Growth and resistance response of eleven eggplant cultivars to infection by the Javanese root-knot nematode – *Meloidogyne javanica* under greenhouse conditions

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Citation: El-Qurashi M.A., Al-Yahya F.A., Almasrahi A.A., Shakeel A. (2025): Growth and resistance response of eleven eggplant cultivars to infection by the Javanese root-knot nematode – *Meloidogyne javanica* under greenhouse conditions. Plant Protect. Sci., 61: 291–300.

Abstract: In Saudi Arabia, root-knot nematodes (RKNs) were found to cause considerable damage to eggplant. These parasites cause significant death of seedlings during nursery production, with infected plants showing the symptoms of chlorosis and wilting, along with the characteristic root galls. Therefore, this work was carried out to find a resistant cultivar of eggplant against RKNs in Saudi Arabia by screening 11 locally available cultivars for two successive seasons. Following Koch's postulates for pathogenicity, RKNs were isolated from infected eggplant, and females were identified morphologically by perineal patterns as Meloidogyne javanica, which was distinguished by clear lateral fields on both sides. Identification was confirmed using two species-specific primers (SCAR), Fjav/Rjav and MjF/MjR, and visualized amplified fragments appeared at 670 bp and 517 bp, respectively. A greenhouse experiment was conducted to screen the cultivars, using five replicates for each cultivar and nematode inoculum (1 000 second-stage juveniles). In response to M. javanica, gall index (GI), egg mass index (EMI), and reproduction factors (RF) were calculated, and all the eggplant cultivars were categorized according to their resistance levels based on RF. Among the 11 eggplant cultivars, four were found resistant to M. javanica including Black Beauty (C5, Bursa Tohum), Melanzana Violetta Difirenze (C6, Zorzi), Melanzana Violetta Lung 2 (C7, Zorzi), and Long Purple (C9, Bursa Tohum) and Violetta Lung 3 (C8, Taj Agri) was found highly resistant. Moreover, two cultivars were found moderately resistant, two susceptible, and two susceptible to M. javanica infection. Therefore, this study provided valuable information to eggplant growers about the resistant cultivars in Saudi Arabia. However, the molecular mechanisms of this resistance need to be evaluated to find novel candidate genes for breeding and CRISPR-Cas9-based gene editing programs.

Keywords: Solanum melongena; susceptibility; reproductive factor, galling index; ecofriendly nematode control

The eggplant, *Solanum melongena* L., is a member of the Solanaceae family. It is typically cultivated in tropical and subtropical regions (Ainurrachmah et al. 2021). This specific vegetable crop

is commonly acknowledged as one of the top ten internationally (Shaaban et al. 2023). According to data from the TRIDGE SA website for 2022 (https://www.tridge.com/intelligences/eggplant/SA/

Supported by the researchers supporting Project No. RSPD2024R875, King Saud University, Riyadh, Saudi Arabia.

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production), the total output of eggplant in Saudi Arabia was 118 850 mt (metric tons) in 2022. During this period, the average price of eggplant was 1 480 USD/mt. According to Food and Agriculture Organization (FAO 2022) figures, the total production area in Saudi Arabia is 3 980 ha, with gross production valued at 174 900 USD. Because of their low-calorie content and high moisture levels, eggplant fruits are often known as diet foods (Kandoliya et al. 2015). Fruits stand out for having a high protein concentration, flavonoids, phenol, anthocyanine, ascorbic acid, and titratable acidity inside the peel. Moreover, eggplant is one of the vegetables with the highest antioxidant activity (Cao et al. 1996).

Eggplants have been demonstrated to be vulnerable to a variety of plant diseases. In Saudi Arabia, eggplant was found to be associated with a wide range of plant-parasitic nematodes (PPNs). The survey conducted by Al-Hazmi et al. (1983), Mokbel (2014), Almohithef et al. (2020), and Mohamed et al. (2023) reported that root-knot nematodes (RKNs) distributed with high density and frequency on eggplant in many geographical regions in Saudi Arabia. However, eggplant cultivars were susceptible to RKNs infections, resulting in severe damage and gradual economic losses (Anwar & McKenry 2010; Ullah et al. 2011; Abdel-Mageed et al. 2023; Shaaban et al. 2023). According to Ghosh (2021), RKNs exploit live cells as a source of nutrients and induce severe symptoms. The stunting of host plants' development and the yellowing and dwarfing of leaves were seen as a consequence of the RKNs infection. The fruits have a diminished size, while the overall output of production has declined. Galls have been shown to develop on the roots of plants (Lehman & Cochran 1991). Moreover, the occurrence of infection during the first phases of plant development has the potential to result in the mortality of seedlings when they are transplanted. The interior root tissues undergo histological modifications, producing giant cells at the feeding site that contribute to the formation of root galls (Di Vito et al. 2004).

