EFFECT OF CRUDE EXTRACT GARNEW ON PEACH AND GRAPE INFECTION WITH ROOT KNOT NEMATODE Moatamed, A. M. H.¹ and Hanaa S. Zawam² 1-Hortculture Research Institute

2- Plant Pathology Research Institute Agric. Res. Center, Giza-Egypt

ABSTRACT

A pot experiment were carried out during two seasons of 2009/2010 and 2010/2011 under greenhouse conditions in the nursery of Hortculture Research Institute. Garnew crude extract of (Artemisia, Garlic, Chrysanthemum, Menthe and Marjoram)

was tested to control the nematode infection of *Meloidogyne incognita* and *Meloidogyne javanica* on peach and grape .Three concentrations of Garnew were used (0.5%, 5% and10%) to control the both nematode species on peach and grape. The most effective concentration of Garnew was 10%. Whereas the nematicide ethoprop decreased the nematode soil population by 98.8% at the recommended dose and oxamyl decreased it by 98.1% at the recommended dose. Garnew decreased the number of galls, egg-masses and the developmental stages of the both nematode species on peach and grape by the same level of reduction. Results of plant growth parameters indicated that the highest concentration of Garnew (10%) has appositive effect on plant growth. Mineral accumulation in the leaves or roots of both cultivars of peach and grape was differed according to the concentration of the treatment, but generally increased than the untreated plants. The total protein electrophoresed on one dimension SDS-PAGE revealed differences in the intensity of the same protein bands between the treated and untreated plants.

Keywords:Non fumigant nematicides, crude extract (Garnew), *Meloidogyne* spp., Peach, Grape, nutrient uptake, one dimension SDS-PAGE, total protein electrophoreses

INTRODUCTION

Nematodes are important pests in grapes and peach trees around the world, and these soil-borne pests can be particularly problematic in Egypt on peach and grape. Two factors intensify the impact of nematodes : the high value of grapes and of vineyard land costs. These factors force growers to ignore the steps of leaving land fallow and rotating crops, both of which reduce nematode build up and delay the selection of adapted strains. Nematicides and fumigants help in control nematodes, but the use of these pesticides has been greatly restricted. Plants are an important source of naturally occurring pesticides. Many compounds with nematicidal activity have been found in plants, including alkaloids, diterpenes, fatty acids, glucosinolates, isothiocyanates, phenols, polyacetylenes, sesquiterpenes and thienyls; (Gommers, 1981; Chitwood, 2002). Many compounds with nematicidal activity have been isolated from species in the family Asteraceae(Gommers, 1981; Chitwood, 2002). Also, Allicin (an active nematicidal principle in garlic) has been isolated by Gupta and Sharmai, 1993 and tested against Meloidogvne incognita infesting tomato, they found that

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juvenile mortality of 87–100% at 2.5–5.0 ppm allicin was recorded within 72 h. Essential oils of some plants and/or their components have been tested for nematicidal activity *in vitro* and in soil(Chatterjee *et al.*,1982; Soler-Serratosa *et al.*, 1996; Oka *et al.*, 2000). Recently, the antifungal and insecticidal activity of the essential oil of *Chrysanthemum coronarium* flowerheads has been reported (Perez and Pascual-Villalobos, 1999; Alvarez-Castellanos *et al.*, 2001). the essential oil from *Mentha spicata* with a high content of carvacrol and thymol, was effective against *M. javanica* (Oka *et al.* 2000).

The aim of the proposed study included;

Evaluate the efficacy of the crude extract compound to reduce the number of nematode in soil and roots of plants

MATERIALS AND METHODS

The present study revealed the comparative efficacy of Garnew compound as crude extract comparing with nematicides (Ethoprop and Oxamyl) with two addition rates.

