Eco-friendly based Integrated Management Practice against Root-Knot Nematode, *Meloidogyne incognita* on Cluster bean (*Cyamopsis tetragonoloba* L.)

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Abstract

Aim

The experiment was carried out during two consecutive *Kharif* seasons to test the combined efficacy of biochemicals *i.e.* salicylic acid, ascorbic acid and L-arginine each at 2 % w/w as seed treatment and botanicals *viz.* neem, lantana and parthenium leaves powder each at 5 g per plant for the management of root-knot nematode, *Meloidogyne incognita* infecting cluster bean..

Method

All treatments applied as soil application at the time of sowing. Before sowing observation on initial nematode population/100 cc soil and at the termination of experiment number of galls/plant, number of egg masses/plant, number of eggs and larvae/egg mass, final nematode population/100 cc soil and yield (q/ha) were recorded.

Result

Among different combinations, minimum nematode population were observed with ascorbic acid at 2 per cent w/w + neem leaves powder at 5 g per plant followed by ascorbic acid at 2 per cent + lantana leaves powder at 5 g per plant and salicylic acid at 2 per cent + neem leaves powder at 5 g per plant.

Conclusion: Results of experiment showed that application of biochemicals along with botanicals significantly reduced galls per plant as compared to untreated check.

Introduction

Cluster bean, *Cyamopsis tetragonoloba* (L.) is a drought and high temperature tolerant deep rooted summer legume of high social and economic significance in India. The qualities of the crop like high adaptation towards erratic rainfall, multiple industrial uses and its importance in cropping system for factors such as soil enrichment properties, low input requirement, etc. have made the cluster bean one of the most significant crop for farmers in the arid region. It is popularly known as '*Guar*' and mainly used as vegetable, fodder, green manuring and seeds. Guar meal, a byproduct of gum industries form concentrate animal feed for immense value as it contains more than 42 per cent protein against 31 per cent in guar seed.

India is the largest producer of cluster bean and contributes 80 percent of total cluster bean production in the world. It occupies about three-fourth of the global cluster bean cultivation and grown in 183.1 thousand ha of area with the production of 268.0 thousand tonnes and average productivity being 146.4 kg/ha during 2014-15 (Anonymous, 2016). Being deep rooted and drought hardiness, this crop has occupied tremendous areas in arid and semi-arid regions encompassing Rajasthan, Gujarat and Haryana state. Rajasthan ranks first with respect to area and production. In Rajasthan it is cultivated in 34.32 lakh ha of area with production and productivity of 12.44 lakh tonnes and 363 kg/ha, respectively during 2017-18 (Anonymous, 2017).

The significantly higher prices of cluster bean in recent time have helped expand the crop to non-traditional regions and seasons. The demand of processed cluster bean in world market is expected to increase with the expansion of shale oil gas tracking to new countries like China and Russia and scaling up in prominent existing countries like USA along with other uses in food and textile industries owing to increased food safety and health concerns.

The crop of such an immense economic value is prone to attacked by several pests and diseases (aphid, white fly, jassids, blight, powdery mildew etc.) including plant parasitic nematode (Sitaramaiah et al., 1971 and Rao et al., 2007). Several plant parasitic nematodes *viz.*, *Meloidogyne incognita*, *M. javanica*, *Rotylenchulus reniformis*, *Helicotylenchus incises*, *Pratylenchus delattrei*, *Tylenchorhynchus capitatus*, *Xiphinema* sp., *Tylenchus* sp., *Criconea* sp. and *Aphelenchus* sp. have been found associated with cluster bean in India and abroad (Bishnoi and Yadav, 1989; Shaukat et al., 2010). Root-knot nematode (*M. incognita*) is considered as most important pest of agronomical and horticultural crops including cluster bean (Seshadri, 1970; Sasser, 1980, Yadav et al., 2009 and Singh and Kumar, 2015) and farmers experience chronic losses because of the high frequency of this nematode (Bhatti and Jain, 1977; Parvatha Reddy, 1986 and Nama and Baheti, 2021). Root-knot nematode received greater attention in India and abroad due to its polyphagous nature, cosmopolitan distribution and adaptability to adverse conditions.