Various approaches have been used for controlling RKNs (*Meloidogyne* spp.), including cultural practices, nematicide treatment, and biological procedures. Chemical treatment is the main strategy for managing diseases and pests in eggplant production fields (Sim et al. 2019). However, this practice led to the development of resis-

tance to pests. The use of nematicides also poses a threat to humans as a result of their lingering impact on plant parts that are ingested, as well as the need for a safety period before harvesting. These concerns have forced various countries and the European Union to sanction regulations against chemical pesticides, necessitating the introduction of novel and eco-friendly pest control methods. Therefore, resistant cultivars can be crucial for preventing or decreasing crop losses due to plant parasitic nematodes (Hill et al. 2012). In this context, the present study was conducted to evaluate the susceptibility of 10 eggplant cultivars distributed in Saudi Arabia and one obtained from Egypt, with prospects of identifying unknown resistance genes in eggplant for breeding and genetic engineering programs.

MATERIAL AND METHODS

Root-knot nematodes

Samples of eggplant roots infected with RKNs were collected from Riyadh, Saudi Arabia. Plant roots were washed by tap water and sterilized with 1.0% sodium hypochlorite solution for 1 min. Then, egg masses were picked up using antiseptic forceps and kept in watch glasses in sterilized distilled water. Under greenhouse conditions, a single egg mass was used to inoculate a pot containing a single 20-day-old eggplant cv. Roomy for producing a pure culture. After 45 days, the plants were carefully removed from the soil and examined for infection. The eggs were then extracted from infected plant roots using 0.5% sodium hypochlorite, as Hussey and Barker (1973) described, and utilized for inoculating other plants. The final densities of RKNS were deemed suitable for use as inoculum in subsequent experiments.

Root-knot nematodes identification

The identification was conducted using a pure culture of RKNs. Adult females were extracted from infected eggplant roots in the Nematology Lab. in the College of Agriculture and Food Science at King Saud University. The perineal patterns were prepared following a technique outlined by Hartman and Sasser (1985). Then, the perineal patterns were examined under a compound microscope (40× magnification), supplemented with a Nikon Digital Sight Ds-5M-L1 camera (Nikon Corpora-

tion, Japan) to capture images. The identification of root-knot nematode species' perineal patterns was achieved based on the description instructions provided by Eisenback et al. (1981) and Hunt and Handoo (2009).

Molecular identification was performed using two species-specific primers (SCAR) of M. javanica to confirm the morphological characteristics. DNA was extracted from two egg masses of two pure cultures according to methods outlined by Holterman et al. (2006). Fjav (5'-GGT-GCGCGATTGAACTGAGC-3'), Rjav (5'- CAG-GCCCTTCAGTGGAACTATAC-3'), and (5'- ACGCTAGAATTCGACCCTGG-3'), MjR (5'-GGTACCAGAAGCAGCCATGC-3') were used for identification (Zijlstra 2000; Meng et al. 2004). The PCR reaction was performed in volume 25 μL, containing 1 μL of DNA, 12.5 μL of 2× Green Master Mix (Thermo Fisher Scientific, United States), 1 μL of each primer, 9.5 µL of PCR water. The PCR program consisted of an initial denaturation at 94 °C for 5 min, followed by 35 cycles of denaturation at 94 °C for 45 s, annealing at 72 °C for 45 s, extension at 55 °C for 1 min, and then the final extension for 10 min. 1 µL of PCR products were loaded on a 0.5× TAE buffer agarose gel (1.5%) stained with 0.1 µL/mL acridine orange. After electrophoresis (100 V for 50 min), the gel was examined under UV trans-illuminator and photographed using the In-Genius LHR gel imaging and analysis system (Syngene, United Kingdom).

Eggplant cultivars sources

Eleven eggplant cultivars were acquired to assess their reactions to RKNs *in vivo*. Ten cultivars' seeds were procured from various marketplaces within the Riyadh region, Saudi Arabia, and one was obtained from Egypt (Table 1).

Greenhouse experiments

The seeds of each cultivar were sown in separate nursing trays under greenhouse conditions (30 ± 2 °C). After 25 days, the plant seedlings were transplanted to plastic pots (20 cm diam) and filled with steam-sterilized soil [sand and peat moss at a ratio of 2:1 (v/v)] after the emergence of actual leave appearance. Three seedlings were transplanted into each pot, and 20 days later, plants were thinned to a single plant per each.

