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Allelopathic effect of dry leaves of lantana and guava for controlling root knot nematode, *Meloidogyne incognita* on cowpea and some associated weeds

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Abstract : Under screen house conditions, powdered dry leaves of lantana (*Lantana camara*) and guava (*Psidium guava*) either alone or in combination were used for investigating their allelopathy effect against root knot nematode, *Meloidogyne incognita* on cowpea (*Vigna unguiculata* (L.) Walp) cv. Baladi and two associated weeds namely *Chorchorus olitorius* and *Echinocloa colonum*. Significant ($P \leq 0.05$) reduction in dry weight of both *Chorchorus olitorius* and *Echinochloa colonum* was obtained by using leaf residues of *Lantana camara* or *Psidium guava* or their mixtures in comparison to unwedded control. The obtained results indicated that the tested treatments reduced the number of second stage juveniles in soil and roots, galls and eggmasses on roots of cowpea (*Vigna unguiculata* (L.) Walp) cv. Baladi compared to unwedded nematode- infected cowpea plants. Subsequently, they increased plant height, fresh weights of shoots and roots, dry weight of shoots and number of nodules compared to unwedded nematode- infected cowpea plants. Also, they increased number of pods/plant, weight of pods/plant, number of seeds/pod, weight of seeds/plant and weight of 100 seeds. Significant increase in carbohydrate content in the seeds of cowpea (*Vigna unguiculata*) was determined when applying leaf residues of *L. camara*, *P. guava* or their mixtures. No nematodes were found in the roots of the tested weeds. The results suggested the use of leaf residues of *Lantana camara* and *Psidium guava* mixtures as natural materials for controlling *Chorchorus olitorius* and *Echinochloa colonum* and root knot nematode, *Meloidogyne incognita.*

Key Words: Allelopathic effect, Lantana and guava plant residues, weeds, *Meloidogyne incognita*, cowpea.

Introduction

Meloidogyne spp. are major problem wherever cowpea is grown in the most parts of the world. The root-knot nematodes were estimated to cause losses ranging from 10% to 69%[1,2.] The nematodes also negatively affect nodulation in cowpea at low population and completely prevent nodulation at high nematode population in cowpea plants[3]. Weeds are considered to be serious problem competing for light, water, nutrients and space. As a result, they reduce plant growth and yield[4]. Weeds act as a reservoir for root knot nematodes[5] and some plant parasitic nematodes can reproduce on weeds[6,7,8,9]. [10]reported that weed removal at 35 days after sowing decreased the number of annual broad and grass weeds and subsequently inhibited the total number of nematodes in maize field. Nematode control is necessary in order to increase crop yield and provide selfsufficiency for food and industrial raw materials. Allelopathy can serve as selective biological pest management by releasing allelochemicals from different parts of living or plant materials[11]. Among the several botanical-based approaches in nematode decomposed management is the use of plant residues [2;13; 14,15,16,17]. The purpose of this research is to investigate the allelopathic potential of powdered dry leaves of guava, *Psidium guava* and lantana, *Lantana camara* on broad leaf weed (*Chorchorus olitorius*) and grass weed (*Echinocloa colonum*) growth and root-knot nematode, *Meloidogyne incognita* infecting cowpea plants.

Materials and Methods

For this investigation, 30-cm-diameter clay pots filled with 5 kg solarized sandy loam soil (1:1 w/w) on 18/5/2014 were prepared on a bench in screen house. Leaf residues were added before sowing for decomposition and other materials were added at the same time.. The following treatments mixed with the soil surface were applied as follows:

1. 40g/ kg powdered dry leaves of Lantana (*Lantana camara*).
2. 40g/ kg powdered leaves of guava (*Psidium guava*).
3. 30g/ kg powdered leaves of Lantana(L) +10g/ kg powdered leaves of guava(P).
4. 20g/ kg powdered leaves of Lantana(L) +20g/ kg powdered leaves of guava(P).
5. 10g / kg powdered leaves of Lantana(L) +30/ kg g powdered leaves of guava(P).
6. A nematicide, Carbofuran 10% G at the rate of 0.05g/pot (equivalent to 10 kg/feddan).
7. An herbicide, Fusilde at the rate of 0.005ml/pot (equivalent to 1L/feddan).
8. Unwedded (Infected)cowpea (control 1).
9. Weed-free (Healthy)cowpea(control 2).

