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CHEMICAL COMPOSITION AND BIOLOGICAL ACTIVITIES OF VOLATILE OILS OF *ARABIS ALPINA* L. SSP. *ALPINA*

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ABSTRACT

This work reports volatile constituents, antimicrobial and antioxidant activity of volatile oils from the air-dried flower and aerial parts (APs) of *Arabis alpina* L. ssp. *alpina*. The volatile components of flower and aerial parts (stem + leaf) were investigated by means of hydrodistillation in Clevenger type apparatus and GC/MS/FID analysis. Fifty-one and fifty-three compounds in the volatile oils of flower and APs of *A. alpina* were identified. Terpene derivatives were the major volatiles in both oils. Hexahydrofarnesyl acetone was the main component of flower oil and APs oil in ratios of 16.27% and 26.94%, respectively. Additionally, both flower and APs essential oils were investigated for their antimicrobial activity against twelve bacteria and five fungi, using agar dilution method and antioxidant activities by using $DPPH^{\bullet}$, $ABTS^{\bullet+}$ and Folin-Ciocalteu assays. Flower oil was effective against *B. subtilis* and *B. cereus* even at 100µg/mL. The amount of total phenolic, %$DPPH^{\bullet}$ scavenging activity and $ABTS^{\bullet+}$ scavenging activity were found as 485.60±7.28 mg/mL GAE, 49.85±1.22% and 166.43±12.05µM Trolox equivalent, respectively. Flower oil exhibited antimicrobial effect against gram positive bacteria especially, and moderate antioxidant activity.

Keywords: Antimicrobial activity, Antioxidant activity, *Arabis alpina* L. ssp. *alpina*, Essential oil, GC-MS/FID, Hexahydrofarnesyl acetone

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INTRODUCTION

Being a significant group of vegetables and considered to be a staple food all over the world, Brassicaceae (also known as Cruciferae) species are highly consumed due to their nutritional value (Singh *et al.*, 2017). Brassicaceae vegetables, which can be used as a salad, fresh or dried as a spice, cooked, fried, baked or fermented, contain many minerals, fibers, vitamins and phytochemical compounds. They contain antioxidant compounds such as carotenoids, ascorbic acid, isothiocyanates and phenolics. There are many species in the Brassicaceae family that are used in traditional medicine and foods these are also recognized as the functional food. Brassicaceae family has received a great deal of attention in recent years because of its antioxidative and antimicrobial properties (Balpinar, 2018). The main metabolite of Brassicaceae is glucosinolates that contain sulfur. The phytochemicals obtained from the flower, seed, leaf and stem of these plants provide a great source for medicinal purposes (Shankar *et al.,* 2019). Consumption of Brassicaceae vegetables has preventive role against a variety of chronic diseases, for example, several cancers etc. (Samec *et al.,* 2017).

Arabis L. is a genus within the family Brassicaceae (Koch *et al.*, 2006). *Arabis* L. (Brassicaceae) consists of nearly 128 species (The Plant List, 2018), and most of them are widespread in Northern Hemisphere, also including some part of Africa (Stevens, 2001) and characterized by rosette leaves at the base of plants. *Arabis alpina* is found mostly subalpine and alpine regions and prefers mesophytic areas (Koch *et al.*, 2006). In Turkey, genus *Arabis* is represented by 22 taxa. Ten of these 22 taxa is endemic to Turkey (Cullen, 1965; Mutlu, 2012). *A. alpina* L. is a very variable taxon, which is previously known as *Arabis caucasica* Willd. in Turkey. With renewed Flora of Turkey, *A. caucasica* is now accepted as synonym of *A. alpina* (Mutlu, 2012). There are economical and culinary uses of *A. alpina*. While *A. alpina* is used as ornamental plant in Erzincan (Turkey) province (Korkmaz *et al*., 2016), shoots and leaves of this plant is eaten freshly in Himalaya (India) region (Bhoyar *et al*., 2011). In addition to these, some *Arabis* species are used for medicinal purposes traditionally. *A. tibetica* is used for wound healing (Kala, 2006), whereas *A. glandulosa* is benefited for treatment of abdominal pain (Ballabh and Chaurasia, 2009).

