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Araştırma
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Effect of *Silybum marianum* (L.) Gaertn. Leaf and Seed Extracts Prepared Using Different Solvents on Root-Knot Nematode

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Abstract

Objective: The nematicidal effect of milk thistle leaves and seeds prepared with different solvents on *Meloidogyne incognita* was investigated.

Materials and Methods: Acetone, ethanol and distilled water were used. The *in vitro* was carried out in 6 cm petri dishes. The extractions were studied with 500 and 1000 µg/ml (ppm). The *in vitro* and pot experiments designed random plots with 5 replications for each extraction, solvent and concentration. Four hundred second stage juvenile larvae (J2) were used as inoculum and dead individuals were counted after 48 hours. Five days after transplantation, nematode inoculation was carried out with 500 J2 per pot. After 24 hours, 30 ml of the solution was applied to the soil at 1000 ppm concentration. The experiment was terminated for 50 days. Then, gall and egg mass counts were made.

Results: *In vitro*, the mortality rate at 1000 ppm was found to be similar in acetone (78.0%) and ethanol (80.8%) solvents in leaf extraction, while the highest was detected in ethanol (94.0%) in the seed extract. In distilled water solvent, 68.0% mortality was determined in the leaf extract and 62.2% mortality in the seed extract. There was no statistically significant difference between the leaf and seed extracts in number of galls and egg masses. No statistical difference could be determined between the solvents in the number of egg masses in seed extraction. While the number of galls in the leaf extract was found to be higher than in acetone (8.8 unit/root) and ethanol (8.0 unit/root) in distilled water (18.0 unit/root) and the difference between them was found to be significant, no statistically significant difference in the number of egg mass between the solvents.

Conclusion: It was observed that all solvents of the leaf and seed extract suppressed galls and egg masses by more than 80% compared to the control.

Keywords: Milk thistle, root-knot nematode, herbal extract, Asteraceae, nematicidal effect

Farklı Çözücüler Kullanılarak Hazırlanan *Silybum marianum* Yaprak ve Tohum Ekstraktlarının Kök Ur Nematoduna Etkisi

Öz

Amaç: Farklı çözücülerle hazırlanan deve dikeni yaprakları ve tohumlarının ekstraktlarının *Meloidogyne incognita* üzerindeki nematisit etkisi araştırılmıştır.

Materyal ve Yöntem: Aseton, etanol ve damıtılmış su kullanılmıştır. *İn vitro* çalışmalar 6 cm'lik petri kaplarında gerçekleştirilmiştir. Ekstraksiyonlar 500 ve 1000 µg/ml (ppm) ile çalışılmıştır. *İn vitro* ve saksı denemeleri, her ekstraksiyon, çözücü ve konsantrasyon için 5 tekrarlı rastgele parsellere göre tasarlanmıştır. İnokulum olarak 400 ikinci dönem larva (L2) kullanılmış ve 48 saat sonra ölü bireyler sayılmıştır. Dikimden beş gün sonra saksı başına 500 L2 ile nematod inokulasyonu yapılmıştır. 24 saat sonra 1000 ppm konsantrasyonda 30 ml çözelti toprağa uygulanmıştır. Deneme 50 gün sonra sonlandırılmıştır. Daha sonra gal ve yumurta paketi sayımları yapılmıştır.

Araştırma Bulguları: *İn vitro* da yaprak ekstraksiyonunda 1000 ppm'deki ölüm oranı aseton (%78.0) ve etanol (%80.8) çözücülerinde benzer bulunurken, en yüksek ölüm oranı tohum ekstraktında etanolde (%94.0) tespit edilmiştir. Distile su çözücüsünde yaprak ekstraktında %68.0,

tohum ekstraktında ise %62.2 ölüm oranı belirlenmiştir. Yaprak ve tohum ekstraktları arasında gal sayısı ve yumurta paketi açısından istatistiksel olarak anlamlı bir fark bulunamamıştır. Tohum ekstraksiyonunda yumurta paketi sayısı açısından çözücüler arasında istatistiksel olarak bir fark tespit edilememiştir. Yaprak ekstraktındaki gal sayısının, distile suda (18.0 birim/kök) aseton (8.8 birim/kök) ve etanolden (8.0 birim/kök) daha yüksek olduğu ve aralarındaki farkın anlamlı olduğu tespit edilmiştir. Çözücüler arasında yumurta paketi sayısı açısından istatistiksel olarak anlamlı bir fark yoktur.

