

Appraisal and *in-vitro* study on anthelmintic effect of *Vernonia cinerea* (*Monerakudumbiya*) against larvae of *Haemonchus contortus* and *Toxocara canis*

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Abstract

This study is an attempt to gather and preserve the knowledge on therapeutic effects of *Vernonia cinerea* (Family: Asteraceae; Sinhala name: *Monerakudumbiya*, Sanskrit name: *Sahadevi*) and to observe anthelmintic effect *in-vitro* as it is used in treatment of worm infestation by villagers. Information was gathered from Ayurveda and Sri Lankan traditional medical texts, traditional physicians, journals and web search. Extracted juice of fresh leaves, herbal gruel and decoction prepared from *V. cinerea* are administered orally to treat stomach-ache, diarrhoea, dysentery, haemorrhoids, jaundice, hepatitis, worm infections, tonsillitis, cough, fever, filariasis, malaria, wounds, snake bites, skin diseases, eczema, leprosy, painful urination and colic, urinary calculi, urinary incontinence in children, arthritis, to increase menstrual flow, to stimulate labour and expedite the expulsion of placenta and in veterinary practice. Anti-inflammatory, antipyretic, anti-diuretic, antihyperglycemic, antioxidant, antimetastatic, antitumor, antifungal, bactericidal, nephrocurative, nephroprotective, and hepatoprotective activities of *V. cinerea* are scientifically proven. *V. cinerea* is used in treatment of worm infections in humans. *In vitro* larvae migratory inhibition assay carried out by the authors with larvae of *Toxocara canis* and *Haemonchus contortus* using water extracts of *V. cinerea* has revealed 89.42% and 86.67% inhibition respectively. *Vernonia cinerea* (*Monerakudumbiya*) is a plant with highly diverse medicinal values and is effective in inhibiting larval migration of *Toxocara canis* and *Haemonchus contortus*.

Keywords: *Vernonia cinerea*, *Monerakudumbiya*, *Toxocara canis*, *Haemonchus contortus*,

Introduction

Vernonia cinerea (Family: Asteraceae; Sinhala name: *Monerakudumbiya*; Sanskrit name: *Sahadevi*) is a common weed with valuable medicinal properties. *Sahadevi* (*V. cinerea*) is a well-known plant; the reference regarding this drug could be traced out in Vedas. The word *Sahadevi* is available in the literatures of Vedic period like Atharvaveda, Samhitha and Garuda purana. Atharvaveda praised *Sahadevi* as *Arundhati*, *Visvarupa*, *Subhaga* and *Jivala*¹. Medicinal values of herb *V. cinerea* were known to people since ancient days. *V. cinerea* is used to treat various ailments including worm infections by Sri Lankan Traditional physicians. Larvae (immature worms) of dog round worm (*Toxocara canis*) causes toxocariasis in humans. *Haemonchus contortus* is one of the nematodes that is responsible for anemia and death of infected goats. Infections of *Haemonchus contortus* in humans are very rarely reported but further studies are needed². Aim of this study is to assimilate existing data on medicinal uses and to evaluate the anthelmintic properties against larvae of *Haemonchus contortus* and *Toxocara canis* through *in vitro* studies.

Material and Method

This study consisted of two components, that is; literal study on medicinal uses of *V. cinerea* and *in vitro* studies on anthelmintic properties against larvae of *Toxocara canis* and *Haemonchus contortus*.

(a) Literal study on medicinal uses of *Vernonia cinerea* Medicinal uses of *Vernonia cinerea* (*Monerakudumbiya*) were gathered from Ayurveda and Sri Lankan traditional medical books, interviewing physicians, research journals and internet.

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(b) *In vitro* studies on Anthelmintic properties against larvae of *Toxocara canis* and *Haemonchus contortus*

Preparation of decoction of *Vernonia cinerea* (*Monerakudumbiya*)

Decoctions were prepared according to Ayurveda norms and rules on preparation of drugs (Ayurveda *Paribhasha*). According to Ayurveda *Paribhasha*, 120 g (24 *Kalan*) of fresh materials are mixed with 1920 ml (8 *Patha*) of water and boiled down to one eighth of it, that is; 240 ml (1 *Patha*) to prepare decoctions.

