Anti helminthic Activity of *Alternanthera pungens* Krunth

Ramesh P., Sai Kumar B., Tabu K., Naga Divya Sree M.,
Irfan Hyder and Uma Devi Parimi*

Department of Organic Chemistry, Gayatri Vidya Parishad College for Degree & PG
Courses (A), Visakhapatnam - 530045, Andhra Pradesh, India.

Abstract : The objective of this work was to evaluate the Anti helminthic activity of the crude extracts of *Alternanthera pungens* kunth against *Pheritima posthuma* (Indian Earth worm). Hexane, chloroform, ethyl acetate and methanol extracts of *Alternanthera pungens* kunth were tested for anti helmentic activity against Indian earth worm (*Pheritima posthuma*). Albendazole was taken as standard reference drug. Chloroform, hexane and ethyl acetate are found to be better Anti helminthic agents in comparison with albendazole at a concentration ranging from 20-100 mg/ml. This is the first report on the Anti helminthic activity of *Alternanthera pungens* kunth. Among all the tested extracts chloroform extract was found to be most potent Anti helminthic agent.

Keywords : *Alternanthera pungens* kunth, Anti helminthic activity, *Pheritima posthuma*, Shexane, chloroform, ethyl acetate and methanol.

Introduction:

Natural products contain metabolites derived from plants that play an essential role in drug discovery due to their vast structural diversity and wide variety of biological activities. Natural products have been used as traditional medicine from ancient times. Medicinal use of herbal medicine in the treatment and prevention of diseases including helmenthes has a long history compared to conventional medicine. The use of herbal medicine is still continuing worldwide and it has been estimated that one fourth of prescription medicine are derived from natural products.

With the advancement in science and technology, remarkable progress has been made in the field of medicines with the discoveries of many natural and synthetic drugs.¹ *Alternanthera pungens* kunth native of central and south America and Asia is a creeping, prostrate perennial plant of the amaranthacea family, locally called as mullu-ponnaganti (Fig-1). phenols, flavanoids, alkaloids, glycosids, steroids, saponins and tannins are reported from *A.pungens* kunth^{2,3}. It is an effective agent to cure abdominal pains in women, as an abortifacient and a galactagogue, used to promote child birth.^{4,5,6} This plant has applications for constipation with gripping, and as an enema for diarrhea Helminthes infections are found mainly in developing countries and are known to

be most acute disease for humans as well as for cattle. Hence, the utmost treatments must be done for helminthes^{7,8} Presently available synthetic drugs are associated with many side effects and natural products are known as indispensable sources for curing many diseases with very less side effects. Previously there are several reports on the anti helmentic activity of different plants.⁹⁻¹² So far there are no reports on the anti helmentic activity of *A.pungens kunth* . This study is focused to study the antihelmentic activity of hexane, chloroform, ethyl acetate and methanol extracts of *A.pungens kunth* .

Fig :1 *Alternanthera pungens kunth*

Materials And Methods:

All the reagents are AR graded and procured from Merck, TLC silica gel 60 F254 (M) is used in the experiments. The TLC plates are developed either in the U.V Chamber or in Iodine chamber.

Collection of the plant material: The Plant material was collected from GVP college for degree and PG courses (A), Rushikonda, Visakhapatnam district ,Andhra Pradesh, India. The plant was identified and authenticated in the department of Botany, Andhra University, and Andhra Pradesh, India.

Preparation of plant extracts: 250 grams of the plant material was shade dried at room temperature for about 3 days and then the plant material was finely powdered with the help of blender. The plant material was successively extracted with hexane, chloroform, ethyl acetate and methanol in a soxhlet apparatus for about 24 hours. The solvent was evaporated in rota evaporator and completely dried. The yields of each extract were noted (**Table 1**)

Table – 1: Yields of extracts in different solvents

Extracts	yields
Hexane	3.18 g
Chloroform	0.41 g
Ethyl acetate	0.88 g
Methanol	13.435 g

Thin Layer Chromatography:

Thin Layer Chromatography (TLC) was performed on different extracts and are tabulated (**Table 2**)

Table 2: Retention factor (Rf's) of different compounds obtained in hexane, chloroform, ethylacetate and methanol extracts.

