Egyptian J. Desert Res., **73**, No. 2, 419-436 (2024)

EFFECT OF POMEGRANATE PEEL EXTRACT (*PUNICA GRANATUM*) AGAINST PLANT PARASITIC NEMATODES

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Plant nematodes of *Meloidogyne incognita* are of great economic importance and lead to major damage to several crops. Therefore, the current study was to assess pomegranate (*Punica granatum*) peel's nematicidal potential through aqueous and ethanolic extracts for nematicidal potential. Chemical analyses were implemented for pomegranate fruit peel ethanolic extract for phenolic, flavonoid, saponins and anthocyanin contents. Additionally, the plant aqueous extracts at varying concentrations of 2.5, 5.0, and 7.5% were screened for egg hatchability and nematicidal activity against second stage juveniles of *M. incognita* in vitro. Results indicated that the alcoholic extract killed second stage juvenile root-knot nematodes and prevented egg hatching more effectively than the aqueous extract. The ethyl alcohol peel extract of *P. granatum* showed a highly promising mortality rate of 76–100% after 48 hours of exposure, and it fully blocked the egg hatching after 4 days at concentration 75 ppm. As extract concentration increased, J2 juvenile mortality and egg hatching gradually decreased. In greenhouse tests, when pomegranate fruit peel powder was used as a soil amendment, Pepper seedlings showed phytotoxicity at treatment rates of 10, 20, and 30 g/kg as compared to the untreated plants. It was noted that there were less larvae, galls, egg masses, and eggs. The growth of the pepper plants greatly enhanced compared to the untreated control plants. These results are promising for the development of eco-friendly natural nematocide agents and potential ways for their integration into sustainable management strategies.

Keywords: pomegranate peel, *Meloidogyne incognita*, phytochemical analysis, soil amendment

INTRODUCTION

Plant parasitic nematodes harm plants in ways that are frequently subtle and might be mistaken for nutritional issues (Trambadiya et al., 2023). Less than a dozen of the hundreds of distinct nematode species that can infect plants

are economically significant root feeding diseases. If there are a lot of damaging nematodes present, plant growth will suffer (Desai, 2007 and Jones et al., 2013). Many plants are impacted by plant nematode *Meloidogyne incognita*. It is extremely harmful to crops, resulting in significant harm and decreased productivity. Nematodes negatively impact the host plant's normal physiology, growth, and development during parasitism. This phenomenon has been linked to the host's direct and indirect reaction to the nematode's mechanical and biochemical activities (Bhargava et al., 2007). Nematode abundances and structural indices varied greatly based on the host weed species, crop types and soil characteristics (AbdelRazek and Balah, 2023). Root-knot nematodes are the most important plant parasitic worms in the world, causing galling, stunting, and yellowing in addition to nutritional inadequacies and production losses in most temperate and tropical crops (Crow, 2019). It is well recognized that chemical nematicides can alter the soil biosphere and provide a risk of contamination. Applying botanicals to nematode management is simple and doesn't harm the environment (Sundarababu, 2000).

For thousands of years, people have utilized plants for food and medicine, and they have learned a great deal about their characteristics (Brouwer et al., 2005). Because they produce a wide range of bioactive chemicals. Globally, there is a steadily increasing need for nature-based biopesticides, especially those made from plants. Biopesticides will provide an alternative to those traditional pesticides because they are more affordable, safer, easily accessible, biodegradable, and environmentally benign (Ranasing, 2007). The dwarf variant of *Panica granatum* is commonly used as a bonsai specimen tree and as an ornamental plant in gardens and bigger containers. Pomegranates represent life, longevity, health, feminity, fecundity, knowledge, morality, and immortality and if not divinity, then spirituality. The pomegranate is regarded as "a pharmacy unto itself" in Aurvedic medicine (Mahdihassan 1984).

Numerous researchers have reported on the pharmacological characteristics of different extracts of different portions of this plant, including antifungal efficacy against plant pathogenic fungi (Tehranifar et al., 2011 and Al-Askar, 2012). The pharmacological properties of various plant components have been connected to their high concentration of bioactive secondary metabolites, such as polyphenols, glycosides, triterpenes, sterols, flavonoids, anthocyanins, triglycerides, tannins, and alkaloids (Tantray et al., 2009).