Compound	Structure	Application rates
Ethoprop	Organophosphate	40 Kg /feddan(1) 4 Kg /feddan(2)
Oxamyl	Oximcarbamate	8 L / feddan(1) 800 Cm ³ /feddan(2)
Garnew	Crude extract of Artemisia, Garlic Chrysanthemum, Menthe and Marjoram	8 L / feddan (1) 800Cm³/ feddan (2)

Table (1) : List of compounds tested in this investigation

Three experiments were carried out to study the effect of crude extract on plant parasitic nematode *Meloidogyne javanica*, and *Meloidogyne incognita*. The first experiment was conducted to test the dosage rates using on peach cultivar (Mit Gammer) and grape cultivar (Superior) seedlings under green house conditions

 $(25-\pm 2^{\circ}C)$ to control the root-knot nematode .The experimental soil was collected from the ARC farm, Giza, Egypt. The soil texture was sandy clay . Black plastic bags 20 cm in diameter were used for our test filled with steam sterilized soil about five kilo per bag

Nematode stock culture

Nematode populations were maintained on tomato plants cv. Castle rock under greenhouse conditions . Plants were infected at 2-3 leaves stage by adding egg-masses to roots (one egg-mass per one plant for making pure culture from *M. incognita* and *M. javanica* in plastic cups) then covered with soil. After 60 days nematode egg-masses collected from each root by a needle, put in Petri dishes and put it in incubator for hatching at $25\pm2^{\circ}$ C for a week. The hatched juveniles were collected daily.

To study the effect of Garnew on development of *Meloidogyne* sp. on peach cultivar Mit Gammer seedling ,forty plastic bags were cultivated by one year

old seedlings of peach in steam sterilized sandy clay soil . Twenty plastic bags were inoculated by 2000 newly hatched larvae of *M. incognita* by boring the nematode suspension in holes around the roots of the peach seedlings. Other twenty plastic bags were inoculated by the same method by 2000 newly hatched larvae of *M.javanica*. The twenty plastic bags which inoculated by *M.incognita* were divided into four groups 3 of them treated by Garnew compound by the dosage of (0.5%, 5% and 10%) and the other 5 bags left without treatment of Garnew and served as inoculated control. The same treatment were made by the plastic bags of *M.javanica* This experiment was repeated on grape cultivar Superior of one year old seedlings. After 90 days the plants were uprooted and the roots were washed free from the adhering soil particles. Number of galls, number of egg-masses per 5gm roots and number of nematodes in 250 cm ³ soil and also the developmental stages inside the roots were determined.

Another pot experiment was conducted to explore the effectiveness of the crude compound Garnew to reduce *Meloidogyne incognit* on peach cultivar Mit Gammer and grape cultivar Superior seedlings comparing with two nematicides (ethoprop and oxamyl) by using two concentrations from each.

Forty plastic bags were cultivated by one year old seedlings of peach in steam sterilized sandy clay soil. The plastic bags were divided into 8 groups, 7 of them inoculated by 2000 newly hatched larvae of *M. incognita* by boring the nematode suspension in holes around the roots of the peach seedlings, and one group left without inoculation and served as control. Other 7 groups were divided into :

- 1-two groups treated by ethoprop by the recommended dose and 1/10 of the recommended .
- 2-two groups treated by oxamyl by the recommended dose and 1/10 of the recommended .
- 3- two groups treated by Garnew by the recommended dose and 1/10 of the recommended .
- 4- one group left without treatment and served as inoculated control.
- This experiment was repeated on grape cultivar Superior of one year old seedlings.

After 90 days the plants were uprooted and the roots were washed free from the adhering soil particles. Number of galls, number of egg-masses per 5gm roots and number of nematodes in 250 cm ³ soil and also the developmental stages inside the roots were determined. Also, plant growth parameters ,shoot length, shoot weight and root weight were determind for both peach and grape cultivars. Also, the number of new branches for peach was recorded . The plant analysis and total protein electrophoresis from this experiment determined according the following methods:

Plant analysis:

Samples of the fourth top leaves and secondary roots were taken and oven dried at 70° C for 48hrs and kept for chemical analysis.

A wet digested according to the methods of Tomas *et al.*, (1967). In the digest solution N,P,K,Fe and Mn were analyzed according to the following methods :

- 1- Total nitrogen % was determined by the distillation in a macrokjeldahl apparatus (Helrich, K. 1990)
- 2- Phosphorus % was calorimetrically determined as described by Ranganna (1979
- 3- Potassium % was determined photometrically using flame photometer, as described by Ranganna(1979)
- 4- Ca, Fe, Mg, Mn, Na and Zn were determined using Atomic Absorption Spectrophotometer PERKIN ELEMER 3300 according to Chapman and Pratt (1981)

All treatments of greenhouse experiments were statistically arranged in a complete randomize design according to Snedecor and Cochran (1989), where mean values were compared using L.S.D. at 5% level.