Although there are lots of studies on plant morphology and Arabis mosaic virus of *Arabis* taxa in the literature (Park *et al.*, 2017; Abelleira *et al.,* 2010), the phytochemical studies are limited. Some phytochemical studies on the *Arabis* taxa have been reported, which describe the isolation and identification of a number of glucosides (Kjaer, 1960; Kjaer and A. Schuster, 1972). In these studies, on glucosinolates in

Arabis, sulfur containing hirsutin, 8-methylsulphinyl-3-oxooctyl isothiocyanate, 8-methylthio-3-oxooctyl glucosinolate, and 9-(methylsulfinyl) nonylisothiocyanate were separated from the seeds of *A. alpina* L. and A. *hirsute* (Kjaer, 1960; Kjaer and A. Schuster, 1972). In a study of GC analyses with 14 wild *Arabis* species seeds, the *Arabis* species were characterized by longer chain homologues (hexyl to decyl) and methoxyphenyl oxazolidine-2-thiones (Daxenbichler *et al*., 1991). 3-methylthiopropyl, 6-methylthiohexyl, 7-methylthioheptyl glucosinolates, all of which have sulfur, were isolated from *A. purpurea* and *A. kennedyae* seeds (Hasapis *et al*., 1981). In another study biological activities of the extracts of ethanol and methanol for *A. alpina* L. subsp. *brevifolia* against different food pathogens were investigated. The extracts were shown to have polyphenols, such as quercetin and rutin, and phenolic acids, such as 2,5-dihydroxybenzoic, vanillic and caffeic acid (Balpinar, 2018).

A previous phytochemical study on *A. caucasica* has shown the presence of different heterosides derived from quercetin and kaempferol. Quercetin derivatives were 3-glucoside, 3-*β*-glucosido-7-*α*-rhamnoside and 3-diglucosido-7-glucoside of quercetin, and kaempferol derivatives were 7-*β*-glucoside, 7-rhamnoside, 3-*β*-glucosido-7-*α*-rhamnoside, 7-arabinosido-glucoside, and 7-xylosidoarabinosido-glucoside of kaempferol (Matlawska *et al*., 1992; Matlawska *et al*., 1991). Five acylated anthocyanins were isolated and identified from flowers of A. *blepharophylla* with HPLC (Ito *et al.*, 2013). In a related study analysis of the HPLC results of *A. alpina* seeds revealed some derivatives of cinnamoyl choline (Bouchereau *et al.*, 1991). Additionally, C_8-C_{10} methylsulfinyl alkyl and methylsulfonyl alkyl glucosinolates were identified in the seed of *A. turrita* by HPLC-ESI/MS analysis of intact glucosinolates (Blažević *et al*., 2015). Only two volatile oil studies on *Arabis* genus have been reported. First report includes six *Arabis* species (*A.holboellii, A. demisa, A. crandalii, A. ligniferia, A. drummondii,* and *A. gunisoniana*) (Raguso and Roy, 1998). According to this report, *Arabis* species contain caryophyllene (*A. holboellii*) as terpene, indole (*A. demisa*) and benzyl alcohol (*A. holboellii*) as aromatics, and especially *n*-hexane derivatives that (Z)-3-hexenylacetate, (Z)-3-hexenyl-3-methylbutylate and (Z)-3-hexenal (*A.holboellii, A. demisa, A. crandalii, A. ligniferia, A. drummondii*) as fatty acid derivatives. Also *A. holboellii, A. demissa* and *A. crandallii* contain isopropyl isothiocyanate. Second report includes volatile oil composition of two *Arabis* species (*A. purpurea* and *A. cypria*) from Cyprus (Polatoğlu *et al.*, 2017). The major ingredients of the essential oils were found as nonacosane, heptacosane, and hexahydrofarnesyl acetone. As mentioned above *Arabis* species contains interesting natural phytochemicals.