Alternative methods of controlling plant parasitic nematodes are now required due to the disadvantage of synthetic nematicides. Thus, the nematicidal activity of biological products, such as chemicals generated from plants and soil amendments, is being studied. Plants are potential sources of biologically based management agents that could be employed or modified for use against phytopathogenic nematodes. The pomegranate (*P. granatum*) is a globose berry that is eaten fresh. It contains edible arils with seeds within (which

contain 75–80% juice and 20–25% seeds), as well as peel, which is a non-edible portion of the fruit that makes up around 40–55% of its weight (Magangana et al., 2020). In addition to preventing the hatching of the eggs of the root-knot nematodes (RKN), pomegranate fruit powder water extracts reduced the motility and viability of the plant-parasitic nematodes *Helicotylenchus dihystera* and *M. incognita* (Korayem et al., 1993).

Polyphenols are the primary elements of the 124 phytochemicals found in pomegranate fruits, albeit the ones that are harmful to plant-parasitic nematodes have not yet been identified. In traditional medicine, pomegranates have also been used as an alcohol extract (Viuda-Martos et al., 2010; Akhtar et al., 2015 and García-Villalba et al., 2015) and as a treatment for intestinal worms (Ismail et al., 2012). Many of the phenolic compounds are nematicidal, meaning they hinder the movement of nematodes or repel them (Ohri and Pannu, 2010 and Ntalli and Caboni, 2012). About 48 flavonoids, including anthocyanins, and phenolics, including tannins, are found in the peel and other fruit parts (Viuda-Martos et al., 2010; Ismail et al., 2012 and Akhtar et al., 2015). Because they contain more polyphenols, especially ellagitannins, than seeds, as waste byproducts of the pomegranate juice industry, pomegranate husk and peel may include substances that could function as biological nematicides (Seeram et al., 2005; Akhtar et al., 2015 and García-Villalba et al., 2015).

Nematocidal compounds found in pomegranate peel and husk suggest that these plants may provide biologically based products or amendments that could help control nematodes. This probability is increased by reports of pomegranate powders suppressing nematodes (Ibrahim, 2011 and Belay 2023). The most significant nematodes in the world are root-knot nematodes *M. incognita* that seriously harm important crops. One of Egypt's most significant vegetable crops is pepper (*Capsicum annuum*). The southern root-knot nematode *M. incognita* can infect the most common pepper types. Therefore, the purpose of this study was to ascertain whether: i) pomegranate peel extracts would prevent RKN eggs from hatching and be deadly to J2. When used as extracts, a soil addition, and dried pomegranate peel powder for suppressing RKN populations in pepper crops.

MATERIALS AND METHODS

1. Preparation of *Punica granatum* Peel Extract

1.1. Aqueous extract

According to Handa et al. (2008), 250 ml of distilled water (1:10) and 25 g of pomegranate peel powder were combined to create the aqueous extract. The combination was shaken for 30 min at a speed of 150 cycles per minute, and it was left to filtrate for 24 h. A concentrated extract (10%) was obtained in the drying jar after the mixture was filtered through several layers to remove the insoluble plant materials and then again using filter papers

(Whatmann No. 2). The extract was then kept in sterile vials at four degrees Celsius. A series of concentrations (25, 5.0, and 7.5%) was prepared for testing.

1.2. Alcoholic extract

The preparation followed the procedure of Gülçin et al. (2003), this involved mixing 250 g of powdered pomegranate peel with 2500 ml of 96% ethanol alcohol, stirring for 24 h on a magnetic stirrer, and then filtering twice using filter paper (Whatmann No. 1). A rotary evaporator was used to concentrate the filtrate, after that, it was placed in sealed bottles, dried in an electric oven set to 40 degrees Celsius, and kept in the refrigerator until it was needed. Serials of 25, 50, and 75 ppm concentration were prepared, and the process was repeated in the same circumstances.

1.3. Preliminary phytochemical screening

1.3.1. Determination of total flavonoids

Separate aliquots of the quercetin solution weighing five to three hundred micrograms were added to a test tube and dried by evaporating in a hot water bath (40 to 50°C). Petroleum ether was used to extract two grams of each defatted powder. Then, ethanol (95%) was added, and the extract's volume was adjusted to 50 ml. After transferring 5 ml of the extract to a test tube, 5 ml aliquots of the 0.1 M $AlCl_3$ reagent were added. In a water bath, the solution was evaporated until it was completely dry. UV light was used to measure the color's absorbance at 266 nm for kaempferol and 445 nm for quercetin (Karawaya and Aboutabl, 1982).

1.3.2. Determination of total phenolic compounds

Two hundred microliters of 70% methanolic extract (1 mg/ml) were mixed with three milliliters of distilled water. 0.5 ml of Folin-Ciocalteu reagent was thoroughly mixed for 3 min, and then 2 ml of 20% (w/v) sodium carbonate was added. After letting the mixture remain for another 60 min in the dark, absorbance was measured at 650 nm. The results were expressed as milligrams of gallic acid equivalent per gram of dry weight, and the calibration curve was utilized to calculate the total phenolic content (Kaur and Kapoor, 2002).

