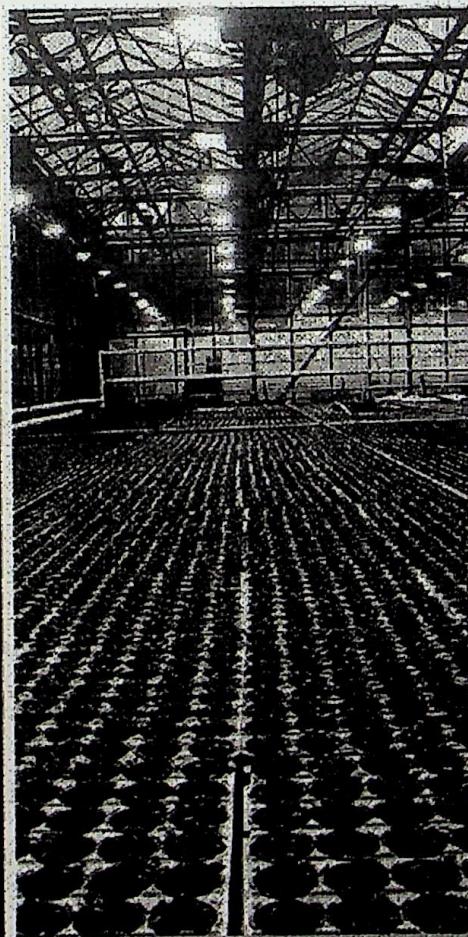




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**Anthelmintic effect of bio-rationals against the root knot nematode,
Meloidogyne incognita colonizing chilli rhizosphere**

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1. Introduction

Chilli is an important cash crop of Sri Lanka (*Capsicum annum*). A variety of biotic-abiotic stresses impair the chilli production particularly in high potential chilli producing areas in dry and intermediate zones of Sri Lanka. Multiple cropping and continuous cultivation of the same crop on the same plot of land increases the problem of soil-borne diseases, specially plant parasitic nematodes (Dhillon et al., 2019). Root-knot nematode causes damage on a variety of crops, including chillies, tomatoes, and potatoes. The Root-knot nematode (*Meloidogyne* spp.) belongs to the family Meloidogynidae is the primary nematode species infesting chili plants. They pierce the root and enter the vascular system, where they cause metabolic changes that contribute to gall formation (Thiyagarajan, 2014).

The root-knot nematodes manage by using various recommended tactics and management strategies. But, synthetic nematicides are preferred by farmers due to their quick knock-down effect. The use of synthetic nematicides is being limited due to the booming number of cancer and chronic kidney disease of unknown etiology (CKDU). Recently, Sri Lankan policy banned the importation of all forms of synthetic agrochemicals. As a result, scientists have been looking for long-term solutions to nematode problems. Many plants are known to produce secondary compounds to safeguard them from harmful pathogens through physiological pathways. Moreover, many higher plants contain nematicidal compounds that can kill, inhibit nematode hatching, or impair nematodes motility (Amora et al., 2017). This study was aimed to evaluate the anthelmintic effect of selected bio-rationals against *M. incognita* attacking chilli.

2. Materials and Methods

Isolation of female of *Meloidogyne incognita*

Root-knot infected root samples were collected from chilli fields in Jaffna District and brought to Nematology Laboratory at the Department of Agricultural Biology, Ariviyal Nagar, Kilinochchi and were washed and disinfected with 2% aqueous solution of NaOCl. Root galls were cut into small pieces and the upper layer of roots was peeled carefully. Pear-shaped females were picked out with the camel hair brush without damaging.

Raising of chilli seedlings

The nursery bed was prepared in sterilized soil to get KA2 chilli seedlings for studies and all the agronomic practices were followed as the Department of Agriculture recommendation. Sieved sterilized (sterilized through Tyndallization process) soil was filled at the ratio of compost: top soil, 1:2 in 6 kg capacity pots made using black polybags. Two weeks old KA2 chilli seedlings were transplanted as two per pots.

