



## Invitro Anti helmentic Activity of aqueous and alcoholic extracts of *Aerva lanata* Seeds and leaves

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### Abstract:

The antiparasitic activity of the seed and leaf extracts of *Aerva lanata* (Amaranthaceae) were tested against a tapeworm and an earthworm, particularly ethanolic extract showing to be better against tapeworms and earthworms than the standard Albendazole, which are used in the treatment of helmentic parasite infections.

**Keywords:** *Aervalanata*; *Antiparasitic*; *Tapeworm*; *Earthworm*.

### Introduction:

Helminth infections are among the most common infections in man, affecting a large proportion of the world's population. In developing countries they pose a large threat to public health, and contribute to the prevalence of malnutrition, anaemia, eosinophilia, and pneumonia. Although the majority of infections due to worms are generally limited to tropical regions, they can occur to travelers who have visited those areas, and some of them can be developed in temperate climates [1]. The limited availability and affordability of pharmaceutical medicines mean that the world's population depends to a great extent on traditional medical remedies, and some 20,000 species of higher plants are used medicinally throughout the world. Many well-known drugs listed in the modern pharmacopoeia have their origins in nature, including for example, quinine from the bark of Cinchona tree for the treatment of malaria, which has been followed by the subsequent development of the synthetic derivatives chloroquine, amodiaquine, primaquine and mefloquine. More recently, the wider recognition of the antimalarial activity of artemisinin from the herb *Artemisia annua* has led current research to focus on the development of a large number of synthetic and semisynthetic compounds, which are more active than artemisinin [2].

*Aerva lanata* Linn (Amaranthaceae) is an herbaceous perennial weed growing wild in

the hot region of India. *Aerva lanata* has been claimed to be useful as diuretic, anthelmintic, antidiabetic, expectorant and hepatoprotective in traditional system of medicine [3]. Antimicrobial and cytotoxicity activity [4], diuretic [5], urolithiasis [6] and anti-inflammatory activity [8] of *Aerva lanata* has been reported. Canthin-6-one and beta-carboline alkaloids were isolated from *Aerva lanata* leaves [8].

### Materials and methods:

#### Standards:

Albendazole is used in the treatment of roundworm and threadworm (Cestoda) infections.

#### Chemicals:

All chemicals and reagents used for the study of pharmacological activity and phytochemical investigations are of analytical grade. Albendazole was purchased from Sigma (BHIMAVARAM). Purification was performed prior to use.

#### Collection and extraction of leaves and stem:

The plant was collected during the month of January from Rajiv Gandhi University Campus Karnataka. The plant was authenticated by Prof. B Anil of General medicine (Pancha karma specialist), Rajiv Gandhi university, Bangalore. The leaves and stem of plant was removed and dried under shade and powdered in a mechanical grinder. The leaf and stem powder was extracted with ethanol (300ml) separately in a soxhlet apparatus for 18 hrs and the

**Table 1:** Organisms used in anti helmentic test

Scientific Name	Group	Common Name	Host	Disease
<i>Taenia solium</i>	Cestoda	Pork tape worm	Pig, man	Taeniasis (adult form) and cysticercosis (larvae form)
<i>Pheretima posthuma</i>	Annelida	Earth worm		

**Table 2:** Anti helmintic activity<sup>a</sup> of *Aerva lanata* leaf extract

Type of Extract	Dose (mg/ml)	Time (min) taken for paralysis of Earth worms (Mean values)	Time (min) taken for death of Earth worms (Mean values)	Time (min) taken for death of tape worms (Mean values)
Ethanolic	2.5	156	125	161
	5	126	114	128
	10	72	74	92
	20	46	35	51
Aqueous	2.5	178	164	198
	5	135	117	154
	10	98	105	134
	20	45	38	52
Albendazole	2.5	188	164	229
	5	135	129	145
	10	111	127	116
	20	53	34	58
Vehicle (1% tween 20)	-	-	-	-

<sup>a</sup> Experiments were carried out at room temperature; <sup>b</sup> In the control group containing 1% tween 20 all the worms were alive even after 24 hrs; <sup>c</sup> See statistical analysis in the text

ethanolic extract was used for pharmacological work.

#### *Anthelmintic bioassays:*

The tapeworm *Taenia solium* (Cestoda, Taeniidae) and the earthworm *Pheretima posthuma* (Annelida, Megascolecidae) were used for evaluating the antiparasitic activity of leaf and stem extracts using a reference substance for comparison.

#### *Activity against earthworms and tapeworms:*

The antihelmentic activity was evaluated on the tapeworm *Taenia solium* (Cestoda, Taeniidae) and the earthworm *Pheretima posthuma* (Annelida, Megascolecidae) collected due to its anatomical and physiological resemblance with the intestinal round worm parasites of human beings.

