NEMATICIDAL ACTIVITY OF ERITREAN WEED PLANTS AGAINST THE ROOT-KNOT NEMATODE *MELOIDOGYNE INCOGNITA*

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ABSTRACT

K.K. Chaudhary, A. Haile, Z. G. Ayresea, G. Semereab, and T. Weldegergish. 2013. Nematicidal activity of Eritrean weed plants against the root-knot nematode *Meloidogyne incognita* (Kofoid and White) Chitwood. Nematropica 43:207-.215

The nematicidal potential of ethanol and aqueous extracts of ten wild plant species distributed on agricultural land of Eritrea against root-knot nematode, *Meloidogyne incognita*, was studied. Three concentrations of each plant extract were tested against egg hatching and mortality of second-stage juveniles (J2) of *M. incognita* at 24, 48 and 72h exposure time. Significant mortality and egg hatching inhibition were observed with aqueous and ethanol extracts of *Datura stramonium*. Similar type of hatch inhibition also occurred with both types of extracts of *Heliotropium indicum*. From tested extracts, J2 mortality ranged from 8 to 100% for hot water extracts and 26 to 100% in for ethanol extracts. The nematotoxicity of the tested plants increased with an increase in concentration and exposure time and vice versa. The hot water and ethanol extracts of *D. stramonium* caused 75-100% mortality and 57-100% inhibition in egg hatch, respectively. This *M. incognita* juvenile mortality was 74-100% in the case of *H. indicum*. Among the tested extracts, hot water and ethanol extracts of *Lantana camara* and *Xanthium strumarium*, respectively, were least effective.

Key words: Eritrea, hot water extracts, ethanol extracts, *Meloidogyne incognita,* egg hatch, mortality.

RESUMEN

K.K. Chaudhary, A. Haile, Z. G. Ayresea, G. Semereab, and T. Weldegergish. 2013. Actividad nematicida de malezas de Eritrea sobre el nematodo agallador *Meloidogyne incognita* (Kofoid y blanco) Chitwood. Nematropica 43:207-215.

Se estudió el potencial nematicida extractos de etanol y acuosos de diez especies de plantas silvestres distribuidas en tierras agrícolas de Eritrea sobre el nematodo agallador, *Meloidogyne incognita*. Se evaluó la eclosión de huevos y la mortalidad de juveniles de segundo estadio (J2) de *M. incognita* frente a tres concentraciones de cada extracto de planta durante 24, 48 y 72 h de exposición. La mortalidad y la inhibición de la eclosión de huevos fue significativa con extractos acuosos y de etanol de *Datura stramonium*. Una inhibición similar en la eclosión también se observó con ambos tipos de extractos de *Heliotropium indicum*. De los extractos evaluados, la mortalidad de J2 osciló entre el 8-100% para los extractos de agua caliente y el 26-100% para los extractos de etanol. La acción nematicida de las plantas testeadas aumentó con el incremento de la concentración y el tiempo de exposición y viceversa. Los extractos de agua caliente y de etanol de *D. stramonium* causaron una mortalidad del 75-100% y el 57-100% de inhibición en la eclosión de huevos, respectivamente. La mortalidad de J2 de *M. incognita* fue de 74-100% en el caso de *H. indicum*. De los extractos evaluados, los de agua caliente y de etanol de *Lantana camara* y *Xanthium strumarium*, fueron menos eficientes, respectivamente.

Palabras clave: Eritrea, extractos de agua caliente, extractos de etanol, *Meloidogyne incognita*, eclosión de huevos, mortalidad.

INTRODUCTION

Meloidogyne spp., the root knot nematodes are the most damaging nematodes in agriculture (Javed *et al*., 2006). *Meloidogyne incognita* (Kofoid and White) Chitwood is considering the most widespread species of this genus (Trudgill and Blok, 2001; Chen *et al*., 2004). Nematode problems may be especially acute in underdeveloped countries such as Eritrea, where 80% of the cultivated land of the Hamelmalo Subzone (Eritrea) is infested by this nematode (Chaudhary *et al*., 2011). A variety of management strategies including cultural practices, resistant cultivars, chemicals, solarization, fumigation, trap crops, organic soil amendments and biological control have been considered effective for reducing nematode damage on various crops. Awareness of the hazards of chemical nematicides to humans and the environment (Taba *et al*., 2008) has spurred increased attention to alternative management approaches and environmentally friendly ways for management of phytonematodes. Identification of natural nematicides from plant materials is a promising approach (Tsay *et al*., 2004; Raina *et al*., 2007; Tariq *et al*., 2007). Chitwood (2002) compiled a list of active compounds such as diterpenes, phenols, polyacetylenes, alkaloids, sesquiterpenes and thyenil derivatives for use in management of plant parasitic nematodes. Consequently, many plants and their tissues have been screened for nematicidal activities. However, little attention has been given to weeds separately or mixed with crop plants on agriculture land. For instance, several plant species regarded as weeds contain useful compounds that can be used as medicines, or as compounds of food, cosmetics and spices. (Stepp and Moerman, 2001; Stepp, 2004). Many of these species also have strong allelopathic activity, in that they discourage the neighbouring growth and activity of other organisms (Fujii, 2000). In the present investigation, wild Eritrean plants were screened for specific nematotoxic activity by testing the effectiveness of plant extracts on *M. incognita* egg hatch and mortality.

