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Research Article

Antimicrobial and Antioxidant Properties of Coriander (*Coriandrum sativ*um L.), Dill (*Anethum graveolens* L.) and Purslane (*Portulaca oleracea* L.) Extracts Prepared with Different Types of Solvent

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Abstract

In the study, the antimicrobial effect of ethanol: water (1:1 v/v), methanol: water (1:1 v/v) and water extracts of coriander (*Coriandrum sativum* L.), dill (*Anethum graveolens* L.) and purslane (*Portulaca oleracea* L.) plants were determined by well diffusion method and antioxidant activity by DPPH• radical removal method. As test microorganisms, *Aspergillus niger* mold and *Salmonella enterica* subsp. *enterica* serovar Typhimurium ATCC 14028, *Escherichia coli* O157:H7, *Staphylococcus aureus* ATCC 25923 bacteria were used, and the inhibition zone was measured only in *S. aureus* ATCC 25923. In DPPH• radical scavenging analysis, the antioxidant capacity of the samples was lower than the controls, and the IC50 values of Trolox, BHA, dill, coriander and purslane were 41.63 µg/mL, 154.15 µg/mL, 683.45 µg/mL, 903.33 µg/mL, 525.99 µg/mL, respectively. Among the plants studied, purslane had the highest antioxidant activity, while coriander had the lowest antioxidant activity. As a result, it was determined that the highest antioxidant and antimicrobial activity values belonged to the purslane plant.

Keywords: Coriander, dill, purslane, antimicrobial activity, antioxidant activity

Farklı Çözücü Tipleriyle Hazırlanan Kişniş (*Coriandrum sativum* L.), Dereotu (*Anethum graveolens* L.) ve Semizotu (*Portulaca oleracea* L.) Ekstraktlarının Antimikrobiyal ve Antioksidan Özellikleri

Öz

Yapılan çalışmada kişniş (*Coriandrum sativum* L.), dereotu (*Anethum graveolens* L.) ve semizotu (*Portulaca oleracea* L.) bitkilerinin etanol: su (1:1 v/v), metanol: su (1:1 v/v) ve su ekstraktlarının kuyu difüzyon metodu ile antimikrobiyal etkisi ve DPPH radikali giderme yöntemi ile antioksidan aktivitesi belirlenmiştir. Test mikroorganizmaları olarak *Aspergillus niger* küfü, *Salmonella enterica* subsp. *enterica* serovar Typhimurium ATCC 14028, *Staphylococcus aureus* ATCC 25923 ve *Escherichia coli* O157:H7 bakterileri kullanılmış ve sadece *Staphylococcus aureus* ATCC 25923'da inhibisyon zonu ölçülmüştür. DPPH• radikal giderme analizinde örneklerin antioksidan kapasitesi kontrollere göre düşük ve Trolox, BHA, dereotu, kişniş ve semizotunun IC50 değerleri sırasıyla 41,63 µg/mL, 154,15 µg/mL, 683,45 µg/mL, 903,33 µg/mL, 525,99 µg/mL olarak belirlenmiştir. Çalışılan bitkiler arasından semizotu en yüksek antioksidan aktiviteye, kişniş ise en düşük antioksidan aktiviteye sahiptir. Sonuç olarak en yüksek antioksidan ve antimikrobiyal aktive değerlerinin semizotu bitkisine ait olduğu belirlenmiştir.

Anahtar Kelimeler: Kişniş, dereotu, semizotu, antimikrobiyal aktivite, antioksidan aktivite

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1. Introduction

Plants are important as raw material sources in the development of new therapeutic agents due to the various chemical components in their content [1,2]. World Health Organization (WHO) reported that, approximately 21,000 plant species are used for medicinal function in the world. Secondary metabolites such as flavonoids, tannins, coumarins, alkaloids, and terpenes formed in various parts of plants have many positive effects on human health as well as antimicrobial and antioxidant activity [1, 3, 4, 5]. Some plants help prevent foodborne diseases by inhibiting microorganisms that can be found in foods and cause various foodborne diseases [6, 7]. Medicinal plants have become a focal point in the research of new antimicrobial products owing to their therapeutic properties, less toxicity and prices [1]. In addition, the genetic resistance of microorganisms to various drugs and the transfer of this resistance genes to other microorganisms have recently increased the interest of researchers in these plants.

