EFFECT OF CERTAIN Aloe vera PLANT PRODUCTS ON ROOT-KNOT NEMATODE, Meloidogyne incognita INFECTED BANANA PLANTS
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ABSTRACT

The efficacy of Aloe vera plants as oil and water leaf extract alone or in combination were singly evaluated against Meloidogyne incognita either under laboratory or greenhouse conditions. With respect to laboratory test, results of the four concentration tested of Aloe vera products i.e. 1:5, 1:10, 1:50 and 1:100ml at $20\pm1^{\circ}$ C and $30\pm1^{\circ}$ C against M.incognita (J₂) showed that all concentration were highly effected second stage (J₂) mortality percentage. Regarding the efficacy of the same plant products at the best concentration 1:5 and 1:10 on M.incognita infecting banana plant under greenhouse, results indicated that root gall index and rate of nematode reproduction were decreased, especially in their application after nematode inoculation, followed by those before nematode inoculation, obviously oxamyl 24% indicated gave the best effect on nematode reduction when compared with treatments of Aloe vera plant products tested.

Keywords: Aloe vera oil, water leaf extract root-knot nematode, *M. incognita*, banana plants, nematode management, oxamyl 24%.

INTRODUCTION

Root-knot nematodes are the most economically important group of plant parasitic nematodes worldwide, attacking nearly every crop grown (Sasser and Freckman, 1987). These highly successful pathogens cause an estimated average crop loss of 5% worldwide and are one of the major obstacles to production of adequate supplies of food in many developing nations. The chemical nematicides which are common and recommended in the current market are highly toxic and expensive. Therefore, the need have crated to use safe and compounds with nematicideal properties such as alkaloids, phenois, saponin, thienyls, diterpenes and polyacetylenes which have been found in plants, (Gammers and Barker, 1988). Aloe plant is one of plants in the family *Liliaceae* have inhibitory effect against pests. This aloe plant is the source of two herbal preparations: aloe gel (AG) and aloe latex (AL). Aloe gel is often called *Aloe vera* and referes to the clear gel or mucilaginous.

Substance produced by parenchymal cells located in the central region of the leaf. The gel is composed mainly of water (99%) and mono- and polysaccharides (25%) of the dry weight of the aloe gel (Shelton 1991). Aloe gel also contains lignan, salicylic acid, saponins, and triterpenoide (Klein and Penneys 1988 and Sabeh and Wright., 1993). The gel also contains, different kinds of vitamins i.e. (A,C,E, and B12) as well as minerals (Shelton) 1991). In addition, anoval anti-inflammatory compounds, C-glucosyl chromone, has recently been isolated from AG (Hutter et al 1996). More over, the effect of vitamins on the activies of nematodes had also demonstrated by many, investigators such as El-Zawohry and Hamada, (1994) and Bekhiet (2003)

who reported a significant reduction on root -knot nematode reproduction due to the application of vitamins. Also polysaccharides had demonstrated to be effective against root-knot nematode (M. javanica) reproduction Bekhiet (2003), as well as El-Deeb et al. (2001) who reported the effect of salicylic acid on root-knot nematode M. incognita. Therefore, these naturally nematicidal compounds involved in such extracts of Aloe vera undoubtedly can suppress the nematade build up in the soil. The greatest reduction in root-knot nematode Meloidogyne incognita was achieved with shopped leaves of Aloe barbadensis (syn. Alae vera). In vitro Randy and Haseeb (1988); Showed that extracts of root and shoot of Aloe barbadensis were tested against M. incognita. There was strong nematicidal activity by all extracts, and highest mortality (100% occurring with the standard dilution of root extracts of A. barbadensis after 12 h. Also, Sweelam (1989); showed that root exudates of aloe (Aloe arborescens) were compared with fenamiphas (15kg/fed) for control of M. javanica on tomatoes in a pot experiment, sixty days after inoculation, nematode populations were reduced by 87.9% and 64.8% for fenamiphas and aloe respectively. The reduction in egg masses was 93.1% and 68.6% for fenamiphas and aloe, Siddiqui and Alam (1997) reported that greatest reduction in root-knot nematode was achieved with Aloe barbadensis. Terasawa et al., (2001); showed that the greatest reduction in root-knot index, juveniles, egg masses, and egg/egg mass were achieved with high antioxsative activity in aloe, Singh et al., (2002) reported that Aloe barbadensis was evaluated against M. incognita. The objective of the present study was to measure the effect of water leaf extract as well as aloe oil plants against M. incognita in laboratory and on banana plants under greenhouse conditions.