In view of disease severity and crop losses caused by these tiny organisms, attempts were made to test various pesticides for the management of root-knot nematodes on vegetables including cluster bean by various researchers but due to health hazards, residual toxicity, environmental pollution and high cost, their adoption at farmer's level has been limited. Therefore, present investigations were emphasized to evolve economical and eco-friendly technologies for the management of plant parasitic nematodes on agri-horticultural crops.

Material And Methods

The experiment was carried out in *Kharif* 2015 and 2016 on farmer's field naturally infested with root-knot nematode, *Meloidogyne incognita* at Behuti near Udaipur having light textured soil. Analysis of soil was done with the help of International Pipette Method (Wright, 1939). The efficacy of biochemicals *i.e.* salicylic acid, ascorbic acid and L-arginine (1, 2 and 4% w/w) were tested against root-knot nematode, *M. incognita* on cluster bean (RGC-936). A treated chemical (acephate 75 SP 2% w/w) and untreated check were also be taken for comparison. The experiment was laid out in randomized block design and all the treatments were replicated five times. Atmost care was taken right from sowing till harvest of experiment for proper growth and development of plants. Observations on initial nematode population/100 cc soil, No. of galls/plant, No. of egg masses/plant, No. of eggs and larvae/egg mass, final nematode population/100 cc soil and Yield q/ha were recorded.

(i) Collection and processing of samples:

Soil samples were collected from each experimental plot, labelled properly and brought to the laboratory and kept in refrigerator till processing. Soil samples were processed by using Cobb's Sieving and Decanting Technique (Cobb, 1918) followed by Baermann's Funnel Assembly Technique (Christie and Perry, 1951). Processed samples were thoroughly examined under microscope to estimate population of test nematode. However, for root population, root samples were soaked in water, rinsed gently to remove adhering soil particles and stained with 0.1% acid fuchsin lactophenol solution at 80°C for 2–3 minutes (Mc Beth et al., 1941) and kept in clear lactophenol at least for 24 hours.

(ii) Counting of galls and egg masses per plant:

After harvesting, root samples were collected from each experimental plot, labelled properly and brought to the laboratory. Roots were gently washed in running tap water to remove adhering soil particles. Well cleaned were taken and observed thoroughly under magnifying glass for counting of galls and egg masses.

(iii) Counting of eggs and larvae per egg mass:

After counting of galls and egg masses, roots were stained with 0.1% acid fuchsin lactophenol solution, rinsed in water to remove excess amount of stain and kept in clear lactophenol at least for 24 hours before examination. Egg masses were randomly selected and detached from stained roots and put in a drop of clear lactophenol on glass slide, covered with cover slip and press gently so that contents of egg mass spread thoroughly. Thereafter, eggs and larvae were counted with the help of telecounter under stereoscopic binocular microscope.

(iv) Identification of root-knot nematode species:

Root samples collected from experimental field, were brought to the laboratory and washed carefully in running tap water to remove adhering soil particles. Egg masses with females were detached from roots with the help of teasing needle and forcep under stereoscopic binocular microscope. Egg masses were kept in water for 24 hours for hatching and females were picked up for identification of species. Posterior cuticular patterns of these females were cut with the help of scaples and the body contents were removed gently with camel brush No. 1 (Taylor and Netscher, 1974). Observation of such several pattern were recorded and the nematode species was identified as *M. incognita* (Eisenback et al., 1981).

Result

Integrated nematode management consisted of the development, use and evaluation of nematode management strategies that result in favorable socio-economic and environmental consequences. It is a system approach to reduce nematode damage to tolerable levels by integration of different management practices. It is now widely recognized as a most desirable approach for managing nematode problems. It involves the design of an appropriate blend of technologies to meet out the needs of the farmers. Therefore, in present investigation, combined efficacy of biochemicals *i.e.* salicylic acid, ascorbic acid and L-arginine (2 % w/w) as seed treatment and botanicals *viz.* neem, lantana and parthenium leaves powder (5 g per plant) as soil application have been tried in combination for the management of root- knot nematode, *M. incognita* infecting cluster bean.

