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Management of Root-knot Nematode *Meloidogyne incognita* on Mulberry with *Passiflora foetida*

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ABSTRACT

Background: The root knot nematode *Meloidogyne incognita* Chit wood infesting mulberry is economically important in silk industry as it affects the leaf yield both quantitatively and qualitatively which in turn affects the silkworm *Bombyx mori* L., growth and cocoon yield because its foliage is the sole food source of silkworm.

Methods: Hence a pot experiment was conducted for the evaluation of efficacy of eco-friendly formulate like botanical extract from *Passiflora foetida* L., to manage nematode disease severity applied as soil amendment and aqueous leaf extract on nematode egg hatching.

Result: After treatment, nematode infestation reduced significantly in terms of number of root knots/g root weight and nematode egg masses/g root weight in mulberry plants and the treated plants showed better growth in shoot and root length compared to control plants. The aqueous extract of *P. foetida* showed maximum inhibition of egg hatchability with an increase in the concentration of plant extract.

Key words: Egg hatchability, *Meloidogyne incognita,* Mulberry, *Passiflora foetida,* Root knots.

INTRODUCTION

Mulberry (*Morus* sps) is highly valuable plant for sericulture, its foliage constitute the main food source for silk worm *Bombyx mori* L. growth and development. Mulberry is prone to a number of diseases caused by virus, fungi, bacteria, mycoplasma, nematodes and some pests which cause 10-30% loss in the leaf yield and reduce nutritive values of mulberry leaves (Chanturia, 1963, Shree *et al;* 1986, Shree and Umeshkumar, 1991). Among the different diseases of mulberry the significant economic loss is caused by the species of nematode *Meloidogyne incognita* (Kofaid and White) Chit Wood (Chitwood, 1949). This is a very serious problem in sandy loamy soils under irrigated conditions Narayanan *et al,* (1966).

Root-knot nematode (*Meloidogyne incognita*) is important constraints on almost all crops plants in the world and effect not only morphological characters but also impaired in biochemical parameters (Amal *et al.* 2020). Which causes galls or knots on roots of its host plants and other symptoms that can be observed above ground include stunted growth, wilting, chlorosis, leaf curling and reduced vigour of the plant (Saha *et al.* 1983).

Once the root-knot nematode gets established in the mulberry garden it is difficult to control due to perennial nature of mulberry and endoparasitic nature of nematode. Using chemical nematicides to manage root knot nematodes always creates serious problems to environment as well as plants, animals and human health. Plants posses a source of effective pesticidal compounds Mohd Asif *et al*., (2017) and may be regarded as an ultimate source of harmless compounds. They are easily biodegradable with low toxicity to plants and human health. Previous studies suggested that, use of botanicals could reduce root knot gall index and prove to be more effective in improving the plant growth characters (Pavaraj *et al*., 2010; Moosavi, 2012; Muniasamy *et al*., 2010; Nelaballe and Mukkara, 2013). Hence, the study was carried out to know the efficacy of important eco-friendly method to manage the nematode *M. incognita* in mulberry using *Passiflora foetida* L., leaf powder as soil amendment and aqueous leaf extract on nematode egg hatching.

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MATERIALS AND METHODS

The study was carried out in the Department of Sericulture, Sri Padmavati Mahila University, Tirupati, Andhra Pradesh, during the period of 2014-2017 (a part of Ph.D work). *Passiflora foetida* is known to have some nematicidal effects (Vijaya Kumari and Lakshmi Devi, 2013). Hence, *P. foetida* dry leaf powder and aqueous extract of leaf were taken as treatments to know their effect on the infestation capacity of juveniles and nematode egg hatching.

a. Efficacy of *Passiflora foetida* aqueous leaf extract on nematode egg hatching

Preparation of leaf extract

Leaves were collected from the *Passiflora foetida* (Plate 1, iii), plants are washed thoroughly with water and were air dried for one hour. 5 grams of *P. foetida* leaf sample was ground well with the addition of 25 ml of acetone in pestle and mortar. The leaf extract was centrifuged at 3000 rpm per 5 min and filter was allowed for evaporation of acetone. The filtered extract was served as 100% stock or crude extract. From stock solution different dilutions *i.e.*, 25%, 50%, 75% and 100% were prepared by adding necessary volume of sterile distilled water (Plate 1, iv).

