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Control of root-knot nematodes using wild plants colonized Sinai, Egypt

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Abstract

The antinematodal activity of some wild plant extracts located in North and South Sinai, Egypt was examined against root-knot nematode (RKN). The selected seven plants viz., *Artemisia judaica*, *A. monosperma*, *Bassia muricata*, *Cornulaca monacantha*, *Salsola kali* and *Zygophyllum album* with different dilutions (25%, 50%, 75% &100%). The potent nematicidal efficacy observed in the extract of *A. judaica* followed by *A. monosperma*. *In vivo* trial post inoculation treatments were effective than pre one, the maximum reduction in nematode population was recorded by *A. judaica* (87.0%) followed by *A. monosperma* (83.0%) then *Z. album*1 (79.0%), while the lowest suppressor was *C. monacantha* (60.4%) at stock concentration. The addition of extracts as soil drench was better than spraying them. The stock solution showed the highest reduction in nematode reproduction but the differences between 100% and 75% concentrations were non-significant. Our finding offers a non-poisonous tool that can insert in combating nematode programs especially for small-holder growers colonized Sinai Peninsula.

Key words: Plant extracts, root-knot nematodes, wild plants, *Artemisia monosperma*, *Artemisia judaica*, *Bassia muricata*, *Cornulaca monacantha*, *Salsola kali*, *Zygophyllum album*.

Plant-parasitic nematodes (PPN) represent the most highly distributed pathogens decreasing global agricultural production i.e., they occur in virtually all soil types. Among the PFN, the most economically damaging soil-borne genus is *Meloidogyne* spp. (Tylenchida: Meloidogynidae).

Root-knot nematode (RKN) is a devastating pathogen that causes tremendous losses on both field and horticultural crops worldwide, RKN being responsible for at least 90% of all damage caused by nematodes. Losses due to RKN reach up to 80% in highly infested fields, also increases the severity of soil-borne diseases (Sikora & Fernandez, 2005; Aissani *et al*., 2015; Hussain *et al*., 2016; Khalil & Darwesh, 2018).

In Egypt, numerous eco-friendly-alternative approaches including plant natural extracts, natural products of microbial origin and organic amendments were applied as nematicide alternatives by many researchers (Nasser, 2018): PGPR, salicylic acid derivatives and ascorbic acid used as biocontrol agents for suppression *M. incognita* (El-Nuby, 2014); Seaweeds alone or combined with nematicides used for the management RKN (Afia & El-Nuby, 2016).

Reduction in egg hatching of RKN by leaf and root extracts of *Azadirachta indica*, *Moringa oleifera*, *Lantana camara* and *Glycyrrhiza glabra* observed by Haroon *et al*., (2018). Dried plant materials at minimum rate

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show promising antinematodal activity for combating *M. incognita* (Ibrahim *et al*., 2018); Radwan *et al*., (2019) found that macrolides-structurally related natural products of microbial origin can regulate RKN population densities and be used against nematicides.

The objectives of the present research were to: evaluate the antinematodal activity of aqueous extracts of some wild plants form Sinai Peninsula, Egypt against root-knot nematode *Meloidogyne incognita*; *in vitro* and *in vivo*.

It is also to determine the best mode (drench or spray) and time of application (pre of post-inoculation) of plant extracts that achieve higher nematode suppressionalso determine the best and economic concentration that achieves maximum quelling of nematode combined with low cost.

Materials and Methods

Collection of plant material: Samples of wild plants (including entire herb, with roots, shoots and rhizosphere soil) were collected from North and South Sinai during 2016/2017. The samples were kept in paper bags to prevent from direct sunlight and heating. The plant specimens were identified and authenticated (by the Head of Plant Taxonomy Unit, Plant Ecology and Ranges Department, Desert Research Center (DRC). A voucher herbarium specimen was deposited in the herbarium of DRC (CAIH). Plants were identified to species levels according to Boulos & El-Hadidi (1994) and Boulos (2002) as presented in Table 1. Samples were processed in the laboratory as the entire herb was air-dried in shade at room temperature (30±2ºC) for two weeks and ground to a fine powder in an electronic grinder.

Table 1. Wild plants collected from different locations in Sinai Peninsula.

Scientific name	Family	Arabic name	Part used
***Artemisia judaica* L. (S)**	Asteraceae (Compositae)	Sheha	Shoots (aerial parts)
***Artemisia monosperma Delile* (N)**	Asteraceae (Compositae)	Ader	Shoots (aerial parts)
***Zygophyllum album* L. f. (N&S)**	Zygophyllaceae	Rotreet	Shoots (aerial parts)
***Salsola kali* L.(N)**	Amaranthaceae (Chenopodiacea)	Eshnana	Shoots (aerial parts)
***Cornulaca monacantha Delile*(N)**	Amaranthaceae (Chenopodiacea)	Shook El-deeb	Shoots (aerial parts)
***Bassia muricata* (L.) *Asch. inchweinf*. (N)**	Amaranthaceae (Chenopodiacea)	Ghobeera	Shoots (aerial parts)

N= North Sinai, S= South Sinai

Preparation of water extract: Plant extracts were prepared according to the procedure described by Al-Manhel & Niamah (2015). The powdered material (5 g) was soaked in 100 ml of distilled water in 250 ml Erlenmeyer flask. The flasks placed on a horizontal shaker and shaken (200 rpm/min for 24hrs) at room temperature (29±1ºC). The extract (suspension) was filtered through cheese cloth, the process was repeated till the aqueous extract of the plant become clear and kept at 4°C till use in the same day in bioassay, the resulted filtrate (5% w/v or 50 mg/ml.) consider stock solution concentration (SS).

