







Comparison of polyphenolic content, radical scavenging activity, and mineral concentrations of *Cuscuta monogyna* VAHL on different host plants

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ABSTRACT

Background and Aims: *Cuscuta* sp. (Dodder), known as “İkşüt”, is a parasitic herbaceous plant that negatively affects the yield and quality of cultivated crops; however, it has been used as an ancient medicinal plant for curing liver problems for centuries in the southeastern part of Turkey. The aim of this study was to investigate the total phenolic and flavonoid contents and mineral compositions of *Cuscuta monogyna* Vahl. subsp. *monogyna* Vahl. (CMM) grown on different host plants, along with their antioxidant activity, and to compare the host plant parts and their collected dodders.

Methods: In this study, dodders were collected from host plants, including vineyardolive, pomegranate, green pepper, and liquorice in Turkey. The dodders were removed from their hosts before the drying process. Mineral contents were determined by Inductively Coupled Plasma Atomic Optical Spectroscopy (ICP-OES), and phytochemical contents were identified by chromatography. The total polyphenolic compositions and antioxidant activities were determined spectrophotometrically.

Results: Because of the phenolic contents in the host plants and dodders, a significant variation was observed, with values ranging from 5.175% to 35.238%.. Different plant extracts and their varied dilutions had radical scavenging activity on 2,2-diphenyl-1-picrylhydrazyl in the ranges of 57.78 - 95.87%. Variations in mineral compositions were also determined in the dodders and their host plants.

Conclusion: Mutualist or semi-parasitic plant communities share many phytochemical compounds and thus have the potential to exhibit similar activities. With regard to the obtained results, the chemical composition and pharmacological activities of dodder plants are closely related to their host plants.

Keywords: *Cuscuta* sp, dodder, host plant, antioxidant, mineral content, polyphenolic

INTRODUCTION

Cuscuta is a climbing-parasitic plant genus that is also generally known as ‘Dodder’. It is a non-chlorophyll and leafless-flowering herbaceous plant that absorbs water, mineral matter, and photosynthetic substances by releasing these haustoriums into the wrapping of the host plant. However, being harmful parasites, they have ecological significance as keystone plant species for biocontrol (Li et al., 2015; Ahmad, Tandon, Xuan, & Nooreen, 2017; Ho & Costea, 2018).

Cuscuta species, which are important parasitic plants for

agricultural production and traditional folk medicine in Turkey, are known as ‘İkşüt’ in Mardin region, and named as ‘bostan-bozan, canavarotu, bagbozan, cinsacı, eftimon, gelinsaçı, kızıl-sarmaşık and seytansacı’ in other regions (Baytop, 1997).

Dodders have a wide range of distribution by nearly 200 species almost all around the world, and 10 different species of them are widely distributed in Turkey (Davis, Mill, & Tan, 1965 Costea, García, & Stefanović, 2015). In Anatolia, three different parasitic species of dodder, including *Cuscuta campestris* Yunck., *C. approximata* Bab., and *C. monogyna* Vahl., were found on common cropped plants (Nemli, 1986). Additionally,

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Submitted: 11.02.2022 • Revision Requested: 21.10.2023 • Last Revision Received: 21.03.2024 • Accepted: 21.03.2024



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C. arvensis has been observed on sugar beet, onion, clover, and other vegetables. These are parasitic crop-damaging plants which causes economically 50 - 90% yield loss in cultivated fields throughout the world. Furthermore, it was found that *Cuscuta* species were mostly damaged to plants such as clover, tomato, carrot, onion, and pepper (Lanini & Kogan, 2005).

In addition to being a harmful parasite, dodder has been prepared as an infusion or maceration for new-born mothers (one or two waterglass) and rarely infants in very small quantities (2-3 teaspoons) in traditional folk medicine in Mardin District in Turkey. In traditional uses, the dried plants are cleaned by passing through cold water, and the tea is prepared in warm water for about one night at room temperature with its lid closed by maceration. Then, the macerated tea is drunk by the mother without sweetener such as sugar or honey. The duration of treatment (3-4 glasses of water per day) is approximately 1 month before and after birth (Şekeröğlu, Koca, & Meraler, 2012).

It has been reported that the flowering branches of *C. europaea* L. have urinary enhancing, laxative, spasmolytic, and kolagogic effects (Baytop, 1999). While its seeds have been mostly used in aphrodisiac recipes in China, herba has been used in western countries to treat different ailments in folk medicine for centuries. Seeds of *C. chinensis* Lam. have also been used to treat liver and kidney problems, improve sexual function, prevent eye complaints such as blurred vision and fatigue; and to prevent clinical ageing (Zheng, Dong, & She, 1998).

According to previous reports, *C. chinensis* was found to possess anticancer, immunostimulant, and antioxidant properties (Nisa, Akbar, Tariq, & Hussain, 1986; Bao, Wang, Fang, & Li, 2002; Umehara et al., 2004). Furthermore, glycosides of *C. chinensis* have been shown anti-ageing and memory-enhancing effects by inducing PC12 cell differentiation (Jian-Hui, Bo, Yong-Ming, & Li-Jia, 2003). Detailed investigations revealed that *C. chinensis* seeds contain one trisaccharide and four new glycosidic acids, acetic acid, propionic acid, methylbutyric acid, tiglic acid, convolvulinic acid, and jalapinic acid, in the ester-insoluble resin glycoside-like fraction (Du, Kohinata, Kawasaki, Guo, & Miyahara, 1998). Studies on *C. japonica*'s polar extract have shown that the active components are caffeoylquinic acid and caffeoylvinyllate derivatives (Oh, Kang, Lee, & Lee, 2002). Subsequent studies have revealed that the methanolic extract of *C. reflexa* has strong antioxidant potential (Pal, Mandal, Senthilkumar, & Padhiari, 2006); and ethylacetate and methanol extracts of *C. chinensis* have antibacterial properties (Yen, Wu, Lin, Cham, & Lin, 2008); and also major components of *C. japonica* have antihypertensive effects (Oh et al., 2002). In addition, studies have shown that the glycoprotein of *C. europaea* has an immunostimulant effect (Stanilova, Zhelev, & Dobрева, 2000), the ethanolic extract of *C. chinensis* has hepatoprotective activity (Yen, Wu, Lin, & Lin, 2007), and the methanolic extract of *C. reflexa* has an ovarian steroido-