Seeds of cowpea (*Vigna unguiculata* (L.) Walp) cv. Baladi seeds were sown at the same pots on 26/5/2014, ten days after adding the tested treatments to soil. Three seeds were sown per pot at a depth of 2 cm; plants were thinned to one per pot six days after emergence to ensure uniform plant vigor. At the same time, all pots were sown with constant weight of weed species (*Chorchorus olitorius* and *Echinocloa colonum*) seeds and the pots were watered regularly once a day.

After seven days of seed emergence, 1,000 freshly hatched juveniles of *Meloidogyne incognita*/pots were added to the pots. and each treatment was replicated five times. All pots were arranged in a completely randomized block design. Plants were uprooted on 9/10/ 2014, 120 days after inoculation. The nematodes in the soil were extracted by sieving and decanting methods[18]. Roots were gently washed by tap water to avoid adhering soil and divided into two halves. The first half was incubated in distilled water according to [19]. Another half was cut into small pieces in Petri dishes and stained by acid fuchsin lactophenol[20]. Numbers of galls and egg masses, on roots of cowpea, were counted using binocular microscope [21]. Plant growth and yield criteria of cowpea and weeds were measured.

Determination of carbohydrate contents*:*

Total carbohydrates were extracted from dry finely ground yielded seeds of cowpea according to [22] and estimated colorimetrically by the phenol-sulphoric acid method[23].

Statistical Analysis*:*

Data were statistically analyzed according to [24]. The least significant differences (LSD) were calculated at $P \leq 5\%$.

Results

Effect of plant residues of lantana and guava on dry weight of the studied weeds:

Table (1) shows significant ($P \leq 0.05$) reduction in dry weight of both *Chorchorus olitorius* and *Echinochloa colonum* due to treatments with leaf residues of *Lantana camara* or *Psidium guava* or their mixtures in comparison to unwedded control. The results reveal that the toxicity of leaf residues of the mixture of both *L. camara* and *P. guava* at 10gL+30gP/pot was higher against *C. Olitorius*, as its dry weight reduction reached about 81.8% of unwedded control. The corresponding result in *E. colonum* was about 66%. It worthy to mention that complete death of *C. olitorius* was obtained with using the herbicide fusilade at the recommended dose (1L/fed).

Table (1): Dry weights of *Chorchorus olitorius* and *Echinocloa colona* grown in association with *Vigna unguiculata* infected by root knot nematode, *Meloidogyne incognita* as affected by leaf residues of *Lantana camara*, *Psidium guava* and their mixtures at different rates.

Treatments	(Rate/pot)	Dry weight(g) of	
		C. olitorius	*E. colona*
Lantana Camara(L)	40g	23.45	22.70
Psidium guafa(P)	40g	20.55	19.10
Lantana Camara(L)+ *Psidium guafa* (P)	30g L+10g P 20g L+20g P 10g L+30g P	15.30 13.75 6.00	15.45 12.25 9.40
Carbofuran 10%G(Nematicide)	0.05g	26.20	27.50
Fusilde (Herbicide)	0.005ml	00.00	10.90
Unwedded (Infected)cowpea	-	32.90	26.85
Weed-free (Healthy)cowpea	-	-	-
LSD at 5%		1.11	1.21

Each value is average of 5 replicates. The least significant differences (LSD) were calculated at $P \leq 5\%$.