Nematode parameters

Results in table 1 and 2 exhibited that all combination of biochemicals and botanicals significantly reduced galls per plant as compared to untreated check (88.70). Among different combinations, minimum galls per plant (35.90) was observed with ascorbic acid at 2 per cent w/w + neem leaves powder at 5 g per plant followed by ascorbic acid at 2 per cent + lantana leaves powder at 5 g per plant (40.30) and salicylic acid at 2 per cent + neem leaves powder at 5 g per plant (43.70). L-arginine at 2 per cent w/w + parthenium leaves powder at 5 g per plant (70.20) was found to be least effective. Maximum reduction (59.53%) in galls per plant was observed with ascorbic acid at 2 per cent w/w + neem leaves powder at 5 g per plant followed by ascorbic acid at 2 per cent + lantana leaves powder at 5 g per plant (54.57%) and salicylic acid at 2 per cent + neem leaves powder at 5 g per plant (50.73%). Similar results were also recorded with respect to egg masses per plant and final soil population per 100 cc soil.

Yield parameter

Data on yield parameters in table 1 and 2 explicit that all the combination enhanced yield of cluster bean over untreated check (6.62 q/ha). Among different combinations, maximum yield (11.78 q/ha) was obtained with ascorbic acid at 2 per cent w/w + neem leaves powder at 5 g per plant followed by ascorbic acid at 2 per cent w/w + lantana leaves powder at 5 g per plant (11.20 q/ha) and salicylic acid at 2 per cent w/w + neem leaves powder at 5 g per plant (10.82 q/ha). Maximum increase in yield (77.95%) was recorded with the application of ascorbic acid at 2 per cent w/w + neem leaves powder at 5 g per plant followed by ascorbic acid at 2 per cent w/w + lantana leaves powder at 5 g per plant (69.18%) and salicylic acid at 2 per cent w/w + neem leaves powder at 5 g per plant (63.44%). Minimum increase in yield (16.01%) was observed with L-arginine at 2 per cent + parthenium leaves powder at 5 g per plant.

Discussion

Nematode parameters

These findings are in accordance with the findings of other research workers (Chawla and Goswami, 2002; Haseeb and Kumar, 2009; Naserinasab et al., 2012 and Mostafanezhad et al., 2014).

Chawla and Goswami (2002) studied the combined effect of neem cake and carbofuran against *M. incognita* on mung bean. The sole application of neem cake and carbofuran was applied at 1 % w/w and 2 kg/ha, respectively, for comparison. Results revealed that the combined application of neem cake and

carbofuran at half the rate of sole application reduced nematode population and found statistically at par with the sole carbofuran treatment. Haseeb and Kumar (2009) observed lowest root-knot index (0.45) when plots of lentil were treated with *P. fluorescens* + *T. viride* + neem seed powder + Carbosulfan. Similarly in a greenhouse experiment, Naserinasab et al. (2012) recorded that combined application of salicylic acid and *T. harzianum* decreased the severity of disease compared with nematode infected treatment as control. Mostafanezhad et al. (2014) used *Arthrobotrys oligospora* as soil drenching coupled with salicylic acid as soil application or foliar spray against root-knot nematode (*Meloidogyne javanica*) on tomato and found that combination of *A. oligospora* and salicylic acid reduced diameter of galls (28%), number of galls per plant (40%) and number of egg masses per plant (53%) compared to control.

Yield parameter

Several workers (Patel and Patel, 1998; Singh and Goswami, 2001; Kumar et al., 2016 and Baheti et al., 2018) also observed better plant growth and yield with integration techniques and supports the findings of present investigation.