1.3.3. Determination of total saponins

Kurkin and Ryazanova (2018) conveyed that a 100 ml conical flask with a ground-glass joint (1 g, precise weight) is filled with a plant powder sample that has been ground to a particle size that can pass through a 3-mm screen. The sample is then treated with 70% EtOH (50 ml). A stopper is used to seal the flask once it has been tared to ±0.01 g. Following 60 min of refluxing on a water bath, the flask is sealed with the same stopper, allowed to cool to room temperature for 30 min, and then weighed again with a reflux condenser attached.

According to Kurkin and Ryazanova (2018), Following 60 min of refluxing on a water bath, the flask is sealed with the same stopper, allowed

to cool to room temperature for 30 min, and then weighed again with a reflux condenser attached. The lost solvent is replaced with 70% EtOH. The working reference standard (WRS) solution is prepared. A 100 ml volumetric flask is filled with 0.05 g of exact, dissolved in 20 ml of purified H_2O, and then adjusted with purified H_2O to the mark. Conc. H_2SO_4 (4 ml) is added to the resultant solution (1 ml), which is then heated and swirled for 15 min in a boiling water bath. A spectrophotometer is used to measure the optical density at 510 nm in a 10-mm cuvette. Purified water is used as the reference solution. The formula Y=10.05X+0.001 is used to determine the total saponin content in percent (X).

1.3.4. Determination of anthocyanins

Anthocyanins were extracted using the method described by Rodriguez and Wrolstad (2001). A stainless-steel Waring blender (Waring Commercial, Torrington, Conn., USA) was used to grind the plant material into a powder. After mixing the powder with 2 L of acetone per kilogram of material, the recovered substance was filtered through a Buchner funnel to separate it from the cake. Twice, 70% (v/v) aqueous acetone was used to re-extract the filter cake residual. The filtrates were combined and separated using chloroform (1: 2 acetone: chloroform, v/v), and then they were stored at 1°C for the duration of the night. The aqueous component containing the ACNs was recovered and separated from the remaining acetone at 40°C in a Buchi evaporator. It was then resolubilized in 0.01% HCl. The extractions were performed four times. All extracts were stored at -70°C before analysis.

2. Bioassays

2.1. Culture for the nematode inoculum

Highly infected eggplant (*Solanum melongena* L.) roots were used to create a pure nematode culture. The eggs were carefully uprooted to prevent the egg masses from separating from the root, and they were thoroughly cleaned in distilled water to remove all the soil debris. They were then put in petri dishes with water that was just deep enough to touch the egg masses, which could benefit the juvenile hatchling, and 15 mesh sieves (8 cm in diameter) with a cross-layered layer of tissue.

2.2. Laboratory assay

2.2.1. Effect of plant extracts on juvenile mortality

Using the methodology outlined by Al-Sayed et al. (1996). The nematicide effects of the defatted ethanolic extract and water extract on the hatchability of eggs and mortality of juveniles of *M. incognita* were evaluated. One milliliter of each nematode, such as *M. incognita*, contains roughly 100 recently hatched juveniles after being individually moved into Petri plates with a diameter of 5 cm and 5 ml of aqueous (2.5, 5, 7.5%) or ethanolic solution of each tested concentration (25, 50, and 75 ppm), the nematode was incubated at 25±°C. Under a stereoscopic microscope, the number of dead and alive juvenile *M. incogentia* was counted after 24 and 48 h, and the mortality

percentage was calculated. Three Petri dishes with one milliliter of distilled water served as the control and three duplicates of each concentration were made.

2.2.2. Effect of plant extracts on hatching test

Five fresh, uniformly sized egg masses were selected from properly cleaned eggplant roots (*S. melongena* infested with the root-knot nematode, *M. incognita*). Under a stereoscopic microscope, the total number of eggs was counted and expressed as the number of eggs per milliliter of water (Jenkins, 1964). After being moved to 40 mm Petri dishes with 5 ml of extract solution of each tested concentration (25, 50, and 75%), the collected egg masses were incubated at 25±°C. There are three duplicates of each treatment. After four days, the number of hatched juveniles was counted using a stereoscopic microscope, and the percentage of inhibition over control was computed.

