



Original article

Chemical compositions by using LC–MS/MS and GC–MS and antioxidant activities of methanolic extracts from leaf and flower parts of *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* L.

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ABSTRACT

The members of the *Scabiosa* genus are one of the traditional medicinal plants used in the treatment of many diseases, in particular the treatment of scabies. In this study, it was aimed to determine antioxidant activities and chemical composition of methanolic extracts of leaves and flowers of *Scabiosa columbaria* subsp. *columbaria* var. *columbaria*. The phenolic contents of both parts of the plant were analyzed by LC–MS/MS. A total of 6 phenolic compounds were determined and chlorogenic acid was the major compound in both flower and leaf parts of the plants, with 5936.052 µg/g and 8021.666 µg/g, respectively. 6 different methods were used to determine the antioxidant activity of the plant parts. Both leaf and flower parts of the plant showed high antioxidant activity in all tested methods and the antioxidant activity values of the leaf part were measured higher than those of the flower part for four tests. The methanol extracts of the plant parts was analyzed with GC–MS and number of the essential oil compounds in the leaf and flower parts were determined as 17 and 13, respectively. Linalool compound was also found to be common in both parts of the plant. The major compounds of the essential oils were identified as 4-Octadecenal (30.01%) in the flower and carvone (35.44%) in the leaf. In addition, terpene derivatives was determined as 90.32% of the highest essential oil group in the leaf, while this value was determined as 1.42% in the flower. For the flower, aromatics were determined as the main component group with 21.31%.

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1. Introduction

Plants have been widely used in the treatment or alleviation of many diseases from past to present (Cai et al., 2004; Maver et al., 2015). Although the synthetic drugs are very effective in the treatment of various diseases, people also prefer to use herbal remedies due to less side effects and easily accessible (Trivedi, 2006). The plants contain secondary metabolites such as flavonoids, phenolic acids, lignans, coumarins and stilbenes (Cosme et al., 2020). These secondary metabolites show many biological activities, for exam-

ple, antioxidant activity, antimicrobial (Chedia et al., 2013; Alqahtani et al., 2019; Akar et al., 2020) antitumor, anti-inflammatory and antiallergic (Pereira et al., 2009).

Turkey has an important place in traditional medicine due to the rich flora (Faydaoğlu and Sürücüoğlu, 2011). Also, one of the plants used in traditional medicine is *Scabiosa* L. (Caprifoliaceae) genus applying in potential treatment of various illnesses such as liver diseases, neurodegenerative diseases, neuromuscular diseases (Pinto et al., 2018) and in particular scabies, skin sores, and other skin infections (Maroyi, 2019). *Scabiosa* genus is represented with 39 taxa in flora of Turkey (Bizim Bitkiler, 2021). One of them is *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* L. taxon, which used in traditional medicine.

According to Matthews (1972) the *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* perennial (sometimes biennial), pubescent, often pilose below, reflexed-pubescent above, 30–80 cm. Leaves of sterile shoots, and lower leaves petiolate, lanceolate, oblong or obovate, crenate, dentate or lyrate, rarely entire, sparsely or densely pubescent; upper leaves sessile, 1–2-pinnatisect, segments ovate to linear, entire or dentate. Involucral bracts

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8–11, linear, usually shorter than flowers. Capitula 1–3–5 cm diam., ± radiant; flowers lilac, pink, outer flowers with entire petals. Fruiting head spherical or ovoid. Involucel 2–3.5 mm, 8-grooved with hairy ribs; corona c. 1–5 mm with 16–22 nerves. Calyx shortly stipitate; setae 3.5–9 mm long.

In the study, antioxidant activities of methanol extracts of the flower and leaf parts *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* were determined using six different antioxidant activity determination methods (DPPH, FRAP, TPC, TFC, CUPRAC and ABTS). In addition, while phenolic contents of both flower and leaf parts were determined with LC-MS/MS essential oil contents of both parts of the plant were detected using GC/MS device.

Although researches have been carried out to determine the chemical composition and biological activities of *Scabiosa columbaria* L. at the species and subspecies category (Horn et al., 2001; Benli et al., 2008; Van Vuuren and Naidoo, 2010; Otang-Mbeng and Sagbo, 2020; Sagbo et al., 2020), no research has yet been carried out to determine the characteristics specified at the variety category.

