Nematicidal Potential of Some Indigenous Plants Against Meloidogyne incognita on Vigna radiata

Hari Raghu Kumar and Irshad Mahmood

Research Journal of Agricultural Sciences
An International Journal

P- ISSN: 0976-1675
E- ISSN: 2249-4538

Volume: 12
Issue: 06

Res. Jr. of Agril. Sci. (2021) 12: 2218–2224

CARAS

Res. Jr. of Agril. Sci. (Nov-Dec 2021) 12(6): 2218–2224
www.rjas.org

Full Length Research Article

Nematicidal Potential of Some Indigenous Plants Against *Meloidogyne incognita* on *Vigna radiata*

Hari Raghu Kumar*[1] and Irshad Mahmood[2]

Received: 10 Sep 2021 | Revised accepted: 18 Nov 2021 | Published online: 13 Dec 2021

ABSTRACT

In the field of agribusiness, the effect of nematodes is exceptionally critical due to its activity upon the yields. A harvest yield misfortune due the small minute nuisances *Meloidogyne incognita* in the different nations is gigantic. They caused yield loss of 157 billion dollars around the world, out of which 40.30 million dollars is accounted for in India. In India, pulse loss is 20-35 percent (8.06-14.12 million dollars) of total loss yearly. The main objectives of this study were to assess the nematicidal potential of different plant extracts and their manure *in vitro* and greenhouse conditions respectively. In this study, the nematicidal efficacy of aqueous extracts and dry manure of five plants, *Azadiracthta indica, Jatropha curcas, Lawsonia inermis, Polyalthia longifolia,* and *Vachellia nilotica*, on J2s of *M. incognita in vitro* and on growth and yield parameters of *Vigna radiata* in greenhouse condition respectively, were assessed. *In vitro* nematicidal efficacy at five doses (0.01%, 0.02%, 0.03%, 0.04% and 0.05%) for mortality at different exposures of times period 24, 48, 72, 96 and 120 hours and for hatching inhibition at two different intervals 168 and 240 hours. Dry manure from leaves of botanicals was prepared for greenhouse experiment and assessed. The in vitro results showed, out of all five plant's extracts and control (distilled water), *A. indica* extract (0.05%) was significantly more effective with 81.80% mortality than the other ones, for all five different exposures viz. 24, 48, 72, 96 and 120 hours. Similarly, the rate of egg hatching inhibition (HI) increases gradually in both intervals 168 and 240 hours. Maximum HI found in 0.05% concentration of *A. indica* with 83.92% and 90.35% on 168 and 240 hours respectively. In the greenhouse experiment, treatment with the 100 gm of dry manure of *A. indica* gave positive implications on growth and yield and physiological parameters of *V. radiata* while negative on pathological parameters of *M. incognita,* which was followed by the *V. nilotica*, *L. inermis*, *J. curcas* and *P. longifolia*. All the plant extracts as well as dry manure were found to be effective against root-knot nematodes in this investigation. In light of their likely job upon the *M. incognita* can be a future substitution of inorganic pesticides.

***Key words*:** *Meloidogyne incognita*, Greenhouse, Juvenile, Manure, Plant extracts, Hatching, Mortality

Plant parasitic nematodes are one of the main biotic constraints in reducing quantitative and qualitative characters of pulse crops including green gram [1]. Among plant parasitic nematodes, root knot forming nematode are considered as notorious pathogen across the world including Indian environment which may cause 20-35% of yield loss [2]. Green gram is assumed to be vulnerable to root-knot nematode and has been accounted with poor yielding ability in infected field. Considerable reduction in plant growth, nodulation, nitrogen and overall yield has been found in *Meloidogyne incognita* infected field [3].

Ecofriendly management of root-knot nematode has been a challenging task for the researchers. To avoid environmental perturbations due to non-judicious applications of pesticides, botanicals are assumed to be very viable, cheap and ecofriendly [4]. Continuous exploitation of wide range of botanicals are find out economically viable option in root-knot management. Application of natural resources or botanical pesticides has shown promising results in various disease management practices. Biopesticides of botanical origin are being tested and taken into consideration for managing plant nematodes. Extracts of botanicals are the emerging facet in the management of crop diseases. Botanical extracts have been found effective against *Meloidogyne* species in a number of prior investigations [5-7]. For example, *Azadirectha indica* contains nematicidal potential [8-9] and *Lawsonia inermis* also [10]. The aim of present study was to evaluate the nematicidal potential of some common botanicals against *M. incognita* under laboratory and greenhouse condition. Potentiality of these botanicals was also observed in plant growth and yield improvement, reduced galling and nematode populations.

* **Hari Raghu Kumar**
✉ raghukumaramu@gmail.com
[1-2] Lab No. 52, Plant Pathology Section, Department of Botany, Aligarh Muslim University, Aligarh - 202 002, Uttar Pradesh, India

MATERIALS AND METHODS

Collection and maintenance of inoculums

Brinjal roots infected with *M. incognita* were collected and brought to the laboratory for isolation of egg masses in order to maintain the inoculation of root-knot nematode. The egg masses were picked up gently from infected plant roots and stored in Petri plates containing sterile water. Petri plates were exposed with 25±2°C. Newly hatched juveniles were collected in a Petri dish and their population was counted in a counting dish under stereo zoom microscope.