Freshly hatched juveniles (J2s) of the *M. javanica* were used to inoculate plants (1 000 J2s/plant, ini-

Table 1. List of eggplant cultivars used in this study

Abbre- viation	Cultivar (company)	Country		
C1	Black King (Seeds Dallas)			
C2	Long Purple (Seeds Dallas)			
C3	Melanzana Black Beauty (Bursa Tohum)			
C4	Melanzana Violetta Lung 2 (Buona Luna)	Saudi Arabia		
C5	Black Beauty (Bursa Tohum)	Saudi Arabia		
C6	Melanzana Violetta Difirenze (Zorzi)			
C7	Melanzana Violetta Lung 2 (Zorzi)			
C8	Violetta Lung 3 (Taj Agri)			
C9	Long Purple (Bursa Tohum)			
C10	Roomy (Mecca Trade)	Egypt		
C11	Pompano Market (Sendian Al Arabia)	Saudi Arabia		

tial population - Pi). Each cultivar was replicated ten times. Five replicates were inoculated with J2s of M. javanica, while an additional five replicates served as control groups without nematodes. The experiment was repeated twice. The experiment was designed following a randomized complete block design (RCBD) on the bench. The plants were irrigated and fertilized as required. Ninety days after RKNs inoculation, flowers and fruit from each plant were counted. Subsequently, the plants were carefully uprooted and washed under tap water. Following this, both plant and nematode parameters were estimated. Shoot and root fresh weight and shoot length were measured. The plant shoots were then dried in an oven at 70 °C for 5 days and weighed. Finally, the reduction percent of plant parameters was computed relying on the untreated control plants following the formula: % reduction = $[(control - infected plant)/control] \times 100.$

The nematode reproduction was conducted by enumerating the number of galls per gram of roots. Also, one gram of roots was used for egg masses assessment after being stained with Phloxine-B, and egg masses were counted (Holbrook et al. 1983) and then computed for the root weight. Approximately 250 cm³ of soil samples were collected from each pot and sent to the nematology laboratory at the College of Food and Agriculture Science, King Saud University. Then, the J2s extraction was conducted using Cobb's wet-sieving and centrifugal sugar flotation procedure (Ayoub 1980), and the number of J2s was counted under

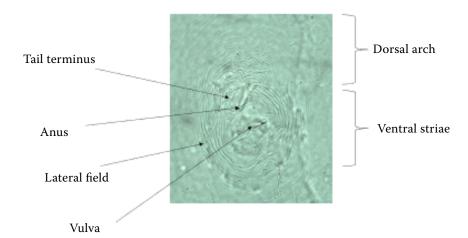


Figure 1 Depicts the perineal pattern of *Meloidogyne javanica*

a stereoscopic microscope at 40× magnify; the final J2s was calculated for whole soil size in the pot. Also, a random selection of five grams of roots was utilized for egg extraction with 1% sodium hypochlorite (Hussey & Barker 1973) and the number of eggs per root was calculated. Subsequently, the final population densities of nematodes (Pf) were determined by calculating both J2s recovered from the soil and eggs extracted from each plant's roots (Zhang & Schmitt 1994). Then, the nematode reproductive factor (RF) was estimated using the following formula: RF = Pf/Pi.

Gall index (Gi) and egg masses index (EMI) were evaluated using a scale from 0-5 where: 0: no galls or egg masses; 1: 1-20 galls or egg masses; 2: 21-100 galls or egg masses; 3: 101-300 galls or egg masses; 4: 301-1 000 galls or egg masses; 5: > 1000 galls or egg masses per root system (Hartman & Sasser 1985 with modification).

The RF averages were transformed to $\log_{10}(x+1)$, and then RF averages were submitted for analysis of variance and compared using the least significant difference (LSD) test at $P \le 0.05$. Finally, cultivars were classified as highly resistant (HR), resistant (R), moderately resistant (MR), susceptible (S) and highly susceptible (HS) according to statistical analysis (Mota et al. 2013).

Data Analysis

An analysis of variance (ANOVA) was conducted using the principles of a randomized complete block design (RCBD). Subsequently, multiple comparisons of means were performed using the least significant difference (LSD) method ($P \le 0.05$). The analysis was conducted using Analytical Statistix software (version 8.1).

RESULTS

Root-knot nematode identification

The perineal patterns of RKNs were related to the species of *Meloidogyne javanica*. The primary characteristic observed was the presence of well-defined lateral lines that divided the perineal pattern into dorsal and ventral sections. The dorsal arch was flattened or rounded (Figure 1). To confirm *M. javanica*, two species-specific primer pairs, Fjav/Rjav and MjF/MjR, gave PCR products at 670 bp and 517 bp, respectively (Figure 2).

Estimation of nematode reproduction

Under greenhouse conditions, eleven eggplant cultivars were evaluated for their response to RKN *M. javanica* infection. Data in Table 2 revealed that all eleven eggplant cultivars were infected with *M. javanica*



Figure 2. The amplification product (517 and 670 bp) generated with the *M. javanica* species-specific Mj-F/Mj-R and F-jav/R-jav SCAR primers

L-100 bp DNA ladder; lane 1, 2 – the two isolates with Mj-F/Mj-R; lane 3, 4 – the two isolates with F-jav/R-jav; lane -ve – water as a negative control (PCR reaction without DNA)

Table 2. Disease indices in terms of egg masses, eggs, galls and J2s of *Meloidogyne javanica* on different cultivars of eggplant