2. Material and methods

2.1. Preparation of plant extraction

Specimens of *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* were collected from locality of Akçaabat-Trabzon province of Turkey (41°02'04" N and 39°30'04" E) 485 m above sea level in September 2019. The leaves and flowers parts of the plant were separated and dried. Then, the dried parts were powdered with grinder and certain amounts powder were weighed from the samples of both flower and leaf and it was extracted with methanol for two hours. At end the process, the extracts were filtered through blue ribbon filter paper to obtain clear solutions and the solvents were removed from solutions with using rotary evaporator. Finally, solutions were prepared by adding methanol so that the concentration of both flower and leaf portions was 40 mg/mL.

For LC/MS-MS analysis, 1 g of each of the samples was added to 10 mL solvent (75% methanol + 25% dichloromethane) and the solutions were extracted on the shaker for two hours. Then, extracts were filtered and injected into the device with passing through a 0.45 µm syringe filter.

Flower and leaf parts of the plant was weighed approximately 35 g for analysis of essential oils in GC-MS device. Essential oils were extracted with the clevenger system. Sodium sulfate was added to remove the water in the oil content dissolved in hexane. Then, the hexane were tested by GC-MS to determine the partial constituents of the essential oils of the plant parts.

2.2. Determination of antioxidant activity

2.2.1. DPPH radical scavenging activity

DPPH • (1,1-diphenyl-2-picrylhydrazyl) radical is commercially available a free radical which shows maximum absorbance at 517 nm. DPPH radical forms intense violet color but the antioxidant compounds decrease the intensity and the absorbance of the formed the color (Brand-Williams et al., 1995). In the study, 100 µM methanolic solution of DPPH radical was used. In addition, the study was carried out in triplicate for each samples and reagent blind. After 50 min, the absorbance values of the tubes mixed with DPPH reagent were read at 517 nm at the spectrophotometer device (UV-1800 (Shimadzu, Japan)). The changes in the absorbance of the DPPH radical treated with different sample concentrations were measured. A graph was drawn based on concentrations corresponding to these absorbance and amount of sample that

halves DPPH concentration in the equation $y = ax + b$ was determined as mg/mL and results were expressed as SC_{50} value.

2.2.2. Ferric reducing antioxidant power (FRAP)

In applying FRAP assay (Benzie and Strain, 1996), the methanolic extracts of both the flower and leaf parts of the plant were analyzed. Also the trolox antioxidant standard were diluted in 6 different concentrations (highest 1000 µM) for calibration graph. Firstly, 50 µL of each samples and standard solution were transferred into tubes as triplicate. In addition, the same amount of sample solution was pipetted into the sample blank tubes. After transferring 50 µL of sample solvents (methanol) to the reagent blind tubes, 1.5 mL newly prepared FRAP reagent was added to the all of the tubes except sample blank tubes. However 1.5 mL of FRAP solvent (pure water with 60% methanol) was added to sample blank tubes. Then tubes were vortexed and kept for 20 min and the absorbance values were read at 595 nm. The results were compared with the standard antioxidant substance trolox and calculated in µM TEAC.

2.2.3. Cupric reducing antioxidant capacity (CUPRAC)

The method (Apak et al. 2004) was modified and applied in the study. Firstly, in this method, equal volumes (500 µL) of Cu (II) chloride solution, neocuproin solution prepared with 96% ethyl alcohol, ammonium acetate buffer (pH = 7) were added respectively. Then analysis solutions were added and the final solutions were completed to 4.1 mL. As in the FRAP method, antioxidant capacities of the samples were calculated as µM TEAC using graphic of standard antioxidant trolox studied in 6 different concentrations.

2.2.4. ABTS⁺ radical scavenging capacity

ABTS antioxidant activity determination method developed by Re et al. (1999) is one of the methods frequently used in antioxidant activity determination studies. To prepare 7 mM ABTS stock solution, ABTS was dissolved in solvent mixture (ethanol : water (5:1)) and ABTS solution was mixed with 2.45 mM potassium persulfate solution in ethanol : water (1:3). The mixture was left at room temperature and in the dark for 18 h to obtain the ABTS radical cation (ABTS⁺). At the end of this period, it was diluted with approximately 1/40 of ethanol: water (3:2) and its absorbance was adjusted to 0.07 at 734 nm. Trolox was used as antioxidant standard and it was pipetted as triplicate in six different concentrations. Simultaneously, the same procedure was performed for reagent and the sample blanks. After 20 min period, the absorbance values of each were measured at 734 nm and the amount of sample that cut ABTS⁺ concentration in half was calculated as mg / mL and the results were expressed as SC_{50} .