Electrophoresis studies:

Total protein analysis

Three grams of plant root samples were ground in precooled mortar and pestle with liquid nitrogen to a fine powder then 0.7 ml of extraction buffer (0.6 ml 1 M Tris HCl $\,$ pH 6.8, 5 ml 50% glycerol , 2 ml 10% SDS , 0.5 ml β mercaptoethanol and 0.9 ml H₂O) was added and the extracts were clarified by centrifugation at 14000xg for 15 minutes under cooling. The supernatants were transferred in fresh ependorf tubes and stored at -20°C. Supernatants containing soluble proteins fractions were transferred to clean tubes and stored at- 20°C. Protein content was estimated according to the methods of Bradford (1976) using Bovine Serum Albumin (BSA) as a standard. Protein content was adjusted to 2 mg / ml per sample. SDS was added to the sample at the rate of 4 mg SDS / 1 mg protein, then 50 µl, ß- mercaptoethanol were added. The mixture was boiled at 100°C in a water bath for 3-5 min. Vertical slab (18x16 cm) gel electrophoresis apparatus was used as marketed by Hoofer (Hoofer SE 600 series Pharmacia). 20 µl of this crude protein solution were resolved on 11 % SDS - PAGE using molecular weight protein marker as a standard. Electrophoresis was carried out at 2 mille ampere per sample at 10 °C for 3 hrs.Gels were stained by silver staining method for protein as described by Sammons et al (1981). This method of staining is sensitive and detects as little as 2 ng of protein in a single band. Gels were scanned for estimation molecular weight by using gel documentation system (AAB Advanced American Biotechnology 1166 E. Valencia Dr. Unit 6C, Fullerton CA, USA 92631). The different molecular weights of bands were determined against protein standard (Peglab) marker.

RESULTS AND DISCUSSION

Plant-parasitic nematodes feed on grapevine roots and cause malformations or necrosis. This leads to destruction of physiologically active roots and an overall reduction in water and nutrient uptake. Above-ground parts of grapevines show no specific visual symptoms on leaves, shoots or fruits, but there is a general reduction in vigour. Similar symptoms could be due to, and confused with, other conditions

such as poor physical characteristics of the soil, mineral excess or deficiency, water stress, or other soil-borne pests and diseases. Soil amended with crude extract Garnew offers a satisfactory and environmentally friendly compound for the control of root-knot nematode

Table (2): Effect of Garnew on development of *Meloidogyne* sp. on peach Mit Gammer seedlings.

			-	
Treatmonte		Nematode p	arameters (M.inc	ognita) /root
Treatments	J2/250cm [*]	No.galls/root	Nematode parameters (M.incognita) /roc galls/root No.egg-masses No.D.S 2300 1700 13 1900* 1200* 98 470* 300* 24 160* 70* 45 97.508 95.946 144. Nematode parameters (M.javanica) /ro 1360 99 1500* 1080* 83 420* 340* 21 180* 92* 53 36.930 159.131 29.5	No.D.S/root
M.incognita	4200	2300	1700	1300
Garnew0.5%	3600*	1900*	1200*	980*
Garnew 5%	900*	470*	300*	240*
Garnew 10 %	350*	160*	70*	45*
LSD 5%	249.737	197.508	197.508 95.946	
		Nematode p	parameters (<i>M.jav</i>	<i>anica</i>) /root
M.javanica	3090	1730	1360	990
Garnew0.5%	2700*	1500*	1080*	830*
Garnew 5%	850*	420*	340*	210*
Garnew 10 %	330*	180*	92*	53*
LSD 5%	75.804	36.930	159.131	29.503

* means there is a significant effect at 5% level

Results in Table (2) indicated that there was a significant effect of the crude extract on all nematode parameters estimated with either *M. incognita* or *M. javanica*.

 Table (3): Effect of Garnew on development of Meloidogyne sp. on grape Superior seedlings.