MATERIALS AND METHODS

Extraction of Plant Components

Ten weed plants and one bionematicide, i.e., Neem (*Azadirachta indica*) were collected from different localities of the Hamelmalo, Eritrea, under the supervision of a botanist in September, 2011. The selections of plants were based on their known microbial and insecticidal properties (Table 1). Healthy leaves of the selected plants were washed thoroughly in running tap water, dried at temperature of 50°C or less for 5 days, then ground to a fine powder with a home mixer and grinder.

Preparation of Ethanol Extract

Thirty grams of dry leaf powder was suspended in 200 ml of 70% ethanol in a parafilm covered conical flask for 72 h in the dark on a mechanical shaker at 200 rpm. The suspension was filtered under vacuum through Whatman No. 1 filter papers. The procedure was repeated with the residue on the filter paper and similarly filtered. The two filtrates were combined and dried in a sterile 10-ml test tube in a rotary evaporator at 45°C, and the resulting extract was stored in a refrigerator until use.

Preparation of Hot Water Extracts

Hot water extract of selected weeds and bionematicide were prepared by suspending 30 grams of dry leaf powder from each of the 11 species in 50 ml of boiling distilled water for one hour in a 500-ml flask. During this period flasks were kept on water bath set at 60°C for maintaining constant temperature for one hour. Extracts were filtered through No. 1 Whatman filter papers and used as a stock solution. These stock solutions were evaporated to dryness at 60°C and stored in a refrigerator until use.

Preparation of Nematode Inoculum

Meloidogyne incognita was collected from naturally infested brinjal (*Solanum melongena* L.) fields in Hamelmalo subzone, Eritrea. Pure cultures were maintained on tomato (*Lycopersicon esculentum* Mill.) roots in pots in the greenhouse at the college campus. Second-stage juveniles (J2) were obtained from hatched eggs by incubating handpicked egg masses in sterile distilled water at 28 ± 1°C. Freshly hatched juveniles were used in all mortality bioassay experiments. A stock suspension of nematodes in sterile water was prepared with a final concentration of 100 ± 5 J2 per ml.

For the hatching bioassay, eggs of *M. incognita* were extracted from egg masses picked from infected tomato (*Lycopersicon esculentum*) roots using 0.4 percent sodium hypochlorite solution (chlorox). The suspension was consecutively passed through 75 mm and 26 mm sieves, and eggs collected from the 26 mm sieve were agitated in water to remove remaining NaOCl and counted under an inverted microscope (Hussey and Baker, 1973). A stock solution of 100 ± 5 eggs/ml sterile water was prepared and used for study.

Experimental Design

Effect of plant extracts on eggs hatching. Two-ml aliquots of sterile water suspension of *M. incognita* eggs (100 ± 5/ml) were placed in watch glasses maintained in a moist chamber at 28 ± 1°C. Each suspension received a plant extract to give final concentrations of 25, 50 or

Table 1. Eritrean plants selected for evaluation of hot water and ethanolic extract against *Meloidogyne incognita*, and there active ingredient against various microorganisms.

