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EVALUATION OF NEEM PLANT PARTS EXTRACT ON EGG HATCHING OF *MELOIDOGYNE INCOGNITA*

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ABSTRACT
Plant parasitic nematode *Meloidogyne incognita* has been recognized as one of the major constraints in vegetable production. Natural products are the alternative approaches to control root knot nematode. An experiment was carried out *in vitro* to test the neem plant parts extract. Different concentration of shade-dried bark, leaves and seeds of neem were tested for egg hatching of *Meloidogyne incognita.* Increase in concentration and exposure period resulted in decreased egg hatching rate of *Meloidogyne incognita.* Neem seed extract (5 ml concentration) proved to be the most effective among used plant parts *viz*. bark, leaves, and seed.

KEY WORDS: *Meloidogyne incognita*, Plant extract, egg hatching.

INTRODUCTION

In plant parasitic nematodes, root-knot nematode *Meloidogyne* species are more damaging one. This genus is considered the most important among plant parasitic nematodes (Sasser and freckmen, 1987). It is widely distributed in India and cause severe damage to vegetable crops. As an alternative to chemical pesticides specially for the purpose of protecting crops against nematode and also for the conservation of biodiversity, botanical may stand as the most promising sources of bioactive products of plant origin. A use of chemicals nematicides to control these nematodes always poses a serious health hazards. The use of botanicals possessing the antifeedant and nematicidal properties, they not only reduce the *nematode* population but also enhance the plant growth (Hussain *et al.*, 2011). Use of neem and neem product has been advocated for the management of root-knot nematode by many workers (Dekha & Rehman, 1998; Jain & Gupta, 1998). Therefore the present investigation was known about the nematicidal effect of neem (*Azadirachta indica*) plant parts aqueous extract on hatching of *M. incognita*.

MATERIALS & METHODS

Preparation of Extracts

The Neem (*Azadirachta indica)* plant parts *viz*. neem bark, neem leaves, neem seeds were shade dried and then oven dry at 60ºC for overnight, powder of plant parts were prepared by using a mixer or blander. Five gm powder of neem bark, neem leaves and neem seeds were soaked in 50 ml water for 2 days.The extract were filtered through four ply muslin cloth and then passed through Whatman filter paper no.1, filtered extract were make up 50 ml with required amount of distilled water and then centrifuged at 4000 rpm for 10 minutes then again filtered through Whatman filter paper no.1. The extract so obtained was put in conical flasks and steam sterilized in an autoclave. The plant part extract *viz*. neem bark, neem leaves and neem seeds designated as stock solution for preparing different dilution *viz*. (5 ml, 2.5ml, 2ml, 1.5ml, 1ml, 0.50 ml, 0.25ml) of each neem plant part extracts were prepared by adding the required amount of distilled water. The sterilized distilled water served as control.

Effect of acetone extracts on juvenile mortality of *M. incognita*

For hatching experiment special PVC tubing were cut in small piece about (1.5 cm diam. and 1.5 cm high). Four plastic legs were attached to elevate each ring which allowed the juveniles to pass through and it also made possible transfer to eggs to fresh test solution every 24 hours. About 5 uniform sized egg masses with an average 250 eggs per egg mass of *M. incognita* were picked from the prepared inoculums. They were transferred to 50ml beaker on PVC tube stand. Seven dilution *viz.* (5 ml, 2.5ml, 2 ml, 1.5 ml, 1 ml, 0.50 ml, 0.25 ml) were prepared from aqueous stock solution of neem plant parts. 5ml of each plant parts extracts were prepared by adding the required amount of distilled water were taken in sterile beaker of 50 ml capacity. Equal numbers of egg mass were transferred to separate beaker containing sterilized water to serve as control. Three replicated were taken completely randomized design (CRD). Mean egg hatching was counted at the intervals of 24, 48, 72, 96, 120 hours after inoculation.