Treatmente		Nematode p	atode parameters (<i>M.incognita</i>) /root				
Treatments	J2/250cm ³	Nematode para cm³ No.galls/root N 0 3250 0 >* 2350* * * 380* * * 210* 9 166.757 Nematode para 0 1340 0 >* 330* * * 130* 94	No.egg-masses	No.D.S/root			
M.incognita	5100	3250	2570	2400			
Garnew0.5%	3800*	2350*	1860*	990*			
Garnew 5%	890*	380*	430*	320*			
Garnew 10 %	370*	210*	65*	38*			
LSD 5%	127.798	166.757	162.329	198.862			
		Nematode p	parameters (M.jav	<i>anica) /</i> root			
M.javanica	3600	1340	1210	1040			
Garnew0.5%	2400*	900*	950*	720*			
Garnew 5%	720*	330*	230*	110*			
Garnew 10 %	230*	130*	60*	55*			
LSD 5%	178.394	66.763	91.373	18.855			

* means there is a significant effect at 5% level

The same trend has been shown in Table (3) the reduction of juveniles in soil numberes of galls, egg-masses and different stages imbedded in the roots was gradually decreased as the concentration of the crude compound Garnew increased either for *M. incognita* or *M. javanica*. The effectivness of the crude compound Garnew was studied by comparing with the nematicides ethoprop and oxamyl using two doses on peach as showen in Table 4

			`			U		
Treatments	J2/250cm ³ soil	R%	No. Galls/root	R%	egg- masses /root	R%	No. D.S /root	R%
Ethoprop1	48	98.8	25	98.3	23	97.6	41	95
Ethoprop2	2620	35.6	1152	23.2	500	48.5	113	86.2
Oxamyl1	76	98.1	42	97.2	31	96.8	50	93.9
Oxamyl2	2700	33.7	1400	6.67	620	36.1	217	73.5
Garnew 1	450	88.9	220	85.3	192	80.2	134	83.7
Garnew 2	700	82.8	340	77.3	221	77.2	161	80.4
Infected plant	4070		1500		970		820	
LSD 5%	900.99		85.58		87.58		6.724	

Table (4) : Effect of some treatments on development of Meloidogyne incognita in Peach (Mit Gammer) seedlings.

J2: number of second stage juvenile in soil

D.S: number of developmental stages inside the roots

Reduction: R%= Nematode number in control_-_nematode number in treatment Nematode number in control

1:recommended dose of the compound

2: 1/10 of the recommended dose

Data in Table (4) showed that highest effect refer to the treatment with ethoprop at recommended dose followed by oxamyl .Also, the crude compound Garnew gave a decrease in root knot nematode on peach seedling compared with the treatment of ethoprop and oxamyl at low concentration (1/10 recommended dose) and untreated control .

Table	(5): Effect c	of some	treatmen	ts on o	develo	opment o	of <i>Meloido</i>	gyne
		incogn	<i>ita</i> on gr	apevine ((Super	ior s	seedling	s.	

Treatments	J2/250cm ³ soil	R%	No. Galls/ root	R%	egg- masses/ root	R%	No. D.S /root	R%
Ethoprop1	200	83.3	300	72.7	321	65.5	117	81.06
Ethoprop2	500	58.3	370	66.4	352	62.2	169	72.7
Oxamyl1	280	76.7	341	69.0	372	60	92	85.1
Oxamyl2	520	56.7	380	65.5	416	55.3	131	78.8
Garnew 1	500	58.3	410	62.7	432	53.5	141	77.2
Garnew 2	600	50	460	58.2	466	49.9	154	75.1
Nematode	1200		1100		930		618	
LSD 5%	97.671		60.516		6.751		6.634	

J2: number of second stage juvenile in soil

D.S: number of developmental stages inside the roots

Reduction : R%= Nematode number in control-nematode number in treatment

Nematode number in control 1:recommended dose of the compound

2: 1/10 of the recommended dose

The nematicide protects the roots from nematode invasion which resulted in sharp reduction in the number of galls, egg-masses in roots, and juveniles in soil. The present results in Table (5) emphasized that the crude extract exhibited potential nematicidal activity against the root-knot nematode and improved growth criteria of vineyard and peach even at low concentrations.