Preparation of plant extracts and manure

Five botanicals (*Azadiracthta indica, Jatropha curcas, Lawsonia inermis, Jatropha curcas Polyalthia longifolia*, and *Vachellia nilotica*) from five distinct botanical families were taken at their vegetative stage from various localities of AMU campus. Plant leaves were wrapped in papers and dried in a laboratory incubator at 58°C for 24 hours before grinding into fine powders with a commercial grinder.

Aqueous extracts were prepared by dissolving 1.0, 2.0, 3.0, 4.0, and 5.0 g of powder in 1000 ml glass flasks containing 100 ml of solvent (DW). The flasks were shaken for 4 hours at 500 rpm using an orbital shaker. To eliminate debris, the mixture was filtered via Whatsmann No.1 filter paper and centrifuged for 15 minutes at 1500 rpm. Final volume was maintained 1000 ml by adding DW. Such solution was used as a 'stock solution' and required percentage of extract solutions were made from this stock solution [11]. Likewise, for preparing the manures, different botanicals were properly chopped and kept in a autoclaved pot. This pot was regularly watered up to watering capacity. Ripened dry compost was collected after three months and nutrient values were analyzed [12].

Biological assays

In vitro evaluation of hatching and mortality

The effect of aqueous extracts at various concentrations was used to examine the mortality of nematodes juveniles as per procedure described by Zaidat *et al*. [13]. Using probit analysis, the lethal concentrations (LC_{50} and LC_{90}) required to kill 50% and 90% respectively were estimated [14]. Similarly, hatching inhibition was observed using the following formula:

$$R\ (HI) = [Nie\text{-}Neh/Nie] \times 100$$

Where Nie denotes the number of eggs lay at the start, and Neh denotes the number of eggs that have hatched [11, 13].

Pot experiment

A pot experiment was carried out to know the growth promoting potential of *A. indica, J. curcas, L. inermis, P. longifolia*, and *V. nilotica* on *V. radiata* 100g botanicals (composted/ manure) was manually applied in the pot before sowing of seeds. After 15 days of germination, 2000 freshly hatched J2 was inoculated around the roots of *V. radiata.*

Data collection and observations

Growth and yield observations

Growth parameters (plant length, fresh and dry weight of plant shoot and root, pods per plant and weight of 100 seeds) were recorded upon termination of experiment. The harvested plants were kept in a laboratory for analyzing different morphological, physiological and biochemical parameters.

Physiological observations

The nitrate reductase activity (NRA) of fresh leaves was determined according to Jaworski [15]. The leaf chlorophyll was estimated by using the procedure of Arnon [16]. Similarly, carotenoid was estimated by using the protocol described by Kirk and Allen [17]. Moreover, protein estimation was done by Lowry *et al*. [18].

Nematological observations

The number of galls per root system, egg masses per root system, and eggs per egg mass were all carefully counted by manual process, Rf was also calculated. The final nematode population per 250 gm of soil was estimated using 40X magnification after they were extracted through the procedure of Cobb's sieving and decanting method [19].

Statistical analysis

Data were subjected to Analysis of variance (ANOVA). Duncan's Multiple Range Test was used to find out significant difference among the treatments. Mean values were considered significant at $P \leq 0.05$ [20]. LC_{50} and LC_{90} were calculated by the AAT Bioquest.

RESULTS AND DISCUSSION

In vitro experiment

Effect of plants extract on the nematodes hatching and mortality

The study revealed that highest hatching inhibition was observed in *A. indica* treated plants followed by *V. nilotica, L. inermis, J. curcas* and *P. longifolia.* The 240 hours exposed plants exhibited highest inhibition (92.6) in *A. indica* followed by 168 hours (154). Similar pattern of hatching inhibition was also observed in rest of plant extracts. Further, minimum inhibition was monitored in *P. longifolia* where 240 hours exposed plants showed 519.8 and 168 hours exposed exhibited 535.8 nematodes (Table 1). To check the nematicidal potential of botanical extracts, different botanicals were used for this purpose; it was observed that *A. indica* exposed plants for 120 hours registered maximum (81.8) mortality followed by 96 hrs (76.2), 72 hrs (59.0), 48 hrs (48.2) and 24 hrs (37.6) in all concentrations. On the other hand, minimum mortality of juveniles was seen in *P. longifolia* treated plants where 120 hrs exposed case exhibited 50.6 juveniles followed by 96 hrs (46.6), 72 hrs (38.6), 48 hrs (32.2), and 24 hrs (30.2) (Table 2).

