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In vitro evaluation of anthelmintic activity of Commiphora caudata leaves extracts on Phertima posthuma

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Abstract

Commiphora caudata is an ethnomedicinal plant which has been used traditionally in the treatment of inflammation and analgesic condition. It also used in the treatment of fever, straungary, vitiated condition of vata and pitta in sidda system of medicine. It also has antioxidant, hepatoprotective, antii nflammatory, antiulcer, and antihyperlipidemic activity were reported from the plant. The aim of the present study was to evaluate the anthelmintic activity of aqueous and ethanolic extracts of 50mg/ml and 100mg/ml using earth worms. The time of paralysis and time of death was evaluated and it is compared with reference standard albendazole 10mg/m and normal saline as a control group. Aqueous and ethanolic extract exhibited the dose dependent activities as evidenced by decreased paralysing time and death time

Keywords: Commiphora caudata, Anthelmintic activity, Pherithima posthuma

Introduction

Helminths is a Greek word meaning "worm". It was supposed to be used only for intestinal worms but now includes tissue parasite free living species and mainly other worms [1]. Helminths have plagued humans since before the era of our earliest recorded history. There are two major phyla of helminths. The nematodes (roundworms) include the major intestinal worms, also known as soil transmitted helminths. Whereas platyhelminths (flatworms) include flukes also known as trematodes and tapeworms [2]. Anthelmintics or anti helminthics are drugs that expel helminth parasitic worms (helminths) from the body, either by stunning or killing them. They may also be called vermifuges (stunning) or vermicides (killing). Anthelmintics are drugs that either kill or expel infesting helminths and the gastrointestinal tract is the abode of many helminths, although some also live in tissues, or their larvae migrate into tissues. However they have shown the development of resistance. The gastrointestinal helminthes becomes resistant to currently available some broad spectrum anthelmintics (benzimidazoles, levamisole, vermectins) and also some narrow spectrum wormers such as the salicylanilides (closantel). Therefore there is a foremost problem in treatment of helminthes diseases [3]. Food supplements like Papaya (Carica papaya), Cinnamon (Cinnamonum Camphora, Cinnamonum Zylenicum), Turmeric (Curcuma longa), Asafoetida (Ferula foeteda), Long pepper (Piper longum) and fresh juices of Pine apple have anthelmintic property. Hence, there is an increasing demand towards natural anthelmintics and to prevent resistance [4]. The anthelmintic assay was carried as per the method of Ajaiyeoba et.al with minor modifications. The assay was performed on adult earthworms. Phertima posthuma due to its anatomical and physiological resemblance with the intestinal round worm parasite of human beings [5].

Material and Methods Collection of Plant material

Commiphora caudata leaves were collected from the Mangadahally village, Ramanagara district, Karnataka India in the month of August. The samples were authenticated by National Institute of Ayurveda and Dietetics institute, Bangalore 560011.

Collection of earth worms: All earth worms were of approximately equal size (15 cm). they were collected from the Karthik vermicompost and earthworms, Hebbala and washed with normal saline.

Preparation of plant extract: Aqueous and ethanolic extracts of *Commiphora caudata* leaves was procured from Green chem and herbal formulations Bangalore. The extraction was done by soxhlet extraction process, percentage yield obtained Aqueous extract - 12.4% w/w and Ethanolic extract- 13.8% w/w

Anthelmintic Activity: The anthelmintic activity was carried as per the method of Ajaiyeoba *et al.*, with minor modification. ^[5] The assay was performed on *Pheretima posthuma* due to its anatomical and physiological resemblance with intestinal roundworm parasites of human being were used in the present study. All earth worm were of approximately equal size (15 cm).

Dose selection: Aqueous and ethanolic doses of *Commiphora caudata* (50 and 100 mg/ml) and albendazole dose (10 mg/ml) were selected based on earlier studies.

Groupings

Earth worms are divided into six groups of six earth worms each

- Group 01: Normal saline.
- **Group 02:** 10 mg/ml albendazole(std) ^[6].
- **Group 03:** 50 mg/ml of aqueous leaves extract of *Commiphora caudata*.
- **Group 04:** 100 mg/ml of aqueous leaves extract of *Commiphora caudata*.
- **Group 05:** 50 mg/ml of ethanolic leaves extract of *Commiphora caudata*.
- **Group 06:** 100 mg/ml of ethanolic leaves extract of *Commiphora caudata*.

The earth worms of respective groups were placed into 10 ml of the respective extract and the time taken for paralysis

and death was recorded. Maximum cut off time to observe or death would be 120 mins. Paralysis is said to occur when the worms do not revive even in normal saline. Death is concluded when the worms lose their motility followed with fading away of their body colour.

