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EFFECT OF CITRUS SINENSIS L. FRUIT PEEL EXTRACT ON IN-VITRO PHYTO-NEMATOCIDAL ACTIVITY ON MELOIDOGYNE INCOGNITA RACE II

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Abstract: Brinjal (*Solanum melongena* Linn) is the fourth most important vegetable cultivated after potato, onion, and tomato in Maharashtra. These crops are grown in all the seasons by small and marginal farmers who get the source of income. The production of brinjal faces a number of problems which causes enormous yield losses. Among them, the *Meloidogyne incognita* Race II root-knot nematode is major pest it reduces significant yield losses up to 30-35%. For managing nematode population infection farmers depends on mainly on the chemical nematicides. The continuous excessive and indiscriminate use of chemical nematicides causing multiple side-effects that include pesticide residues, environmental pollution, and destruction of natural enemies of pests, the resurgence of pest population and increased cost of cultivation. To overcome the above-mentioned side effects. The ability of the treatment schedule of peel extracts *Citrus sinensis* L. were tested against Root-knot nematode *Meloidogyne incognita* Race-II associated with Brinjal (*Solanum melongena* L.) by *In-vitro* method. It causes 62.2% larval mortality in 24 hrs. The *Citrus sinensis* L. were highly active phytonematicide as compared with the control standard neem cake and Carbofuran.

Keywords: words: Brinjal (*Solanum melongena* L.), Carbofuran., *Citrus sinensis* L., *In-vitro* nematicidal activity and Neem cake

I INTRODUCTION

Brinjal or eggplant (*Solanum melongena* L.) belongs to the *solanaceous* family, It is most popular vegetable crop grown in the subtropical and tropical regions in all the seasons so it is called as king of vegetables. It is preferred by both vegetarians as well as non-vegetarians to its nutritive and medicinal values. In India immature fruits are consumed as cooked vegetable in various forms (Rai et al., 1995). Fruits are used in daily cooking curry, roasted, fried, and pickled. The fruits are to be a rich source of ascorbic acid and phenolic (Vinson et al., 1998). It has also used in some of disease preventions haemorrhoids; reduce the blood cholesterol levels, ulcerations, diabetes, asthma, cholera, bronchitis, dysuria, otitis, toothache (Sutarno et al., 1993). One hundred grams of brinjal contains 40gm carbohydrate, 1.4gm proteins, 0.3gm minerals, (including phosphorus 47mg, calcium 18mg, potassium 2.0mg and iron 0.9mg) and vitamins A, B and C (Arycord, 1983).

The area under brinjal cultivation in India is 711.3 thousand hectares with estimated annual production of

13557.8 thousand metric tons with productivity of 19.1 metric tons per hectare. In Maharashtra, brinjal is grown in an area of 0.02 m hectare, with an annual production of 0.69 metric tons and productivity of 23 metric tons the production is very less due to infection (Arycord, 1983).

II MATERIALS AND METHODS

Collection of nematode from infected soil sample

Soil sample from infected fields of tomato, brinjal, and okra of 1 kg soil collected out of 200 cc soils was washed thoroughly and processed using Cobb's sieving and Decanting method (Cobbs, 1918). Followed by modified Baermann's funnel methods. It was used to study seasonal incidence, prevalence of *M. incognita* race II.

Identification of root-knot nematode species

Meloidogyne incognita race II Species was identified on the basis of perennial pattern method described by (Eisenbackett et al., 1981). The species of *Meloidogyne incognita* race II were identified by Dr. Hole U. B. and Dr. Kadm D. B. Nematology section Entomology Department M.P.K.V. Rahuri (M.S.) India.

Taxonomic Identification of Plants

The collected plants were authenticated from plant taxonomist at a Department of Botany, D. B. F. Dayanand College of Arts and Science Solapur, and the selected plants Herbarium sheets were deposited in the Department of Zoology, D.B.F. Dayanand College of Arts and Science, Solapur.

Fractions separated by Column chromatography

Extracts obtained were mixed with silica gel (60-120 mesh) and eluted by using Hexane and Acetone. By using column chromatography techniques different fractions A, B, C, D, E and F were collected (Dama, 1998). Eluted bioactive

fractions were further used for *in-vitro* phyto-nematicidal activity.

***In-vitro* phyto-nematicidal testing method**

In vitro phyto-inematicidal activity of the plant material were used method described (Dama et al., 1999; Dama, 2002).

Statistical Analysis

Data were statistically analyzed by using Standard Statistical Methods (SAS, 1992; 1999). The present work consists of mean, SD, p-value and t-test and correlation coefficient. Observations were made for death time.

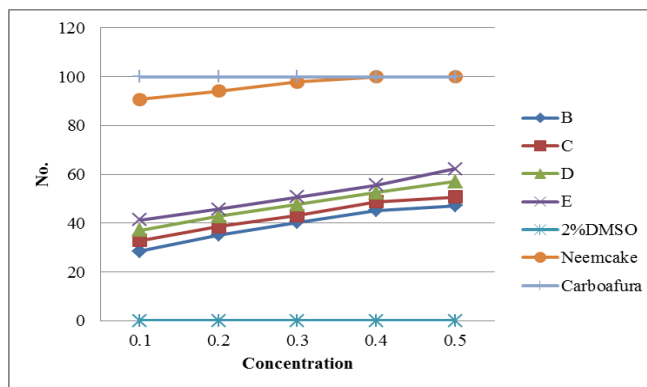
III RESULT AND DISCUSSION

Table 1. Shows the Mean ± SD values of the effect of *Citrus sinensis*L. plant extract, fraction B, C, D and E with control 2% DMSO, Neemcake and Carboafuran on mortality of *M. incognita* race II associated with brinjal at 24 hrs

