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Identification of Root-knot Nematode (*Meloidogyne* spp. Goeldi, 1887) (Tylenchida: Meloidogynidae) Species in Celery (*Apium graveolens* L.) (Apiaceae) Growing Areas of Çanakkale Province

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ABSTRACT
In this study, the species and extensiveness of root-knot nematodes (*Meloidogyne* spp. Goeldi, 1887) (Tylenchida: Meloidogynidae) causing yield loss in the celery (*Apium graveolens* L.) (Apiaceae) production areas of Çanakkale were determined. For this purpose, celery plant roots and soil samples were taken by making non-periodical surveys of a total of 75 different celery growing areas in Çanakkale province and districts in 2020-2021. Females of root-knot nematodes and second-stage juveniles (J2s) from egg masses were obtained from celery samples brought to the laboratory. Morphological identification of root-knot nematode species was done by making sections obtained from perineal regions of female individuals and morphometric measurements were performed from J2s for each population. For molecular identification, DNA was then extracted from samples and analyzed by species-specific primers referring to the most common *Meloidogyne* spp. *Meloidogyne javanica* (Treub, 1885) Chitwood,1949 in 9 samples, and *M. arenaria* (Neal, 1889) in 5 samples were determined based on morphologic -morphometric and molecular methods. The result of the study indicated that the rate of root-knot nematode infestation in celery cultivation areas in Çanakkale was 18,6%. The infestation rates of *M. javanica* and *M. arenaria* determined in celery growing areas in the province were 12% and 6.6%, respectively. In this study, *M. javanica* and *M. arenaria* species were detected for the first time in the celery fields of Çanakkale province.

Plant Protection

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Keywords
Celery
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Çanakkale İli Kereviz (*Apium graveolens* L.) Yetiştirilen Alanlardaki Kök-ur Nematod (*Meloidogyne* spp. Goeldi, 1887) (Tylenchida: Meloidogynidae) Türlerinin Tanılanması

ABSTRACT
Bu çalışmada Çanakkale ili kereviz (*Apium graveolens* L.) (Apiaceae) üretim alanlarında verim kaybına neden olan kök-ur nematodlarının (*Meloidogyne* spp. Goeldi, 1887) (Tylenchida: Meloidogynidae) türleri ve yaygınlıkları belirlenmiştir. Bu amaçla, 2020-2021 yıllarında Çanakkale il ve ilçelerinde bulunan toplam 75 farklı kereviz üretim alanlarına periyodik olmayan arazi çıkışları yapılarak kereviz bitki kök ve toprak örnekleri alınmıştır. Laboratuvara getirilen kereviz örneklerinden kök-ur nematodlarının dişi bireyleri ve yumurta paketlerinden ikinci dönem juveniller (J2s) elde edilmiştir. Kök-ur nematodlarının morfolojik tür teşhisleri her popülasyon için dişi bireylerin perineal bölgelerinden elde edilen kesitler ve ikinci dönem larvaların morfometrik ölçümleri yapılarak belirlenmiştir. Daha sonra moleküler tanılama için örneklerden DNA elde edilmiştir ve en yaygın *Meloidogyne* spp. türlerine istinaden türe özgü primerler ile analiz edilmiştir. Yapılan morfolojik ve moleküler tanılama çalışmalarına göre 9 örnekte *Meloidogyne javanica* (Treub, 1885) Chitwood, 1949 türü, 5 örnekte ise *M. arenaria* (Neal, 1889) Chitwood, 1949 türü belirlenmiştir. Çalışma sonucunda Çanakkale ili kereviz yetiştiriciliği yapılan alanlarda kök-ur nematodu bulaşıklık oranı

Bitki Koruma

Araştırma Makalesi

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Yaygınlık
Kök-ur nematodu

%18,6 olarak belirlenmiştir. İlde kereviz yetiştirilen alanlarda belirlenen *M. javanica* ve *M. arenaria*'nın bulaşıklık oranı sırası ile %12 ve %6,6 olarak tespit edilmiştir. Bu çalışma ile Çanakkale ili kereviz alanlarında *M. javanica* ve *M. arenaria* türleri ilk defa tespit edilmiştir.

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INTRODUCTION

The main winter vegetables grown in Çanakkale province are cabbage (*Brassica oleraceae* L. var. *capital*), lettuce (*Lactuca sativa* L.), cauliflower (*Brassica oleraceae* L. var. *botrytis*), spinach (*Spinacia oleracea* L.), celery (*Apium graveolens* L.) and leek (*Allium porrum* L.). In 2020, 2.750.000 tonnes of winter vegetables were grown in 1,700.000 decares cultivation areas of Türkiye. Among the winter vegetables with the highest production rate, celery belonging to the Apiaceae family, comes to the forefront with 230 tonnes in Çanakkale (TUIK, 2020).

