

## EFFECT OF CERTAIN MEDICINAL PLANT EXTRACTS AND ESSENTIAL OIL FOR CONTROLLING THE ROOT-KNOT NEMATODE *MELOIDOGYNE JAVANICA* ON POTATO PLANTS

Bekhiet, M.A

Nematology Dept., Plant Pathology Institute, ARC, Giza, Egypt.

Accepted 30 / 3 / 2004

**ABSTRACT :** Water extracts of two medicinal plants namely : the mint (*Mentha microphylla*) and holywormwood (*Artemisia santonicum*) as well as kamphor leaf oil (*Eucalyptus globolus*) were tested for controlling the root-knot nematode *Meloidogyne javanica* on potato plants cv. Nicole under greenhouse condition during spring season 2002 and open field conditions during spring season 2003. Results revealed that kamphor leaf oil and holywormwood extract gave the highest reduction in number of galls and nematode population and increased plant growth. While the least reduction of galling and nematode population was occurred with the mint treatment increasing also the plant growth. Kamphor leaf oil greatly reduced numbers of galls and nematode population with all tested concentrations, but one concentration (1%) of kamphor leaf oil improved plant growth parameters, whereas the other concentrations appeared to be highly toxic for plant growth. Vydate 10%G gave the best results in this respect in comparison with all other treatments.

**Key words:** Medicinal, plants, leaf water extracts, essential oil, root-knot nematode, *Meloidogyne javanica*, potato, cv. Nicola, Vydate 10% G.

### INTRODUCTION

Several plants were recorded as antagonistic for phytoparasitic nematodes. This practice has many advantages, i.e.,

sheep, avoid soil pollution with chemicals, don't have any hazardous residual effects, safe and non toxic to plants, animals and human health, and very easily apply. The inhibitory effect of

kamphor on development and reproduction of some nematode species root-knot nematodes *Meloidogyne incognita* and *M.javanica* as well as the reniform nematode *Rotylenchulus reniformis* was demonstrated by many investigators (Mani *et al.*, 1986; Akhtar and Alam, 1989; Montasser, 1991; Akhater and Mahmood, 1993; El-Naggar *et al.*, 1993; Farahat *et al.*, 1994; Abadir *et al.*, 1996; Shawky-Samaa 2001; and Robin *et al.*, 2001; on the other hand Shawky-Samaa 2001; reported that kamphor *Eucalyptus* dry leaf implement gave very good results in increase and improve the yield of fruits of olive tree. Whereas the use of mint either as plant extract or intercropping plant had been demonstrated to be effective against root-knot nematode reproduction, (Singh *et al.*, 1955; Ibrahim *et al.*, 1986 Mahmood *et al.*, 1982; Bettini 1993; and Ali *et al.*, 1998). Hammad-Eman 2003; demonestrated that holywormwood extract was effective against root-knot nematodes *M.incognita* and *M.javanica*. Ali *et al.*, 1998. cited that the effect of the nematicide Vydate was significantly higher than the effect of antagonistic plants to reduce egg-masses on

tomato plants, also Shawky-Samaa 2001; reported that Vydate gave significant results to reduce nematode reproduction and root gall endix. This research aimed to study the effect of some medicinal plant extracts and essential oil, on the root-knot nematode *M.javanica* and potato plant growth under greenhouse and field conditions.

## MATERIALS AND METHODS

### 1) Greenhouse Experiment:-

Two medicinal plant extracts (holywormwood and mint) and essential oil (kamphor) were tested for controlling the root-knot nematode *M.javainca* parasitizing potato plants under greenhouse condition during spring season 2002. One sprout of potato tuber cv. Nicola was planted in 20cm diam, clay pots containing mixture of clay and sand soils (2:1 w/w). After two weeks, the potato seedlings were inoculated with 3000 newly hatched juveniles (J2) of *M.javanica* for each plant. Each treatment was replicated three times. All medicinal plant extracts and essential oil were added weekly as soil drench at the concentration of 1, 2 and 3% for

kamphor leaf oil, while 2,4 and 6 gram by percentage 2, 4 and 6% for mint extract and holywormwood extract. All amendments were added after inoculation directly. Three inoculated pots were left without adding any extracts and served as control. Pots were kept in greenhouse at  $25\pm 5^{\circ}\text{C}$  in randomized block design. Fifty days after inoculation, the plants were harvested. Data on plant growth, nematode population in the soil and roots, and number of galls were counted and recorded.

