



## Anthelmintic potential of some selected plants having nutritional value used by Tribals of Ganjam district, Odisha-India

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### ABSTRACT

This study was undertaken to determine the anthelmintic activity of some of the selected plants like *Eleusine coracana*, *Cephalandra indica* and *Colocasia esculenta* commonly consumed by different tribals of Ganjam, Odisha-India for nutritional purposes using *in vitro* biological models like tapeworms (*Raillietina spiralis*), roundworms (*Ascaridia galli*) and earthworms (*Pheretima posthuma*) where Albendazole and Piperazine citrate were used as reference standards. It was revealed that the ethyl acetate extract of *Eleusine coracana* was most potent which was well comparable with both standard drugs followed by ethyl acetate extract of other two plants, but at little higher doses. Methanolic and petroleum extracts of all these plants were endowed with minute anthelmintic property, which were not up to the standards. The present study enlightens the potential usefulness of those selected plants as good anthelmintic agents.

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### Introduction

Infections with helminths, or parasitic worms, affect more than two billion people worldwide. They are the most common infectious agents of humans in developing countries and produce a global burden of disease and contribute to the prevalence of malnutrition, anaemia, eosinophilia and pneumonia. People living in poverty in developing countries often suffer from helminth infections, which more often physically impair their hosts than kill them. 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 develop in temperate climate<sup>1</sup>.

The origin of many effective drugs is found in the traditional medicine practices and in view of this several researchers have undertaken studies to evaluate folklore medicinal plants for their proclaimed anthelmintic efficacy<sup>2</sup>. Plants have been found to rich botanical for anthelmintic activity. The traditional medicines used by various tribes hold a great promise as source of easily available effective anthelmintic agents to the people, particularly in developing countries, including India<sup>3</sup>.

*Colocasia esculenta* has a wide reputation among natives of being curative for intestinal worm infections<sup>4,5</sup>. *Cephalandra indica* is a established medicine used to treat helminth infection since ancient times in India by traditional practitioners of Ayurveda<sup>6,7</sup>. *Eleusine coracana* is one of the important foods of many tribes throughout the world. It is also used as very good anthelmintic agent traditionally<sup>8,9</sup>. As after through literature survey it has been found that no such extensive research have been done in this area yet, we attempted to investigate these medicinal plants for their claimed anthelmintic activity.

### Materials and methods

#### Plant material

These plants were identified and authenticated by taxonomist of Khallikote Auto. College, Berhampur-Odisha, India. The voucher herbarium specimens (no.-CPS/HS-268, 269 & 270) were deposited in the herbarium of College of pharmaceutical Sciences, Berhampur-Odisha, India for future reference. After authentication, respective seeds and leaves were collected separately (during April-June 2011) in bulk from young matured plants cultivated by tribals of rural hill area of Mohuda, Ganjam-Odisha.

#### Extraction and fractionation

After collection, these plant materials were washed thoroughly, shade dried and then milled in to coarse powder by using mechanical grinder. All the powders of those different plants were passed through sieve number 40 and used for further studies. Powder of seeds of *Eleusine coracana* (250 gm), leaves of *Cephalandra indica* and *Colocasia esculenta* (250 gm each) were separately extracted successively with petroleum ether (60-80°C), ethyl acetate and methanol using Soxhlet apparatus. The solvents were then removed under reduced pressure to obtained sticky residues.

#### Chemicals & Drugs used

Petroleum Ether (60-80°C), Ethyl acetate and Methanol (Merck Ltd.); saline water (Claris Lifesciences Ltd.). Albendazole (Cipla Ltd.) and Piperazine citrate (Glaxo Smithkline Ltd.) were used as reference standards for anthelmintic study.

#### Preliminary phytochemical screening

Preliminary phytochemical screening of all the extracts was done by using standard methods<sup>10</sup> to know the nature of phytoconstituents present in it (Table. 1).