genesis suppressant effect (Gupta, Mazumder, Pal, & Bhattacharya, 2003). Hepatoprotective effects of methanolic extract and anti-inflammatory and analgesic effects of both ethanolic and aqueous extracts of *C. arvensis* were determined in mice in our previous studies (Koca, Küpeli-Akkol, & Sekeroglu, 2011; Koca-Caliskan et al., 2018). Although biological studies of the extracts obtained from *C. arvensis* and other *Cuscuta* species have been performed, no phytochemical studies have been conducted on *C. arvensis* except in our study in Turkey. Previous studies have been conducted to elucidate the phytochemical content of different *Cuscuta* species, and flavonoids, lignins, quinic acid, and polysaccharides were determined, particularly in *C. chinensis* (Du et al., 1998; Wang, Fang, Ge, & Li, 2000; Ye, Li, Yan, Liu, & Ji., 2002; Ye, Yan, & Guo 2005). Thus, this study was assumed to be the first report to determine antioxidant activity, chemical and polyphenolic contents, as well as mineral compositions of *C. monogyna* VAHL subsp. *monogyna* VAHL collected from different host plants and evaluate each plant by comparing the plants and their dodders to each other.

MATERIAL AND METHOD

Plant materials

Cuscuta monogyna VAHL subsp. *monogyna* VAHL along with their hosts used as materials in this study were supplied from the agricultural production areas and natural flora of Kilis and Gaziantep, located in the southeastern part of Turkey. *Cap-sicum annum* L. (green pepper) was collected from Bulamaçlı Village of Kilis in September; *Vitis vinifera* L. (vineyard) and *Olea europea* L. (olive) were collected from Akçabağlar Village of Kilis in July; and *Punica granatum* L. (pomegranate) and *Glycyrrhiza glabra* L. (licorice) were collected from Nizip District of Gaziantep in July 2014 (Figure 1).

Cuscuta species, which naturally grows on garden plants in Kilis and Gaziantep provinces, were identified as "*Cuscuta monogyna* VAHL subsp. *monogyna* VAHL" by Prof. Dr. Nazim Sekeroglu, and the herbarium specimens were kept with KHB-79 code in the Herbarium of Kilis 7 Aralik University, Department of Biology.

Chemicals

Chemicals used in the analyses were gallic acid, 2,2-diphenyl-1-picrylhydrazyl (DPPH), ferric chloride, butylated hydroxy anisole (BHA), trichloroacetic acid (TCA), acetic acid, quercetin, potassium ferricyanide, sodium phosphate, sodium carbonate, methanol, ethanol, aluminium chloride purchased from Merck® (Darmstadt, Germany), ascorbic acid from Jt Baker, and Sigma (St. Louis, MO, USA).



Fig. 1A.



Fig. 1B.

Figure 1. A. *Cuscuta monogyna* VAHL subsp. *monogyna* VAHL, B. Dodder on different host plants such as vineyard, olive, liquorice, pomegranate, and green pepper

Preparation of extracts and extraction of plants

Cuscuta species and their host plants were separated, cleaned, dried, and powdered. Each plant sample (7-15 g) was mixed with a ten-fold methanol as solvent (about 100 mL), and they were held for 24 h at room temperature by shaking. At the end of the shaking period, the mixture was filtered with filter paper, and the same procedure was repeated twice by adding solvent to the wet plant again, and the solvents were then evaporated in rotavator. The following chemical analyses were performed on the dried extracts.

Determination of the total phenolic compounds

The total phenolic contents of 15 different extracts were determined by using Folin-Ciocalteu method (Singleton & Rossi, 1965) with minor modifications (Şenol, Şekeroglu, Gezici, Kilic, Orhan, 2018; Gezici & Sekeroglu, 2019). According to the calibration curve, the total phenolic concentration was calculated as gallic acid equivalents using the absorbance of the samples at 765 nm.

Determination of the total flavonoids

To determine the total flavonoids, 2 mg mL⁻¹ of each extract sample and the reference quercetin solutions were mixed with 75% EtOH, 10% AlCl₃, 1M sodium acetate and 2,800 µL distilled water separately. All procedures and reagents were the same as those described in our previous publication (Gezici & Sekeroglu, 2019). According to the calibration curves, total flavonoids were calculated as quercetin equivalents using

the absorbance of the samples at 415 nm (Woisky & Salatino, 1998).

