Anthelmintic potential of various extracts of the rhizomes of *Curcuma amada* Roxb.

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**ABSTRACT**

**Objective:** To investigate the anthelmintic potential of various extracts of the rhizomes of *Curcuma amada* Roxb.

**Methods:** Cyclohexane, ethyl acetate, methanol and aqueous extracts of *Curcuma amada* Roxb. against *Pheretima posthuma*, an Indian species of earthworm. Various concentrations (25, 50 and 100 mg/mL) of these extract were evaluated for anthelmintic activity by recording the time required for paralysis and death of worms. Albendazole was used as a reference standard drug.

**Results:** Methanol and aqueous extracts shows most significant anthelmintic activity compared with standard drug.

**Conclusions:** Since methanol and aqueous extracts were tested by different chemical tests. It shows presence of carbohydrates, steroid, alkaloids, glycosides and flavanoids. These phytoconstituents may be responsible for the said activities.

**KEYWORDS**

Anthelmintic activity, *Curcuma amada*, Albendazole, *Pheretima posthuma*

1. Introduction

*Curcuma amada* Roxb. (*C. amada*) (Family: Zingiberaceae) is perennial rhizomatic aromatic herb which is known as mango ginger, ambha haldi and is available from November to April. The genus originated in the Indo–Malayan region, and is widely distributed in the tropics of Asia to Africa and Australia. Mango ginger (*C. amada*) is a unique spice having morphological resemblance with ginger but imparts a raw mango flavor[1]. The rhizomes are useful in vitiated conditions of pitta, anorexia, dyspepsia, flatulence, colic, bruises, wounds, chronic ulcers, skin diseases, pruritus, fever, constipations, stranguary, hiccough, cough, bronchitis, sprains, gout, halitosis, otalgia, cooling. They possess carminative, digestive, stomachic, demulcent, vulnerary, febrifuge, alexertic, aphrodisiac, laxative, diuretic, expectorant, anti-inflammatory and antipyretic effects. The main use of mango ginger rhizome is in the manufacture of pickles, culinary preparations, salads as a source of raw mango flavour for foods and therapeutic purpose. Ayurveda, the oldest system of medicine in India, has given importance to rhizome as an appetizer, alexiteric, antipyretic and laxative. According to Unani systems of medicine, it is a diuretic, maturant, emollient, expectorant, antipyretic and appetizer. It is useful against inflammation in the mouth, ear, as well as gleet, ulcers on the male sex organs, scabies, lumbar and stomatitis[2–6]. It is also used as antimicrobial, antifungal, antibacterial and anti-inflammatory agent as well as traditionally for their medicinal and functional properties[7–8].
2. Materials and methods

2.1. Plant material

The fresh plants rhizomes of *C. amada* were collected in the month of November 2011 from the local areas of Shrigonda, Ahmednagar district, Maharashtra. The plant was authenticated by Botany Department, Chatrapati Shivaji College, Shrigonda, Ahmednagar, India.

2.2. Preparation of extracts

The collected plants rhizomes of *C. amada* were separated and air dried in shade at room temperature, then they were crushed in electric blender powder and subjected to extraction by using Soxhlet’s extractor. By using successive solvent extraction, dried powdered rhizomes material was extracted with cyclohexane, ethyl acetate, methanol and aqueous solvents. The extract was dried at low temperature under reduced pressure.

2.3. Worm collection and authentication

Indian adult earthworms *Pheretima posthuma (P. posthuma)* were collected from moist soil of the vermiculture plant, Government Agriculture Department Farm, Kashi, Taluka Shrigonda, District Ahmednagar, Maharashtra and authenticated from the Zoology Department, Chatrapati Shivaji College, Shrigonda, Ahmednagar, India. Then all collected worms were washed with normal saline to remove all the faecal matter and used for the anthelmintic study. The earthworms of 3–5 cm in length and 0.1–0.2 cm in width were used for all the experimental protocol.