Sonuç: Yaprak ve tohum ekstraktındaki tüm çözücülerin gal ve yumurta paketlerini kontrole göre %80'den fazla baskıladığı gözlenmiştir.

Anahtar Kelimeler: Devedikeni, kök ur nematodu, bitkisel ekstrakt, Asteraceae, nematisidal etki

Introduction

Root-knot nematodes (*Meloidogyne* spp.) are very common species of greenhouse-growing plants in the world (Mateille et al., 2020. They have sedentary endoparasites feeding and cause damage to the vascular system (Subedi et al., 2020). This ultimately causes stunting, galled root formation, and early death in the plant (Palomares-Rius et al. 2017. Plants infected with root-knot nematode become susceptible to secondary microorganisms (Greco & Vito,2009). Although 105 species of root-knot nematodes have been reported so far, *Meloidogyne incognita* (Kofoid & White, 1919) Chitwood, 1949, *Meloidogyne arenaria* (Neal, 1889) Chitwood, 1949, *Meloidogyne javanica* (Treub, 1885) Chitwood, 1949, and *Meloidogyne hapla* Chitwood, 1949 are particularly significant species in the World (Karssen et al., 2013; Maleita et al. 2021). In Türkiye, ten *Meloidogyne* species *M. incognita, M. javanica, M. arenaria, M. hapla, M. chitwoodi, M. artiellia, M. acrita, M. luci, M. exiqua* and *M. thamesi*, have been recorded (Çetintaş & Çakmak, 2016; Aydınlı, 2018; Gürkan et al., 2019; Arslan & Elekçioğlu, 2022). *Meloidogyne incognita* is one of the most important species from this genus (Regmi and Desaeger 2020; Vendruscola et al., 2024).

The common controlling methods of nematodes is sanitation and selecting plant varieties that exhibit resistance. However, not every plant has nematode-resistant varieties, and virulent populations are also observed in resistant varieties and resistance is broken (Bernard et al., 2017). Soil fumigant is generally used in chemical control of nematodes. Several highly effective and widely used nematicides (Class 1 pesticides) have faced severe restrictions under pestilent substances (Oka, 2020). Recently, plant extracts have become an environmentally friendly alternative to chemical pesticides in pest control (Aji, 2024). Plants are a good source for revealing new pesticide molecules. Numerous nematicidal compounds have been isolated from various plant families, including Asteraceae (Oka, 2010). Nematicidal sesquiterpenic acids and monoterpenes reported that Asteraceae (Chitwood, 2002; D'Addabbo et al. 2013). Tsay et al. (2004) reported that of the 56 species and 43 genera of Asteraceae tested, 9 were highly resistant or immune to *M. incognita*. *Tagetes minuta, Artemisia annua, Bidens pilosa* and *Chrysanthemum cinerariaefolium* of Asteraceous plants are repellent to *M. incognita* (Sydney, 2016). In an Asteraceae plant, Golden Crown-Beard [*Verbesina encelioides*], the strongest nematicidal activity was found in the aqueous extracts of the fresh flower, followed by the leaf and stem extracts, respectively (Oka, 2012).

Milk thistle (*Silybum marianum* (L.) Gaertn.) is a spiny herb into the *Asteraceae* family and found throughout the World (Porwal et al., 2019). It is known that milk thistle has been used for medicinal purposes for over 2000 years (Polyak et al., 2010), since no health hazards or side effects are known (Med. Economic Company, 2000). Medicinal and aromatic plants produce various secondary metabolites during their development. These compounds are generally antioxidant phenolic compounds with redox properties (El-Abbassi et al., 2012). The most important secondary metabolite of milk thistle is a flavonoid complex called silymarin, which consists of a mixture of silybin A and B, isosilybin A and B, silychristin and silydianin (Anthony & Saleh, 2012). Milk thistle seeds generally contain 1-5% silymarin. The extracts produced from its seeds contain 70-80% silymarin. Other organs of milk thistle (leaf, flower, root) do not contain silymarin (Çelik & Yüksel, 2013). However, different flavonoids (taxifolin, quercetin, dihydrokemferol, kemferol, apigenin, naringin, eriodictyol, chrysoeriol) and other compounds (fixed oils, sterols, dihydroconiferyl alcohol, proteins and mucilage) found in milk thistle structure (Tajmohammadi et al., 2018). Previously, some extracts of *S. marianum* were reported to have antibacterial and antifungal activities (Zaouia et al., 2010; Ahmad et al., 2015; Mohammed et al., 2019).