Anthelmintic properties against larvae of *Toxocara canis*

Collection of *Toxocara canis* eggs

Eggs of *Toxocara canis* were obtained from the faeces of young puppies and were embryonated for larval preparations as described by Rajapakse *et al.*, (1992)³. Before collection of samples from the puppies, faeces were screened by direct smear method. The faecal samples were collected from all the positive animals. All positive samples were mixed in one litre of water containing 0.05% Teepol (Lankem Ceylon Ltd., Colombo) in a measuring cylinder and washed five times by sedimentation method to remove all fat and other fine materials.

Thereafter, the sediment was re-suspended in 500ml of saturated salt solution and the suspension was centrifuged (at 1000g for 10 minutes) in order to separate the eggs. The surface layer of the supernatant solution containing the *T. canis* eggs was collected using a Pasture pipette and washed with water through a filter of 100µm pore size in order to remove the coarse fibrous matter. The filtrate was then poured through a filter of pore size 50µm where *T. canis* eggs remained on the filter.

Development of infective eggs of *Toxocara canis*

Freshly harvested eggs were stored in 0.1 N H₂SO₄ at a depth of 0.5 cm in Petri dishes (10 cm x 1.5 cm) in an incubator (Lindberg and May Pvt. Ltd., Australia) at 14.5 °C. At this temperature the development of eggs was arrested without any substantial reduction of their viability. The eggs could be stored in this manner for 60 days. Whenever infective eggs were required, Petri dishes containing the required number of eggs were placed at room temperature (22°C - 24°C). In the course of this second incubation, the

culture was rocked gently once a day to ensure aeration. Eggs reach infective stage within 30-40 days. Thereafter eggs were washed twice by centrifugation at 150g for 15 minutes with 0.15 M Phosphate Buffered saline (PBS) (pH 7.2) to remove H₂SO₄ and the other organic matter and the eggs were recounted at 1:100 dilutions by the McMaster technique. Viability of the *T. canis* embryonated eggs was assessed by the light stimulation method before use as described by O'Lorcain *et al.* (1995).

The storage and maintenance of larval cultures

The storage and maintenance of larval cultures of *Toxocara* larvae to be used for experimental purposes often have to be stored, and this was done satisfactorily in a shallow layer of water. Forty millilitres of a suspension containing not more than 3000 larvae per ml were placed in a tissue culture flask and kept in an incubator maintained at 10 °C. As the storage at low temperatures would induce inhibition of some population, care was taken not to use larvae while they were being conditioned. This means that the usage before 4 weeks of storage (larvae had been stored for 2 weeks to ensure a normal establishment rate) or after 16 weeks, was avoided.

***In vitro* larval migration inhibition assay**

The larvae migration inhibition (LMI) bioassay developed by Wagland *et al.* (1992) and modified by Rabel *et al.* (1994)⁴ was used to determine the effectiveness of the twenty five plant extracts against infective larvae.

Decoction of *Vernonia cinerea* (*Monerakudumbiya*) was diluted by adding Phosphate Buffered Saline (PBS). One millilitre of the solution was taken and diluted with 29 ml of PBS so as to obtain a transparent solution. Then the density was measured in these solutions. As the positive control levamisole 200 µg/ml was used, whereas phosphate buffered saline (PBS) was used as the negative control.

Then 200 µl of larval suspensions were added to wells containing 800 µl of either controls (positive and negative) or plant extract and were incubated at 37°C in the wells of tissue culture plates. Three wells (replicate samples) were run for each concentration of each decoction and for the controls.

All the incubations were carried out in 24 well tissue culture plates overnight (18 hours), at 37 °C and pH 7.2. Following day solutions were transferred to sieves (20 µm mesh at one end) and left for 24 hours at room temperature for active larvae to migrate through the sieves, which were counted later.

On the next day, sieves were removed, Lugol's iodine (0.1 ml) was added to the well and the number of larvae which had migrated was counted under the microscope. The viability and activity of the post migratory larvae with different plant remedies were observed and recorded as follows.

Grade 0 = Dead; No recovery after prolonged immersion in saline

Grade 1 = Inactive but occasional movement can be observed;

Grade 2 = Inactive but intermittent movement can be observed clearly;

Grade 3 = Slow moving;

Grade 4 = Active.