It has been reported that the celery plant is known as a host of many plant-parasitic nematodes, such as *Pratylenchus penetrans* Cobb, 1917 (Townsend & Wolynetz, 1991); *P. hamatus* Thorne & Allen (Townshend, 1962); *Hemicycliophora Arenaria* Raski, 1958 (Franklin & Stone, 1974); *Longidorus plus* Lamberti & Zacheo, 1977 (Bleve-Zacheo et al., 1979; Wyss, 1980); *Cactodera cacti* Filipjev & Schuurmans Stekhoven, 1941 (Esser, 1992); *Paratylenchus* sp. Micoletzky, 1922, (Lownsbery et al., 1952); *Nacobbus aberrans* (Thorne) Thorne & Allen,1944 (Doucet, 1999); *Ditylenchus dipsaci* Kühn, 1857 (Di Benedetto, 2005) and *H. poranga* Monteiro & Lordello, 1978 (Emilse et al., 2011). Root-knot nematodes (RKNs) have a wide host range, and it was reported that they feed on 5500 different plant varieties, including vegetables, fruit trees, ornamental plants, weeds, and medicinal plants (Trudgill & Block, 2001; Karssen et al., 2013; Ataş et al., 2021). Hitherto 105 root-knot nematodes species (*Meloidogyne* spp. Goeldi, 1887) (Tylenchida: Meloidogynidae) were identified all over the world (Ghaderi & Karssen, 2020; Maleita et al., 2021), and the most common species are *Meloidogyne incognita* (Kofoid & White, 1919), *M. javanica* (Treub, 1885) Chitwood, 1949, *M. arenaria* (Neal, 1889) Chitwood, 1949, *M. Chitwood* (Golden et al., 1980), *M. fallax* (Karssen, 1996), and *M. hapla* Chitwood, 1949 (Adam et al., 2007). In the studies conducted in Türkiye, 10 root-knot nematodes species were detected; *M. incognita*, *M. javanica*, *M. arenaria*, *M. hapla*, *M. Chitwood*, *M. Thames*, *M. Martinelli*, *M. exiqua*, *M. luci* and *M. graminis* species (Yüksel, 1974; Elekçioğlu & Uygun, 1994; Elekçioğlu et al., 1994; Mennan & Ecevit, 1996; Söğüt & Elekçioğlu, 2000; Devran & Söğüt 2009; Özarslandan et al., 2009; Akyazı & Ecevit, 2010; Özarslandan & Elekçioğlu, 2010; İmren et al., 2014; Kepenekçi et al., 2014; Çetintaş & Çakmak, 2016; Devran et al., 2017; Aydınlı, 2018; Uysal et al., 2023).

Meloidogyne species cause significant yield losses and more serious damage to celery compared to other plant-parasitic nematodes. Some studies indicated that the most dominant root-knot species infesting celery are *M. incognita*, *M. incognita* race-1, *M. javanica*, *M. hapla*, and *M. arenaria* in the world (Incer & Lopez, 1979; Doucet, 1999; Chaves, 2002; Vovlas et al., 2008; Malakeberhan et al., 2012). There is also, a study on the presence of root-knot nematodes in celery growing areas of Türkiye. *Meloidogyne incognita* and *M. arenaria* were reported on the celery plant in the Black Sea Region of Türkiye (Yüksel 1974). This present study aimed to determine root-knot nematode species collected from Çanakkale celery cultivation areas by using morphometric-morphologic and molecular methods.

MATERIAL and METHOD

Survey

Overall, 75 root samples were collected from celery cultivated fields of Çanakkale in 2020 and 2021. The sampling locations of the celery cultivation areas of Çanakkale are given in Figure 1. Celery plants infested with root-knot nematodes in an ice box were brought to the laboratory. In both years, adult females, egg masses, and second-stage juveniles (J2s) of root-knot nematodes were extracted from samples.

Culture of the root-knot nematodes

To obtain pure cultures, egg masses on infected celery roots were collected by using a small needle. Each root-knot nematode isolate was cultured from a single egg mass taken from galled celery roots and multiplied on the susceptible tomato cv. Çanakkale F1 in a growth chamber at 25±1 □C and 65% RH with a 16:8 L:D photoperiod.

Morphologic-morphometric identification

Perineal patterns: Adult root-knot nematode females were dissected from the roots of the celery plants with a needle and scalpel under the binocular microscope.

Perineal patterns of the extracted females were cut in 45% lactic acid and their preparations were made in glycerin (Hooper, 1986). Morphological identification of Meloidogyne species was made according to Jepson (1987) and Karssen (2002).

Figure 1. Sampling locations of the celery cultivation areas in Çanakkale.
Şekil 1. Çanakkale kereviz ekim alanlarındaki örnekleme noktaları.

Morphometric characters of second-stage juveniles: J2s hatched from the egg masses multiplied as pure cultures were fixed in TAF fixative and permanent preparations were done according to Seinhorst's (1959) method. Measurements of approximately 25 J2s were made according to Karssen (2002) under the Leica DM1000 stereomicroscope.

Molecular identification

DNA isolation

DNA was extracted from pure cultures of J2s by using the High Pure PCR Template Preparation Kit (Roche Diagnostics, Germany) according to the manufacturer's guidelines.