## II) Open Field Experiment :-

A heavy infested sandy soil area with *M.javanica* was selected to carry out this open field experiment during spring season 2003 in Nobaria, Behera governorate. The best medicinal plant extract and essential oil were added weekly as soil drench two weeks after planting for three times by concentration 1, 2 and 3% for holywormwood extract, whereas 0.25, 0.50 and 1.0% for kamphor leaf oil alone. Combination of kamphor leaf oil and holywormwood extract (1:1) as well as one nematicide, oxamyl Vydate 10%G by rate 0.25, 0.50,

1.0 g per plant were applied. The nematicide which was added for one time directly before planting. Three microplots were chosen for each treatment including the check without any addition. Each microplot includes sixty potato plant in three rows. Each row was treated by one concentration. Three months and twenty days after planting, all plants in each microplot were harvested, and the number of juveniles in 250g soil was counted by means of Oostenbrink elutration (Goody, 1963). The number of galls and egg masses per root and number of eggs per egg mass were counted for each treatment, and comparison with the check. The root of each plant was stained in lactophenol acid fuchsin (Goody, 1957). On the other hand tuber yield was counted and calculated to compare with the check. Data were subjected to statistical analysis using F test and means were compared by Duncan's multiple-range test (Duncen, 1955).

## RESULTS AND DISCUSSION

### I) Greenhouse Experiment :-

Two medicinal plant

Extracts (holywormwood and mint) and one leaf oil (kamphor) were evaluated for their efficacy to control *M.javanica* on potato. Data presented in Table (1) revealed that the tested materials were effective in reducing the nematode galling on the root system and inhibited the nematode reproduction on potatoes. The adverse effect of such treatments was increased as the dosage increased. The highest concentration of kamphor leaf oil extract was highly toxic for plant growth, ( Table 2), generally showed that kamphor gave the best results when compared with other treatments, the highest reduction in root galling and juveniles in soil as well as numbers of egg-masses per root were associated with treatments of kamphor leaf oil extract (Table 1). While moderate reduction obtained with the plants treated by holywormwood extract. However least reduction was occurred with the treatment of mint extract. Data in (Table 2) revealed that the increment in fresh weight of both shoot and root of potato plants was varied greatly with the type of treatment. Such improvement in the plant growth of fresh weight was correlated positively with the increase in dosage of holywormwood extract.

The least increment was noticed by mint extract (Table 2).

## II) Open Field Experiment :-

Data in (Table 3) show results of the open field treatments of the medicinal plant extract of *Artemisia* and essential oil of *Eucalyptus* applied alone and combined for controlling *M.javanica* on potato plants as compared with the nematicide Vydate 10%. Data revealed that all treatments gave a positive effect in suppressing root galls and egg masses on the root system of potato plants. Single application of kamphor leaf oil extract gave the highest reduction of root-galls, egg-masses and number of eggs/egg mass, followed by the mixture of kamphor leaf oil and holywormwood extracts and holywormwood alone. While the nematicide Vydate gave best results when compared with all treatments in reducing the rate of nematode reproduction. Data in (Table 3) showed the effect of kamphor leaf oil and holywormwood extract alone and combined under field conditions. It was found that there were many differences between field and greenhouse applications.

Table (1): Effect of some medicinal plant extracts and essential oil as soil drench on the reproduction of the root-knot nematode *Meloidogyne javanica* infected potato cv. Nicola during spring season 2002 under greenhouse conditions.

