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First Record of *Meloidogyne javanica* (Treub, 1885) (Chitwood, 1949) on Lettuce (*Lactuca sativa* L.) Growing Areas in the Southern Marmara Region

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ABSTRACT
Root-knot nematodes are the most important plant parasitic nematode group due to their wide host range and difficulties in control and cause significant quality and yield losses in vegetables including lettuce plants. Root-knot nematodes are causing symptoms such as yellowing, wilting, and stunting on lettuce plants. For monitoring lettuce fields for detecting nematode-infected areas, surveys were made to the lettuce growing areas of Çanakkale province in the winter months of 2021-2022 and the plants infected with root-knot nematodes were examined and pure nematode cultures were obtained in the laboratory. A total of 16 plant and soil samples were taken and 3 of them were found to be infected with root-knot nematodes. Species identification was made using morphological methods and morphometric measurements from second-stage infective juvenile and female individuals obtained from pure cultures. As a result, the first record of *Meloidogyne javanica* in lettuce-growing areas in the Southern Marmara Region was made in this study.

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Güney Marmara Bölgesi Marul (*Lactuca sativa* L.) Yetiştirilen Alanlarda *Meloidogyne javanica* (Treub, 1885) (Chitwood, 1949)'nın İlk Kaydı

ÖZET
Bitki paraziti nematodlar arasında yer alan Kök-ur nematodları, geniş konukçu yelpazesi ve mücadelesindeki zorluklardan dolayı en önemli bitki paraziti nematod grubunu oluşturmaktadır. Sebzelerde önemli kalite ve verim kayıplarına sebep olan Kök-ur nematodları marul bitkisinin de önemli bir konukçusudur. Kök-ur nematodları marul üzerinde sararma, solma ve bodurlaşma gibi belirtiler meydana getirmektedir. Marul yaprakları tüketilen bir sebze olmasından dolayı üreticiler tarafından bu durum istenmemektedir. Bu amaçla Çanakkale ili marul yetiştirilen alanlara 2021-2022 yılları kış aylarında periyodik olmayan arazi çıkışları yapılmış ve Kök-ur nematodlarıyla bulaşık bitkiler incelenmiş, ardından saf kültürleri oluşturulmuştur. Toplam 16 bitki ve torpak örneği alınmış 3 tanesinin kök-ur nematodları ile bulaşık olduğu tespit edilmiştir. Morfometrik ölçümler ve morfolojik yöntemlere göre, saf kültürlerden elde edilen ikinci dönem larva ve dişi bireylerden tür teşhisi yapılmıştır. Bu çalışma Güney Marmara Bölgesi Marul yetiştirilen alanlardaki *Meloidogyne javanica*'nın ilk kaydıdır.

Bitki Koruma

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INTRODUCTION
Nematodes have adapted to almost all environments, including terrestrial and aquatic habitats; and belong to the "Nematoda" clade, which are distributed all over the world and live as parasites on plants, animals, and humans (Cavaness & Jensen, 1955; Boag et al., 1998). The Tylenchida (Nematoda) order constitutes the most important group among the

Plant Parasitic Nematodes (PPNs), containing most of the harmful species.

Root-knot nematodes (RKNs) are in first place among PPNs in terms of their prevalence in the world, difficulties in control, and the level of economic damage they cause to plants (Taylor & Sasser, 1978; Whitehead, 1998; Karssen & Moens, 2006; Kalaiarasan, 2009; Jones et al., 2013). About 100 RKN species have been identified in different hosts in the world to date (Trinh et al., 2019), but most commonly, *M. incognita* (Kofoid & White, 1919) Chitwood, 1949, *M. arenaria* (Neal, 1889) Chitwood 1949, *M. javanica* (Treub, 1885) Chitwood 1949 ve *M. hapla* Chitwood 1949 (Netscher & Sikora, 1990; Eisenback & Triantaphyllou, 1991; Karssen, 2000; Hunt et al., 2005) are known all over the world.

Root-knot nematodes are among the most important organisms in vegetable production because they are found in 52% of cultivated agricultural lands and have a wide host range (Sasser, 1977; Taylor, 1987; Trudgill & Blok, 2001). This is because RKNs are polyphagous organisms, having about 5500 host plants, and cause significant yield losses in vegetables reaching up to 15-85% of total production (Trudgill & Block, 2001; Anonymous, 2008).