The percent mortality and inhibition of hatching of *M. incognita* were found to be directly proportional to the concentration of lignans and their various derivatives from *S. marianum*. At 24 hours, silybin pentabenzoate showed the highest nematicidal activity at 500 µg/ml, causing a 99% mortality rate (Pandey, 2011). Although there are limited studies in the world on the nematicidal effect of milk thistle, no detailed study has been found in Türkiye. The aim of this study was to investigate the nematicidal effect of milk thistle leaves and seeds, extracts prepared with acetone, ethanol and distilled water solvents *in vitro* and on tomatoes under controlled conditions.

Material

The material of the study consists of extracts obtained from milk thistle leaves and seeds. It was collected in July, 2022 from Deregümü, the central village of Isparta Province. Sampling was done from adjacent fields within the same region (N: 37°47'6.19'', E:30°30'9.78''). They were brought to the laboratory (23°C) and left to dry up to 20 days. Then the leaves and purple flowers are separated with pruning shears. Seeds appear when you pull the yellow hairs from the dried purple flowers.

Preparation of leaf extractions

The separated leaves were ground into flour using a spice grinder. Ethanol, acetone and distilled water were used as solvents. 500 g of the floured leaf extract was weighed and separated, and 500 ml of different solvents (acetone, ethanol and distilled water) were added and kept at room temperature for 48 hours. At the end of this period, it was filtered using filter papers. Drying was carried out in a rotary evaporator to evaporate the solvents in the obtained filtrates (Adekunle et al., 2007).

Preparation of seed extractions

The seeds were ground into flour using a spice grinder. Ethanol, acetone and distilled water were used as solvents. 20 g of the floured seed extract was weighed and separated, and 200 ml of different solvents (acetone, ethanol and distilled water) were added and kept at room temperature for 24 hours. At the end of this period, it was filtered using filter papers. Drying was carried out in a rotary evaporator to evaporate the solvents in the resulting filtrates (Kabil &Adam, 2020).

Nematode ınoculum

Meloidogyne incognita ISP isolate, which was maintained in mass production tomato variety of Tueza F1under climate chamber conditions (24±1 °C, 60±5% humidity), was used in the experiment. Egg masses were removed from tomato roots under a binocular microscope, placed in sieves in a 9 cm petri dish containing distilled water, and incubated at 28°C for three days. In this way, second stage juvenile larvae (J2) emerged from the egg masses. J2 counts were made under a light microscope and placed in Eppendorf tubes ® and stored at +4 °C to be used in the experiment (Göze Özdemir et al., 2022).

***In vitro* experiment**

The experiment was carried out in 6 cm petri dishes. 400 J2 was used to determine nematicidal activities of different extracts on J2 (Kabil & Adam, 2020). Stock solutions were prepared by diluting evaporator-dried floured leaf and seed extracts with distilled water containing 0.3% Tween 20, separately. Then, concentrations of 500 and 1000 µg/ml (ppm) were prepared from the stock solution. 0.3% tween 20 was used as a control. The study was set up with 5 replications in a randomized plot trial design for 500 and 1000 ppm concentrations of each extraction (Kabil & Adam, 2020). After the experiment was set up, petri dishes were stored at 25 ± 1°C. Dead J2s were determined 48 hours later under a 40x binocular microscope. Nematodes were considered dead if they did not move when examined with a fine needle. The experiment was repeated twice and percentage mortality values were calculated with the abbott formula (m) (Finney, 1978). Then, their averages were taken and subjected to statistical analysis.

m = 100 (1- (nt/nc)) (m = percent mortality, nt = number of alive nematodes after application, and nc = number of alive nematodes in water control)

Pot experiment

The study was carried out in pots under climate chamber conditions (24±1 °C, 60±5% humidity) with thirty-five-day-old Özkan F1 tomato seedlings, which are known to be sensitive to nematodes. The experiment was set up in a randomized plot design with 5 replications for each concentration. Tomato seedlings obtained from Olympos Fide (Kumluca, Antalya) were transplanted into 250 ml plastic pots containing a sterile 300 g (68% sand, 21% silt and 11% clay) soil mixture, with 1 tomato seedling per replicate. Five days after transplate, nematode inoculation was carried out with 500 J2 in each pot (Vinodhini et al., 2019). Stock solutions were prepared by diluting the evaporator-dried floured leaf and seed extracts with distilled water containing 0.3% Tween 20, separately.