Table (2): Effect of leaf residues of *Lantana camara*, *Psidium guava* and their mixtures at different rates on root-knot nematode, *Meloidogyne incognita* infecting cowpea, *Vigna unguiculata*.

Treatment	Rate /pot	J_2 in soil /pot	Red. %	J_2 in roots/plant	Red. %	No. of galls	Red. %	No. of egg-masses	Red. %	General average of reduction %
Lantana camara (L)	40g	920	59.4	107	83.0	19	65.5	12	71.4	69.8
Psidium guava (P)	40g	520	77.1	173	72.5	16	70.9	10	76.2	74.2
L. camara + P. guava	30gL+10gP 20gL+20gP 10gL+30gP	677 758 550	70.1 66.6 75.7	60 83 117	90.5 86.8 81.4	12 13 11	78.2 76.4 80.0	8 6 5	81.0 85.7 88.1	80.0 78.9 81.3
Carbofuran10%G	0.05g	800	64.7	70	88.9	17	69.1	8	76.2	74.7
Fusilde herbicide	0.005ml	450	80.1	93	85.2	14	74.5	9	78.6	79.6
Unwedded (Infected)cowpea	-	2267	-	630	-	55	-	42	-	-
Weed-free(Healthy)cowpea	-	-		-	-	-	-	-	-	-
LSD at 5%	-	222.36	-	49.41	-	6.81	-	5.48	-	-

-Each value is average of 5 replicates. The least significant differences (LSD) were calculated at $P \leq 5\%$.

Effect of leaf residues of lantana and guava on root knot nematode:

Dry leaf residues of lantana and guava either alone or in combination reduced population density of root knot nematode, *M. incognita* as indicated by the number of second stage juveniles(J_2) in soil and roots, gall and eggmasses on roots of cowpea (Table 2). No nematodes were found in roots of the studied weeds. By calculating the general average of nematode reduction, it was found that the highest percentage nematode reduction (81.5%) was achieved by using the combined treatment of 10g *Lantana camara* (L) +30g *Psidium guava* (P) followed by that (80%) achieved by the combined treatment of 30gL+10gP, herbicide (79.6%) and the combined treatment of 20gL+20gP. Lantana as single treatment had the least general percentage nematode reduction t (69.8%). The other treatments were on bar in reducing nematode criteria.

Effect of leaf residues of lantana and guava on plant growth criteria:

Data in Table (3) illustrate the effect of the tested plant residues on the studied plant growth criteria. In general, the plant residues either alone or in combination increased plant height, fresh weights of shoots and roots, dry weight of shoots and number of nodules compared to unwedded nematode- infected cowpea plants. On the basis of the percentage plant growth vigor index potential, it was found that the highest percentage potential (196.3%) was achieved by using the combined treatment of 10g *Lantana camara*+30g *Psidium guava* followed by those of Fusilde herbicide, dry leaves of *Lantana camara*, Carbofuran and combined treatment of 20gL+20gP.

Table (3): Plant growth of cowpea, *Vigna unguiculata* infected by root knot nematode, *Meloidogyne incognita* as affected by leaf residues of *Lantana camara* and *Psidium guava* or their mixtures at different rates.

Treatment	Rate /pot	Plant height (cm)	Fresh weight of shoots (g)	Dry weight of shoots (g)	Fresh weight of roots (g)	no. of nodules	Plant vigor index	Pant growth vigor index potential %
Lantana Camara(L)	40g	77.0	123.7	44.0	19.5	33.0	59.4	167.3
Psidium guava(P)	40g	75.0	83.1	30.4	11.1	21.0	44.1	124.2
L. camara + P. guava	30gL+10gP	72.0	113.3	42.9	10.4	22.0	52.1	146.8
	20gL+20gP	82.0	116.0	46.8	12.4	29.0	57.2	161.1
	10gL+30gP	87.0	166.7	53.1	10.8	31.0	69.7	196.3
Carbofuran 10%G	0.05g	77.0	128.6	42.5	13.8	26.0	57.6	162.3
Fusilde herbicide	0.005ml	81.0	148.7	37.6	13.3	23.0	60.7	171.1
Unwedded(Infected) cowpea	-	63.0	62.4	25.9	8.4	18.0	35.5	100.0
Weed-free(Healthy)cowpea	-	93.0	117.7	29.8	14.3	43.0	41.0	115.5
LSD at 5%	-	14.9	59.6	18.2	4.5	4.6	2.08	-