Determination of the radical scavenging activity

The method of diphenyl picrylhydrazyl (DPPH) was utilised to determine *the in vitro* radical scavenging activity (Blois, 1958; Lee, Chung, Chang, & Lee, 2007). The extracts were prepared in methanol with different concentrations in the range of 1 mg/mL⁻¹, 0.5 mg/mL⁻¹, 0.25 mg/mL⁻¹, 0.125 mg/mL⁻¹ to 0.0625 mg/mL⁻¹, and butylated hydroxy anisole (BHA) was used as a reference. 1 mL of each extract and BHA were mixed with 2 mL of DPPH (0.1mM in 70 % methanol) and incubated for 30 min in the dark at room temperature, and their absorbances were measured at 517 nm. The antiradical activity was calculated as follows;

$$\text{Scavenging activity (\%)} = (1 - A1/A0) \times 100$$

[A0: absorbance of control; A1: absorbance of sample]

Thin-layer chromatography

Chloroform and methanolic extracts (0.05 g) were dissolved in 5 mL of methanol. An ultrasonic bath was used to ensure complete dissolution of the extracts. On the aluminium plate, 80 µL of each extract was applied as a 0.5-cm-long line. The Chloroform: Methanol: Water (61: 32: 7) solvent system drifted up to 10 cm. The fluorescent stains were examined under ultra-violet light at 254 and 366 nm and marked. The revelator was then sprayed and exposed for 5 min at 120°C. The stains were disambiguated after this period.

Determination of the Concentration of Minerals

Preparation of the samples

The dodder, separated from the host plants, was washed with pure water and dried in the shade for 72 h. The plant material was dried again for 48 h at 70°C, and 2 mL of distilled water, 2 mL of H₂O₂ (30%) (Merck®, Darmstadt, Germany), and 4 mL of HNO₃ (65%) (Merck®, Darmstadt, Germany) were added to 0.2 g of the samples. The mixture was burned for 5 min at 200°C in a microwave (HP-500 CEM MARS 5 crop, Matthews NC, USA).

These incinerated samples were cooled to room temperature and filtered with a blue band filter paper containing pure water until the volume reached 25 mL. The extracts were stored at 4°C in polyethylene boxes until ICP-OES analysis. For accuracy, the analysis was repeated three times for each plant sample using standard reference samples (Corn bran, Standard Reference Material, 1547) supplied by NIST (International Institute of Standards and Technology). Read errors in the results of analysis were determined to be below 1% compared with the reference samples.

Preparation of the Standards

The polyethylene and glass materials used in the study were washed with 2-4% HCl before being passed through pure water three times. Merck® standards (R1 and R2 groups) were used as references. Cd, Cu, Fe, Mn, Zn, and HNO₃ were used in the analyses at a concentration of 1% in 1000 mg per stock solution.

Conditions of the ICP-OES Device

The ICP-OES device, supported by a magnetic field, atomised and stimulated in the sample, is a high-temperature plasma technique. Light is sent to the detector at the selected wavelength, and the light intensity is determined. According to the analytical method, the wavelengths of Al, B, Ca, Cd, Co, Fe, K, Mg, Mn, Mo, Na, Ni, Pb, S, and Zn were 396.152, 208.889, 370.602, 214.439, 2390.786, 238.204, 404.721, 383.829, 257.610, 203.846, 588.995, 216.555, 220.353, 181.972, and 213.857 nm, respectively. The cadmium concentration was found 0.1 µg/mL with ICP-OES (Inductively Coupled Argon Plasma-Optical Emission Spectrometer) U-5000 AT + Ultrasonic Nebuliser at 214.438 nm wavelength.

Statistical Analysis

The experiments were carried out in triplicate, and all the data are presented as the detected values ± standard deviation values (mean ± SD) for total amounts of phenolic and flavonoid substances for *Cuscuta* species and their host plants. The DPPH

free radical scavenging activity of *Cuscuta* species and their host plants is given as a percentage of inhibition.

RESULTS AND DISCUSSION

Determination of the total phenolic compounds

Total phenolic compounds were determined from methanolic extracts of *Cuscuta* species and their host plants. The total amounts of the phenolics varied in a wide range from 5.175% to 35.237% (Table 1). As presented in Table 1, the total phenolic content also showed variations depending on the host plants and their used parts. As the highest total phenolic content was found in the liquorice leaf, the lowest value was determined in the pepper branches.

Considering the extracts prepared from *Cuscuta* species by their host plants; the highest value for total phenolic content was found in the dodder grown on liquorice. The lowest total phenolic content was determined in the extracts obtained from dodders on pepper plants. According to different hosts, the total phenolic contents in dodder extracts could be classified from higher levels to lowest values as liquorice, vineyard, pomegranate, olive, and pepper, respectively.

The amounts of total phenolic substances in the leaves of host plants (liquorice, vineyard and pomegranate) were higher than those in the branches of these plants and their parasites. The total amount of phenolic compound emerged in the highest value in the extract of olive's branch, while the lowest value in the olive leaf extract despite the total phenolic content differing partially in the pepper.

When the phenolic content of *C. reflexa* and *C. europea* plants collected from different hosts (*Acacia nilotica*, *Acacia nilotica*, *Lycium barbarum*, *Azadirachta indica*, *Calotropis procera* and *Ziziphus jujuba*) was examined, the highest value was found to be 189.68 (mg/100g) in the methanolic extract of *C. europea* obtained from *Ziziphus jujuba* host plants and the lowest value was 97.68 (mg/100g) in the methanolic extract of *C. europea* collected from *Calotropis procera* host plant (Perveen, Hussain Bukhari, Qurat-Ul-Ain, Kousar, & Rehman, 2013).

Determination of the total flavonoid compounds

Total flavonoid compounds were determined in the methanolic extracts of *Cuscuta* species and their host plants. The total amount of flavonoid compounds varied between 0.308% and 15.731% (Table 1). The highest total phenolic content was found in the vineyard leaf compared with that of the others.