2.2.5. Total phenolic content (TPC)

According to the Slinkard and Singleton (1977), the total phenolic content of flower and leaf parts of the plant was determined using the Folin-Ciocalteu reagent. 50 µL of sample solution was diluted with 2.5 mL of distilled water and 250 µL of 0.2 N Folin-Ciocalteu reagent was added. After that the 750 µL Na₂CO₃ (7.5%) was added to the mixture and it was vortexed. The reaction solutions were incubated for 2 h at room temperature and the absorbance values were determined at 765 nm at the spectrophotometer device. The standard calibration graph of gallic acid (1000–500–250–125–62.5–31.25 µg / mL) prepared in six different concentrations was drawn and the amount of phenolic compound in the samples was calculated based on function of the line obtained from the graph as gallic acid equivalent (GAE µg / mL).

2.2.6. Total flavonoid content (TFC)

Total flavonoid content of the plant parts was determined according to the method of Fukumoto and Mazza (2000). The method was carried out in triplicate for all samples and reagent and sample blanks. Equal amounts of samples were pipetted into test tubes and methanol was added to all tubes. 1 M ammonium acetate ($\text{CH}_3\text{COONH}_4$) and 10% aluminum nitrate ($\text{Al}(\text{NO}_3)_3 \cdot 9\text{H}_2\text{O}$) were added to the tubes, except for sample blank tubes, respectively. At the end of the incubation period (40 min) absorbance values were read at 415 nm. Quercetin was used as antioxidant standard, simultaneously. The standard (0.25 mg / mL) was prepared in six different concentrations. The absorbance values were read and the graph was plotted based on the absorbance values corresponding to the concentrations. The total flavonoid amount of the extracts was determined as quercetin equivalent (QAE μg / mL).

2.3. LC-MS/MS analysis

Phenolic component analysis was performed with LC-MS/MS Thermo Scientific/Dionex Ultimate 3000-TSQ Quantum device. LC-MS/MS analyzes of methanolic extracts of the plant parts were carried out by Hitit University Scientific Technical Application and Research Center. The analyses were performed using ODS Hypersil 4.6 * 250 mm, 5 μm column and applied a gradient program including formic acid, water and methanol. Gradient program with 0.1% formic acid (pure water) in reservoir A and 100% methanol in reservoir B was applied. In addition, it is optimized that the injection volumes of the standards and samples to 20 μL , the flow of mobile phase to 0.7 mL / min, and temperature of the column to 30 °C were fixed. The gradient program was optimized by passing through the reservoir 100% A for 0–1 min, 5% A for 3 min in 22 min, and 100% B for 8 min in 26 min.

2.4. GC-MS analysis

Thermo Scientific™ gas chromatography mass spectrometry (GC-MS) was used to determine the essential oil content of the plant. The analyzes of the samples were carried out by Hitit University Scientific Technical Application and Research Center. In the study, the flow rate was determined as 1 mL / min-He (Helium). In addition, TG-5MS column was used and the process was run at 250 °C (the extraction block temperature) and for 55 min.

3. Results

3.1. LC-MS/MS analysis

Six phenolic compounds were detected in LC-MS-MS analysis of both flower and leaf parts of the *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* (Table 1). The same phenolic compounds were detected in different amounts in both parts of the plants. The amounts of gallic acid, 4-OH benzoic acid, caffeic acid in flow-

Table 1
Phenolic content of flower and leaf parts of *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* using LC-MS/MS.

Phenolic Compound	Flower ($\mu\text{g/g}$)	Leaf ($\mu\text{g/g}$)
Gallic acid	1.698	0.957
Catechin	0.296	0.480
Chlorogenic Acid	5936.052	8021.666
4-OH benzoic acid	42.207	19.542
4-OH benzaldehyde	2.064	2.675
Caffeic acid	26.278	7.324

ers were measured as higher than those in the leaves. On the contrary, chlorogenic acid, 4-OH benzaldehyde, catechin were measured higher in leaf parts. However, chlorogenic acid was determined as the major phenolic compound in leaves (8021.666 $\mu\text{g/g}$) and flowers (5936.052 $\mu\text{g/g}$).