Table 1 Effect of different concentrations of various botanicals upon the *Meloidogyne incognita* mortality *in vitro*

Botanicals	Duration of Exposure (in hrs)	No. of J2 dead ± SE (± % of SE) in extracts of various concentrations				
		0.01%	0.02%	0.03%	0.04%	0.05%
Azadiracthta indica	24	10.2 ±1.69 (±16.53%)	17.4 ±1.33 (±7.64%)	24.4 ±1.82 (±7.45%)	31.6 ±2.11 (±6.68%)	37.6 ±2.53 (±6.72%)
	48	12.2 ±1.14 (±9.37%)	21.2 ±1.14 (±5.39%)	31.6 ±2.11 (±6.68%)	38.6 ±2.53 (±6.54%)	48.2 ±3.24 (±6.73%)
	72	19.2 ±1.69 (±8.78%)	31.6 ±0.99 (±3.16%)	41.4 ±2.37 (±5.72%)	48.4 ±2.95 (±6.09%)	59.0 ±3.39 (±5.75%)

	96	21.4 ±1.59 (±7.44%)	40.4 ±1.82 (±4.50%)	53.2 ±3.24 (±6.10%)	63.8 ±3.64 (±5.70%)	76.2 ±4.08 (±5.36%)
	120	24.4 ±1.82 (±7.45%)	46.8 ±2.79 (±5.98%)	57.6 ±3.69 (±6.42%)	69.8 ±3.99 (±5.71%)	81.8 ±4.40 (±5.38%)
Jatropha curcas	24	5.6 ±0.48 (±8.57%)	12.2 ±0.73 (±6.01%)	20.2 ±1.14 (±5.66%)	26.8 ±2.43 (±9.08%)	32.4 ±1.82 (±5.61%)
	48	8.6 ±0.99 (±11.62%)	16.2 ±0.73 (±4.53%)	25.8 ±1.44 (±5.58%)	30.6 ±2.11 (±6.90%)	36.6 ±2.53 (±6.90%)
	72	14.8 ±0.73 (±4.96%)	24.6 ±1.33 (±5.40%)	25.8 ±1.44 (±5.58%)	38.4 ±1.82 (±4.73%)	45.2 ±2.09 (±4.63%)
	96	16.8 ±1.30 (±7.74%)	27.6 ±1.18 (±4.26%)	36.2 ±1.90 (±5.25%)	43.8 ±2.27 (±5.18%)	50.8 ±2.79 (±5.51%)
	120	18.4 ±0.99 (±5.43%)	30.2 ±1.57 (±5.19%)	34.2 ±1.57 (±4.58%)	46.4 ±3.01 (±6.49%)	53.2 ±2.73 (±5.13%)
Lawsonia inermis	24	6.4 ±0.99 (±15.62%)	13.4 ±0.99 (±7.46%)	21.8 ±1.99 (±9.17%)	27.2 ±1.57 (±5.76%)	33.4 ±1.82 (±5.44%)
	48	9.2 ±0.73 (±7.97%)	18.0 ±1.39 (±7.70%)	26.2 ±1.57 (±5.98%)	31.6 ±2.11 (±6.68%)	38.4 ±2.37 (±6.17%)
	72	16.0 ±1.39 (±8.66%)	26.4 ±1.82 (±6.88%)	33.4 ±1.82 (±5.44%)	39.6 ±2.67 (±6.75%)	46.6 ±3.69 (±7.94%)
	96	18.2 ±0.73 (±4.03%)	32.6 ±2.37 (±7.26%)	45.0 ±2.56 (±5.68%)	52.2 ±2.93 (±5.62%)	58.8 ±3.12 (±5.31%)
	120	20.6 ±1.33 (±6.45%)	35.6 ±2.67 (±7.51%)	48.0 ±2.56 (±5.32%)	56.8 ±3.12 (±5.50%)	64.0 ±3.39 (±5.30%)
Polyalthia longifolia	24	4.6 ±0.99 (±21.73%)	10.4 ±2.02 (±19.40%)	19.0 ±1.39 (±7.29%)	24.4 ±2.02 (±8.27%)	30.2 ±2.73 (±9.04%)
	48	6.8 ±1.14 (±16.81%)	14.8 ±1.30 (±8.78%)	23.6 ±0.99 (±4.23%)	27.8 ±1.44 (±5.18%)	32.2 ±2.51 (±7.80%)
	72	13.0 ±1.39 (±10.66%)	22.2 ±1.57 (±7.06%)	29.2 ±2.51 (±8.60%)	33.8 ±2.27 (±6.71%)	38.6 ±1.82 (±4.71%)
	96	15.4 ±0.99 (±6.49%)	25.2 ±1.57 (±6.22%)	33.4 ±6.09 (±18.24%)	40.4 ±1.82 (±4.50%)	46.6 ±2.53 (±5.42%)
	120	17.0 ±1.39 (±8.15%)	27.4 ±1.82 (±6.63%)	35.8 ±1.69 (±4.71%)	42.4 ±2.67 (±6.30%)	50.6 ±2.95 (±5.82%)
Vachellia nilotica	24	7.2 ±0.73 (±10.19%)	14.2 ±0.73 (±5.16%)	22.8 ±1.90 (±8.33%)	28.8 ±1.90 (±6.60%)	35.2 ±1.57 (±4.45%)
	48	10.6 ±1.33 (±12.54%)	18.8 ±1.44 (±7.66%)	28.8 ±1.44 (±5.00%)	34.6 ±1.82 (±5.25%)	43.4 ±2.67 (±6.16%)
	72	17.8 ±1.68 (±9.47%)	28.6 ±2.02 (±7.06%)	39.2 ±1.90 (±4.85%)	48.0 ±2.97 (±6.19%)	56.2 ±2.73 (±4.86%)
	96	19.8 ±1.44 (±7.27%)	32.8 ±1.90 (±5.79%)	49.6 ±3.69 (±7.46%)	59.4 ±2.67 (±4.50%)	69.6 ±3.19 (±4.59%)
	120	22.4 ±1.31 (±5.86%)	37.6 ±2.11 (±5.61%)	52.6 ±3.19 (±6.08%	63.8 ±2.73 (±4.28%)	72.8 ±3.24 (±4.46%)
Control (in DW)		00.0 ±0.00 (±0.00%)	00.0 ±0.00 (±0.00%)	00.0 ±0.00 (±0.00%)	00.0 ±0.00 (±0.00%)	00.0 ±0.00 (±0.00%)

