In vitro and in-vivo efficacy of Eucalyptus citriodora Leaf in gastrointestinal nematodes of goats

Sushmita Sastya, Rajeev Ranjan Kumar and Stuti Vatsya

Abstract

The study was undertaken to evaluate the anthelmintic activity of crude powder (CP), crude aqueous (CAE) and crude methanolic (CME) extracts of Eucalyptus citriodora leaves. In vitro anthelmintic activity was evaluated against freshly collected adult Haemonchus contortus using Adult Motility Test (AMT). However, against eggs and larvae of gastrointestinal nematodes of goats using Egg Hatch Assay (EHA) and Larval Paralysis Test (LPT) at 0.156%, 0.312%, 0.625%, 2.5%, 5% and 10% concentration. In-vivo trial was conducted in goats naturally infected with GI nematodosis by using Faecal Egg Count Reduction Test (FECRT%). In AMT an average highest corrected motility of 100% was observed from 1.25–10% followed by 66.66%, 46.66%, 46.66% at 0.625%, 0.312% and 0.156% concentration, respectively. In EHA, CAE showed better ED₅₀=0.997mg/ml and ED₉₀=105.033 mg/ml values than CME (ED₅₀= 1.820 and ED₉₀= 52.182 mg/ml). Both CAE and CME showed highest (100%) inhibition of egg hatching at 50 and 100 mg/ml and minimum (60% and 54%) at 0.625%, 0.312% and 0.156% concentration, respectively. In LPT, CAE (ED₅₀= 5.305 and ED₉₀= 157.326mg/ml) and CME (ED₅₀= 5.503 and ED₉₀= 165.447 mg/ml) showed highest 100% and 100% and lowest 25% and 22% paralysis of third stage larvae of gastrointestinal nematodes at 100mg/ml and 1.56 mg/ml concentration, respectively. In in-vivo anthelmintic activity, CME showed highest (35%) reduction in faecal egg count followed by CAE (21.05%) and then CP (10.34%) @ 4gm/kg b.wt. on 7, 14 and 14 day post-treatment, respectively.

Keywords: Eucalyptus citriodora, anthelmintic activity, GI nematodosis, Goats

Introduction

Gastrointestinal (GI) nematodes are major problem and primary cause for affecting small ruminant production throughout the worldwide including India. They produce various pathogenic effect like anaemia, villous atrophy, pimply gut, enteritis and is a major cause of morbidity (Tyasi et al., 2015) leading to retarted growth, weight loss, reduce food consumption, lower milk production, impaired fertility and animal becomes more prone to secondary infections due to relaxation in immunity. Among these GI nematodes, H. contortus is most common and most pathogenic parasite because of its voracious blood feeding ability (Jabbar et al., 2006). Currently GI parasite control programs in small ruminants mainly depend through the use of anthelmintics. However, these are very expensive and often not easily available to the farmers in rural or remote areas and also the frequent use of the anthelmintics results in emergence of resistance (Jaiswal et al., 2013; Rialch et al., 2013) because of increase in severity of anthelmintic resistance, increase public awareness towards the drug residues and toxicity problem. Therefore there is urgent need, not only to develop alternative or complementary control method but also extends the life and preserve efficacy of current anthelmintics available in the market (Hoste et al., 2002). This has given a new area of research to find out alternative therapeutic agents for the control of GI nematodosis. Several plants have been identified having anthelmintic properties are available in India (Kumar et al., 2014 and 2016) These herbal plants are cheaper or affordable, easily available, easy to prepare and administer (Tariq et al., 2009), also are less toxic, biodegradable, and ecofriendly (Melo et al., 2003) so it is a good option for ordinary or small scale animal farmer. Use of traditional plants not only reduces the conventional anthelmintic treatment it also extends the life of anthelmintics (Chagas et al., 2008).

Eucalyptus spp. commonly called Safeda in Hindi and Blue Gum in English is basically a native plant of Australia. It is mainly cultivated for paper production, cosmetic and pharmaceutical purposes (Hasegawa et al., 2008).