MATERIALS AND METHODS

Water leaf extract:

After the execution of spines from the both sides of aloe leaves, 1Kg of fresh leaves were cut into small pieces, moved into glass jar of a blender with 100 ml water and hit in the blender under the highest speed for four minutes. Suspension was filtrated through a muslin cloth. The stock solution of leaf extract was kept in plastic bottles in a refrigerator at(2-4°C). Aloe oil:

In this test *Aloe* vera oil is a commercial product was used at different concentrations (1:5,1:10,1:50 and 1:100) in the vitro while (1:5 and 1:10) in the vivo experiments.

Nematicide treatment Trade name vydate, ingrediate 24% (oxamyl), added by rate 0.5 ml before planting. In vitro tests:

The solution of leaf extract was diluted with water to different concentrations (1:5,1:10,1:50 and 1:100 ml.) the effect of concentration of leaf extract and oil were studied using petri plates, containing about 100 of second stage juveniles of M. incognita in 1 ml Water. Nematode suspension in distilled water (100 J₂/1 ml) was taken as control. The treatments were replicated six times. Half petri dishes were incubated at $20\pm1^{\circ}$ C as group and second half of petri dishes were incubated at $30\pm1^{\circ}$ C. Observation on

juvenile mortality were taken daily up to five days. The dilution of leaf extract and oil were added to dishes daily to prevent drying.

Determination and fractation of polysaccharides wich extracted from aloe leaves plants (Aloe vera):

Polysaccharides was determined in one gram fresh weight of *Aloe vera* as 26.5 mg/gm fresh weight, while reducing and non-reducing sugar content were 19.3 and 7.2 mg/gm frish weight, respectively. All analysis was done in Bio-technology lab., Plant Pathology institute, ARC, Egypt. The fractation and qulification of these polysaccharides should that seven types of ten tested types as follows; Glucose, Arbinose, Trehalose, Riffanose, Maltose, Mannose and Fractose (2.20, 1.11, 10.15, 19.35, 2.23, 5.39 and 3.39mg/gm fresh weight respectively).

Analysis of aloe leaves plants:

(Table 1) according Shelton (1991) and Sabeh and Wright (1993), Khein and Penneys (1998).

Table (1): Analysis of Aloe leaves plant

Contents of Aloe vera	Aloe gel
Formulation	Liquid _
Water	99%
Mono- and polysaccharides	25% of the dry weight of the gel
Monosaccharides	Mannase-6-phasphate
Common polysaccharide	Gluco-mannons (β-1,4 acetylated mannan Glucose and mannase Gluco alain
Anotehr contains	Lignan-salicylic acid, saponin, sterois and triterpenoids
Fresh gel contain	Protealytic enzyme carboxy-peptidase, glutathiane peroxidase, several isoymes of superoxide dismutase.
Vitamins	A.C,E, and B12
Minerals	Sodium, Potassium, calcium, magnesium manganes, copper, zine chramium and iron

Pot experiment:

The experiment was carried out under greenhouse conditions. The two formula of aloe plants (Aloe vera) (aloe oil and aloe leaf extract) as well as the nematicide oxamyl 24% E.C. were evaluated against M. incognita parasitizing banana plants cv. grandnan during spring season 2005. Two months old seedlings were transplanted into 25cm diam. clay pots filled with sterilized sandy soil. After three weeks, the pots were treated at two times of applications:

- 1.Before nematode inoculation, plants were assigned to single or combined treatments of aloe oil at rates 1:10 and 1:5 ml/plant by concentrations 10% and 20% respectively or leaves extract at the same rates and concentration. (Table 3).
- 2.After inoculation, the same treatments and concentration were repeated. All pots were inoculated with 2000 freshly hatched second stage juveniles of M. incognital pot, ten days before or after inoculation and treatments in concerning that the pervious treatments were added three time weakly as soil drench, but (oxamyl) 24% was added one time before planting at rate 0.5ml/pot. Each treatment was replicated three times. Pots were inoculated

and kept without any additions and severed as control. Pots were watered regularly and kept in a greenhouse at $25\pm5^{\circ}$ C in randomized block design sixty days after inoculation. The plants in the pots of each treatment were gently removed from pots. Root gall index values were estimated according to the following scale:0:0galls,1= 1-2 galls, 2 = 3-10 galls, 3 =11 - 30 galls, 4 = 31 - 100 galls and 5 = >100 galls (Taylor and Sasser, 1978). The numbers of juveniles in soil, galls, egg masses per root system, egg per egg mass, rate of reproduction and fresh weights (shoots and roots) of banana plants were recorded. The data were statistically analyzed by the method of Ducan's multiple range test (Duncans,1955).