Primary screening *in vitro*: Nematotoxicity of extracts concerning 21 wild plants against *M. incognita* was investigated. The nematodes

(obtained from the maintained culture of tomato cv. Castel Rock) were harvested according to the method as described by Barker (1985) in terms of second-stage juveniles (J_2) using 4 dilutions (25%, 50%, 75%, and 100%). 50 J_2 were transferred to Petri plates (5cm diam.), 2 ml of water extract of each plant was added then all plates were incubated at 27ºC arranged in a completely randomized design. Each treatment was replicated four times, distilled water served as a negative control. After 24hrs the dead and life larva were counted under the stereoscopic-zoom microscope. Live and dead nematodes were recorded as well as mortality percentages were calculated [(No. dead J_2/ No. of sum live + dead J_2) x100]. The juveniles were considered dead if they remain static or immobile (paralyzed) after probing them with a fine needle (Cayrol *et al*., 1989; Abbasi *et al*., 2008). The recovery test was done by transferred the juveniles to distilled water and check them after 24 hrs.

***In vivo* study (Pre vs post inoculation application):** Tomato seedlings (cv. Super strain-B) 4 weeks was transplanted in 15 cm diam. an earthen pot filled with steam-sterilized soil (3:1 sand- clay V: V), after ten days plants were divided to two sets: the first was inoculated with 1500 freshly hatched J_2 by pouring the nematode suspension above the roots after temporary removing the soil layer; the second set of pots received 100 ml from each plant extract. After one week the first group was drenched with different plant extracts and the other one was inoculated by *M. incognita* similar to the first group. Each treatment consisted of 4 replicates arranged in randomized blocks on clean benches of Plant Protection Department greenhouse, DRC. Plants were horticulturally treated the same and watered when needed. One month and a half later, the plants were uprooted and the growth parameters viz., root and shoot weights and lengths, and dry shoot weights were recorded. After staining the roots with acid fuchsin lactophenol stain nematode criteria including gall numbers, egg-masses numbers, developmental stages, fecundity (number of egg/egg-mass), the total number of eggs, equal mean number of eggs/egg-mass x total egg-masses/roots (after picked 10 egg masses similar in size and count them separately under stereo zoom and dissecting microscope).

The final population (FP) was calculated by gathering the galls + egg-masses + developmental stages + total number of eggs.

***In vivo* study (Effect of different concentrations):** Four dilutions (1/4SS, 1/2SS, 3/4SS, and SS that expressed as 25, 50, 57 & 100%, respectively) were applied as soil drench after one week from inoculation. Four earthen pots were used in such treatment and contain the same soil composition as mentioned before. Also, the plant inoculated with 1500 freshly hatched J_2 of *M. incognita* from the source which established for all experiments. The same numbers of replicates were served as the negative control (nematode only in distilled water instead of plant extracts) and positive or comparable control (Vydate or Oxamyl 24% liquid) was used at the rate of 0.6% (i.e. 6 ml/liter of water).

Pots were kept on greenhouse benches in DRC, cared and irrigated as the same. After six weeks each pot was soaked in the water pan to easily remove the adhering soil particles with minimum root damage.

The impact on growth was assayed by recoding shoot and root heights and weights. Nematode parameters as gall numbers, egg-masses, developmental stages and fecundity were registered in stained roots. Total eggs/ root was counted, also the final population/ root was calculated by summation of the egg-masses + developmental stages + mature females + total eggs.

Statistical analysis: The differences between means were tested using Duncan's multiple ranged test at the 5% significance level (Duncan's, 1955).

Table 2. Effect of aqueous extracts of 21 wild plant species on mortality percentages of second stage juveniles of *M. incognita* at different concentrations after 24 hrs exposure time.

No.	Plant species	% Juveniles mortality			
		Concentrations			
		100% (S)	75%	50%	25%
1.	*Artemisia judaica*	100.0 a	100.0 a	96.7 a	85.7 a
2.	*Artemisia monosperma* 1	100.0 a	99.3 ab	96.0 ab	85.3 a
3.	*Artemisia monosperma* 2	100.0 a	98.0 a-d	95.3 b	85.0 a
4.	*Artemisia monosperma* 3	100.0 a	93.0 fg	84.0 c	68.7 e
5.	*Artemisia monosperma* 4	100.0 a	93.0 e-g	85.3 c	73.0 c-e
6.	*Bassia muricata*1	100.0 a	97.3 a-e	84.3 c	72.0 c-e
7.	*Bassia muricata* 2	100.0 a	92.0 fg	85.0 c	72.7 c-e
8.	*Cornulaca monacantha* 1	100.0 a	97.7 a-e	87.0 c	71.0 de
9.	*Cornulaca monacantha* 2	100.0 a	98.3 a-c	86.0 c	70.7 de
10.	*Salsola kali*	100.0 a	95.0 b-g	83.3 c	69.7 e
11.	*Zygophyllum album* 1N	100.0 a	98.0 a-d	84.0 c	78.0 b
12.	*Zygophyllum album* 2N	100.0 a	97.4 a-e	83.2 c	64.3 f
13.	*Zygophyllum album* 3N	100.0 a	86.0 h	74.0 d	71.7 c-e
14.	*Zygophyllum album* 4N	100.0 a	84.3 h	76.7 d	72.3 c-e
15.	*Zygophyllum album* 5N	100.0 a	94.0 c-g	86.3 c	76.7 bc
16.	*Zygophyllum album* 6N	100.0 a	91.0 g	85.3 c	69.0 e
17.	*Zygophyllum album* 1S	100.0 a	96.3 a-f	88.0 c	75.3 b-d
18.	*Zygophyllum album* 2S	100.0 a	93.7 c-g	83.3 c	73.0 c-e
19.	*Zygophyllum album* 3S	100.0 a	93.0 d-g	74.0 d	72.0 c-e
20.	*Zygophyllum album* 4S	100.0 a	94.3 c-g	86.0 c	75.3 b-d
21.	*Zygophyllum album* 5S	100.0 a	93.0 e-g	84.0 c	72.0 c-e
	Control (Distilled water)	0 b	0 i	0 e	0 g

