Anti helminthic Activity of *Alternanthera pungens* Kunth

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**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 Alternantera pungens kunth were tested for anti helminthic 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 helminthes 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. *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*. It is an effective agent to cure abdominal pains in women, as an abortifacient and a galactogogue, used to promote child birth. 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

Uma Devi Parimi *et al* /International Journal of ChemTech Research, 2019,12(4): 214-218.

DOI= [http://dx.doi.org/10.20902/IJCTR.2019.120426](http://dx.doi.org/10.20902/IJCTR.2019.120426)
be most acute disease for humans as well as for cattle. Hence, the utmost treatments must be done for helminthes. 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 helminthic activity of different plants. So far there are no reports on the anti helminthic activity of A.pungens kunth. This study is focused to study the antihelminthic 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 soxhelet 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         | 1st spot | 2nd spot | 3rd spot | 4th spot | 5th 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 helmintic activity:

The antihelmintic activity was carried out as per the method of Ajaiyeba.et.al. Anti helmintic 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 helmintic 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          |
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 20 s 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 50 s, 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 20 min 01 s, 16 min, 01 min 30 s and 01 min 14 s and the death was observed at 28 min 50 s, 22 min, 09 min 30 s 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.

Acknowledgement:

All the authors contributed equally .The authors are thankful to Gayatri Vidya Parishad College for
Degree and PG courses (A) for providing all the facilities to carry out this research work .There is no conflict of
interest.

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