RESULTS

In vitro test:

Observation on effect of oil and water leaf extracts of *Aloe vera* plant on mortality *M. incognita* juveniles is illustrated in Table (2). Data showed that the percentage of mortality increased by increasing concentration, exposure time and temperature degree. However the both concentrations 1:5 and 1:10 of each tested materials were more effective than the rest other concentrations. The highest concentration of oil and water leaf extracts gave pronounced to mortality through the exposure period ranging between 82 to 100 % at 20°C, while 100% mortality was observed during the exposure period at 30°C. Likely the concentration 1: 10 of both extracts gave ranging of mortality between 60 to 100% after 72 hours at 20°C whereas these materials gave 80-100% mortality after 48 hours at 30°C. In 1:50 concentration of oil aloe plant, mortality was ranging between 35 to100 after 96 hours at 20°C and 50 –100% mortality after 72 hours at 30°C. While the same concentration (1: 50) of water leaf extract mortality was 40 to 100% after 72 hours and 66 to 100% after 48 hours at 20°C and 30°C respectively.

Table (2): Percentage of mortality of juvenile root-knot nematode Meloidogyne incognita when treated with different concentration of Aloe vera.

Concentration of Albe yeld.											
Treatments	Temperature	% mortality after (hours)					% alive after (hours)				
Aloe vera	°C	24	48	72	96	120	24	48	72	96	120
1:5 ml	20	82	100	100	100	100	18	0	0	0	0
Oil: water	30	100	100	100	100	100	0	0_	0	0	0
1:10 ml	20	60	90	100	100	100	40	10	0	0	0
Oil: water	30	80	100	100	100	100	20	0	0	0	0
1:50 ml	20	35	75	85	100	100	65	25	15	0	0
Oil: water	30	50	85	100	100	100	50	15	0	0	0
1:100 ml	20	20	58	83	100	100	80	42	17	0	0
water leaf extract	30	25	60	90	100	100	75	40	10	0-	0
1:5 ml	20	86	100	100	100	100	14	0	0	0	0
water leaf extract	30	100	100	100	100	100	0	0	0	0	0
1:10 ml	20	75	90	100	100	100	25	10	0	0	0
water leaf extract	30	83	100	100	100	100	17	0	0	0	0
1:50 ml	20	40	90	100	100	100	60	10	0	0	0
water leaf extract	30	66	100	100	100	100	34	0	0	0	0_
1:100 ml	20	20	50	70	80	100	80	50	30	20	0
water leaf extract	30	40	70	90	100	100	60	30_	10	0	0
Control	20	0	0	0	0	0	100	100	100	100	100
Conto	30	0	0	0	0	0	100	100	100	100	100

Inoculum with 100 (J₂) for pateri dishes

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The lowest concentration 1:100 of both oil and water extracts showed delayed effaicacy against second stage juveniles, where the percentage of mortality reached 100% after 96 hours of exposure time. Nematode kept in distilled water (check) remained active over the period of 96 hours.

in pot experiment :

Data presented in Table (3) indicated that the tested concentration of oil and water leaf extracts had significant depressive effect on juveniles numbers in soil, number of egg mass, eggs / egg mass and root-gall index; when added to the soil singly and in combined treatments and whatever the treatments applied before or after nematode inoculation. However the tested nematicide (oxamyl 24%) was more suppressive on nematode development and gall formation in comparison to check treatment (nematode only). Data also revealed the reproduction rate was higher at soil treatment before nematode inoculation (3.94, 2.66, 2.01 and 1.21% respect.) than after nematode inoculation treatment (1.40, 0.70, 1.24 and 0.72 respect.).