-Each value is average of 5 replicates. The least significant differences (LSD) were calculated at $P \leq 5\%$.
- Plant vigor index= an average plant growth and nodule criteria.
-Plant growth vigor index potential%= plant vigor of each treatment /plant vigor of the highest one multiplied by 100.

Effect of leaf residues of lantana and guava on plant yield criteria:

Data in Table (4) illustrate the effect of the tested plant residues on the studied plant yield criteria. In general, the plant residues either alone or in combination increased number of pods/plant, weight of pods/plant, number of seeds/pod, dry weight of seeds/plant and weight of 100 seeds compared to unwedded nematode-infected cowpea plants. On the basis of the percentage yield vigor index potential, it was found that the highest percentage yield vigor potential (144.80%) was achieved by using the combined treatment of 20g Lantana+20g guava followed by that (139.78%) achieved by powdered dry leaves of Lantana. The least percentage yield vigor potential (108.36%) occurred in nematicide, Carbofuran treatment.

Table (4): Yield of cowpea, *Vigna unguiculata* infected by root knot nematode, *Meloidogyne incognita* as affected by leaf residues of *Lantana camara*, *Psidium guava* and their mixtures at different rates.

Treatment	Rate /pot	No. of pods/ plant	Weight of Pod /plant (g)	No. of seeds /pod	Dry weight of seeds/ Pod(g)	Dry weight of 100 seeds(g)	Plant yield vigor index	Plant yield vigor index potential%
Lantana camara(L)	40g	14.0	1.39	9	1.060	12.15	7.52	139.78
Psidium guava (P)	40g	10.0	1.50	8	1.020	11.43	6.39	118.77
L. camara + P. guava	30gL+10Pg	11.0	1.36	8	1.000	10.35	6.34	117.84
	20gL+20gP	17.0	1.28	8	0.990	11.68	7.79	144.80
	10gL+30gP	12.0	1.04	7	0.810	10.29	6.23	115.80
Carbofuran10%G	0.05g	10.0	1.09	7	0.830	10.25	5.83	108.36
Fusilde	0.005ml	16.0	0.91	6	0.760	10.76	6.89	128.07
Unwedded (Infected) cowpea	-	8.0	1.01	7	0.720	10.15	5.38	100.00
Weed-free(Healthy)cowpea	-	14.0	1.06	8	0.990	11.10	7.03	130.67
LSD at 5%	-	3.3	NS	NS	0.023	0.89	0.97	-

- Each value is average of 5 replicates. The least significant differences (LSD) were calculated at $P \leq 5\%$.
- Plant yield vigor index= an average plant yield criteria. Plant yield vigor index potential%= plant vigor of each treatment /plant vigor of the highest one multiplied by 100. NS= not significant.

Table (5). Carbohydrate content in cowpea, *Vigna unguiculata* seeds infected by root knot nematode, *Meloidogyne incognita* as affected by leaf residues of *Lantana camara* and *Psidium guava* and their mixtures at different rates.

Treatments	Rate /pot	Carbohydrates %
Lantana Camara (L)	40g	30.85
Psidium guava (P)	40g	34.99
Lantana Camara (L)+ *Psidium guafa* (P)	30g L+10g P 20g L+20g P 10g L+30g P	37.69 48.18 56.59
Crbofuran 10%G(Nematicide)	0.05g	26.93
Fusilde(herbicide)	0.005ml	45.68
Unwedded(Infected)cowpea	-	21.38
Weed-free(Healthy)cowpea	-	59.49
LSD at 5%	-	1.69

-Each value is average of 5 replicates. The least significant differences (LSD) were calculated at P ≤ 5%.