Patel and Patel (1998) recorded that carbosulfan 25 ST @ 0.75% as seed treatment coupled with soil application of organic amendments viz., pressmud (3.0 t/ha), poultry manure (3.0 t/ha), neem cake (1.0 t/ha) and mustard cake (1.0 t/ha) significantly enhanced height of plants, grain and fodder yield of chickpea. Singh and Goswami (2001) reported that combination of neem cake and carbofuran at reduced doses significantly increased growth of cowpea and reduced infection of root-knot nematode, *M. incognita* and wilt fungus, *F. oxysporum*. Kumar et al. (2016) conducted a cage house study to assess the combined efficacy of bio-agents (*T. viride*, *P. lilacinus* and *P. fluorescens*) as soil treatment at 1.5g/kg soil and plant extracts viz., jatropha, aak, and neem as root deep treatment at 20 per cent concentration against root-knot nematode, *Meloidogyne incognita* infecting Chilli (*Capsicum frutescens* L.) and found that *P. lilacinus* + neem leaves extracts was most effective to increase plant growth characters as compared to other combinations. Baheti et al. (2018) tested chemical inducers viz. salicylic acid (250 ppm) and ascorbic acid (500 ppm) as seed soaking (12 hours) and foliar spray (30 and 60 days after sowing) treatments in combination in okra infected with root-knot nematode, *Meloidogyne incognita* in field and recorded that seed soaking + foliar spray of ascorbic acid was most effective to increase 27.66–29.81% crop yield followed by salicylic acid as seed soaking + foliar spray (21.15–23.40%) over untreated control during Ist and IInd year, respectively.

These findings clearly indicated that integration of biochemicals (seed dressing) and plant leaves powder (organic amendment) at reduced dose provides good alternatives for management of root-knot nematode, *M. incognita* while augmenting the limitations of bulk quantity of plant leaves powder required for sole application of organic amendment. It is logistic from these findings that biochemicals and plant leaves powder when used in combination at reduced dose proved effective, eco-friendly and economical to enhance crop productivity in nematode prone areas.

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Tables

Table-1 : Management of root-knot nematode, *Meloidogyne incognita* on cluster bean through biochemicals as seed treatment and botanicals as soil amendment

Treatments		Galls/plant			Egg masses/plant			Eggs and larvae/egg mass			Final Nematode Population/100 cc soil			Yield (q/ha)	
		1 st year	11 nd year	Pooled	1 st year	11 nd year	Pooled	1 st year	11 nd year	Pooled	1 st year	11 nd year	Pooled	1 st year	11 th year
Salicylic Acid @ 2% w/w + Neem Leaves Powder 5g/plant	T ₁	46.00	41.40	43.70	38.20	34.20	36.20	158.25	153.50	155.88	708.60	692.00	700.30	10.71	10.71
Salicylic Acid @ 2% w/w + Lantana Leaves Powder 5g/plant	T ₂	54.20	48.60	51.40	44.40	40.60	42.50	201.75	198.00	199.88	772.80	740.40	756.60	9.89	10.71
Salicylic Acid @ 2% w/w + Parthenium Leaves Powder 5g/plant	T ₃	60.40	56.00	58.20	49.00	43.40	46.20	223.00	216.25	219.63	819.40	789.80	804.60	9.34	9.34
Ascorbic Acid @ 2% w/w + Neem Leaves Powder 5g/plant	T ₄	38.80	33.00	35.90	29.40	24.20	26.80	126.75	120.50	123.63	651.00	630.60	640.80	11.65	11.65
Ascorbic Acid @ 2% w/w + Lantana Leaves Powder 5g/plant	T ₅	42.40	38.20	40.30	33.00	29.80	31.40	141.50	134.00	137.75	687.40	659.20	673.30	11.01	11.01
Ascorbic Acid @ 2% w/w + Parthenium Leaves Powder 5g/plant	T ₆	52.00	46.80	49.40	41.80	37.00	39.40	180.50	175.25	177.88	735.20	718.80	727.00	10.36	10.36
L-arginine @ 2% w/w + Neem Leaves Powder 5g/plant	T ₇	63.60	59.40	61.50	52.40	47.80	50.10	238.25	232.00	235.13	848.00	821.40	834.70	9.30	9.30
L-arginine @ 2% w/w + Lantana Leaves Powder 5g/plant	T ₈	69.00	60.60	64.80	59.00	53.60	56.30	255.00	242.50	248.75	889.20	850.00	869.60	9.04	9.04
L-arginine @ 2% w/w + Parthenium Leaves Powder 5g/plant	T ₉	72.40	68.00	70.20	63.80	60.40	62.10	263.75	255.25	259.50	924.00	895.60	909.80	7.51	7.51
Acephate 75 SP @	T ₁₀	32.20	28.80	30.50	22.60	18.00	20.30	118.25	111.00	114.63	629.40	602.80	616.10	12.30	12.30