Effect of leaf extract on egg hatching

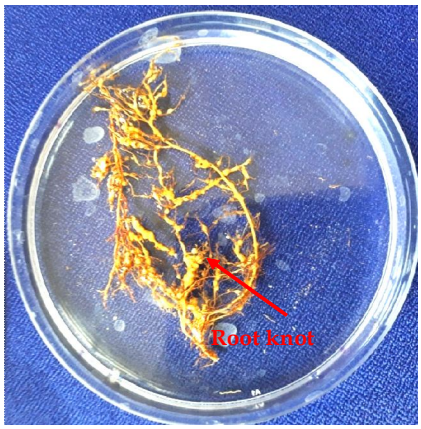
Three uniform sized egg masses of *M. incognita* were picked from infested mulberry roots and were transferred to petri plates containing 5 ml of different concentrations of the leaf extract and made three batches. Egg masses placed in distilled water served as control. Egg hatching was observed in each batch at the intervals of 24 hrs, 48 hrs and 72 hrs.

b. Efficacy of *Passiflora foetida* dry leaf powder as soil amendments on nematode inhibition

For this experiment, mulberry cuttings were planted in pots containing sterilized soil. After three months of establishment of cuttings only one cutting was retained in each pot. Then 1000 juveniles/plant were inoculated as ECL (Economic Threshold Level and Crop Loss) (Govindaiah *et al.*, 1991b; Sharma, 1999a) along with one gram of *Passiflora* leaf powder for treatment. Sixty days after application, the nematode pathogencity was observed by studying the number of root galls/g of root weight, number of egg masses/ g root weight, simultaneously plant growth parameters like shoot and root length were calculated by comparing treated plants with control.

Statistical analysis

All the experimental data collected in three replicates has been subjected to statistical analysis (SPSS-2.0 Version). The analysis of variance has been done to find out the significant differences between the control and infested plants for the T-test and Two way ANOVA test following the procedures laid down in Agricultural statistics (Rangaswamy, 2000).



Nematode infested mulberry roots

Passiflora foetida treated mulberry roots

Passiflora foetida L.

Effect of *Passiflora foetida* leaf extract on nematode egg hatching

Plate 1: Nematode management.

RESULTS AND DISCUSSION

a. Effect of *Passiflora foetida* leaf extract on nematode egg hatching

The egg hatching was recorded in treatments of leaf extraction of *Passiflora foetida* leaf at different concentrations (25%, 50%, 75% and 100%) in different periodical intervals (24, 48 and 72 hours). The results are presented in the Table 1 (Fig 1).

i. At 24 hours

The number of nematode juveniles hatched at 25% concentration was 9.33, whereas in control the number of nematode juveniles hatched was 69.33. The percentage of hatching inhibition was 86.54 over the control. In 50% concentration the number of nematode juveniles hatched was observed as 6.00, while in control it was 69.33. The percentage of hatching inhibition was 91.34 over the control.

In 75% concentration the number of nematode juveniles hatched was observed as 5.66, while in control it was 69.33. The percentage of hatching inhibition was 91.83 over the control. The number of nematode juveniles hatched in 100% concentration was recorded as 3.66 and in control it was 69.33. The percentage of hatching inhibition was 94.72 over the control.

ii. At 48 hours

The number of nematode juveniles hatched in 25% concentration was observed as 13.00, whereas in case of control it was 75.00. The percentage of egg hatching inhibition was 82.66 over the control. In 50% concentration

Table 1: Efficacy of *Passiflora foetida* leaf extraction on nematode egg hatching.