2.3. Soil amendments experiment

The effect of dried powdered pomegranate (*P. granatum*) fruit peel on the *M. incognita* population was investigated in a greenhouse experiment. One-month-old peppers of the Khayrat variety of *Capsicum annuum* were transplanted individually in clay pots measuring 15 cm and filled with sandy clay soil (2:1, v:v) at rates of 10, 20, and 30 g/kg. After a week, 2000 newly hatched juvenile *M. incognita* were added to each pot. Three duplicates of each treatment were used. Each treatment was set up in a greenhouse using a completely randomized block design. Untreated plants control and the bio-product Micronema (*Pseudomonas* sp. and *Bacillus* sp.) applied at a rate of 2 m/pot. Pots were watered every three days on a periodic basis. The plants were collected after the inoculation for 45 days. The % decrease in nematode counts and the calculation of plant growth parameters.

The Baerman-pan method was used to prepare the soil in each pot for nematode extraction (Southey, 1970). The number of second stage juveniles (J2) in the soil of each pot was counted using a stereoscopic microscope using a Hawksley counting slide. To determine the average number of eggs/egg masses, four randomly selected egg masses per root system of each replicate were also rinsed in 1% sodium hypochlorite to separate the eggs from the egg matrix.

The released eggs were then counted under a stereoscopic microscope while suspended in water. Juveniles collected were tallied. Egg masses and galls were categorized as indices. The following formula was used to determine the reduction in nematode parameters, including the percentage decrease in gall development, egg mass production, and juvenile number (Raddy et al., 2013).

$$\text{Reduction (\%)} = (\text{Control} - \text{Treatment})/(\text{Control}) \times 100$$

For each treatment, the fresh weight and length of the roots as well as the fresh and dried shoot weights were used to measure plant growth response. The statistical analysis was conducted using the "Anova" Test (Snedecor and

Cochran, 1980). At the ninety-five percent probability level, treatment means were compared using Duncan's Multiple Range Test. These methods were completed using SPSS program version16.

RESULTS

Table (1) shows the PPE's antioxidant effects (the total phenols, flavonoids, saponins and anthocyanin. It was noted that the content of phenols and flavonoids saponins and Anthocyanin in the alcoholic extract increased, followed by the aqueous extract.

Table (1). Phytochemical constituents test indicated the presence of some important compounds in the alcoholic extract of *Punica granatum* (peels).

Extract	Total phenolics conc. (mg/g)	Total flavonoids conc. (mg/g)	Total saponins conc. (mg/g)	Total anthocyanins conc. (µg/g)
Alcoholic extract	150	126.83	20.12	285.52
Aqueous extract	140	118.00	14.50	265.33

Table (2) and Fig. (1) present the results of an in vitro investigation into the nematicide effect of ethanol and water extract on egg hatchability and juvenile *M. incognita* nematode mortality. The aqueous extract demonstrated ovacidal and larvicidal properties at concentrations between 2.5 to 7.5%. Generally, the extract with a higher concentration exhibited greater activity in egg hatchability and mortality after 24 and 48 h by 74, 70 and 82.0% as compared with its control. This finding demonstrated that a rise in concentration indicates an additional input of several active substances in this water extract.

As for ethanol extract, the effect of the extract on the hatching of *M. incognita* eggs (Table 2) indicated that the highest concentration of ethanol solution was found to completely suppress the hatching (100%) and achieved complete mortality after 24 and 48 h (100%). While, 75 and 50 ppm decreased egg-hatching by 95.0 and 70.0% as compared with its control. The ethanolic dilutions of 75 ppm exhibited mortality reached 79 and 83% after 24 and 48 h exposure, respectively. While the extract at 50 ppm exhibited mortality by 61.0 and 76.0% after 24 and 48 h from exposure respectively as compared with the control.

In the greenhouse, Table (3) and Fig. (2) show that when pepper plants were cultivated in soil infested with *M. incognita*, it was discovered that the bioactive components of *P. granatum* led to a decrease in the number of nematodes, larvae, galls, egg masses, and eggs. By increasing the concentration of the evaluated bioactive components from 10 to 20 g and beyond, this impact amplified the plant growth. Conversely, pepper plants cultivated in soil that were infected with each of *M. incognita* and unamended with the bioactive

components of pomegranate powder showed negligible variations in soil nematodes J2 counts.

Table (2). Effects of dried, ground powders of *Punica granatum* (peels) on egg hatching and nematode mortality of *Meloidogyne incognita* in vitro.