RESULTS & DISCUSSION

Result exhibited that inhibition of hatching of egg was reduced with the increase in concentration of stock solution of used all the neem bark, neem leaves and neem seed and increased exposure period presented in table & fig. (1, 2 & 3). Maximum inhibition of egg hatching was recorded in 5ml concentration and maximum hatching 88.00 per cent recorded at its lowest con. (0.25ml) after 120 hrs of exposure period. Hatching per cent 67.20 was observed in Neem bark extract) with 5 ml. con. at 120 hrs exposure period (Table 1, figure 1). Treatment of neem leaves extract showed complete inhibition of hatching for 48 hrs. but after 48 hrs 15.20% hatching was recorded at 5ml. con. and 120 hrs exposure period and 83.20 per cent

hatching recorded at its lowest con. (0.25ml) after 120 hrs of exposure period (Table 2, figure 2). Neem seed extract on the other hand was found to be the most effective in inhibition of egg hatching (Table 3, figure 3), maximum cent percent inhibition of hatching percent was obtained in neem seed extract having 5ml. con. at exposure just 24 hrs and its lowest con. had 60.80 per cent hatching over untreated control 89.33 per cent after 120 hrs of exposure period (Table 3, Fig. 3). Cumulative percent mortality of *M. incognita* was maximum in neem seed extract and minimum in neem bark extract which could be due to the fact that Neem bark does not possess sufficient active ingredient which are nematicidal. Neem leaves and neem seed having more concentrated nematicidal/nematostatic properties.

TABLE :1 Effect of Neem Bark Extract (aqueous) on egg hatching of *Meloidogyne incognita* at different time intervals (Observations are mean of three replicates)

Extract Con.	% hatching at different exposure period				
	24 hrs.	48 hrs.	72 hrs.	96 hrs.	120 hrs.
0.25 ml	64.000	66.400	72.800	81.600	88.000
0.50 ml	61.600	64.733	72.533	86.000	87.500
1 ml	46.400	55.200	65.600	76.800	80.333
1.5 ml	40.800	51.333	57.600	75.200	80.000
2 ml	39.267	48.000	55.200	64.800	80.000
2.5 ml	35.200	42.200	47.133	63.200	79.200
5 ml	18.467	31.200	34.400	53.600	67.200
Control	64.000	69.333	76.333	88.000	89.333
SEM ±	0.685	0.728	0.864	0.757	1.167
C.D. at 5%	1.579	1.679	1.993	1.746	2.691
C.D. at 1%	2.297	2.442	2.899	2.540	3.916