3.2. Antioxidant activity

The antioxidant activity of the plant parts was determined using six different antioxidant activity tests. Flowers and leaves of the plant showed significant antioxidant activity values (Table 2). The measured antioxidant activity values in the leaf part were determined much higher than the flowers for CUPRAC (0.462), ABTS (0.064), TPC (640.111) and TFC (13.527) assays. However there is not much difference between the activity values of the flowers and leaf for DPPH and FRAP tests (Table 2).

3.3. GC-MS analysis

The essential oil contents of the flower and leaf parts of the plant determined with GC-MS instrument were reported in Table 3 and Table 4. While 13 compounds were identified in the flower, it was determined as 17 in the leaf part of the plant. The area of the flower essential oil was ranged from 1.00 (5-Octadecenal) to 30.01 % (4-Octadecenal). These values were determined as 0.64 % (Docosane) and 35.44 (Carvone) in the leaf. While other major compounds in flower were stearic acid (15.51), 1-methoxy-3,4,5,7-tetramethylnaphthalene (14.58%) and 2-octadecoxyethanol (11.96), major compounds in the leaf were α -curcumene (21.09%) and 6,10,14-trimethyl-2-pentadecanone (11.76%). Essential oils of leaf and flower parts are completely different from each other both in terms of quantity and content. Both parts of the plant contain only linalool in common.

In addition, the compound groups of essential oils were formed and the total area of each was calculated (Table 5). The groups having the highest area (%) in the flower and leaf parts are aromatics and terpene derivatives, respectively. Unlike the leaves, the flower parts also contain aldehydes and fatty acids. However, leaf part of the plant has approximately 64 times more terpenes than the flower.

4. Discussion

4.1. LC-MS/MS analysis

Recently, foods produced with edible chemical additives to inhibit microbial growth and to extend shelf life are consumed in large quantities (Sridhar et al., 2021). However, the consumption of these foods can cause various health problems in the humans. Phenolic compounds in plants have recently attracted attention of consumers as functional foods due to their biological activities (Naveed et al., 2018). Chlorogenic acid, major compound of flowers and leaf parts of the *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* is a family of esters formed between caffeic acid and quinic acid, which constitute the majority of hydrocinnamic acids, which are major phenolic components in almost all plants (Clifford, 1999; Olthof et al., 2001). It is found in many foods of plant origin (herbal tea, apples, pears, berries, sweet potato, tomato, artichoke and aubergines, anis, plum, peach and brassica vegetables), especially in coffee (Rice-Evans et al., 1996; Clifford, 1999; Wan et al., 2013). Chlorogenic acid has biological activities such as antibacterial (Lou et al., 2011), anticancer (Yan et al., 2017), and antioxidant (Kono et al., 1997) and is used to reduce cholesterol and fatty liver attenuating properties (Wan et al., 2013). In addition plants containing chlorogenic acid are used as

Table 2
Antioxidant activity values of the flower and leaf parts of the *Scabiosa columbaria* subsp. *columbaria* var. *columbaria*.

	DPPH IC ₅₀ (µg/mL)	FRAP TEAC (µM)	CUPRAC TEAC (µM)	ABTS SC ₅₀ (mg/mL)	TPC GAE (µg/mL)	TFC QAE (µg/mL)
Flower	0.0114 ± 0.004	267.381 ± 0.012	0.113 ± 0.005	0.160 ± 0.011	269.833 ± 0.010	6.060 ± 0.014
Leaf	0.0138 ± 0.008	242.857 ± 0.003	0.462 ± 0.007	0.064 ± 0.001	640.111 ± 0.003	13.527 ± 0.028

Table 3
GC–MS analysis of essential oil in flower parts of *Scabiosa columbaria* subsp. *columbaria* var. *columbaria*.