Anthelmintic properties against larvae of *Haemonchus contortus* (in vitro)

Collection of the eggs of *Haemonchus contortus*

Fecal samples were collected directly from the rectum of goats in Kekirawa veterinary range, Sri Lanka. Fecal egg count was determined using the modified McMaster technique (Cringoli, 2011). Faeces of high eggs per gram (EPG) of >5000 from each animal were collected for this study. All positive samples were then subjected to fecal culture for collection of infective larvae.

Fecal culture and isolation of *Haemonchus contortus* larvae

Faecal cultures were prepared using faeces collected from infected goats. The faeces were broken up finely, using a large pestle and mortar, mixed with sterile dung or sawdust in 1:1 ratio, and dampened with distilled water until the mixture was moist and crumbly. Then the mixture was kept in wide-mouthed glass jars or enamel trays and incubated at room temperature for 10-14 days. The cultures were maintained by aerating the lower layers every day and, to prevent drying, by adding a few drops of water in order to maintain moisture. After 14 days, cultures were baermannized using wide-mouthed glass jars. The larvae were counted and assessed for viability

and identification was carried out to the level of genus before being stored at 10 °C.

The storage and maintenance of larval cultures

The storage and maintenance of larval cultures were carried as described under *Toxocara canis*.

In vitro larval migration inhibition assay

In vitro larval migration inhibition assay was carried out as described under *Toxocara canis* but infective larvae in unsheathed forms were used. The *Haemonchus* infective larvae that were subjected to test were in unsheathed forms. Sheathes were removed by incubating the larvae in sodium hypochlorite solution (0.025% available chlorine) for 10 minutes at room temperature, washing several times and concentrating to approximately 2500 larvae/ml PBS.

Results

Review on *Vernonia cinerea* (*Monerakudumbiya*)

Scientific classification of *Vernonia cinerea* (*Monerakudumbiya*)

Kingdom:	Plantae
Subkingdom:	Tracheobionta
Division:	Magnoliophyta
Class:	Magnoliopsida
Subclass:	Asteridae
Order:	Asterales
Family:	Compositae
Genus:	<i>Vernonia</i>
Species:	<i>cinerea</i>

Synonyms of *Vernonia cinerea* (*Monerakudumbiya*)

Sinhala name:	<i>Monerakudumbiya</i>
Sanskrit name:	<i>Sahadevi, Uttamkanyaka, Dandopalaa.</i>
English name:	Purple Fleabane, Ash coloured Fleabane
Tamil name:	<i>Naichotte Poonde, Seedeviyar shenkaluneer</i>
Botanical name:	<i>Vernonia cinerea, Cyanthillium cinereum</i> (L.) H. Rob.

Description of *Vernonia cinerea* (*Monerakudumbiya*)

An annual herb with slightly branched, stiff, erect, cylindrical, more or less pubescent stem 15-60 cm tall; Leaves simple, alternate, distant, the lower ones 4-5 cm long, 3-3.5 cm broad, gradually becoming smaller upwards. Broadly oval to linear-lanceolate, tapering to the base, sub-obtuse, apiculate, coarsely and shallowly crenate-serrate, more or less hairy on both sides, Petioles 0.6-1.8 cm long; Flowers regular, bisexual, pinkish violet, all tubular, sessile on long, stalked, small heads in divaricate, terminal corymbs, involucre-bracts linear to oblong. 1.5-2.5 mm long, mucronate, silky outside, flowers 20-25 in a head; sepals reduced into long bristles with a shorter outer row; petals 5, fused into a long, tubular corolla about 4 mm long, segments deep and narrow; stamens 5, on the corolla tube, anther not tailed at the base; ovary 1mm long; hairy, inferior, 2-carpellary, unilocular with a single basal ovule, style stout, 3.5 mm long, stigma bilobed; fruit a hairy achene, 1.5-2 mm long but not ribbed, with a white pappus the outer row of which is short⁵ (Figure 1 and 2).



Figure 1: Plant of *Vernonia cinerea*



Figure 2: Flowers of *Vernonia cinerea*

Distribution of *Vernonia cinerea* (*Monerakudumbiya*)

Occurs throughout Sri Lanka, India, tropical Asia, Africa and Australia (Figure 3). In Sri Lanka, it is a very common weed everywhere⁵. It can be seen in roadside, open waste places, dry grassy sites and in perennial crops during plantation.