Name of Weed	Family	Vernacular Name in Tigrina	Antimicrobial Chemicals of selected plant	References
Azadirachta indica (BNC)	Meliaceae	Neem	Azadiradion; Azadirachtin and Nimbin	Kraus *et al*., 1994
Acanthospermum hispidum	Asteraceae	Melhas Sebeyti	Loliolide; sesquiterpene lactones and acanthospermal B	Kraus *et al*., 1994
Aerva persica	Amaranthaceae	----	5,4-hydroxy-3,6,7-trimethoxyflavone; 5-hydroxy-3,6,7,4-tetramethoxyflavone; apigenin7-O-β-D-glucoside; lupeol and 5-hydroxy2',3,5',6,7-pentamethoxyflavone	Ahmed *et al*., 2008
Bidens pilosa	Asteraceae	Tsegogo	Flavonoids; terpenes and phenylpropanoids	Adedapo *et al*., 2011
Cassia tora	Fabaceae	Abake Harmaz	Parthenin; pyrazoline; anhydroparthenin and hemiacetal	Chandan *et al*., 2011
Datura stramonium	Solanaceae	Mezerbae	Scopolamine; hyoscyamine; atropine meteloidine; apoatropine; terpenoids and Flavonoids	Pavela, 2004
Flaveria trinervia	Asteraceae	Diha-Nekel	1-methyl-3-(methylthio)-benzene, 3-methylbencil mercaptan, tannins, flavonoids, leucoanthocyanidins, steroids and triterpenoids	Umadevi *et al*., 2005
Helianthus indica	Asteraceae	"SUF" Bereka	Lactones, diterpenoic acid and flavonoid (nevadensin)	Mullin *et al*., 1991
Heliotropium indicum	Boraginaceae	Amamgemel	5, 8, 11, 14, 17-Eicosapentenoic acid; methyl ester, 2, 4-ditertbutyl phenol; p-Mentha-6, 8-dien-2-one-semicarbazone; 1, 2-Benzenedicarboxylic acid and diisooctyl ester.	Oluwatoyin *et al*., 2011
Lantana camara	Verbanaceae	Bun tilian	Lantanilic acid; camaric acid and oleanolic acid	Qamar *et al*., 2005
Xanthium strumarium	Asteraceae	Eshok Mergem	deacetyl xanthumin; xanthanolide; xanthanol; isoxanthanol, sesquiterpene lactones (lactone, 2-hydroxytomentosin- 1β,5β-epoxide; 1,3,5-tri-O-caffeoylquinic acid and 3,5-di-O-caffeoylquinic acid); carboxyatractyloside; hydroquinone and Xanthatin	Kim *et al*., 2002

Table 2. Effect of hot water extracts of Eritrean weed plants on the *Meloidogyne incognita* eggs hatching inhibition (corrected cumulative percentage)

	Hot Water Plant Extracts (mg/ml)								
	24 hrs Exposures			48 hrs Exposures			72 hrs Exposure		
Name of weed	25[yz]	50[yz]	100[yz]	25[yz]	50[yz]	100[yz]	25[yz]	50[yz]	100[yz]
Azadirachta indica (BNC)	42.72 ± 3.12b	50.51 ± 2.83b	74.41 ± 0.75b	61.85 ± 1.45b	71.47 ± 3.18b	82.58 ± 1.86b	78.13 ± 1.11b	90.96 ± 1.83b	93.42 ± 2.02ab
Acanthospermum hispidum	19.49 ± 2.44d	27.73 ± 2.60d	54.61 ± 2.46d	44.61 ± 2.40cd	63.19 ± 3.45bc	72.10 ± 3.57cd	66.02 ± 2.34c	79.03 ± 1.19c	79.42 ± 4.46c
Aerva persica	18.23 ± 1.19def	24.68 ± 1.88de	45.05 ± 3.98e	37.59 ± 3.62de	50.57 ± 3.95de	66.13 ± 0.49de	69.69 ± 0.46bc	76.80 ± 3.50c	77.53 ± 3.78cd
Bidens pilosa	31.84 ± 2.83c	40.54 ± 3.52c	65.53 ± 0.38c	54.23 ± 3.83bc	70.39 ± 0.84b	79.87 ± 2.91bc	72.66 ± 1.12bc	90.16 ± 1.19b	90.32 ± 1.02b
Cassia tora	13.56 ± 0.98def	18.54 ± 1.24e	36.35 ± 1.92f	37.38 ± 1.35de	51.73 ± 1.89de	54.25 ± 1.94f	51.95 ± 2.35d	60.77 ± 4.30d	63.96 ± 2.93e
Datura stramonium	75.39 ± 1.84a	89.74 ± 1.01a	98.27 ± 0.27a	92.99 ± 3.05a	100.0 ± 0.00a	100.0 ± 0.00a	100.0 ± 0.00a	100.0 ± 0.00a	100.0 ± 0.00a
Flaveria trinervia	19.99 ± 4.96de	27.30 ± 3.08d	44.45 ± 1.83e	36.16 ± 2.62de	49.86 ± 0.81de	61.41 ± 0.95ef	51.08 ± 1.33d	62.58 ± 1.92d	70.76 ± 0.87de
Helianthus indica	38.40 ± 2.33bc	45.16 ± 2.11bc	63.84 ± 1.31c	50.34 ±2.25bc	57.41 ± 3.38cd	78.04 ±1.99bc	65.12 ± 2.06c	79.47 ± 1.67c	79.93 ± 1.49c
Heliotropium indicum	77.92 ± 4.01a	93.97 ± 1.52a	97.98 ± 0.69a	88.25 ± 4.24a	95.69 ± 1.84a	100.0 ± 0.00a	95.81 ± 1.36a	100.0 ± 0.00a	100.0 ± 0.00a
Lantana camara	10.12 ± 0.44ef	17.61 ± 0.45e	40.03 ± 1.34ef	29.48 ± 3.09e	51.04 ± 4.56de	58.28 ± 2.18ef	47.69 ± 3.18e	62.16 ± 0.85d	68.54 ± 2.05e
Xanthium strumarium	8.87 ± 0.44f	18.75 ± 2.32e	37.84 ± 1.36f	33.14 ± 2.86e	43.80 ± 3.22e	53.31 ± 4.34f	49.31 ± 1.60de	59.59 ± 1.34d	65.23 ± 2.06e
$P = 0.05$	9.44	8.04	6.46	11.09	11.01	8.16	6.43	7.34	8.34