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Correspondence

Sushmita Sastya
College of Veterinary and Animal Sciences G.B. pant University of Agriculture & Technology Pantnagar, Uttarakhand, India
Leaves have febrifuge, carminative, stimulant, expectorant, antifungal, antibacterial, anti-inflammatory, antiseptic, antimalarial and mosquito repellent properties. It increases the flow of saliva, gastric and intestinal juices and increases the digestion and appetite and also it has anthelmintic properties (Kuamr et al., 2015) [18]. To concentrate on anthelmintic property of E. citriodora, the present was aimed to evaluate the anthelmintic activity against GI nematodes of Goats.

Materials and Methods

Plant material

Eucalyptus citriodora was selected, on the basis of their documented anthelmintic properties or literature survey. Leaves of E. citriodora were collected (July 2015 to April 2016) from the plants situated in G.B. Pant University of Agriculture & Technology, Pantnagar. Plant were identified and authenticated by Department of Biological Sciences, College of Basic Science & Humanities, G.B. Pant University of Agriculture and & Technology, Pantnagar.

Preparation of extract

Leaves of E. citriodora were cleaned manually, dried in a well aerated room protected from sun and then they were dried in the oven at 50 °C. Powder was prepared using electric mixer grinder and stored in airtight container. About 30 gm of powdered sample was soaked separately in 400 ml of distilled water and 400 ml methanol in 1 lit conical flask, covered with aluminium foil and stirred one hour interval for 3-2 times and left undisturbed for overnight at room temperature and then filtered through Whatman paper No.1 and separating funnels. Then after, filtrate was transferred into separate glass tray which then placed in an oven at 40°C to dry the extract into powder. The extract residues were kept in airtight glass petridishes at 4 °C (refrigerator) for further use (Ombasa et al., 2012) [30]. Seven different dilutions of each extract were prepared viz. 0.156% (1.56mg/ml), 0.312% (3.125mg/ml), 0.625% (6.25mg/ml), 1.25% (12.50mg/ml), 2.5% (25mg/ml), 5% (50mg/ml) and 10% (100mg/ml). Thiabendazole dissolved in Dimethyl Sulfoxide (DMSO) at same concentrations as above and used as positive control and distilled water was used as negative control.

In vitro evaluation of anthelmintic activity of E. citriodora

Adult Motility Test

Collection of Haemonchus contortus worm

Adult H. contortus worms were collected from the abomasii of freshly slaughtered goats at Lalkuan, Nainital. The worms were washed and cleared off debris with distilled water. Then after, they were kept in lukewarm normal saline and brought to the laboratory, Department of Parasitology, College of Veterinary & Animal Sciences, Pantnagar. The cleaned worms were transferred with the help of needle in beaker containing Lock’s solution and incubated at 37 °C for acclimatize before beginning test (Bhatnagar et al., 1961) [2]. Adult motility test for anthelmintic activity of CAE, CME and CP of plant leaves were conducted on adult H. contortus of goats. Ten worms were exposed to 15 ml of seven different dilutions in separate Petri dishes and then incubated at 37 °C±1 °C for 24 hrs. Lock’s solution was used as positive and distilled water as negative control. The motility was observed after 1, 2, 4, 6, 12, 18 and 24 hrs intervals. The dead worms were easily recognized by their straight flat appearance without movement of head and tail region but for conformation the viability of the worms was determined by pinch technique (the absence of motility for an observation period of 5-6 second) as described by Neogi et al. (1964) [29]. The number of dead worm was recorded for each treatment. Corrected mortality was calculated as per the formula given by Sangwan and Sangwan (1998) [31].

% Corrected mortality = Total mortality - Control mortality

Egg Hatch Assay

Collection of GI nematode eggs and Egg hatch assay were conducted according to the standard protocol described by World Association for the Advancement of Veterinary Parasitology guidelines (Coles et al., 1992) [6]. Faecal samples were collected from goats naturally infected with GI nematodes and obtain a pooled sample by mixing several samples. Sample was stored anaerobically in screw top plastic bottle filled with water upto brim. Faecal sample was broken in mortar-pestle with water to make homogenised suspension. It was filter through tea strainer of 0.15 mm in size and filtrate was transferred into 15 ml conical centrifuge tubes. Then after it was centrifuge for 2 min. at 300xg and then supernatant was discarded. The sediment was resuspended in saturated salt solution and after gentle mixing, the suspension was re- centrifuged for 3 min at 130xg. Then tubes were filled with saturated salt solution until a meniscus was formed above the tube. A cover slip was placed gently above the meniscus for few minutes. The cover slip was plucked off with the help of artery forceps and the eggs were washed twice with deionised water into a conical centrifuge tube. Isolated egg suspension was counted in 100 µl and numbers of eggs were estimated per ml and diluted to 100-150/100µl. The assay was performed in 24 well plates as per Le Jambre (1976) [21]. 100µl of fresh egg suspension were exposed to 1ml of each prepared dilution of extract. The plate was incubated at 27 °C for 48 hours. Thereafter, two drops of Lugol’s iodine were added to each well to stop further hatching. A total of hundred eggs and hatched larvae (L1) were counted under compound microscope and percent inhibition of egg hatching was calculated.