Twenty four hours after nematode inoculation, for each extract from the stock solutions, 30 ml of solution was applied to the soil around the plant with the help of a measuring tape, at a concentration of 1000 ppm in each pot (Kabil & Adam, 2020). Distilled water containing 0.3% Tween 20 was used as a negative control. The experiment was terminated 50 days after nematode inoculation. Then, the plants were removed and washed with water to remove soil from the roots of the plant, and after the roots were color with acid fuchsin, galls and egg masses were counted (Moltmann, 1988).

Statistical analyzes

SPSS (version 20.0) program was used for statistical analysis of the data obtained. Analysis of variance (ANOVA) was used to test differences between means compared with Tukey HSD test at $P \leq 0.05$.

Results and Discussion

The mortality effect of 500 and 1000 ppm concentrations of milk thistle leaves acetone and ethanol extracts on J2 was similar but higher than that of sterilled water extract. At 500 ppm, the mortality effect of acetone, ethanol and distilled water extracts was 45.2%, 53.0% and 39.0%, respectively, while at 1000 ppm, it was 78.0%, 80.8% and 68.0%, respectively. As the concentration increased, the mortality effect increased. There is no statistical difference between acetone and ethanol extract on J2 mortality at 1000 ppm concentration (P≤0.05) (Table 1).

Table 1. Percentage mortality effect of leaf extracts on second stage juvenile larvae

	%Mortality±Standard error Concentration		
Solvent	**500 ppm**	**1000 ppm**	**%0.3 Tween 20 (Control)**
Acetone	45.2± 2.6 b AB*	78.0± 2.0 a A	0.6± 0.2 c
Ethanol	53.0± 2.8 b A	80.8± 1.7 a A	0.6± 0.2 c
Sterilled water	39.0± 1.3 b B	68.0± 1.1 a B	0.6± 0.2 c

*Uppercasel letters in the same column indicate the difference between solvents, and lowercase letters in the same row indicate the statistical difference between concentrations (P≤0.05).

While the highest number of galls and egg masses was in control, it was determined that the extracts of milk thistle leaves significantly reduced the number of galls and egg masses. As the concentration increased, the number of galls and egg masses decreased. At 1000 ppm concentration, there was no statistical difference in the gall number between acetone and ethanol (P≤0.05), but it was determined to be higher than the sterile water extract. No statistical difference was found between acetone, ethanol and distilled water in the number of egg masses (P≤0.05) (Table 2).

Table 2. Effect of leaf extracts on the number of galls and egg masses on tomato roots

	Mean of galls number±Standard Error Concentration		Mean of egg masses number± Standard Error Concentration	
Solvent	**500 ppm**	**1000 ppm**	**500 ppm**	**1000 ppm**
Acetone	20.2± 1.8 a * B	8.8± 0.5 ab A	17.6± 2.2 a B	7.2± 0.5 a A
Ethanol	18.0± 2.4 a B	8.0± 0.8 a A	15.8± 2.0 a B	7.2± 0.7 a A
Sterilled water	37.6± 1.8 b B	18.0± 0.9 b A	33.0± 1.1 b B	14.4± 1.2 a A
%0.3 Tween 20 (Control)	138.2± 4.4 c	138.2± 4.4 c	130.0± 5.4 c	130.0± 5.4 b

*Lowercase letters in the same column indicate the difference between solvents, and uppercase letters in the same row indicate the statistical difference between concentrations (p≤0.05).

The mortality effect of solvents on J2 in seed extracts is significantly different from each other (P≤0.05). The lowest percentage of mortality was found in the sterilled water extract, and the highest effect was found in the ethanol extract. At 1000 ppm, the mortality effect of acetone, ethanol and distilled water extracts was 81.6%, 94.0% and 62.2%, respectively (Table 3).

Table 3. Percentage mortality effect of seed extracts on second stage juvenile larvae

	%Mortality±Standard Error Concentration		
Solvent	**500 ppm**	**1000 ppm**	**%0.3 Tween 20 (Control)**
Acetone	58.6±2.0 b B	81.6±2.3 a B	0.8±0.3 c
Ethanol	78.2±2.0 b A	94.0±2.5 a A	0.8±0.3 c
Sterilled water	46.8±2.8 b C	62.2±1.3 a C	0.8±0.3 c

*Uppercase letters in the same column indicate the difference between solvents, and lowercase letters in the same row indicate the statistical difference between concentrations (P≤0.05).