Effect of plant residues of **lantana and guava on carbohydrate content of the yielded seeds:**

The results in Table (5) indicate significant increase in carbohydrate contents in the seeds of cowpea when applying leaf residues of *L. camara*, *P. guava* or their mixtures compared to the contents in seeds of unwedded control. Maximum increase in carbohydrate contents was recorded in seeds yielded from the plants treated with the mixture of *L. camara* and *P. guava* at 10g+30g/kg soil. As expected, the seeds of weed -free plants recorded the highest amount of carbohydrate content.

Discussion

The results of the present study indicate significant reduction in dry weights of *C. olitorius* and *E. colonum* by using leaf residues of *L. camara* or *P. guava* especially with their mixtures in comparison to unwedded control. The allelopathic activity of *P. guava* leaves was documented by many workers [25, 26, 27,28.] In addition, [29,30,31,32,33]reported inhibition, occurred by *L. camara* leaves, in growth of some weeds including *E. colonum* The allelopathic activity of *P. guava* leaf residue may be attributed to terpenoids, flavonoids, coumarins and cyanogenic acids[34; 35]. [28] found that the extract of *P. guava* leaves contains some phenolic acids e.g. ferulic, coumaric and chlorogenic acids suggesting that these phenolic acids may be the causative factors of allelopathic activity of *P. guava* leaves. Moreover, [31] attributed the allelopathic activity of *L. camara* leaves to the presence of salicylic acid which was found at high amount.

Numerous plant parts caused reduction in number of root knot nematode population as indicated by previous studies [13,14,36, 37, 38]. Greenhouse studies carried by [28] showed significant inhibition in Purslane growth and number of nematode galls and egg masses by using guava extract. Also, [14]reported that the chopped leaves of lantana significantly reduced nematode criteria of *M. incognita* infecting sunflower. In addition, [39] reported that using dried ground leaves of lantana at rates of 5 and 10 g/kg soil significantly suppressed nematode population in soil and root galling of tomato. [40,41,42]attributed the reduction in *M. incognita* by aerial parts of lantana to the presence of two new constituents namely lantanoside (1) and lantanone (2) and the known compounds, linaroside (3) and camarinic (4). Moreover, [34, 43,44, ,45]attributed the nematicidal activities against the second stage juveniles of *M. incognita* to the presence of pentacyclic triterpenoides which known as camaric acid, lantanilic acid and lenoleic acid. On this basis, it is assumed that before *L. camara* is applied under field conditions to inhibit plant parasitic nematodes, optimal concentrations or rates should be determined that may be toxic to nematodes but neither to plants nor to any associated beneficial microorganisms. The nematicidal activity due to nitrogenous by-products is more effective when the C: N ratio of the amendment is less than 20:1[46] causing more influence of toxic by-products of plant parts on the nematode population. Other factors reported to increase the activity of plant parts decomposition include very thorough powdered or mashed plant tissues before its incorporation into soil and sufficient soil moisture at the time of tissue incorporation [47].Also, suitable soil temperature at the time of incorporation must be provided[48,49]. The secondary decomposed products of organic amendments may be directly toxic to nematodes[50], or they have benefits for increasing plant growth and for increasing natural enemies[51] which may explain their mode of action.

The results, also, reveal that controlling weeds and nematodes was concomitant with increasing in growth and consequently yield of *Vigna unguiculata*. Several workers reported that controlling weeds increased the competition of the crop plants against weeds and so, increased the crop yield[4;52,53,54]. In addition, many workers obtained subsequent increase in growth and yield of crop plants by controlling weeds and subsequently the nematodes[10; 27,28,55,56;]. The increase in plant growth is accompanied by different metabolic processes that may explain increase in carbohydrate content in the yielded seeds [53].

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