[y]Percentage are average of three replicates; mean ± SE value followed by the different letter in same vertical column are significantly different according to Duncan's multiple range test ($P = 0.05$).
[z]Hatching inhibition = (percentage hatching inhibition in extract – percentage egg hatching inhibition in sterile distilled water control) × 100%/(100 − percentage egg hatching inhibition in sterile distilled water control) (Abbott, 1925).

Table 3. Effect of Ethanolic extracts of Eritrean weed plants on the *Meloidogyne incognita* eggs hatching inhibition (corrected cumulative percentage).

	Hot Water Plant Extracts (mg/ml)								
	24 hrs Exposures			48 hrs Exposures			72 hrs Exposure		
Name of weed	25[yz]	50[yz]	100[yz]	25[yz]	50[yz]	100[yz]	25[yz]	50[yz]	100[yz]
Azadirachta indica (BNC)	54.59 ± 1.90b	70.05 ± 2.36b	83.27 ± 1.20cd	63.00 ± 1.18c	75.19 ± 0.67bc	93.01 ± 2.41a	92.60 ± 1.65b	95.49 ± 1.83b	99.07 ± 0.39a
Acanthospermum hispidum	35.46 ± 2.29d	47.60 ± 1.05d	62.66 ± 1.66e	43.09 ± 1.54d	58.03 ± 2.39c	74.08 ± 2.48c	67.39 ± 3.72d	79.80 ± 1.37c	81.57 ± 2.87c
Aerva persica	35.73 ± 2.65d	48.04 ± 1.65d	64.74 ± 4.37e	40.93 ± 0.67d	54.02 ± 1.84d	72.52 ± 3.89c	64.78 ± 1.10d	79.67 ± 0.45c	81.07 ± 3.38c
Bidens pilosa	49.47 ± 1.37bc	69.26 ± 1.71bc	87.64 ± 0.87bc	69.40 ± 1.59bc	83.12 ± 1.26b	96.23 ± 0.49a	98.96 ± 2.5a	100.00 ± 0.00a	100.00 ± 0.00a
Cassia tora	46.16 ± 1.56c	62.69 ± 1.18c	79.16 ± 1.30d	68.08 ± 0.68bc	75.36 ± 2.36bc	84.20 ± 0.84b	72.27 ± 3.55c	81.70 ± 1.49c	87.27 ± 1.16b
Datura stramonium	80.04 ± 0.98	94.70 ± 1.56a	97.56 ± 1.06a	100.00 ± 00.65a	100.00 ± 0.00a	100.00 ± 0.00a	100.00 ± 0.00a	100.00 ± 0.00	100.00 ± 0.00a
Flaveria trinervia	29.63 ± 1.91de	43.73 ± 2.11d	61.21 ± 3.02	40.83 ± 1.59d	53.01 ± 1.37d	66.83 ± 2.49cd	41.55 ± 1.09e	57.74 ± 1.05d	76.97 ± 3.80e
Helianthus indica	49.14 ± 1.35bc	63.91 ± 0.72bc	85.32 ± 1.80cd	71.80 ± 1.84b	89.17 ± 1.32b	97.80 ± 1.30a	96.96 ± 2.46a	100.00 ± 0.00a	100.00 ± 0.00a
Heliotropium indicum	74.74 ± 1.89a	93.51 ± 1.39a	95.75 ± 1.18ab	78.60 ± 0.77b	100.00 ± 0.00a	100.00 ± 0.00a	98.98 ± 0.46a	100.00 ± 0.00a	100.00 ± 0.00a
Lantana camara	26.40 ± 1.17e	42.73 ± 1.18d	61.84 ± 3.42e	30.20 ± 1.86e	52.23 ± 1.01d	67.12 ± 0.84cd	41.46 ± 1.02e	53.93 ± 1.33	77.87 ± 1.83de
Xanthium strumarium	26.81 ± 2.29e	42.07 ± 0.99d	59.27 ± 2.66e	30.24 ± 3.73e	50.59 ± 1.07e	63.58 ± 2.03d	39.92 ± 3.52e	55.51 ± 1.02de	77.59 ± 2.24de
$P = 0.05$	7.22	6.62	8.35	8.1	6.66	7.68	3.58	3.83	3.68