Rf's	1 st spot	2 nd spot	3 rd spot	4 th spot	5 th spot
Hexane	0.19140.	5319	0.7021	0.8936	0.9574
Chloroform	0.235	0.6588	0.9294	0.964	
Ethyl acetate	0.3727	0.7272			
Methanol	0.119	0.190	0.285	0.880	

Evaluation of anti helmentic activity:

The antihelminthic activity was carried out as per the method of Ajaiyeba.et.al.¹³ Anti helminthic activity was performed on Indian earth worms *Pheretima posthuma* as its anatomical and physiological resemblance is similar with the intestinal round worm parasites of human beings. Earth worms were collected from the water logged areas of soil, the earth worms of size 10cm were washed with tap water for the removal of dirt. A stock solution of each extract 10mg/ml was prepared by dissolving in double distilled water. Working standard solutions of concentrations, 40, 60, 80, and 100mg/ml (hexane extract), 10, 20, 30mg/ml (chloroform extract), 20, 30, 40, 60mg/ml (ethyl acetate extract) . The formulations containing different concentrations of the extracts are taken in petri dishes and five earth worms of approximately same size were placed in petri dishes. Albendazole solution was taken as standard reference drug with four different concentrations (40, 60, 80,100mg/ml). Time taken for paralysis and time taken for death of worms were noted respectively and the results are tabulated (**Table-3**) and presented in **Figure-2**.

Table 3: Anti helminthic activity of *Alternanthera pungens kunth*

Plant extract	mg/ml	time taken for paralysis	time taken for death
Hexane	40	20 min 01 s	28 min 5 s
	60	16 min	22 min
	80	1 min 30 s	9 min 30 s
	100	1 min 14 s	9 min
Chloroform	10	11 min	12 min 20 s
	20	3 min 50 s	4 min 39 s
	30	52 s	2 min 30 s
Ethyl acetate	20	20 min 33 s	28 min 3 s
	30	16 min 50 s	23 min 07 s
	40	10 min	15 min
	60	06 min 52 s	10 min
Methanol	120	22 min 24 s	47 min 49 s
	140	13 min 45 s	40 min 12 s
	160	13 min 19 s	15 min 58 s
Albendazole	40	15 min 24 s	18 min 43 s
	60	14 min 30 s	16 min 31 s
	80	07 min 40 s	11 min 52 s
	100	05 min 20 s	09 min 11 s

Fig-2 : Anti helminthic activity of *A. Pungens Kunth*

Results and Discussion:

Hexane, chloroform, ethylacetate and methanol extracts of *A. pungens kunth* have been evaluated for their Anti helminthic activity. Chloroform extract, at a concentration of 10mg/ml, 20mg/ml and 30mg/ml, the time taken for paralysis was 11 min, 3 min 50 s and 52 s respectively and the corresponding times for death are 12 min 20 s, 07 min 20s and 02 min 30 s. In case of ethyl acetate extract, at concentrations of 20mg/ml, 30mg/ml, 40mg/ml and 60mg/ml, the paralysis time was 20 min 33 s, 16 min 50s, 10 min and 06 min 52 s respectively and the time taken for death was at 28 min 30 s, 23 min 07 s, 15 min and 10 min. For hexane extract, at a concentration of 40mg/ml, 60mg/ml, 80mg/ml and 100mg/ml the time taken for paralysis was at 20min 01 s, 16 min, 01 min 30s and 01 min 14 s and the death was observed at 28 min 50 s, 22 min, 09 min 30s and 9 min respectively. It is noticed that the Anti helminthic activity of methanol is at higher concentrations *i.e.*, at 120mg/ml, 140mg/ml and 160mg/ml. At concentrations lower than these no effect was observed. The activity of chloroform, ethyl acetate and hexane extracts may be due to flavanoids, tannins, saponins, glycosides and sterol-triterpenes.

Conclusion:

This is the first report for the Anti helminthic activity of *Alternanthera pungens kunth*. It was noticed that the Anti helminthic activity is concentration dependent. Among all the tested extracts, chloroform extract showed the best Anti helminthic activity, at a concentration of 30mg/ml. The time for paralysis is only 52 s and

time taken for death is 2 min 30 s, where as for the standard drug albendazole the time taken for paralysis is more than 1 hr. Hexane extract showed maximum potency at a concentration of 80mg/ml. Ethyl acetate extract at a concentration of 60mg/ml showed the best potency against earthworms. Methanol extracts was found to be inferior among all the tested extracts as well as the standard drug albendazole. In conclusion, all the extracts except methanol are superior and more potent than the standard drug albendazole and *A.pungens kunth* may be used as an alternative for the present Anti helminthic drugs. This is the first report for the Anti helminthic activity of *Alternanthera pungens kunth*.

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