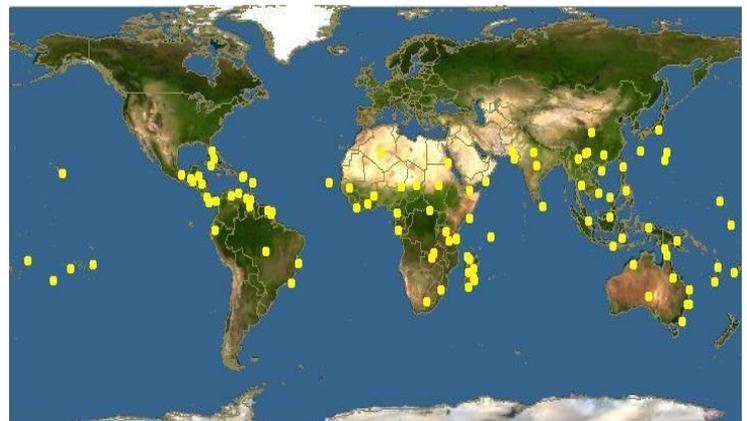


Figure 3: Distribution of *Vernonia cinerea*

Ayurveda pharmacodynamic properties of *Vernonia cinerea* (*Monerakudumbiya*)⁶

Rasa: Tikta

Guna: Laghu, Ruksha

Veerya: Ushna

Vipaka: Katu

Dosha Karma: Kapha Vata Shamaka

Part use in Medicine: Leaves, flowers, seeds, root, entire plant

Medicinal uses of *Vernonia cinerea* (*Monerakudumbiya*)

Vernonia cinerea (*Monerakudumbiya*) has been used externally and internally to treat a number of disorders, it is used singly or in combination with various medicaments. Some selected formulae prepared with *V. cinerea* are given below.

- i. Juice is extracted from fresh leaves and 10-20 ml is given to treat dysentery, colic and piles⁵.
- ii. Decoction is prepared with entire fresh plant and 120 ml of the decoction is given to treat cough, bronchitis and bronchial asthma⁵
- iii. A paste prepared with leaves is applied to reduce pain and swelling (*Shota*)⁶.
- iv. Paste prepared from leaves is applied to the eye in conjunctivitis⁶.
- v. Paste prepared from leaves is applied in disorders in nervous system⁶.
- vi. The crushed leaves are applied externally on wounds and sores⁵.
- vii. In fever, a root is tied on the head and juice is applied on the body⁶.
- viii. Chronic fever and periodically recurrent fever are managed with decoction prepared with entire plant of *Vernonia cinerea*⁶.
- ix. A paste prepared with leaves is applied for Ringworm⁷
- x. 120ml of decoction prepared from entire plant is given twice a day in treatment of diarrhoea, stomach-ache and intestinal colic⁸.
- xi. To treat urinary incontinence in children, 10-20 ml of fresh juice of entire plant is given⁸.
- xii. In Dysuria and renal calculi, 120 ml of decoction of entire plant is given⁸.
- xiii. 120ml of decoction of entire plant is given twice a day in treatment of psoriasis, vitiligo and other ailments in skin⁸.
- xiv. Worm infections (round worm and thread worm) are treated by giving 120 ml of decoction prepared with fresh entire plant⁸.
- xv. Seeds are ground into a paste, mixed with lime juice and applied to destroy Pediculi⁸.
- xvi. In snake bites, 4 gm of roots of *V. cinerea* is boiled in water, strained through a piece of cloth and taken orally 3-4 times a day⁹.
- xvii. In gynecological disorders, *V. cinerea* is used orally to treat leucorrhoea and control excessive menstruation¹⁰
- xviii. Roots and leaves of *V. cinerea* are chewed raw or entire plant is boiled in water and

drunk to cure sexual impotency and erectile dysfunction¹¹.

- xix. Herbal gruel prepared with entire plant of *Vernonia cinerea* is administered in jaundice and hepatitis, as a home remedy in Sri Lanka.
- xx. Fifteen grams each of entire plant of *Vernonia cinerea* and *Phyllanthus debilis*, stem of *Tinospora cordifolia* and dried fruits of *Phyllanthus emblica* are added to 1920 ml of water and boiled down to 240ml. 120ml of this decoction is given twice a day to treat epistaxis¹².

Veterinary uses

Seeds are given to the animals to treat food poisoning. Infusion of seeds is given to the livestock animals to cure fever¹³.