Cultivar	No. of egg masses per root		No. of eggs per root		No. of galls per root			No. of J2s per pot				
	S1	S2	Mean**	S1	S2	Mean	S1	S2	Mean	S1	S2	Mean
C1	597 ^c *	611 ^c	604 ^c	21 668 ^b	22 840 ^b	22 388 ^b	538 ^{cd}	566 ^{cd}	552°	7 488°	7 520°	7 504°
C2	573°	635°	$604^{\rm c}$	$21\ 668^{\mathrm{bc}}$	$20~954^{bc}$	$21\ 311^{\rm b}$	491^{cd}	$516^{\rm cd}$	$503^{\rm cd}$	8 946 ^b	8 176 ^{bc}	8 561 ^b
C3	$1810^{\rm b}$	791°	$1\ 301^{\rm b}$	46 086 ^a	25 961 ^b	36 023 ^a	$1714^{\rm b}$	$750^{\rm c}$	$1\ 232^{\rm b}$	5 920 ^d	$5888^{ m d}$	$5904^{\rm c}$
C4	$349^{\rm c}$	362^{c}	$356^{\rm cd}$	$20~839^{bc}$	$22\ 352^{\rm b}$	$21\ 596^{b}$	326^{cd}	$352^{\rm cd}$	$339^{\rm cde}$	19 000a	18 672 ^a	18 836 ^a
C5	$84^{\rm c}$	79^{c}	$81^{\rm cd}$	$6\ 104^{\rm fg}$	6586^{efg}	6345^{de}	$60^{\rm d}$	60 ^d	60^{de}	$1~846^{\rm efg}$	$1~846^{efg}$	$1\ 846^{\rm f}$
C6	71 ^c	76°	$73^{\rm cd}$	$4439^{\rm fg}$	$4.952^{\rm fg}$	4695^{de}	$51^{\rm d}$	55 ^d	53^{de}	$1~640^{fgh}$	$1636^{\rm fgh}$	$1638^{\rm f}$
C7	$405^{\rm c}$	$472^{\rm c}$	$439^{\rm cd}$	$6\ 166^{efg}$	$5~039^{fg}$	$5\ 602^{\mathrm{de}}$	379^{cd}	$408^{\rm cd}$	$394^{\rm cde}$	$1~880^{\rm efg}$	1 917 ^{efg}	$1898^{\rm f}$
C8	$24^{\rm c}$	$52^{\rm c}$	$38^{\rm d}$	$4385^{\rm fg}$	$2701^{\rm g}$	3 543 ^e	$18^{\rm d}$	28^{d}	23 ^e	$1446^{\rm gh}$	$1434^{\rm gh}$	$1\ 440^{\rm fg}$
C9	2 762 ^a	3 149ª	2 957ª	$4361^{\rm fg}$	$7\ 444^{\rm efg}$	5 903 ^{de}	$2\ 183^{\rm b}$	2 916 ^a	2 550 ^a	840 ^h	902 ^h	871 ^g
C10	492^{c}	532^{c}	512^{cd}	13 272 ^{de}	14 755 ^{cd}	$14\ 014^{\rm c}$	392^{cd}	420^{cd}	$406^{\rm cde}$	2 000 ^{efg}	1 958 ^{efg}	1 979 ^{ef}
C11	$330^{\rm c}$	419^{c}	$374^{\rm cd}$	9281^{defg}	$9914^{\rm def}$	$9598^{\rm cd}$	279^{cd}	$334^{\rm cd}$	$307^{\rm cde}$	$2~644^{\rm e}$	$2\ 397^{ef}$	2 520 ^e
Mean	682ª	653ª	_	14 413ª	13 045ª	_	585 ^a	582ª	_	4 877ª	4 758ª	_
LSD	$C \times S = 781$		C 550	$C \times S = 7 \ 114$		C 5 021	$C \times S = 665$		C = 470	C × S = 816		C 577
value***	S = 235		C = 552	S = 2	C=5 031		S = 201			S =	246	C = 577

^{*}means followed by the same letter in both 1st and 2nd season are not significantly different

at different levels. The results provided information on the number of galls per root, number of juveniles per pot, no. of egg masses per root, and no. of eggs per root. There were notable variations among the evaluating cultivars regarding the no. of galls, with statistical significance ($P \le 0.05$). Cultivar C9 had the greatest infectivity, with an average of 2 550 galls per plant root, followed by C3 with an average of 1 232. On the other hand, the C8 cultivar had the lowest average of galls per plant root (23), and the C6 and C5 cultivars had an average of 53 and 60, respectively. For the comparison between the two seasons, overall, there were no significant differences between the averages of both seasons at $P \le 0.05$. Also, eggplant cultivars' response among both seasons was similar in the no. of galls except C3 and C9, which exhibited significant differences.