Another factor that threatens human health is the oxidative stress caused by harmful components known as free radicals in the body. As a result of oxidative stress, serious diseases like cancer, hypertension, heart failure and asthma can occur. Oxidation also occurs in fatty foods, causing various quality deterioration and the production of toxic oxidation components. Synthetic antioxidants like butylated hydroxyanisole (BHA), propyl gallate, and butylated hydroxytoluene (BHT) are widely used to prevent oxidation in foods. However, the reliability of synthetic antioxidants has recently been argued due to hepatotoxicity, carcinogenic and mutagenic effects. In general, natural antioxidants are good alternatives to reduce oxidative stress since they have properties such as biodegradation and low toxicity [1, 8, 9].

Coriander (*Coriandrum sativum* L.), which is used both as a vegetable and as a spice, is an annual medicinal plant belonging to the Umbelliferae/Apiceae family [10, 11, 12]. It contains 0.2–1.5% essential oil in its fruits, but this rate can reach up to 2.7% in some varieties. Its essential oil mainly contains linalool and it also contains *y*-terpinene, *a*-pinene, geraniol, geranyl acetate and camphor. Due to the active components in coriander, it is used in cosmetics, perfumery and as a conventional medicine in the treatment of various diseases [13, 14]. Coriander has antimicrobial, antioxidant, antifungal, hypolipidemic, anticonvulsant, hypocholesterolemic, antipyretic, analgesic, sedative and diuretic properties [11, 15].

Dill (*Anethum graveolens* L.**)**, an annual herb that grows in Asia, Europe, and Mediterranean region, belongs to the genus *Anethum*, family Apiaceae/Umbelliferae. It has also been used in Ayurvedic medicine for the treatment of various gastrointestinal diseases, as a carminative, diuretic, and also for its antifungal, antibacterial, hypoglycemic, antispasmodic, and antisecretory effects. Authors reported that dill contains alkaloids, tannins, anthocyanins and flavanols [16, 17].

Purslane (*Portulaca oleracea* L.) is an annual herb belonging to the family of *Portulaceae*, which is widely found in India and the Mediterranean region [18]. It has an antioxidant effect due to β-carotene, vitamin C, vitamin E and high amount of omega-3, α-linolenic acid, linolenic acid, oxalic acid and other compounds in its content [18, 19]. In addition, purslane has pharmaceutical properties such as anti-carcinogenic, anti-cardiovascular and antidiabetic [19].

In the present study, it was aimed to investigate the antimicrobial effects and antioxidant activities of extracts obtained from dried coriander, dill and purslane plants using different types of solvents.

2. Material and Method

2.1 Preparation of Plant Extracts

Fresh coriander (*Coriandrum sativum* L.), purslane (*Portulaca oleracea* L.), and dill (*Anethum graveolens* L.) plants were obtained from local markets of Erzurum city Türkiye. After being brought to the laboratory, the above-ground parts were thoroughly cleaned and rinsed with pure water. The plants were evenly dissected and dried at room conditions. After the plants dried, they were grinded and stored at +4 ℃. The plant samples were weighed (5 g) and ethanol: water (1:1 v/v), methanol: water (1:1 v/v) and water solvents (50 mL) were added. Samples were kept in a shaking water bath (JSSB-30T, JSR, Korea) at 90 rpm at 40 ℃ for 20 hours. The mixture was filtered through general purpose filter paper, and obtained supernatant was centrifuged at 4500 rpm for 15 minutes. The solvents were then evaporated by a vacuum rotary evaporator (Heidolph Laborota 4000 Efficient, Germany) at 40 °C. The samples were then dried by lyophilization. Dry samples were kept in the dark at -20°C. The yield of all extracts was calculated according to the following equation [20].