2.4. Sample preparation

Test sample for *in vitro* study were prepared separately by dissolving and suspending 2 g of cyclohexane, ethyl acetate, methanol and aqueous extract respectively in minimum amount of 0.2% v/v of Tween 20 as a suspending agent. The volume was adjusted to 20 mL with normal saline so as to obtain concentration of 25 mg/mL, 50 mg/mL and 100 mg/mL for each type of extract.

2.5. Anthelmintic activity

The anthelmintic activity was performed on Indian adult earthworms (*P. posthuma*) as it has anatomical and physiological resemblance with the intestinal roundworm parasites of human being. *P. posthuma* worms are easily available and used as suitable model for screening anthelmintic drugs. The 25 mg/mL, 50 mg/mL and 100 mg/mL of normal saline concentration of each group of cyclohexane, ethyl acetate, methanol and aqueous extracts and standard (20 mg/mL) were prepared and six earthworms with approximately equal size were released in each group. Observations were made for the time taken for paralysis or death of individual worms. Paralysis was said to occur when the worms lose their motility followed by fading away of their body color. Albendazole (20 mg/mL) was used as standard while normal saline as control. All the results were expressed as Mean±SD of six animals in each group.

### 3. Results

Experimental data showed that, the methanol and aqueous extract of the rhizomes of *C. amada* has shown more significant anthelmintic activity as compare to cyclohexane and ethyl acetate extract in dose dependent manner as shown in Table 1.

#### Table 1

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Group</th>
<th>Concentration (mg/mL)</th>
<th>Time of paralysis (min) (mean±SEM)</th>
<th>Time of death (min) (mean±SEM)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>I</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Albendazole</td>
<td>II</td>
<td>20</td>
<td>64.49±0.1±0.062</td>
<td>0.01±0.010±0.057</td>
</tr>
<tr>
<td>Cyclohexane extract</td>
<td>III</td>
<td>25</td>
<td>22.51±0.0±0.12</td>
<td>35.45±0.1±0.13</td>
</tr>
<tr>
<td>Ethyl acetate</td>
<td>IV</td>
<td>50</td>
<td>15.30±0.1±0.39</td>
<td>21.69±0.1±0.06</td>
</tr>
<tr>
<td>Methanol extract</td>
<td>V</td>
<td>100</td>
<td>07.00±0.1±0.139</td>
<td>11.37±0.1±0.169</td>
</tr>
<tr>
<td>Aqueous extract</td>
<td>VI</td>
<td>50</td>
<td>23.58±0.0±0.396</td>
<td>31.77±0.1±0.356</td>
</tr>
<tr>
<td>XII</td>
<td>50</td>
<td>14.45±0.1±0.291</td>
<td>19.59±0.1±0.460</td>
<td>15.41±0.1±0.364</td>
</tr>
<tr>
<td>Methanol extract</td>
<td>VII</td>
<td>100</td>
<td>09.57±0.1±0.174</td>
<td>35.45±0.1±0.13</td>
</tr>
<tr>
<td>Aqueous extract</td>
<td>VIII</td>
<td>100</td>
<td>17.40±0.1±0.175</td>
<td>35.45±0.1±0.13</td>
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<tr>
<td>Methanol extract</td>
<td>IX</td>
<td>50</td>
<td>14.45±0.1±0.066</td>
<td>21.76±0.1±0.945</td>
</tr>
<tr>
<td>Aqueous extract</td>
<td>X</td>
<td>100</td>
<td>04.50±0.1±0.300</td>
<td>06.14±0.1±0.102</td>
</tr>
<tr>
<td>Methanol extract</td>
<td>XI</td>
<td>50</td>
<td>26.10±0.1±0.355</td>
<td>26.39±0.1±0.359</td>
</tr>
<tr>
<td>Aqueous extract</td>
<td>XII</td>
<td>100</td>
<td>11.27±0.1±0.177</td>
<td>16.59±0.1±0.106</td>
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<tr>
<td>Methanol extract</td>
<td>XIII</td>
<td>50</td>
<td>04.51±0.1±0.235</td>
<td>06.11±0.1±0.635</td>
</tr>
<tr>
<td>Aqueous extract</td>
<td>XIV</td>
<td>100</td>
<td>26.10±0.1±0.355</td>
<td>26.39±0.1±0.359</td>
</tr>
</tbody>
</table>