Larval Parasitology Test

Faecal sample collected from each group was pooled and faecal sample was broken in mortar with help of pestle. The faeces were transfer into petridish and covered it. Then petridish was incubated at 27 °C in incubator upto 7 days. The infective Larvae (L3) were procured according to MAFF (1986) [24] using Baerman’s apparatus. Test was performed in 24 well plate in which 100µl of larvae suspension containing 100-150 larvae/100µl were exposed to different dilutions of extract residues. Thiabendazol and distilled water were used as positive and negative control, respectively. The plates were kept at room temperature (27 °C) for 24 hours. A total of hundred dead and live larvae were counted under compound microscope and percent larval paralysis was calculated.

In-vivo Evaluation of Anthelmintic Activity of E. citriodora

Location of Study Area

Evaluation of anthelmintic efficacy of CAE, CME and CP was carried out in goats from Department of Livestock Production & Management, College of Veterinary and Animal Sciences, Pantnagar and also in local areas, Sanjay colony and Jha colony of Pantnagar.
**Grouping and dosing of animals**

Total 42 adult goats age of 1-3 years of either sex naturally infected with mixed infections of gastrointestinal nematodes having >200 faecal egg count and history of not using any kind of anthelmintic treatment since last three months were selected, numbered and weighed during each trial. These animals were randomly divided into seven group viz. GI, GII, GIII, GIV, GV, GVI and GVII with 6 animals in each. The GI, GII and GIII groups were treated with CP, CAE and CME of *E. citriodora* leaves @ 2gm/kg b.wt. orally, while GIV, GV and GVI were treated @ 4gm/kg b.wt. orally. However, animals of GVII kept as untreated infected control.

**Faecal Egg Count Reduction Test (FECRT)**

Faecal samples were collected directly from rectum from individual animals of each treated and control group on 0 day (before) of treatment, 7 and 14 day post-treatment (7 and 14 DPT) and placed separately in plastic bag, marked them and transfer to laboratory. Intensity of egg per gram of faeces (EPG) of each faecal sample was determined by modified McMaster Technique (MAFF, 1971) [23] briefly 1g of fresh faeces was accurately weighed and suspended in 14ml saturated sodium chloride NaCl. The suspension was strained through a fine sieve. The larger particles of faeces were removed and the residue was left to pass through. The suspension was stirred well in order to obtain a completely homogenous distribution of eggs in the liquid. Using a Pasteur pipette, compartment of the McMaster slide was filled, avoid air bubbles. After a few minutes eggs from the concentration solution floats up and sticks to the cover glass. Number of eggs was counted in the compartment under low magnification. The egg per gram of faeces was calculated as follows:

\[
\text{Egg per gram (EPG)} = \frac{\text{Number of egg in the chamber}}{50}
\]

**Determination of Anthelmintic Efficacy**

The anthelmintic efficacy of the extracts residue was calculated using the following formula given by Dash et al. (1988) [27].

\[
\text{% efficacy} = \frac{\text{Pre - treatment EPG - Post treatment EPG}}{\text{Pre - treatment EPG}} \times 100
\]

**Coproculture**

Larvae were cultured according to MAFF (1986) [24] using Baermann’s apparatus. Gastrointestinal nematode larvae were identified on the basis of their morphological characters as described by Soulsby (1965) [36].

**Phytochemical analysis of extracts**

Extracts residue obtained from CAE and CME were tested for the presence of their phytoconstituents such as alkaloid, antraquinones, tannins, flavonoids, saponins, glycoside, resins, triterpencs, reucing sugars, proteins and coumarins by standard procedures (Sofowara, 1982) [38].