1% w/w + Neem cake 5g/plant															
Untreated Check	T ₁₁	90.00	87.40	88.70	79.80	73.20	76.50	289.50	272.50	281.00	1250.40	1231.00	1240.70	6.46	6.7
SEm ±		1.295	1.544	0.986	1.093	1.283	0.825	4.582	5.686	3.553	17.899	22.430	13.945	0.210	0.3
CD at 5 %		3.701	4.414	2.774	3.123	3.666	2.323	13.096	16.252	9.999	51.160	64.111	39.245	0.599	0.9

Data are average value of five replications

Initial Nematode Population: 670 larvae/100 cc soil (2015)

610 larvae/100 cc soil (2016)

Table-2: Per cent change of root-knot nematode parameters and yield of cluster bean through seed treatment with biochemical and soil application with botanicals

Treatments		Galls/plant			Egg masses/plant			Eggs and larvae/egg mass			Final Nematode Population/100 cc soil			Yield (q/ha)	
		I st year	II nd year	Pooled	I st year	II nd year	Pooled	I st year	II nd year	Pooled	I st year	II nd year	Pooled	I st year	II nd year
Salicylic Acid @ 2% w/w + Neem Leaves Powder @ 5g/plant	T ₁	48.89	52.63	50.73	52.13	53.28	52.68	45.34	43.67	44.53	43.33	43.79	43.56	65.79	61.1
Salicylic Acid @ 2% w/w + Lantana Leaves Powder @ 5g/plant	T ₂	39.78	44.39	42.05	44.36	44.54	44.44	30.31	27.34	28.87	38.20	39.85	39.02	53.10	48.1
Salicylic Acid @ 2% w/w + Parthenium Leaves Powder @ 5g/plant	T ₃	32.89	35.93	34.39	38.60	40.71	39.61	22.97	20.64	21.84	34.47	35.84	35.15	44.58	42.1
Ascorbic Acid @ 2% w/w + Neem Leaves Powder @ 5g/plant	T ₄	56.89	62.24	59.53	63.16	66.94	64.97	56.22	97.80	56.00	47.94	48.77	48.35	80.34	75.1
Ascorbic Acid @ 2% w/w + Lantana Leaves Powder @ 5g/plant	T ₅	52.89	56.29	54.57	58.65	59.29	58.95	51.12	50.83	50.98	45.03	46.45	45.73	70.43	67.1
Ascorbic Acid @ 2% w/w + Parthenium Leaves Powder @ 5g/plant	T ₆	42.22	46.45	44.31	47.62	49.45	48.50	37.65	35.69	36.70	41.20	41.61	41.40	60.37	56.1
L-arginine @ 2% w/w + Neem Leaves Powder @ 10g/plant	T ₇	41.51	47.14	30.67	34.34	34.70	34.51	17.70	14.86	16.32	32.18	33.27	32.72	43.96	41.1
L-arginine @ 2% w/w + Lantana Leaves Powder @ 5g/plant	T ₈	23.33	30.66	26.94	26.07	26.78	26.41	11.92	11.01	11.48	28.89	30.95	29.91	39.94	35.1
L-arginine @ 2% w/w + Parthenium Leaves Powder @ 5g/plant	T ₉	19.56	22.20	20.86	20.05	17.49	18.82	8.89	6.33	7.65	26.10	27.25	26.67	16.25	15.1
Acephate 75 SP @ 1% w/w + Neem cake @ 5g/plant	T ₁₀	64.22	67.05	65.61	71.68	75.41	73.46	59.15	59.27	59.21	49.66	51.03	50.34	90.40	85.1
Untreated Check	T ₁₁	-	-	-	-	-	-	-	-	-	-	-	-	-	-