S= Stock solution or standard (5mg/ml); Numbers (1,2,3,…) = mean that the same plant species collected from different locations at North (N) & South (S) Sinai

In the same column numbers followed by same letter (s) are not significant different (P≥ 0.5) according to Duncan's Multiple Range Test.

Results

Based on results shown in Table (2) there were no immobile juveniles in controls with distilled water after 24 hrs. It was observed that all tested plant extracts were toxic to *M. incognita* juveniles in Petri dish experiments by efficacy range from 64.3-100%. The lethal effects of different extracts on *M. incognita* juveniles were observed. Mortality was concentration-dependent, *i.e.* the mortality of the nematode was increased by increasing of extract concentration. Stock solution (SS or 100%) in all tested plants achieved complete death of juveniles (J_2s) as their percentages of mortality (M %) were 100%. At 3/4SS (75%) concentration *A. judaica* showed the same nematicidal activity as in-stock solution (100%), followed by *A. monosperma* 1 (99.3%). The lowest M% (84.0 and 86.3%, consequently) was observed in extract of *Z. album* 3, 4- collected from different sites located in North Sinai-without statistical differences. At 1/2 SS (50%) concentration, *A. judaica* showed the highest M% (96.7%) followed by *A. monosperma* 1 & 2 (96.0 and 95.3%, respectively). The least effective extracts were *Z. album* 3N, 3S (74.0% M for both). The last concentration 1/4SS (25%) had a minor effect on the trend of lethality, so *Artemisia* genus was considered the best toxicant for nematode juveniles in this preliminary experiment with death percent was circled about 85%, followed by 78.0% in *Z. album.* Other tested genera that showed highly nematotoxic were *Bassia muricata* 1, *Cornulaca monacantha* and *Salsola kali* (72.0, 71.0, 69.7 M %, respectively) although they were less effective than the twogenera, *Artemisia* and *Zygophyllum*. They were selected to complete further bioassay in the lab and then *in vivo* to investigate their behavior in nematode-infected plants. The mortality % of J_2 affected by the lowest concentration, 1/4SS or (25%), was the criterion of selecting plants for further evaluations.

Some techniques for enhancing the extraction efficacy were tested (Heating aqueous extract previously prepared in water bath, Microwave oven and Sonicator) for improving nematicidal activity *in vitro.* After 24 or 48hrs of exposure, cold water extraction without any assisting protocol was exhibited the highest efficacy than others, so the cold water extract was used *in vivo* experiments (data not shown). In this study, the drench application was proper and effective than foliar spray (data not shown).

Application time, pre or post-inoculation showed to be effective in the efficacy of botanical extracts and post-application was better than pre one (Table 3). Reduction in the total population of *M. incognita* was highest in post addition of *A. judaica* (87.5%) followed by *A. monosperma* (83.0%) then *Z. album* (81.3 and 77%, consequently), while the minimum reduction was observed in *S. kali* (71.4%). Pre inoculation treatment achieved the greatest reduction in total population by *A. judaica* (71.1%) and the lowest reduction (47.9%) was observed in *S. kali* pre addition.

Data from Table (4) collectively demonstrated that all tested botanical extracts have a positive impact on growth criteria of shoots and roots. Root length of *A. judaica* was increased in pre application similar to *A. monosperma* (66.67 and 60.61%, respectively) but without significant difference. Shoot lengths of all treatments were higher than controls. Generally, *Artemisia* plant, *C. monacantha*, *S. kali* in pre addition were not significantly varied in shoot length parameter but statistically differed as compared to control plants. Major treatments showed not significant differences between root weight values and the check treatment. Shoot fresh weights in half treatment are statistically similar to control plants, the maximum increase was registered as *S. kali*, *Z. album* and *A. monosperma* but these values were non significance. Dry shoot weight of all botanical extracts in both application times was similar to control from statistical view (Table 4), while the highest increase (31.15%) was observed in pre inoculation addition with *A. judaica* followed by *A. monosperma* (28.42%) at the same time of application.

Table 3. Effect of aqueous extracts of selected wild plants applied pre and post-inoculation on *Meloidogyne incognita* development and reproduction infected tomato plants.