Regarding the number of J2s per plant, overall, there is a significant difference between the average of cultivars ($P \le 0.05$). C4 recorded the highest number of J2s (18 836), followed by C2 (8 561), C1 (7 504) and C3 (5 904). While C9 exhibited the lowest number of J2s with an average of 871, followed by C8 (1 440) with a non-significant difference.

In addition, there are non-significant differences between the average of 1^{st} and 2^{nd} seasons and among the means of each cultivar during both seasons.

The number of egg masses per root was counted for each plant. Generally, over the average of cultivars, C9 exhibited a high number with 2 957 egg masses/plant root, followed by C3 (1 301). At the same time, C8 was determined as the lowest cultivar in no. of egg masses (38). There are non-significant differences among the rest of the cultivars at $P \le 0.05$. Non-significant differences have been observed between the average of both seasons and the means of each cultivar in the 1st and 2nd seasons, except C3.

The RKN eggs were extracted from 5 g roots for each plant by sodium hypochlorite (1%). Then, the number of eggs for each plant was determined after examination under the compound microscope at 10x magnification. Data in Table 2 revealed that there is a significant difference ($P \le 0.05$) over the average of cultivars. However, C3 was the greatest in no. of eggs (36 023), followed by C1 (22 388), C2 (21 311), and C4 (21 596), while C8 was the lowest with an average of 3 543, followed by C7 (5 602), C9 (5 903), and C5 (6 345). On the other hand, non-significant differences were found

^{**}average of cultivars, the mean followed by the same letter for each column are not significantly different from each other at $P \le 0.05$

^{***}LSD value, S is used to compare the average of 1^{st} and 2^{nd} seasons; $C \times S$ is used to compare the mean of interaction between the season and cultivars; C is used to compare the average of cultivars

between the average of both seasons and the means of each cultivar in 1st and 2nd seasons except C3.

Evaluation of plant growth for finding resistant cultivar

The reduction percent of plant biomass in terms of plant height, shoot and root fresh weight, and shoot dry weight were determined for plants at 90 days after inoculation for two successive seasons (Table 3). Significant differences ($P \le 0.05$) were found between the average eggplant cultivars regarding root fresh weight. The most reduction was found in C11 (53.7%), followed by C5 (44.7%) and C7 (35.1%). At the same time, C3 recorded the lowest reduction percent (-14.5), followed by C1 (0.2). No significant differences were found between the average of both seasons or between the means of each cultivar in the 1st and $2^{\rm nd}$ seasons.

In the case of shoot dry weight, C11 had the highest reduction percent (23.5%), while C3 had the lowest, with an average of -14.3 %. However, non-significant differences were recorded between the average of both seasons and among 1^{st} and 2^{nd} seasons for each cultivar means at $P \le 0.05$.

C6 and C11 were the cultivars that exhibited the most reduction in shoot fresh weight, with an average of 34.6% and 34.2%, respectively, while C3 was the lowest, with an average of -22.9%. For the comparison between the 1st and 2nd seasons, no significant differences were found among the average of both seasons and each cultivar means among the two seasons as well ($P \le 0.05$).

C11 recorded the highest reduction percent in plant height, with an average of 33%, followed by C5, C6, C7, and C8, with an average of 27.7%, 29%, 20%, and 29.2%, respectively. On the other hand, no significant differences ($P \le 0.05$) were found between the average of each season and the mean of each cultivar during 1^{st} and 2^{nd} seasons.

Estimation of reduction percent in plant production. The reduction percent in plant fruiting and flowering for each treated plant was calculated and compared to the control (Table 4). No significant differences were found between the average of eggplant cultivars except C3, which recorded the lowest percent with an average of -72 in plant flowering ($P \le 0.05$). The means of reduction percent in plant flowering for each cultivar did not vary in the 1^{st} and

Table 3. Pathogenic effects of Meloidogyne javanica on different eggplant cultivars