Root-knot nematodes are distinguished from other plant parasitic nematodes (PPNs) by the typical galls they form on the roots, which gives the species its name. Root-knot-type root galls occur as a result of hypertrophy (cell growth) and hyperplasia (increase in the number of cells) of the cells on which the nematode creates a feeding site (Bridge & Starr, 2007). Above-ground symptoms (yellowing, wilting, stunting) caused by root-knot nematodes on vegetables produce significant quality and yield losses, especially for vegetables whose leaves are consumed.

The lettuce plant (Balkaya et al., 2017), which originated from Anatolia, Caucasus, Iran, and Turkistan region, is among the vegetables whose leaves are consumed in the Asteraceae family. The optimum growth temperature of lettuce, which is a one-year cool climate vegetable, is around 15-18 °C, and the temperature exceeding 20 °C negatively affects its development. In addition, lettuce can withstand temperatures as low as -2 or -3 °C below 0 °C for a short time (Splittstoesser, 1990; Günay, 1992).

As a result of the studies, aphids (*Aphis gossypii*, *Myzus persicae*), thrips (*Thrips tabaci*, *Frankliniella occidentalis*), leafhoppers (*Empoasca decipiens*, *Asymetresca decedens*), whitefly (*Bemisia tabaci*) and leaf worms (*Spodoptera littolarigera*) are among the species that cause significant damage in lettuce cultivation. Apart from these, RKNs are also reported to cause significant yield and quality losses in lettuce cultivation (Akyazı & Ecevit, 2011; Uzunoğulları et al., 2022).

Therefore, in this study, it was aimed to determine the distribution of root-knot nematodes in lettuce-growing areas in Çanakkale and to determine the species found in the areas by morphological and morphometric methods.

MATERIAL and METHOD

The main material of the study consists of root and soil samples taken from lettuce cultivation areas of Çanakkale province and its districts and female and second-stage infective juveniles of *Meloidogyne* spp. obtained from these samples.

Figure 1. Lettuce plant roots infected by *Meloidogyne javanica* a: Sample No: 85, b: Sample No: 107, and c: Sample No: 115.

Şekil 1. Meloidogyne javanica ile infekteli marul bitkisi kökleri a: Örnek 85, b: Örnek 107 ve c: Örnek 115.

In the winter months of 2021-2022, non-periodic land surveys were made to the lettuce-growing areas in Çanakkale. A total of 16 plant and soil samples were taken from lettuce growing areas in Çanakkale province and its districts, including 7 in the center, 3 in Ezine, 2 in Ayvacık, 2 in Biga, 1 in Çan, and 1 in Lapseki.

The plants found in these areas were examined, and the roots of the plant samples showing signs of infestation with root-knot nematode were brought to the laboratory and examined under a binocular microscope (Figure 1). After the roots infested with root-knot nematodes were detected, an egg pack was taken from the root of each sample with forceps under a binocular microscope. The collected egg packets were used to infect the root-knot nematode-sensitive tomato cultivar "Troy F1" and pure nematode cultures were formed ahead. After about 60 days, female individuals of root-knot nematodes grown in pure cultures for diagnostic studies and second-stage infective juveniles (J2s) were obtained from the egg packages laid by these females using the sieve method.

J2s obtained from egg packs were fixed in TAF solution and then fixed on a slide in pure glycerin according to the method of Seinhorst (1959) and made ready for species identification. Twenty-five root-knot nematode second-stage infective juveniles were measured for each infected sample. Standard identification characters used in the morphological and morphometric diagnosis of second-stage infective juveniles were made using a Leica DM1000 light microscope according to Jepson (1987), and species-level diagnosis was made according to Whitehead (1968) and Özarslandan (2009). After the female individuals were obtained, they were kept in 45% lactic acid. After that step, 1/3 of the vulva region was cut and put in pure glycerin between the slide and the coverslip (Hooper, 1986). Morphological identifications of female individuals were made according to Jepson (1987) and Karssen (2002).

RESULTS and DISCUSSION

The results of the diagnostic studies performed with morphological and morphometric methods showed that the species infecting the roots of the lettuce plant was *M. javanica* (Table 1; Figure 2,3).

