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Original article

Controlling of *Meloidgyne incognita* (Tylenchida: Heteroderidae) using nematicides, *Linum usitatissimum* extract and certain organic acids on four peppers cultivars under greenhouse conditions

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A B S T R A C T

Organic acids and plant extracts, which have a nemacidal action and may be used instead of nematicides that pollute the environment, are one way for controlling the pepper root-knot nematode. We provide in this study for a first time a new strategy for management *Meloidgyne incognita* (Kofoid and White) by using organic acids and plant extract compared to nematicides on four peppers cultivars (Super amarr, Super mard, Super noura and Werta) under greenhouse conditions compared to nematicides. This study aimed to evaluate 0.1% of organic acids (humic and salicylic acid) and 0.1% of *Linum usitatissimum* extract on plant parameters of pepper varieties (Super amarr, Super mard, Super noura and Werta) and control of *M. incognita* under greenhouse conditions compared to four nematicides (Oxamyl 24% SL, Fosthiazates 75% EC, Ethoprophos N40% EC and Fenamiphos 40% EC). Our data obtained four nematicides were more effectiveness than other treatments in reduced galls and egg masses of *M. incognita*. Whilst, humic and salicylic acids have remarkably higher nematicidal activity than *L. usitatissimum* in all lines of pepper. Therefore, plant extract and organic acids may be used a best alternative of nematicides to control PPNs and caused the longitudinal growth of plant. Also, ultimately reduce environmental risk from nematicide pollution.

1. Introduction

Pepper (*Capsicum annum*) is a major vegetable crop in Egypt, both for domestic use and export (Wesolowska et al., 2011) and it is a member of the solanaceous vegetables group. Peppers are high in phenolics and flavonoids (Bae et al., 2012), as well as carotenoids (Ha et al., 2007), vitamin C, vitamin E (García-Closas et al., 2004), and alkaloids (Srinivas et al., 2009), all of which are beneficial to human health. Throughout its growing stage, pepper output is seriously challenged by a variety of pests and diseases, with nematodes being one of the most troublesome pests, inflicting extensive root damage and considerable financial losses (Djian-Caporalino et al., 2007). Plant-parasitic nematodes (PPNs) are a serious agricultural concern, producing with an estimated annual decreased productivity of 12.3% ($157 billion) over the world (Singh et al., 2015), considerably more than invading insects (about US$70 billion; Bradshaw et al., 2016). *Meloidogyne* spp., root knot nematodes (RKNs), is one of the most devastating nematode pests in agricultural producing regions, inflicting significant economic damage worldwide (Rehman et al., 2012). RKN-infected plants display symptoms both above and belowground. Aboveground, infected plants show poor development, fewer and small pale green leaves that wilt in high temperature. The symptoms emerge as large galls on the roots that are two to several times the diameter of a healthy root and interfere with the plant's ability to

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absorb and translocate water and dissolved nutrients. The worms prey on roots, injuring them and allowing other soil-borne illnesses to enter (Agrios, 2005). Escobar et al., (2015) reported that *Meloidogyne incognita* infection has a significant impact on pepper root development, resulting in root galls, yellowed and stunted leaves, and potentially the loss of entire plants.

Chemical toxins, resistant cultivars, crop rotation, and biological management are some of the approaches used to manage and control this nematode, and these methods are sometimes combined (Katooli et al., 2010). As population expansion necessitates increased agricultural output, the traditional method of nematode management is completely reliant on nematicides. However, the loss of pesticides as a result of EU rules (EC No1107/2009) has increased the need for effective nematode resistance, which state that they are dangerous to human health and a pollutant to the environment (Zhang et al., 2017). As a result, there is a critical need for a nematode control option that is both ecologically acceptable and effective, such as *Linum usitatissimum* extract (Ismail et al., 2014), organic acids like humic acid (HA) (Nagachandrabose and Baidoo, 2021) and salicylic acids (SA) (Martínez-Medina et al., 2017). Many crops have been displayed to benefit from HA, a byproduct formed during the breakdown of organic waste (Khattab et al., 2012; Khan et al., 2013). In vitro, HA impacts the hatching of eggs and the survival of *M. incognita* second stage juveniles (Seenivasan and Senthilnathan, 2017). Salicylic acid (SA) is a phytohormone that controls many aspects of plant development and resilience to different stressors, including herbivores that feed on phloem. It governs many aspects of plant development and resilience to different challenges, including phloem-feeding herbivores, and causes a wide spectrum of metabolic and physiological responses in plants (Kawazu et al., 2012). Moreover, SA increases plant tolerance to a variety of biotic and abiotic stresses (Horváth et al., 2007).