Table (3): Effect of oil and water leaf extract for aloe added as soil drench on reproduction of the root-knot nematode Meloidogyne incognita infection banana c.v. grandnan under greehouse conditions.

under greehouse conditions. Nematode population Nemat-									
Treatments and time of application	Concentration/ plant (ml)	Root gall index (0-5)	Juvenile in soil/pot	No. of egg masses/ root	No. of egg/ Egg mass	Nemat- ode final popula- tion (PF)	Rate of nemotode reproduction (Pf/Pi)		
Aloe oil B A	1-10	3.7 B 2.6 C	28 BC 126 E	30B 12C	253BCD 220 D	7870 2766	3.94 1.40		
Aloe oil B A.	1-5	2.3 CD 1.6 D	260 BCD 86 E	22BC 6 C	230 BCD 213 D	5320 1364	2.66 0.70		
Water leaf extract B A	1-10	3.3 B 2.3 CD	293 B 146 DE	13 C 6 C	287 B 259 BCD	4024 2477	2.01 1.24		
Water leaf extract B A	1-5	2.2 CD 1.8 D	160 CDE 66 E	8 C 6 C	283 B 230 CD	2024 1446	1.21 0.72		
Oil and water leaf extract B A	1-10 +1-10	3.4 B 2.7 C	166 CDE 86 E	11 C 8 C	280 BC 263 B CD	3246 2190	1.62 1.10		
Oil and water leaf extract B A	1-5+1-5	2.7 C 2.3CD	100 E 60 E	9 C 6 C	260 BCD 220 D	2440 1380	1.22 0.69		
Oxymal 24%	0.5 ml.	1.3 D	53 E	6 C	210 D	1313	0.66		
Ckeck	0	4.8 A	933 A	122 A	542 A	67057	33.53		

Values in a columns followed by the same letter (s) are not significant (P= 0.05) according

to Dunco's multiple range test.

B = Before nematode inoculation

A= After nematode inoculation

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In single treatment:

In is apparent from observation that the higher concentration of both materials (1:5) ranked the first in reducing root-gall index as nematicidal then followed by water leaf extracts at concentrations 1: 5 of water extract before nematode inoculation and 1: 10 after nematode inoculation.

In combined treatment:

The reduction in juvenile numbers, egg mass, eggs / egg mass and rate of reproduction was observed in the higher concentration (1: 5 + 1: 5) was the most effective then followed by the next concentration (1: 10 + 1: 10).

Oxamyl treatment:

The nematicide treatment surpassed the all treatments of oil and water leaf extracts inducing the height reduction in numbers of the nematode in roots and soil.

Data in Table (4) show the effect of oil and water leaf extracts on banana growth in comparison with the nematicide, (oxamyl) 24%. It could be seen that, aloe plant extracts increased plant growth with increasing concentration in single treatments and gave significant increase in fresh weight of banana shoots and roots than the check. On contrary, the combined treatments in shoots and roots effect the less than in single treatment especially at treatment before nematode inoculation. On the other hand, (oxamyl) 24% gave the highest effect to plant growth than all aloe plant treatments.

Table (4): Growth response of banana c.v grandnan infected with the root-knot nematode *M.incognita* as influenced by application of oil and water leaf extract aloe added as soil drench under greenhouse condition.

Treatments and time of	Concentr-	Fresh weight (gm)						
Treatments and time of application	ation/plant (mi)	Shoot	% increase	Root	% increase			
Aloe oil	1-10			1				
B.	1	49.00 G	4.25	40.00 E	6.21			
A.	1	66.33 CDE	41.13	57.00 BC	51.35			
Aloe oil	1-5							
В.	}	53.00 EFG	7.10	49.66 CDE	31.86			
A.	1	71.00 BCD	51.10	64.00 AB	69.94			
Water leaf extract	1-10							
B.	1	79.00 ABC	68.10	67.00 AB	77.90			
A.	1	83.00 AB	76.59	68.33 AB	81.43			
Water leaf extract	1-5			1				
В.	1	82.00 AB	74.46	68.66 AB	82.31			
A .	1	88.00 A	87.23	71.66 A	90.28			
Oil and water leaf extract	1-10+1-10							
В.	1	50.00 FG	6.38	41.66 DE	10.62			
A.	<u> </u>	61.66 DEF	31.19	55.00 BCD	46.04			
Oil and water leaf extract	1-5+1-5							
B.	1	54.66 EFG	16.29	47.66 CDE	26.55			
A.	}	70.00 BCD	48.93	60.00 ABC	59.32			
Oxamy! 24 %	0.5 ml	90.00 A	91.48	66.00 AB	15.25			
Check	0	47.00 FG		37.66E				
Jalue in a column follower	by the same	letter (stare no	at cianifican	t (P= 0.05) a	ccording			

Value in a column followed by the same letter (s) are not significant (P= 0.05) according to Duncnm's multiple range test. B = Before nematode inoculation A = After nematode inoculation.