Treat- ments	Reduction in root fresh weight (%)			Reduction in shoot dry weight (%)		Reduction in shoot fresh weight (%)			Reduction (%) in plant height			
	1 st	2 nd	Mean**	1 st	2 nd	Mean	1 st	2 nd	Mean	1 st	2 nd	Mean
C1	0.5 ^{d-g} *	-0.04 ^{efg}	0.2 ^{ef}	9.3 ^{a–e}	9.3 ^{a-e}	9.3 ^{bc}	16.2 ^{abc}	2.3 ^{bc}	9.2 ^b	-1.2 ^{fgh}	3.5 ^{efg}	1.1 ^{de}
C2	10.9^{c-f}	12.4^{c-f}	11.6 ^{de}	5.2^{a-e}	5.5 ^{a-e}	5.4^{bc}	13.6^{abc}	19.6 ^{abc}	16.6 ^{ab}	-12.7^{gh}	$3.1^{\rm efg}$	-4.8^{ef}
C3	-21.2^{g}	-7.8^{fg}	-14.5^{f}	-5.8^{ef}	-22.8^{f}	-14.3^{d}	$-8.9^{\rm cd}$	-36.8 ^d	-22.9^{c}	-11.6gh	-23.7^{h}	-17.7^{f}
C4	14.2^{b-f}	14.6^{b-f}	14.4^{cde}	8.1^{a-e}	8.1^{a-e}	8.1^{bc}	19.2^{abc}	25.1^{ab}	22.1^{ab}	4.6^{d-g}	9.6 ^{b-g}	$7.1^{\rm cde}$
C5	44.9^{ab}	$44.5^{\rm ab}$	44.7^{ab}	18.5^{ab}	2.6^{b-e}	10.6^{ab}	21.1^{abc}	22.7^{abc}	21.9^{ab}	26.8^{a-d}	$28.5^{ m abc}$	27.7^{ab}
C6	32.2^{a-d}	32.2^{a-d}	32.2^{a-d}	13.2 ^{a-d}	13.2^{a-d}	13.2^{ab}	36.2^{a}	32.9^{ab}	34.6 ^a	33.4^{a}	24.6^{a-e}	29^{ab}
C7	28.8^{a-e}	$41.3^{ m abc}$	35.1^{abc}	12.0^{a-e}	15.6 ^{a-d}	13.8^{ab}	8.9^{abc}	26.7^{ab}	17.8^{ab}	15.6^{a-f}	24.5^{a-e}	20^{abc}
C8	11.6^{c-f}	13.7^{b-f}	$12.7^{\rm cde}$	19.1^{ab}	$17.4^{ m abc}$	18.3^{ab}	31.4^{ab}	31.4^{a}	31.4^{ab}	30.0^{ab}	$28.3^{ m abc}$	29.2^{ab}
C9	17.2^{b-f}	39.0^{abc}	28.1^{bcd}	9.2^{a-e}	22.1^{ab}	15.6 ^{ab}	16.5^{abc}	30.1^{ab}	23.3^{ab}	14.7^{a-f}	14.5^{a-f}	14.6^{bcd}
C10	14.9^{b-f}	10.0^{c-g}	12.5 ^{de}	$-0.8^{\rm cde}$	-1.7^{de}	-1.3 ^{cd}	17.9^{abc}	28.7^{ab}	23.3^{ab}	5.9 ^{c-g}	15.3^{a-f}	$10.6^{\rm cde}$
C11	52.8 ^a	54.6 ^a	53.7 ^a	23.5^{a}	23.5^{a}	23.5 ^a	32.9^{ab}	35.5 ^{ab}	34.2^a	30.6 ^{ab}	35.3^{a}	33.0^{a}
Mean	18.8ª	23.1ª	_	10.1 ^a	8.5 ^a	_	18.6ª	19.8ª	_	12.4ª	14.9 ^a	_
LSD value***	S = 9.6		C = 22.5	S = 8.7		C = 13.3	S = 10.2		C = 23.9	S = 6.9		C 16.1
	$S \times C = 31.8$		C = 22.5	$S \times C = 18.9$			$S \times C = 33.8$		C = 23.9	$S^* \times C$	= 22.7	C = 16.1

^{*}means followed by the same letter in both 1st and 2nd season are not significantly different

^{**}average of cultivars, the mean followed by the same letter for each column are not significantly different from each other at $P \le 0.05$

^{***}LSD value, S is used to compare the average of 1st and 2nd seasons; $C \times S$ is used to compare the mean of interaction between the seasons and cultivars; C is used to compare the average of cultivars

Table 4. Reduction percentage in eggplant cultivars production (fruiting and flowering) due to the *Meloidogyne javanica* infection

						-	
Treat- ment		Reductio		Reduction			
	in f	lowering	(%)	in fruits (%)			
ment	1 st *	$2^{\rm nd}$	mean**	1^{st}	2 nd	mean	
C1	25 ^a	14^a	19 ^a	-73^{b}	-73^{b}	-73°	
C2	77 ^a	73^{a}	75 ^a	30^{a}	43 ^a	37^{ab}	
C3	11 ^a	-155^{b}	-72^{b}	40^{a}	20^{a}	30^{ab}	
C4	50 ^a	50 ^a	50 ^a	30^{a}	60 ^a	45^{ab}	
C5	73 ^a	73^{a}	73 ^a	60 ^a	80 ^a	70 ^a	
C6	67 ^a	70^{a}	68 ^a	0^{ab}	60 ^a	30^{ab}	
C7	20^{a}	40 ^a	30^{a}	35 ^a	55 ^a	45^{ab}	
C8	0^a	0^a	0^{ab}	0^{ab}	0^{ab}	$0_{\rm p}$	
C9	-10^{a}	30^{a}	10 ^a	20^{a}	60 ^a	40^{ab}	
C10	20^{a}	24^{a}	22^a	50 ^a	66 ^a	58 ^a	
C11	65 ^a	65 ^a	65 ^a	40 ^a	80 ^a	60 ^a	
Mean	36 ^a	25 ^a	_	21 ^a	41 ^a	_	
LSD	S =	35	C 00	S =	C 55		
value***	$C \times S = 116$		C = 82	$C \times S$	C = 57		

^{*}means followed by the same letter in both 1st and 2nd season are not significantly different

***LSD value, S is used to compare the average of 1^{st} and 2^{nd} seasons; $C \times S$ is used to compare the mean of interaction between the season and cultivars; C is used to compare the average of cultivars

 2^{nd} seasons; C3 showed a high variation among the mean in the 1^{st} and 2^{nd} seasons, with 11% and 155%, respectively. No significant differences were observed between the average of both seasons.