Till today, there is only one report on the volatile oil composition of two *Arabis* species (*A. purpurea* and *A. cypria*) in scientific publications. Literature survey did not reveal any publication related to the chemical composition, biological activities of the volatile oil of the aerial parts of *A. alpina* plant.

Nowadays, 80% of the materials used in the treatment of diseases in developed countries are of plant origin. In order to take advantage of the properties of plants such as antimicrobial and antioxidant many researchers conduct research to discover new active substances. In this paper, we have reported volatile constituents and biological properties of the volatile oil of the flower and aerial parts (stem + leaf) of *Arabis alpina* L. ssp. *alpina* wild-growing in Turkey. This study is the first attempt in the literature on antioxidant and antimicrobial properties of essential oil of any *Arabis* species.

MATERIALS AND METHODS

Plant Material: *Arabis alpina* L. ssp. *alpina* plant at the blooming stage were gathered from nearby Karaçukur village of Torul, Gümüşhane: (40°36'27"N, 39°17'08"E at 960 m above sea level) in Turkey (A7) during March 2017. The taxonomic identification of plant materials was done by Assoc. Prof. Mutlu Gültepe, in Programme of Forestry, Dereli Vocational School, Giresun University, Giresun, Turkey. Flowers and aerial parts (stem + leaf) of *A. alpina* were separated from plant and shade-dried at room temperature. The voucher specimen has been deposited with the number KTUB744 in the Department of Biology, KTU, Trabzon-Turkey.

Volatile Oil Isolation: The volatile oils from minced and shade-dried plant parts (flower-157g, leaf+stem-181g) of *A. alpina* were obtained by hydrodistillation in Clevenger apparatus (4h, yields: 0.04 and 0.03 %(v/w), respectively). The tests and analyses were performed with GC-FID/MS (İskender *et al.*, 2009; Pino *et al.*, 2005).

GC/FID/MS Analysis of the Volatile Oil: The GC analysis of the volatile oils was performed using an Agilent-5975 Network Gas Chromatography System equipped with a FID and MS Detectors. HP-5MS capillary column (30 m × 0.25 mm ID, film thickness 0.2 μm) was used for GC-FID and GC-MS analyses. Helium was used as carrier gas at a flow rate of 1.2 mL/min. 1 µL volatile oil solution in hexane was injected and analyzed with HP-5MS column. Each sample was injected at split ratio 1:1. Operation conditions were as follows: oven temperature at 50°C (5 min), 50 to 260°C at 4°C/min and 260°C (15 min), manual injection 250°C, and MS detector temperature 230°C (Pino *et al.*, 2005).

Identification of Volatiles: Retention indices (RI or Kovats Index, KI) of all of the volatiles were determined

through the retention times (RT) of *n*-alkanes C_6-C_{32} with linear interpolation. Identification of essential oils compositions was completed through comparing of retention indices(RI) values obtained with the published values and with data of mass spectral libraries (Wiley 275,NIST 05 and Adams Essential Oil Mass Spectral Library) (Pino *et al.*, 2005; Adams 2007; Andriamaharavo 2014; Zhao *et al.*, 2009; Fanaro *et al.*, 2012; Forero *et al.*, 2008; Hammami *et al.*, 2011; Saroglou *et al.*, 2006; Pérez *et al.*, 2007; Demyttenaere *et al.*, 2002; Zhao *et al.*, 2006; Kallio *et al.*, 2006; Sarikurkcu *et al.*, 2008; Kukic *et al.*, 2006). The essential oil samples were analyzed twice and the percentages of components calculated from the GC results. Chemical compositions of essential oils are presented in Table 1 and Table 2.

Antimicrobial activity: Strains of twelve bacteria, which are four gram-positive bacteria and eight gram-negative bacteria and five fungi, were provided by Food Engineering Laboratories of Gümüşhane University. The antimicrobial activities of the volatile oils were determined against bacteria and fungi with agar-well diffusion method (Maksimovic and Mraovic, 2005; Sağdıç and Özcan, 2003). Antimicrobial results of essential oils were presented Table 3.