Treatments	Amount added %	No. of galls/root	Nematode population				Nematode final population (Pf)	Rate of nematode reproduction (Pf/Pi)
			Juveniles in soil/pot	Nematode developmental stages/root	No. of egg masses/root	No. of eggs-egg mass		
<i>Eucalyptus globolus</i> (kamphor)	1	30 D	80	21 BC	15 FG	50 F	851	0.283
	2	0 F	0	0 F	0 G	0 G	0.00	0.00
	3	0 F	0	0 F	0 G	0 G	0.00	0.00
<i>Artemisia santonium</i> (holy wormwood)	2	37 CD	187	16 CD	51 D	242 C	12545	4.181
	4	18 E	160	6 EF	23 EF	220 CD	5226	1.742
	6	12 E	60	5 EF	14 FG	209 DE	2991	0.997
<i>Mentha microphylla</i> (mint)	2	119 B	820	34 B	143 B	390 A	56624	18.874
	4	68 C	440	27 BC	69 C	370 A	25997	8.665
	6	41 CD	287	17 CD	65 CD	360 A	23704	7.901
check	0	253 A	2533	148 A	158 A	369 A	60983	20.327

Values in columns followed by the same letter(s) are not significantly different ( $P=0.05$ ) according to Duncan's multiple-range test.

Table (2): Effect of some medicinal plant extracts and essential oil as soil drench application on the growth of potato cv. Nicola infected with the root-knot nematode *Meloidogyne javanica* during spring season 2002 under greenhouse condition.

Plant growth Treatments	Amount added %	Fresh weight in (g)			
		Shoot	Increase %	Root	Increase %
<i>Eucalyptus globolus</i> (kamphor)	1	33.67 BC	65.61	26.33 A	71.75
	2	26.33 D	29.51	19.00 CDE	23.93
	3	00.00 F	00.00	00.00 G	00.00
<i>Artemisia sontonicum</i> (holywormwood)	2	25.67 D	26.26	16.00 F	4.37
	4	26.00 D	27.88	18.33 DEF	19.56
	6	27.00 CD	32.80	19.33 CDE	26.09
<i>Mentha microphylla</i> (mint)	2	32.00 BCD	57.40	18.33 CDEF	19.65
	4	34.67 AB	67.24	21.67 BC	41.35
	6	35.67 AB	75.45	24.00 AB	56.55
Check	0	20.33 E	0	15.33 F	

Values in columns followed by the same letter(s) are not significantly different ( $P=0.05$ ) according to Duncan's multiple-range test.

Concerning the tuber yield of potato, data in (Table 4) revealed that all treatments under investigation increased the tuber yield. Vydate and kamphor leaf oil had the highest effect to increase the yield, followed by the combination of kamphor leaf oil and holywormwood extract. It has been also observed that during planting in spring season the multiplication rate of *M. javanica* increased and subsequently the population density also increased, which gave the chance for the third generation of nematode to infect tubers at the end of plant age. The effect of *Eucalyptus globolus* leaf oil and *Artemisia santonicum* extract under greenhouse condition was more sharp and obvious in greenhouse than under the field conditions.

From the above mentioned data, it can be concluded that the antagonistic effect of the kamphor leaf oil, holywormwood (*Artemisia santonicum*), and mint (*Mentha microphylla*) against *Meloidogyne javanica* inhibit the nematode infection (No. of galling) and reproduction. This inhibition action may be attributed to the accumulation of toxic compounds which produced during the decomposition of such materials in

the soil (Alam *et al.*, 1978; Siddiqi and Alam, 1988). Such materials can also encourage the microbial activities and may generate suppressive soil against plant-parasitic nematodes (Kloepper *et al.*, 1991, 1992). Also the naturally nematicidal compounds involved in such extracts undoubtedly suppressed the nematode build up in the soil. Mohamed-Bassyonia, 2001 and Shawky-Samaa, 2001 reported that adding of kamphor leaf as green matter into the soil gave a very good results in reducing number of galls as well as the reproduction factor of root-knot nematode on plants. Ali *et al.*, 1998 cited that mint (*Mentha spicata*) was reported as resistant crop against *Meloidogyne javanica* they reported also that mint as intercrop with tomato plants reduced the infection of tomato with root-knot nematode. Reynolds, 1982 cited that mint contain a.i called carvone ( $C_{10}H_{14}O$ ), which has nematicidal effect. To concern holywormwood (*Artemisia santonicum*) many investigators demonstrated nematicidal effect of plants related to the same genus as *Artimisia cinae* and *Artimisia judaica*. Ali *et al.*, 1997 reported that the species extracts of *Artemisia judaica* and

Table (3): Numbers of root galls, egg masses and eggs per egg mass as affected by some medicinal plant extracts and essential oil on potato cv. Nicola Infected with *M.Javanica* in open field during spring season 2003.