RT (min)	Compound Name in Flower	Molecular Formula	Molecular Weight (g/mol)	Nature of Compound	Area %
13.62	Linalool	C ₁₀ H ₁₈ O	154.253	Monoterpenoid	1.42
16.54	p-Anisyl isonitrile	C ₈ H ₈ NO	134.15	Aromatic	1.27
23.12	o-Toluidine, 5-isopropyl-	C ₁₀ H ₁₅ N	149.23	Aromatic	3.61
28.16	1-Tetradecanol	C ₁₄ H ₃₀ O	214.393	Fatty alcohol	3.06
29.78	4-Hexylacetophenone	C ₁₄ H ₂₀ O	204.31	Aromatic	2.35
32.02	5-Octadecenal	C ₁₈ H ₃₄ O	266.46	Aldehyde	1.00
33.76	1-methoxy-3,4,5,7-tetramethylnaphthalene	C ₁₅ H ₁₈ O	214.21	Aromatic	14.08
37.61	4-Octadecenal	C ₁₈ H ₃₄ O	266.46	Aldehyde	30.01
38.14	Cytisine	C ₁₁ H ₁₄ N ₂ O	190.24	Others	3.26
46.24	2-Octadecoxyethanol	C ₂₀ H ₄₂ O ₂	314.55	Fatty alcohol	11.96
47.99	Nonacosane	C ₂₉ H ₆₀	408.799	Hydrocarbone	2.71
49.65	Stearic acid	C ₁₈ H ₃₆ O ₂	284.484	Fatty Acid	15.51
50.25	1,2-Diphenylacenaphthylene	C ₂₄ H ₁₆	304.384	Hydrocarbone	2.90

Table 4
GC–MS analysis of essential oil in leaf parts of *Scabiosa columbaria* subsp. *columbaria* var. *columbaria*.

RT (min)	Compound Name in Leaf	Molecular Formula	Molecular Weight (g/mol)	Nature of Compound	Area %
9.22	2-Ethyl-2-hexene-1-ol	C ₈ H ₁₆ O	128.21	Fatty alcohol	0.73
13.60	Linalool	C ₁₀ H ₁₈ O	154.253	Monoterpenoid	1.37
16.17	Verbenene	C ₁₀ H ₁₄	134.22	Monoterpenoid	2.04
17.53	α-Terpineol	C ₁₀ H ₁₈ O	154.25	Monoterpenoid	2.50
19.79	Carvone	C ₁₀ H ₁₄ O	150.22	Monoterpenoid	35.44
23.02	Thymol	C ₁₀ H ₁₄ O	150.221	Monoterpenoid	3.01
25.24	p-Eugenol	C ₁₀ H ₁₂ O ₂	164.20	Aromatic	3.32
25.57	β-Damascenone	C ₁₃ H ₁₈ O	190.286	Monoterpenoid	3.63
28.13	Tetradecane, 1-chloro-	C ₁₄ H ₂₉ Cl	232.83	Others	1.27
28.90	β-Ionone	C ₁₃ H ₂₀ O	192.30	Monoterpenoid	2.35
30.77	Caryophyllene oxide	C ₁₅ H ₂₄ O	220.35	Sesquiterpenoid	3.40
31.34	Palustrol	C ₁₅ H ₂₆ O	222.37	Sesquiterpenoid	2.57
32.23	8-Cedren-13-ol	C ₁₅ H ₂₄ O	220.35	Sesquiterpenoid	1.16
33.02	Benzophenone	C ₁₃ H ₁₀ O	182.22	Aromatic	1.67
33.73	α-Curcumene	C ₁₅ H ₂₂	202.22	Sesquiterpenoid	21.09
37.62	6,10,14-trimethyl-2-Pentadecanone	C ₁₈ H ₃₆ O	268.48	Sesquiterpenoid	11.76
46.23	Docosane	C ₂₂ H ₄₆	310.6	Hydrocarbone	0.64

Table 5
Compound groups of essential oils in the flowers and leaves of *Scabiosa columbaria* subsp. *columbaria* var. *columbaria*.

Constituents	Flower oil % Area	Leaf oil % Area
Terpene derivatives	1.42	90.32
Hydrocarbones	5.61	0.64
Aldehydes	31.01	–
Fatty acids	15.51	–
Fatty alcohols	15.02	0.73
Aromatic	21.31	4.99
Others	3.26	1.27
Total	93.14	97.95

food. It has been reported that some taxa of *Scabiosa* contain significant amounts chlorogenic acid as in *Scabiosa columbaria* subsp. *columbaria* var. *columbaria*, for example 57.55 mg / g in *Scabiosa stellata* L. (Rahmouni et al., 2018), in 9.15 mg / g in *Scabiosa comosa* Fisch. and 13.58 mg / g *Scabiosa tschilliensis* Grunning (Ma et al., 2016). In another study, the chlorogenic acid content of the *Scabiosa tschilliensis* at different growth stages varied between 4.23 and 45.35 mg / g and it was major components at some growth stage of this species (Wang et al., 2017). In addition to chlorogenic