The main hypothesis of this research is that the plant extract and organic acids would control PPNs and ultimately reduce environmental risk from nematicide pollution. This study aimed to evaluate *Linum usitatissimum* extract and organic acids (humic acid and salicylic acids) on plant parameters of pepper varieties (Super amarr, Super mard, Super noura and Werta) and control of *Meloidogyne incognita* under greenhouse conditions compared to four nematicides (Oxamyl 24% SL, Fosthiazates 75% EC, Ethoprophos N40% EC and Fenamiphos 40% EC).

2. Material and methods

2.1. Preparation of Root-knot Nematode, Meloidogyne incognita Culture:

According to (El-Ashry et al., 2021), susceptible tomato cultivar (Super Strain B) was used to propagation of *M. incognita* by using a single egg mass of previous identified pure inoculum of *M. incognita* under greenhouse conditions. Infected tomato roots were cut to small parts then eggs of *M. incognita* extracted by using 0.5% sodium hypochlorite (180 ml water +20 ml Clorox). Collected eggs were immediately washed by distilled water and incubated in Petri dishes at 26 ± 2 °C until hatching. A micropipette was used to gather newly hatched juveniles.

2.2. Pesticides

The recommended application rates of four commercial formulations of registered nematicides (Oxamyl 24% SL, Fosthiazate75% EC, Ethoprophos 40% EC and Fenamiphos 40% EC) (Table 1) were used to conducted current experiments to compare with co-friendly materials in controlling RKN. The registered nematicides were purchased from the Central Laboratory of Pesticides, Dokki, Giza, Egypt. As well as, a pot experiments were conducted in the Plant Protection Department, Faculty of Agriculture, Zagazig University.

2.3. Preparation of tested organic acids and plant extract:

Two organic acids (salicylic and humic) were obtained from were obtained from Gomhoria company, Egypt, besides linseed oil extract or flaxseed oil is made from the flax plant's dried, matured seeds (*Linum usitatissimum*). Linseed oil is dried oil that reacts with oxygen in the air to polymerize into a solid state (Brendel, 2013).

Two organic acids (salicylic and humic) besides plant extract, *L. usitatissimum* were separately prepared by using 1 g from two organic acids and 1 ml of stock solution of *L. usitatissimum* that added to 1 L distilled water, respectively.

2.4. Greenhouse experiment setup and procedures

Plastic pots 13 cm in diameter were employed in the greenhouse, filled with 1300 g sterilized sandy soil (72.5% sand, 12.5% clay, 12.1% silt), 120 g peat moss, and 3 mg urea fertilizer per kilogram of soil. After one week of planting pepper (*Capsicum annuum* L.) cultivars (Super mard, Super amarr, Super noura and Werta), The experimental pots were put in a random pattern with nine different treatments: (1) plants without infection with *M. incognita* (negative control), (2) plants infected with *M. incognita* (positive control), (3) plants infected with *M. incognita* + 10 ml of RD from oxamyl (24% SL), (4) plants infected with *M. incognita* + 10 ml of RD from Fosthiazate (75% EC), (5) plants infected with *M. incognita* + 10 ml of RD from Ethoprophos (40% EC), (6) plants infected with *M. incognita* + 10 ml of RD from fenamiphos (40% EC), (7) plants infected with *M. incognita* + 10 ml of stock solution of plant extract *L. usitatissimum*, (8) plants infected with *M. incognita* + 10 ml of humic acid, (9) plants infected with *M. incognita* + 10 ml of Salicylic acid. Every plant seedling in treatments (2–9) was infected with a combination of recently J2 hatched juveniles of *M. incognita* by pipetting 3 ml of the inoculum suspension into proper holes around each pepper cultivar root system. Stock solution of plant extract, *Linum usitatissimum* and two organic acids, humic and salicylic acid were applied twice in 5 cm of top area of Pot soil around plants roots, one of them after three days of seed plants and other one at the inoculation of nematode suspension. Each treatment was replicated five times. Clean tape water was used to irrigate pepper plants as needed, and the glasshouse bench was stored at a temperature of 25 °C. The test was ended after sixty days of nematode inoculation, and pepper plants were carefully removed from pots and bathed in clean water for 30 min. Then, plants warped by tissue papers to conducted the following plant and nematode parameters. Plant growth parameters data, involving fresh and dry shoot weight (g) and root length (cm) were estimated. Moreover, number of galling/plant and number of egg masses/plant were assessed. Nematode extraction was performed on 100 g soil samples using a combination of sieving and the Baermann trays approach (Hooper, 1986). Reproduction factor (RF) was assessed according to the following equation:

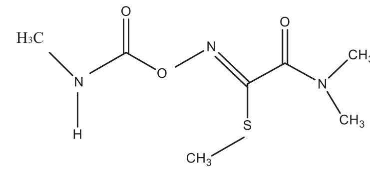
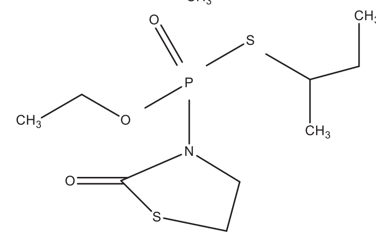
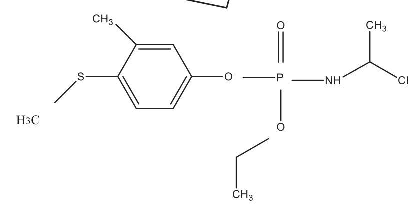
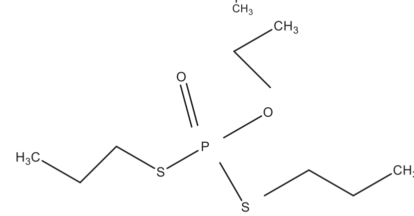
$$RF = FP/FI$$

whereas, FP is final population and IF is an initial population.

The present equations and the percentage of decrease or rise in the parameters imputed to "negative or positive" control values were used:

$$\text{Reduction }(\%) = \frac{\text{Control - Treated}}{\text{Control}} \times 100$$

Table 1
Tested nematicides against *Meloidogyne incognita.*

Nematicide	Trade name	Chemical class	Chemical structure
Oxamyl	Vydate 24% SL	Oxime carbamate	
Fosthiazate	Crinkle 75% EC	Organophosphorus	
Fenamiphos	Lagona 40% EC	Organophosphorus	
Ethoprophos	Smart N 40% EC	Organothiophosphate	

$$\text{Increase } (\%) = \frac{\text{Treated - Control}}{\text{Control}} \times 100$$

2.5. Statistical analysis

Statistically, the fully randomized design was implemented for laboratory experiments. The data were analyzed using MSTAT version4, and the analysis of variance and means were compared using Duncan's multiple range test at a probability of $\leq$0.05.

3. Results

Effects of four nematicides, plant extract *L. usitatissimum* and organic acids (humic and salicylic acids) on reproduction of *M. incognita* and plant growth parameters in four lines of pepper.