[y]Percentage are average of three replicates; mean ± SE value followed by the different letter in same vertical column are significantly different according to Duncan's multiple range test ($P = 0.05$).
[z]Hatching inhibition = (percentage hatching inhibition in extract – percentage egg hatching inhibition in sterile distilled water control) × 100%/(100 − percentage egg hatching inhibition in sterile distilled water control) (Abbott, 1925).

100 mg/ml. Each treatment was replicated three times and sterile distilled water served as a control. The watch glasses were arranged on incubator shelves in a Completely Randomized Design (CRD). Hatching was determined after 24, 48 and 72 h from respective treatments. After the first observation, the extract of watch glass was carefully replaced with sterile distilled water. Following the water replacement hatched J2 were counted up to 15 days each after 2 day intervals in treatments as well in controls (distilled water). Data on hatching were converted to percentage cumulative hatching inhibition, corrected by Abbott's formula (Abbott, 1925). Data were statistically analyzed with ANOVA followed by Duncan's multiple range test (DMRT). $P<0.05$ was considered as significant.

Effect of plant extracts on the mortality of J2. Mortality of J2 was determined following the same procedure as used in case of data collection on effect of plant extracts on eggs hatching as described above, with slight modification having initial volume of 1 ml of a sterile water suspension of *M. incognita* J2, instead of 2 ml. The test tubes were arranged on incubator shelves in completely randomized design (CRD). Immobile nematodes were counted after 24 and 72 h incubation at 28°C ± 1. Second stage juveniles that did not move when touched with a drawing-brush hair were transferred to distilled water. They were considered dead if they still failed to react to probing with a bristle 2 hours later. Mortality data were converted to percentage cumulative mortality and corrected with Abbott's formula (Abbott, 1925) with reference to distilled water control. Data were statistically analysed with ANOVA, followed by DMRT. $P<0.05$ was considered as significant.

RESULTS

Effect of Plant Extracts on Hatching

The number of hatching nematodes increased with elapsed days in the water control. Time of exposure and concentration of plant extracts were positively correlated. All three extract concentrations inhibited J2 hatching at each exposure period. Generally, inhibition of egg hatching was time and dose-dependent. Exposure to 25 mg/ml of all plant extracts for 24 h was least effective in inhibiting egg hatch while exposure to 100 mg/ml of all plant extract for 72 h produced significantly greater inhibition compared to exposures of to 25 and 50 mg/ml concentrations (Tables 1, 2).

Extracts of *D. stramonium* and *H. indicum* had the highest cumulative inhibition of egg hatching (100%) at 48 and 72-h exposures to 50 and 100 mg/ml concentrations. Neem is a certified bionematicide but it produced significantly lower inhibition at all the exposure times and concentrations in present study. Likewise, other plant extracts, though significant in cumulative inhibition of egg hatching, were less effective compared to those of *D. stramonium* and *H. indicum* (Table 2). The least effective extracts were those of *Xanthium strumarium* and *Lantana camara*, which had no effect on egg hatch.

Inhibition by alcohol extracts was higher than that caused by hot water extracts in all plant species at all concentrations and durations of exposure. The maximum cumulative inhibition (100%) was observed at three concentrations and two exposure times (25, 50 and 100 mg/ml for 48 and 72 h) for *D. stramonium* (Table 3). For *H. indicum* 100% inhibition occurred at concentrations of 50 and 100 mg/ml at 48 and 72 h exposures, and was not different from that of *D. stramonium*. Complete (100%) inhibition occurred with extracts of *B. pilosa* and *Helianthus indica* at concentrations of 50 and 100 mg/ml for 72-h exposures. The inhibition caused by Neem extract at 100 mg/ml for 72 did not differ significantly from that of *D. stramonium* and H. indicum at same concentration and exposure time. Cumulative inhibition of M. incognita egg hatching in all the plant extracts was between 26 and 80% at a concentration of 25 mg/ml for 24 h. Amongst all the tested alcoholic extracts, those from *X. strumarium* and *L. camara* were found to be the least effective in reducing egg hatch. The range of cumulative inhibition in egg hatching was 26 to 77% in these two plant extracts.