$$\text{Extraction Yield (\%)} = (W1/W2) \text{ X } 100$$

W1; Weight after extraction, W2; Total weight before extraction

2.2 Antimicrobial Activity of Plant Extracts

Aspergillus niger, *Salmonella enterica* subsp. *enterica* serovar Typhimurium ATCC 14028, *Staphylococcus aureus* ATCC 25923, and *Escherichia coli* O157:H7 were obtained from Atatürk University Food Engineering Department Culture Collection (AUFECC). Antimicrobial activity of plant extracts was determined using the agar well diffusion method [21, 22]. Test bacteria were growth on Nutrient Agar (NA, Merck, Darmstadt, Germany) and incubated at 30°C for 24-48 hours, mold was incubated on Potato Dextrose Agar (PDA, Merck, Darmstadt, Germany) at 30°C for 72-96 hours [23, 24, 25]. Fresh overnight 0.5 McFarland (~10^8 CFU/ml) cultures of bacteria and mold (100 µL) were spread onto NA and PDA respectively and wells (5 mm) were made with a sterile pipette tip. Filter sterilized 10% DMSO (Dimethyl sulfoxide) was used to dissolve the all plant extracts [26, 27, 28]. Each well was loaded with 100 µL of 120 mg/mL extract solution [29]. Extracts were allowed to dry by keeping them at room temperature for 1 hour. Bacteria were incubated at 30°C for 24 hours and mold at 30°C for 72 hours [21, 22].

Following the incubation, the inhibition zones formed on the medium were measured in millimeters (mm). Antibiotics/antifungals were used as positive control (Gentamicin (10 µg) for *E. coli* O157:H7, Ampicillin (10 µg) for *S. aureus* and *S.* Typhimurium, and Amphotericin B (5 µg) for *A. niger* and 10% DMSO was used as negative control [23].

2.3 Antioxidant Capacity by 1, 1-diphenyl-2-picrylhydrazyl (DPPH•) of Plant Extracts

Extract samples were weighed at a concentration of 100, 300, and 600 µg/mL and put into test tubes. DPPH• solution (1 mM) prepared with ethanol was used as free radical. After transferring 0.3 mL of DPPH• solution to each test tube, the total volume was made up to 3 mL with ethanol. Samples were kept in the dark conditions for 30 min at room temperature. Absorbance were measured with a spectrophotometer (PG Instruments Ltd., Lutterworth, United Kingdom) at a wavelength of 517 nm versus a blank. Samples were compared with the standard antioxidants BHA and Trolox. The control consists of 0.3 mL of DPPH• and 2.7 mL of ethanol solution [30]. The inhibition % was calculated using the absorbance values with the following formula [31, 32, 33].

$$\text{DPPH• radical scavenging activity (\%)} = [(A_{control}-A_{sample}) / A_{control}] \times 100$$

Results are expressed as the IC_{50} value. The IC_{50} value is the amount of antioxidant necessary to reduce the DPPH• concentration by 50% [34]. The results were expressed as µg on dry weight basis.

2.4 Statistical Analysis

All microbiological and chemical analyses were carried out on three parallels. Data were obtained as mean ± standard deviation. The data were evaluated by ANOVA using the IBM SPSS Statistics, version 20.0 (IBM Corporation, New York, USA) and the means were compared by Duncan test for multiple comparisons at a 0.05 significance level.

3. Results and Discussion

3.1 Extraction Yield

Extract abbreviations and extraction yield are depicted in Table 1. There was no statistical difference ($p>0.05$) between the extraction yields of the samples. Solvent viscosity, dipole moment, polarity and dielectric constant can affect the rate of diffusion and extraction. Solvents with high dielectric constant (δ) also have high polarity. Since the dielectric constant of water is higher than ethanol and methanol, the extraction efficiency is also higher [35]. Extraction efficiency is also affected by particle size, nature of phytochemicals and method [36]. Nile et al. [37] reported the extraction yield of ethanol extract obtained from purslane plant by maceration method as 9.6 ± 0.50 g/100 g. The reason why the extraction yield is lower than this study may be that the ethanol:water mixture has a higher polarity index than ethanol.