3.1. Soil and root parameters

Humic and salicylic acids with nematicides exhibited strong nematicidal activity against *M. incognita* J2 in pots soils, number of galls/plant and number of egg masses/plant after 60 days of application by recommended rates and stock solution of tested materials in soil of all tested lines of *C. annuum* L (Table 2). In Super Mard Cultivar, The treatment with 4 ml/plant of each organic acids (humic and salicylic acid) and 40 μL/plant of linseed oil extract, *Linum usitatissimum,* showed comparable nematicidal activity with 44.83%, 70.33%; 48.27%, 80.54% and 46.20%, 65.46% galls and egg masses per plant, respectively. As well as the reduction percentage of J2/100 g number soil in pots treated with humic acid, salicylic acid and plant extract, *L. usitatissimum* were 46.83%, 54.91% and 36.43% respectively. Whereas, the percentage of galls, egg masses and J2 number reduction were 77.94%, 90.51%, 79.15%; 52.41%, 81.27%, 73.98%; 51.04%, 79.81, 69.37% and 68.96%, 87.35%, 78.04% with oxamyl, fosthiazate, ethoprophos and fenamiphos, respectively (Table 2). Whilst, in the Super Amarr cultivar, humic acid, salicylic acid, and linseed oil exhibited nematicidal activity with 75.28%, 38.85%, 59.51%; 80.83%, 47.92%, 68.46%; 65.00%, 19.83%, 52.86% galls, egg masses and number of J2/100 g per plant, respectively. Whilst, tested four nematicides were more effectiveness against *M. incognita* reproductive parameters than organic acids. Number of galls, egg masses and J2 number were 92.22%, 76.86%, 84.04%; 85.28%, 54.54%, 77.95%; 81.39%, 52.88%, 76.01% and 88.61%, 67.76%, 81.15% with oxamyl, fosthiazate, ethoprophos and fenamiphos, respectively. Moreover, in Super Noura cultivar**,** high numbers of J2/100 g soil were found in soil pots treated with humic acid, salicylic acid, and *L. usitatissimum.* Galls numbers, egg masses and J2/100 g soil were 38.00, 36.00, 41.00; 26.33, 29.00, 32.33 and 20.00, 24.66, 21.88 with *L. usitatissimum,* humic and salicylic acids, respectively. Whereas the same parameters in pots treated with oxamyl, fosthiazate, ethoprophos and fenamiphos were 11.00, 18.66, 8.66; 19.66, 11.33, 16.00; 22.00, 21.66, 21.33 and 13.66, 14.66, 11.33, respectively. Whereas, Oxamyl achieved the maximum effect in reducing number of gall (6.66) by 93.88%, number of egg masses (8.00) by 76.91% and J2/100 g soil (6.68) by 89.28%, respectively in Werta cultivar. The equivalent values in treatments of humic and salicylic acids were 24.33 by 77.67, 24.66 by 28.85%, 21.66 by 65.24 and 20.00 by 81.65%, 22.66 by 34.62%, 18.66 by 70.06%, respectively.

On the other hand, the soil parameters reprehensive in the reproduction factor of nematode. Table 3 showed that humic acid, salicylic acid and plant extract, *L. usitatissimum* are 0.531, 0.450 and 0.635 in Super Mard Cultivar. Whilst, the reproduction factor of nematode are 0.208, 0.260, 0.306, and 0.219 with oxamyl, fosthi-

Table 2
Individual effects of four nematicides (Oxamyl, Fosthiazate, Ethoprophos and Fenamiphos), plant extract (*Linum usitatissimum*) and organic acids (humic and salicylic acid) on galls, egg masses and J2 of *Meloidogyne incognita* of tested pepper cultivars.