Effect of Plant Extracts on M. incognita mortality

Juvenile mortality increased with increases in exposure time and concentration (Figs. 1–4), but mortality differed significantly among different concentrations and exposure times. Hot water extracts of *D. stramonium*, *H. indicum* and Neem were equally effective (56-57% mortality) against J2 after 24 hours of incubation at 25 mg/ml concentration. The other extracts had lower effects on mortality, with *L. camara* (29.2 ± 1.8%) the least effective. However, higher extract concentrations increased J2 mortality in almost all the extracts assayed. For example, mortality caused by extracts of *D. stramonium* reached 71.5 ± 1.4% at 50 mg/ml and 88.7 ± 2.6% at 100 mg/ml after 24 h of exposure. Extracts from *H. indicum* and Neem gave results similar to those for *D. stramonium*. The other treatments also gave more than 50% mortality at 50 and 100 mg/ml (Fig. 1). Percent mortality of J2 at after 72 h exposure was generally higher than mortality after 24 h (Fig. 2). Extracts of *D. stramonium*, *H. indicum* and Neem gave mortalities close to or equal to 100%. The least effective extracts were *X. strumarium* and *L. camara* at 25 mg/ml concentration after 72 h of exposure (Fig. 2).

Mortality was significantly higher for ethanol extracts than for hot water extracts at all three concentrations and two exposures times (Figs. 3, 4). In general, at 25 mg/ml some ethanol extracts induced less than 50% mortality while this mortality increased from 50 to 95% at concentrations of 50 mg/ml and up to 99% at 100 mg/ml for 24 h exposures. The best

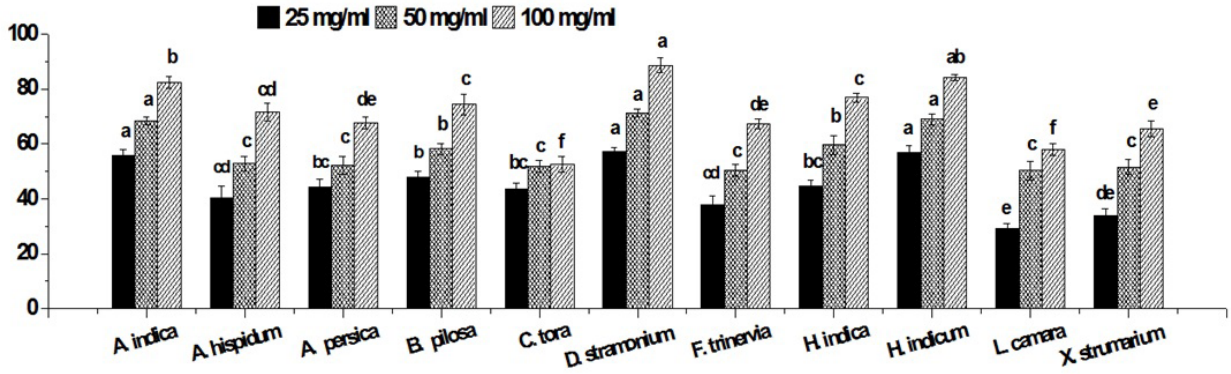

Fig.1. Corrected percentage cumulative mortality of *Meloidogyne incognita* J2 exposed to different concentrations of hot water extract of Eritrean plants at 24 hrs of exposures. The bars corresponded to the standard error which represents mean ± SE. Different letters indicate statistically significant difference (Duncan's multiple range test, $P = 0.05$) within concentrations. Each treatment has three replications.