Fractions and Standard Conc. mg/ml of 2% DMSO	Fraction B	Fraction C	Fraction D	Fraction E	2% DMSO	Neemcake	Carboafuran
	Mean± SD	Mean± SD	Mean± SD	Mean± SD	Mean± SD	Mean± SD	Mean± SD
0.1	28.6± 0.5	33.0± 0.7	37.2± 0.4	41.4± 0.9	0.0± 0.0	90.8± 0.4	100.0± 0.0
0.2	35.2± 0.4	38.6± 0.5	42.8± 0.4	45.8± 0.4	0.0± 0.0	94.2± 0.4	100.0± 0.0
0.3	40.4± 0.5	43.2± 0.4	47.6± 0.5	50.8± 0.4	0.0± 0.0	97.8± 0.4	100.0± 0.0
0.4	45.2± 0.8	48.8± 0.4	52.6± 0.5	55.6± 0.5	0.0± 0.0	100.0± 0.0	100.0± 0.0
0.5	47.2± 0.4	50.8± 0.4	57.0± 0.7	62.2± 0.4	0.0± 0.0	100.0± 0.0	100.0± 0.0

Table 1. Shows the methanolic peel extracts of *Citrus sinensis*L. shows phyto-nematicidal activity in concentration dependent manner. Among all the grades the least mortality were recorded in fraction B 47.2 % mortality and highest in Fraction E 62.2% mortality, neem cake and carbofuran show 100% mortality in 24 hrs.

Statistically average values of immobilized *M. incognita* race II treated with *Citrus sinensis* L. Shows in Table 1, in B fraction, C fraction, D fraction and E fraction are significantly more than 2% DMSO positive control and significantly less than Neemcake and Carboafuran negative control at 0.1, 0.2, 0.3, 0.4 and 0.5 concentrations (P<0.01) at 24 hrs.



Graph 1. Shows the immobilized *M. incognita* race II of *Citrus sinensis*L.at 24 hr.

Table 2. Comparison of values of immobilized *M. incognita* race II, treated with *Citrus sinensis* L. fraction B, C, D, E, with Control 2% DMSO, Neemcake and Carboafuran at 24 hrs.

Fraction	Fraction and Standard Conc. mg/ml of 2% DMSO	2% DMSO		Neemcake		Carboafuran	
		t test	P value	t test	P value	t test	P Value
B	0.1	104.43	P<0.01	-175.93	P<0.01	-260.72	P<0.01
	0.2	157.42	P<0.01	-186.57	P<0.01	-289.79	P<0.01
	0.3	147.52	P<0.01	-162.35	P<0.01	-217.63	P<0.01
	0.4	108.05	P<0.01	-131.00	P<0.01	-131.00	P<0.01
	0.5	211.08	P<0.01	-236.13	P<0.01	-236.13	P<0.01
C	0.1	93.34	P<0.01	-138.17	P<0.01	-189.50	P<0.01
	0.2	140.95	P<0.01	-157.26	P<0.01	-224.20	P<0.01
	0.3	193.20	P<0.01	-172.66	P<0.01	-254.02	P<0.01
	0.4	218.24	P<0.01	-228.97	P<0.01	-228.97	P<0.01
	0.5	227.18	P<0.01	-220.03	P<0.01	-220.03	P<0.01
D	0.1	166.36	P<0.01	-169.50	P<0.01	-280.85	P<0.01
	0.2	191.41	P<0.01	-162.54	P<0.01	-255.81	P<0.01
	0.3	173.81	P<0.01	-141.99	P<0.01	-191.34	P<0.01
	0.4	192.07	P<0.01	-173.08	P<0.01	-173.08	P<0.01
	0.5	161.22	P<0.01	-121.62	P<0.01	-121.62	P<0.01
E	0.1	92.57	P<0.01	-98.80	P<0.01	-131.03	P<0.01
	0.2	204.82	P<0.01	-153.05	P<0.01	-242.39	P<0.01
	0.3	227.18	P<0.01	-148.63	P<0.01	-220.03	P<0.01
	0.4	203.02	P<0.01	-162.13	P<0.01	-162.13	P<0.01
	0.5	278.17	P<0.01	-169.05	P<0.01	-169.05	P<0.01

IV DISCUSSION

Plant produces secondary metabolites allelochemicals compounds it is toxins to organisms that acts as attractants or deterrents (Dodds, 1996; Brown and Morra, 1997).

In the present study *Citrus sinensis* L. peel extracts shows 62% and 68 % juvenile larval mortality in 24 hr. and 48 hr. respectively and the present research findings, *Citrus sinensis* L. fractions shows the very high phyto-nematicidal activity as compared to findings of other researcher (Sellami and Moufarrah, 1994). This observation agrees with those of earlier researchers. The findings from the study corroborate those of earlier researchers like Kumar and Singh (1972), Pandey (1990), Mani and Al Hinai (1998), reported the toxicity of various plant extracts used in their experiments. Essential oils peel extracts inhibits egg hatching, migration and mortality of *Meloidogyne incognita* (Ibrahim et al., 2006).

V CONCLUSION

botanicals plants *Citrus sinensis* L. peel extracts significantly increases larval mortality in second juveniles of *M. incognita in vitro*. In 24 h it shows 62 % larval mortality, increase time period it mortality rate also increases. These findings are significant in the identification and development new alternative approaches in controlling root-knot nematode populations. Bio-pesticides isolated from natural plants to replace the synthetic hazardous and expensive chemicals used at present.

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