All the tested materials significantly suppressed root-galling, the number of egg-masses and subsequently the final population. However, the natural compound Gar- new seemed to have toxic action

The nematicidal effect of the tested natural compound may possibly be attributed to high contents of certain oxygenated compounds which are characterized by their lipophilic properties that enable them to dissolve the cytoplasmic membrane of nematode cells and their functional groups interfering with the enzyme protein

structure (Knoblock *et al.*,1989). The mechanisms of plant extracts action may include denaturing and degrading of proteins, inhibition of enzymes and interfering with the electron flow in respiratory chain or with ADP phosphorylation(Konstantopoulou *et al.*, 1994).

	parameters of	Mit Gammer peach cultivar.						
Treatn	nents	Shoot length Cm	Shoot weight gm	Root weight gm	No.of new branches			
Infected plant +N+	Ethoprop1	88.3	53.7	40.1	3			
Infected plant +N+	Ethoprop2	78.7	45.3	35.7	3			
Infected plant +N+	Oxamyl1	78.3	50.1	38.3	4			
Infected plant +N+	Oxamyl2	69.7	39.3	28.7	3			
Infected plant +N+	Garnew1	96.3	60.3	43.7	7			
Infected plant+N+	Garnew2	89.1	49.7	39.3	7			
Infected nematode p	lant	45.1	21.7	13.3	2			
Non-infected plant		47.3	30.3	19.7	3			
LSD 5%		5.6	3.4	4.5	1.6			

 Table (6): Effect of Garnew and two nematicides on some growth parameters of Mit Gammer peach cultivar.

It is clear from the data in Table (6) that applying the nature compound Garnew at high rate (8 L/feddan) recorded the highest shoot length and shoot and root weights. Also the number of new branches significantly was increased by 2.5% as compared with untreated plants. The lowest values of vegetative growth were associated with the treatment of nematode only without any treatment. Increasing ethoprop and oxamyl doses increased significantly root and shoot criteria. Nyczepir *et al* 2000 found that plant growth of peach cultivar Lovell was suppressed by both *M. incognita* and *M. javanica*.

Table (7): Effect of Garnew and two nematicides on	some growth
parameters of Superior grape cultivar.	

Treat	tments	Shoot length	Shoot weight	Root weight
		Cm	gm	gm
Infected plant +N+	Ethoprop1	58.6	30.5	20.3
Infected plant +N+	Ethoprop2	46.6	28.7	18.1
Infected plant +N+	Oxamyl1	60.2	30.7	20.7
Infected plant +N+	Oxamyl2	45.0	25.3	17.3
Infected plant +N+	Garnew1	87.7	43.2	31.3
Infected plant+N+	Garnew2	75.2	40.7	29.1
Infected nematode	plant	33.7	12.3	15.3
Non-infected plant		36.3	23.7	13.7
LSD 5%		2.23	4.8	4.16

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Data in Table (7) showed that the same trend of increasing in shoot length ,shoot and root weights of grape. The highest values recorded from Garnew treatments followed by ethoprop and oxamyl at the dose 1 and finaly the lowest treatment values was recorded with the ethoprop and oxamyl at the dose two.

Table	(8)	:	Effect	of	Garnew	and	two	nemati	cides	on	min	eral
			accun	nula	tion of	Leaf :	sampl	es from	peach	cul	tivar	Mit
			Gamn	ner i	noculaled	d with	Meloi	idoavne	incoar	nita.		

Treatments			Concentration% concentration ppm									
	Ν	Ρ	κ	Mg	Са	Na	Zn	Mn	Fe			
Infected plant +N+	Ethoprop1	2.61	0.16	3.20	908.09	0.182	0.250	36.93	8.37	296.3		
Infected plant +N+	Ethoprop2	2.53	0.15	2.20	905.68	0.126	0.241	25.81	7.69	282.1		
Infected plant +N+	Oxamyl1	2.76	0.23	3.00	908.09	0.172	0.259	25.54	7.63	297.2		
Infected plant +N+	Infected plant +N+ Oxamyl2		0.22	2.77	901.56	0.144	0.213	25.40	7.48	280.3		
Infected plant +N+	Garnew1	2.57	0.16	4.15	1206.5	0.247	0.286	86.40	10.8	347.3		
Infected plant+N+	Garnew2	2.45	0.18	3.08	909.43	0.209	0.259	26.80	8.77	315.4		
Infected nematode plant		2.20	0.15	2.89	806.84	0.107	0.222	24.62	6.73	252.7		
Non-infected plant		2.30	0.15	2.89	901.15	0.182	0.227	24.73	7.0	277.3		
LSD 5%		0.41	0.01	0.43	15.1	0.08	0.006	2.62	1.2	2.883		