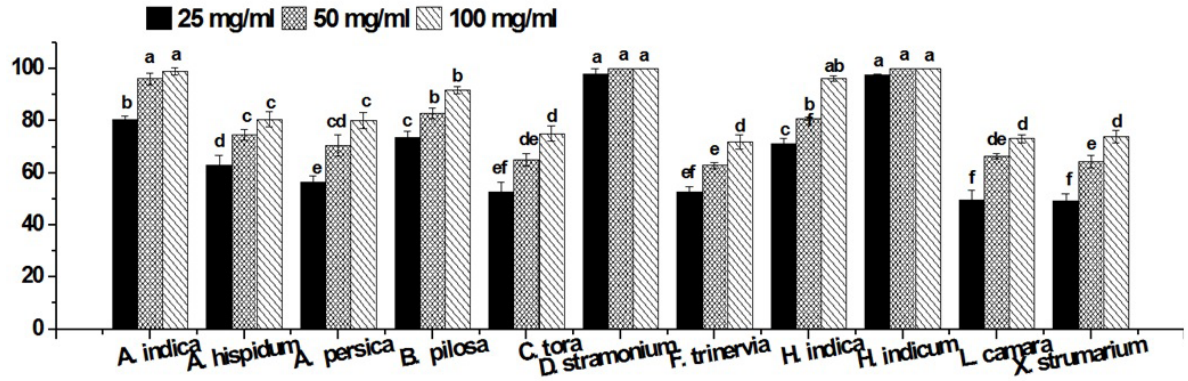

Fig.2. Corrected percentage cumulative mortality of *Meloidogyne incognita* J2 exposed to different concentrations of hot water extract of Eritrean plants at 72 hrs of exposures. The bars corresponded to the standard error which represents mean ± SE. Different letters indicate statistically significant difference (Duncan's multiple range test, $P = 0.05$) within concentrations. Each treatment has three replications.

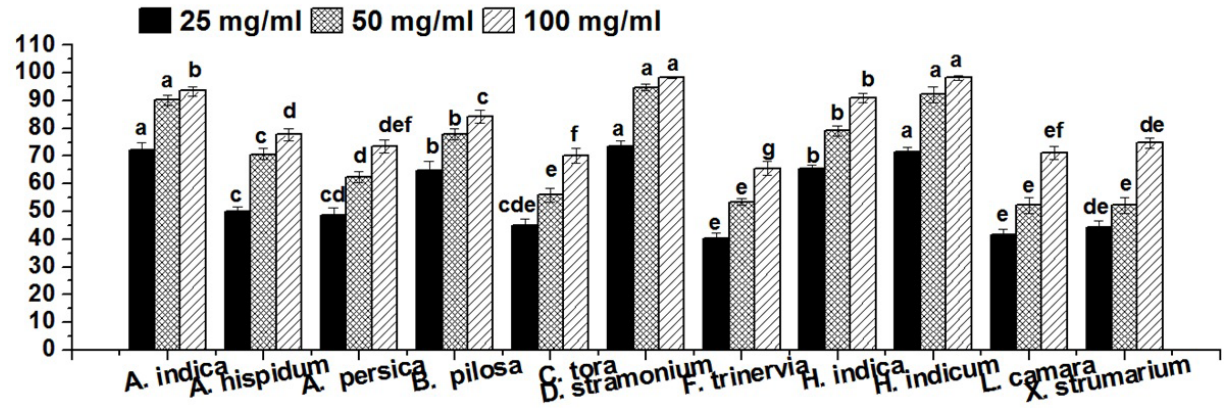

Fig.3. Corrected percentage cumulative mortality of *Meloidogyne incognita* J2 exposed to different concentrations of ethanolic extract of Eritrean plants at 24 hrs of exposures. The bars corresponded to the standard error which represents mean ± SE. Different letters indicate statistically significant difference (Duncan's multiple range test, $P = 0.05$) within concentrations. Each treatment has three replications.