Each value is the mean of five replicates
SE- Standard error, DW- Distilled water
Values of percent of standard errors are given in brackets

Table 2 Effect of different concentrations of various botanicals upon the *Meloidogyne incognita* (J2) mortality *in vitro*

Botanicals	Duration of exposure	LC50 value in percent (95% CL)	LC90 value in percent (95% CL)
Azadiracthta indica	24	0.072	0.267
	48	0.053	0.127
	72	0.040	0.102
	96	0.027	0.067
	120	0.023	0.061
Jatropha curcas	24	0.091	NaN
	48	0.075	0.463
	72	0.062	0.234
	96	0.049	0.162
	120	0.043	0.132
Lawsonia inermis	24	0.083	0.639
	48	0.072	0.249

	72	0.051	0.150
	96	0.036	0.089
	120	0.032	0.092
Polyalthia longifolia	24	0.121	NaN
	48	0.084	NaN
	72	0.075	0.425
	96	0.057	0.265
	120	0.050	0.149
Vachellia nilotica	24	0.077	0.490
	48	0.061	0.170
	72	0.042	0.129
	96	0.032	0.077
	120	0.028	0.069

LC_{50}- Lethal concentration caused 50% mortality after 24, 48, 72, 96 and 120 hours at 95% confidence limit
LC_{90}- Lethal concentration caused 90% mortality after 24, 48, 72, 96 and 120 hours at 95% confidence limit
CL- Confidence limit

Effect of lethal concentration (LC_{50} and LC_{90})

The botanicals with least LC_{50} and LC_{90} values revealed highest mortality and higher LC_{50} and LC_{90} values showed lesser mortality of *Meloidogyne incognita*. It was observed that least LC_{50} and LC_{90} values were recorded in *Azadirectha indica* treated samples for 120 hours and followed by 96, 72, 48 and 24 hours (Table 3).

Table 3 Effect of different concentrations of various botanicals upon the *Meloidogyne incognita* hatching *in vitro*

Botanicals	Duration of exposer (in hrs)	No. of J2 hatched ± SE (± % of SE) in extracts of various concentrations (after D_7 and D_{10})					
		0.01%	0.02%	0.03%	0.04%	0.05%	Control (in DW)
Azadiracthta indica	168	743.2 ±28.19 (±3.79%)	443.4 ±13.92 (±3.14%)	305.8 ±10.30 (±3.37%)	221.4 ±8.65 (±3.91%)	154.4 ±6.69 (±4.33%)	960
	240	724.2 ±22.45 (±3.10%)	402.6 ±15.05 (±3.74%)	254.8 ±7.83 (±3.07%)	164.2 ±6.89 (±4.19%)	92.6 ±3.99 (±4.32%)	960
Jatropha curcas	168	883.4 ±34.81 (±3.94%)	733.2 ±24.62 (±3.36%)	641.4 ±19.49 (±3.04%)	515.8 ±16.02 (±3.11%)	403.4 ±12.95 (±3.21%)	960
	240	878.6 ±32.70 (±3.72%)	722.2 ±22.45 (±3.11%)	626.6 ±16.00 (±2.55%)	495.8 ±12.22 (±2.46%)	378.6 ±9.75 (±2.58%)	960
Lawsonia inermis	168	822.4 ±33.83 (±4.11%)	526.6 ±17.55 (±3.33%)	481.4 ±14.12 (±2.93%)	341.2 ±12.48 (±3.66%)	262.6 ±8.24 (±3.14%)	960
	240	813.8 ±31.89 (±3.92%)	503.6 ±14.24 (±2.83%)	456.0 ±11.74 (±2.58%)	309.0 ±7.67 (±2.48%)	226.6 ±6.86 (±3.03%)	960
Polyalthia longifolia	168	902.2 ±37.28 (±4.13%)	775.6 ±31.59 (±4.07%)	683.4 ±18.83 (±2.75%)	602.2 ±14.41 (±2.39%)	535.8 ±15.83 (±2.95%)	960
	240	898.8 ±36.03 (±4.01%)	767.8 ±25.41 (±3.31%)	672.6 ±17.38 (±2.58%)	588.6 ±13.49 (±2.29%)	519.8 ±13.47 (±2.59%)	960
Vachellia nilotica	168	805.8 ±29.55 (±3.67%)	533.2 ±15.39 (±2.89%)	414.6 ±13.04 (±3.15%)	307.8 ±11.50 (±3.74%)	232.2 ±7.75 (±3.34%)	960
	240	794.8 ±26.27 (±3.31%)	507.2 ±14.22 (±2.80%)	381.8 ±11.47 (±3.00%)	269.2 ±9.33 (±3.46%)	189.4 ±7.68 (±4.06%)	960

Each value is the mean of five replicates
SE- Standard error, DW- Distilled water
Values of percent of standard errors are given in brackets

Pot experiment
Effect on growth and yield parameters

This experiment was performed to examine the impact of botanicals on growth and yield, physiological and nematological parameters infesting *V. radiata* crop. Applied manures significantly increased the growth and yield, and physiological parameters of the crop plants.