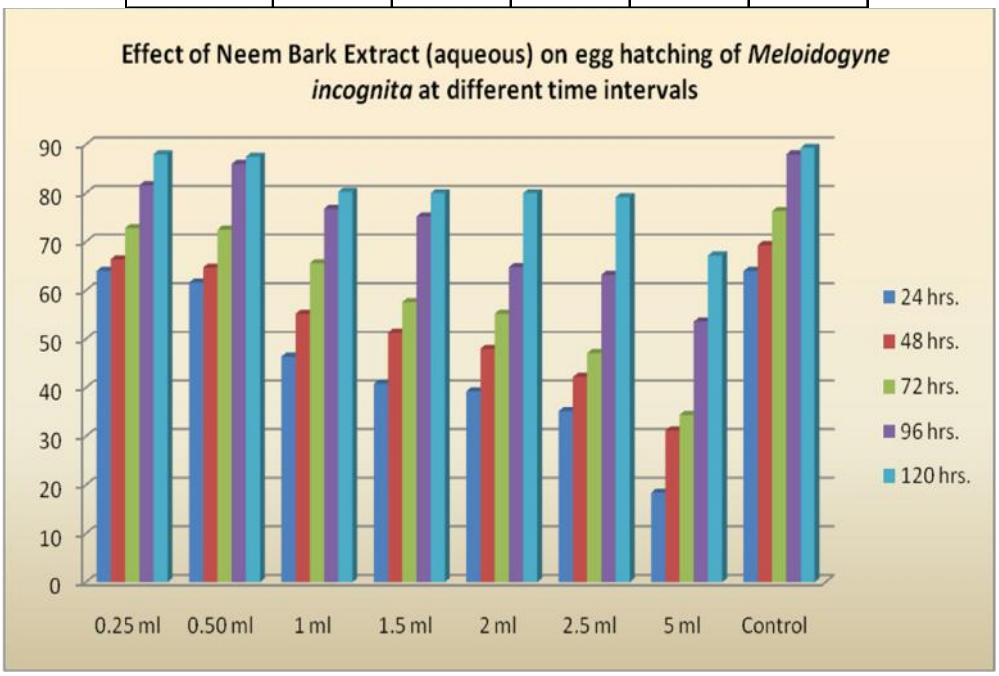

FIGURE : 1

The present investigation is in confirmation with the finding of Gowda and Setty (1979) who observed that hatching of larvae from fresh eggs of *M. incognita* soaked for 24 hrs. in water extract of *A. indica* cake was significantly reduced with 1 : 1 dilution of extract. Neem producte (Neem Kernel, Neem leaf, Neem bark extract) are known to posses nematicidal activity against nematode population (Zaki & Bhatti, 1989, Derkar *et al*., 1990). Krishna and Dabur (2004) tested different con. of shade dried leaves, bark and kernel of neem for their inhibitory role in egg hatching of *M. incognita*. (Haseeb *et al*. (2007) reported that *A. indica* seeds powder were found effective against *M. incognita*. In general with the increase in con. and exposure period, there was reduced egg hatching. Azadirachtin the principal active ingredient from neem seed kernel and a well known nematicidal action. The neem bitters (Teranotriterpinoid) such as epinimbin, sulanin, deacetylsalanin, azadiirachin, and many active principle like nimbidine, thionemone and limonoide from leaves have been reported the be highly nematicidal and nematostatics these neem chemicals are known to act against plant parasitic nematode (Mojumder, 2002). In view of general awareness about hazardous effect of chemical pesticides. This can be further used as non chemical nematode management technologies.

TABLE 2: Effect of Neem Leaf Extract (aqueous) on egg hatching of *Meloidogyne incognita* at different time intervals (Observations are mean of three replicates)

Extract Con.	% hatching at different exposure period				
	24 hrs.	48 hrs.	72 hrs.	96 hrs.	120 hrs.
0.25 ml	54.400	59.600	63.200	70.400	83.200
0.50 ml	49.600	55.200	58.400	67.867	72.000
1 ml	40.800	45.600	54.400	56.800	63.200
1.5 ml	24.000	38.400	35.200	39.200	41.600
2 ml	21.600	29.600	32.800	40.467	38.667
2.5 ml	18.400	24.800	27.200	29.600	32.800
5 ml	0.000	0.000	9.600	11.200	15.200
Control	64.000	69.333	76.333	88.000	89.333
SEM ±	0.927	0.338	0.685	0.682	0.765
C.D. at 5%	2.138	0.780	1.579	1.573	1.765
C.D. at 1%	3.111	1.135	2.297	2.289	2.567