Treatments	Amount added %	No. of galls/root	No. of egg masses/root	No. of egg/egg mass	% root galls reduction	% egg masses reduction	% egg-eggmasses reduction
<i>Eucalyptus globolus</i> (kamphor)	0.25	65	50	216	81.42	71.42	34.00
	0.50	53	36	192	85.00	79.42	41.00
	1.00	34	24	169	90.28	86.28	48.00
<i>Artemisia santonium</i> (holy wormwood)	1	112	89	280	68.00	49.14	14.00
	2	93	74	240	73.43	58.00	26.15
	3	81	63	230	77.00	64.00	32.30
<i>Artemisia and Eucalyptus</i>	0.25	59	38	205	83.14	78.28	37.00
	0.50	30	21	186	91.42	88.00	43.00
	1.00	23	14	160	93.42	92.00	51.00
Vydate 10%G	0.25 G	35	39	120	90.00	77.71	69.23
	0.50 G	17	28	90	95.14	84.00	76.92
	1.00 G	16	20	60	95.42	88.57	84.61
Check	0	350	175	325	0	0	0

$$\text{Reduction \%} = \frac{\text{No of galls (egg masses) or eggs/egg mass (treated)}}{\text{No of galls (egg masses) or eggs/egg mass (check)}} \times 100 = \text{Resultant} - 100 = \text{Reduction\%}$$

Table (4) : Effect of some medicinal plant on the root-knot nematode *Meloidogyne javanica* infected potato c.v Nicola and yield during spring season 2003 in open field condation.

Tuber yield Treatments	Amount added %	Total weight for tubers (g)	Yield for tubers as treated weight/plant		%Tuber	
			Infested	Healthy	% Infested	% Healthy
<i>Eucalyptus globolus</i> (kamphor)	0.25	0.816	0.183	0.633	22.43	77.57
	0.50	0.880	0.080	0.800	9.10	90.90
	1.00	0.740	0.040	0.700	5.40	94.60
<i>Artemisia sontonicum</i> (holywormwood)	1.0	0.899	0.366	0.533	40.71	59.29
	2.0	0.968	0.316	0.652	32.64	67.36
	3.0	0.800	0.200	0.600	25.00	75.00
<i>Artemisia and Eucalyptus</i>	0.25	0.803	0.233	0.570	29.01	71.00
	0.50	0.866	0.166	0.700	19.16	80.84
	1.0	0.870	0.070	0.800	8.04	91.96
Vydate 10%G	1 g	0.652	0.130	0.522	19.93	80.07
	2 g	0.855	0.103	0.752	12.04	87.96
	3 g	0.833	0.000	0.833	00.00	100.00
Check	0	0.660	0.500	0.160	68.00	32.00

$$\% \text{ Infested} = \frac{\text{healthy}}{\text{total}} \times 100 = \text{resultant} - 100.$$

$$\% \text{ Healthy} = \frac{\text{infested}}{\text{total}} \times 100 = \text{resultant} - 100.$$

*Artemisia cinae* highly affected the rate of egg-hatching of *Meloidogyne hapla*, as well as ability of newly hatched larvae to cause galling on the plants. Data also revealed that the highest dosage of kamphor had a phytotoxicity on potato plants. These results can be explained by the findings of Abadir *et al.*, 1994 (who reported that phytotoxicity was obviously noticed with *Oryza sativa* or *Triticum aestivum* especially at the higher doses). This results of phytotoxicity may be due to the accumulation of toxic compounds in the soil as a result of the decomposition of such materials. To concern the differences between the antagonistic effect of the two extracts of kamphor leaf oil *Artemisia santonicum* in greenhouse and under field conditions it has been observed that the effect was more sharp with greenhouse than under field conditions. These results can be explained as a result of the delusion effect in the soil under field conditions moreover the antagonistic effect of the soil microorganisms which may be delute the drastic effect of extracts during the dicomposing of the toxic material in such extracts.

From the above mentioned data it can be concluded that such extracts can replace the nematicides in controlling the plant-parasitic nematodes. These materials are inveronmentaly safe, cheep and available for applied treatment.

