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Short Communication Comparative study of Withania somnifera and Ocimum sanctum for Anthelmintic Activity

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Abstract

The hydroalcoholic extracts of *Withania sominifera* (Ws) and *Ocimum sanctum* (Os) were assayed against adult earthworms (*Pheretima posthuma*) for the evaluation of anthelmintic activity. Various concentrations of both extracts were tested and results were expressed in terms of time for paralysis and time for death of worms. Piperazine citrate (10 mg/ml) was used as a reference standard. The result showed that in both of the extracts (i.e. Os and Ws) dose of 40 mg/ml possesses more wormicidal activity. The time of paralysis was 2.5 ± 0.6 and 2.8 ± 0.8 whereas the time of death was 6.5 ± 0.7 and 7.1 ± 0.9 in the case of Os and Ws respectively. The comparative study showed that out of the two, Os proves to be a better anthelmintic remedy.

Keywords: Withania somnifera, Ocimum sanctum, anthelmintic activity.

Introduction

The development of anthelmintic drug-resistance in helminthes against synthetic drugs have been reported in number of countries¹⁻⁴ which gives a clear indication that control programs based exclusively on their use are not sustainable. Even though the majority of infections due to worms are limited to tropical regions, these can develop in individuals who had visited such infected areas and then, can transmit the disease in temperate regions⁵ Some anthelmintic drugs such as praziquantel and albendazole, are contraindicated for certain groups of patients like pregnant and lactating woman⁶. This has led to the increase in interest of ethnomedical practices across the world for the use of medicinal plants in treatment of helmintic diseases⁷. Plants are known to provide a rich source of herbal anthelmintics, antibacterials and insecticides remedies^{8,9}. A number of medicinal plants have been used in the treatment of parasitic infections in man as well as in animals¹⁰⁻¹².

Ocimum sanctum (Os) is a widely grown and sacred plant of India, belonging to the Labiateae family. When applied locally, Os helps in eradicating ringworms and other skin diseases. *Withania somnifera* (Ws) also known as Indian ginseng is a plant belonging to Solanaceae family. Ws has many applications such as in the treatment of inflammation, fevers, and to protect against infection or illness¹³. In search of compounds with anthelmintic activity, earthworms have been used widely for the initial evaluation of anthelmintic compounds *in vitro* because they resemble intestinal worms in their interaction with anthelmintics and their easy availability. It has been demonstrated that all anthelmintics are toxic to earthworms also and thus support their use in the investigation of anthelmintic activity¹⁴. The present research is thus motivated by the need to find new substances of natural origin which possess anthelmintic activity with a low degree of toxicity for humans.

Materials and Methods

Plant Material: The whole plant of Os and roots of Ws were collected, shade-dried, grinded and stored in room temperature in a closed container for further use.

Preparation of Extract: Both of the plant materials (350g) were repeatedly extracted in a 2000 ml round bottomed flask with 1500 ml solvent of 70% hydro-alcoholic solution. The reflux time for each solvent was 40 cycles. The extracts were cooled at room temperature, filtered, and evaporated to dryness.

Anthelmintic Bioassays: The earthworm *Pheretima posthuma* (Annelida, Megascolecidae) was used for evaluating the anthelmintic activity of crude extract using the peprazine citrate as reference substance for comparison.

Activity against Earthworms: Anthelmintic activity was assessed using earthworms *Pheritima posthuma*, as it has anatomical and physiological resemblance with the intestinal roundworm parasites of humans^{15,16} by the reported methods with slight modification^{17,18}. Four groups of approximately equal sized Indian earthworms, consisting of five earthworms each, were released into 20 ml of desired formulation. The groups were prepared as reference i.e. peprazine citrate (20mg/ml) and that of extracts of Ws and Os (20 and 40 mg/ml for each). Observations were made for the time taken to paralyze or death of individual worms. Paralysis occurred, when the worms do not retrieve even in normal saline. Death was

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concluded when the worms lost their motility followed with fading away of their body colors¹⁹. Results are shown in table-1.

 Table -1

 Comparative study of Ws and Os for Anthelmintic Activity

Sample	Conc. (mg/ml)	Time taken for paralysis (min)	Time taken for death (min)
Os	20	6.0±0.5	13.5±1.2
	40	2.5±0.6	6.5±0.7
Ws	20	6.5±0.5	13.9±1.2
	40	2.8±0.8	7.1±0.9
Peprazine citrate	20	7.0±0.6	10.9±1.3

All values represent Mean +SD; n= 6 in each group

Results and Discussion

Subsequent to a short stimulant effect, earthworms lost their motility on exposure to crude extracts of Os and Ws. Each crude extract containing 20 and 40 mg/ml, produced dose-dependent paralysis ranging from loss of motility to loss of response to external stimuli, which eventually progressed to death. In the case of Os extract paralysis was evident at 6.0±0.5 and 2.5±0.6 minutes, while death occurred within 13.5 ± 1.2 and 6.5 ± 0.7 for each 20 and 40 mg/ml concentration. In the case of Ws extract paralysis was evident at 6.5±0.5 and 2.8±0.8 minutes respectively, while death occurred within 13.9±1.2 and 7.1±0.9 for each 20 and 40 mg/ml concentration. The higher concentrations of crude extract produced paralytic effect much earlier and the time to death was shorter. Haemorrhagic and necrotic spots were observed externally on the worms, with the higher concentrations. The effect of each crude extract was compared with Peprazine citrate (20 mg/ml), which was found to produce paralyses within 7.0±0.6 min, while death occurred in 10.9 ± 1.3 minutes.

Conclusion

The comparative study of both the plant extracts showed that Os proves to be a better anthelmintic agent than Ws, as Os has taken lesser time in terms of paralysis and death both. Future scope involves need of isolation of phytoconstituents responsible for activity.

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