These include eugenol, linalool, fernesol, nerol, tryptanthrine, indigo, indirubin, isatin, linoleic acid,  $\alpha$ ,  $\beta$ -amirins, carotenoids, sterols and some acidic and phenolic compounds. Traditionally, the leaves of this plant have been used in the treatment of skin diseases, stomachache, and intestinal gas formation, antithrombotic and vasodilatory actions [5].

#### MATERIALS AND METHODS

##### Plant material

Fresh leaves were collected from the vicinity of adamangalam village, India during the month of January-February 2013. The plant was verified in the department of pharmacognosy, Anurag Pharmacy College, India for its authenticity by comparing with the standard herbarium before the study.

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### Experimental worms

All the experiments were carried out in Indian adult earthworms (*Pheretima posthuma*) due to its anatomical resemblance with the intestinal roundworm parasites of human beings. They were collected from moist soil and washed with water to remove all fecal matters.

### Preparation of Extracts

The leaves of *Couroupita guianensis* were dried under shade and crushed in an electric blender to form coarse powder and subjected to Soxhlet extraction (Continuous hot extraction) by using methanol and water as solvent. The extracts were concentrated by rotary evaporator and used for testing anthelmintic activity. Preliminary phytochemical screening was carried out to assess the presence of phytoconstituents in the extract.

### Administration of Albendazole

Albendazole (20 mg/ml) was prepared by using 0.5% w/v of CMC as a suspending agent as administered as per method of extract.

### Administration of extract

The suspension of Methanolic and aqueous extract of leaves of *Couroupita guianensis* of different concentration (50,100mg/ml) were prepared by using 0.5%

w/v of CMC as a suspending agent and final volume was made up to 10 ml for respective concentration. Albendazole was used as standard. Groups of approximately equal size worms consisting of two earthworms individually in each group were released into in each 10 ml of desired concentration of drug and extracts in the petridish.

### Experimental Design

The anthelmintic activity was performed according to the method[6]. On adult Indian earth worm *Pheretima posthuma* as it has anatomical and physiological resemblance with the intestinal round worm parasites of human beings. *Pheretima posthuma* was placed in petridish containing two different concentrations (50 &100mg/ml) of methanolic & aqueous extract of leaves of *Couroupita guianensis*. Each petridish was placed with 2 worms and observed for paralysis or death. Mean time for paralysis was noted when no movement of any sort could be observed, except when the worm was shaken vigorously; the time death of worm (min) was recorded after ascertaining that worms neither moved when shaken nor when given external stimuli. The test results were compared with Reference compound Albendazole (20 mg/ml) treated samples.

**Table 1. Anthelmintic potency of methanolic and aqueous extract of *Couroupita guianensis***

GROUP	CONCENTRATION USED (mg/ml)	TIME TAKEN FOR PARALYSIS (min)	TIME TAKEN FOR DEATH (min)
Control (0.5% CMC)	-	-	-
Standard (Albendazole)	25	28.71 ± 1.86	58.90 ± 6.85
Methanol extract	25	54.22 ± 2.95	104.3 ± 3.76
	50	44.90 ± 2.59	89.03 ± 2.25
Aqueous extract	25	63.25 ± 8.10	105.2 ± 11.97
	50	45.55 ± 2.05	86.66 ± 4.69

All Values represent Mean± SD; n=6 in each group. Comparisons made between standard versus treated groups, P<0.05 was considered significant

### RESULTS AND DISCUSSION

Preliminary phytochemical analysis of methanolic extracts showed the presence of Flavonoids, Saponins, Tannins, Steroids, Terpenoids & Alkaloid whereas aqueous revealed the Tannins, Steroids & Alkaloid active phytoconstituents. The data revealed that the methanol extract showed anthelmintic activity at a concentration of 100 mg/ml, whereas the aqueous extract also showed paralysis and death at similar concentrations. The other test concentrations of both the extracts showed marked degree of anthelmintic activity. The anthelmintic effect of extracts is comparable with that of the effect produced by the standard drug albendazole. Parasitic helminths affect animals and man, causing considerable hardship and stunted growth. Hundreds of millions if not billions of human infections by helminthes exist worldwide and increased world travel and immigration from the

developing countries. However tremendous advances has been made during the previous decade and substantial number of synthetic precursors have been derived to cope up the damage caused by parasite, but unfortunately no effective medicine has been developed so far. Moreover the problems associated with the use of such drugs like some serious side effects and development of resistance drives the severity of infection to the next level. These factors paved the way for herbal remedies as alternative anthelmintics. Evaluation of activities of medicinal plants claimed for possessing the anthelmintic property is getting the attention these days. Screening and proper evaluation of the claimed medicinal plants could offer possible alternatives that may be both sustainable and environmentally acceptable. The results of this study have shown promising anthelmintic activity suggesting the possible use of *Couroupita guianensis* extracts in intestinal

nematode control. The anthelmintic activity of methanol extracts could be due to the constituents present. The present study suggested that the methanol extract was more effective than the other extracts, even though all the extract were endowed with anthelmintic property. The activity was concentration dependent of the different extracts. The activity of the extracts was found to be inversely proportional to the time taken for paralyse / death of the earth worms.

### CONCLUSION

The results of the present study clearly indicated that the crude methanol extract of *Couroupita guianensis* did produce anthelmintic activity against Indian earthworm

*Pheretima posthuma*. The plant possesses significant anthelmintic activity at 100 mg/ml concentration measured by time taken for paralyse / death of the earth worms. The current investigation leads to conclusion that the leaves of *Couroupita guianensis* have potent anthelmintic activity when compared with the conventionally used drug. The results did not, however, exclude the possibility that doses of the extract with lower anthelmintic activity in this study might be efficacious against other species of helminths. Further studies using in vivo models and to isolate active constituents from extract are required to carry out and established the effectiveness and pharmacological rational for the use of *Couroupita guianensis* as an anthelmintic drug.

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