Regarding the reduction percent of plant fruiting, C5, C10, and C11 showed the most reduction, averaging 70%, 58%, and 60%, respectively. At the same time, C1 had the lowest reduction (averaging -73%), followed by C8 (0.0%). Non-significant differences were recorded between the means of each cultivar during $1^{\rm st}$ and $2^{\rm nd}$ seasons and among the averages of $1^{\rm st}$ and $2^{\rm nd}$ seasons ($P \le 0.05$).

Category eggplant cultivars

For the galling index (GI) (Table 5), C8 was the lowest (averaging 1.6), followed by C5 and C6 (2.0 for each). On the other hand, C9 had the highest galling index, followed by C3, C10, C1, and C2, with averages of 4.4, 4.0, 3.7, and 3.7, respectively.

Table 5. Estimation of gall index (GI), egg masses index (EMI) reproduction factor (RF) of *Meloidogyne javanica* on eleven eggplant cultivars

Cultivars	GIª	EMIª	RF*	Category**
C1	3.7	4.0	1.48^{b}	S
C2	3.7	3.7	1.49^{b}	S
C3	4.4	4.5	1.60^{a}	HS
C4	3.6	3.6	1.62 ^a	HS
C5	2.0	2.3	0.91^{d}	R
C6	2.0	2.2	0.86 ^{de}	R-HR
C7	3.6	3.6	0.91 ^d	R
C8	1.6	1.7	0.77^{e}	HR
C9	4.9	5.0	0.86 ^{de}	R-HR
C10	4.0	3.9	$1.21^{\rm c}$	MR
C11	3.5	3.6	1.11 ^c	MR

average value of GI or EMI. 0 = no galls or egg masses, 1: 1–20 galls or egg masses, 2: 21–100 galls or egg masses, 3: 101–300 galls or egg masses, 4: 301–1 000 galls or egg masses, 5: > 1 000 galls or egg masses (Hartman & Sasser 1985 with modification).

*average values transformed in $\log_{10}(x+1)$. Means followed by different letters are significantly different ($P \le 0.05$) according to the LSD test (LSD value = 0.1)

**Hs – highly susceptible, S – susceptible, MR – moderately resistant, R – resistant, HR – highly resistant

The egg masses index (EMI) was assessed for each cultivar (Table 5). Cultivars were variable in EMI, where cultivar C8 was the lowest with an average of 1.7, followed by C6 and C5 with an average of 2.2 and 2.3, respectively. On the other hand, C9 was the highest, with an average of 5.0, followed by C3 and C1, averaging 4.5 and 4.0, respectively.

For two seasons, eleven eggplant cultivars were evaluated in a greenhouse to determine their response to M. javanica infection based on the reproductive factor (RF) significantly ($P \leq 0.05$). As shown in Table 5, cultivar C8 showed a high resistance to infestation with non-significant differences with C6 and C9. cultivars C5, and C7 were resistant. High susceptibility was observed in C3 and C4, while C1 and C2 were susceptible. On the other hand, C10 and C11 were moderately resistant.

DISCUSSION

Eggplant is recognized as one of the most important vegetable crops globally, growing in tropi-

^{**}average of cultivars, the mean followed by the same letter in the column are not significantly different from each other at $P \le 0.05$

cal and subtropical regions like Saudi Arabia. Its fruits, as indicated by Cao et al. (1996), are characterized by a high concentration of nutritional components, notably protein, flavonoids, phenols, anthocyanins, ascorbic acid, and antioxidants. In Saudi Arabia, eggplants were cultivated in greenhouses and open fields. Plant-parasitic nematodes, particularly the root-knot nematode, significantly negatively impact the yield and quality of eggplant fruits (El-Qurashi et al. 2023). The worldwide crop yield losses from RKNs infection were estimated at 30 bil. USD (Eisenback et al. 1981). In Saudi Arabia, RKNs infect eggplants with high density and frequency (Al-Hazmi et al. 1983; Mokbel 2014; Almohithef et al. 2020; Mohamed et al. 2023). In our study, RKNs were isolated from eggplant and identified using the perineal pattern and molecular methods with two SCAR primers. The result showed that *M. javanica* was the species infecting eggplant. These results were comparable to El-Qurashi et al. (2017) and Eisenback et al. (1981). To choose management strategies, it is essential to identify rootknot nematodes (El-Qurashi et al. 2017) accurately. According to ElNesr et al. (2010), Saudi Arabia is one of the nations situated in tropical and subtropical climates that has an annual temperature range of 27–43 °C, and *M. javanica* (tropical nematode) is mostly found in the warm and temperate regions (Moens et al. 2009). Moreover, M. javanica was discovered to be a high species that was widely distributed over Saudi Arabian crops and vegetables, according to Al-Hazmi et al. (1983).