Extraction medium (Solvent)	Conc.	Egg hatching inhibition after 4 days	% of egg hatching inhibition after 4 days	Nematode mortality			
				24 h		48 h	
				Nematode mortality	% of nematode mortality	Nematode mortality	% of nematode mortality
Water extract	2.5%	182.33± 0.33[f]	48	46.00± 3.46[d]	46	54.00± 2.89[c]	54
	5.0%	213.00± 3.79e	56	57.00± 2.31[d]	57	68.00± 2.89[b]	68
	7.5%	282.00± 3.51[c]	74	70.00± 1.15[b]	70	82.00± 1.15[a]	82
Ethyl alcohol	25 ppm	265.00± 3.21[d]	70	61.00± 1.73[c]	61	76.00± 3.46[b]	76
	50 ppm	361.33± 3.76[b]	95	79.00± 4.04[b]	79	93.00± 1.73[a]	93
	75 ppm	388.00± 0.00[a]	100	100.00± 0.00[a]	100	100.00± 0.00[a]	100
Distilled water)		388.00± 0.00[a]	0	0.00	0	00	0
F value P-value		18.785 0.000		15.936 0.000		27.972 0.000	

The values represent mean ± SE. Different letters within the same column indicate significant differences ($p < 0.05$) according to Duncan's multiple range test.

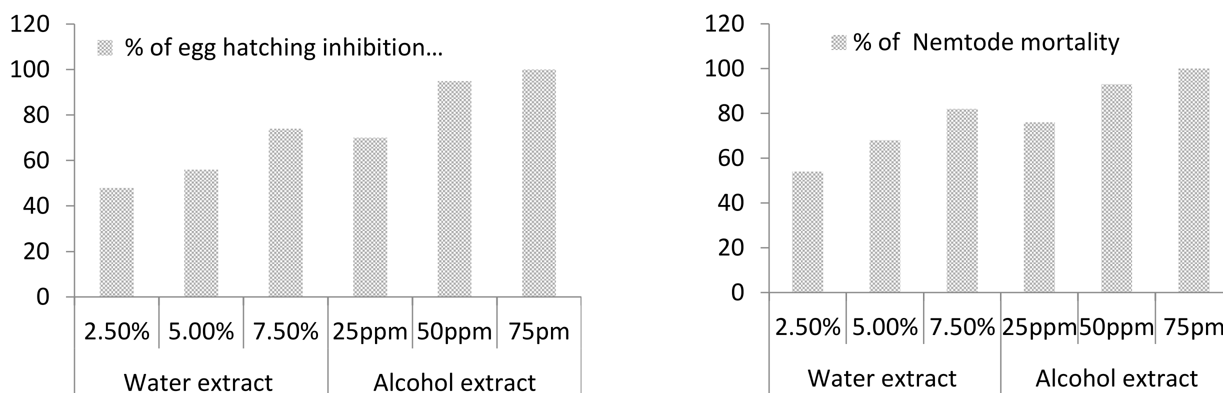

Fig. (1). Compression between water and alcohol extract of *Punica granatum* (peels) on mortality and egg hatching of *Meloidogyne incognita* in vitro.

The higher the dose achieved the highest the reduction in nematode number related to the maximum dose 30 g/kg which gave the best results. There was a clear decrease in nematode juveniles (J2) number per soil among the treatments, regardless, to concentration levels 30 g/kg. For instance, the highest concentration level of in dose 30 g/kg significantly reduced nematode juvenile's percentage (80.31%), followed by the bioproduct Micronema (69.00%), while the lowest reduction reached 61.13%, respectively in juveniles count and was obtained in concentration dose of 10 g/kg. A few galls increasingly decreased as concentration increased. The highest reduction (86.11%) resulted in concentration dose of 30 g/kg, while the lowest reduction was 73.61 resulted in concentration dose of 10 g/kg. As a result, the egg mass counts per plant for most treatments were much lower than those of the control. The greater percentage decreased in the generation of egg masses in concentration doses of 30 g/kg were 76.92%, followed by the bioproduct Micronema by 70.08%, respectively. The concentration level of 30 g/kg resulted in a more remarkable inhibition of egg production by 44.58 over the control (Table 3).

P. granatum treatments at several doses resulted in varying and proportional improvements in plant growth parameters, including shoot and root length and weight (Table 4). Increments for the dose from 10 to 30 g/kg produced the best results. The treatments had the greatest influence on plant height, root length, root weight, shoot weight, dry weight, and fruit weight. All treated plants had shoot weight values that were noticeably greater than the untreated control. When compared to the control, the fresh weight of infected plants treated with 30 g of *P. granatum* greatly were suppressed by 34.10%. However, plants treated with 10 g of *P. granatum* showed the lowest fresh shoot weight values, with the bioproduct Microne*ma* recording 8.74 and 8.82%, respectively. All treated plants had shoot weight values greater than the untreated control.