Plants species	Time of application	No. galls /roots	R%	No. develop-mental stages/roots	R%	No. adult females /roots	R%	No. egg-masses/ roots	R%	No. eggs/ egg-mass	R%	Final population	R%
Artemisia judaica	**Pre**	46.3 de	45.5	43.3 def	45.8	41.7 def	44.4	41.7 cde	40.5	105.0 ef	51.9	4460.0 ef	71.1
	Post	30.0 g	64.7	27.7 g	65.4	28.7 hi	61.8	26.7 g	61.9	70.0 h	67.9	1923.0 h	87.5
Artemisia monosperma	**Pre**	58.3 bc	31.4	53.3 bc	33.3	49.7 bc	33.8	50.0 bc	28.6	115.0 e	47.3	5853.0 cd	62.1
	Post	40.0 ef	52.9	36.7 efg	54.2	32.7 ghi	56.4	30.0 g	57.1	85.0 g	61.1	2619.3 gh	83.0
Bassia muricata	**Pre**	65.0 b	23.5	60.0 b	25.0	55.0 b	26.7	43.3 cd	38.1	130.0 cd	40.4	5748.3 cd	62.8
	Post	41.0 ef	51.8	36.7 efg	54.2	32.7 c	56.4	33.3 efg	52.4	105.0 ef	51.9	3569.3 fg	76.9
Cornulaca monacantha	**Pre**	65.0 b	23.5	60.0 b	25.0	55.0 b	26.7	50.0 bc	28.6	135.0 bc	38.2	6865.0 c	55.5
	Post	50.7 cd	40.4	45.3 cde	43.3	40.0 defg	46.7	42.7 cd	39.0	100.0 f	54.2	4352.0 ef	71.8
Salsola kali	**Pre**	65.0 b	23.5	60.0 b	25.0	55.0 b	26.7	56.7 b	19.0	140.0 b	35.9	8048.3 b	47.9
	Post	48.3 de	43.1	43.0 def	46.3	38.3 efg	48.9	40.0 def	42.9	108.3 ef	50.4	4414.7 ef	71.4
Zygophyllum album **1N**	**Pre**	52.0 cd	38.8	46.7 cd	41.7	43.0 cde	42.7	40.0 def	42.9	125.0 d	42.7	5089.7 de	67.0
	Post	35.0 fg	58.8	30.0 g	62.5	26.0 i	65.3	27.0 g	61.4	105.0 ef	51.9	2891.0 gh	81.3
Zygophyllum album **5N**	**Pre**	55.0 cd	35.3	50.0 cd	37.5	46.3 cd	38.2	39.3 def	43.8	130.0 cd	40.4	5209.7 de	66.3
	Post	40.0 ef	52.9	35.0 fg	56.3	35.0 i	53.3	31.7 fg	54.8	110.0 ef	49.6	3553.3 fg	77.0
Control (Water only)		85.0 a	0.0	80.0 a	0.0	75.0 a	0.0	70.0 a	0.0	218.3 a	0.0	15438.3 a	0.0

In the same column numbers followed by common letter (s) are not significant different (P≥ 0.5) according to Duncan's Multiple Range Test
Pre= 7 days Pre inoculation, Post= 7 days post inoculation, %R= Reduction percentage

Table 4. Effect of some wild plants water extracts applied pre and post-inoculation with *Meloidogyne incognita* on tomato plant growth.

Plant species	Time of application	Root length (cm)	I%	Shoot length (cm)	I%	Root weight (g)	I%	Shoot fresh weight (g)	I%	Shoot dry weight (g)	I%
Artemisia judaica	**Pre**	36.67 a	66.67	37.67 a-c	46.73	7.43 a	36.89	32.50 b	36.17	4.80 a	31.15
	Post	31.33 bc	42.42	36.33 a-e	41.54	5.90 a-d	8.66	29.73 c	24.58	4.07 a	11.11
Artemisia monosperma	**Pre**	35.33 ab	60.61	40.00 a	55.82	6.97 a-d	28.30	25.53 d	6.98	4.70 a	28.42
	Post	28.67 cd	30.30	32.00 f	24.66	6.30 a-d	16.02	25.53 d	6.98	3.77 a	2.91
Bassia muricata	**Pre**	27.67 cd	25.76	34.67 b-e	35.05	7.40 a	36.28	35.90 a	50.42	4.33 a	18.40
	Post	22.67 e	3.03	32.33 d-f	25.96	5.60 cd	3.13	24.80 d	3.91	3.83 a	4.74
Cornulaca monacantha	**Pre**	32.33 a-c	46.97	40.33 a	57.12	7.33 ab	35.05	30.73 bc	28.77	4.03 a	10.20
	Post	22.33 e	1.52	37.00 a-d	44.14	6.30 a-d	16.02	25.67 d	7.54	4.07 a	11.11
Salsola kali	**Pre**	34.00 ab	54.55	38.67 ab	50.63	7.27 a-c	33.82	38.13 a	59.77	4.07 a	11.11
	Post	29.00 cd	31.82	33.33 c-e	29.85	6.20 a-d	14.18	25.27 d	5.86	3.67 a	0.18
Zygophyllum album **1N**	**Pre**	27.67 cd	25.76	34.00 c-e	32.45	6.13 a-d	12.95	33.27 d	39.38	3.77 a	2.91
	Post	25.33 de	15.15	32.33 d-f	25.96	5.67 cd	4.36	37.30 a	56.28	3.67 a	0.18
Zygophyllum album **5N**	**Pre**	25.67 de	16.67	31.67 ef	23.36	7.27 a-c	33.82	25.37 d	6.28	3.93 a	7.47
	Post	22.33 e	1.52	27.67 fg	7.78	5.93 a-d	9.27	24.27 d	1.67	3.73 a	2.00
Control (Water only)		22.00 e	0.0	25.67 g	25.67	5.43 d	0.0	23.87 d	0.0	3.67 a	0.0

In the same column numbers followed by common letter are not significant different (P≥ 0.5) according to Duncan's Multiple Range Test.