Eleven eggplant cultivars were obtained from Saudi and Egyptian markets and were evaluated for their response against M. javanica infection under controlled greenhouse conditions for two trials. Screening these cultivars for susceptibility is particularly interesting in detecting the resistant cultivars available and overcoming using nematicides to control RKNs. Resistance refers to the plant's capacity to inhibit the development and/or reproduction of RKNs. However, A susceptible plant provides a condition in which rootknot nematodes may multiply without any obstacle (Hussey & Janssen 2002). On the other hand, tolerance refers to the capacity of plants to endure infection of RKNs without showing any reduction in plant production.

The plant growth response to *M. javanica* exhibited varies across all eggplant cultivars. In general, cultivar C3 was not affected by infection and showed

no reduction in average plant growth measurement (root fresh weight, shoot dry and fresh weight, and plant height). On the other hand, C11 had the highest average plant reduction percentage. For plant productivity, C3 and C1 had the lowest percentages of reduction in the average productivity of flowering and fruit, respectively. According to Begum et al. (2014), the degree of damage is thus determined by the host plant's response (tolerant or sensitive).

In our study, Violetta Lung 3 C8 was highly resistant, followed by Melanzana Violetta Difirenze C6 and Long Purple C9. Moreover, Black Beauty C5 and Melanzana Violetta Lung 2 C7 were resistant to *M. javanica* infection. In 1991, Lehman and Cochran reported the eggplant cultivar Black Beauty exhibited a high level of resistance against *M. javanica* and a moderate level of resistance against *M. incognita*. Akhter and Khan (2018) and Ghosh (2021) reported that the Black Beauty cultivar was highly susceptible to infection with RKNs.

The reproduction rate was used to describe the ultimate population of nematodes. A high initial inoculum rate of *Meloidogyne* spp. inhibited and decreased the development of eggplant (Bakr et al. 2022). Conversely, the greatest initial inoculum level may lead to a decline in the final population because of competition among the nematodes for food (Shaaban et al. 2023).

Plant resistance may manifest in several ways, such as the inability of juveniles to enter the roots of resistant cultivars due to physical barriers. Additionally, the phenomenon being discussed may pertain to the response of roots in terms of their ability to either attract or repel juveniles. Occasionally, infective individuals invade the root system and then abandon it. The variation in eggplant cultivars' reactions to M. javanica infection may be attributed to genetic diversity, resulting in the synthesis of several compounds crucial in regulating the pathogen (Ullah et al. 2011; Devi & Sumita 2015). The cultivars that exhibit tolerance may lack the capacity to generate nematode-feeding sites in plants after invasion due to hypersensitive reactions triggered by resistant genes, ultimately failing nematode development. According to Colak-Ates et al. (2018), if the host does not establish a feeding site, the RKNs cannot absorb nutritional components, resulting in decreased development and reproduction. The tolerance demonstrated by eggplant cultivars may be attributed to post-infection resistance occurring before nematode penetration of the roots, which is believed to be facilitated by toxic compounds (Tan-

imola et al. 2015). Chemicals are essential for plants to be resistant to RKNs infection. Pegard et al. (2005) proposed that phenolic chemicals, particularly chlorogenic acid, cause host plant resistance. The most vulnerable and resistant tomato cultivars were used in an HPLC examination, and the results showed that the total phenolic compounds were more highly concentrated in the resistant cultivars than in the susceptible ones (Shaaban et al. 2023).

CONCLUSION

We conclude that cultivating eggplant cultivars resistant to *M. javanica* infection is an alternative method to overcome using nematicides and losses caused by RKNs infestation. Violetta Lung 3 C8 was highly resistant to *M. javanica* infestation. Moreover, Black Beauty C5, Melanzana Violetta Difirenze C6, Melanzana Violetta Lung 2 C7, and Long Purple C9 were resistant to infestation with *M. javanica* so they should be cultivated alternatively to other cultivars. In future, the resistance genes in these cultivars need to be identified for eggplant breeding and genetic engineering programs towards RKNs resistance.

Funding acknowledgement statement:

The authors extend their appreciation to the Researchers Supporting Project No. RSPD2024R875, King Saud University, Riyadh, Saudi Arabia.

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Received: September 27, 2024 Accepted: March 25, 2025 Published online: April 22, 2025