3. Pearson's Correlation Analysis

Pearson's correlation analysis revealed that the larvae and plant height, root length, root weight and fresh weight had a negative correlation with larvae (-0.1, -0.3, -0.2 and -.03) whereas galls was negatively correlated with root length and root weight (-0.5 and -0.1) as well as the egg masses was negatively correlated with plant height, root length, root weight and fresh weight (-0.2, -0,3, -0.2, -0.1), and eggs was negatively correlated with plant height, root length, root weight and fresh weight (-0.1, -0.3, -0.2 and -0, -.1).

Table (3). Effect of dried, ground powders of *Punica granatum* (peels) for management of *Meloidogyne incognita* in pepper plants under greenhouse conditions.

Treatment	No. of larvae in soil	Reduction %	No. of galls/ root system	Reduc-tion %	No. of egg masses/ root system	Reduction %	No. of eggs/root system	Reduction %
Control	965.00± 23.63[a]		288± 1.85[a]		117.00± 1.47[a]		157.00± 12.79[a]	
Micronema	299.00± 9.57[c]	69.00	50± 1.04[c]	82.63	35± 1.55[c]	70.08	98.00± 1.08[cd]	37.57
Punca 10	375.00± 9.57[b]	61.13	76± 0.96[b]	73.61	57.00± 0.95[b]	51.28	124.00± 2.02[b]	21.01
Punca 20	315.00± 9.57[c]	67.35	48± 0.85[c]	83.33	36.00± 1.22[c]	69.23	108.00± 1.44[bc]	31.31
Punca 30	190.00± 5.77[d]	80.31	40± 1.31[d]	86.11	27.00± 0.85[d]	76.92	87.00± 0.71[d]	44.58
F. value	544.08		7080.45		875.78		21.49	
***P*. value**	00.00		00.00		00.00		00.00	

The values represent mean ± SE. Different letters within the same column indicate significant differences ($p < 0.05$) according to Duncan's multiple range test.

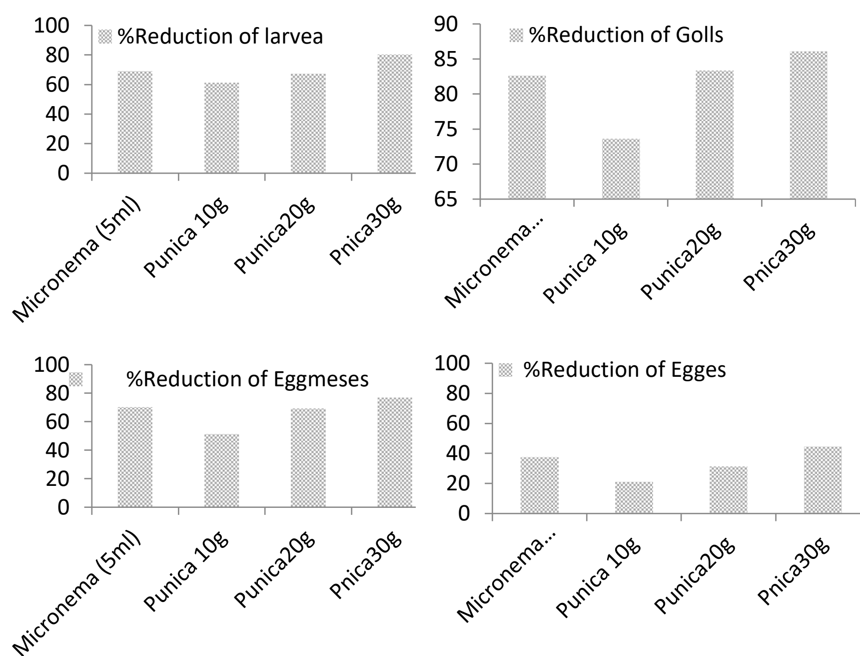

Fig. (2). Effect of dried peel powders of *Punic granatum* (conc./ pot) for management of *Meloidogyne incognita* in pepper plants under greenhouse conditions.

Table (4). Growth parameters of pepper plants infected with *Meloidogyne incognita* and treated with dried, ground powders *Punica granatum* (peels).