According to the data given in Table 1, the total phenolic content was found to be the highest value in vineyard leaves by comparing that of the other samples investigated. On the other hand, the lowest total flavonoid content was detected in the *Cuscuta* extracts obtained from the pomegranate branches (0.308

Table 1. Total amounts of phenolic and flavanoid substances of *Cuscuta* species and their host plants

Host plants	Parts of the plant	Total phenolic substances (%) ^{a,b}	Total flavonoid substances (%) ^{b,c}
Liquorice	Leaf	35.238 ± 1.096	7.638 ± 0.694
	Branch	11.438 ± 0.247	0.812 ± 0.145
	Dodder	27.138 ± 0.141	3.158 ± 0.183
Vineyard	Leaf	33.363 ± 0.282	15.731 ± 0.318
	Branch	25.713 ± 0.919	3.301 ± 0.694
	Dodder	26.388 ± 0.035	5.797 ± 0.463
Olive	Leaf	14.138 ± 0.494	2.667 ± 0.048
	Branch	29.013 ± 1.025	4.106 ± 0.019
	Dodder	19.700 ± 1.007	4.419 ± 0.019
Green pepper	Leaf	5.175 ± 0.265	4.297 ± 0.135
	Branch	6.180 ± 0.017	1.945 ± 0.068
	Dodder	7.237 ± 0.141	2.933 ± 0.135
Pomegranate	Leaf	14.100 ± 0.583	1.917 ± 0.087
	Branch	28.175 ± 0.583	0.308 ± 0.068
	Dodder	23.600 ± 1.361	2.845 ± 0.106

^a mg equivalent of gallic acid (GAE) per g of extract.

^b SD: Standard deviation (n=3).

^c mg equivalent of quercetin (QE) per g of extract.

± 0.068), followed by liquorice (0.812 ± 0.145). The variation in the total flavonoid content of the host plant parts varied considerably. The highest values were found in the leaves of vineyard (15.731 ± 0.318), which is followed by liquorice and peppers (7.638 ± 0.694 and 4.297 ± 0.135, respectively). Although there is limited host plant-dodder comparison in terms of flavonoid content, there are studies evaluating the flavonoid content of different *Cuscuta* spp. seeds. In one study, it was shown that 80% methanolic extract of *C. reflexa* seed contains high flavonoids (Noureen et al., 2018). In a study examining *C. campestris* seeds, which are the hosts of thyme, basil, and onion, the highest flavonoid content was found in onion (Ramezan et al., 2023).

DPPH Free Radical Scavenging Activity

The DPPH free radical scavenging activity of methanolic extracts of *Cuscuta* species and their host plants was determined as a concentration-dependent variable (Table 2). The highest free radical scavenging activity was found at 1 mg/mL⁻¹ dose of extract obtained from dodder on olive, whereas the lowest activity was found at 0.125 mg/mL⁻¹ dose of extract prepared from dodder on pepper.

It was determined that the free radical scavenging activity of extracts obtained from *Cuscuta* species and their host plant's parts showed variation in accordance with doses of the extracts. As seen in Table 2, the highest antioxidant capacity was found in the olive, whereas the lowest effect was found in the pepper at 1 mg/mL⁻¹ concentration.

Even though a remarkable variation was not observed among the extracts obtained from different host plants and their parts,

extracts of *Cuscuta* species from the olive, liquorice, and vineyard showed higher DPPH scavenging activity than the others, even at lower concentrations. When antioxidant capacities of dodder were examined according to host plants, it was stated from high DPPH inhibition to low as olive'dodder>vineyard'dodder ≥ pomegranate'dodder ≥ liquorice'dodder >pepper'dodder.

In our study, the highest activity was found in the concentration of 1 mg/mL⁻¹ solution of branch of olive and liquorice at 95.56%, and the lowest activity was found in the concentration of 0.125 mg/mL⁻¹ in the leaf of pepper at 87.30% among the plant parts. The highest antioxidant activity was detected in the branches of each host plant, except for pepper. It is observed that the host plants and dodders have the highest activity at a dose of 1 mg/mL⁻¹ and the lowest activity at a dose of 0.125 mg/mL⁻¹. The data obtained from the presented work were found to be in agreement with results reported by Koca et al. (2011) previously. Yen et al. (2007) also reported that ethanolic extracts of *Cuscuta chinensis* had hepatoprotective properties because of their high antioxidant capacities. Methanolic and n-hexane extracts of *C. reflexa* and *C. europea* from different host plants had different antioxidant capacities as DPPH (Perveen et al., 2013).

Thin-layer chromatography

Thin layer chromatography allows comparison of plants and hosts in terms of their phytochemical content. Because of this preliminary study, by ultraviolet spectroscopy, bluish-greenish and dark fluorescence belong to flavonoids or coumarin; and the red fluorescence at the upper part belongs to chlorophyll. After the vanillin-sulphuric acid revelator, which is used to di-

Table 2. DPPH free radical scavenging activity of *Cuscuta* species and their host plants