**Statistical Analysis**

The data of *In vitro* trials viz. Egg Hatch Assay and Larval Paralysis Test were analyzed statistically using probit analysis for calculation of ED$_{50}$ and ED$_{90}$ by SPSS version 16. However, for *in-vivo* trial multivariat two ways ANOVA was used to analyzed the data for calculation of F-value and difference between the mean were considered significant at $p > 0.05$.

**Results**

**In-vitro anthelmintic activity of *E. citriodora***

**Adult Motility Test**

Crude powder showed 40, 40, 100, 100, 100 and 100% mortality at 0.156%, 0.312%, 0.625%, 1.25%, 2.5%, 5% and 10% concentration respectively. CAE showed 100% mortality at all tested concentrations whereas CME and TBZ showed 0, 0, 0, 100, 100 and 100% mortality at 0.156%, 0.312%, 0.625%, 1.25%, 2.5%, 5% and 10% concentration respectively. Overall, CAE showed maximum (100%) adulticidal activity against *H. contortus* followed by CP (82.86%) and minimum (57.14%) with CME and TBZ (Table-1). *E. citriodora* showed overall 79.99% corrected mortality. An average highest corrected motility of 100% was observed from 1.25–10% followed by 66.66%, 46.66% and 46.66% at 0.625%, 0.312 and 0.156% concentration, respectively (Table-1).

**Egg Hatch Assay**

CAE and CME of *E. citriodora* caused significant dose dependent inhibition of egg hatching (Table-2). CAE (ED$_{50}$ = 0.997 and ED$_{90}$ = 105.003 mg/ml) showed 100% inhibition @ 50 and 100mg/ml concentration. However, at 25, 12.5, 6.25, 3.125 and 1.56 mg/ml concentration, inhibition of egg hatching was 92, 86, 79, 76 and 60%, respectively. However, CME (ED$_{50}$ = 1.820 and ED$_{90}$ = 52.182 mg/ml) extract showed 100% inhibition of egg hatching @ 50 and 100mg/ml while at 25, 12.5, 6.25, 3.125 and 1.56 mg/ml concentration it was 98, 91, 73, 60 and 54%, respectively (Table-2 and 3). In EHA, TBZ showed 100% inhibition in egg hatching at all concentrations (Table-2).

**Larval Paralysis Test**

In LPT, both CAE and CME of *E. citriodora* also showed significant dose dependent anthelmintic activity (Table-2). CAE (ED$_{50}$ = 5.305 and ED$_{90}$ = 157.326mg/ml) at 100 mg/ml concentration showed 100% paralysis of L$_3$ larvae, however, at 50, 25, 12.5, 6.25, 3.125 and 1.56 mg/ml concentration, paralysis of L$_3$ were 98, 78, 71, 50, 36 and 25%, respectively. However, CME (ED$_{50}$ = 5.503 and ED$_{90}$ = 165.447 mg/ml) also showed 100% paralysis of L$_3$ larvae at 100mg/ml concentration. However, it was 89, 83, 79, 52, 30 and 22% at 50, 25, 12.5, 6.25, 3.125 and 1.56 mg/ml concentration respectively (Table-2 and 3). TBZ caused 100% paralysis of third stage larva (L$_3$) of gastrointestinal nematodes in all tested concentrations (Table-2).

**In-vivo anthelmintic efficacy of *E. citriodora***

Coproculture examination revealed the presence of *Haemonchus contortus* (68-85%) as predominant GI nematode followed by *Oesophagostomum columbianum* (8-21%), *Trichostrongylus colubriformis* (5-8%) and *Strongyloides* spp. (2-4%) in animals. CME showed highest (34.11%) reduction in faecal egg count followed by CAE (30.55%) and lowest (7.5%) against CP @ 2gm/kg b.wt. on 7 DPT, however, it was 12.56, -27.08 and -12.57%, respectively on 14 DPT. CME showed highest (35%) reduction in faecal egg count followed by CAE (13.15%) and lowest (6.89%) against CP @ 4gm/kg b.wt. on 7 DPT, however, it was 33.80, 21.05 and 10.34%, respectively on 14 DPT (Table-4). Non-significant difference was found at different doses between CP, CAE and CME.
Phytochemical Analysis of Extracts

On phytochemical analysis of E. citriodora, CAE revealed the presence of alkaloids, flavonoid, reducing sugar, saponins, tannins and triterpenes. However, CME contained alkaloids, coumarin, reducing sugar, saponin, tannins and triterpenes.