Antioxidant activity test:

DPPH$^{\cdot}$ assay: The DPPH (2,2-diphenyl-2-picryl-hydrazyl) radical scavenging test is the most common method among antioxidant determination assays. The antioxidant activities of volatile oils were determined by DPPH method with some modifications (Ekici and Özaltın, 2018). Pure methanol was used for blank instead of essential oils of *A. alpina*. Both reaction mixture and reactive blank measurements were repeated in three parallels and averaged. Antioxidant activity was shown through scavenging the DPPH radical percentages. Trolox and ascorbic acid were used as standard antioxidants at 200 µg/mL concentration for comparison.

The result of % scavenging was calculated using the following formula:

%DPPH Radical Scavenging = $((A_{blank} - A_{sample}) / A_{blank})$ x 100

A_{blank}: Absorbance value of the blank DPPH solution

A_{sample} : Absorbance value of the sample tube

Determination of Total Phenolic: The amount of total phenolic contents of essential oils was determined using Folin-Ciocalteu reagent according to the method from the literature, with some modification (Agbor *et al*., 2014). The total phenolic content was calculated from the calibration curve (obtained with gallic acid standard), and the results were expressed as gallic acid equivalent (GAEmg/mL).

ABTS$^{\cdot+}$ radical cation scavenging assay: The radical scavenging capacity of essential oils was evaluated by using ABTS radical cation scavenging assay according to the method from the literature, with some modification (Miller and Rice-Evans, 1997). Results were expressed as µM Trolox equivalent.

For this analysis, 200 µL samples were added to the tubes and mixed thoroughly with 2850 µL ABTS$^{\cdot+}$ solutions and vortexed. This mixture was let to stand for 120 minutes, and absorbance values were measured at 734 nm. The same procedures were performed with standard solutions of ascorbic acid and Trolox. Instead of samples, 150 µL pure methanol was used as a blank. Antioxidant activity was expressed as scavenging percentage of the ABTS$^{\cdot+}$radical (Miller and Rice-Evans, 1997).

%Scavenging =$((A_{blank} - A_{sample})/A_{blank})\times 100$

A_{blank}: Absorbance value of the blank ABTS$^{\cdot+}$solution

A_{sample} : Absorbance value of the sample tube

Statistical analysis: The data were analyzed using the SPSS 17.0 statistical version software program (SPSS Inc., Chicago, IL, USA) for Windows. All the experimental results were presented as mean values ± SD of triplicate measurements. The data were evaluated by using the one-way analysis of variance (ANOVA). Significant differences in groups were indicated at $p < 0.05$.

RESULTS AND DISCUSSION

The chemical composition, antimicrobial and antioxidant activities of essential oils from the flower and aerial parts (leaf + stem) of *A. alpina* ssp. *alpina* were investigated for the first time. According to our literature survey, the present study is the second report on the essential oil composition of any *Arabis* species.

In this study, we evaluated the chemical composition, antimicrobial and antioxidant activities of essential oils from flower and aerial parts (stem and leaf) of *Arabis alpina* L. ssp. *alpina*, which is used as a culinary and ornamental plant. Fifty one and fifty three components were identified constituting 86.64 and 92.98% of the essential oil composition of the flower and APs of *A. alpina*, respectively. Chemical compositions of essential oils are presented in Table 1. Identified volatile compounds were classified as terpene derivatives, hydrocarbons, aldehyde and ketones, fatty acids, alcohols and others (Table 2). The major components of the oils of *A. alpina* were hexahydrofarnesyl acetone (flower 16.27 and APs 26.94%), 2,6,10-trimethyltridecane (flower 9.98%), nonanal (flower 3.19 and APs 4.06%) and geranylacetone (APs 3.70%). Terpene derivatives were the major volatiles in both flower (38.58%) and APs (46.76%). These compounds trigger pharmacological

activities through antioxidant action (González-Burgos and Gómez-Serranillos, 2012).