## REFERENCES

- Abadir, S.K.; A.E., Ismail, and A.M, Khier, (1994). Efficacy of some plant wastes as soil amendments agaist *Meloidogyne incognita* on sunflower. Annals of agric. Sc., Moshtohor 32 (2) : 1027 - 1033.
- Abadir, S.K.; A.M. Khier and Ekram. F. Hashim, (1996). Nematieidel activity of some ornamental plants on the root-knot nematode, *Meloidogyne incognita*. Egypt. J. Appl. Sci; 11 (4) 241-249.
- Akhtar, M.; and M.A. Alam (1989). Evaluation of nematicidal potential in some medical plants. Int. Nematol. Network Newsl., 6 (1) : 8 - 10.
- Akhtar, A and I.Mahmood (1993). Control of plant parasitic nematode with nimin and some oils by bare-root dip treatment, Nematol. medit. 21:89-92.
- Alam, M.M.; Khan, A.M. and Saxena, S.K. (1978). Mechanism

- of control of plant parasitic nematodes as a result of the application of organic amendments to the soil. IV: Role of formaldehyde and acetone. *Indian J.Nematol.*, 8:172-174.
- Ali, H.H.Ali; M.H.El-Hamawi; and A.Kamel., 1997. Nematicidal action of some Egyptian plants. *Egypt. J.Appl. Sci*; 12:245-254.
- Ali, E.M.; M.H. El-Hamawi and Mohamed-Basyonia.E. (1998). Control of root-knot nematode *Meloidogyne javanica* by intercropping of some antagonistic plants. *Egypt. J. Appl. Sci*; 13 (6) 39 – 56.
- Al-Sayed, A.A.; S S. Ahmed and S.H. Abdel-Hamed (1992). Influence of decomposition dry leaf powders on *Rotylenchulus reniformis* reproduction and growth response of soybean. *Annals of Agric., Sci., Moshtohor*, 30 (1):615 – 620.
- Betteni, G. (1993). *Heterodera schachtie* Schmidt; the sugarbeet cyst nematode. *Informatore Agrario*. 49:2, 69 – 74.
- Duncan, D.B., (1955). Multiple range and F-test *Biometric*, 11:1-42.
- El-Naggar, H.I.;S.H. Abdel-Hameed; A.A. Farahat and A.A. Osman (1993). The role of dry ground leaves of some plants in controlling the reniform nematode *Rotylenchulus reniformis* infecting sunflower. *Bull. Fac. Agric, Cairo Univ.*, 44 (1):205 – 216.
- Farahat, A.A.;H.H., Hendy; H.I., El-Naggar and A. El-Ghonemy (1994). Time of application of dry ground leaves in relation to the nematode reproduction on sunflower. *Egypt. J. Appl. Sci.* 9 (3):79 – 97.
- Goody, L.B. (1957). Laboratory methods for work with plant and soil nematodes. *Bull. No. 2, Min. Agric. Fish and food, London.* 47pp.
- Goody, L.B. (1963). Soil and fresh water nematodes. London. 544pp.
- Hammad Eman, E. (2003). The efficacy of some nematode, antagonistic plant against the root-knot nematode, *Meloidogyne javanica* ph.D Thisi, Fac. Agric. Cairo Univ. Cairo Egypt. 219pp.
- Ibrahim,I.K.A.; M.A. Resk; H.A.A. Khalil and M.A. El-Saed, (1986). Occurrence and host range of root-knot nematode, *Meloidogyne* spp. in northern Egypt. *Alex. J. Agric. Res.* 13. 267.
- Kloepper, J.W., R.Rodriguez-Kabana, J.A.McInroy, and D.J.Collins, 1991. Analysis of