According to the species identification, taxonomy studies related to root-knot nematodes conducted in Turkey, 11 different root-knot nematode species (*M. incognita*, *M. javanica*, *M. arenaria*, *M. hapla*, *M. chitwoodi*, *M. artiellia*, *M. Arita*, *M. luci*, *M. exiqua*, *M. Thames* and *M. Ethiopia*) were identified (Kepenekçi et al., 2002; Devran & Söğüt, 2009; Özarslandan & Elekçioğlu, 2010; Aydınlı et al., 2013; İmren et al., 2014; Aydınlı & Mennan, 2016; Aydınlı, 2018; Ataş, 2021; Yılmaz et al., 2023). In previous studies, *M. javanica* species were detected in tomatoes, cucumber, eggplant, bean, celery, okra plants, and lettuce (Uysal et al., 2017; Yağcıköse, 2021).

In previous studies in our country, *M. incognita* species was detected in lettuce-growing areas (Akyazı & Ecevit, 2011: Uzunoğulları et al., 2022). Uysal et al. (2017) detected the *M. javanica* species in the lettuce-growing areas of the Lake District. However, *M. javanica* has not been detected in lettuce-growing areas in the Southern Marmara Region.

In this study, a survey was made in the lettuce-growing areas of the Southern Marmara Region and it was confirmed that the species obtained from the lettuce plant was *M. javanica* by morphological and morphometric methods.

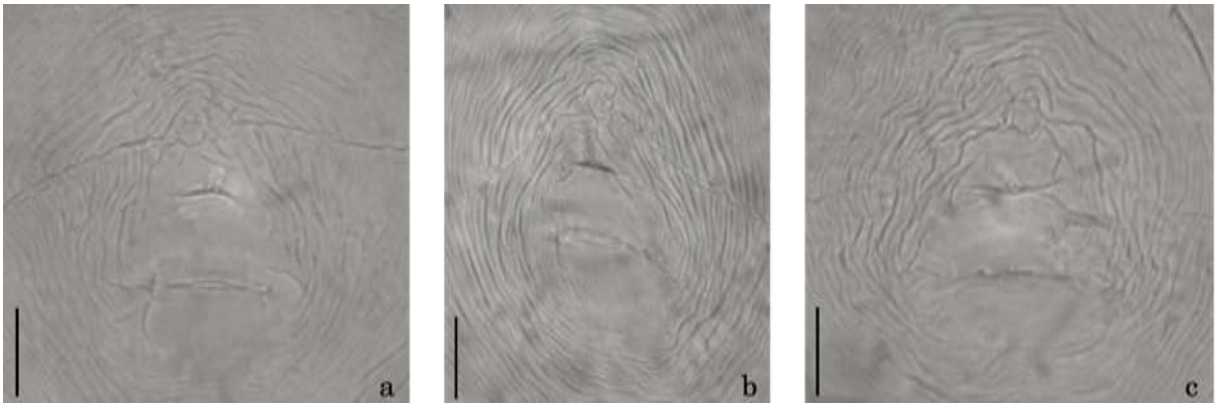

Figure 2. The perineal pattern of *Meloidogyne javanica* from lettuce a: Sample No: 85, b: Sample No: 107, and c: Sample No: 115 (Scale Bar: 20 µm).

Şekil 2. Maruldan elde edilen Meloidogyne javanica'ya ait perineal pattern a: Örnek 85, b: Örnek 107 ve c: Örnek 115 (Ölçek Çubuğu: 20 µm).