Table 1. Extract abbreviations and extraction yield

Plant	**Extract**	**Abbreviation**	**Extraction Yield (%)**
Coriander	Ethanol: water (1:1 v/v)	CEW	16.66 ± 2.13
	Methanol: water (1:1 v/v)	CMW	15.40 ± 1.70
	Water	CW	17.33 ± 4.62
Dill	Ethanol: water (1:1 v/v)	DEW	14.53 ± 2.60
	Methanol: water (1:1 v/v)	DMW	15.00 ± 3.99
	Water	DW	17.86 ± 2.86
Purslane	Ethanol: water (1:1 v/v)	PEW	14.06 ± 0.99
	Methanol: water (1:1 v/v)	PMW	14.33 ± 1.22
	Water	PW	16.86 ± 1.86

3.2 Antimicrobial Activity of Plant Extracts

The antimicrobial activity results of the coriander, dill and purslane plant extracts are given in Table 2. While plant extracts showed varying degrees of inhibitory effect on *S. aureus* ATCC 25923, no inhibition effect was observed against *E. coli* O157:H7, *Salmonella* Typhimurium ATCC 14028 and *Aspergillus niger*.

Table 2. Antimicrobial activity of plant extracts against various food borne pathogens

Extract (120 mg/mL)	**Inhibiton Zone (mm)**			
	E. coli O157:H7	*S.* Typhimurium ATCC 14028	*S. aureus* ATCC 25923	*A. niger*
CEW	-	-	13.99 ± 0.87	-
CMW	-	-	12.99 ± 0.57	-
CW	-	-	-	-
DEW	-	-	13.10 ± 1.34	-
DMW	-	-	10.55 ± 0.50	-
DW	-	-	-	-
PEW	-	-	14.86 ± 4.45	-
PMW	-	-	10.22 ± 3.29	-
PW	-	-	12.99 ± 1.15	-
Negative Control	-	-	-	-
Positive Control	(Gentamicin) 15.33±1.00	(Ampicillin) 10.66±1.00	(Ampicillin) 21.33±1.00	(Amphotericin B) 21.66±1.00

-, no inhibition zone.

While no effect was observed in coriander water extract on *S. aureus*, it was determined that methanol:water extract formed a 12.99 ± 0.57 mm inhibition zone, and ethanol:water extract formed a 13.99 ± 0.87 mm inhibition zone. In a research examining the antimicrobial activity of the water extract of the coriander, it was stated that while an inhibitory activity was formed on *S. aureus*, it had no activity on the growth of *E. coli* [38]. Salma et al. [39] were found that the water extract of the coriander did not have any activity on *E. coli* and *S.* Typhi, while it formed an 8 mm inhibition zone on *S. aureus*. On the other hand, ethanol extract (70%) of coriander formed 10 and 9 mm inhibition zones on *E. coli* and *S.* Typhi, respectively, but had no activity on *S. aureus*. In another study, it was determined that methanol and ethanol extracts (150 mg/mL) obtained from the coriander plant did not exhibit antimicrobial effect against *Candida albicans* RSKK02029, *Listeria monocytogenes* ATCC 7644, *Salmonella* Typhimurium RSSK19, and *Escherichia coli* ATCC 11229 [30]. Al-Jedah et al. [40] stated that coriander extracts showed strong inhibitory activity against diverse Gram (+) and Gram (-) bacteria. Antimicrobial activities of plant extracts vary according to the strain and load of the tested microorganism, the species of plant and, concentration and composition of the extract [41].

While the water extract of the dill plant did not show any inhibition effect, it was determined that the ethanol:water extract formed 13.10 ± 1.34 mm inhibition zone on *S. aureus*, and the methanol:water extract formed 10.55 ± 0.50 mm inhibition zone. Similarly, Nair *et al.* [42] reported that ethanol extract of the dill formed 2 mm inhibition zone on *S. aureus* and the aqueous extract showed no effect. On the contrary, Rasheed *et al.* [43] reported that the ethanol extract of dill leaves had no effect on *C. albican*s, *S. aureus,* and *E. coli*. Ünsal et al. [44] revealed that ethanol extract of dill leaves had no inhibitory activity on *Staphylococcus aureus* subsp. *aureus* Rosenbach ATCC 6538P, while it showed an inhibitory effect with a diameter of 7 mm inhibition zone on *E. coli* ATCC 8739. According to Bagamboula *et al.* [45], the results may vary even when different strains of the same microorganism are used.

