ABSTRACT
The ethanolic extract of the leaves of Coleus aromaticus was evaluated for its anthelmintic efficacy in vitro. Graded doses of the extract (10, 50, 100, 200 μg/mL) showed significant anthelmintic activity, with their sensitivity when compared with the standard. Ivermectin and levamisole were used as reference drugs.

Key words: Anthelmintic, Ivermectin, levamisole, Coleus aromaticus.

I. INTRODUCTION
Helminthiasis is among the most important animal diseases inflicting heavy production losses. The disease is highly prevalent particularly in third world countries due to poor management practices. Chemical control of helminthes coupled with improved management has been the important worm control strategy throughout the world.

The leaves of Coleus aromaticus Bentham were used in the East Indian archipelago, mainly in cases of aphthous stomatitis. For this purpose the Dutch Pharmacopoeia Ed.V, introduced the Species antiaphthosae with Coleus leaves as an active component, because of their antiseptic qualities. Older research ascribed this activity to carvacrol and perhaps thymol. Coleus aromaticus Bentham., (Lamiaceae), syn. Coleus amboinicus (Lour.) Spreng. is commonly known as Indian / country borage.

It is a large succulent herb with aromatic leaves, found abundantly in India. The leaves of this plant are traditionally used for the treatment of severe bronchitis, asthma, diarrhea, epilepsy, renal and vesicle calculi and fever, it has been reported to exhibit antilithiotic, chemopreventive, antiepileptic and antioxidant properties.

II. EXPERIMENTAL
2.1 Plant material and extraction
The plant material was collected from the plantation in the Medicinal Garden, School of pharmacy, Devi Ahilya Vishwavidhyalaya, Indore. The plant material was identified by a botanist, Dr.A.B.Sheerwani (Retd. Prof. and Head), Department of botany, Holkar Science College, Indore, and their voucher specimens were deposited in the Department of Pharmacognosy (No.C-04/HF), School of Pharmacy, Devi Ahilya Vishwavidhyalaya, Indore.

The dried leaves were powdered mechanically and extracted 95% ethanol by using soxhlet extractor. After 72h extract was concentrated under reduced pressure (22-26 mmHg) at 50-60°C (yield 6.2%). The presence of phytochemicals, alkaloids (Dragendorff’s), flavonoids (Shibata’s reaction), saponins (Frothing test), tannins (5% ferric chloride), terpenoids (2,4 dinitrophenylhydrazine), glycosides (fehling’s solution), steroids (Liebermann’s Burchard test) and anthraquinones (Borntrager’s test) were evaluated.

A Preliminary phytochemical screening gave positive test for glycoside and flavonoids.

2.2 Evaluation of anthelmintic activity
Anthelmintic activity (in vitro) by microwell plate assay. Levamisole and ivermectin were used as reference standard. The extract was dissolved in 1% DMSO.

The free living roundworm Rhabditis pseudoe longata (strain L.Lamy) was used for the anthelmintic screening. Roundworm strain was
collected from Choithram Hospital and Research Centre, Indore. The worms were cultivated at 25°C in a moist atmosphere in darkness in a solid medium constituted of autoclaved rabbit faeces. Three replicates were used for each concentration of ethanol extract.

III. RESULTS
The ethanolic extract of the seeds of *Coleus aromaticus* (10-200 μg/mL) exhibited potent anthelmintic activity (Table I). This result may lend support for the traditional use of the plant as an anthelmintic.

Preliminary phytochemical screening shows presence of glycoside and flavonoids. Secondary metabolite like glycoside and flavonoids, occur in several plants have been reported to show anthelmintic property by several investigators. Glycosidic and flavonoidal compounds, are shown to interfere with energy generation in helminth parasites by uncoupling oxidative phosphorylation or, binds to the glycoprotein on the cuticle of parasite, and cause death [7-9]. Further research is to be carried out to fractionate and purify the extract, in order to find out the molecule responsible for the anthelmintic activity observed.

<table>
<thead>
<tr>
<th>S. No.</th>
<th>Tested material</th>
<th>Conc.(µg/mL)</th>
<th>ED₅₀ (µg/mL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td><em>Coleus aromaticus</em> extract</td>
<td>10, 50, 100, 200</td>
<td>82.2 (60.2-130.8)</td>
</tr>
<tr>
<td>2.</td>
<td>levamisole*</td>
<td>1, 2, 4, 8</td>
<td>3.9 (2.8-5.2)</td>
</tr>
<tr>
<td>3.</td>
<td>Ivermectin*</td>
<td>0.5, 1, 2, 4</td>
<td>2.7 (2.1-3.2)</td>
</tr>
</tbody>
</table>

*aAll determination were done in triplicate.

*Reference standards

REFERENCES