PCR analyses

The PCR reactions were conducted on the SimpliAmp™ Thermal Cycler (Applied Biosystems, CA, USA) using the reaction conditions in a total volume of 25 µ L: 20 ng of DNA, 2 mM MgCl2, 2.5 µ L 10X PCR buffer, 200 µ M dNTPs, 0.4 µ M of each primer, 1 U Taq DNA Polymerase (ABM), and molecular double distilled water. To screen samples; Inc-K14F/Inc-K14R primers (Randig et al., 2002) and MincF1/MincR1 (Devran et al., 2018) for *M. incognita*; Far/Rar primers (Zijlstra et al., 2000) for *M. arenaria*; Fjav/Rjav primers (Zijlstra et al., 2000) for *M. javanica* and JMV primers for *M. hapla* (Wishart et al., 2002) were used in the PCR reactions. Then, PCR products were electrophoresed on a 1.5% agarose gel in 1X TAE and visualized under UV light with Xpert Green DNA Stain by using the Gel iX Imager (Intas Science, Germany).

RESULTS and DISCUSSION

Morphologic-morphometric identification

Root samples taken from celery cultivation areas revealed that there was an 18.6% prevalence of root-knot nematodes in Çanakkale. In other words, root-knot nematode was detected in 14 of 75 celery samples collected, and galled symptoms of plant roots were shown in Figure 2.

Figure 2. Celery plant roots infested with root-knot nematode.
Şekil 2. Kök-ur nematodları ile infekteli kereviz kökleri.

From a total of 14 root-knot nematode populations, 9 samples were identified as *M. javanica*, while 5 samples were identified as *M. arenaria* by perineal pattern and morphometric measurement of J2s. Locations and coordinates of root-knot nematode species in infested areas recorded with Global Positioning System (GPS) were given in Table 1 and Figure 3.

Meloidogyne javanica

Similar to the findings obtained in this study, distinct lateral ridges that divide the dorsal and ventral striae, and lateral lines extended on both sides of the tail terminus are typically as clear as the perineal patterns of *M. javanica (Figure 4).*

Table 1. Location, Root-knot nematodes species, and coordinates of celery samples collected from Çanakkale
Tablo 1. Çanakkale'den toplanan kereviz örneklerine ait konum Kök-ur nematodu türleri ve koordinatları

Sample No	Location	Species*	Latitude (N)	Longitude (E)
1	Kepez	*Mj*	40° 5′ 31"	26° 23′ 2"
3	Kepez	*Mj*	40° 5′ 41"	26° 22′ 8"
7	Kepez	*Mj*	40° 5′ 21"	26° 22′ 42 "
9	Saraycık	*Mj*	40° 8′ 17 "	26° 28′ 6 "
12	Çıplak	*Ma*	39° 57′ 31 "	26° 16′ 16 "
23	Halileli	*Ma*	39° 58′ 13 "	26° 16′ 55 "
27	Umurbey	*Ma*	40° 14′ 30 "	26° 38′ 26 "
30	Çardak	*Ma*	40° 24′ 40 "	26° 45′ 17 "
44	Halileli	*Mj*	39° 57′ 47"	26° 17′ 59"
62	Umurbey	*Mj*	40° 14′ 27"	26° 39′ 35"
63	Umurbey	*Mj*	40° 14′ 41"	26° 39′ 14"
65	Biga[a]	*Mj*	40° 17′ 10"	27° 17′ 7"
69	Biga[a]	*Mj*	40° 14′ 50"	27° 12′ 34"
72	Eceabat[a]	*Ma*	40° 15′ 44"	26° 23′ 24"

*Ma: *Meloidogyne arenaria*, Mj: *Meloidogyne javanica*, a: Distinct of Çanakkale