### Table 1: Average% corrected mortality of CP, CAE and CME of E. citriodora and TBZ against adult H. contortus

<table>
<thead>
<tr>
<th>Extracts</th>
<th>Average% mortality at different% concentration</th>
<th>Average% Corrected mortality</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.156</td>
<td>0.312</td>
</tr>
<tr>
<td>CP</td>
<td>40</td>
<td>40</td>
</tr>
<tr>
<td>CAE</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>CME</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Average% corrected mortality</td>
<td>46.66</td>
<td>46.66</td>
</tr>
</tbody>
</table>

CP- Crude Powder, CAE- Crude aqueous extract, CME- Crude methanol extract

### Table 2: Percent efficacy of Eucalyptus citriodora extracts and TBZ on eggs hatching inhibition and larvae paralysis of GI nematodes of Goats

<table>
<thead>
<tr>
<th>Concentration (mg/ml)</th>
<th>% inhibition of egg hatching</th>
<th>% larvae paralysis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CAE</td>
<td>CME</td>
</tr>
<tr>
<td>1.56</td>
<td>60</td>
<td>54</td>
</tr>
<tr>
<td>3.125</td>
<td>76</td>
<td>60</td>
</tr>
<tr>
<td>6.25</td>
<td>79</td>
<td>73</td>
</tr>
<tr>
<td>12.5</td>
<td>86</td>
<td>91</td>
</tr>
<tr>
<td>25</td>
<td>92</td>
<td>98</td>
</tr>
<tr>
<td>50</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
</tbody>
</table>

### Table 3: ED₅₀ and ED₉₀ (mg/ml) of crude aqueous and methanolic extracts of E. citriodora in Egg Hatch Assay and Larval Paralysis Test

<table>
<thead>
<tr>
<th>E. citriodora</th>
<th>Tests</th>
<th>ED₅₀ mg/ml</th>
<th>ED₉₀ mg/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Lower limit</td>
<td>Upper limit</td>
</tr>
<tr>
<td>CAE</td>
<td>EHA</td>
<td>0.997</td>
<td>0.315</td>
</tr>
<tr>
<td></td>
<td>LPT</td>
<td>5.305</td>
<td>3.337</td>
</tr>
<tr>
<td>CME</td>
<td>EHA</td>
<td>1.820</td>
<td>0.984</td>
</tr>
<tr>
<td></td>
<td>LPT</td>
<td>5.503</td>
<td>3.818</td>
</tr>
</tbody>
</table>

### Table 4: In-vivo anthelmintic efficacy of Eucalyptus citriodora against mixed infection of gastrointestinal nematodes in goats

<table>
<thead>
<tr>
<th>Groups</th>
<th>GI</th>
<th>GII</th>
<th>GIII</th>
<th>GIV</th>
<th>GV</th>
<th>GVI</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CP @ 2gm</td>
<td>CAE @ 2gm</td>
<td>CME @ 2gm</td>
<td>CP @ 4gm</td>
<td>CAE @ 4gm</td>
<td>CME @ 4gm</td>
</tr>
<tr>
<td>% FECRT</td>
<td>(7DPT)</td>
<td>7.5</td>
<td>30.55</td>
<td>34.11</td>
<td>6.89</td>
<td>13.15</td>
</tr>
<tr>
<td></td>
<td>(14DPT)</td>
<td>-12.57</td>
<td>-27.08</td>
<td>12.36</td>
<td>10.34</td>
<td>21.05</td>
</tr>
</tbody>
</table>

**Discussion**

In the present study of In vitro trial of Eucalyptus citriodora, crude aqueous extract showed maximum (100%) adulticidal activity against H. contortus followed by crude powder (82.86) and minimum (57.14%) with crude methanolic extract (Table-6). Kuamr et al. (2015) also reported adulticidal activity of Eucalyptus globulus against H. contortus. They also observed that methanolic extract had maximum efficacy. In EHA both extracts showed 100% inhibition of egg hatching @50 and 100mg/ml, similar observation has also been reported by Kanojiya et al. (2015). However, Kuamr et al. (2015) also observed dose-dependent inhibition of egg hatching with maximum (100%) against methanolic extract @50mg/ml. In contrast to the present findings, Macedo et al. (2010) and Macedo et al. (2011) reported 99.27% and 98.80% inhibition of egg hatching against Eucalyptus staigeriana and Eucalyptus citriodora essential oil @ 1.35mg/ml and 5.3mg/ml, respectively. The variations in inhibition of egg hatching at lower concentration might be due to the differences in method, type of extraction, collection time, maturity of leaves, soil type and geographical areas in which plant is grown (Peach and Tracey, 1955). It might also be due variation in species of plants which contain different phytochemical constituents responsible for anthelmintic activity.