Treatment	Super Mard cultivar			Super Amaar cultivar			Super Noura cultivar			Werta cultivar		
	Root parameters (% reduction)											
	No. of galls/plant	No. of egg masses/plant	No. of IJs/ 100 g	No. of galls/plant	No. of egg masses/plant	No. of IJs/ 100 g	No. of galls/plant	No. of egg masses/plant	No. of IJs/ 100 g	No. of galls/plant	No. of egg masses/plant	No. of IJs/ 100 g
Healthy plants	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Control + *M. incognita* 1000 IJs	57.67^{a}	137.00^{a}	48.33^{a}	68.33^{a}	40.33^{a}	120.00^{a}	71.33^{a}	41.00^{a}	126.66^{a}	62.33^{a}	34.66^{a}	109.00^{a}
Oxamyl	12.02^{bc} (79.15)	13.00^{ef} (90.51)	10.66^{c} (77.94)	10.90^{e} (84.04)	9.33^{f} (76.86)	9.33^{f} (92.22)	8.66^{f} (87.85)	18.66^{e} (54.48)	11.00^{e} (91.31)	6.68^{f} (89.28)	8.00^{d} (76.91)	6.66 ^{g} (93.88)
Fosthiazate	15.00^{bc} (73.98)	25.66^{de} (81.27)	23.00^{b} (52.41)	15.06^{de} (77.95)	18.33^{d} (54.54)	17.66^{de} (85.28)	16.00^{e} (77.56)	11.33 ^{g} (72.36)	19.66^{d} (84.47)	11.66^{e} (81.29)	13.66^{c} (60.58)	17.33^{e} (84.10)
Ethoprophos	17.66^{bc} (69.37)	27.66 ^{cd} (79.81)	23.66^{b} (51.04)	16.39^{de} (76.01)	19.00^{d} (52.88)	22.33^{d} (81.39)	21.33^{d} (70.09)	21.66^{de} (47.17)	22.00^{d} (82.63)	13.33^{e} (78.61)	16.33^{c} (52.88)	19.33^{de} (82.26)
Fenamiphos	12.66^{bc} (78.04)	17.33^{de} (87.35)	15.00^{c} (68.96)	12.88^{de} (81.15)	13.00^{e} (67.76)	13.66^{ef} (88.61)	11.33^{f} (84.11)	14.66^{f} (64.24)	13.66^{e} (89.21)	9.00^{f} (85.56)	8.66^{d} (75.01)	11.00^{f} (89.90)
Linum usitatissimum	36.66^{ab} (36.43)	47.33^{b} (65.46)	26.00^{b} (46.20)	32.21^{b} (52.86)	32.33^{b} (19.83)	42.00^{b} (65.00)	41.00^{b} (42.52)	36.00^{b} (12.19)	38.00^{b} (69.99)	30.66^{b} (50.81)	33.33^{a} (3.83)	34.33^{b} (68.50)
Humic acid	30.66^{b} (46.83)	40.66^{bc} (70.33)	26.66^{b} (44.83)	27.66^{bc} (59.51)	24.66^{c} (38.85)	29.66^{c} (75.28)	32.33^{c} (54.67)	29.00^{c} (29.26)	26.33^{c} (79.21)	21.66^{c} (65.24)	24.66^{b} (28.85)	24.33^{c} (77.67)
Salicylic acid	26.00^{b} (54.91)	26.66^{cde} (80.54)	25.00^{b} (48.27)	21.55 ^{cd} (68.46)	21.00^{d} (47.92)	23.00^{d} (80.83)	21.88^{d} (69.32)	24.66^{d} (39.85)	20.00^{d} (84.20)	18.66^{d} (70.06)	22.66^{b} (34.62)	20.00^{d} (81.65)

Each value is a mean of five replicates.
Means with the same letter are not significantly different ($p \le 0.05$).
$\text{Reduction } (\%) = \frac{\text{Control - Treated}}{\text{Control}} \times 100$

azate, ethoprophos and fenamiphos, respectively (Table 3). Whereas, in the Amarr cultivar, the reproduction factor of nematode for humic acid, salicylic acid and linseed oil are 0.404, 0.315 and 0.471. Whilst, the reproduction factor of nematode are 0.016, 0.22, 0.247, and 0.188 with oxamyl, fosthiazate, ethoprophos and fenamiphos, respectively. Also, the reproduction factor of nematode for humic acid, salicylic acid and linseed oil are 0.453, 0.306 and 0.574. Whilst, the reproduction factor of nematode are 0.121, 0.224, 0.299 and 0.158 with oxamyl, fosthiazate, ethoprophos and fenamiphos, respectively in Super Noura. Finally, in Werta cultivar, the reproduction factor of nematode for humic acid, salicylic acid and linseed oil are 0.347, 0.299 and 0.491. Whilst, the reproduction factor of nematode are 0.107, 0.187, 0.213 and 0.144 with oxamyl, fosthiazate, ethoprophos and fenamiphos, respectively.