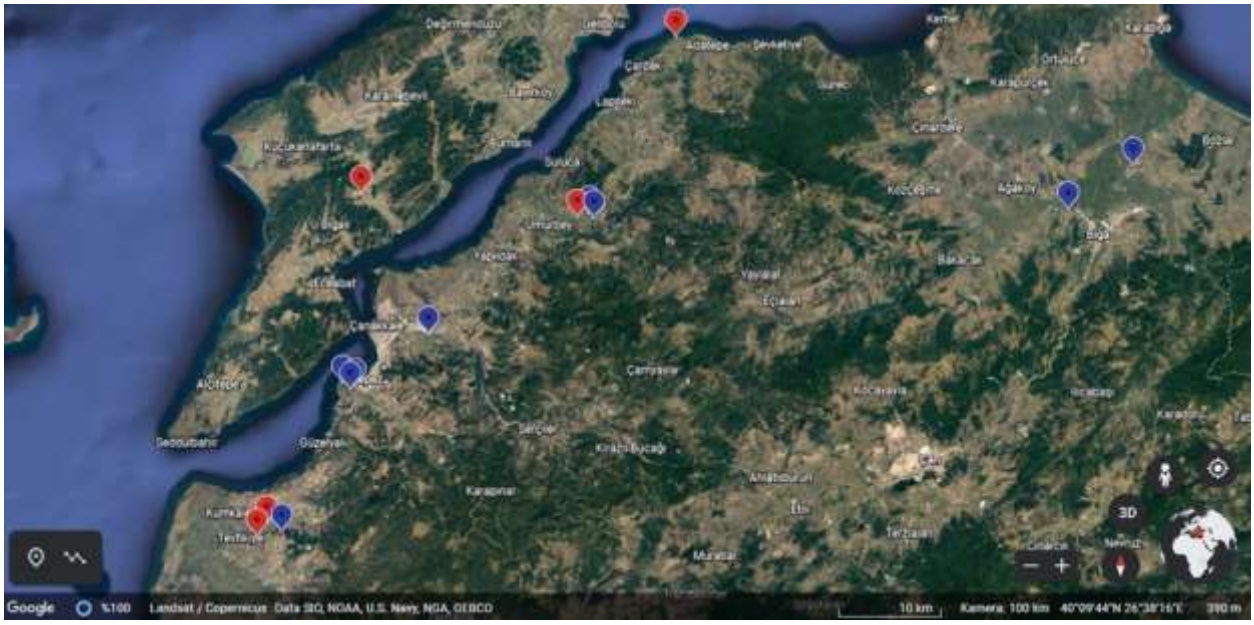

Figure 3. Locations of Root-knot nematode populations collected from celery growing areas in Çanakkale.
Şekil 3. Çanakkale kereviz alanlarından toplanan Kök-ur nematodu popülasyonlarının lokasyonları.
(Places where *Meloidogyne arenaria* was identified on celery- red color)
(Places where *Meloidogyne javanica* was identified on celery- blue color)

In the study, stylet lengths in all populations were long, and their DGO was relatively shorter than those reported by Whitehead (1968). Conversely, the ratios of body length to body width (a) in all populations were relatively short. All other lengths aligned with the findings of Whitehead (1968) (Table 2).

Meloidogyne arenaria

These patterns showed a typically rounded to flattened low dorsal arch near the lateral field with irregular forks (Figure 5).

The body length of all populations of *M. arenaria* was shorter than those reported by Whitehead (1968). The style length and the ratio of body length to tail length (c) of all populations were relatively long according to the Whitehead (1968) (Table 3).

Molecular identification

Molecular identification of a total of 14 DNA samples was done by using four different species-specific primer sets. *Meloidogyne arenaria-specific* Far/Rar and *M. javanica-specific* Fjav/Rjav primers were produced in expected amplicons of approximately 420 bp and 670 bp, respectively (Figure 6, 7).

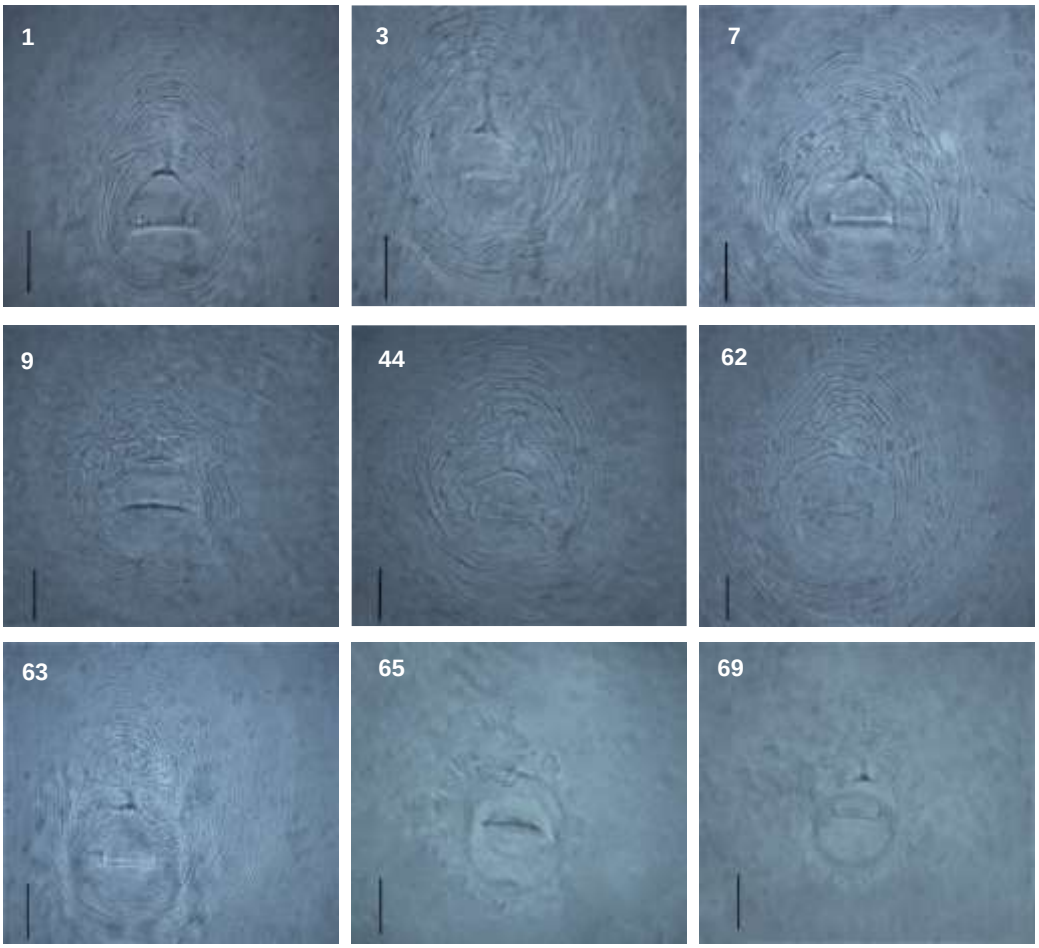

Figure 4. Perineal patterns of *Meloidogyne javanica* isolates collected from the celery fields of Çanakkale. Scale bar: 20 µm.
Şekil 4. Çanakkale'de kereviz alanlarından toplanan Meloidogyne javanica izolatlarının perineal bölge kesitleri. Ölçek çubuğu 20 µm.