DISCUSSION

From the above mentioned data it can be concluded that usage of aloe, either as water leaf extract for aloe or aloe oil was successfully effective to suppress the development and reproduction of M. incognita on banana. These findings are in accordance with the findings of Randy and Hasseeb (1988), Sweelam (1989), Saleem et al., (1997), Sheals et al., (1998); and Singh et al., (2002); This nematicidal activity of some medicinal plants Saleem et al., (1997); Showed that Aloe vera (Aloe barbadensis) was tested for mematicidal activity against M. javanica in laboratory experiments and showed the highest nematicidal activity. Also Terasawa et al., (2001) showed that the antioxidative activity of water extracts from freash leaves of Aloe (Aloe nabilis), Singh et al., (2002) who found a aqueaus extract obtained from Aloe barbadensis was against M. incognita and showed that Aloe barbadensis exhibited the greatest nematicidal property. This treatments were tested on nematode larvae isolated from tomato were transferred to petri dishes containing extract solutions (1:10 or 1:100). Larval mortality was calculated after 24,48 and 72 h. Extract for Aloe barbadensis exhibited the greeted nematicidal activity.(Singh et al., 2002)

Also Sweelam (1989). Showed that root exudates of Aloe (Aloe arborescens) were compared with fenamiphas (15 Kg/fed) for control of M. javanica on tomato in a pot experiment. Nematode populations were reduced by 87.9% and 64.8% for fenamiphas and aloe respectively. While the reduction in egg masses was 93.1% for for fenamiphas and 68.6% for aloe, and the increasing in the effectiveness by the increasing of dosage or by the increasing in decomposition period may be attributed to accumulation of toxic compound of such material in the soil and subsequently become more effective against nematodes. Concerning the simulative effect of aloe oil and water leaf extract on the plant growth parameters (weight of shoots and roots), it can be explain as a result to the controlling of root infection by nematodes and subsequently improved the root uptake of elements and nutrients from the soil, as well as it's effect as a source of organic matter and to encourage the beneficial and antagonistic microorganisms in the soil. These findings are in accordance with findings of Terasawa et al., (2001). And this could be aloe to a nematicidal effect of these vitamins, salicylic acid and polysaccharides on this nematode (El-Zawohry and Hamada, (1994), El-Deeb, (2001) and Bekhiet; (2003) respectively. As a final conclusion, it can be observed that aloe plants include natural nematicidal compounds which is adequate to produce sufficient natural control to the plant parasitic nematodes and improve the plant growth when used as fresh organic amendments, or as a natural aloe oil. Also the present investigation mentioned promising data may be gave more attention to the nonchemical methods for the control of the nematode problems by safe, economic and less dangerous methods.

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تأثیر بعض منتجات نبات الصبار البلدی آلوی فیرا علی نیماتودا تعقد الجذور میلودوجینی انکوجنیتا التی تصیب نباتات الموز محمد علی بخیت محمد علی بخیت معهد بحوث أمراض النباتات مرکز البحوث الزراعیة -الجیزة - مصر

تم تقييم فعاليه نبات الصبار البلدى آلوى فيرا باستخدام كل من مستخلص الأوراق وزيت الصبار فى حاله منفرده أو مجتمعة ضد نيماتودا تعقد الجذور ميلودوجينى انكوجينيا تحت ظروف المعمل والصوبة وذلك باستخدام تركيزات مختلفة منهما ١٠٥ و ١٠٠١ و ١٠٠٠ وتحت درجات حرارة مختلفة تحت ظروف المعمل وتم استخدام الفضل المعاملات فى المعمل ١٠٥ و ١٠١٠ مقارنة مع المبيد النيماتودى الاوكساميل ٢٤% تحت ظروف الصوبة .

أظهرت النتائج في اختبارات المعمل أن كل المتركيزات الثرت بدرجة ملحوظة على الطور البيرقى الثاني لنيماتودا تعقد الجذور وكان لاستخدام افضل التركيزات تحت ظروف الصوبة تأنيرا واضحا في خفض عدد العقد النيماتودية ومعدل التكاثر وخاصة عند اضافه المعاملة بعد العدوى مباشرة يليها المعاملات قبل العدوى مباشرة بينما أعطى مبيد الاوكساميل ٢٤% أعلى تأثير بالمقارنة بجميع المعاملات المستخدمة لمنتجات الصبار البلدى.