<i>Cuscuta</i> parts of the host plants	1 mg/mL ⁻¹	0.5 mg/mL ⁻¹	0.25 mg/mL ⁻¹	0.125 mg/mL ⁻¹	0.0625 mg/mL ⁻¹
VL (Vineyard Leaf)	93.65 ± 0.03	87.94 ± 0.19	87.84 ± 0.12	87.62 ± 0.21	53.55 ± 0.31
VB (Vineyard Branch)	94.94 ± 0.06	94.60 ± 0.06	94.29 ± 0.09	93.65 ± 0.04	72.99 ± 0.11
VD (Vineyard Dodder)	94.60 ± 0.05	94.29 ± 0.08	94.19 ± 0.08	94.09 ± 0.05	67.77 ± 0.24
GPL (Green Pepper Leaf)	92.70 ± 0.05	92.06 ± 0.13	90.48 ± 0.05	87.30 ± 0.14	37.60 ± 0.36
GPB (Green Pepper Branch)	90.16 ± 0.11	87.94 ± 0.17	79.37 ± 0.25	75.24 ± 0.18	22.59 ± 0.45
GPD (Green Pepper Dodder)	93.33 ± 0.04	88.25 ± 0.21	80.32 ± 0.17	57.78 ± 0.29	40.76 ± 0.27
LL (Liquorice Leaf)	94.29 ± 0.07	93.65 ± 0.09	93.33 ± 0.06	93.02 ± 0.05	54.50 ± 0.27
LB (Liquorice Branch)	95.56 ± 0.08	94.29 ± 0.05	93.97 ± 0.03	92.38 ± 0.07	75.04 ± 0.10
LD (Liquorice Dodder)	94.60 ± 0.03	93.97 ± 0.10	93.65 ± 0.05	93.33 ± 0.03	56.08 ± 0.26
PL (Pomegranate Leaf)	94.60 ± 0.02	93.65 ± 0.07	93.02 ± 0.05	91.43 ± 0.08	56.08 ± 0.21
PB (Pomegranate Branch)	95.24 ± 0.05	94.60 ± 0.05	93.65 ± 0.08	91.11 ± 0.06	75.20 ± 0.14
PD (Pomegranate Dodder)	94.60 ± 0.04	93.97 ± 0.06	87.30 ± 0.12	86.67 ± 0.11	42.97 ± 0.13
OL (Olive Leaf)	95.24 ± 0.09	94.60 ± 0.07	94.29 ± 0.08	93.97 ± 0.03	82.46 ± 0.09
OB (Olive Branch)	95.56 ± 0.09	93.97 ± 0.09	93.65 ± 0.04	88.57 ± 0.05	82.46 ± 0.07
OD (Olive Dodder)	95.87 ± 0.04	95.60 ± 0.05	94.60 ± 0.09	93.65 ± 0.07	75.20 ± 0.14
BHA ^b	94.29 ± 0.07	93.65 ± 0.03	92.70 ± 0.04	89.21 ± 0.09	87.68 ± 0.11

^a Values are presented as percentages of inhibition

^b Butylated hydroxy anisole; commercial standard

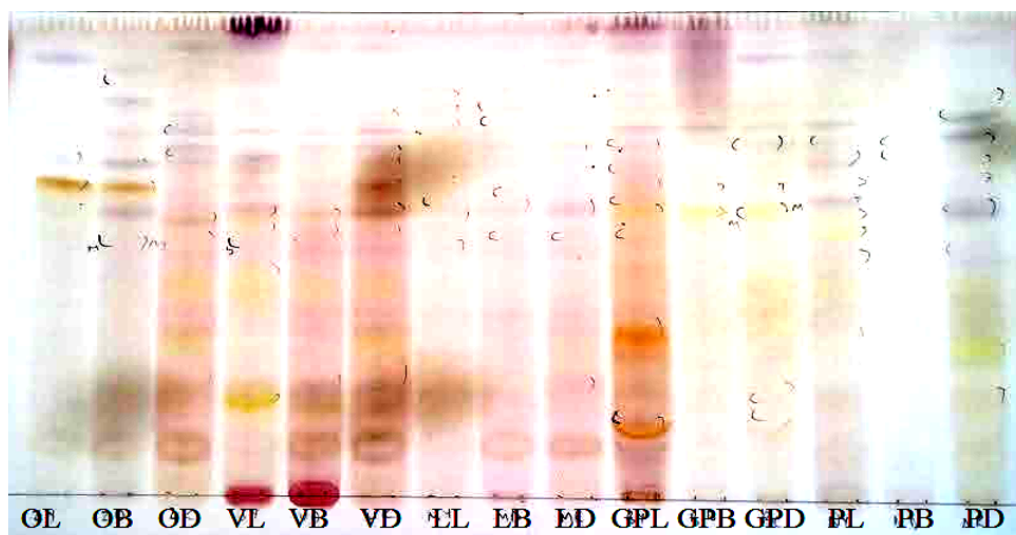
agnose metabolites such as steroids, highly structured alcohols, phenols, volatile oils, and terpenic compounds, was sprayed, it is thought that the pink purple spots belong to diterpenes, the yellowish spots belong to flavonoids or other phenolic compounds; orange colours belong to alkaloids or iridoid. It has been observed that compounds thought to be flavonoid derivatives are more concentrated in the host plant, olive, grape and green pepper, and are not taken up as much in dodder. On the contrary, it is thought that terpenic compounds are found more in cuscutea, which is a semi-parasitic plant, according to the TLC plate results (Figure 2).

Mineral Analysis

The amounts of 14 different minerals including aluminium, boron, calcium, cadmium, copper, iron, potassium, magnesium, manganese, sodium, nickel, phosphorus, sulphur, and

zinc, were determined in the leaves, branches, and doddres of green pepper, grape, liquorice, pomegranate, and olive plants, and *Cuscuta* species on these plants. The mineral contents of *Cuscuta* species and their host plants were summarised in Table 3.

The aluminium (Al) contents of plants showed a wide variation between 28 and 747 mg/kg. The highest value was determined in the leaves of vineyard and the lowest value was determined in the branches of pomegranate according to plant parts. Aluminium concentrations of dodder samples collected from different plant species ranged from 152 to 285 mg/kg. The highest aluminium content was found in the pomegranate dodder, and the lowest value was found in the liquorice dodder. It was determined that the boron (B) minerals of *Cuscuta* species and parts of their host plants were in the range of 16-92 mg/kg in this study (Table 3). The amount of boron in the plant parts was found in the lowest branches of the pepper, and the



Olive Leaf (OL), Olive Branch (OB), Olive Dodder (OD), Vineyard Leaf (VL), Vineyard Branch (VB), Vineyard Dodder (VD), Liquorice Leaf (LL), Liquorice Branch (LB), Liquorice Dodder (LD), Green Pepper Leaf (GPL), Green Pepper Branch (GPB), Green Pepper Dodder (GPD), Pomegranate Leaf (PL), Pomegranate Branch (PB), and Pomegranate Dodder (PD).