Bark of babul, seeds of *Trachyspermum ammi* and *V. cinerea* mixed with jaggery is given to the livestock. Bark of babul, seeds of *Trachyspermum ammi* and *V. cinerea* mixed with jaggery is given to the livestock wise a day for one month as a tonic to cure overall weakness¹³.

Some animals such as wild chimpanzees are believed to ingest *Vernonia cinerea* when suffering from cancer.

Chemical constituents

Preliminary phytochemical screening revealed the presence of flavonoids, glycosides, tannins, and carbohydrates in *Vernonia cinerea*.¹⁴ It also contains flavonoids, saponins, alkaloids, and terpenoids.¹⁵ *V. cinerea* contains vernolide-A and vernolide-B (two novel sesquiterpene lactones); β -amyryn, lupeol and their acetates; and β -sitosterol, stigmasterol, α -sp inasterol and phenolic resin in the whole plant. The roots contain δ -amyryn acetate, α -amyryn acetate, β -amyryn acetate, β -amyryn and α -amyryn. In addition, the leaves contain urticifolene (new polyene), lutein (carotenoid) and sitosterol (triterpenoid). The stem, bark and leaves contain lupeol, 12-oleanen-3-ol-3 β -acetate and stigmasterol.¹⁶

Research

- i. Reddy *et al.*, (2012) evaluated the anticataleptic efficacy of ethanol extract of *Vernonia cinerea* L. in haloperidol induced catalepsy in rats. Scientific evaluation of this claim using experimental model Anticataleptic activity using block method, Locomotor activity in actophotometer and

- Exploratory behavior in hole board apparatus. From the observations of above studies, it could be envisaged, that the protective effect of ethanol extract of *Vernonia cinerea* L. against symptoms of Parkinson's disease (catalepsy) may be due to regulation in neurotransmitters such as dopamine, serotonin, glutamate which are playing an important role in protection of catalepsy and antioxidant properties¹⁷.
- ii. Ganesh *et al.*, (2011) further confirmed the antidiarrhoeal activity of methanolic extract of *Vernonia cinerea* L., (Less) and reported dose dependent¹⁸. For evaluation of antidiarrhoeal efficacy of methanolic extract of the plant, rats were used as test animal. The time of onset of first wet faeces increased significantly and dose dependently by the extract. It was excellent at higher doses (100 and 200 mg/kg body wt., orally).
 - iii. Ngbolua *et al.*, (2011) detected moderate antiplasmodial activities in *V. cinerea* subsp *vialis*; a plant species not previously reported as antimalarial in the traditional medicinal knowledge of Madagascar¹⁹.
 - iv. Latha *et al.*, (1998), tested anti-inflammatory effect of an alcoholic extract from the flower of *Vernonia cinerea* (Asteraceae) in adjuvant arthritic rats. Changes in paw volume, body and tissue weights and, serum and tissue enzyme activities of ALT, AST, ACP and cathepsin-D in adjuvant rats were reversed by oral administration of 100 mg of the flower extract per kg of body weight (BW). The extract also reversed the major histopathological changes in the hind paws of the arthritic rats²⁰.
 - v. Bashar *et al.*, (2014) reported antipyretic, analgesic and anti-inflammatory activities of the methanol extract of whole plant of *V. cinerea* Less. Antipyretic activity was assessed by the yeast-induced hyperthermia in mice. The analgesic property was evaluated by formalin-induced writhing test. Acetyl salicylic acid (ASA) was used as standard in in-vitro anti-inflammatory activity test²¹.
 - vi. Ushasri *et al.*, (2013) evaluated the alcoholic, ethereal and chloroform extracts were obtained from the roots of plant *Vernonia cinerea* by soxhlet extraction or continuous hot percolation methods for their respective anthelmintic activity, against locally available earth worms (*Pheretima posthuma*). Three concentrations (10, 30, 60 mg/ml) were prepared from each extract and used for the study over earth worms. The study involves the determination of time of paralysis and time for death of the earth worms tested. The results obtained from the study revealed the fact that chloroform and alcoholic extracts from the roots of *Vernonia cinerea* possess significant anthelmintic effect²².
 - vii. Toyang and Verpoorte (2013) found that *Vernonia cinerea* has potential against cancer and inflammatory conditions according to reported literature. Vernolide A is so far the most promising single agent from a *Vernonia* species that has potential for development into an anticancer agent²³.
 - viii. Dakshini *et al.*, (1992) found that the concentrations of heavy metals i.e. cobalt, copper, nickel, manganese and zinc in dried material of *Vernonia cinerea* are much higher than in the soil samples. Accumulation of these metals is greater in pink than in purple or mauve flowered forms²⁴.
 - ix. Asha and Abraham (2015) had evaluated the efficacy of methanolic extract of *Vernonia cinerea* (MEVC) in selenite induced cataract using Sprague Dawley rats. MEVC was administered as orally from 8th day up to 21st day at the concentration 5 µg/g body weight. The findings, suggest that *V. cinerea* has the therapeutic potential of lens against selenite induced cataract. It is possible that *V. cinerea* might be useful against lens damage caused by ROS generation under oxidative stress. It is also relatively nontoxic when given in small doses. Hence, these findings are considered pharmacologically significant; evaluation of active component from *V. cinerea* will certainly uncover novel therapeutic possibilities²⁵.