In Larval Paralysis Test, crude aqueous showed better ED₅₀ (5.305 mg/ml) and ED₉₀ (157.326mg/ml) values than crude methanolic extracts (ED₅₀= 5.503 and ED₉₀= 165.447 mg/ml). However, Kanojiya et al. (2015) observed that methanolic extract revealed better ED₅₀ (6.352 mg/ml) and ED₉₀ (305.48mg/ml) in comparison to aqueous extract (ED₅₀=6.878 and ED₉₀= 878.027mg/ml) of Eucalyptus globulus. It might be due to differences in plant used in the experiment. In the present study, crude aqueous extract showed better dose-dependent larval paralysis. The dose-dependent larvicidal activity of H. contortus has also been reported by Macedo et al. (2010). In in-vivo trial, crude methanolic extract of Eucalyptus citriodora showed better anthelmintic efficacy than crude aqueous extract and crude powder. Anthelmintic activity of Eucalyptus spp. against gastrointestinal nematodes has also been reported by several workers (Mesquita et al., 2013; Macedao et al., 2010 and Kanojiya et al., 2015). In general, higher anthelmintic efficacy of herbal plants have been observed at higher doses by several workers, but in this trial, crude powder and crude aqueous extract of Eucalyptus citriodora showed higher efficacy at low dose (@2gm/kg b.wt.) than higher dose (@4gm/kg b.wt.). It might be due to drug metabolism could have taken longer time with increase in doses and due to early saturation of aqueous extract of the
individual plants during preparation of extracts (Kimani et al., 2014) [17].

The result of coproculture indicated that E. citriodora had lower anthelmintic activity against H. contortus than other parasites. The results are in agreement with the findings of Kanojiya et al. (2015) [16].

The phytochemical results are partially in agreement with the finding of Javed et al. (2012) [15] where they reported alkaloids, flavonoids, tannins, glycosides, saponins, triterpenoids in aqueous and in addition to steroids and phenols in methanolic extract. However, presence of tannins, flavonoids and steroids in crude aqueous and methanolic extracts have been observed by Sharma et al. (2009) [13] while Bhagat et al. (2012) [1] found the presence of anthraquinon, saponins, tannins, flavonoids and cardiac glycosides as phytoconstituents of the methanolic extract.

E. citriodora showed anthelmintic activity against mixed infection of gastrointestinal nematodes of goats might be due to presence of tannins, alkaloids, saponins, reducing sugars, coumarins, flavonoids and triterpenes. Condensed tannins can impair vital process like feeding and reproduction of the parasites or may bind and disrupt the integrity of cuticle of the parasites (Dave et al., 2009; Zafar et al., 2009) [8, 9]. Min and Hart (2003) [28] reported that condensed tannins can improve protein nutrition by binding to plant proteins in the rumen that results into preventing microbial degradation and increasing the flow of amino acids in upper part of intestine. Alkaid plays an important role in the interaction of plants with their environment (Fester, 2010) [9]. Alkaid inhibits essential enzymatic system in parasites and also plays an important role in inhibition of mobility of larvae. In several studies, alkaid isolated from higher plants has been used as anthelmintic agents (Watts et al., 2010) [39]. Havsteen (2002) [11] reported that many plant extracts which showed a high anthelmintic activity revealed the presence of flavonoids along with other phytoconstituents. It is possible, that different developmental stages of helminths might possess different susceptibility to selected flavonoids as observed by Braguine et al. (2012) [4]

Conclusion

Results of present study indicate that E. citriodora showed low to moderate anthelmintic activity and did not reach the required therapeutic dose to controlling gastrointestinal nematodosis. But, it may be included in integrated parasite management to achieve sustainable parasite control in goats.

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