Figure 2. Plate appearance after vanillin-sulphuric acid revelator spraying

Table 3. Mineral values of *Cuscuta* species and their host plants (mg/kg)

Cuscuta/ Plant ^a	Minerals (mg/kg) ^b													
	Al	B	Ca	Cd	Cu	Fe	K	Mg	Mn	Na	Ni	P	S	Zn
VL	747 ± 66	48 ± 1.3	33237 ± 658	0.07 ± 0.00	3.88 ± 0.05	140 ± 5.8	3817 ± 298	6205 ± 231	63.1 ± 2.0	208 ± 8.0	5.09 ± 0.30	1025 ± 98	1527 ± 65	16.3 ± 2.80
VB	117 ± 6	25 ± 0.8	8979 ± 203	0.17 ± 0.02	6.49 ± 0.02	139 ± 4.1	8694 ± 245	2350 ± 125	19.6 ± 0.8	214 ± 8.1	1.04 ± 0.10	773 ± 42	746 ± 47	21.9 ± 3.70
VD	206 ± 14	43 ± 2.1	3847 ± 125	0.02 ± 0.00	4.83 ± 0.06	220 ± 6.3	11508 ± 356	1043 ± 98	10.4 ± 0.3	129 ± 5.2	1.29 ± 0.20	990 ± 65	784 ± 32	3.9 ± 0.50
GPL	563 ± 42	92 ± 3.3	32868 ± 496	0.44 ± 0.07	22.76 ± 2.10	151 ± 3.0	26458 ± 375	1050 ± 88	132.1 ± 26	109 ± 2.5	10.01 ± 0.80	1776 ± 47	6568 ± 84	19.9 ± 0.40
GPB	277 ± 8	16 ± 0.4	9260 ± 236	0.18 ± 0.05	8.48 ± 0.08	265 ± 2.7	17314 ± 425	2350 ± 159	16.7 ± 0.8	121 ± 7.0	3.70 ± 0.10	662 ± 66	3018 ± 29	5.0 ± 0.90
GPD	157 ± 12	12 ± 0.5	3311 ± 125	0.09 ± 0.03	9.37 ± 1.10	190 ± 3.2	24721 ± 541	2445 ± 287	15.9 ± 1.4	118 ± 3.2	4.22 ± 0.30	2465 ± 75	2237 ± 54	13.9 ± 2.60
LL	268 ± 15	32 ± 2.3	17789 ± 326	0.05 ± 0.00	4.01 ± 0.02	237 ± 2.6	12524 ± 453	2644 ± 178	61.5 ± 1.4	74 ± 0.4	2.91 ± 0.40	785 ± 37	2044 ± 49	12.8 ± 1.80
LB	65 ± 5	16 ± 1.1	14045 ± 287	0.01 ± 0.00	3.10 ± 0.02	68 ± 0.8	8451 ± 125	1217 ± 101	12.7 ± 2.8	101 ± 0.9	1.62 ± 0.10	441 ± 24	881 ± 37	8.6 ± 0.50
LD	152 ± 11	26 ± 0.6	5115 ± 106	0.03 ± 0.00	4.50 ± 0.06	157 ± 2.4	13864 ± 264	1048 ± 76	19.3 ± 3.2	59 ± 0.8	4.22 ± 0.60	1208 ± 41	913 ± 19	15.3 ± 1.20
PL	145 ± 7	26 ± 0.8	17385 ± 456	0.05 ± 0.00	4.57 ± 0.01	147 ± 1.1	18351 ± 279	2434 ± 245	19.4 ± 1.1	48 ± 1.3	2.18 ± 0.30	1463 ± 32	1428 ± 47	19.3 ± 3.40
PB	28 ± 3	20 ± 0.5	10771 ± 258	0.01 ± 0.00	5.44 ± 0.02	32 ± 0.2	7332 ± 357	893 ± 65	8.6 ± 0.5	63 ± 1.1	1.69 ± 0.30	1092 ± 71	596 ± 39	18.1 ± 3.80
PD	285 ± 9	97 ± 2.5	4074 ± 125	0.06 ± 0.01	7.43 ± 0.04	217 ± 3.4	23173 ± 519	1226 ± 98	14.6 ± 0.7	57 ± 0.9	3.80 ± 0.20	2965 ± 59	1546 ± 40	19.4 ± 2.60
OL	645 ± 38	23 ± 0.2	16477 ± 546	0.05 ± 0.01	5.32 ± 0.03	55 ± 0.9	12176 ± 129	2123 ± 106	67.2 ± 3.7	129 ± 3.0	6.34 ± 0.70	1000 ± 47	1432 ± 41	11.7 ± 2.00
OB	83 ± 5	23 ± 0.2	6715 ± 312	0.02 ± 0.00	6.93 ± 0.02	87 ± 0.4	11750 ± 264	6205 ± 231	12.4 ± 0.8	153 ± 2.0	0.83 ± 0.10	845 ± 28	548 ± 38	7.8 ± 1.40
OD	282 ± 11	37 ± 0.4	3608 ± 211	0.03 ± 0.00	7.48 ± 0.03	145 ± 1.9	13233 ± 340	2350 ± 125	15.0 ± 1.0	164 ± 5.6	2.24 ± 0.40	1076 ± 54	834 ± 17	8.2 ± 1.00

^a Vineyard Leaf (VL), Vineyard Branch (VB), Vineyard Dodder (VD), Green Pepper Leaf (GPL), Green Pepper Branch (GPB), Green Pepper Dodder (GPD), Liquorice Leaf (LL), Liquorice Branch (LB), Liquorice Dodder (LD), Pomegranate Leaf (PL), Pomegranate Branch (PB), Pomegranate Dodder (PD); Olive Leaf (OL), Olive Branch (OB), Olive Dodder (OD).