Inhibition zones of ethanol:water, methanol:water and water extracts of purslane plant on *S. aureus* were 14.86 ± 4.45 mm, 10.22 ± 3.29 mm and 12.99 ± 1.15 mm, respectively. In a study, antimicrobial activity of purslane plant on *E. coli* and *S. aureus* was determined using different concentrations of ethanol and water extracts. It was observed that width of the inhibition zone increased in direct proportion with increase in concentration. Inhibition zones formed by water extract on *E. coli* at 0.125 g/mL, 0.25 g/mL, and 0.5 g/mL concentrations were 14.5 ± 0.17 mm, 19.2 ± 0.19 mm, 22.7 ± 0.24 mm, respectively. Inhibition zones formed by the ethanol extract at the same concentrations were determined as 16.6 ± 0.16 mm, 18.4 ± 0.27 mm and 22.5 ± 0.18 mm respectively. For *S. aureus,* it was determined that water extracts formed 0 ± 0.00 mm, 8.4 ± 0.17 mm, and 12.2 ± 0.18 mm inhibition zones respectively and ethanol extracts formed 12.3 ± 0.15 mm, 15.6 ± 0.19 mm, and 18.3 ± 0.23 mm inhibition zones respectively [46].

The reason why Gram (+) *S. aureus* is more susceptible to plant extracts than Gram (-) bacteria is the presence of lipopolysaccharides in the outer membranes of Gram (-) bacteria that prevent the entry of antimicrobial compounds into the cell. In addition, enzymes in the periplasmic space of Gram (-) microorganisms catabolize the degradation reactions of these compounds [36]. Therefore, antimicrobial activity was observed only against *S. aureus* among the test microorganisms.

3.3 Antioxidant Activity

IC_{50} values of plant extracts prepared with different types of solvents are depicted in Figure 1. Solvent type significantly ($p<0.05$) effected the IC_{50} values of extracts. BHA (146.76 ± 13. 89 µg/ml) (41.79 ± 0.39 µg/ml) and Trolox antioxidants have lower IC_{50} values than all extracts.

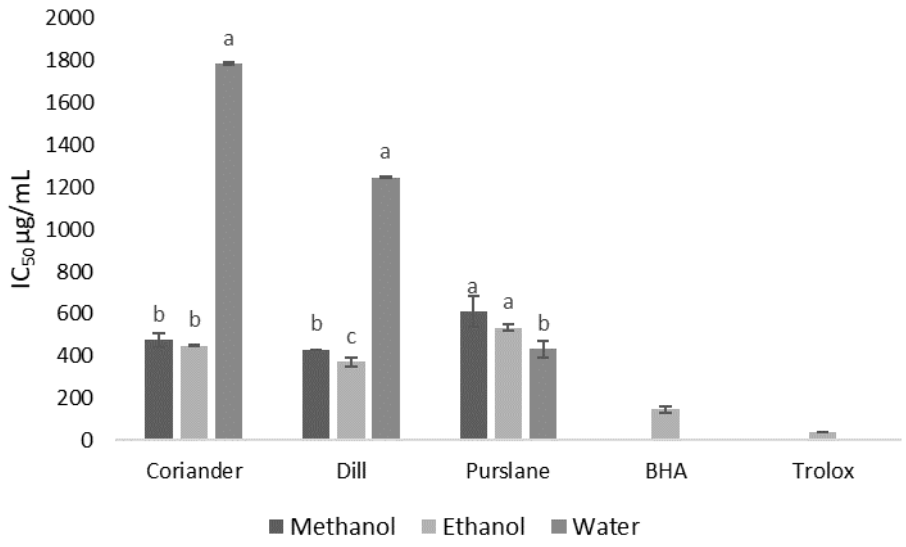

Figure 1. Extracts, BHA, and Trolox's IC_{50} values of DPPH• radical removal activity. Mean values with different letters within each group are significantly different ($p<0.05$)