3.2. Plant growth parameters

Table 4 displayed that humic and salicylic acid overwhelmed *L. usitatissimum* in fresh and dry weight of pepper plants. Moreover, two organic acids surpassed four nematicides in enhancement of plant growth parameters. For instance, the increasing percentage of fresh and dry shoot weight for Super Mard cultivar treated with humic, salicylic acids and *L. usitatissimum* were 40.22%, 20.49%; 40.59%, 29.65% and 31.64%, 7.91% compared with 8.58%, 11.49%; 32.46%, 7.91%; 32.83%, 9.62% and 34.47%, 8.54% and fenamiphos for oxamyl, fosthiazate and ethoprophos, respectively. Furthermore, in positive control pots (infected pepper plants), the parallel values of fresh and dry shoot weight were 13.40 and 6.44 g, respectively. These data indicated that humic and salicylic acids have remarkably higher nematicidal activity than plant extract and while four nematicides were the most efficiency. Whilst, fresh and dry weight of pepper shoot Super Amarr cultivar treated with two organic acids were 18.96, 8.20; 16.12, 6.72; 14.83, 7.90 g with humic, salicylic acid and linseed oil compared with 15.00, 7.71; 14.68, 7.53; 14.90, 7.69; 18.01,7.06 g with oxamyl, fosthiazate, ethoprophos and fenamiphos, respectively. Furthermore, parallel values of fresh and dry shoot weight in infected pepper plants with *M. incognita* (positive control) were 14.41 and 6.52 g, respectively. Current data indicated that four nematicides were more effectiveness than humic and salicylic acids in reduced galls and egg masses of *M. incognita* whereas extract *L. usitatissimum* was the least effective one. The type of organic acids and the application of nematicides affected the pepper shoot weights at each of recommended concentration in Super Noura whereas, Application of humic and salicylic acids resulted in enhance in fresh and dry shoot weight as compared with positive control treatment. However, compared to the non-treated Super Noura cultivar (positive control), adding humic and salicylic acids always resulted in a higher shoot weight more than those of nematicides. Fresh and dry pepper shoot in treatments of humic, salicylic acids and *L. usitatissimum* were 18.85, 7.66; 18.86, 7.98 and 17.82, 7.61, whilst, in treatments of oxamyl, fosthiazate, ethoprophos and fenamiphos were 18.47, 7.59; 18.06, 7.33; 17.95, 7.15 and 18.38, 7.48, respectively compared with 17.42 and 6.15 in infected pepper plants. Moreover, fresh and dry weight of pepper shoot in treatments received two organic acids were 19.43 g (23.60%), 7.89 g (37.45%) and 20.97 g (33.39%), 8.39 g (46.16%) with humic and salicylic acid compared 19.26 g (22.51%), 8.00 g (39.37%); 18.38 g (16.92%), 7.85 g (36.75%); 17.93 g (14.05%) 7.71 g (34.32%) and 18.87 g (20.03%),7.88 g (37.28%) with oxamyl, fosthiazate, ethoprophos and fenamiphos, respectively. Likewise, equivalent values of fresh and dry shoot weight in infected Werta cultivar with *M. incognita* were 15.72 and 5.74 g, respectively. These data indicated that four nematicides were more effectiveness than humic and salicylic acids which achieved considerable reduction of root galls, egg masses numbers and J2 numbers in pots soils.

Table 3
Evaluation of four nematicides (Oxamyl, Fosthiazate, Ethoprophos and Fenamiphos), plant extract (*Linum usitatissimum*) and organic acids (humic and salicylic acid) on final population and reproduction factor of *Meloidogyne incognita* in different pepper cultivars.

Treatment	Super Mard cultivar			Super Amaar cultivar			Super Noura cultivar			Werta cultivar		
	Soil parameters											
	Finial population	% reduction	RF (FP/IP)	Finial population	% reduction	RF (FP/IP)	Finial population	% reduction	RF (FP/IP)	Finial population	% reduction	RF (FP/IP)
Healthy plants	0.00	0.00	–	0.00	0.00	–	0.00	0.00	–	0.00	0.00	–
Control + *M. incognita* 1000 IJs	749.71	0.00	–	888.29	0.00	–	927.29	0.00	–	810.29	0.00	–
Oxamyl	156.26	79.15	0.208	141.70	84.04	0.016	112.58	87.85	0.121	86.84	89.28	0.107
Fosthiazate	195.00	73.98	0.260	195.78	77.95	0.220	208	77.56	0.224	151.58	81.29	0.187
Ethoprophos	229.58	69.37	0.306	219.70	75.26	0.247	277.29	70.09	0.299	173.29	78.61	0.213
Fenamiphos	164.58	78.04	0.219	167.44	81.15	0.188	147.29	84.11	0.158	117.00	85.56	0.144
Linum usitatissimum	476.58	36.43	0.635	418.73	52.86	0.471	533	42.52	0.574	398.58	50.81	0.491
Humic acid	398.58	46.83	0.531	359.58	59.51	0.404	420.29	54.67	0.453	281.58	65.24	0.347
Salicylic acid	338.00	54.91	0.450	280.15	68.46	0.315	284.44	69.32	0.306	242.58	70.06	0.299