Statistical Analysis" The results were expressed in terms of mean \pm standard error of mean (SEM). The results were subjected to statistical analysis by using one way ANOVA followed by Dunnet t test.

p<0.05 was considered as statistically significant.

Results and discussion

The preliminary phytochemical analysis of different plant extracts showed the presence of chemical constituents in the extract. The results revealed the presence of flavonoids, tannins, steroids, saponin, terpenes and phenolic compounds in table no 1.

The results of anthelmintic activity of the aqueous and ethanolic extracts of *Commiphora caudata* leaves summarised in table 2.

It is evident that aqueous and ethanolic leaves extracts of *Commiphora caudata* exhibited anthelmintic activity in a dose dependent manner by reducing the time of paralysis and time of death. In aqueous extract of 50mg/ml and 100mg/ml concentration shows dose dependent activity on paralysis time and death time. The aqueous extract at 50mg/ml caused paralysis at 45.33min and death time at 50.1min. in 100mg/ml concentration time taken for death and paralysis 33.17min and 37.67 min.

While ethanolic extract at the concentration of 50 and 100 mg/ml revealed the paralysis time and death time in a dose dependent manner. 19.83 and 26.83 min in 50 mg/ml and in the concentration of 100 mg/ml paralysis and death time was 17.5 and 22.83 min.

Flavonoids are polyphenolic compounds that disrupt the parasites energy production by uncoupling the oxidative phosphorylation. This disturbance, along with their interaction with cell surface glycoproteins, ultimately leads to the death of the parasites [8].

Table 1: Phytochemical investigation of leaves extracts of *Commiphora caudata*.

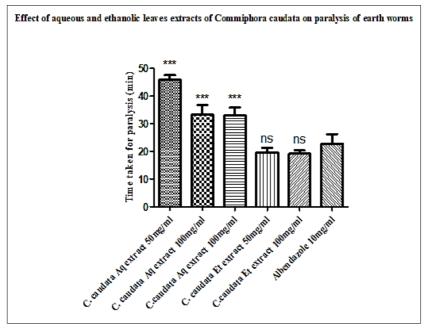
| Active constituents | Aqueous extract | Ethanolic extract |
|---------------------|-----------------|-------------------|
| Alkaloids | - | - |
| Saponins | + | + |
| Flavonoids | + | + |
| Steroids | + | + |
| Carbohydrates | - | - |
| Tannins | + | + |
| Proteins | + | + |
| Amino acids | + | + |
| Glycosides | 1 | - |
| Phenolic compounds | + | + |
| Anthraquinones | - | = |

Table 2: Effect of aqueous and ethanolic leaves extracts of Commiphora caudata on paralysis and death time of earth worms

| Treatment drug | Concentration used | Time taken for paralysis (min) | Time taken for death (min) |
|-----------------------|--------------------|--------------------------------|----------------------------|
| Normal saline | - | - | - |
| C. caudata Aq extract | 50mg/ml | 45.33 | 50.17 |
| C. caudata Aq extract | 100mg/ml | 33.17 | 37.67 |
| C.caudata Aq extract | 100mg/ml | 33.17 | 37.67 |
| C. caudata Et extract | 50mg/ml | 19.83 | 26.83 |
| C.caudata Et extract | 100mg/ml | 17.5 | 22.83 |
| Albendazole | 10mg/ml | 22.33 | 25.33 |

All values are expressed in mean \pm SEM, one way ANOVA followed by Dunnett t test.

p<0.05 * represents significant, p<0.01 ** represents medium significant and p<0.001***represents highly significant when compared to standard group.



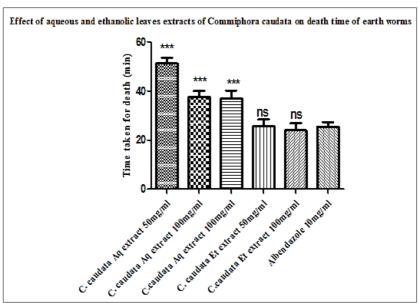


Fig 1: Effect of aqueous and ethanolic leaves extracts of Commiphora caudata on Pheretima posthuma

Conclusion: From the results, it was concluded that both aqueous and ethanolic extracts of *Commiphora caudata* shows significant anthelmintic activity. But the ethanolic extract shown potent anthemintic activity when compared to aqueous extract. *In vitro* anthelmintic activity was confirmed by the presence of active constituent's flavonoids and tannins exhibit to interfere with energy generation in helminth parasites by uncoupling oxidative phosphorylation. Further studies have to be done to isolate the active principles responsible for activity.

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