Values are expressed as mean±SEM, One way ANOVA followed by Dunnett’s test, n=6 in each group. **P<0.01.

The shortest time required for paralysis and death was observed with concentration of 100 mg/mL of methanol and aqueous extract when compared to standard drug. Higher concentration of methanol extract showed maximum effect [4.63±0.30 min in paralysis and 6.41±0.10 min in death] and aqueous extract showed maximum effect [4.51±0.21 min in paralysis and 6.14±0.06 min in death]. Standard albendazole showed the paralysis at (64.49±0.10 min) and death at (108.33±0.06) min after release of worms in it. Albendazole by increasing chloride ion conductance of worms muscle membrane produced hyperpolarization and reduced excitability that led to muscle relaxation and flaccid paralysis. The rhizomes extract of plant *C. amada* not only showed paralysis but also caused death of worms at different concentration. It can be concluded that the active phytoconstituents may be responsible for anthelmintic activity present in the extract of *C. amada*.

### 4. Discussion

From the results it conclude that cyclohexane, ethyl acetate, methanol and aqueous extracts of various concentrations of *C. amada* gives dose dependant ant
amada show dose dependent anthelmintic activity with compared to standard drug albendazole. But among them methanol and aqueous extracts show most significant activity as compare to other extracts of cyclohexane and ethyl acetate. This response may be due to presence of phytoconstituents in methanolic and aqueous extracts. There is an insufficient quantity of data about the phytochemical constituents which prompted us to carry out further study concerning the phytochemical constituents. Very less documentation and information is available related to the C. amada. Therefore, according to this study it is concluded that, the genus Curcuma represents many species; most of them are fully explored but C. amada is not much studied. Traditional claims of this crude drug are yet to be pharmacologically explored to develop new compounds, which may be beneficial for future studies. C. amada is of very less documented data as well as almost untouched drug although it has high potential value related to botanical, economical and pharmacological point of view. This research paper may provide consolidated information to researchers, as well as practitioners to plan future studies in isolation of phytochemical as well as elucidate the mechanisms behind its traditional effects under pharmacological activities. Finally this work may prove that C. amada will be natural gift to human being.

Conflict of interest statement
We declare that we have no conflict of interest.

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Comments
Background
Nowadays helminthes infection is a cause of many acute as well as chronic diseases related to the various human beings. The aim of this study is to act against these helminthics worms with the help of a traditional Indian spice.

Research frontiers
Few extracts of this species were very effective in causing death of earthworms. But more specific type of data was not available on this type of activity. Since, the present work shows that all four extracts of the rhizomes gives dose dependent anthelmintic activity.

Related reports
Gill et al. reported the anthelmintic activity of ethanol and dichloromethane extracts and which was very effective in causing death of earthworms. These related reports were the preface to the study.

Innovations & breakthroughs
This research study may prompt new phytochemical constituents from this particular plant extracts which was not reported till today’s dates from a natural Indian spice against stomach disorders.

Applications
The research paper is an amazing contribution to the field of anthelmintic agents and disorders of stomach. This study can be used as a foundation for further articulation of research in the active chemical constituents useful in the treatment of intestinal disorders caused by helminthics.

Peer review
The chemical constituents useful in acting against the intestinal microbes have been listed and proved by this study. This paper has enormous findings in the field of botanical, pharmacological, phytochemical and pharmacognostic point of view. This particular research can be improved by in depth survey of chemical constituents and research.

References