acid, 4-OH benzoic acid and caffeic acid were detected as secondary major components in the flower and leaf parts of the *Scabiosa columbaria* subsp. *columbaria* var. *columbaria*. Caffeic acid and its derivatives, present in plant foods that are widely consumed in the human diet, are compounds with a wide range of biological activity and this compounds have potential therapeutic applications (Silva et al., 2014). Caffeic acid was detected in *Scabiosa* species in some studies. Wang et al. (2017) determined phenolic content at the different developmental stages of *Scabiosa tschilliensis*. Caffeic acid were present at some developmental stages of the *Scabiosa tschilliensis* and it was measured the as highest values 6.12 mg / g. In another study, caffeic acid was detected in *Scabiosa tschilliensis* together *Scabiosa comosa* as 0.19 and 0.23 mg / g, respectively (Ma et al., 2016). The other major component , 4-OH benzoic acid was not detected in *Scabiosa* species.

4.2. Antioxidant activity

The high antioxidant activity of leaf extracts may be associated with higher phenolic substance in the leaves (Table 1). Ranilla et al. (2010) stated that the total phenolic content in medicinal plants is directly proportional to the their antioxidant activity. Thus, the

measured high antioxidant activity values in *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* may be related to the high content of chlorogenic acid. Chlorogenic and caffeic acids, which are diphenolics have a higher radical scavenging ability than monophenolics (p coumaric acid) (Rice-Evans et al., 1996). Otang-Mbeng and Sagbo (2020) reported that the FRAP test of the leaves of *Scabiosa columbaria* had high antioxidant activity values due to presence of polyphenolic compounds. Hafsa et al. (2009) concluded that extracts (acetonic, ethanolic and aqueous) obtained using different solvents from *Scabiosa arenaria* Forssk showed high antioxidant activity with different activity tests such as DPPH scavenging capacity (highest inhibition percentages 93.84%), radical cation ABTS⁺ scavenging activity (after 5 min from 7.50% to 93.67% for ethanolic extract), TEAC assay (2.25 mM trolox/g extract) and total phenolic content (138.36 mg catechin/ g extract).

4.3. GC–MS analysis

GC–MS studies on members of the genus *Scabiosa* are limited in the literature. Sagbo et al. (2020) determined the essential oil composition of ethanol extracts from leaves of *Scabiosa columbaria* using GC–MS. The composition of essential oil in *Scabiosa columbaria* (Sagbo et al. 2020) significantly differed than *Scabiosa columbaria* subsp. *columbaria* var. *columbaria*. Unless terpene derivatives in *Scabiosa columbaria* subsp. *columbaria* var. *columbaria*, they determined flavonoids (40.84%) as the major component in *Scabiosa columbaria* (Sagbo et al. 2020). Essential oil composition of *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* showed little similarity with those in *Scabiosa* genus members such as *Scabiosa flavida* Boiss. and Hausskn. (Javidnia et al., 2006), *Scabiosa arenaria* (Besbes et al., 2012), *Scabiosa prolifera* L. (Al-Qudah et al., 2016).

Essential oils of plants are not stable qualitatively and quantitatively (Stevović et al., 2011) and there are some factors that affect chemical composition of plants, such as individual genetic diversity, variations between different plant parts, different developmental stages of the plants and environmental modifications (Barra, 2009). Al-Qudah et al. (2016) reported that chemical composition of essential oil at different flowering stages (fresh and air-dried aerial parts of pre-flowering and flowering) of *Scabiosa prolifera* showed large variations. When the terpenes in both parts of the plant are compared, leaf part of the in *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* has approximately 64 times more terpenes than the flower. Chemical variations between different parts of a plant are related to the its anatomical and physiological characteristics (Barra, 2009).

5. Conclusions

In this study, the chemical composition and antioxidant activity of *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* plant were reported for the first time. Extracts of leaf and flower parts of the plant showed strong antioxidant activity. However, antioxidant activity values in the leaf were higher than the flower. This situation may be associated with the higher quantity and number of phenolic compounds in the leaf parts; especially the high chlorogenic acid content. Determination of the phenolic and essential oil contents and antioxidant activity of leaf and flower extracts of *Scabiosa columbaria* subsp. *columbaria* var. *columbaria* may contribute to in vitro and in vivo biological activity tests.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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