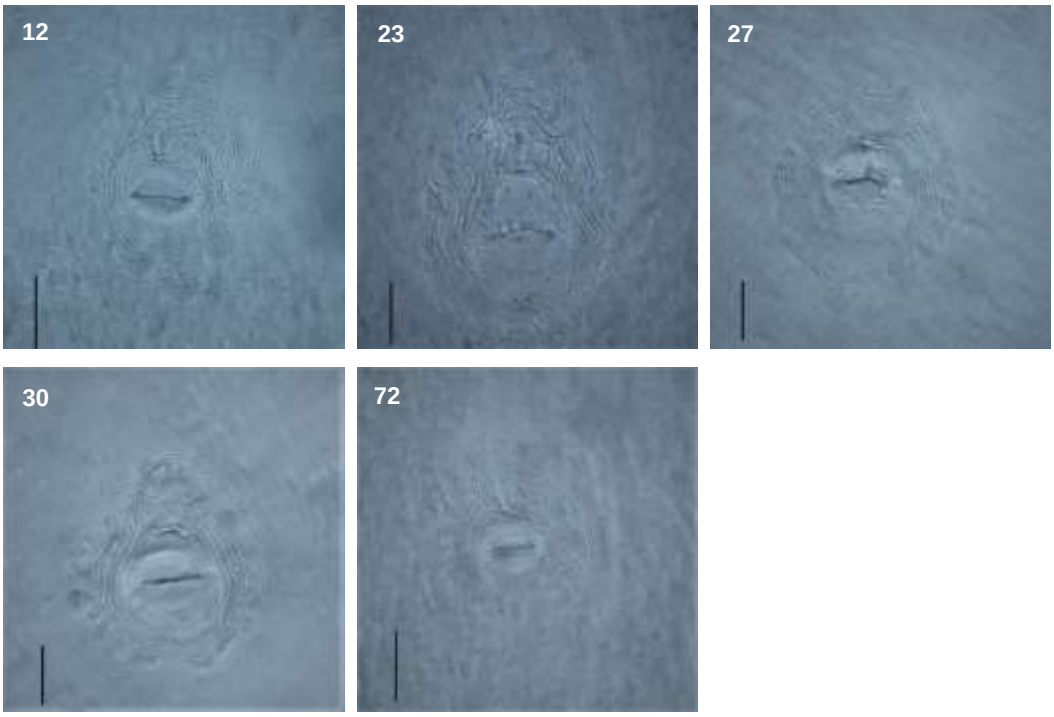

Figure 5. Perineal patterns of *Meloidogyne arenaria* isolates collected from the celery fields of Çanakkale. Scale bar: 20 µm.
Şekil 5. Çanakkale'de kereviz alanlarından toplanan Meloidogyne arenaria izolatlarının perineal bölge kesitleri. Ölçek çubuğu 20 µm.

Table 2. Diagnostic characters of second stage juveniles (J2) of *Meloidogyne javanica* on celery
Tablo 2. Kereviz üzerindeki Meloidogyne javanica'nın ikinci dönem juvenillerinin (J2s) teşhis karakterleri

Diagnostic characters	1	3	7	9	44	Whitehead (1968)
Body length	411.9±17.8 (385.1-445.7)	395.1±14.8 (371.7-419.9)	404.3±12.4 (380.6-431.3)	402.0±14.1 (378.8-429.8)	414.9±15.3 (380.2-449.7)	387-459
Body width	14.7±0.6 (13.3-15.8)	14.9±0.5 (14.1-16.0)	15.4±0.4 (14.7-16.2)	15.7±0.5 (14.6-16.6)	15.7±0.6 (14.8-16.9)	
Body width at stylet base	8.5±0.6 (7.0-9.8)	8.6±0.5 (7.9-9.8)	8.6±0.4 (7.0-10.0)	8.8±0.4 (7.9-10.8)	8.7±0.5 (7.8-10.0)	
Body width at the anus	9.2±0.6 (8.2-10.4)	9.7±0.6 (8.9-11.0)	9.8±0.6 (8.8-11.0)	9.8±0.7 (8.7-11.7)	9.3±0.7 (7.9-10.8)	
Stylet length	13.0±0.7 (11.6-14.2)	12.6±0.5 (10.7-12.8)	11.7±0.7 (10.0-13.6)	12.6±0.7 (11.1-13.6)	12.7±1.0 (10.8-14.1)	9.4-11.4
DGO	3.6±0.3 (2.7-4.1)	3.3±0.4 (2.3-4.0)	3.3±0.4 (2.4-4.0)	3.3±0.4 (2.5-4.0)	3.4±0.5 (2.5-4.1)	4
Tail length	50.3±1.8 (47.9-55.9)	47.8±2.0 (44.4-51.0)	50.9±3.2 (45.9-58.0)	50.0±2.3 (45.6-55.0)	50.7±2.8 (45.4-55.5)	36-56
Excretory pore to head end	79.6±3.3 (74.4-86.2)	77.2±3.9 (67.7-87.7)	76.1±4.2 (68.7-84.1)	81.4±3.9 (74.4-88.2)	80.1±4.8 (70.5-88.3)	
Body width at the excretory pore	11.6±0.7 (10.4-13.0)	11.8±0.5 (10.7-12.8)	11.0±1.1 (9.3-13.2)	11.2±0.9 (9.2-13.7)	11.9±2.8 (45.4-55.5)	
a	26.9±0.7 (26.5-29.2)	26.4±1.2 (24.9-29.6)	26.1±0.8 (24.6-27.9)	25.5±0.8 (24.0-27.0)	26.4±0.9 (24.6-27.8)	27.1-35.9
b	4.3±0.2 (3.9-4.8)	4.0±0.2 (3.7-4.5)	4.4±0.2 (4.0-4.8)	4.4±0.3 (3.8-5.0)	4.5±0.2 (4.0-5.2)	
c	8.1±0.4 (7.4-9.1)	8.2±0.5 (7.3-9.3)	7.9±0.6 (6.6-8.9)	8.0±0.4 (7.2-8.8)	8.2±0.6 (6.8-9.2)	7.3-11.1
c'	5.4±0.3 (4.8-6.2)	4.9±0.3 (4.3-5.4)	5.1±0.4 (4.4-6.05)	5.1±0.4 (4.2-5.7)	5.4±0.4 (4.9-6.4)	