The obtained results in Table (8) showed that the uptake and accumulation of minerals by Mit Gammer leaves that reflected the improvement of plants according to the treatments comparing with nematode treated plants in some minerals

Table	(9):Effect	of	Garne	ew a	and	two	nemat	icides	on	min	eral
	accum	ulati	on of	root	t sa	mples	from	peach	culti	var	Mit
	Gamm	er in	oculale	d wi	th M	eloido	qyne in	cognita	-		

Trea	Cone	centrat	ion%	concentration ppm							
			Ν	Р	κ	Mg	Ca	Na	Zn	Mn	Fe
Infected	plant	+N+	1.76	0.17	0.71	808.45	0.182	0.232	28.17	28.09	352.1
Ethoprop1											
Infected	plant	+N+	1.59	0.16	0.60	800.97	0.135	0.222	23.59	29.05	317.0
Ethoprop2											
Infected	plant	+N+	1.67	0.19	0.71	706.73	0.228	0.222	32.17	53.60	322.7
Oxamyl1											
Infected	plant	+N+	1.59	0.16	0.57	706.73	0.135	0.204	26.65	24.82	303.5
Oxamyl2											
Infected	plant	+N+	1.82	0.22	0.67	1200.6	0.219	0.305	27.78	28.93	355.3
Garnew1											
Infected plant+N+		1.70	0.19	0.54	1106.4	0.219	0.277	26.56	25.72	336.4	
Garnew2											
Infected nematode			1.16	0.14	0.37	701.11	0.163	0.204	24.03	44.18	218.7
plant											
Non-infected plant			1.56	0.15	0.44	704.40	0.228	0.222	26.36	20.7	265.1
LSD 5%			0.08	0.012	0.02	12.2	0.003	0.003	3.31	3.2	2.955

Our data in Table (9) showed that the roots of treated plants accumulated more N, P ,K and small or minor elements than the untreated controls. Since the treated roots were heavier than those of the controls, this higher amount of nutrients is probably a consequence of an increased root

system absorbing surface, although gall formation would have contributed significantly to the final root mass.

	Č	accu	mula	tion	01	Lear	sampie	s nor	n gra	ipe (Juitivai
		Supe	rior i	nocu	lated	with <i>l</i>	Meloido	gyne	incog	nita.	
Trootmonto		Concentration %						concentration ppm			
rieatments			Ν	Р	κ	Mg	Ca	Na	Zn	Mn	Fe
Infected	plant	+N+	1.73	0.29	3.86	703.93	0.246	0.250	28.27	9.03	285.1
Ethoprop1											
Infected	plant	+N+	1.66	0.27	3.07	604.05	0.246	0.241	25.44	8.80	272.3
Ethoprop2											
Infected	plant	+N+	1.78	0.30	3.02	609.11	0.225	0.259	26.73	7.97	281.9
Oxamyl1											
Infected	plant	+N+	1.65	0.28	2.55	508.24	0.219	0.213	23.13	7.89	275.7
Oxamyl2											
Infected	plant	+N+	1.78	0.35	2.34	708.66	0.274	0.286	42.42	8.73	336.5
Garnew1											
Infected	plant+N+		1.65	0.32	2.34	562.87	0.246	0.259	36.77	8.38	307.2
Garnew2											
Infected nematode			1.41	0.20	2.17	400.17	0.163	0.222	24.42	6.40	243.6
plant											
Non-infected plant			1.60	0.32	2.57	405.94	0.181	0.247	24.67	6.55	265.7
LSD 5%			0.09	0.06	0.33	13.42	0.02	0.006	1.36	0.79	0.920

 Table (10):
 Effect of Garnew and two nematicides on mineral accumulation of Leaf samples from grape cultivar Superior inoculated with *Meloidogyne incognita*.

Most elements were within sufficiency levels (Jones *el al.*, 1991). Leaves of plants inoculated with the nematode alone were low in N and Fe. The addition of nematicides and biofertilizer resulted in increased plant growth and gave the highest Ca, Fe, Mg, Mn and Na values. Also, P and K increased with the addition of defferent treatments with some exceptions

Table (11) : Effect of Garnew and two nematicides on mineral accumulation of root samples from grape cultivar Superior inoculated with *Meloidogyne incognita*.