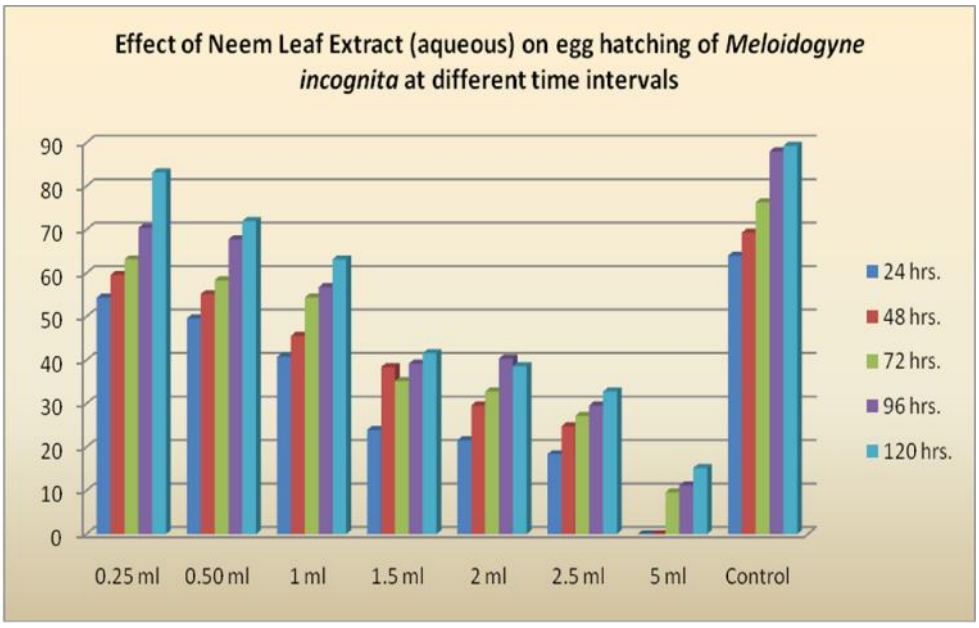

FIGURE: 2

TABLE 3: Effect of Neem Seed Extract (aqueous) on egg hatching of *Meloidogyne inco*gnita at different time intervals

Neem plant parts extract on egg hatching of *Meloidogyne incognita*

(Observations are mean of three replicates)

Extract Con.	% hatching at different exposure period				
	24 hrs.	48 hrs.	72 hrs.	96 hrs.	120 hrs.
0.25 ml	24.800	33.600	39.200	49.600	60.800
0.50 ml	22.400	29.933	33.600	41.600	51.267
1 ml	17.600	23.200	30.400	38.400	46.400
1.5 ml	16.000	20.800	24.000	28.000	32.000
2 ml	12.400	17.600	21.600	24.800	25.000
2.5 ml	9.600	14.400	16.000	17.600	23.400
5 ml	0.000	0.000	0.000	0.000	0.000
Control	64.000	69.333	76.333	88.000	89.333
SEM ±	1.083	0.844	0.845	0.907	0.621
C.D. at 5%	2.498	1.946	1.949	2.092	1.432
C.D. at 1%	3.634	2.831	2.836	3.044	2.083

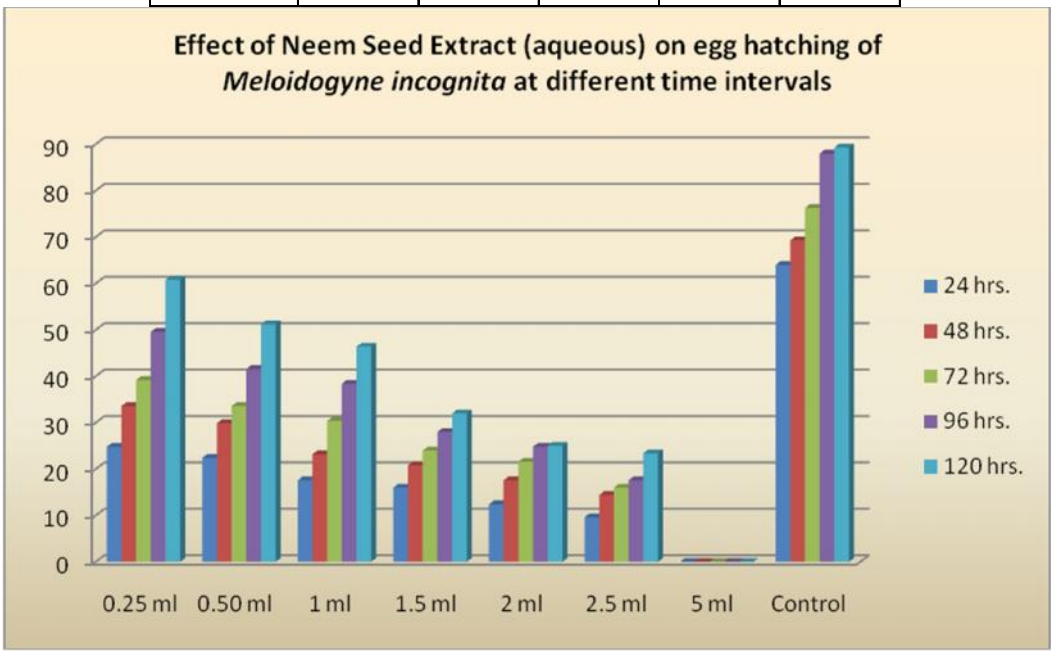

FIGURE: 3

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