**Table 3:** Anti helminthic activity<sup>a</sup> of *Aerva lanata* stem extract

Type of Extract	Dose (mg/ml)	Time (min) taken for paralysis of Earth worms (Mean values)	Time (min) taken for death of Earth worms (Mean values)	Time (min) taken for death of tape worms (Mean values)
Ethanolic	2.5	142	135	156
	5	119	92	131
	10	82	81	99
	20	42	28	55
Aqueous	2.5	172	166	183
	5	128	103	143
	10	92	97	101
	20	28	23	33
Albendazole	2.5	188	164	229
	5	135	129	145
	10	111	127	116
	20	53	34	58
Vehicle (1% tween 20)	-	-	-	-

<sup>a</sup>Experiments were carried out at room temperature; <sup>b</sup> In the control group containing 1% tween 20 all the worms were alive even after 24 hrs; <sup>c</sup> See statistical analysis in the text.

The method of Mathew *et al*<sup>9</sup>. were followed for anti helminthic screening. Emulsions of the leaf and stem extract in Tween 20 (1%) were prepared, and further diluted to give doses of 2.5, 5, 10, 20 mg/ml emulsions. Albendazole solutions of the same concentrations were prepared using distilled water, and used as reference standard. Two ml each of the emulsion and the solution were diluted to 10 ml each using physiological solution, and further poured into Petri dishes. The antiparasitic activity was determined in duplicate. Six worms of about the same size per Petri dish were used. The death and/or total paralysis time were

recorded at room temperature. The death of the worm was ascertained by transferring it into a beaker containing hot water at 50°C, which stimulated and induced movements if the worm was alive. Five independent experiments were carried out for each observation to confirm the results.

#### Results:

##### Statistical analysis:

The precision of this test methodology was determined by statistical analysis of all results on paralysis (P) and/or death (D) time. Repeatability was considered as the difference between successive results obtained by the same operator with the same

chronometer under constant conditions on identical test worm, in the normal and correct operation of the test methodology. Reproducibility was considered as the difference between two single and independent results obtained by different operators working in different laboratories on identical test worm in the longest assay in the normal and correct operation of the test methodology.

**Discussion:**

In order to validate scientifically the use in traditional medicines in antiparasite screens, the extract of the leaves and stem of *Aerva lanata* was studied. The species assayed, common name, host, and disease produced are shown in Table 1. One species of Cestodes and one species of Annelids were used. The cestode *Taenia solium* causes taeniasis and cysticercosis in man and pig. The earthworm *Pheretima posthuma* is one of the most important soil invertebrate in promoting soil fertility. Its feeding and burrowing activities break down organic matter and release nutrients and improve aeration, drainage, and aggregation of soil. Earthworms are also important components of the diets of many higher animals<sup>10</sup>. However, it was used in order to investigate the activity of *Aerva lanata* against annelids, since there are human parasitic annelida such as *Limnatis* sp. and *Haemadipsa* sp. that primarily affect the skin<sup>11</sup>.

The results in Table 2 and Table 3 were subjected to statistical analysis. The precision of this test methodology of all results on paralysis (P) and/or death (D) time is as follows.

**Repeatability:** The difference between successive results obtained by the same operator with the same chronometer under constant conditions on identical test worm in the normal and correct operation of the test methodology exceeds the following values in one case in twenty:  $0.10 (x + 3 \text{ min})$ . Where x, the mean replicate data in minutes.

**Reproducibility:** The difference between two single and independent results obtained by different operators working in different laboratories on identical test worm, in the longest assay, in the normal and correct operation of the test method, would exceed the following values only in one case in twenty:  $0.22 (x + 3 \text{ min})$ . Where x are the mean data obtained, in minutes, by two different laboratories.

The statistical analysis of the results showed that the methodology used is in agreement with validation test criteria.

The Anti-helmentic activity was evaluated. The Aqueous extract of leaf and stem of *Aerva lanata* at high concentration showed good anti helmentic activity. The alcoholic extract of leaf and stem of *Aerva lanata* at normal concentration only showed good anti-helmentic activity and this is compared with the effect produced by the reference standard drug Albendazole. The results in Table 2 and Table-3 indicate that the Aqueous and Alcoholic extracts obtained from the leaves and stems of *Aerva lanata* are active against both the tapeworms and the earthworms tested. It is worth to mention that the leaf and stem alcoholic extracts are comparatively more active than Albendazole against *Taenia solium*. These findings support the use of *Aerva lanata* as anti-helmentic drug in the traditional medicine. The present study reveals that the Ethanolic extract was more potent than the aqueous extract even though both the extracts were endowed with antihelmentic activity.

**Conclusion:**

In conclusion, it is revealed that the Aqueous and Alcoholic extracts obtained from the leaf and stem of plant *Aerva lanata* possess good anti helmentic activity. Work is still under progress to explore the chemical nature of the active constituents and other pharmacological investigations are also under evaluation.

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