Treatments			Conce	entratio	on %		concentration ppm						
			Ν	Р	κ	Mg	Са	Na	Zn	Mn	Fe		
Infected Ethoprop1	plant	+N+	0.95	0.26	1.21	804.8	0.219	0.289	40.03	20.95	335.4		
Infected Ethoprop2	plant	+N+	0.85	0.24	1.10	709.59	0.209	0.286	38.98	20.58	297.3		
Infected Oxamyl1	plant	+N+	0.95	0.24	1.14	701.85	0.217	0.286	39.85	20.86	315.3		
Infected Oxamyl2	plant	+N+	0.94	0.21	0.78	690.85	0.200	0.277	31.02	19.59	289.7		
Infected Garnew1	plant	+N+	1.25	0.33	1.65	902.11	0.293	0.350	70.3	26.06	344.5		
Infected Garnew2	plant+N	1+	1.09	0.30	1.50	802.72	0.228	0.323	40.88	22.31	327.1		
Infected nematode plant			0.67	0.21	0.75	409.5	0.172	0.236	27.01	16.94	226.3		
Non-infected plant			0.83	0.24	1.00	505.39	0.200	0.268	30.75	17.39	255.1		
LSD 5%			0.06	0.06	0.15	15.36	0.33	0.04	1.93	2.49	1.264		

Our findings in Table (11) indicated that some nutrient elements decrease(N and Fe) while others increase notably in leaf tissues (Mg, Mn, Zn, and Na) in nematode inoculated treatments. In the first case, absorption and transport of Fe and N to aerial parts would seem to be impaired by the destruction of the root cortical tissues caused by the nematode probably due to the loss of the capacity for differential permeability which reduces nutrient element transport (Kirkpatrick, 1964). In contrast, Mg, Mn, Zn and Na, seem to be absorbed continuously and accumulate in leaf tissues as a result of reduced growth, thus their increasing concentration. The lower concentrations in leaf tissues of these same elements in treatment without the nematode is explained by a growth dilution effect (Kleinschmidt & Gerdemann, 1972; Granger *et al.*, 1983).

A similar pattern for these elements (increase in Zn, Mg, and Mn and reduction in Fe and Cu in foliar apple (**Pinochet** *el al.*, **1993** *a*) in plants infected with nematode.

Protein profile of peach and vineyard infected with *Meloidogyne* spp.:

To find the biochemical differences between the infected and treated plants with Gar new and nematicides, total protein was extracted and electrophoresed on one dimension Sodium dodecyle sulphate, polyacrylamide gel electrophoresis (SDS-PAGE).

SDS-PAGE analysis of infected plants revealed a clear differences in the intensity of the same protein bands between the infected and treated plants.

Data presented in Fig. (1& 2) showed the protein profile of plants infected with *Meloidogyne incognita* and treated with biofertelizer Gar new and nonfumegant nematicides ethoprop and oxamyl this reflected the possible physiological differences among the treatments . The present results are in harmony with those of Farahat *et al* 2012, who reported that treating infected plants with fertilizers improve the performance of infected plants by enabling them to recompense root losses of soluble sugars and total carbohydrate and brought phenol contents back to be almost near to those in untreated healthy plants, raising tannins content, diminishing root contents of amino acids to be around those in healthy plants.

This study was designed to evaluate the nematicidal activity of the organic compound from several family species of the Asteraceae on the rootknot nematode *M. incognita* in planta experiments clearly demonstrated that J_2 survival and reproduction rate of the nematode were significantly reduced on grape and peach compared to the nonamended treatment. other researchers found that the population density of *Meloidogyne* spp. was reduced when host plants were grown in soil amended with *Chrysanthemum spp.* or *Artemisia, Mentha, Garlic* and / or *Marjoram* the results of the present study regarding the effects of nematode parasitism on plant growth under artificial conditions may be are not in agreement with the results of other researchers under field conditions. Differences in the susceptibility of the plant cultivars or differences in environmental conditions could be responsible for this. Essential oils from several plant species have been shown to have nematicidal activity on root-knot nematodes *in vitro* and in soil This compound is easily used into organic, conventional and integrated control



growing system. Given obvious benefits and government may consider it as a promoting practice

Fig. (1). Protein profile analysis and dendrogram of peach plants infected with *Meloidogyne incognita* using SDSpolyacrylamide gel electrophoresis stained with silver nitrate.