Pre= 7 days Pre inoculation, Post= 7days post inoculation, I %= Increment percentage

Data in Table (5) indicated that all tested plant extracts, significantly affected root populations of *M. incognita* infecting tomato, cv. Super strain-B. In general, the highest concentration (stock or 100%) of each material achieved the highest R% in total population compared to nematode only or untreated or water control (R%) of nematode in roots. The greatest nematode reduction (90.7%) in final population (FP) was achieved by Oxamyl, comparable treatment, but within the botanical extracts 100% of *A. judaica* recorded the maximum reduction in FP (87.0%) that were not statistically differs from Oxamyl followed by *A. monosperma* (83.4%) at the same concentration. The lowest reduction in FP in the stock concentration (60.4%) occurred in *C. monacantha*. At the lowest concentration (25%) it was found the plants of *Artemisia* showed their suppression ability with highest reduction (61.2 & 57.8%) in FP. *C. monacantha* at this concentration possessed the lowest ability to suppress nematode reproduction (42.3%) followed by *S. kali* and *B. muricata* (48.0 & 49.2%, respectively). Nematode fecundity (eggs/egg-mass) was suppressed in all tested concentrations of wild plants, the lowest numbers of eggs (60) were formed in tomatoes roots treated by nematicide superseded *A. judaica* (62) and *A. monosperma* (65) but they didn't statically different. The development of *M. incognita* juveniles to mature females was highly suppressed by both species of *Artemisia* at all concentrations and *Z. album* 1N at the highest concentration (55.6%) whereas *B. muricata* was the lowest inhibitor (33.9%) at highest concentration.

Results presented in Table (6) showed that aqueous extracts of different wild plants had some positive effects on the fresh and dry shoot weight of tomato. Statistical analysis confirmed that fresh shoot weight was non-significantly affected by different concentrations in most cases, compared to the control (P=0.05), except *A. judaica* at 100 & 75% concentration, *A. monosperma* and *Z. album* 100%, respectively compared with water or Oxamyl controls. The highest increased in fresh shoot weight was in *A. judaica* (23.86%). Fresh shoot weight was statically differed in most treatments except all concentrations of *S. kali* and the lower concentration of *B. muricata* and *Z. album* 5N. The impact on root weight was maximized by the stock concentration of *A. judaica* (27.63%) followed by *A. monosperma* (26.84%) but there are no significance differences in most cases compared with controls, while the minimum increment (3.3%) was recorded by the lowest concatenation of *S. kali*. Shoot length significantly increased after addition of plant extracts, *A. judaica* and *A. monosperma* possessed the highest increment (25.74% & 23.47, consequently) without significant differences between both. The length of root was increased by all extract treatments, maximum increase was registered by *A. judaica* succeeded by *A. monosperma* and *Z. album* 1 & 5 (32.6, 27.92, 26.9 and 25.86%, respectively).

Discussion

Plants are a source of naturally occurring pesticides. Several nematicidal compounds have been isolated from plants, mainly from members of the family Asteraceae (Gommers, 1981). Other nonvolatile compounds from plants, such as polyacetylenes and benzofuran derivatives, also have antinematodal activity (Gommers & Bakker, 1988), however, none has been developed as a commercial nematicide. Allelochemicals are plant-produced compounds, ecofriendly, that affect the activity of other organisms and are thought to be toxins and secondary metabolites that act as attractants or deterrents (Dodds, 1996; Brown & Morra, 1997). Plant extracts of antagonistic plants or their byproducts for managing plant-parasitic nematodes have been used globally against conventional chemical nematicides and have been reported by many investigators (Pandey & Dwivedi, 2000; Pandey *et al*., 2001; Abdel-Rahman *et al*., 2017; Haroon *et al*., 2018).

Table 5. Effect of selected wild plant extracts at different concentrations applied post inoculation on *M. incognita* development and reproduction infecting tomato plants in pot trial.