Duration (hrs)	% of leaf concentration	Nematode egg hatching		% of decrease over control
		Control (Distilled water)	Treated	
1st batch after 24 hrs	25%	69.33	9.33	86.54
	50%	69.33	6.00	91.34
	75%	69.33	5.66	91.83
	100%	69.33	3.66	94.72
2nd batch after 48 hrs	25%	75.00	13.00	82.66
	50%	75.00	10.00	86.66
	75%	75.00	6.33	91.56
	100%	75.00	5.33	92.89
3rd batch after 72 hrs	25%	90.67	17.66	80.52
	50%	90.67	12.66	86.03
	75%	90.67	8.00	91.17
	100%	90.67	7.00	92.27

Anova Table

Source	Type III sum of squares	df	Mean square	F	Sig.
Time	540.717	2	270.359	160715.201	0.000
Group	35230.982	4	8807.746	5235780.063	0.000
Time * Group	389.747	8	48.718	28960.759	0.000
Error	0.05	30	0.002		

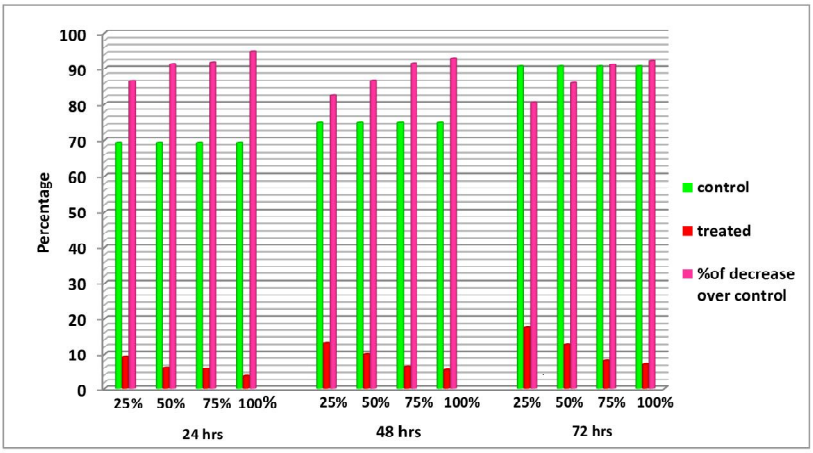


Fig 1: Nematode egg hatching treated with *Passiflora foetida* leaf extract.

the number of nematode juveniles hatched was found as 10.00, but in control it was 75.00. The percentage of egg hatching inhibition was 86.66 over the control.

In 75% concentration the number of nematode juveniles hatched was recorded as 6.33, against the control which was 75.00. The percentage of egg hatching inhibition was 91.56 over the control. The number of nematode juveniles hatched in 100% concentration was observed as 5.33 and in control it was 75.00. The percentage of hatching inhibition was 92.89 over the control.

iii. At 72 hours

The number of nematode juveniles hatched in 25% concentration was observed as 17.66, however in control it was 90.67. The percentage of hatching inhibition was 80.52 over the control. In 50% concentration the number of nematode juveniles hatched was recorded as 12.66 and in the control it was 90.67. The percentage of hatching inhibition was 86.03 over the control.

In 75% concentration the number of nematode juveniles hatched was observed as 8.00, whereas in the control it was 90.67.The percentage of hatching inhibition was 91.17 over the control. The number of nematode juveniles hatched in 100% concentration was showed as 7.00, while in the control it was 90.67.The percentage of hatching inhibition was 92.27 over the control.

In the present study it was revealed that the leaf extract concentrations and exposure time significantly affected nematode hatching. The inhibitory effect of the extracts could be due to the presence of toxic compounds in the leaf extracts that might be responsible for nematicidal effect.

b. Efficacy of *Passiflora foetida* dry leaf powder as soil amendments on nematode inhibition

The results pertaining to the nematode pathogenicity parameters *viz.* number of root galls/g root weight, number of egg masses/g root weight and plant morphological parameters *viz.* shoot length and root length in control (infested) and treated plants were recorded and presented in the Table 2 (Fig 2).

1. Nematode pathogenicity parameters

i. Number of root galls/g root weight

The number of root galls/ g root weight recorded in the control mulberry plants was 68.33 and in the *Passiflora foetida* treated plants, it was observed as 13.33 with a percentage of reduction 80.49 over control (Plate 1, i and ii).

ii. Number of egg masses/g root weight

The number of egg masses/g root weight in *Passiflora foetida* treated plants was 10.33 compared to control plants wherein it was 59.67 with a percentage of reduction was 82.68.