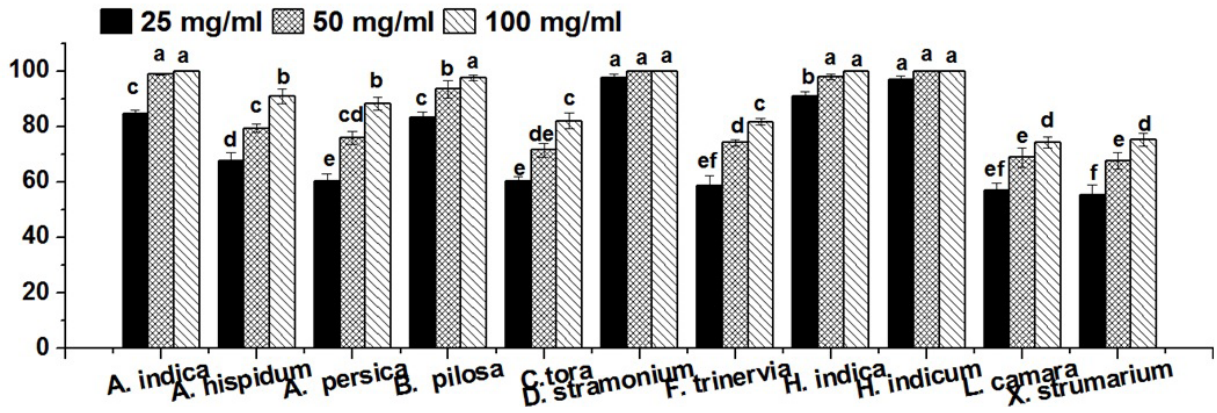

Fig. 4. Corrected percentage cumulative mortality of *Meloidogyne incognita* J2 exposed to different concentrations of ethanolic extract of Eritrean plants at 72 hrs of exposures. The bars corresponded to the standard error which represents mean ± SE. Different letters indicate statistically significant difference (Duncan's multiple range test, P = 0.05) within concentrations. Each treatment has three replications.

results were obtained with extracts of *D. stramonium* (98.34 ± 0.27), *H. indicum* (98.12 ± 0.74) and Neem (93.43 ± 1.65) at 100 mg/ml and 24 h, with mortality caused by *D. stramonium* significantly greater than with *H. indicum* and Neem. At the same concentration and time of exposure, the least effective treatment was *L. camara* (Fig. 3).

At 72 h exposure duration, nematode mortality ranged from 55 to 100% for the all tested plant extract. Amongst all the tested extracts, *D. stramonium* and *H. indicum* exhibited the highest mortality at 72 h of exposure at both the concentrations (50 mg/ml and 100 mg/ml). One hundred percent mortality was recorded at 72 h exposure with 100 mg/ml concentration of *Helianthus indica* and Neem. At 100 mg/ml concentration and 72 h of exposure time, *L. camara* extract caused 74.8% mortality. The range of nematode mortality in other extracts was between 55 and 98% at a concentration of 25 mg/ml at same exposure time, which increased from 67 to 100% at a concentration of 50 mg/ml. *X. strumarium* and *L. camara* had the least effect on nematode mortality.

DISCUSSION

Present findings highlight the potential of indigenous wild plant species of Hamelmalo agricultural areas and their active ingredients on immobilization and toxic effects on *M. incognita*. The nematotoxicity of the tested plants increased with an increase in concentration and exposure time. The inhibitory effects of leaf extracts of various weed plants has been reported by a number of researchers (Tiyagi and Ajaz, 2003; Raina *et al*., 2007). Adegbite and Adesiyan (2005) suggested that the inhibitory effect observed in egg hatching might be due to ovicidal or larvicidal chemicals present in the extracts. In this aspect, the findings of the present study are in conformity with the finding of the previous researchers (Chattopadhyay, 1991; Radwan *et al*., 2007). In the present study, *D. stramonium* and *H. indicum* were found to be superior amongst the all tested plant extracts. The nematicidal value of the *D. stramonium* is well reported and the findings of the present study are in conformity with the findings of Nandal and Bhati (1986); Sharma and Trivedi (2002) and Ahmad *et al.* (2004) who reported superiority of *Datura* over the other tested plants in increasing nematode mortality and inhibition in egg hatching. The nematicidal properties of the *H. indicum* was not well known but the insecticidal properties of this plant was reported by Kamruzzaman *et al*. (2005) and Dolui and Debnath (2010).

Amongst all tested plant species *L. camara* and *X. strumarium* were reported as least effective extract, but some previous studies (Ali *et al*., 2001; Shaukat and Siddiqui, 2001; Begum *et al*., 2001) reported high nematicidal potential of *L. camara* against *M. incognita*, which requires further confirmation. Although the toxicity of *X. strumarium* was reported by Prakash and Rao (1997) but in present study it was found to be the least effective treatment.

Neem (*Azadirachta Indica*) was certified as a bio nematicide by various researchers and the product of this tree Azadirachtin as a nematicide is in vogue in the various parts of the words (Akhtar and Malik, 2000; Chitwood, 2002; Adegbite, 2011). Nevertheless, most of the tested plants extracts exhibited almost equal or superior nematotoxic effects then the well reported bionematicide Neem, which shows the considerable potential of tested plants as source of nematicidal compounds, which needs further elucidation. Our study is a first report on the evaluation of *A. persica*; *C. tora*; *F. trinervia* and *H. indicum* extracts for their nematicidal activity against the root knot nematode *M. incognita*. Overall, the current study highlights the potential of certain indigenous weed plants for their

use as nematicides which can serve as cheaper and environmentally benevolent alternatives to chemical nematicides for resource poor farmers.

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