



IN-VITRO EVALUATION OF ANTHELMINTIC ACTIVITY OF ETHANOLIC EXTRACT OF *ALTERNANTHERA SESSILIS* LINN

Mrinmay Das*¹ and Yadgiri Phalgun¹

¹Department of Pharmacy, Bharat Institute of Technology, Mangalpally, Ibrahimpatnam, Hyderabad, Telengana, India.

*Corresponding Author: Mrinmay Das

Department of Pharmacy, Bharat Institute of Technology, Mangalpally, Ibrahimpatnam, Hyderabad, Telengana, India.

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ABSTRACT

Alternanthera sessilis Linn. ('Joyweed'), a member of Amaranthaceae family is a weed and occurs in both wetlands and uplands on variety of soil types. *Alternanthera sessilis* is a popular leafy vegetable in Sri Lanka and also used as traditional medicine in China, India and Sri Lanka. The herb has been reported to be used as galactagogue, cholagogue, febrifuge and indigestion. The present study was aimed to investigation of the anthelmintic potential of crude ethanolic extract of *Alternanthera sessilis* Linn. on Indian earth-worm (*Pheretima posthuma*). Three concentrations (25, 50, 100 mg/ml) of each extract were studied in activity which involved the determination of time of paralysis (vermifuge) and time of death (vermicidal) of the worms. Albendazole in 25mg/ml concentration was included as standard reference and normal saline with 1% CMC as control. The ethanolic extracts exhibited significant anthelmintic activity at a concentration of 100 mg/ml. The present investigation confirms that, the ethno-medicinal claim of anthelmintic activity of this plant is genuine.

KEYWORDS: *Alternanthera sessilis*, Indigestion, Anaemia, Diarrhoea, Inflammation, Hair Tonic.

INTRODUCTION

Parasitic infection including Helminthiasis is a critical serious problem in the tropical regions including the Asian countries which affects more than two billions of people worldwide. Helminthes produce serious problem in human and other animals around the world specifically to the third world countries. Different type of helminthes infects the human and animals out of which intestinal round worms (*Ascaridia* sp.) are most common. Approximately 300 million people suffer severe morbidity associated with these parasites and half of which are school-going children affected by massive infections. Variety of several clinical symptoms arises due to this infection include dysentery, diarrhoea, nausea-vomiting, loss of appetite and weight, acidity and sometimes anaemia. Other manifestations of helminthic infections include respiratory symptoms, dermatological consequences and epilepsy as a result of neurocysticercosis. Helminthic infections may also subvert immune responses to pathogens of other diseases such as tuberculosis, HIV, and malaria.^[1]

Alternanthera sessilis Linn. (Amaranthaceae) is an annual or perennial prostrate herb with several spreading branches, bearing short petioled simple leaves and small white flowers, found throughout the hotter part of India, ascending to an altitude of 1200m. The plant spreads by seeds, which are wind and water-dispersed and by

rooting at stem nodes. Young shoots and leaves are eaten as a vegetable in Southeast Asia.^[2] It is a weed of rice throughout tropical regions and of other cereal crops, sugarcane and bananas. Although it is a weed, it has many utilities. The leaves were used in eye infections, cuts, antidote to snake bite and skin diseases.^[3] It was also reported that the plant *Alternanthera sessilis* shown wound healing property.^[4] The degenerative and necrotic changes in the liver and kidney in Swiss mice, caused by oral administration of water extract of *Alternanthera sessilis* in high doses through histopathological test were revealed.^[5] Thus, the present investigation was aimed to evaluate the anthelmintic activity of ethanolic extracts of aerial parts of *Alternanthera sessilis* Linn.

MATERIALS AND METHODS

Plant materials

The fresh *Alternanthera sessilis* Linn. plant was collected nearly 2.0 kg from the rural belt of the Jangalakandriga Village, Nellore Dist., Andhra Pradesh, India. The plant was authenticated by Dr. C.V.S. Bhaskar, Ph. D, Director and Principal, Department of Botany, Venkata Raja's College, Nellore Dist, Andhra Pradesh, India. A voucher specimen number 12 (*Alternanthera sessilis* Linn.) had kept in Department of Pharmacognosy for future references.