Treatment	Plant height (cm)	Increase %	Root length (cm)	Increase %	Root weight (g)	Increase %	Shoot fresh weight (g)	Increase %	Shoot dry weight (g)	Increase %	Fruit weight (g)	Increase %
Control	46.50±1.85[ab]	11.37	18.50± 0.96[b]	23.33	10.50± 0.92[bc]	16.27	15.00± 1.45[a]	28.53	2.18± 0.05[b]	45.33	61.20± 2.88[a]	73.96
Control with nematode	41.75±0.85[c]	----	15.00± 0.71[c]	---	9.03± 0.43[c]	---	11.67± 0.21[b]	---	1.50± 0.03[c]	---	35.18± 2.05[c]	---
Micronema	44.25±1.25b[c]	5.98	21.25± 0.85[ab]	41.66	11.34± 0.45[b]	25.58	12.70± 0.28[ab]	8.82	2.06± 0.34[b]	37.33	58.67± 1.36[ab]	66.77
Punca 10	41.85±0.48[c]	0.23	19.75± 0.85[ab]	31.66	9.89± 0.30[c]	9.52	12.69± 0.51[ab]	8.74	1.88± 0.09[b]	25.33	51.58± 3.81[b]	46.61
Punca 20	44.75±1.25[abc]	7.18	21.25± 0.75[ab]	41.66	10.59± 0.32[bc]	17.27	13.68± 0.61[ab]	18.76	2.17± 0.10[b]	44.66	57.67± 3.14[ab]	63.92
Punca 30	48.25±1.18[a]	15.56	22.25± 1.25[a]	48.33	13.39± 1.18[a]	48.28	15.65± 1.56[a]	34.10	2.80± 0.13[a]	86.66	62.70± 3.22[a]	78.22
F. value	4.89		8.36	---	5.89	---	2.61	---	10.39	---	12.74	---
P. value	0.00		0.00	-	0.00		0.00	--	0.00	--	0.00	--

The values represent mean ± SE. Different letters within the same column indicate significant differences ($p < 0.05$) according to Duncan's multiple range test.

DISCUSSION

Pomegranate peel extracts suppressed the activity of hatched J2 and the hatching of RKN eggs. Even at low doses, the pomegranate extract proved efficient, lowering egg hatch by up to 94%. Additionally, aqueous and ethanol extracts were nematotoxic and nematostatic to J2. at 7.5% and 75 ppm, respectively and greater concentrations, caused a complete death to J2 of *M. incognita*. Several authors have reported the effects of pomegranate extract on the root-knot nematode, including Korayem et al. (1993), Youssef et al. (2014) and El-Nagdi and Youssef (2015). Water extracts as direct treatment suppress the nematodes via the precursor allelopathic constituents and may be used as effective natural pesticides (Balah and AbdelRazek, 2020). The investigated bioactive components of *P. granatum* var. nana may work by directly causing toxicity, activating disease-resistant enzymes, or causing systemic acquired resistance to plant parasite nematodes (Ibrahim et al., 2006; Tehranifar et al., 2011 and Abd El-Salam et al., 2012). A few phytochemicals with nematoidal properties have also been documented in the literature including Calvet et al. (2001), Belal et al. (2007) and Shakil et al. (2008). The highly

exciting information gathered suggests that the bioactive components of *P. granatum* could be used as a safe alternative treatment for nematode and soil-borne fungus infections. The outcomes are in line with those found in different plant extracts by Hernandez et al. (1999), Osorio et al. (2010) and Abd El-Salam et al. (2012). Pomegranates can be consumed fresh or processed to make jams, jellies, fruit juices, and other goods. However, as the peels account for around half of the fruit's weight and are usually discarded as garbage with no value, eating this fruit generates a lot of waste. However, it has been observed that peels are a desirable natural antioxidant source since they have higher concentrations of bioactive compounds and even stronger biological activity than juice (Sood and Gupta, 2015). According to Zhu and Liu (2013), pomegranate peels are a good source of dietary antioxidants that may have antibacterial, anti-cancer, anti-obesity, anti-diabetic, anti-ulcerogenic, anti-hypertensive, and anti-mutagenic properties, among other potential therapeutic effects. *M. javanica* on tomatoes was controlled by adding crushed pomegranate peels to the soil (Ismail, 2015). Pomegranate fruit has been found to contain approximately 124 phytochemicals, with polyphenols being the main constituents (Viuda-Martos et al., 2010; Akhtar et al., 2015 and García-Villalba et al., 2015).

