



## ANTIHELMINTIC ACTIVITY OF VARIOUS FRACTIONS OF ETHANOLIC EXTRXCT OF *OCIMUM SANCTUM*

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### ABSTRACT

The ethyl acetate, the n-butanolic and the aqueous fractions of the ethanolic extract of *Ocimum Sanctum* were evaluated separately for anthelmintic activity on adult Indian earthworms, *Eudrilus Eugenia*; the n-butanolic fraction being more potent. The activities were compared with the reference drug piperazine citrate and albendazole, respectively.

**Key words:** Anthelmintic, *Ocimum sanctum*, Albendazole, Piperazine citrate.

### INTRODUCTION

Helminthiasis infections are most common in peoples of tropical and sub-tropical regions. The world health assembly, in a number of resolutions has emphasized the need to the use of natural products with therapeutically proven efficacy particularly in patients residing in tribal areas, who are very much prone to attack of several infections due to lack of knowledge about proper sanitation. Search for anthelmintic factor in plants therefore remains a potential area of investigation.

*Ocimum Sanctum* (family- *Labiatae*), commonly known as 'Tulsi' in India, worshipped every morning by most Hindus. It is perhaps the most sacred plant in India and is referred to in Ayurveda for its healing and life giving properties such as stimulant, aromatic, anticitarrhal, spasmolytic, diaphoretic etc. It is an upright, 30-60 cm tall plant covered with soft hairs. The stems are square in transection, and the leaves are opposite, elliptical-oblong with relatively long petioles and serrated leaf margins.

In view of pharmacological evidence showing sacred basil to possess multiple

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pharmacological effects including immuno-modulating, anti-stress, hepatoprotective, chemopreventive, anti-inflammatory and antioxidant activity<sup>1,2</sup>. The tribes of many regions use the juice of fresh aerial parts of *Ocimum sanctum* for treating intestinal roundworm parasites<sup>3,4</sup>. Hence in the present investigation, the anthelmintic activity of the different fractions (ethyl acetate, n-butanol and water) of ethanolic extract of whole plant of *Ocimum Sanctum* has been reported.

## EXPERIMENTAL

### Plant material

The whole plant of *Ocimum sanctum* was collected from Uttara Kannada district of Karnataka in September 2006 and was authenticated by the taxonomist of the Department of Botany, Gokhale Centuary College of Science, Ankola (Uttara Kannada), Karnataka. After authentication, fresh young matured plants were collected in bulk from the rural belt of Uttara Kannada district during early summer, washed, shade dried and then milled in to coarse powder by a mechanical grinder.

### Preparation of extracts

The powdered plant material was defatted with petroleum ether (60-80°) and then extracted with ethanol using Soxhlet apparatus. The solvent was then removed under reduced pressure, which was obtained as a dark green colored sticky residue. The residue was then partitioned successively between ethyl acetate-water system and then between n-butanol water system. The ethyl acetate, the n-butanolic and the aqueous fractions of the ethanolic extract are referred to as OS-I, OS-II and OS-III, respectively. The dried extracts were suspended in 1% gum acacia in normal saline (vehicle) and used for anthelmintic activities.

### Evaluation of antihelmintic activity

The anthelmintic activity was carried out on adult Indian earthworm, *Eudrilus Eugenia* due to its anatomical and physiological resemblance with the intestinal roundworm parasites of human beings<sup>5-7</sup>. The activity was carried out by using *Grag's* method<sup>8</sup>. Eighteen group of approximately equal sized Indian earthworm consisting of six earthworms in each group were released into 50 mL of desired formulation containing in a petri plate of 4 inches diameter. Each group was treated with one of the following: vehicle (1% gum acacia in normal saline), piperazine citrate (15 mg/mL), albandazole, (10 mg/mL)

or different fractions (2.5, 5, 10, 25 and 50 mg /mL), respectively. Observations were made for the time taken to paralysis and/or death of individual worms for 4 h. Paralysis was said to occur when the worms do not revive even in normal saline. Death was concluded when the worm's loss their motility followed with fading away of their body color.

## RESULTS AND DISCUSSION

Data in Table 1 reveals that all the extracts of *Ocimum Sanctum* possess anthelmintic activity, when compared with the standard drug piperazine citrate and albandazole.

**Table1: Anthelmintic activity of different fractions of ethanolic extract of *Ocimum Sanctum***

Group (n = 6)	Treatment	Conc. used (mg/mL)	Time taken for paralysis (min)	Time taken for death (min)
I	Vehicle	-	-	-
II	Piperazine Citrate	15	13.30 ± 0.23	28.45 ± 0.56
III	Albendazole	10	26.48 ± 0.43	50.35 ± 0.40
IV	OS-I	2.5	164.48 ± 1.39	-
V	OS-I	5	96.38 ± 0.56	-
VI	OS-I	10	43.35 ± 0.28	94.27 ± 0.23
VII	OS-I	25	20.32 ± 0.54	51.26 ± 0.82
VIII	OS-I	50	14.28 ± 0.36	34.14 ± 0.65
IX	OS-II	2.5	120.35 ± 1.04	-
X	OS-II	5	72.38 ± 0.42	160.26 ± 0.72

Cont...

Group (n = 6)	Treatment	Conc. used (mg/mL)	Time taken for paralysis (min)	Time taken for death (min)
XI	OS-II	10	32.36 ± 0.64	72.32 ± 0.32
XII	OS-II	25	17.24 ± 0.69	40.12 ± 0.40
XIII	OS-II	50	7.47 ± 0.19	20.21 ± 0.62
XIV	OS-III	2.5	132.32 ± 0.38	-
XV	OS-III	5	102.28 ± 1.49	-
XVI	OS-III	10	51.38 ± 0.84	109.24 ± 0.44
XVII	OS-III	25	32.43 ± 0.43	68.18 ± 0.76
XVIII	OS-III	50	10.44 ± 0.48	28.10 ± 0.15

All values are expressed as Mean ± SEM ; OS-I, OS-II and OS-III are the ethyl acetate, the n-butanolic and the aqueous fractions of ethanolic extract respectively.

The n-butanolic fraction (OS-II) was more potent than the other two fractions (OS-I and OS-III) of ethanolic extract of whole plant of *Ocimum Sanctum*. The study reveals that none of the fractions at a dose level of 2.5 mg/mL was lethal to worms. Potency of the fraction was found to be inversely proportional to the time taken for paralysis/death of worms.

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