- x. Muir (1981) reported that the aqueous extract of *Vernonia cinerea* contains a depressant agent whose primary effect is that of analgesia. In Malaysia, *Vernonia cinerea* is included in several traditional herbal preparations used for insomnia and related ailments. Most preparations of *Vernonia cinerea* are concoctions where the plant is boiled in water. Any active ingredient is therefore presumably water soluble and heat stable. Though these results are preliminary in nature, they may suggest that the use of aqueous extracts of *Vernonia cinerea* for its sedative effect may entail hitherto unknown dangers since effective sedative actions in mice appear to occur only at relatively high doses but that the plant may contain an agent which might be of use at relatively lower (and therefore safer) doses for the control of pain²⁶.
- xi. Dhanalakshmi *et al.*, (2013) reported that ethyl acetate extract of *Vernonia cinerea* exhibited excellent antidandruff activity against *Pityrosporum ovale* and *Pityrosporum folliculitis*. The antifungal activity of *V. cinerea* leaf extracts showed positive results against all the tested fungal pathogens; *C. albicans*, *C. parapsilosis* and *C. tropicalis*²⁷.
- xii. Leelarungrayub *et al.*, (2010) carried out a study to evaluate the effects of *Vernonia cinerea* Less. (VC) supplementation and exercise on oxidative stress biomarkers, beta-endorphin release, and the rate of cigarette smoking. 20gm of dried entire plant of *V. cinerea* mixed with 390ml of water and boiled in an earthen pot until water evaporated down to 130ml. Condensed VC juice was then preserved in a clean bottle and was provided to subjects to drink prior to smoking each, three days per week for two months. Supplementation with *V. cinerea* Less and exercise provided benefit related to reduced smoking rate, which may be related to oxidative stress and beta-endorphine levels²⁸.
- xiii. Sreedevi (2011) studied nephroprotective activity of *V. cinerea*. The alcoholic extract of *Vernonia cinerea* showed promising nephrocurative activity, whereas ethyl acetate extract of *Vernonia cinerea* possessed significant nephroprotective activity in the rat model of cisplatin induced renal toxicity. These results suggest the therapeutic utility of herbal *Vernonia cinerea* extracts in renal injury²⁹.
- xiv. An *in vivo* study showed that *V. cinerea* had an equivalent antipyretic effect to paracetamol when the extract was taken at a dose of 500mg/kg in rats (Gupta, 2003)³⁰.

Physio-chemical composition of *Vernonia cinerea* (*Monerakudumbiya*)

Madanayaka *et al.*, (2015) studied physio-chemical composition of root of *Vernonia cinerea* in 100gm. It contains 68.8 gm moisture. Ash value is 2.81g. The researchers reported that it also contains crude protein (2.36gm), crude fibre (13.73 gm), crude fat (0.81 gm) and water soluble sugars (38.5gm)³¹.

Anthelmintic properties against larvae of *Toxocara canis* and *Haemonchus contortus* (*in vitro*)

As shown in the table 1, decoction of *V. cinerea* was 89.4 % effective in inhibiting *Toxocara* larval migration and 86.7% effective in inhibiting *Haemonchus* larval migration. Whereas larval migration inhibition of *Toxocara canis* and *Haemonchus contortus* with Levamisole were 99.7% and 96.6% respectively.

