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IN-VITRO ANTHELMINTIC ACTIVITY OF *HOLARRHENA ANTIDYSENTERICA* BARK

Patil Reshma^{1*},
Devkar Swati¹, Pawar Prashant¹, Pattewar Aditya¹

¹Rajarambapu College of Pharmacy, Kasegaon, Tal. Walwa, Dist Sangli 415404, MS, India.

ABSTRACT

The present study aimed at the in-vitro comparative study of anthelmintic activity of aqueous and ethanolic extracts of leaves of *Holarrhena antidysenterica* using *Pheretima posthuma* at three different concentrations (100, 50, 25 mg/ml) respectively.

The study involved the determination of time of paralysis (P) and time of death (D) of the worms. At the concentration of 100 mg/ml both the ethanolic and the aqueous extracts showed very significant activities as compared to the standard drug piperazine citrate (10 mg/ml). In case of aqueous extract the time of paralysis and death time was observed as 4.30 ± 0.13 and 10.23 ± 0.09 and in case of ethanolic extracts 14.30 ± 0.13 and 22.59 ± 0.61 respectively. In conclusion, the use of leaves of *Holarrhena antidysenterica* as an anthelmintic have been confirmed and further studies are suggested to isolate the active principles responsible for the activity.

Keywords *Holarrhena antidysenterica*, Anthelmintic, *Pheretima posthuma*, Piperazine citrate.

Correspondence to Author



Patil Reshma

Rajarambapu College of Pharmacy,
Kasegaon, Tal. Walwa, Dist Sangli
415404, MS, India.

Email: patil.reshma1990@gmail.com

INTRODUCTION

Parasitic diseases cause ruthless morbidity affecting principally population in endemic areas. The gastro-intestinal helminthes becomes resistant to currently available anthelmintic drugs; hence there is an increasing demand towards natural anthelmintics. Among other alternative methods, there is considerable and expanding interest in traditional herbal dewormers. *Holarrhena Antidysenterica*, commonly called Kurchi bark is deciduous shrub belonging to Apocynaceae family.

The bark has astringent, antidiarrhetic, anthelmintic, stomachic, febrifugal and tonic properties. It is used in the treatment of amoebic dysentery and diarrhoea.¹

OBJECTIVE

The objective of the study is to evaluate the anthelmintic activity of *Holarrhena Antidysenterica* bark.

MATERIALS AND METHODS

Plant material

The bark of *Holarrhena antidysenterica* were collected from the local market and authenticated by authority of Botany Department, Y C M Science college, Karad.

Preparation of Extract

Bark of *Holarrhena antidysenterica* were shade dried and powdered mechanically. The powdered plant material (500g) was first defatted with pet ether and then extracted with ethanol by soxhlet extraction process. Solvents were removed at temperature below 50°C in an oven.

The aqueous extract was obtained by maceration for 24 hrs. The aqueous extract obtained was filtered and concentrated on hot plate.

Animals

Indian adult earthworms (*Pheretima posthuma*) were collected from water logged areas of Kasegaon village and the worm types were identified at the Department of Zoology, Y C M Science College, Karad

Phytochemical screening: Preliminary phytochemical screening of extract was performed for presence of carbohydrates, diterpenoids, flavonoids, tannins, glycosides.

Evaluation of anthelmintic activity²

The anthelmintic assay was carried as per method of Ajaiyeoba et al with minor modifications the anthelmintic activity was evaluated on adult Indian earthworm *Pheretima posthuma* worm due to its anatomical and physiological resemblance with the intestinal round worms' parasites of human beings: Fifty milliliter of formulation containing three different concentrations, each of crude alcoholic and aqueous extract (25, 50, 100 mg/ml in distilled water) was and six worms (same type) were placed in it. Observation was made for the time taken to cause paralysis and death of the individual worms. mean time for the paralysis (P) in min was noted when no movement of any sort could be observed, except when the worm was shaken vigorously; time of death (D) in min was recorded after ascertaining the worms neither moved when shaken vigorously nor when dipped in warm

water (50° C). Piperazine citrate (10 mg/ml) was included as reference compound.

Statistical analysis

Data were analyzed using one way factorial ANOVA tests followed by Dunnett's *t*-tests on each group. P values under 0.01 were considered highly significant. (shown as *).

RESULTS AND DISCUSSION

Results shown in the Table No1. signifies that aqueous & alcoholic extract of Bark of *Holarrhena antidysenterica* exhibited anthelmintic activity in dose dependent manner giving shortest time of paralysis and deaths compared to Piperazine citrate, especially with 100 mg/ml concentration.. Reported anthelmintic effect of tannins is that they can bind to free proteins in the gastrointestinal tract of host animal or glycoprotein on the cuticle of the parasite and may cause death.

Synthetic phenolic anthelmintics interfere with the energy generation in the helminth parasites by uncoupling the oxidative phosphorylation. Another possible mechanism of action is that they bind to free proteins in the gastrointestinal tract of the host animal or to glycoprotein on the cuticle of the parasite and causes death.

The possible mechanism of action of tannins may be:

- Interfere with energy generation by uncoupling oxidative phosphorylation
- They may interfere with glycoprotein of cell surface
- They can bind to free proteins in the gastrointestinal tract of host animal or glycoprotein on the cuticle of the parasite and cause death. [10].

Alkaloids may act on central nervous system and caused paralysis of the earthworm [11]. The effect would be due to presence of the steroidal alkaloid oligoglycosides which may suppress the transfer of sucrose from the stomach to the small intestine together with its antioxidant effect which is capable of reducing the nitrate generation which could interfere in local homeostasis which is essential for the development of helminthes [12].

Groups	Concentration mg/ml	<i>Pheretima Posthuma</i> (Earthworm)	
		Time taken for paralysis (P) in min. (Mean & SEM)	Time taken for death (D) in min. (Mean & SEM)
Control	-----	-----	-----
Alcoholic extract	25	44.35 ± 0.16	72.15 ± 0.12
	50	29.59 ± 0.23	54.15 ± 0.17*
	100	14.30 ± 0.13*	22.05 ± 0.61*
Aqueous extract	25	34.55 ± 0.6	52.15 ± 0.05
	50	9.59 ± 0.23*	32.05 ± 0.07*
	100	4.30 ± 0.13*	10.23 ± 0.09*
Standard	10	3.9 ± 0.06	41.15 ± 0.05

CONCLUSION

Phytochemical screening of extracts revealed presence of alkaloids, saponins, flavonoids, triterpenes, tannins and steroids. It is possible that tannins contained in the extracts of Bark of *Holarrhena antidysenterica* produced the effects. Further studies are in process to identify the possible phytoconstituents responsible for anthelmintic activity.³

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