Unlike *in vitro*, in the study conducted under controlled conditions, there was no significant difference between the solvents on the number of galls and egg masses of the seed extracts. However, a significant decrease was determined compared to the control (P≤0.05). The number of galls and egg masses was found to be less at 1000 ppm concentrations of the solvents than 500 ppm. While the number of galls and egg masses was found to be lower in ethanol seed extract than in acetone and distilled water, the difference between them was found to be statistically insignificant (P≤0.05) (Table 4)

Table 4. Effect of seed extracts on the number of galls and egg masses on tomato roots

	Mean of galls number±Standard Error Concentration		Mean of egg masses number± Standard Error Concentration	
Solvent	**500 ppm**	**1000 ppm**	**500 ppm**	**1000 ppm**
Acetone	12.2± 0.8 a * B	5.4± 0.5 a A	9.6± 0.8 a B	4.8± 0.3 a A
Ethanol	7.2± 0.8 a B	1.4± 0.5 a A	6.8± 0.7 a B	1.4± 0.5 a A
Sterilled water	16.6± 1.5 a B	7.2± 0.7 a A	14.6± 1.5 a B	5.6± 0.6 a A
%0.3 Tween 20 (Control)	120.2± 6.9 b	120.2± 6.9 b	110.0± 6.6 b	110.0± 6.6 b

*****Lowercase letters in the same column indicate the difference between solvents, and uppercase letters in the same row indicate the statistical difference between concentrations (P≤0.05).

Although there was a difference between the solvents at 1000 ppm concentration in the percent mortality effect *in vitro* and the suppressive effect on the gall and egg mass in tomato roots, no statistically significant difference was found between the leaf and seed extracts of the same solvent (Table 5). *In vitro*, the mortality rate at 1000 ppm was found to be similar in acetone and ethanol solvents in leaf extraction, while the highest was detected in ethanol in the seed extract. No statistical difference could be determined between the solvents in terms of the number of eggs number in seed extraction (Table 5).

Table 5. Effect of leaf and seed extractions at 1000 ppm concentration on *Meloidogyne incognita*

	Mortality Effect (%) on J2 Extraction		Mean of galls number Extraction		Mean of egg masses number Extraction	
Solvent	**Leaf**	**Seed**	**Leaf**	**Seed**	**Leaf**	**Seed**
Acetone	78.0± 2.0a A	81.6±2.3b A	8.8± 0.5ab A	5.4± 0.5a A	7.2± 0.5a A	4.8± 0.3a A
Ethanol	80.8± 1.7a A	94.0±2.5 a A	8.0± 0.8a A	1.4± 0.5a A	7.2± 0.7a A	1.4± 0.5a A
Sterilled water	68.0± 1.1 b A	62.2±1.3c A	18.0± 0.9 bA	7.2± 0.7a A	14.4± 1.2a A	5.6± 0.6a A
%0.3 Tween 20 (Control)	0.6± 0.2c	0.8±0.3d	138.2± c 4.4	120.2± 6.9a	130.0± 5.4b	110.0± 6.6 b

* Lowercase letters in the same column indicate the difference between solvents, and uppercase letters in the same row indicate the statistical difference between leaf and seed extraction (P≤0.05).

Consequently, Milk thistle leaf and seed extract had a nematicidal effect on *M. incognita* compared to the control. The nematicidal effects of seed and leaf extracts are very similar. *In vitro*, solvents were found to be important in terms of nematicidal effect. The nematicidal effect of leaf and seed extracts made using ethanol and acetone solvents was found to be higher than that of distilled water extract. This may be due to differences in the composition of phenolic compounds in leaf and seed extracts. However, the same results were not achieved in the pot experiment. The suppressive effect of the extract prepared from distilled water on the number of galls and egg mass in tomato roots was found in the same statistical group as acetone and ethanol. This is important for a more environmentally friendly control on root-knot nematode. Water is a good polar solvent (Bonnaillie et al., 2012). Ethanol has been reported to be the best extractor of flavonoids in decoction (Bourgou et al., 2016). Bettaieb et al. (2016) investigated the effect of different solvents on antiradical activity and found that antioxidants were most active in ethanol. Benchaachoua et al. (2018) reported that phenolic compounds from *S. marian*um leaves were strongly influenced by the type of solvent and the extraction method.