- populations and physiological characterization of microorganisms in rhizospheres of plants with antagonistic properties to pytopathogenic nematodes. *Plant soil* 136:95-102.
- Kloepper, J.W., R.Rodriguez-Kabana, J.A.McInroy, and R.W.Young. 1992. Rhizosphere bacteria antagonistic to soybean cyst (*Heterodera glycines*) and root-knot (*Meloidogyne incognita*) nematodes: Identification by fatty acid analysis and frequency of biological control activity. *Plant soil* 139:75-84.
- Mahmood, I.; A., Masood; S.K. Saxena; and S.I., Hussin, (1976). Effect of some plant extracts on the mortality of *Meloidogyne incognita* and *Rotylenchulus reniformis* *Acta Bort. India* 7:129 - 132.
- Mahmood, I; S.K.Saxena, and K.A. Kiuddin, (1982). Effect of certain plant extracts on the mortality of *Rotylenchulus reniformis* and *Meloidogyne incognita*, *Bangladesh J. Bot.* 11 (2):154 - 157.
- Mani, A.; S.N. Ahmed; P.K. Puo, and V. Dakshinamusti, (1986). Plant products toxic to the atrus nematode, *Tylenchulus semipenetrans* Cobb., *Int. Nematol. Network Newsl.* 3(2): 5:173 - 177.
- Mohamed E. Basyonia., 2001. Efficacy of different organic amendments against *Meloidogyne incognita* on broad bean. *Egypt. J. Appl. Sci;* 16:30-39.
- Montasser, S.A. (1991). effect of decomposing dry leaf powders on the incidence of root galls and the growth of tomato. *Annals of Agric. Sci Moshtohor*, 29 (4): 1739 - 1751.
- Pondey, R. (1990). Studies on phytonematotoxic properties in the extracts of some medical plants, *Inti. Nematol. Network Newel.* 7 (3):19 - 20.
- Reynolds, J.E.F. (1982). "MARTINDALE" The exto pharmacopooia, twenty-eighth Editoin. The pharaceutical press, London, 672 - 675 and 683.
- Robin, D.L., C. Jean and C. Patrice (2001). The effect of organic amendments on the interactions between nematophagous fungus *Arthorbatrys oligaspor*, and root-knot nematode *Meloidogyne mayaguensis* parasitizing tomato plants. *Biol Fertil Soils*, 34:1 - 6.
- Shawky Samaa, M. (2001). Studies on the management of some plant parasitic nematodes on

- olive Ph.D Thisis, Fac. Agric., Cairo Univ., Cairo. Egypt. 189pp.
- Siddiqi, M.M; and Alam, M.M. (1988). Studies on the nematotoxicity of root exudates of certain species of Tagetes. Indian J.Nematol, 18:335-337.
- Singh, A.J, D. Kohoi, and B.D. Parihor (1955). Search for antielmentics among indigenous remedies. I. Action of acetyl saponin from *Anagallis arvensis* on annelids and helminthes. Ind. J. Vet. Sci, 25:25 – 29.

### تأثير بعض مستخلصات النباتات الطبية والزيوت العطرية في مكافحة نيماتودا تعقد الجذور (ميلودوجيني جافاتيكا) على نباتات البطاطس

محمد علي بخيت

قسم بحوث النيماتودا - معهد بحوث أمراض النباتات

مركز البحوث الزراعية - الجيزة - مصر

تم اختبار المستخلص المائي لأوراق كل من الشيح والنناع بالإضافة إلى زيت أوراق الكافور بجرعات مختلفة لمكافحة نيماتودا تعقد الجذور (ميلودوجين جافاتيكا) على نبات البطاطس صنف نيقولا تحت ظروف الصوبة خلال موسم ربيع ٢٠٠٢ وظروف الحقل خلال موسم ربيع ٢٠٠٣. أظهرت النتائج أن زيت أوراق الكافور ومستخلص الشيح أعطيا أعلى خفض في عدد العقد وتكاثر النيماتودا بالإضافة لإحداث زيادة في نمو النبات ، بينما أعطى مستخلص أوراق النناع تخفضاً قليلاً في عدد العقد وتكاثر للنيماتودا وزيادة جيدة في نمو النبات. هذا وأحدث زيت أوراق الكافور بجميع التركيزات المستخدمة تخفضاً كبيراً في عدد العقد وتكاثر النيماتودا في حين أعطى التركيز الأول ١% تحسن في نمو النبات بينما أظهرت التركيزات الأخرى سمية عالية للنباتات ، في حين أظهر المبيد النيماتودي الفاويديت ١٠% محبب أفضل النتائج بالمقارنة بجميع المعاملات الأخرى المستخدمة.