^b Values are expressed as means ± standard deviation.

highest in the leaves of the green pepper. The boron minerals in the *Cuscuta* species, which are full parasites that grow on host plants, range from 12 to 97 mg/kg.

It was found that the highest boron value was in *Cuscuta* sp. collected from green pepper, and the lowest boron value was in *Cuscuta* sp. collected from pomegranate.

The amount of calcium (Ca) showed a variance in the range of 6715-33237 mg/kg (Table 3). The highest calcium value was observed in the leaf of the vineyard, whereas the lowest value was found in the olive branch. Calcium found in the dodders

showed variance in the range of 3311-5115 mg/kg. As the lowest value was found in the green pepper dodder, the highest amount of calcium was found in the liquorice dodder.

Heavy metal cadmium (Cd), which limits plant growth, blocks and slows down the physiological processes of plants, and reduces their productivity, was determined in host plants and their dodders. Higher doses and long-term exposure to cadmium can cause plant death (Temmerman, Bouma, Govers, & Lauwaet, 2005; Marschner, 1995). World Health Organisation (WHO) explained that the limit value of Cd for medicinal plants

is 0.300 mg/kg (Şekeroğlu, Ozkutlu, Kara, & Ozguven, 2008). In parts of the plant, the highest value of cadmium was found to be 0.44 mg/kg in green pepper's leaf and the lowest value was found to be 0.01 mg/kg in pomegranate's leaf. While the lowest value (0.03 mg/kg) was found in the liquorice's dodder and in the olive's dodder, the highest value (0.09 mg/kg) was found in the green pepper's dodder (Table 3).

Copper (Cu) mineral in the dodder on host plants was determined to be in the range of 4.50–9.37 mg/kg. While the highest value was found in the green pepper dodder; the lowest value was found in the liquorice dodder. In host plants, copper was found in the range of 3.10–22.76 mg/kg (Table 3). It is stated that the lowest amount was observed in the branch of liquorice, and the highest amount in the green pepper's leaf.

Iron (Fe) in plants is one of the important elements required for chlorophyll synthesis. Iron acts not only as a cofactor for peroxidase and some other enzymes but also in the content of ferredoxin and cytochromes (Ozen and Onay, 2007). The iron mineral content in the dodders varied in the range of 145–220 mg/kg. As the highest iron value was found in the vineyard dodder, the lowest value was determined in the olive dodder. In the host plants, the iron mineral content varied from 32 to 265 mg/kg over a wide range. Among the host plants, the highest iron content was in the branch of green pepper, whereas the lowest iron content was in the branch of pomegranate (Table 3). It can be seen that the iron values obtained in this study are considerably lower than those in previous studies. Although it varies according to different parts of the plant, it is estimated that the low iron ratio is caused by the genetic structure and growth conditions of the plant. It plays a role in the opening and closing of plant stomata, the synthesis of starch, the activation of some enzymes, and the regulation of osmotic pressure (Basgel & Erdemoglu, 2006).

The content of potassium (K) ranged from 3817 to 26458 mg/kg in the plant parts. While the highest value was found in the green pepper's leaf, the lowest value was found in the vineyard's leaf. It was found that the highest potassium value (24701 mg/kg) was in the green pepper's dodder, and the lowest potassium value (11508 mg/kg) was found in the vineyard's dodder (Table 3). The potassium values determined in this study are in agreement with the results of previous studies on this base (Sekeroğlu, Ozkutlu, Devci, Dede, & Yilmaz., 2006; Koca, Özkutlu, Şekeroğlu, 2009; Sekeroğlu, Ozkutlu, & Kilic, 2017).

Magnesium (Mg) is an essential element in the basic structure of chlorophyll and is necessary for the synthesis of many enzymes and proteins (Ozen & Onay, 2007). Magnesium content varied from 837 to 2445 mg/kg in the present study (Table 3). The highest value was found in green pepper plants and the lowest value in olive plants. Magnesium values obtained from parts of the plant showed variance in the range of 893–6205 mg/kg. The branch of pomegranate had the lowest magnesium content, whereas the leaf of vineyard had the highest magne-

sium mineral content. The detected magnesium values in the analyzed plant samples were in the normal ranges declared by FAO/WHO (1993) and WHO (1999), and these values are similar to those reported in previous studies (Akgunlu et al., 2016; Sekeroglu et al., 2017). Manganese (Mn) is important for photosynthesis and necessary for electron transfer (Özen and Onay, 2007). While the highest value (132.1 mg/kg) was found in the green pepper leaf, the lowest value (8.6 mg/kg) was found in the pomegranate branch. It was determined that the highest manganese value (19.3 mg/kg) in the liquorice dodder, the lowest manganese value (10.4 mg/kg) in the vineyard's dodder (Table 3). Similar results were reported for investigated medicinal and aromatic plants by Sekeroglu et al. (2006) and Basgel and Erdemoglu (2006).

