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Comparative In vitro Anthelmintic Activity of Centratherum Anthelminticum (L.) and Mebendazole

Santosh J Gutte*

Department of Pharmacology, Dattakala College of Pharmacy, India

Abstract

Helminthiasis is the most common cause of the intestinal infestation. Anthelmintic from natural sources may play a key role in the treatment of parasitic infestation. Aqueous extracts from the seed of Centratherum Anthelmintic (L). were investigated for their anthelmintic activity against Earthworm (Eisneia Fetida). Mebendazole is used as the Standard drug. Extract was studied in the bioassay a 100 mg/ml, which involved determination of time of paralysis and time of death of the worms.

Keywords: Centratherum anthelmintic (L); Mebendazole; In vitro anthelmintic activity

Introduction

The plant Centratherum anthelmintic (Wild) Kuntze called as 'Kalijiri' in Hindi is reported to be a medicinally important plant; this species has a wide variety of application in traditional medicine, especially for treatment of fever, cough, and diarrhoea.

The various part of Centratherum anthelmintic are documented to possess medicinal properties such as, Centratherum anthelmintic Kuntz (Hindi- Kalijiri) previously known as Vernonia anthelmintica belongs to family composite (1). It has a good anthelmintic property and used for the treatment of various skin infections. It is also reported to be used in asthma, kidney troubles, cough and also used to remove blood from liver (2). But no work has been done until now to establish its antidiabetic potential. The major classes of chemical constituent present in this plant are glycosides(3), carbohydrates(4), phenolic compounds and tannins(5), flavonoids(6), proteins, saponins(7), sterols(8,9), lipids(10) and fats(11).

Chemical constituent

The major chemical constituent present in C. Anthelminticum is Vernodalin, Butein, Daucosterol, Vernolic acid, Vernodalol, Vernovan, Stigmastadienol, Lupeol and Beta-Sitosterol. Other chemical constituents are Vernolic acid, Linoleic acid, Oleic and Palmitic acid, Stearic acid, Stigmasterol, Vernosterol, Avenasterol, D-lactose, L-sorbose, D-arabinose, Protein, Lipids and Fats [1-3]. The anthelmintic activity of C. Anthelminticum is attributed to the presence of Anthraquinone [4].

Materials and Methods

Plant materials

The Centratherum Anthelminticum Plant Seed are collected from botanical garden of gunmala Herbal botany, Indore, India. The Seeds were air-dried and grinded to fine powder.

Preparation of extracts

Centratherum Anthelminticum (L.) Shows the More Anthelmintic Activity in Aqueous Compare to Methanolic and Ethanolic Extract. The anthelmintic activity of C. Anthelminticum is attributed to the presence of Anthraquinone [4].

Animals

India adult earthworms, which were collected from moist soil of swami-Chincholi and washed with normal saline to remove all faecal matter, were used for anthelmintic study. The earthworms (Eisenia Fetida) of 5–7 cm in length and 0.1–0.2 cm in width, 500- 600 mg.

Weight was used for all the experimental protocol due to its anatomical and physiological resemblance with the intestinal roundworm parasites of human beings.

The anthelmintic activities have been reported by using adult earthworm Eisenia foetida by a number of references due to its anatomical and physiological resemblance with the intestinal parasite of human beings [5-8]. Its easy availability and maintenance makes it one of the most commonly used models [1].

Drugs and chemicals

The following drugs and chemicals were used.

Standard Drug: Mebendazole, distilled water.

Bioassay for Centratherum Anthelminticum (Kalijiri)

Preparation of Standard Plot: To prepare standard plot, C. Anthelminticum (100 mg) was accurately weighted and transferred to 100 mL volumetric flasks and volume was adjusted with distilled water to get the concentration of 1 mg/mL. Further dilutions were made from stock solution (1 mg/mL). were transferred from stock solution into Volumetric flask 10 mL volumetric flask and volume was adjusted with distilled water to get the concentration of 5, mg/mL. Different weights i.e. 50, mg were accurately weighed and transferred to 10 mL volumetric flasks and volume was adjusted with distilled water to get the concentration of 5 mg/mL. Sample (10 mL) from each dilution was transferred to petri plate containing one earthworm each. The study was conducted in triplicate and ADT was noted (Table 1).