Table 1. Preliminary phytochemical screening of *E. coracana*, *C. indica* and *C. esculenta*

TEST FOR	<i>Eleusine coracana</i>			<i>Cephalandra indica</i>			<i>Colocasia esculenta</i>		
	PE	EE	ME	PE	EE	ME	PE	EE	ME
Alkaloids	+	-	-	+	-	-	+	-	-
Carbohydrates	-	-	+	-	-	+	-	-	+
Glycosides	-	-	+	-	-	-	-	-	+
Triterpenoids	-	+	-	-	+	-	-	+	-
Tannins-phenolic compounds	-	-	+	-	-	-	-	+	+
Protein & amino acids	+	+	-	-	-	-	-	-	-
Gum & mucilage	-	-	-	-	-	+	-	-	-
Flavones & flavonoids	-	+	+	-	+	-	-	+	-
Saponins	-	-	-	-	-	+	-	-	+
Steroids & sterols	+	-	-	+	-	-	+	-	-

+ stands for present and - stands for absent; PE -Pet. Ether Extract; CE-chloroform Extract; ME-Methanol Extract

Table 2. Evaluation of anthelmintic Activity of *E. coracana*, *C. indica* and *C. esculenta* Time taken for paralysis (P) and death (D) of worms in min

		Conc <sup>n</sup>	<i>Raillietina spiralis</i>		<i>Ascaridia galli</i>		<i>Pheretima posthuma</i>	
		Mg/ml	P	D	P	D	P	D
<b>Control</b>		-	-	-	-	-	-	-
<i>Eleusine coracana</i>	Pet..Ether Extract	10	68±0.11	98±0.75	68±0.33	90±0.87	75±0.71	98±0.21
		20	58±0.61	89±0.51	55±0.45	82±0.92	64±0.44	89±0.32
		40	44±0.22	84±0.76	48±0.76	71±0.13	55±0.29	81±0.86
	Ethyl acetate Extract	10	45±0.32	69±0.38	40±0.72	67±0.65	35±0.17	67±0.19
		20	36±0.81	63±0.87	35±0.12	58±0.14	30±0.31	62±0.14
		40	25±0.76	57±0.62	24±0.56	50±0.06	23±0.32	57±0.22
	Methanolic Extract	10	52±0.23	81±0.55	55±0.26	73±0.58	50±0.12	67±0.04
		20	39±0.12	77±0.34	47±0.08	63±0.51	42±0.12	53±0.14
		40	29±0.12	60±0.48	27±0.57	55±0.72	33±0.49	47±0.26
<i>Cephalandra indica</i>	Pet..Ether Extract	10	81±0.61	--	89±0.87	95±0.25	91±0.11	--
		20	73±0.56	98±0.75	76±0.47	87±0.22	83±0.40	96±0.08
		40	57±0.33	83±0.38	53±0.12	78±0.23	67±0.67	89±0.55
	Ethyl acetate Extract	10	53±0.19	75±0.79	48±0.23	73±0.13	43±0.31	71±0.54
		20	45±0.17	68±0.91	39±0.6	66±0.11	37±0.6	65±0.07
		40	33±0.31	61±0.11	30±0.32	58±0.25	29±0.04	60±0.6
	Methanolic Extract	10	51±0.64	87±0.27	58±0.02	91±0.59	59±0.17	88±0.22
		20	42±0.23	69±0.09	45±0.33	78±0.09	47±0.68	68±0.11
		40	32±0.55	60±0.02	33±0.17	68±0.28	35±0.92	59±0.35
<i>Colocasia esculenta</i>	Pet..Ether Extract	10	97±0.83	--	94±0.13	--	--	--
		20	88±0.22	--	88±0.2	95±0.08	93±0.22	--
		40	79±0.33	94±0.45	77±0.36	89±0.57	83±0.23	92±0.8
	Ethyl acetate Extract	10	59±0.27	80±0.92	53±0.23	78±0.98	57±0.26	79±0.73
		20	47±0.98	72±0.44	44±0.36	69±0.24	46±0.88	70±0.24
		40	39±0.17	65±0.33	35±0.76	61±0.78	37±0.31	62±0.37
	Methanolic Extract	10	63±0.54	98±0.18	68±0.12	98±0.13	65±0.47	92±0.13
		20	49±0.12	83±0.04	49±0.29	88±0.51	57±0.07	83±0.02
		40	38±0.45	67±0.18	38±0.23	77±0.42	49±0.28	71±0.09
Albendazole		10	22±0.64	54±0.26	20±0.14	48±0.84	22 ±0.28	56± 0.78
Piperazine citrate		10	27±0.48	58±0.54	25±0.18	53±0.78	26±0.68	61±0.28