Diagnostic characters	62	63	65	69	Whitehead (1968)
Body length	410.8±17.2 (378.0-440.8)	400.5±13.3 (375.7-426.8)	399.7±13.4 (375.6-426.7)	402.9±11.6 (377.6-421.6)	387-459
Body width	15.6±0.6 (14.5-16.9)	15.5±0.5 (14.4-16.8)	15.3±0.6 (14.1-16.8)	15.5±0.5 (14.5-16.5)	
Body width at stylet base	9.1±0.9 (7.4-11.0)	8.7±0.6 (7.6-10.0)	8.8±0.6 (7.4-10.0)	8.5±0.5 (7.6-9.6)	
Body width at the anus	9.3±0.7 (8.1-10.8)	9.5±0.7 (8.2-10.8)	9.7±0.8 (8.2-11.7)	9.7±0.7 (8.4-11.2)	
Stylet length	11.2±0.7 (10.7-11.6)	11.7±1.0 (10.4-11.8)	11.1±0.7 (10.6-11.4)	11.6±0.6 (10.9-11.9)	9.4-11.4
DGO	3.4±0.4 (2.6-4.1)	3.3±0.4 (2.6-4.1)	3.1±0.4 (2.4-4.0)	3.2±0.4 (2.4-4.0)	4
Tail length	52.1±3.7 (43.7-61.0)	50.3±3.0 (44.6-56.1)	50.9±2.7 (45.9-56.4)	49.6±2.9 (44.5-55.8)	36-56
Excretory pore to head end	80.3±3.2 (75.6-86.9)	73.5±5.0 (64.3-86.9)	79.0±3.6 (70.5-85.1)	74.9±5.8 (64.1-85.8)	
Body width at the excretory pore	12.2±0.9 (10.7-13.6)	12.0±0.8 (10.8-13.9)	12.0±0.8 (10.7-13.2)	11.9±0.5 (10.9-13.0)	
a	26.2±0.9 (24.3-27.7)	25.7±1.0 (24.2-27.9)	26.0±0.8 (25.0-27.6)	26.0±0.9 (24.3-27.9)	27.1-35.9
b	4.5±0.3 (3.9-5.3)	4.6±0.3 (3.9-5.5)	4.3±0.3 (3.9-5.1)	4.6±0.4 (3.7-5.4)	
c	7.9±0.6 (6.8-9.5)	7.9±0.5 (6.9-9.2)	7.8±0.4 (6.9-8.7)	8.1±0.4 (7.3-8.9)	7.3-11.1
c'	5.5±0.4 (4.5-6.4)	5.2±0.3 (4.6-5.9)	5.2±0.4 (4.4-6.1)	5.1±0.4 (4.4-6.0)	

Note: Note: All measurements are in µm Body length/Body width, b: Body length/ Intestine to the head end, c: Body length/Tail length, c': Tail length/Body width at the anus) and in the form: mean ± s.d. (range) n: 25

Table 3. Diagnostic characters of second-stage juveniles (J2) of *Meloidogyne arenaria* on celery
Tablo 3. Kereviz üzerindeki Meloidogyne arenaria'nın ikinci dönem juvenillerinin (J2) teşhis karakterleri