Previous studies on milk thistle, including in Türkiye, have shown that its antibacterial, antioxidant, antifungal and antibacterial properties have been investigated. Yaldız (2017) reported that n-hexane extract of seeds of *S. marianum* collected from Türkiye was reported to be effective against *Pseudomonas aeruginosa*, *Escherichia coli, Staphylococcus aureus, Salmonella typhi, Aspergillus niger* and *Candida albicans* at different concentrations. Aqueous and methanol extracts of S*. marianum* collected from Algeria were reported to be effective against *S. aureus, E. coli, Klebsiella pneumoniae, P. aeruginosa, Enterobacter aerogenes* and *C. albicans* at different concentrations (Zaouia et al., 2010).

Ahmad et al. (2015), found to be effective against *E. coli, Salmonella* spp.*, Shigella* spp.*, S. aureus* and *V. cholerae* at different concentrations of methanol, n-hexane and chloroform extracts of *S. marianum* collected from 10 different regions of Pakistan. Mohammed et al. (2019) determined that antioxidant and antibacterial activities in *S. marianum* ethanol, methanol and dichloromethane extracts of the fruit part which collected from Duhok (Iraq). With this study, the nematical effect of *S. marianum* (Astereceae) collected from Türkiye was demonstrated for the first time. Many plants in the Astereceae family have already been reported to have nematicidal effects. An aqueous extract of the leaves and stem of *Eupatorium odoratum* which is an herbaceous plant belonging to the Asteraceae family emerged as an effective nematode management on *Radopholus similis* (Tran & Quac, 2024). Bay biscaine, tridax daisy, French marigold, wild sunflower, buttercup and Peruvian zinnia species of Asteraceae were highly resistant to the *M. incognita* (Ferreira et al., 2013). *Gaillardia pulchella* could be used to manage *M. incognita* as a rotation crop, a co-planted crop, or a soil amendment for control of root-knot nematode (Tsay et al., 2004).

Milk thistle may have killed the root-knot nematode with the synergistic or antagonistic effect of the flavonoid compounds it contains. Silymarin is a polyphenolic flavonoid derived from milk thistle which is found in the leaves, seeds, and fruits which it has the highest therapeutic effects compared with other flavonolignans (Emadi et al., 2022). Flavonoids are polyphenol secondary metabolites that can have various structures. They are found in aglycone or glycoside form in many plants. Flavonoids include flavonols, flavones, flavonoids, flavanones, anthocyanidins, and isoflavones (Queiroz Ferreira et al., 2015; Li et al., 2017). These compounds are pigments of different colors that are known to be widely found in the leaves, seeds, bark and flowers of plants. Functions; protection from ultraviolet light, defense against abiotic stresses and from bacterial and fungal phytopathogens. They have also been found to serve as endogenous regulators of auxin movement in plants (Li et al., 2017; Sandoval-Yañez et al., 2018). Silybin pentabenzoate caused 99% mortality at 500 µg/ml after 24 hours *in vitro* on *M. incognita* (Pandey, 2011). Massuh et al. (2017) reorted that the main component of *Tagetes minuta* (Asteraceae) essential oil and nematicidal activity against *M. javanica* as E)-Ocimenone. *Artemisia pedemontana* subsp. *assoana* (Asteraceae) hydrosols have strong nematicidal activity against *M. javanica* (Sainz et al., 2019). Thujone and camphor are regarded as the major contributors of nematicidal activity against *M. incognita* in *Artemisia herba-alba* (Avato et al., 2017).

Milk thistle has different uses and benefits which has antioxidant, anti-inflammatory, anticancer, antifungal, immunomodulatory, and other properties. It has no negative effects on human health and is widely used in the treatment of liver diseases (Abenavoli et al., 2010; Gillessen &Schmidt, 2020). This feature offers an advantage in terms of its use in the control against root-knot nematodes.

Conclusion

This is the first study conducted in Türkiye on the use of milk thistle in nematode control. It has been determined to be promising in controlling root knot nematode. Although, silymarin is thought to be effective on root-knot nematodes, the compound(s) that have nematicidal effects are unknown. Therefore, nematicidal compounds of milk thistle should be determined and purified. Once identified, they or their derivatives can be artificially synthesized. Their use will add another approach to nematode control.

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