Plant species	Concen-tration	No. galls/ roots	%R	No. devlop. stage/roots	%R	No. adult females/roots	%R	No. egg-masses/roots	%R	No. eggs/ egg-mass	%R	Final population	%R
Artemisia judaica	100	22.3 l-n	68.1	19.0 o	68.0	20.0 h	66.7	15.7 l	57.2	62.0 p	60.0	1026.0 qr	87.0
	75	30.0 d-g	57.1	21.0 n-o	64.6	22.0 h	63.3	20.7 jk	48.9	72.0 no	53.5	1551.7 pq	80.3
	50	31.7 c-f	54.8	25.0 l-n	57.9	24.0 h	60.0	24.7 g-j	42.2	74.7 mn	51.8	1915.4 m-o	75.7
	25	34.0 cd	51.4	26.7 k-m	55.1	25.7 h	57.2	33.0 cd	28.3	90.0 g-k	41.9	3055.3 f-i	61.2
Artemisia monosperma	100	17.7 n	74.8	27.0 kl	54.5	23.0 h	61.7	19.0 kl	62.0	65.0 op	58.1	1304.0 pq	83.4
	75	19.7 mn	71.9	21.7 m-o	63.5	25.0 h	58.3	21.3 i-k	57.3	71.7 no	53.8	1596.9 n-p	79.7
	50	21.7 l-n	69.0	22.3 l-o	62.4	27.0 h	55.0	27.7 e-g	44.7	94.7 h-j	38.9	2696.1 g-j	65.7
	25	24.3 h-m	65.2	23.3 l-o	60.7	28.3 h	52.8	31.7 c-e	36.7	102.3 d-h	34.0	3323.9 d-f	57.8
Bassia muricata	100	21.7 l-n	69.0	38.7 f-h	39.9	39.7 d-g	33.9	21.7 i-k	36.7	92.7 i-k	40.2	2107.8 l-n	73.2
	75	23.3 l-n	68.1	43.7 d-f	31.5	46.7 b-f	22.2	25.0 f-i	32.0	98.7 g-j	36.3	2582.0 h-l	67.2
	50	23.0 k-n	67.1	45.7 c-e	27.0	47.3 b-e	21.1	27.7 e-g	28.7	111.0 cd	28.4	3191.7 e-g	59.4
	25	24.0 i-m	65.7	49.0 bc	19.1	50.3 bc	16.1	32.3 cd	19.3	119.7 b	22.8	4000.9 c	49.2
Cornulaca monacantha	100	25.0 h-m	64.3	35.7 h-i	34.8	38.7 e-g	35.6	31.7 c-e	56.7	95.0 h-j	38.7	3114.3 f-h	60.4
	75	28.7 d-j	59.0	40.7 e-g	26.4	42.7 c-g	28.9	34.0 c	50.0	108.3 c-e	30.1	3800.7 cd	51.7
	50	29.7 d-h	57.6	43.3 d-f	23.0	46.7 b-f	22.2	35.7 c	44.7	114.3 bc	26.2	4203.6 bc	46.6
	25	32.7c-e	53.3	48.0 b-d	17.4	52.7 ab	12.2	40.3 b	35.3	109.0 cd	29.7	4537.3 b	42.3
Salsola kali	100	28.3 e-k	59.5	41.3 e-g	30.3	37.0 g	38.3	22.7 h-k	54.7	98.0 g-j	36.8	2322.3 j-m	70.5
	75	34.0 c	51.4	47.7 b-d	19.7	40.3 d-g	32.8	24.3 g-j	51.3	100.0 e-i	35.5	2545.7 i-l	67.6
	50	36.0 b	48.6	51.7 b	12.9	48.0 b-d	20.0	26.3 f-h	47.3	111.3 c	28.2	3057.8 f-i	61.1
	25	39.7b	43.3	56.3 a	5.1	50.3 bc	16.1	32.3 cd	35.3	122.3 b	21.1	4094.4 bc	48.0
***Zygophyllum album* 1N**	100	22.3 l-n	68.1	24.3 l-n	59.0	26.7 h	55.6	21.0 i-k	58.0	75.3 mn	51.4	1654.0 n-p	79.0
	75	23.3 j-m	66.7	32.0 i-j	46.1	37.0 g	38.3	25.0 f-i	50.0	85.3 kl	44.9	2227.3 k-m	71.7
	50	24.7 h-m	64.8	41.3 e-g	30.3	43.3 c-g	27.8	29.0 d-f	42.0	93.3 i-k	39.8	2820.3 f-j	64.2
	25	31.7c-f	54.8	45.7 c-e	23.0	42.7 c-g	28.9	33.3 c	33.3	107.0 c-f	31.0	3688.3 c-e	53.1
***Zygophyllum album* 5N**	100	21.3 m-n	69.5	30.0 jk	49.4	38.3 fg	36.1	20.7 jk	58.7	81.7 lm	47.3	1776.8 m-p	77.4
	75	23.3 j-m	66.7	36.3 j-i	38.8	41.7 c-g	30.6	23.7 g-j	52.7	91.0 g-k	41.3	2255.3 j-m	71.3
	50	29.3 d-i	58.1	41.0 e-g	30.9	40.3 d-g	32.8	29.0 d-f	42.0	102.3 d-h	34.0	3078.0 f-i	60.9
	25	27.1 h-l	61.2	45.0 c-e	24.2	48.0 b-d	20.0	35.0 c	30.0	106.0 c-g	31.6	3838.0 cd	51.2
Control	Water	70.0 a	0.0	59.3a	0.0	60.0 a	0.0	50.0 a	0.0	155.0 a	0.0	7919.3 a	0.0
	Oxamyl	11.0 o	84.3	10.7 b	82.0	9.3 i	84.4	11.7 m	63.9	60.0 p	61.3	731.7 r	90.7

Concentrations100 % = stock solution, R% = Reduction percentage

Table 6. Effect of selected plant extracts at different concentrations on growth criteria of tomato plantsinfected with *M. incognita* in pot trial.