Diagnostic characters	12	23	27	30	72	Whitehead (1968)
Body length	449.5±16.3 (423.8-454.7)	442.6±9.8 (431.7-452.9)	441.2±17.0 (433.7-452.8)	442.5±14.2 (429.5-450.7)	448.9±12.0 (437.3-453.5)	450-490
Body width	15.8±0.4 (14.7-16.9)	15.7±0.5 (14.9-17.0)	15.7±0.6 (14.1-17.0)	15.0±0.6 (13.7-16.0)	15.6±0.4 (14.8-16.4)	
Body width at stylet base	8.5±0.6 (7.7-9.7)	8.8±0.5 (8.0-10.0)	8.8±0.7 (7.6-10.0)	9.1±0.8 (7.5-10.9)	8.7±0.5 (8.0-9.6)	
Body width at the anus	10.1±0.7 (8.7-11.1)	9.8±0.7 (8.5-11.2)	10.4±0.5 (9.0-11.7)	10.0±0.7 (8.5-11.1)	9.8±0.5 (8.5-10.9)	
Stylet length	10.5±0.7 (10.1-12.0)	11.2±1.0 (10.8.12.7)	11.2±0.6 (10.8-12.2)	11.0±1.0 (10.3-12.0)	11.0±0.7 (10.4-12.4)	10
DGO	3.1±0.3 (2.5-4.0)	3.1±0.4 (2.1-4.0)	3.2±0.4 (2.4-4.0)	3.2±0.4 (2.5-4.0)	3.1±0.4 (2.3-3.8)	3
Tail length	48.6±3.3 (44.3-58.5)	49.1±2.9 (42.9-54.3)	49.0±2.5 (44.7-53.7)	50.1±2.7 (45.6-55.3)	49.6±2.4 (45.6-54.5)	
Excretory pore to head end	78.0±4.9 (65.1-96.1)	77.6±4.7 (67.6-85.7)	77.8±6.5 (67.5-89.8)	76.9±4.4 (69.6-83.7)	78.5±3.7 (67.9-85.2)	
Body width at the excretory pore	12.2±0.7 (10.9-13.1)	11.6±0.9 (9.2-13.1)	12.7±0.7 (11.0-12.8)	11.5±0.8 (9.8-13.6)	12.2±0.6 (10.5-13.7)	
a	25.8±0.7 (24.6-27.4)	24.9±0.8 (23.6-26.8)	26.1±0.8 (24.5-27.6)	25.4±0.7 (24.2-26.6)	26.0±0.8 (24.4-27.3)	26-32
b	4.4±0.3 (3.8-5.1)	4.1±0.3 (3.8-4.9)	4.4±0.3 (3.9-5.0)	4.1±0.2 (3.7-4.7)	4.5±0.2 (4.1-5.0)	
c	7.6±0.5 (7.1-8.4)	7.8±0.5 (7.1-8.4)	7.7±0.1 (6.6-8.2)	7.6±0.4 (6.7-8.8)	7.9±0.4 (7.3-8.4)	6-7.5
c'	4.8±0.4 (4.2-5.9)	5.0±0.4 (4.2-6.1)	4.0±0.4 (4.0-5.5)	5.0±0.5 (4.1-6.3)	5.0±0.4 (4.5-6.1)	

Note: Note: All measurements are in µm Body length/Body width, b: Body length/ Intestine to the head end, c: Body length/Tail length, c': Tail length/Body width at the anus) and in the form: mean ± s.d. (range) n: 25

However, both *M. incognita* primer sets and JMV primers did not give any DNA bants in analyzed samples. As a result of PCR studies, *M. arenaria* and *M. javanica* were found in 5 and 9 samples, respectively. These results show that this is the first report of root-knot nematode infecting celery in Çanakkale of Türkiye.

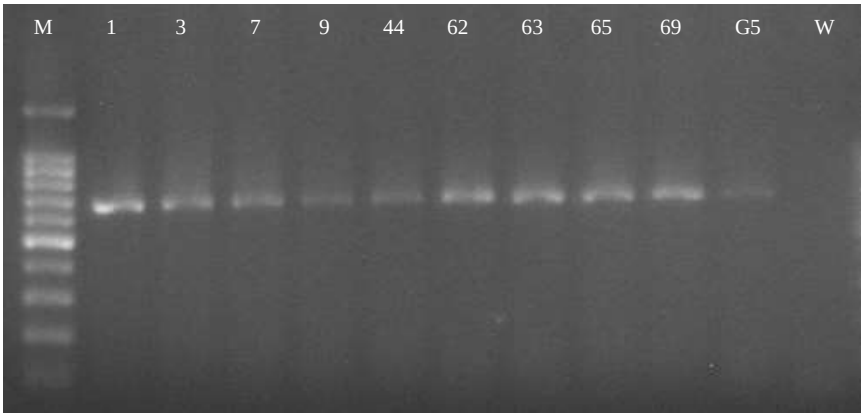

Figure 6. PCR products of amplified DNA using *Meloidogyne javanica*-specific primer Fjav/Rjav, M: 100 bpDNA Ladder (Hibrigen); Samples:1, 3, 7, 9, 44, 63, 65 and 69; G5: *M. javanica* (positive control); W: Water.
Şekil 6. Meloidogyne javanica'ya özgü primer Fjav/Rjav primer kullanılarak coğaltılmış DNA ya ait PCR ürünleri, M: 100 bpDNA Ladder (Hibrigen); Örnekler:1, 3, 7, 9, 44, 63, 65, ve 69; G5: M. javanica (pozitif kontrol); W: Su.