With regard to host plants, the amount of sodium (Na) mineral was found the highest (214 mg/kg) in the leaf of vineyard, which was found the lowest (48 mg/kg) in the leaf of pomegranate. In addition, the highest sodium value (164 mg/kg) was observed in the olive dodder, whereas the lowest sodium value (57 mg/kg) was observed in the pomegranate dodder. Sodium values obtained in this study were similar to those reported for different plants in previous studies (Akgunlu et al., 2016). Lower or higher values obtained in some plants are thought to be due to genetic structures, growth conditions, and different parts of the plants (Ozkutlu, Kara, & Şekeroğlu, 2007; Sekeroglu et al., 2006).

As for nickel (Ni), it helps to provide the absorption of iron and the germination of seeds of plants. It was found that the highest Ni value (4.22 mg/kg) in the green pepper dodder and liquorice dodder, and the lowest Ni value (1.29 mg/kg) was found in the vineyard dodder. In host plant parts, the amount of Ni mineral was identified as the highest (10.01 mg/kg) in the leaf of green pepper and the lowest (0.83 mg/kg) in the branch of olive. According to a report released by FAO/WHO, the permitted Ni limit for edible parts of plants is 1.63 mg/kg (FAO/WHO, 1984). Our results showed that the detected Ni levels were lower in wild crops and their hosts. When an agricultural system with chemicals is applied, Ni contents may increase by chemical uptake.

In the host plant's parts, the amount of phosphorus (P) mineral was found the highest (1776 mg/kg) in the leaf of green pepper, and that was the lowest (441 mg/kg) in the branch of liquorice. Furthermore, the highest P mineral value (2965 mg/kg) was determined in the pomegranate dodder, which was found to be the lowest (990 mg/kg) in the vineyard dodder. According to the analysis results, the highest P mineral values were detected in the leaves of the host plants and in the pomegranate dodder. According to sulphur (S) mineral content, it was found to be the highest with a value of 2237 mg/kg in the green pepper's dodder. In host plant parts, the amount of S mineral was determined to be the highest (6568 mg/kg) in the leaf of green pepper and the lowest (548 mg/kg) in the branch of

olive (Table 3). As previously reported, the sulphur source can be absorbed by plants as the elemental sulphur, SO_4^{2-} ions, SO_2 gas, and mineral matter contained in organic substances. This mineral plays a role in the composition of plant proteins, in the formation of chlorophyll, and in the content of some vitamins (Sekeroğlu et al., 2018).

Zinc (Zn) plays an important role in pollen formation and auxin synthesis. It also preserves the ribosome structure (Özen and Onay, 2007). The amount of Zn mineral was found in the range of 3.9-19.4 mg/kg in the dodder collected from the host plants. The highest Zn mineral value was determined in the pomegranate's dodder, when the lowest Zn value was found in the vineyard's dodder. With respect to host plant parts, the amount of Zn mineral was observed the highest (21.9 mg/kg) in the branch of vineyard and that was the lowest (5.0 mg/kg) in the branch of green pepper (Table 3).

According to the analysis results, the mineral concentrations of the dodders and their host plants were highly variable. This variation could be explained by the amount of minerals reaching the plant body and tissues. Moreover, this variability could result in the genetic structure and ecological and growing conditions of the investigated plants. Dodder mineral compositions are directly related to their host plants behaviour.

CONCLUSIONS

However, there has been a significant variation in the total polyphenolic contents of the extracts of *Cuscuta monogyna* VAHL subsp. *monogyna* VAHL plant parts and their host plants. The total amount of phenolic substance was observed to be the highest in the leaf of liquorice extract, which was closely followed by the leaf of vineyard, and the total amount of flavonoids was also determined to be the highest in the leaf of vineyard extract. According to the antioxidant capacity, almost all the extracts obtained from the host plants were found to have remarkable radical scavenging activities on DPPH. As for the thin layer chromatographic method, the general chemical profiles in terms of secondary metabolites, phenolic and terpenic compounds, especially flavonoids of dodders and host plants, were similar. In addition to the determination of the general chemical profiles, the morphogenetic variability of Al, B, Ca, Cd, Cu, Fe, K, Mg, Mn, Na, Ni, P, S, and Zn minerals was determined in this study.

It was concluded that the chemical content of *Cuscuta* species and its biological activity are closely related to the host plant because of parasitism. Despite its harmful effect on the parasitic direction, the results obtained from our laboratory studies could offer a positive scientific insight to obtain the desired and effective compounds from the plant. To the best of our knowledge, this is the first assessment conducted on *Cuscuta* species to reveal their interactions with host plants in Turkey

Viscum album L., which although semi-parasitic, has signif-

icant biological activity and phytochemical content, inspired this study. This preliminary study comparing the parasitic *Cuscuta* species with the host plant is considered to be a guide for future research. Further studies on different host plants and *Cuscuta* species are required.

Peer-review: Externally peer-reviewed.

Author Contributions: Conception/Design of Study- G.K.Y., N.Ş.; Data Acquisition- G.K.Y., C.D., U.K.Ç.; Data Analysis/Interpretation- G.K.Y., C.D.; Drafting Manuscript- S.G.; Critical Revision of Manuscript- N.Ş.; Final Approval and Accountability- G.K.Y., C.D., U.K.Ç., S.G., N.Ş., F.Ö.

Conflict of Interest: The authors have no conflict of interest to declare.

Financial Disclosure: The authors declared no financial support.

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How cite this article

Şekeroğlu, N., Korkmaz Yaşar, G., Koca Çalışkan, U., Dönmez, C., & Gezici, S., & Özkutlu, F. (2024). Comparison of polyphenolic content, radical scavenging activity, and mineral concentrations of *Cuscuta monogyna* VAHL on different host plants. *İstanbul Journal of Pharmacy*, 54(2), 154–164. DOI: 10.26650/IstanbulJPharm.2024.1071442