Fig. (2) :Protein profile analysis and dendrogram of grape plants infected with *Meloidogyne* incognita using SDSpolyacrylamide gel electrophoresis stained with silver nitrate.

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تأثير المستخلص الطبيعى جارنيو على اصابة الخوخ والعنب بنيماتودا تعقد الجذور عاطف معتمد حسين معتمد¹ و هناء سيدهم زوام² 1- معهد بحوث البساتين 2- معهد بحوث امراض النباتات - مركز البحوث الزراعية – جيزة

تمت هذه الدراسة تحت ظروف الصوبة لدراسة تأثير الستخلص الطبيعى جارنيو (مستخلص مخلوط مجموعة نباتات الاقصوان والنعناع والثوم والدمسيسة) بتركيزات 5,0%و5%و10%على نوعى نيماتودا تعقد الجذور ميلوديجينا انكوجنيتا وميلوديجينا جافانيكا على كل من الخوخ ميت غمر والعنب صنف سبيريور وقد اظهرت النتائج انه بزيادة تركيز المستخلص يزداد الانخفاض فى اعداد الطور اليرقى الثانى فى التربة وكذلك اعداد العقد النيماتودية وأكياس البيض على الجذور كما قلت بوضوح اعداد الاطوار بداخل لجذور لكل من نوعى النيماتودا على كلا النياتين

وفى تجربة لمقارنة تأثير المستخلص بمبيدين نيماتوديين هما الايثوبروب والاوكساميل بتركيزات 8 لتـر / للفـدان و800سم³/ للفـدان للمسـتخلص و40 كجـم /فـدان و4كجـم /فـدان للايتُـوبروب و8 لتر/فدان و 800سه³/فدان للاوكساميل لنوع النيماتودا ميلوديجينا انكوجنيتا على الخوخ والعنب اظهرت النتائج نسبة انخفاض لأعداد اليرقات في التربة تراوحت بين 8 89% -1 89% للتركيز ات المرتفعة لكل من الايثوبروب والأوكساميل في حين كانت نسبة الانخفاض 9 88% للتركيز المرتفع مسن المستخلص وذليك مقار نسسة بساكنترول المعسدي بالنيمساتودا فقسط وصارت نسبة الانخفاض في اعداد العقد النيماتودية وأكياس البيض والاطوار بداخل الجذور حول نفس المعدل تقريباً وذلك عند معاملة الخوخ ميت غمر

أما فى حالة صنف العنب سبيريور فقد انخفضت أعداد اليرقات فى التربة الى 3,83% و7,77%و3,85% لكل من الايثوبروب والأوكساميل والمستخلص جارنيو على الترتيب فى التركيزات المرتفعة وتفوق الأوكساميل على الايثوبروب والجارنيو فى التأثير على الاطوار بداخل الجذور حيث كانت نسبة الانخفاض 1,85%و 06,81%و2,77% للمركبات الثلاثة على الترتيب مقارنة بالكنترول المعدى بالنيماتودا فقط

كما أوضحت القياسات النباتية تأثير المركبات الثلاثة المحفز للنمو سواء للخوخ او العنب كما زاد عدد النموات الحديثة في الخوخ المعامل بالمستخلص النباتي الطبيعي عن المبيدات النيماتودية مقارنة بالكنترول المعدى بالنيماتودا فقط كما زادت النسبة المؤية لتركيزات العناصر الكبرى والمتوسطة وكذلك التركيزات بالجزء في المليون للعناصر الصغرى سواء في الأوراق او الجذور لكلا النوعين النباتيين

كذلك اوضح التفريد الكهربى للبروتين المستخلص من نباتات الخوخ والعنب المصابة بنيماتودا تعقد الجذور والمعاملة بالإيثوبروب والأوكساميل والجارنيو بتركيزين ان هناك اختلافات بيوكيميائية تظهر من خلال عدد الباندات وكذلك كثافتها باختلاف المعاملة والتركيز .

قام بتحكيم البحث

أ.د / عبد البديع عبد الحميد غانم
 أ.د / محمود حسن الحموى

كلية الزراعة – جامعة المنصورة مركز البحوث الزراعية