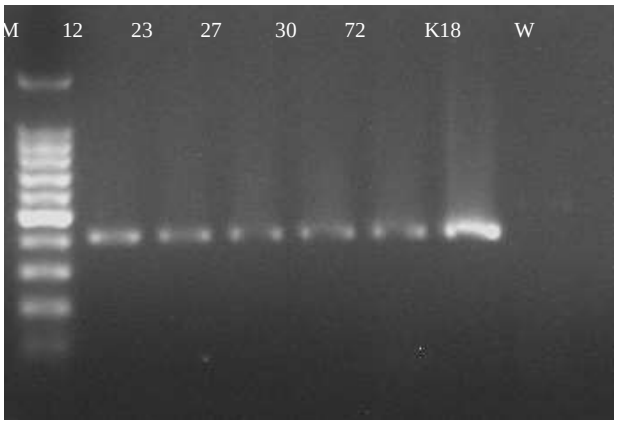

Figure 7. PCR products of amplified DNA of using *Meloidogyne arenaria*-specific primer Far/Rar, M: 100 bp DNA Ladder (Hibrigen); Samples: 12, 23, 27, 30 and72; K18: *M. arenaria* (positive control); W: Water.

Şekil 7. Meloidogyne arenaria'ya özgü primer Fjav/Rjav primeri kullanılarak coğaltılmış DNA'ya ait PCR ürünleri, M: 100 bp DNA Ladder (Hibrigen); Örnekler: 12-72r; K18: M. arenaria (pozitif kontrol); W: Su.

Morphologic-morphometric and molecular data were evaluated together. In the provincial center, 46 samples were taken from celery growing areas and it was determined that 5 of them were infected with *M. javanica* and 2 of them were infected with *M. arenaria*. Ten samples were taken from the fields in Lapseki and they were infected with *M. javanica* (2) and *M. arenaria* (2) respectively. Twelve samples were taken from the fields in Biga and 2 of them were found to be infected with *M. javanica*. Five samples were taken from the fields in Eceabat and one of them was found to be infected with *M. arenaria*. Single samples taken from each of Ezine and Bayramiç district's cultivation areas were not infected with RKNs. It has been determined that 12% and 6.6% of the celery-growing areas of Çanakkale are infested with *M. javanica* and *M. arenaria*, respectively (Table 4).

As an important edible vegetable, celery plants ought to be protected from soil-borne pests including plant parasitic nematodes. Especially RKNs have a wide host range, and their complete control is quite costly and difficult. Root-knot nematode infestation has been reported in many other important plant cultivars with edible tubers, such as carrot (Singh, 2009; Evlice et al., 2020), potato (Özarslandan et al., 2009; Maleita et al., 2018), sugar beet (Yu, 1995; Maareg et al., 1998), sweet potato (Rutter et al., 2019; Yigezu, 2021). Accurate identification is of great importance in the control of RKNs. Therefore, control methods such as the use of resistant cultivars and crop rotation can significantly reduce the population density of the pest.

Table 4. Rates of finding of root-knot nematode species in Çanakkale provincial center and districts

Tablo 4. Çanakkale il merkezi ve ilçelerinde kök-ur nematodu türlerinin bulunma oranları

Districts	Number of Samples	Number of Infected Samples and Finding Rate			
		Meloidogyne javanica	Finding Rate (%)	*Meloidogyne arenaria*	Finding Rate (%)
Center	46.0	5.0	10.9	2.0	4.4
Ezine	1.0	0.0	0.0	0.0	0.0
Bayramiç	1.0	0.0	0.0	0.0	0.0
Lapseki	10.0	2.0	20.0	2.0	20.0
Biga	12.0	2.0	16.6	0.0	0.0
Eceabat	5.0	0.0	0.0	1.0	20.0
Total	75.0	9.0	12.0	5.0	6.6

In the present study, in total, 14 of 75 celery samples (18,6%) collected from celery cultivation areas in the Çanakkale were found to be infested with RKNs. It was determined that *M. javanica* is the most dominant species with a 12% infestation rate in the region. In the international *Meloidogyne* project in Malawi, Saka (1981) reported the presence of *M. javanica* on celery. *Meloidogyne javanica* has been found also in different vegetable-growing areas of Türkiye, (Devran & Söğüt, 2009; Aydınlı & Mennan, 2016; Devran et al., 2017; Uysal et al., 2017; Gürkan et al., 2019; Aslan & Elekçioğlu, 2022).

In the present study, the infestation rate of *M. arenaria* in celery cultivation areas was found to be 6.6%. *Meloidogyne arenaria* was identified in some districts. Celery cultivation areas in Halileli and Umurbey regions were found to be infested with both RKN species. In Türkiye, the first and only study on the presence of RKNs in celery cultivation areas in the Black Sea Region was conducted and *M. arenaria* and *M. incognita* species were reported (Yüksel, 1974). Previous studies also reported that *M. arenaria* has been found on cultivated plants (Devran & Söğüt, 2009; Aydınlı & Mennan, 2016; Devran et al., 2017; Uysal et al., 2017; Gürkan et al., 2019; Aslan & Elekçioğlu, 2022). However, it was reported that *Meloidogyne hapla* is the most prevalent RKN species

in celery cultivation areas in some parts of the world (Anita, 2012; Malakeberhan & Wei, 2012). In this study, *M. hapla* was not detected in the survey areas.

In conclusion, the survey was carried out for the identification of RKNs in celery-growing areas of Çanakkale. *Meloidogyne javanica* and *M. arenaria* were identified on celery in the region. This study is the first report of *M. javanica* in celery cultivation areas in Türkiye.

Author's Contributions

The contribution of the authors is equal.

Statement of Conflict of Interest

The authors declare no conflict of interest.

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