**Invitro** anthelmintic activity of different extracts of *Coldenia procumbens*

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**ABSTRACT**

In recent times there is an exponential growth in herbal medicine due to high safety and low cost. The drugs such as anthelmintics are very cost effective and this made researchers in search of alternatives. In the present study, invitro experiments were conducted to evaluate the possible anthelminitic effects of different extracts (petroleum ether, chloroform, ethanol, aqueous) of the leaves of *Coldenia Procumbens* on Pheretima Posthuma worms. Various concentrations (25, 50, 100mg/ml) of all extracts were tested and results were expressed in terms of time for paralysis and time for death of worms. Albendazole used as a reference standard and saline as a control group.

**Key words.** Herbal medicine, anthelmintics, *coldenia procumbens*, Albendazole, *Pheretima Posthuma*.

**INTRODUCTION**

The word Helminth has been derived from the Greek which means worm [1]. Helminth infections are among the most common infections in man, affecting a large proportion of the world’s population[2]. The parasites can be acquired by contact with a) infected water b) infected meal c) infected animal. Anthelmintics are drugs that may act locally to expel worms from the GIT or systemically to eradicate parasites [3]. Gastro intestinal Helminthes are resistant to currently available drugs[4]. So the discovery and development of new chemical substances for helminth control is greatly needed and has promoted studies of traditionally used anthelmintic plants, which are generally considered to be very important sources of bioactive substances[5].

*Coldenia Procumbens* Linn (Boraginaceae) is an annual herb, common weed in India[6,7]. *Coldenia Procumbens* is only species of its genus has a place both in the Hortus Bengalensis and Moon’s Catalogue of ceylon plants[8]. In the traditional system of medicine, the plant was used as anti inflammatory[9], anti microbial[10], analgesic[11], anti diabetic[12], CNS depressant[13]. Fresh leaves of *Coldenia Procumbens* ground and applied to Rheumatic Swellings, equal parts of dried powder mixed with seeds of fenugreek causes Suppurations of boils[14]. The active constituent of plant is wedelolactone which is a derivative of coumestans[15] This plant is widely used in traditional medicines in India, Africa, Malaysia. Acetone, water, methanolic extract of dried aerial parts shows weak angiotensin-converting enzyme inhibition in vitro[16,17]. Present study made to investigate the anthelmintic potency of *Coldenia Procumbens*.
MATERIALS AND METHODS

Plant material
Fresh aerial parts of *Caldenia Procumbens* Linn (Boraginaceae) was collected from Thanjavur (India) and it was identified and authenticated by Dr. G. V. S. Murthy, Scientist ‘F’ & Head of Botanical Survey Of India, Coimbatore. Specimen number BSI/SRC/5/23/2011-12/Tech-1058. The aerial parts of *Caldenia Procumbens* was dried in the shade and it was milled into coarse powder by a mechanical grinder and it was stored in closed vessel for further use.

Extraction of plant drug,
Aqueous extraction (Decotion method)
150 g of coarse powder of *Caldenia Procumbens* leaves was boiled with 600 ml of distilled water. Then it was cooled to room temperature and filtrate was filtered. The percentage of yield was found to be 8.1%.

Solvent Extraction,
100 gm of powder was subjected to successive soxhlet extraction by various solvent such as petroleum ether, chloroform and ethanol. The percentage of yield were found to be as 2.6 %, 3.6%, 6.9% respectively.

Collection of worms
Indian adult Earthworms (*Pheretima Posthuma*) were collected from the moist soil of PRIST University. Selected earthworms are 5-7 cm in length and 0.1-0.3 cm in width. The earthworms were washed with normal saline to remove all the fecal matter.

Preparation of test samples
Samples for in-vitro study were prepared by dissolving and suspending 2.5 g of each extract in 25 ml of distilled water to obtain a stock solution of 100 mg/ml. From this stock solution, different working dilutions were prepared to get concentration range of 25, 50 and 100 mg/ml final volume made with saline solution.