The IC_{50} value of coriander was high in the water (1783.78 ± 6.34 µg/mL) extract, ethanol:water (450.19 ± 0.28 µg/mL) and methanol:water (476.04 ± 34.49 µg/mL) extracts values were close to each other. Solvent type had a significant ($p<0.05$) effect on the DPPH• radical scavenging activity of the coriander. Similar to our results, Wong and Kitts [47] determined that the DPPH• radical scavenging activity of the ethanol extract of coriander leaves and stems was higher than water extract. Similar to ethanol extract of purslane (450.19 ± 0.28 µg/mL), Wangensteen *et al.* [48] reported the IC_{50} value of coriander leaf ethanol extract as 389.0 ± 5.0 µg/mL.

The IC_{50} value was found to be the highest in the water extract (1247.50 ± 2.68 µg/mL) of the dill plant, and the lowest in the ethanol:water (372.19 ± 22.40 µg/mL) extract. Solvent type had a significant ($p<0.05$) effect on DPPH• radical scavenging activity of the dill plant. Ratananikom and Premprayoon [49] determined the IC_{50} value of dill extracts obtained by classical extraction as 4.94 ± 0.03 mg/mL. In the study conducted by İşbilir and Sağıroğlu [50], contrary to our results, the IC_{50} value of dill water extract (1.93 ± 0.53 mg/mL) was determined to be lower than the IC_{50} value of ethanol extract (4.75 ± 1.35 mg/mL).

The highest IC_{50} values of purslane plant was found in methanol:water (610.69 ± 15.42 µg/mL) extract and lowest in the water extract (432.43 ± 38.91 µg/mL). Solvent type had a significant ($p<0.05$) effect on the DPPH• radical scavenging activity of purslane. Similar to the IC_{50} value of the purslane water extract, You Guo *et al.* [51] determined the IC_{50} value of water extract of the purslane polysaccharides as 350 µg/mL. Besides, Erkan [52] determined the IC_{50} value of methanol extract of purslane plant as 511.8 ± 5.3 µg/mL. The study conducted by Güngören *et al.* [53], antioxidant capacity of wild purslane samples collected from different provinces and cultivated samples were investigated. It was determined that the difference between the inhibition % values varied according to the provinces and in general, water extract demonstrated higher antioxidant activity than ethanol [53].

The differences between the IC_{50} values of the studies may be due to the plant growing conditions, harvesting method and time, light condition and temperature of the storage environment as well as the use of different extraction methods [5].

4. Conclusion

Results of the present research demonstrated that the antioxidant activity of coriander, dill, and purslane plant depends on the extraction solvent. Statistically significant differences were determined between different solvent extracts of the same plant. In terms of DPPH radical scavenging activity, ethanol:water was the suitable solvent for coriander and dill plants, and water was the appropriate solvent for purslane plant. Furthermore, highest antioxidant and antimicrobial activity values belonged to the purslane plant. Antimicrobial activity analysis results revealed that methanol:water and ethanol:water extracts of coriander, dill plant and all extracts of purslane plant showed inhibition activity against Gram positive *S. aureus* ATCC 25923. The most suitable solvent type for all plants in terms of antimicrobial activity was ethanol:water. It is thought that this study will make significant contributions to the scientific literature in determining the antimicrobial and antioxidant activities of these plants, which have been frequently used among the public since ancient times, and will increase the knowledge and competence of researchers working in this field. The possibilities of using these plants and their extracts in food preservation against *S. aureus* should be investigated in further studies.

Ethics in Publishing

There are no ethical issues regarding the publication of this study.

Author Contributions

Conceptualization, A.E., M.G., H.M.A.; methodology, S.T.; validation, S.T., H.M.A. and A.E.; investigation, S.T.; data curation, S.T.; writing-original draft preparation, S.T; writing-review and editing, H.M.A.; visualization, S.T.; supervision, A.E., M.G.

Conflict of Interest

The authors confirm that they have no conflicts of interest with respect to the work described in this manuscript.

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