Preparation of extract

Extraction is a process where the main focus is on the materials to be extracted and the type(s) of the compound that is being isolated. The best way to get the extract is boiling the plants parts in ethanol. The classical chemical procedure for obtaining organic constituents from dried plant tissue is to continuously extract powdered material in a soxhlet apparatus within a range of solvent, starting in turn with Hexane, Petroleum ether, Chloroform, Ethyl acetate, Ethanol, Methanol etc. This method is useful when working on the gram scale. But it is rarely possible to complete separation of constituents and the same compounds may be recovered in several fractions.^[6]

Extraction of aerial parts of *Alternanthera sessilis* Linn.

Alternanthera sessilis Linn. plant was collected, then they were separated out from unwanted ingredients and washed with plenty of water. Then the plants were dried in shade and then milled in to coarse powder by a mechanical grinder. The powder was kept in air tight container for further use. The powdered material was then packed in Soxhlet Apparatus and continuous hot extraction was carried out using Ethanol as solvent. The extract was filtered through Whatman filter paper to remove any impurities if present. The extract was then concentrated by using vacuum distillation unit. The concentrated extract was placed in vacuum desiccators to remove the excess moisture. The dried extract was weighed and the yield of the extract was calculated as 14.8% w/w. The extracts were then kept in airtight container for further use.

Anthelmintic activity study

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.

Table: 1 Percentage Yields of Ethanolic Extract of *Alternanthera sessilis* Linn.

Sl. No.	Name of the Extract	Percentage Yield (%w/w)
1.	Ethanol	14.8%

Anthelmintic Activity study

The anthelmintic activity was performed on adult Indian earth worm *Pheretima posthuma* as it has anatomical and physiological resemblance with the intestinal round worm parasites of human beings.

Table 2: Anthelmintic activity study of *Alternanthera sessilis* Linn.

Sl. No.	Compounds	Concentration	<i>Pheretima posthuma</i>	
			Paralysis	Death
1.	Control	--	--	--
2.	Ethanolic extract of <i>Alternanthera sessilis</i> Linn.	25 mg/ml	41.66 ± 0.661	65.01 ± 1.061
3.		50 mg/ml	21.50 ± 0.421	42.16 ± 0.793
4.		100 mg/ml	12.56 ± 1.491	15.83 ± 1.641
5.	Standard (Albendazole)	25 mg/kg	25.01 ± 1.155	64.06 ± 0.881

All values represent Mean ± SD; n = 2 in each group, Comparisons made between standard Vs treated groups, p < 0.005 was considered significant

Administration of extract

The suspension of Ethanolic extract of *Alternanthera sessilis* Linn. of different concentration (25, 50 & 100 mg/ml) were prepared by using 1.0 % w/v of CMC as a suspending agent and final volume was made up to 20 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 20 ml of desired concentration of drug and extracts in the petridish.

Administration of Albendazole

Albendazole (25 mg/ml) was prepared by using 1.0 % w/v of CMC as a suspending agent as administered as per method of extract.

Experimental design

The anthelmintic activity was performed according to the method.^[7] *Pheretima posthuma* was placed in petridish containing three different concentrations (25, 50 & 100 mg/ml) of ethanolic extract of *Alternanthera sessilis* Linn. 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 (25 mg/ml) treated samples.

RESULTS

Percentage yield of ethanolic extract of *Alternanthera sessilis* Linn.

The powdered material of *Alternanthera sessilis* Linn. was extracted in Soxhlet apparatus by using ethanol and the solvent was evaporated by vacuum distillation process. The percentage yields of extract was obtained had been tabulated in the following Table No. 1.

Ethanolic extract of *Alternanthera sessilis* Linn. of different concentration and Albendazole as standard were used for the study. The results had tabulated in Table No. 2.

DISCUSSION

From the observations made, a dose dependent paralytic effect much earlier and the time of death was observed (Table no. 2). Although both the test extracts showed significant anthelmintic activity in a dose dependent manner but the ethyl acetate fraction appeared to be more effective. Evaluation of anthelmintic activity was compared with reference standard albendazole.

Preliminary phytochemical analysis of test extracts revealed the presence of tannins & flavonoids.^[8] Tannins have been reported to produce anthelmintic activities^[9, 10] as they can bind to free proteins in the gastrointestinal tract of host animal^[11] or glycoprotein on the cuticle of the parasite and thereby cause deaths.^[12] The potent wormicidal activity of ethyl acetate fraction against earthworms suggests that it is effective against parasitic infections of humans.

CONCLUSION

The ethanolic extract of *Alternanthera sessilis* Linn. exhibited significant anthelmintic activity against earthworms in dose dependent manner. The observed activity may be due to their phenolic content (flavonoids) which is worth for further investigations on isolation of the specific constituents.

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