Anthelmintic Assay
Anthelmintic activity was carried as per the method reported by Rajesh R et al., with minor modifications. All the extracts and the standard drug solution were freshly prepared before starting the experiments. *Pheretima posthuma* was placed in petridish containing three different concentrations (25, 50, 100 mg/ml) of *Caldenia Procumbens* (petroleum ether, chloroform, ethanol, and water extract) solutions. Albendazole is used as a standard reference. Each petridish was placed with 6 worms and observed for paralysis (or) death. Observations were made for the time taken to paralyze and/or death of individual worms. Paralysis was said to occur when the worms do not move even in normal saline. Death was concluded when the worms lost their motility followed with fading away of their body colour.

RESULTS
From the observations in comparision with standard drug all the extracts of *Caldenia Procumbens* has showed the potent anthelmintic activity. All extracts had been showed the dose dependent activity.

The Ethanolic extract at concentration 25 and 50 mg/ml shows paralysis at 15.9±0.19 and 9.7±0.16 min and death at 21±0.33±0.37 and 35.23±0.18 min respectively. But mortality with 100 mg/ml concentration is at 21±0.33 min (Fig 1).

The aqueous extract at concentration 25, 50 and 100 mg/ml shows paralysis at 19.43±0.25, 11.8±0.17, and 7.66±0.18 and death at 48.6+0.48, 38.3±0.26 and 23.97±0.23 respectively (fig 2).

The chloroform extract at concentration 25, 50 and 100 mg/ml shows paralysis at 25.33±0.22, 17.97±0.21, and 11.9±0.18 and death at 51.47+0.18, 39.55±0.16 and 31.8±0.14 respectively (fig 3).

The petroleum ether extract at concentration 25, 50 and 100 mg/ml shows paralysis at 26.87±0.21, 16.83±0.13, and 10.5±0.12 and death at 45.63+0.19, 39.72±0.17 and 29.43±0.18 respectively (fig 4).
The different extracts of *Caldenia Procumbens* has been showed potent anthelmintic activity in comparision with standard drug Albendazole. Albendazole shows paralysis at 8.9 min and death after 28.87 min. The anthelmintic activity of the different extracts were indicated as ethanol>aqueous>petroleum ether>chloroform. Ethanolic and aqueous extracts having the equipotent activity with standard drug Albendazole. All values were expressed as mean SEM (n=6).

**Figure 1**, Effect of *Caldenia procumbens* ethanolic extract on *pharetima posthuma*

*Caldenia procumbens* Ethanol extract effect on worms

- **Paralysis and death time (min)**
- **groups**
- albendazole 20mg/ml
- ET 25mg/ml
- ET 50mg/ml
- ET 100mg/ml

**Figure 2**, Effect of *Caldenia procumbens* aqueous extract on *pharetima posthuma*

*Caldeiana procumbens* aqueous extract effect on worms

- **Paralysis and death time (min)**
- **groups**
- albendazole 20mg/ml
- AQ 25mg/ml
- AQ 50mg/ml
- AQ 100mg/ml

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Figure 3, Effect of *Coldenia procumbens* chloroform extract on *Pheretima posthuma*

![Graph showing the effect of *Coldenia procumbens* chloroform extract on paralysis and death time of *Pheretima posthuma*.

Figure 4, Effect of *Coldenia procumbens* petroleum ether extract on *Pheretima posthuma*

![Graph showing the effect of *Coldenia procumbens* petroleum ether extract on paralysis and death time of *Pheretima posthuma*.
DISCUSSION

The leaves extract of *Coldenia Procumbens* has been showed anthemic activity. ethanolic and aqueous extract at dose 100 mg/ml shows potent anthelmintic activity compared with Albendazole. *Coldenia Procumbens* is having alkaloids, flavonoids, carbohydrates, phenolic compounds [19]. However Further invivo studies needed to find out the potential pharmacological profile of *Coldenia Procumbens* as an anthelmintic drug. The plant may be further explored for isolation of the active constituent responsible for anthelmintic activity.

REFERENCES