The husk and peel of pomegranates contain higher levels of polyphenols called ellagannins, which may contain physiologically active substances that could function as biological nematicides (Seeram et al., 2005; Akhtar et al., 2015 and García-Villalba et al., 2015). Pomegranate-derived gallic and ellagic acids demonstrated nematoidal activity against the animal-parasitic nematodes *Caenorhabditis elegans* and *Haemonchus contortus* (Ndjonka et al., 2013 and Mondal et al., 2015). *Rotylenchulus reniformis* and *M. incognita* were inhibited by triterpene saponins that were extracted from the fruit pericarp of *Sapindus mukorossi* (Saha et al., 2010). These results are consistent with earlier research that demonstrated that pomegranate fruit rind methanol extracts reduced the survivability of second-stage juveniles J2 of *M. incognita* in an in vitro experiment and impeded egg hatching (Meyer et al., 2016). The vitro experiment showed that the organic solvent extracts of all the plants under test were generally more efficient than the aqueous extracts at inhibiting nematode reproduction when compared to the control treatment (Abdel-Baset and Abdel-Monaim, 2020). Plants with high polyphenol content are generally resistant to a range of plant diseases, according to the in vitro experiment (Malik and Singh, 1980). Their function in enhancing plants' resistance to infectious diseases and pathogen growth may account for the elevated amount of total phenols. It was discovered that the harmful phenolic chemicals in plant cells work in two ways: first, by altering the structure of the link formed with plant tissue cell wall components (Mahadevant and Sridhar, 1986); and second by boosting host defensive mechanisms and increasing host resistance (Subba-Rao et al., 1988). According to the current study's findings, pomegranate fruit

pericarps and pericarps for nematode control not only offer a substitute for chemical nematodes but also aid in the disposal of agro-industrial waste. All treatments decreased all the nematode parameters evaluated. They had a positive effect on plants parameter and reduced population and damage of *M. incognita* infected plants and enhanced plant growth due to their fertilizing ability the pomegranate powder then provided a good substrate for growth (Ismail, 2015). Therefore, this may be dependent on the soil and other environmental factors. The results indicate that peel of pomegranates can be used as a fertilization and used as part of an integrated strategy for the control of *M. incognita.*

CONCLUSION

The goal of this study is to take advantage of pomegranate peel (*P. granatum*) waste as a source for bioactive constituents containing phenols, flavonoids, saponins, and anthocyanins. Additionally, the peel of *P. granatum* aqueous, and ethyl alcohol extracts as well as when used as powder for soil, it can suppress second-stage root-knot nematodes and prevent egg hatching. Whereas these effects increased gradually with increasing concentration, which increases pepper plant growth and yields under greenhouse conditions. These results are promising for use as natural nematicides that could potentially be integrated into sustainable nematode management strategies.

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تأثير مستخلص قشر الرمان ضد النيماتودا المتطفلة نباتيًا

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النيماتودا المتطفلة نباتيًا *Melodogine incognita* لها أهمية اقتصادية كبيرة حيث تؤدي إلى أضرار جسيمة للعديد من المحاصيل. لذلك استهدفت الدراسة الحالية تقييم إمكانات وكفاءة قشر الرمان (*Punica granatum*) من خلال المستخلصات المائية والإيثانولية ضد هذه النيماتودا. حيث تم تنفيذ التحليلات الكيميائية لمستخلص قشر فاكهة الرمان الإيثانولي لمحتويات الفينول والفلافونويد والسابونين والأنثوسيانين. بالإضافة إلى ذلك، تم فحص المستخلصات النباتية للتأكد من قابلية فقس البيض ومعدل موت يرقات العمر الثاني *M. incognita* في المختبر بتركيزات متفاوتة. أشارت النتائج إلى أن المستخلص الكحولي قتل يرقات نيماتودا تعقد الجذور وقلل من فقس البيض وكان أكثر فعالية من المستخلص المائي. أظهر مستخلص قشر الكحول الإيثيلي لـ *P. granatum* معدل وفيات واعد للغاية يتراوح بين ٧٦-١٠٠٪ بعد ٤٨ ساعة من التعرض، كما أنه منع فقس البيض بالكامل بعد ٤ أيام عند التركيز ٧٥٪. ومع زيادة تركيز المستخلص، زاد معدل يرقات العمر الثاني وانخفض الفقس تدريجيًا. تم اختبار مسحوق قشر فاكهة الرمان في الصوب الزجاجية أيضًا كإضافة للتربة المنزرعة بنباتات الفلفل بمعدلات ١٠، ٢٠، ٣٠ جرام/كجم للتربة من مسحوق قشور ثمار الرمان مقارنة بالنباتات غير المعاملة. وقد لوحظ أن هناك عددًا أقل من اليرقات العمر الثاني، والعقد الجذرية، وكتل البيض والبيض. وأظهرت النتائج زيادة نمو نباتات الفلفل معنويًا مقارنة بنباتات الكنترول الغير معاملة. تعتبر هذه النتائج واعدة وتشير لإمكانية تطوير عوامل إبادة طبيعية صديقة للبيئة وكما يحتمل دمجها في استراتيجيات الإدارة المستدامة للنيماتودا.