Preparation and Application of treatments

Anthelmintic plants leave namely *Cassia alata* (Candle Bush), *Vernonia anthelmintica* (Ironweed), *Aristolochia bracteolata* (Worm killer), *Moringa oleifera* (Drumstick tree) were collected from herbal garden maintained at the same department and ground thoroughly using an electric grinder and water extract was prepared. Extracts were vacuumed in a rotary

evaporator at 50 °C- 55 °C. 40 ml of each extract and 200 ppm solution of fluopyram (400 g/l SC) were used as for root dipping of chilli seedlings (15-20 min) before transplanting. 20 Pear-shaped females of *M. incognita* were inoculated three days after transplanting. Soil application treatments (40 ml) were repeated at two weeks' intervals also.

Experimental design, data collection and Statistical analysis

This experiment was carried out under the experimental design of CRD in the green house with six treatments and four replicates. Plant height, number of galls according to scaled (Sasser et al., 1984) where 0=No galls; 1=1-2 galls; 2=3-10 galls; 3=11-30 galls; 4=31-100 galls and 5=>100 galls, length and diameter of galls were measured after uprooting of seedlings. The statistical analysis was performed by using ANOVA, and Tukey's HSD multiple comparison test was administrated to identify the best treatment at $P < 0.05$ using SAS 9.1.

3. Results and Discussion

Influence of anthelmintic plant extract on plant growth

Initially all the chilli plants were selected in equal height (Fig. 1). Four weeks after planting, significantly different plant height of 12.2 cm was observed in *C. alata* treated trail but in control where plant height was 9.8 cm at $P < 0.05$. Plant height was insignificant in other treatment combinations.

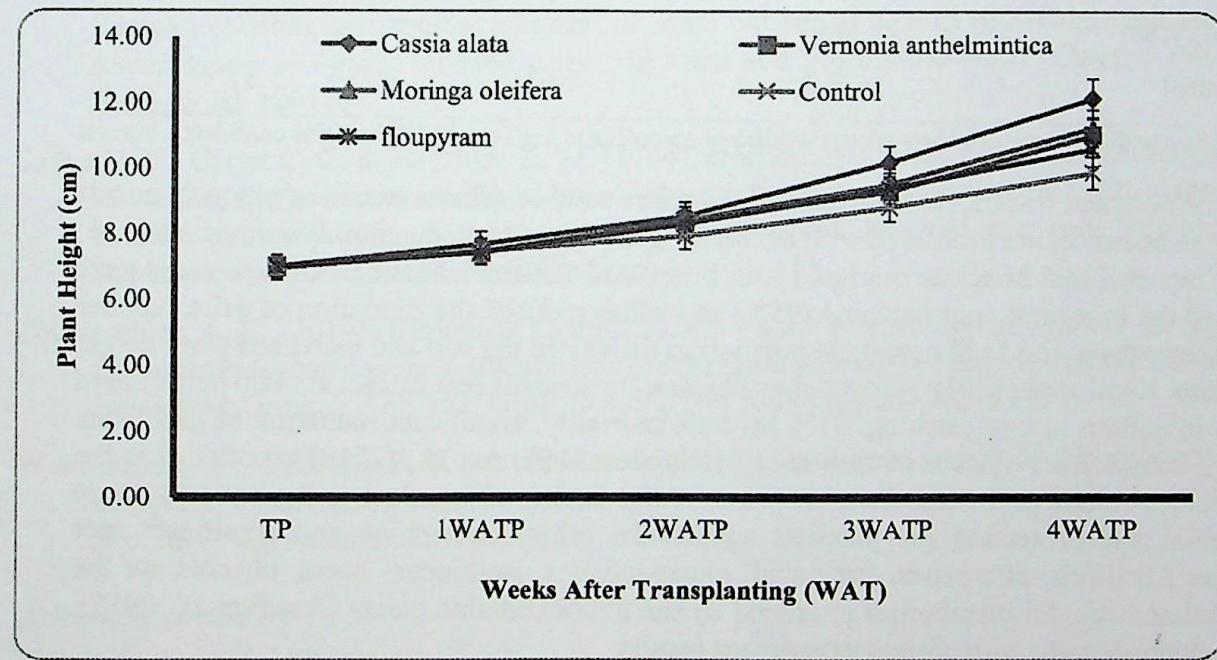


Figure 1. Plant height of KA2 chilli in different treatments.