The viability of post-migratory larvae of *Toxocara canis* and *Haemonchus contortus* with controls and with decoction of *V. cinerea* are presented in Table 2. Maximum number of migrated larvae of *Toxocara* and *Haemonchus* was observed in the negative control PBS. Least number of migrated larvae was observed in the positive control Levamisole and all the larvae were dead after migration. 6.7% of *Toxocara* and 4.4% of *Haemonchus* larvae were migrated in decoction of *V. cinerea*. All the migrated *Toxocara* larvae were dead or in Grade 1, 2, 3 or 4 and in *Haemonchus* larvae were dead or in Grade 1.

Table 1: Percentages of *in vitro* larval migratory inhibition of *Toxocara canis* and *Haemonchus contortus* infective larvae with decoction of *Vernonia cinerea*

Treatment	Percentage (%) of larval migration inhibition (LMI)	
	<i>Toxocara canis</i>	<i>Haemonchus contortus</i>
Levamisole 200 µg / ml in PBS (Positive Control)	99.7	96.6
Phosphate buffered saline (Negative control)	0	0
<i>Vernonia cinerea</i> (<i>Monerakudumbiya</i>)	89.4	86.7

Table 2: Viability of post-migratory larvae of *Toxocara canis* and *Haemonchus contortus* larvae with decoction of *Vernonia cinerea*

Treatment	Percentage (%) of viability of <i>Toxocara canis</i> larvae						Percentage (%) of viability of <i>Haemonchus contortus</i> larvae					
	Grade 0	Grade 1	Grade 2	Grade 3	Grade 4	Total	Grade 0	Grade 1	Grade 2	Grade 3	Grade 4	Total
Levamisole (Positive control)	55	0	0	0	0	55	0.1	0	0	0	0	0.1
Phosphate buffered saline (Negative control)	0	0	0	0.2	61.5	61.7	0	0	0	0	72.5	72.5
<i>Vernonia cinerea</i> (<i>Monerakudumbiya</i>)	0.2	0.1	0.9	1.3	4.2	6.7	2.2	2.2	0	0	0	4.4

Grade 0 = Dead; No recovery after prolonged immersion in saline; Grade 1 = Inactive but occasional movement can be observed; Grade 2 = Inactive but intermittent movement can be observed clearly; Grade 3 = Slow moving; Grade 4 = Active

Discussion

Vernonia cinerea (*Monerakudumbiya*) is used to treat oedema, fever, jaundice, skin diseases, epistaxis, urinary incontinence, dysuria, renal calculi and Parkinsonism. Antipyretic, anti-inflammatory, analgesic, antidiarrhoeal, anti-cancer, antitumor, antidandruff, antiplasmodial, anticataleptic, nephroprotective activities and anthelmintic activity against earth worms (*Pheretima posthuma*) are scientifically proven.

Decoction of *V. cinerea* was 89.4 % effective in inhibiting *Toxocara* larval migration and 86.7% effective in inhibiting *Haemonchus* larval migration. According to Ayurveda *V. cinerea* possesses *Tikta Rasa*, *Laghu* and *Ruksha Guna*, *Ushna Veerya* and *Katu Vipaka*. These properties lead to reduction of

Kapha Dosha. Ayurveda describes three methods to treat *Krimi Roga* (worm infection)⁸. One of them is *Prakruti Vighata*, that is making the environment unfriendly for worms. *V. cinerea* makes the surrounding unfriendly to worms by reducing *Kapha Dosha* in the environment. Further, Ayurveda describes *Krimighna* (wormicide) property of *V. cinerea* which leads to killing of the worms. Therefore, *V. cinerea* is effective in inhibiting *Toxocara* larval migration and *Haemonchus* larval migration. Iqbal et al., (2007) reported that tannins has anthelmintic effect³². β -sitosterol possess *in vitro* anthelmintic properties against sheep GIS.³³ Alkaloids, Phenols and tannins are accountable for anthelmintic activity.³⁴ As *Vernonia cinerea* contain these phytochemicals, it possesses anthelmintic properties and it inhibits *Toxocara* larval migration and *Haemonchus* larval migration.

Conclusion

It is concluded that *Vernonia cinerea* (*Monerakudumbiya*) is effective in inhibiting larval migration of *Toxocara canis* and *Haemonchus contortus* and also has multi-faceted medicinal values.

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