2. Morphological parameters

i. Shoot length (cm)

The shoot length of control mulberry plants was observed as 54.50 cm whereas it was 58.02 cm in plants treated with *Passiflora foetida* with a percentage of increased 6.45 over control.

Table 2: Efficacy of *Passiflora foetida* dry leaf powder as soil amendments on nematode inhibition.

Parameters	Control (Infested)	Treated (*Passiflora foetida*)	% of decrease over control	T-Test	p-value	Level of significance
No. of root knots/g of root weight	68.33	13.33	80.49	1868.257	0.000	**
No. of egg masses/g of root weight	59.67	10.33	82.68	285.819	0.000	**
			% of increase over control			
Shoot length (cm)	54.50	58.02	6.45	84.548	0.001	**
Root length (cm)	25.33	34.08	34.54	200.307	0.000	**

Note: p<0.01=**Significant at 0.01 level.

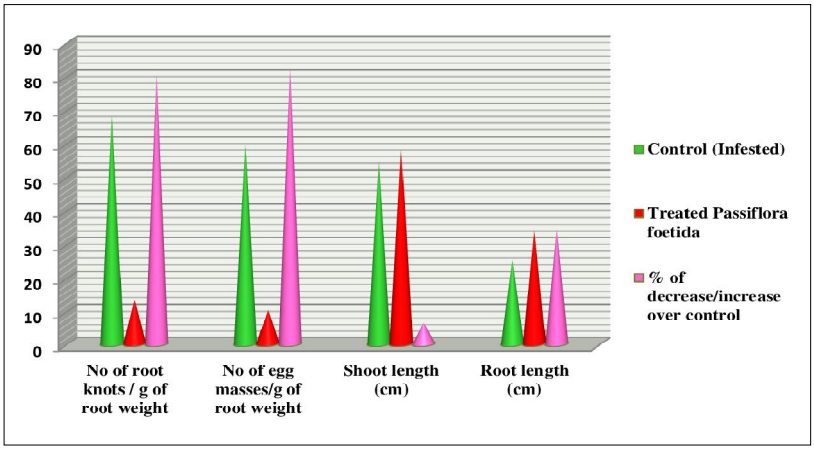


Fig 2: Efficacy of *Passiflora foetida* dry leaf powder as soil amendments on nematode inhibition.

ii. Root length (cm)

In case of control mulberry plants, the root length was observed as 25.33 cm and in *Passiflora foetida* treated plants it was 34.08 cm with a percentage of increased 34.54 over control.

In the present study, *Passiflora foetida* leaf powder reduced nematode infestation considerably and the plant growth parameters were increased. *Passiflora foetida* leaf powder was used as soil amendment to control root knot nematode in mulberry. It was found that maximum percentage of reductions was observed in root knots/galls (80.49) and in egg masses (82.68) per gram root weight when compared to nematode infested plants.

Passiflora foetida constitute the major bioactive phytochemicals like passifloricins, polyketide, alpha-pyrones, alkaloids, phenols, glycosides, flavonoids and cyanogenic compounds (Echeverri *et al.,* 2001 and Dhawan *et al.,* 2004). The *Passiflora foetida* leaf powder during the process of decomposition in the soil releases some biochemical metabolites which are known to have nematicidal properties and contributing in reducing the nematode pathogenicity and accelerated rapid root and shoot growth development and overall plant growth consequently help the plants to develop resistance against nematode attack.

CONCLUSION

Root knot disease caused by nematode *Meloidogyne incognita* is a most serious problem in mulberry and is found in almost all sericulture practicing countries. Infestation of root knot nematodes reduces the yield and quality of leaves which in turn affects the silk worm growth and quality cocoon production. And finally it leads to economic loss in silk farmers. Use of synthetic nematicides to manage nematodes, cause very harmful effects on human health and environment. Therefore, it is better to the use of cheap, eco-friendly and harmless methods of plant extracts and products. The *Passiflora foetida* leaf powder has nematicidal properties, contribute in reducing the nematode pathogenicity.

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