Efficacy of anthelmintic plant extract on root-knot nematode

The results show that the mean number of galls produced, length of the gall and the diameter of the galls were significantly different among the treatments (Table 1). The lowest gall number of 4.75, gall length of 1.19 mm and gall diameter of 0.57 mm were observed in *C. alata* treated trail. The number of galls counted in *C. alata* extract treated trail was significantly different from all other treatments except nematicide Floupyram applied trail. Number of galls comparatively less in *V. anthelmintica* and *M. oleifera* treated plants with the root-knot index value of 3 than control where the nematode index value was 4, but they were not significant than control treatment.

Mean length of gall is significantly lower in all the treatments than control. The lowest gall length of 1.19 mm was recorded in *C. alata* treated trail whereas second lowest mean gall length

of 1.33 mm was recorded in *V. anthelmintica* which was insignificant when compared with *C. alata* at $P < 0.05$.

Mean diameter of gall was less than 1mm in all the treatments except control. Diameters of galls were not significantly on par among them except in control where the highest diameter of gall (1.37 mm) was recorded. The gall diameter in *V. anthelmintica*, *M. oleifera* and floupyram treatments were 0.96 mm, 0.90 mm and 0.75 mm, respectively.

All the chilli plants treated with the *A. bracteolata* were dead few days after treatment. That extract may contain some toxic components; it may have negatively affected the plant's survival.

Table 01. Effect of different treatments on Root-knot nematode

Treatment	Number of galls	Length of gall	Diameter of gall
		(mm)	(mm)
<i>Cassia alata</i>	4.75 ^b	1.19 ^c	0.57 ^b
<i>Vernonia anthelmintica</i>	23.50 ^a	1.33b ^c	0.96 ^b
<i>Moringa oleifera</i>	24.00 ^a	1.45 ^b	0.90 ^b
Floupyram	6.00 ^b	1.45 ^b	0.75 ^b
Control	33.25 ^a	1.63 ^a	1.37 ^a

Mean values with the same alphabets are not significant according to the Tukey's HSD at 95% confidence interval

From the obtained results, it was clear that the water soluble nemato-toxins or phytochemicals present in botanical are interfered with nematode growth and reproduction. Wondimeneh et al., (2013) reported that Mexican marigold leaf, bitter leaf, lantana leaf and baker tree seeds were inhibited the root-knot eggs hatching (95%) as well as reduced the formation of galls, number of eggs/egg-mass and final nematode population density in the soil and increased plant height of tomato. Khan et al., (2019) proved that *Phyllanthus amarus* leaf extract at 5000 ppm caused 86.5% inhibition in egg catching, 91% juvenile mortality, significant reduction of gall index and egg masses/root system of root-knot nematodes. Dahlin et al., (2019) reported that the nematicide Velum protected the plants from *M. incognita* by reducing the soil nematode population and protecting the plantlets against the initial penetration and significant root damage. Alkaloids, diterpenes, fatty acid, glucosinolates, isothiocyanates, phenols are the potential nematicidal metabolites produced by the antihelmenthic plants (Asadi et al., 2015). These evidence tally with current experiment results.

4. Conclusions

The study revealed the potential of anthelmintic plant extracts in the management of root-knot nematode. Among the plant extracts, *C. alata* reduced the infestation, gall-forming ability, number of galls (4.75), length (1.19 cm) and diameter of gall (0.57 mm), even superior to the nematicide. Therefore *C. alata* extract could be used for the eco-friendly management of root-knot nematode in the chilli. The active ingredients present in the *C. alata* plant parts can be a potential source of new organic nematicidal compounds. However, further investigations are needed for recommendation.

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