Present experiment revealed that shoot, root, and plant length significantly increased by 32.53 cm (50.53%), 14.66cm (52.07%) and 47.19 (52.40%) respectively. In fresh weight of shoot, root, and total fresh weight was also increased by 22.54g (52.40%), 8.60g (50.88%), and 31.14g (51.97%) respectively. Dry weight of shoot, root, and total dry weight of plant increased by 6.77g (48.79%), 1.33g (44.57%), and 5.47g (48.08%) respectively in pots treated with *A. indica*. Similarly, pods formation per plant was also increased by 17.48 (52.66%) and the weight of 100 seeds increased by 5.56g (68.99%). Plants treated with *P. longifolia* manure showed the minimum improvement in growth and yield variables. Growth variables like shoot, root, and total plant length was increased by 25.19 cm (16.57%), 11.16 cm (15.77%), and 36.35 cm (16.32%) respectively; fresh weight of shoot, root, and total fresh weight increased by 17.38 gm (17.51%), 6.63 gm (16.32%), and 24.01 gm (17.18%); dry weight of shoot, root, and total dry weight of plant increased by 5.40 gm (18.68%), 1.11 gm (20.65%), and 6.15 gm (12.43%) respectively. In comparison to the infected control, the number of pods per plant increased by 13.51 (17.99%) and the weight of 100 seeds increased by 4.25g (29.18%) (Table 4).

Table 4 Effect of various botanicals (100g/kg of soil) upon the *Meloidogyne incognita in relation to the* plant length (shoot length and root length), weight (fresh and dry weight), number of pods/plant and weight of 100 seeds of *Vigna radiata*

Treatments	Length (cm)			Fresh weight (g)			Dry weight (g)			Pods plant^{-1}	Weight of / 100 seeds
	Shoot	Root	Total	Shoot	Root	Total	Shoot	Root	Total		
Control	44.01^{a}	19.76^{a}	63.77^{a}	30.40^{a}	11.62^{a}	42.02^{a}	9.25^{a}	1.87^{a}	11.12^{a}	23.69^{a}	7.33^{a}
Meloidogyne incognita	21.61^{f}	9.64^{f}	31.25^{e}	14.79^{f}	5.70^{g}	20.49^{d}	4.55^{g}	0.92^{f}	5.47^{g}	11.45^{e}	3.29^{f}
Meloidogyne incognita + Azadiracthta indica	32.53^{b}	14.66^{b}	47.19^{b}	22.54^{b}	8.60^{b}	31.14^{b}	6.77^{b}	1.33^{b}	8.10^{b}	17.48^{b}	5.56^{b}
Meloidogyne incognita + Jatropha curcas	26.25^{de}	12.01^{d}	38.26^{cd}	18.24^{e}	6.97^{e}	25.21c^{d}	5.59^{e}	1.16^{cd}	6.75^{e}	14.22^{d}	4.32^{e}
Meloidogyne incognita + Lawsonia inermis	29.39^{cd}	13.34^{c}	42.73^{bcd}	20.27^{d}	7.79^{d}	28.06^{bc}	6.20^{d}	1.25^{bc}	7.45^{d}	15.67^{c}	4.98^{d}
Meloidogyne incognita + Polyalthia longifolia	25.19^{e}	11.16^{e}	36.35^{de}	17.38^{f}	6.63^{f}	24.01^{cd}	5.40^{f}	1.11^{d}	6.15^{f}	13.51^{e}	4.25^{f}
Meloidogyne incognita + Vachellia nilotica	30.61^{bc}	14.03^{bc}	44.64^{bc}	21.33^{bc}	8.14^{c}	29.47^{bc}	6.67^{c}	1.50^{b}	8.17^{c}	16.37^{c}	5.21^{c}

Each value is the mean of five replicates

The values in each column followed by same letter are not significantly different according to Duncan's Multiple Range Test (DMRT) at $P \leq 0.05$. C- Control

Effect of botanicals on the physiology of V. radiata

Observed data revealed that *A.indica* treated plants registered highest NRA 0.342 (55.45%), over nematode inoculated control followed by *V. nilotica* 0.332 (50.90%), *L.inermis* 0.294 (32.72%), *J. curcus* 0.254 (14.54%) and *P. longifolia* 0.258 (17.27%). Other examined (Total chlorophyll, carotenoids and proteins) parameters exhibited similar pattern of response (Table 5).