RF = Reproduction factor, FP = final population, IP = initial population.
$\text{Reduction } (\%) = \frac{\text{Control - Treated}}{\text{Control}} \times 100$

Table 4
Changes in tested lines of pepper plant growth parameters after treatment four nematicides (Oxamyl, Fosthiazate, Ethoprophos and Fenamiphos) plant extract (*Linum usitatissimum*) and organic acids (humic and salicylic acid) on galls, egg masses and J2 of *Meloidogyne incognita* under greenhouse condition.

Treatment	Super Mard cultivar		Super Amaar cultivar		Super Noura cultivar		Werta cultivar	
	Plant growth parameters (% increase)							
	Fresh shoot weight (g)	Dry shoot weight (g)	Fresh shoot weight (g)	Dry shoot weight (g)	Fresh shoot weight (g)	Dry shoot weight (g)	Fresh shoot weight (g)	Dry shoot weight (g)
Healthy plants	20.40^{a}	8.66^{a}	20.75^{a}	8.93^{a}	20.60^{a}	8.78^{a}	21.97^{a}	9.19^{a}
Control + *M. incognita* 1000 IJs	13.40^{c}	6.44^{f}	14.41^{b}	6.52^{eg}	17.42^{de}	6.15^{g}	15.72^{e}	5.74^{e}
Oxamyl	14.55^{bc} (8.58)	7.18^{d} (11.49)	15.00^{b} (4.09)	7.71^{d} (18.25)	18.47^{c} (6.02)	7.59^{c} (23.41)	19.26^{cd} (22.51)	8.00^{c} (39.37)
Fosthiazate	17.75^{abc} (32.46)	6.95^{e} (7.91)	14.68^{b} (1.87)	7.53^{e} (15.49)	18.06^{d} (3.67)	7.33^{e} (19.18)	18.38^{ef} (16.92)	7.85^{c} (36.75)
Ethoprophos	17.80^{abc} (32.83)	7.06^{de} (9.62)	14.90^{b} (3.40)	7.69^{d} (17.94)	17.95^{e} (3.04)	7.15^{f} (16.26)	17.93^{f} (14.05)	7.71^{c} (34.32)
Fenamiphos	18.02^{abc} (34.47)	6.99^{de} (8.54)	18.01^{ab} (24.98)	7.06^{f} (8.28)	18.38^{c} (5.51)	7.48^{d} (21.62)	18.87^{e} (20.03)	7.88^{c} (37.28)
Linum usitatissimum	17.64^{abc} (31.64)	6.95^{e} (7.91)	14.83^{b} (2.91)	7.90^{c} (21.16)	17.82^{e} (2.29)	7.61^{c} (23.73)	18.00^{f} (14.50)	7.77^{c} (35.36)
Humic acid	18.79^{ab} (40.22)	7.76^{c} (20.49)	18.96^{ab} (31.57)	8.20^{b} (25.76)	18.85^{b} (8.20)	7.66^{c} (24.55)	19.43^{c} (23.60)	7.89^{c} (37.45)
Salicylic acid	18.84^{ab} (40.59)	8.35^{b} (29.65)	16.12^{ab} (11.86)	6.72^{g} (3.06)	18.86^{b} (8.26)	7.98^{b} (29.75)	20.97^{b} (33.39)	8.39^{b} (46.16)

Each value is a mean of five replicates.
Means with the same letter are not significantly different ($p \leq 0.05$).
$\text{Increase } (\%) = \frac{\text{Treated - Control}}{\text{Control}} \times 100$

4. Discussion

Noteworthy aspects of these experiments were that both organic acids (humic and salicylic acids) and tested nematicides effectively reduced the galls formation, egg masses and number of J2 in pots soil; only oil plant extract (*L. usitatissimum*) was tended to be less effective versus RKN (*M. incognita).*

These results suggest that eco-friendly materials particularly humic and salicylic acids besides *L. usitatissimum* may play varying roles in reducing reproduction of *M. incognita* across pots soils that are favorable to specific pepper cultivars. Nevertheless, it would be valuable to study the effects of resistance or highly susceptible pepper cultivars as main option on RKN, *M. incognita*, reproduction in the absence and presence of eco-friendly materials to select proper cultivars of pepper plants.