P=paralysis; D= death, each value represents mean ± SEM (N=6).

### Anthelmintic activity

The *in-vitro* anthelmintic study was done by using tapeworms (*Raillietina spiralis*), roundworm *Ascaridia galli* Schrank (Nematode) and adult earthworms *Pheretima posthuma* L.Vaill (Annelida)<sup>11</sup>. Use of *Raillietina* and *Ascaridia* species as a suitable model for screening of anthelmintic drug was advocated earlier<sup>12,13</sup>. The adult earthworms were selected for this study due to their anatomical and physiological resemblance with the intestinal roundworm parasites *Ascaris lumbricoides* of human beings<sup>14,15</sup>. Roundworms and tapeworms were obtained from infested intestines of fowls *Gallus spadiceus* (Phasianidae) of local slaughter house and washed with normal saline solution to remove all the faecal matter. These intestines were then dissected and worms were collected and kept in normal saline

solution. The earthworms were collected from moist muddy soil from the water logged areas. The average size of round worm was 5-6 cm, average size of tapeworm was 6-7 cm and average size of earthworm was 8-9 cm. These helminths and earthworms were identified in Zoology Department, Berhampur University- Berhampur, Odisha and services of veterinary practioners were also utilized to confirm the identity of worms.

The anthelmintic assay was carried out as per some established methods previously followed<sup>11,14</sup>. Test samples of each extract was prepared at the concentrations, 10, 20 and 40 mg/ml in distilled water and six helminths/worms i.e. *Raillietina spiralis*, *Ascaridia galli* and *Pheretima posthuma* of approximately equal size (same type) were placed in each nine

cm Petri dish containing 25 ml of above test solution of extracts. Albendazole (10 mg/ml) and Piperazine citrate (10 mg/ml) was used as reference standard and saline water as control<sup>14,16,17</sup>. All the test solution and standard drug solution were prepared freshly before starting the experiments. Observations were made for the time taken for paralysis was noted when no movement of any sort could be observed except when the worms were shaken vigorously. Time for death of helminths/worms were recorded after ascertaining that worms neither moved when shaken vigorously nor when dipped in warm water (50°C). All the results were shown in Table 2 and expressed as a mean  $\pm$  SEM of six worms in each group.

### Results and discussion

In our study it was observed that the ethyl acetate extracts of all the plants (EEEC, EECI and EECE) were more potent which is well comparable with both standard drugs followed by Methanolic extracts (MEEC, MECI and MECE), but at higher doses. Petroleum extracts (PEEC, PECE and PECE) of all the three plants were endowed with minute anthelmintic property, which were not upto standards.

The activity revealed concentration dependence nature of the different extracts. Potency of the extracts was found to be inversely proportional to the time taken for paralysis/death of the worms (Table. 2). Ethyl acetate extracts of all the selected plants (showing most potent anthelmintic activity) contains mainly flavones and flavonoids, triterpenoids and phenolic compounds may be responsible for the anthelmintic activity<sup>18,19,20,21</sup>. Moderate anthelmintic activity of the methanolic extracts of different plants may be due to the presence of glycosides, phenolic compounds, saponins and flavonoids<sup>18,19,20</sup> present in it. Our results from the present study indicate the potential usefulness of *Eleusine coracana*, *Cephalandra indica* and *Colocasia esculenta* in the treatment of helminthiasis. Attempts for the isolation and characterisation of the active constituents responsible for such activities are currently under progress. Further studies are necessary to understand the exact mechanism of action.

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