Table 5 Effect of various botanicals upon the *Meloidogyne incognita* in relation to the physiological parameters of *Vigna radiata*

Treatments	NRA	Total chlorophyll	Carotenoids	Proteins
Control	0.460^{a}	2.017^{a}	0.353^{a}	5.150^{a}
Meloidogyne incognita	0.220^{g}	1.000^{g}	0.170^{g}	2.520^{g}
Meloidogyne incognita + Azadiracthta indica	0.342^{b}	1.515^{b}	0.267^{b}	3.815^{b}
Meloidogyne incognita + Jatropha curcas	0.254^{e}	1.292^{e}	0.229^{e}	3.254^{e}
Meloidogyne incognita + Lawsonia inermis	0.294^{d}	1.362^{d}	0.241^{d}	3.437^{d}
Meloidogyne incognita + Polyalthia longifolia	0.258^{f}	1.226^{f}	0.227^{f}	3.064^{f}
Meloidogyne incognita + Vachellia nilotica	0.332^{c}	1.423^{c}	0.251^{c}	3.588^{c}

Each value is the mean of five replicates

The values in each column followed by same letter are not significantly different according to Duncan's Multiple Range Test (DMRT) at $P \leq 0.05$. C- Control

Effect on the nematode related parameters

Highest suppression in nematode related parameters like galls, eggmasses, egg per eggmass and total nematode population was observed in *A indica* inoculated plants followed by *V. nilotica*, *L. inermis*, *J. curcas*, and *P. longifolia* (Table 6).

Table 6 Effect of various botanicals upon the *Meloidogyne incognita* in relation to the nematode related parameters of *Vigna radiata*

Treatments	Number of galls / root system	Number of egg masses / root system	Number of egg / egg mass	Reproduction factor (Rf)	Final nematode population
Control	-	-	-	-	-
Meloidogyne incognita	161.00^{a}	69.00^{a}	143.00^{a}	5.53^{a}	11067.00^{a}
Meloidogyne incognita + Azadiracthta indica	93.00^{e}	39.00^{e}	109.00^{e}	3.75^{f}	7494.00^{f}
Meloidogyne incognita+ Jatropha curcas	112.00^{b}	47.00^{b}	132.00^{b}	4.55^{c}	9095.00^{c}
Meloidogyne incognita+ Lawsonia inermis	105.00^{c}	43.00^{c}	122.00^{c}	4.29^{d}	8574.00^{d}
Meloidogyne incognita+ Polyalthia longifolia	113.00^{b}	47.00^{b}	133.00^{b}	4.63^{b}	9250.00^{b}
Meloidogyne incognita+ Vachellia nilotica	101.00^{d}	42.00^{d}	119.00^{d}	4.14^{e}	8280.00^{e}

Each value is the mean of five replicates

The values in each column followed by same letter are not significantly different according to Duncan's Multiple Range Test (DMRT) at $P \leq 0.05$. C- Control

From the above experiment it is clear that botanicals used for treatments contains some growth promoting as well as nematicidal compounds. These chemicals are toxic in nature to the nematodes. Such chemicals are not required by the individual plant but used in defense system. These metabolites have the nematicidal capacity against the phytoparasitic nematodes [21-22]. Plants have some chemicals which have been proven proved growth promoting in nature such as *D. inoxxia, L. camara, A. mexicana, H. integrifolia, A. scholaris* and *P. dulce* have plant promoting

ability in *V. radiata* [23]; *Ulva lactuca* treated plants showed significant improvement in growth and biochemical parameters of *V. radiata* [24]. Leaves extracts of some indigenous plant species like *A. indica, Allium sativum, Polygonum lanatum, Allamanda cathartica, Terminalia arjuna and P. longifolia* was found effective against the disease of *V. radiata* [25]. *A. indica* have secondary metabolites like Azadirachtin [26] and Nimbin, Salanin [8], 3-acetyl-1-tigloylazadirachtinin and 3-tigloylazadirachtol were collected from its underground parts [27] which are nematicidal in nature. Ferulic, gallic, and tannic acids were found in the bark of *A. indica* [28], alkaloids, phenolics, flavonoida, glycosides, coumarins, saponins and tannins in *J. curcas* [29-31], Naphthoquinone derivatives, coumarins, xanthones, tannins, flavonoids, triterpenes, and sterols were also identified in *L. inermis* [32], *P. longifolia* contains diterpenoides, alkaloids, tannins, and mucilage [33-35] and *V. nilotica* contains tannins (astringent) [36].

In the present study, aqueous extract of *A. indica, J. curcas, L. inermis, P. longifolia and V. nilotica* showed nematicidal potential in terms of nematode hatching and mortality. Out of 5 botanicals, *A. indica* was found highly effective in all concentration as compared to the remaining botanicals that were used in the experiment.

The results of in vitro mortality showed that the plant extract of *A. indica* have highest nematicidal potential against *M. incognita* at D5. Extracts of *A. indica*, after the 24, 48, 72, 96, and 120 hours of exposure gave different values of LC_{50} as well as LC_{90} compared to control. Similar, results were also observed by Zaidat *et al*. [13]. Researchers also found that at lesser value of LC_{50} and LC_{90} exhibited most toxic and more value of LC_{50} and LC_{90} exhibited least toxic [37].