HA and SA have higher nematicidal activity contra *M. incognita* J2 and eggs than oil of plant extract, *L. usitatissimum*. Current results are harmony with El-Sherif et al. (2015), used HA and SA at two concentration 0.1& 0.05% compared to oxamyl versus *Meloidogyne incognita* infecting tomato cv.9065FI and found that salicylic acid outperformed other treatments in terms of percentage increases in total plant fresh weight and shoot dry weight, as well as the lowest percentage reductions in final nematode population and number of egg masses.

The SA elicitor controls the activity of enzymes like polyphenol oxidase and peroxidase, The SA elicitor controls the activity of enzymes like polyphenol oxidase and peroxidase, that are key part of plant defenses versus biotic and abiotic stressors (Hayat et al., 2009; Zhao et al., 2009; War et al., 2011). Increase tested plants resistance treated with organic acids because of the enhancement activity of phenylalanine ammonialyase and this reaction enhances phenolic production, which is linked to increased plant defense resistance (Zinovieva et al., 2011; Díaz-Rivas et al., 2018; Khoshfarman-Borji et al., 2020). Moreover, Abd El-Kareem, 2007 mentioned to increased chitinase activity aids HA in combating the negative effects of chocolate spot and rust infections in faba bean plants, as well as enhanced plant growth indices and nutrient and water absorption (Chen et al., 2004).

Nemours studies evaluated essential plant oils efficacy against RKN, *M. incognita*, in vitro (eggs and infective juveniles) and in vivo on vegetable plants (such as tomato, pepper, and cucumber). Previous research has shown that essential oils comprising major components have fumigant toxicity that varies depending on the chemical makeup of the plant species. The nematicidal mechanisms of essential oils of medicinal plants as nematicides are not clear. Various plant extracts exhibited acetylcholinesterase inhibition. Others maybe cause disruption in cell membrane of nematodes and affected its permeability (Kayani et al., 2012, Abd El-Aal et al., 2021).

Refaat et al. (2020) In vitro experiments on egg hatching and juvenile mortality of the root-knot nematode Meloidogyne incognita revealed that the oils investigated had a nematicidal effect on egg masses, free eggs, and juveniles of M. incognita. El-Ashry et al. (2021) reported that researchers employed a mixture of abamectin, Purpureocillium lilacinum, rhizobacteria, and botanicals to control Meloidogyne incognita on tomato compared to oxamyl. Various applications by stock solution of seed oils and botanicals only maybe achieve proper phytonematodes control management as displayed from current study under greenhouse conditions which agreement with El-Ashry et al., 2021 and Refaat et al., 2020.

Organic acids have low molecular weights and are found in low quantities in the soil, and low molecular weight organic acids tend to vanish quickly (McBride et al., 2000). As a result, the transitory character of organic acids appears to impair their efficiency in preventing plant pathogen-caused illnesses (Tabarant et al., 2011; Seo and Kim, 2014; El-Sherif et al., 2015). As a result, the use of organic acids to combat RKNs is not actively encouraged.

Given the amount of organic acids used in our study, the high control cost of using a mixture of organic acids would be roughly equal to the cost of a low-effective agrochemical nematicide, which could result in economic disadvantages for organic acids and plant extracts, which have generally failed to provide adequate nematode control (Upadhyay and Rai, 1988, Aioub et al., 2021).

5. Conclusion

Humic and salicylic acid were found to reduce gall formations, egg masses and number of J2 of *M. incognita*. When employed in correct proportions with good climatic circumstances and resistant plant cultivars, it can be regarded a viable alternative to chemical nematicides, as well as the creation of stable formulations of active organic acids for RKN control. However, further RKN control studies in the field are needed to establish the effectiveness of organic acids or plant extracts, as well as the right practical concentration and control cost, as compared to nematicides (oxamyl, fosthiazate, ethoprophos, and fenamiphos).

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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