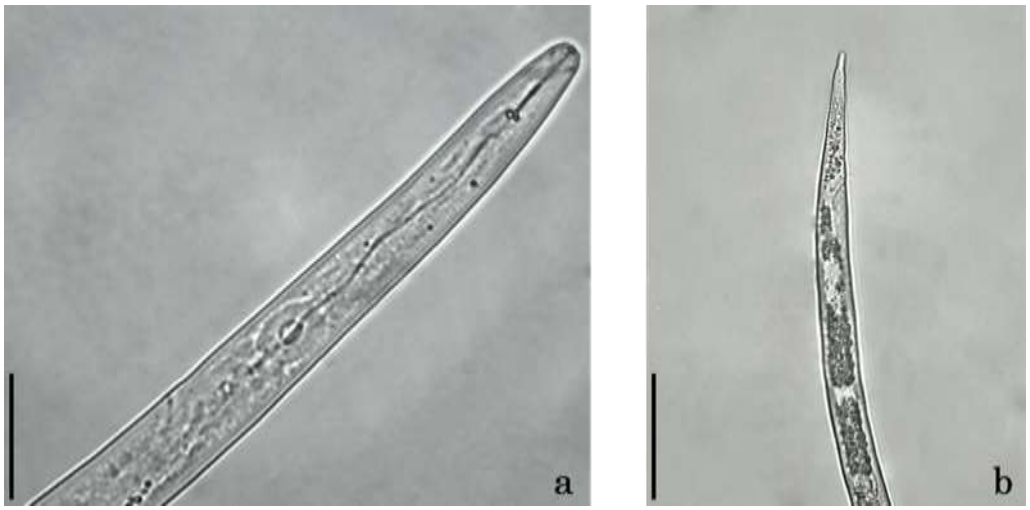

Figure 3. J2s of *Meloidogyne javanica* from lettuce a: anterior region of Sample No: 107, b: posterior region of specimen 107 (Scale Bar: 20 µm).

Şekil 3. Maruldan elde edilen Meloidogyne javanica'ya ait J2s a: Örnek 107 anterior bölgesi, b: Örnek 107 posterior bölgesi (Ölçek Çubuğu: 20 µm).

Table 1. Morphometric measurements of J2s of *Meloidogyne javanica* obtained from lettuce plant roots

Çizelge 1. Marul bitkisi köklerinden elde edilen Meloidogyne javanica'ya ait J2s'lerin morfometrik ölçümleri

Diagnostic Characters	This Study Sample 85	This Study Sample 107	This Study Sample 115	Whitehead (1968)	Özarslandan (2009)
Body length	416.93±19.14 (392.92-456.24)	428.97±17.94 (401.30-450.00)	431.33 ±13.01 (405.15-450.80)	387-459	408-454.4
Greatest body width	15.32±0.76 (14.07-16.86)	15.89±0.37 (15.10-16.54)	15.66±0.49 (14.46-16.54)		
Body width at stylet base	9.75±0.37 (9.13-10.63)	9.94±0.34 (9.11-10.50)	9.99±0.54 (8.48-10.85)		
Body width at the anus	9.80±0.52 (8.60-10.63)	9.93±0.45 (9.00-10.73)	10.17±0.52 (9.21-10.91)		
Stylet length	15.66±0.42 (14.80-16.30)	15.54±0.38 (15.01-16.19)	15.42±0.37 (14.60-16.09)	9.4-11.4	11.20-14.40
DGO	2.98±0.36 (2.15-3.58)	2.78±0.37 (2.04-3.37)	2.93±0,41 (2,27-3.98)	4	3.2-4
Tail length	52.14±2.05 (48.39-56.28)	54.22±1.57 (51.30-57.84)	53.66±2.10 (49.63-57.22)	36-56	46.40-59.20
Excretory pore to head end	84.54±2.01 (81.71-89.79)	84.79±1.21 (82.94-87.83)	84.45±1.62 (81.55-87.45)		
Body width at the excretory pore	13.32±0.65 (12.03 -14.38)	14.51±0.46 (13.35-15.33)	13.93±0.69 (12.57-15.13)		
a	27.24±1.23 (24.92-30.38)	26.98±1.16 (25.18-28.91)	27.46±1.10 (25.36-29.27)	27.1-35.9	30.33
b	3.90±0.19 (3.63-4.21)	4.08±0.14 (3.81-4.34)	4.06±0.14 (3.80-4.34)		
c	8.00±0.37 (7.39-8.81)	7.91±0.36 (7.39-8.71)	8.01±0.37 (7.39-8.72)	7.3-11.1	8.31
c'	5.33±0.34 (4.83-6.13)	5.46±0.29 (5.02-5.97)	5.27±0.28 (4.83-5.97)		

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.

This study is the first record of *M. javanica* detected on lettuce in the Southern Marmara Region. Above-ground (yellowing, wilting, and stunting) and root symptoms caused by *M. javanica* in the lettuce plant

cause significant yield and quality losses in this plant. For this reason, it is recommended to carry out more comprehensive studies in the future, in lettuce growing areas to monitor and conduct control strategies against root-knot nematodes such as rotation, use of resistant varieties, and solarization applications.

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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