Plant species	Concen-tration	Root length (cm)	I%	Shoot length (cm)	I%	Fresh shoot weight (g)	I%	Root weight (g)	I%	Dry shoot weight (g)	I%
Artemisia judaica	100	25.7 a	32.60	33.7 a	25.74	33.9 a	23.86	5.7 a	27.63	3.5 a	29.56
	75	21.9 a-e	23.68	31.7 a-c	21.88	32.1 a-c	16.62	5.3 a-e	23.90	3.1 ab	23.53
	50	21.7 b-g	20.15	31.9 a-e	19.35	32.4 a-e	15.37	5.4 a-e	22.83	3.1 a-f	18.75
	25	20.0 e-g	13.50	27.7 b-h	9.64	32.8 a-e	15.26	5.5 a-e	22.04	3.3 b-f	14.43
Artemisia monosperma	100	24.0 ab	27.92	32.7 ab	23.47	33.4 ab	21.42	5.5 ab	26.48	3.2 a-c	22.49
	75	21.7 c-g	20.15	31.0 a-e	19.35	32.4 a-c	19.81	5.4 a-c	24.21	3.0 a-f	18.66
	50	20.7 c-i	16.29	31.0 a-e	19.35	31.2 a-e	18.30	5.3 a-c	21.29	3.0 a-f	17.11
	25	19.3 g-j	10.52	30.3 a-f	17.58	31.3 a-e	13.97	5.3 a-e	21.63	2.8 b-f	12.62
Bassia muricata	100	18.3 g-j	5.64	28.0 b-g	14.77	30.1 a-f	8.52	5.0 c-e	18.69	2.8 b-f	14.63
	75	21.0 b-h	17.62	28.3 c-h	11.76	29.2 a-g	7.87	4.7 d-e	15.99	2.7 b-f	7.61
	50	20.3 g-ij	14.92	27.7 d-g	9.64	28.1 c-g	5.53	4.6 c-e	12.81	2.6 c-f	4.88
	25	20.0 e-g	13.50	29.0 b-h	13.79	26.9 e-g	4.49	4.5 e	8.81	2.5 d-f	2.76
Cornulaca monacantha	100	21.0 b-h	17.62	29.3 b-g	14.77	29.5 a-f	13.57	5.0 a-e	16.94	2.9 b-f	13.64
	75	19.0 g-j	8.95	29.7 a-g	15.73	30.8 a-e	10.29	4.8 b-e	20.44	2.8 b-f	11.15
	50	18.7 g-j	7.32	27.0 e-h	7.41	30.7 a-e	8.07	4.7 c-e	20.10	2.7 b-f	8.52
	25	18.3 g-j	5.64	25.7 d-h	2.60	28.0 c-g	17.78	5.3 a-d	12.50	2.6 d-f	4.14
Salsola kali	100	20.0 e-g	13.50	31.0 a-e	19.35	28.6 b-g	15.10	5.1 a-e	14.13	2.7 b-f	8.41
	75	19.0 g-j	8.95	27.9 b-g	14.77	28.1 c-g	12.23	5.0 b-e	12.60	2.7 d-f	4.26
	50	18.0 h-j	3.89	27.0 e-h	7.41	27.3 d-j	15.10	5.1 a-e	10.15	2.5 c-f	2.88
	25	17.7 ij	2.08	25.7 b-h	2.60	25.4 fg	12.82	5.0 b-e	3.30	2.5 e-f	1.72
***Zygophyllum album* 1N**	100	23.7 a-c	26.90	30.0 a-f	16.67	32.2 a-d	13.34	5.0 a-e	23.74	3.1 a-d	21.09
	75	23.1 d-f	22.54	28.3 c-h	11.76	32.5 a-f	14.76	5.1 a-e	18.77	3.2 a-f	16.27
	50	21.0 b-h	17.62	31.3 a-f	18.48	32.5 a-f	13.28	5.2 a-e	17.68	3.2 a-f	18.21
	25	20.0 e-g	13.50	30.9 a-c	21.05	32.4 a-f	12.64	5.2 b-e	17.13	3.2 a-f	17.76
***Zygophyllum album* 5N**	100	23.3 a-d	25.86	30.0 a-f	16.67	32.5 a-c	18.51	5.3 a-c	24.45	3.1 a-e	19.89
	75	21.7 b-g	20.15	29.7 a-g	15.73	32.3 a-c	15.76	5.1 a-e	23.98	3.0 a-f	18.48
	50	21.3 b-g	18.91	27.7 d-h	9.64	28.2 c-g	15.10	5.1 a-e	13.01	2.9 b-f	14.14
	25	20.7 c-i	16.29	26.7 f-h	6.25	26.9 e-g	10.97	4.9 b-e	8.92	2.5 e-f	2.50
Control	Water	11.3 j	0.00	25.0 h	0.00	4.8 g	0.00	24.5 b-e	0.00	2.5 f	0.00
	Oxamyl	23.7a-c	26.90	29.7a-g	15.73	4.8 c-g	9.16	28.0 c-e	12.39	2.8 b-f	12.31

I%= increment percentage= [(reading of a treatment – reading of check)/reading of check] x100

Nematicidal activity of aqueous extract at different dilutions (100, 75, 50, 25%) of certain twenty-one wild plants collected from different locations in North and south Sinai were tested *in vitro* against *M. incognita* juveniles. All botanical extracts were lethal to the nematode larva. Seven medicinal plants selected from preliminary experiments at 25% concentration namely: *Artemisia judica, A. monosperma, Bassia muricata, Cornulaca monacantha, Salola kali, Zygophyllum album* and *Z. album*, were evaluated in this study.