***Corresponding author:** Santosh J Gutte, Department of Pharmacology, Dattakala College of Pharmacy, India, Tel: 07499032342; E-mail: santoshgutte1052@gmail.com

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Table 1: Bioassay of Centratherum Anthelminticum (Kalijiri) Using Earthworm (*Eisneia fetida)* APT and ADT was noted.

Concentration	APT (min)	ADT (min)
5 mg/ml	8-9	20-21

Anthelmintic Activity

Anthelmintic activity of C. Anthelminticum was evaluated using Eisenia foetida. Different concentrations of spray dried aqueous decoction of C.

Anthelminticum were tested in the bioassay. APT and ADT were noted. Mebendazole is used as standard drug for investigation of biological activity [9]. Spray dried powder of C. Anthelminticum was weighed is quantities of 0, 50,100,150 mg. Weighed amounts were transferred to 10 mL volumetric flask and volume was adjusted with distilled water to get the concentration of 0, 5, 10, 15 mg/ mL.

Observations were made for the time taken until the paralysis and death of an individual worm. The paralysis was said to occur when the worms were not able to move even in normal and. Death was concluded when the worms lost their motility followed with fading away of their body colors 15 .

Experimental work

Comparative Study between C. Anthelminticum with Standard drug Mebendazole: Anthelmintic activity of C. Anthelminticum was evaluated using Eisenia foetida. Different concentrations of spray dried aqueous decoction of C. Anthelminticum were tested in the bioassay. APT and ADT were noted. Mebendazole is used as standard drug for investigation of biological activity [10-13]. Spray dried powder of C. Anthelminticum was weighed is quantities of 0, 50,100,150 mg. Weighed amounts were transferred to 10 mL volumetric flask and volume was adjusted with distilled water to get the concentration of 0, 5, 10, 15 mg/ mL. The same concentration of standard drugs i.e. Mebendazole was also prepared [14-17]. Distilled water was used as control. One earthworm was added to each petri plate containing different concentration of test sample. Each study was conducted in triplicate [18-21]. ADT and APT were noted and mention in table. The Comparative Study between Centratherum Anthelminticum with Mebendazole Using the Earthworm *(Einstein foetida)* the result has been shows as the following: Where, APT- Actual Paralysis Time & ADT- Actual Death Time (Table 2).

Table 2: Comparative Study between C. Anthelminticum with Mebendazole Using Earthworm *(Eisneia foetida)* at different Conc.

Sr No.	Drug	Conc. (mg/ml)	APT (min)	ADT (min)
1	C. Anthelminticum	5	15-16	27-28
2	C. Anthelminticum	10	11-12	23-24
3	C. Anthelminticum	15	8-9	19-20
4	Mebendazole	5	20-21	50-51
5	Mebendazole	10	18-19	47-48
6	Mebendazole	15	14-15	38-39

Results and Conclusion

Based on the reports of limiting side effects and development of resistance to conventional anthelmintic therapy herbal formulation was selected. An herbal formulation is in clinical use for its anthelmintic activity for last few decades. However, no systematic study on its therapeutic/pharmacological effect is reported. The current research work was under taken to evaluate the anthelmintic property of this Centratherum Anthelminticum. Another aim of the study was to attribute the pharmacological effects to individual constituent as the formulation is of polyhedral nature. Suitable modifications in the existing formula based on the inferences of the study may render it more effective. On comparison of the anthelmintic activity of the decoction of all constituents (prepared in our lab), decoction Centratherum Anthelminticum and Standard drug Mebendazole.

- Decoction of Centratherum Anthelminticum of the formulation is slightly more effective than the marketed formulation (Mebendazole) indicating some role of excipients like sweeteners, flavours, preservatives etc.

- The anthelmintic activity of the decoction containing all constituent as well as that containing Centratherum Anthelminticum (L.) most effective constituents is less than that of the decoction Mebendazole. Based on above results, we prepared the dried powder of the decoction of C. Anthelminticum. Another interesting observation was made. Spray dried powder of decoction showed an enhanced anthelmintic activity indicating the role of amorphization and particle size reduction, particularly in light of the fact that the obtained decoctions were slightly turbid in nature.

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