Pot experiments revealed that botanical amended plants registered good crop growth and yield. Pots treated with *A. indica* manure showed maximum crop growth followed by *V. nilotica, L. inermis, J. curcus* and *P.longifolia*. Enhanced growth and yield of *V. radiata* in amended soil may be due to poor nematode population and fortified soil. Improvement in the biochemical parameters may be due to healthy nature of crop after amending the manure. Nematode related parameters were diminished significantly due to presence of some nematicidal compounds in the used botanicals [8], [27].

CONCLUSION

This research have clearly indicates that use of five botanicals in the form of aqueous extract in vitro increase the mortality rate of J2s and decrease the rate of egg hatching of *M. incognita* in each level of concentration. This inhibitory effect may be due to presence of various chemical compounds in the botanicals that were used. Green house experiment gave results with significant improvement with *A. indica*. Plant length, plant weight in fresh and dry condition, number of pods per plant and biomass of seeds increase with significant difference compare to the control. These growth factors directly associated with the improvement in the biochemical parameters mainly protein contents. The results obtained from the use of *A. indica, J. curcas, L. inermis, P. longifolia and V. nilotica* indicate that further study can be done on the specific substance/components which affect the *M. incognita* population, improve the protein contents in the seeds and balanced the soil properties. Alternative treatments can be used in the further study which are available in access but not used by our farmers. For example, paddy straw produced in million tonnes each year by the farmers of Haryana, Punjab and Western Uttar Pradesh causes pollution, if burned to clear the field for next crop, in New Delhi and nearby areas of India.

Abbreviations

NaN: Not a number, LC_{50}: Lethal concentration 50, LC_{90}: Lethal concentration 90.

Competing interests

There are no competing interests declared by the authors.

Acknowledgements

The author wishes to express his gratitude to the University Grants Commission of India for financial assistance, and Department of Botany, AMU, Aligarh. Dr. Athar Ali Khan deserves special thanks for his assistance during the experiment.

LITERATURE CITED

1. Askary TH. 2017. Diversity of plant parasitic nematodes in pulses. *In*: (Eds) Ansari AA, Gill SS, Abbas ZK, Naeem M. Plant biodiversity: monitoring, assessment and conservation, CABI, Wellingford, U.K. pp 239-274.
2. Gaur HS, Singh RV, Kumar S, Kumar V, Singh JV. 2001. Search for nematode resistance in crops. AICRP on nematodes, Division of Nematology, IARI, New Delhi Publication. pp 4.
3. Hussaini S, Seshadri R. 1975. Interelationship between *Meloidogyne incognita* and *Rhizobiume* sp. on mungbean (*Phaseolus aureus*). *Indian Journal of Nematology* 5: 189-199.
4. Odeyemi IS, Afolamia SO, Adekoyejoa AB. 2013. Integration of *Glomus mosseae* with *Chromolaena odorata* powder for suppression of *Meloidogyne incognita* on maize (*Zea mays* L.). *Arch. Phytopatho.l Plant Protection* 46: 1589-1597.
5. Hassan MA, Pham TH, Shi H, Zheng J. 2013. Nematodes threats to global food security. Acta Agriculturae Scandinavica, Sect. B- *Soil and Plant Science* 63(5): 420-425.
6. Curto G, Dongiovanni C, Sasanelli N, Santori A, Myrta A. 2014. Efficacy of dimethyl disulfide (DMDS) in the control of the root-knot nematode *Meloidogyne incognita* and the cyst nematode *Heterodera carotae* on carrot in field condition in Italy. *Acta Horticulturea* 1044: 405-410.
7. Xia Y, Li S, Liu X, Zhang C, Xu J, Chen Y. 2019. *Bacillus halotolerans* strain LYSX1-induced systemic resistance against the root-knot nematode *Meloidogyne javanica* in tomato. *Ann. Microbiology* 69: 1227-1233.
8. Akhtar M. 2000. Nematicidal potential of the neem tree *Azadirachta indica* (A. Juss). *Integrated Pest Management Reviews* 5: 57-66.
9. Gamal E, Lee D, Park J, Yu H. 2008. Eavaluation of various plant extracts for their nematicidal efficacies against juveniles of *Meloidogyne incognit*. *Journal of Asia-pacific Entomology* 11: 99-102.