In the current study, the examined wild plant extracts of *A. judica, A. monosperma, B. muricata, C. monacantha, S. kali, Z. album* proved to be effective against RKN, *Meloidogyne* spp. These results are in agreement with previous work with Asteraceae (Al-Obaedi *et al*., 1987; Oka *et al*., 2000; Al-Banna *et al*., 2003; Costa *et al*., 2003; Tsay *et al*., 2004; Abdel-Rahman & Saleh, 2006; Zouhar *et al*., 2009; D'Addabbo *et al*., 2013; Nour El-Deen *et al*., 2014; Kepenekci *et al*., 2016; Samaliev *et al*., 2017). It was also observed that mortality increased with increasing concentration of the extract, the highest mortality was recorded in a concentration of 100% (Stock). A similar finding was reported by Ameer-Zareen *et al*., 2003; Haroon *et al*., 2018. Furthermore, the toxicity of *Artemisia* plants is due to some nematode-lethal compounds viz., caffeic acid, chlorogenic acid, and artemisinin (D'Addabbo *et al*., 2013). The superiority of *Artemisia* spp., over the rest examined plants also support the previous finding of Al-Obaedi *et al*., (1987) as they recorded mortality up to 100% on *M. javania* juveniles and also in a recent study of Abdel-Rahman *et al*., 2017 that *A. judaica* water extract killed 100% of *M. incognita* juveniles. Abdel-Rahman & Saleh (2006) recorded a high mortality rate (78%) of water extract from *A. monosperma* against *M. incognita*; they also reported nematicidal activities of phytochemicals from some arid land plants including *Z. album* against *M. javanica* juveniles and eggs. *Salsola* sp. has nematicidal activities reported by Abbasi *et al*., (2010); they found that halophytes significantly reduced hatching and enhanced mortality of J_2s *in vitro*.

The pre-inoculation application was found to be less effective than post inoculation addition. These findings are not matched with the results of Al-Obaedi *et al*., (1987) as they found pre-application was better than post-application without significant differences at the specific concentrations.

Nematicidal activity of genus *Zygophyllum* was proved by many authors: Abdel-Rahman & Saleh (2006) found that second-stage larvae of *M. incognita* incubated in *Z. album* extract for 48 hours, then inoculated to healthy tomato seedlings reduced pathogenicity of *M. incognita*. El-Sherbiny & Al-Yahya (2010) observed that powders aerial parts of *Zygophyllum migahidii* achieved relative nematicidal efficacy against *M. javanica*. Based on our results, we concluded that pesticide treatment decreased the nematodes infection, followed by 100% aqueous extract from all seven medicinal plants.

Among the seven tested plants in the current study, aqueous extract of *A. judaica* was the most effective treatment to decrease the nematodes infection in tomato crops. The nematicidal effect of the tested extracts may be attributed to their high contents of certain oxygenated compounds which are characterized by the lipophilic properties that enable them to dissolve the cytoplasmic membrane of nematode cells and their functional groups interfering with the enzyme protein structure (Knoblock *et al*., 1989). From the economic point of view, these results indicated that *A. judica* was the most effective plant against *M. incognita* development and reproduction on highly susceptible tomato cultivar that was used in this study. The normal aqueous extract at concentration 100% was not significantly different from 75%, besides, the 50% gave a satisfactory effect in reducing nematode population (75.7%), so it can recommend by using them in the lower concentration in South Sinai by Bedouins or farmers colonized these areas in managing PPN

and enhancing plant growth. Also, *Z. album* plants, native in South Sinai, can be used in this respect. *A. monosperma* at concentration 100 or 75% are statically equal, furthermore, it is considered abundant species in some habitats of North Sinai, so the application of these plants as an extract or soil amendment could be beneficial in managing nematode. *Artemisia* species are abundant in many habitats and highly distributed in Egyptian deserts (El Maggar, 2012), which facilitate their application by farmers and can be employed in controlling root-knot nematodes according to their abundance in such location.

Conclusion

In conclusion, this study has shown that extracts of some wild plants located in Sinai Peninsula (in particular *Artemisia* and *Zygophyllum*) are very beneficial in the management of root-knot nematodes (RKN). These materials are common and are found in abundance easily. This finding could have the promise for managing RKN without the use of chemical pesticides and seem to have the potential to be developed into new nematicidal formulate or biopesticides (BP), though their activity should be validated under actual field conditions. Sustained ecofriendly strategies like the use of plant powders as a soil amendment or plant extracts in the cheapest solvent, water, can help growers especially smallholder farmers in combating root-knot diseases in plants as well as improving the nutritive states of soil and its health. So we recommend and encourage these approaches as they are almost cheap with no negative effects on the environment for sustainable agriculture. The control of *M. incognita* by the botanical extracts used in this study might be probably based on a complex mode of action involving multiple mechanisms. Therefore, further studies are needed to characterize the active compounds in the tested plants that have antinematodal activity and possessing complex modes of action. Also, research should be frequent in many areas such as production, formulation, delivery, and commercialization of biopesticides.

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