10. Fabiyi OA, Osunlola OS, Olatunji GA. 2015. *In vitro* toxicity of extracts from *Hyptis suaveolens* (L.) point on eggs and second stage juveniles of *Heterodera sacchari*. *Agrosearch* 15(1): 89-99.
11. Baba Ali D, Roeb J, Hammache M, Hallmann J. 2017. Nematicidal potential of aqueous and ethanol extracts gained from *Datura stramonium*, *D. innoxia* and *D. tatula* on *Meloidogyne incognita*. *Jr. Plant Dis. Prot.* 124: 339-348.
12. Malik MMR, Akhtar MJ, Ahmad I, Khalid M. 2014. Synergistic use of *Rhizobium*, compost and nitrogen to improve growth and yield of mungbean (*Vigna radiata*). *Pak. Jr. Agric. Sciences* 51: 393-398.
13. Zaidat SAE, Mouhouche F, Babaali D, Abdwssemed N, Cara MD, Hammache M. 2020. Nenaticidal activity of aqueous and organic extracts of local plants against *Meloidogyne incognita* (Kofoid and White) Chitwood in Algeria under laboratory and greenhouse conditions. *Egypt Jr. Biol. Pest Control* 30: 30-46.
14. Bliss CI. 1934. The method of probits. *Science* 79(2037): 38-39.
15. Jaworski EG. 1971. Nitrate reductase assay in intact plant tissues. *Biochem. Biophys. Res. Commun*. 43(6): 1274-1279.
16. Arnon D. 1949. Copper enzymes isolated chloroplasts, polyphenoloxidase in *Beta vulgaris*. *Plant Physiology* 24: 1-15.
17. Kirk JTO, Allen RL. 1965. Dependence of chloroplast pigments synthesis on protein synthetic effects on actilione. *Biochem. Biophysics Res. Jr. Canada* 27: 523-530.
18. Lowry OH, Rosebrough NJ, Farr AI, Randall RJ. 1951. Protein measurement with folin phenol reagent. *Journal of Biological Chemistry* 193: 265-275.
19. Cobb NA. 1918. Estimating the nema population of the soil. *Agric. Tech. Circ. Bur. Pl. U.S. Dep. Agriculture*. pp 1:48.
20. Gomez KA, Gomez AA. 1984. *Statistical Procedures for Agricultural Research*. (2 Edition). John Wiley and Sons, New York. pp 680.
21. Chitwood DJ. 2002. Phytochemical based strategies for nematode control. *Annu. Rev. Phytopathology* 40: 221-249.
22. Andres MF, Rossa GE, Cassel E, Vargas RMF, Santana O, Diaz CE, Gonzalez CA. 2017. Biocidal effects of *Piper hispidinervum* (Piperaceae) essencial oil and synergism among its main components. *Food and Chemical Toxicology* 109(2): 1086-1092.
23. Kadoo M, Badere RS. 2012. Effect of aqueous extract of commonly available plants on early growth of *Vigna radiata*. *Hislopia Journal* 5: 1-5.
24. Bakker J, Gommers FJ. 1978. Activity of the nematicidal principal alpha-terthienyl. *Proceedings of the International Congress of Plant Pathology*, Berlin. pp 144.
25. Gowda ND, Setty KGH. 1972. *In vitro* studies of neem cake (*Azadirachta indica*) extract on hatching of eggs and survival of larvae of root-knot nematode, *Meloidogyne incognita*. *Current Science* 8: 78-79.
26. Sundaram KMS, Curry J. 2016. Effect of some UV absorbers on the photostablization of azadirachtin, a neem-based biopesticide. *Chemosphere* 32(4): 649-659.
27. Khan MB, Alam MM, Khan AM, Saxena SK. 1975. Effect of water-soluble fractions of oil cakes and bitter principles of neem on some nematodes. *Acta Botany* 2: 120-128.
28. Khan AM, Alam MM, Ahmed R. 1974. Mechanism of control of plant parasitic nematodes as a result of the application of oil cakes to the soil. *Indian Jr. Nematology* 4: 93-96.
29. Balaji R, Rekha N, Deecaraman M, Manik L. 2009. Antimetastatic and antiproliferative activity of methanolic fraction of *Jatropha curcas* against B16F10 melanoma induced lung metastasis in C57BL/6 mice. *African Jr. Pharm. Pharmacology* 3(11): 547-555.
30. Abugre S, Quashie-Sam SJ. 2010. Evaluating the allelopathic effect of *Jatropha curcus* aqueous axtract on germination, radicle and plumule length of crop. *Int. Jr. of Agric. and Biology* 12(5): 769-772.
31. Rejila S, Vijaykumar N, Jayakumar M. 2012. Chromatographic determination of alleochemicals (phenolic acids) in *Jatropha curcus* by HPTLC. *Asian Jr. Plant Sci. Research* 2(2): 123-128.
32. Varghes JK, Silvipriya K, Resmi S, Jolly C. 2010. *Lawsonia inermis* (Henna): A natural dye of various therapeutic uses- A review. *Inventi. Rapid: Cosmeceuticals* 1: 1-5.
33. Phadnis AP, Patwardhan SA, Dhaneshwar NN, Tavale SS, Row TNG. 1988. Clerodane diterpenoids from *Polyalthia longifolia*. *Phytochemistry* 27: 2899-2901.
34. Faizi S, Khan R, Mughal N, Malik M, Sajjadi KS, Ahmad A. 2008. Antimicrobial activity of various parts of *Polyalthia Longifolia* var. pendula: isolation of active principles from the leaves and the berries. *Phytotherapy Research* 22: 907-912.
35. Lee TH, Wang MJ, Chen PY, Wu TY, Wuche W, Tsai FU, Lee CK. 2009. Constituents of *Polyalthia longifolia* var. Pendula. *Jr. of Nat. Products* 72: 1960-1963.
36. Duke JA, Duke PK. 1983. *Medicinal Plants of the Bible*. Trado-Medic Books, Owerri New York.
37. Zasada IA, Klassen W, Meyer SLF, Codallo M, Abdul-Baki AA. 2006. Velvetbean (*Mucuna pruriens*) extracts: Impact on *Meloidogyne incognita* survival and on *Lycopersicon esculentum* and *Lactuca sativa* germination and growth. *Pest Management Science* 62(11): 1122-1127.