Table 1. Identified components and chemical class distribution of the essential oils *A. alpine.*

No	Compounds	Flower[a]	APs[a]	Exp.RI[b]	Lit. RI/MS[(Lit)]
1	2-Ethyl furan		2.47	702	702(Pino *et al.*, 2005)
2	Octane[c]	0.69	1.50	800	800(Adams 2007)
3	Hexanal	3.58		802	800(Pino *et al.*, 2005)
4	(*E*)-2-Hexenal	0.47	0.68	853	854(Pino *et al.*, 2005)
5	Heptanal	0.53	1.39	902	899(Pino *et al.*, 2005)
6	*α*-Pinene[c]	2.21	0.87	936	939(Pino *et al.*, 2005)
7	5-Methyl hexanenitrile	0.55	1.31	945	942(Andriamaharavo 2014)
8	1-Octen-3-ol	1.28	3.62	980	978(Pino *et al.*, 2005)
9	4-Isothiocyanato-1-butene		0.48	983	983(Andriamaharavo 2014)
10	6-Methyl-5-hepten-2-one		2.39	987	985(Pino *et al.*, 2005)
11	2-Pentyl furan	0.60	1.19	992	992(Pino *et al.*, 2005)
12	2,4,5-Trimethyl thiazole		0.48	998	MS
13	(*E*)-2-Pentenylfuran	1.22		1001	1001(Zhao *et al.*, 2009)
14	Octanal	0.20		1004	1001(Pino *et al.*, 2005)
15	(*E,E*)-2,4-Heptadienal		0.47	1011	1011(Fanaro *et al.*, 2012)
16	*p*-Cymene	0.52	0.68	1024	1026(Pino *et al.*, 2005)
17	2-Ethyl-1-hexanol		2.83	1030	1031(Forero *et al.*, 2008)
18	Benzaldehyde	1.00	1.03	1043	1044(Pino *et al.*, 2005)
19	(*E*)-2-Octenal		0.44	1060	1063(Pino *et al.*, 2005)
20	1-Octanol	0.24	0.87	1072	1070(Pino *et al.*, 2005)
21	(Z)-Linalooloxide	1.12		1078	1066(Hammami *et al.*, 2011)
22	(E)-Linalooloxide	0.69		1091	1087(Saroglou *et al.*, 2006)
23	Nonanal	3.19	4.06	1105	1103(Pino *et al.*, 2005)
24	Benzeneacetonitrile	0.21		1140	1140(Pino *et al.*, 2005)
25	(*E*)-2-Nonenal	0.26		1163	1162(Pino *et al.*, 2005)
26	1-Nonanol	0.13	0.76	1172	1173(Andriamaharavo 2014)
27	Azulene	3.23		1189	MS
28	2,5-Thiophenedicarboxaldehyde	3.49	1.03	1195	MS
29	Safranal	0.21	0.80	1201	1197(Saroglou *et al.*, 2006)
30	Decanal	0.63	1.11	1206	1205(Pino *et al.*, 2005)
31	*β*-Cyclocitral	0.21	0.72	1222	1220(Pino *et al.*, 2005)
32	Benzenepropanenitrile	0.33	2.75	1241	1243(Andriamaharavo 2014)
33	*p*-Menth-4-en-3-one		0.96	1250	1251(Pérez *et al.*, 2007)
34	Vitispirane	0.78		1281	1281(Demyttenaere *et al.*, 2002)
35	Carvacrol	1.13	0.44	1303	1298(Pino *et al.*, 2005)
36	2-Methoxy-4-vinylphenol	0.30	0.71	1315	1312(Pino *et al.*, 2005)
37	(*E,E*)-2,4-Decadienal	0.64	1.11	1318	1317(Zhao *et al.*, 2009)
38	Dehydro-ar-ionene	0.53		1355	1354(Zhao *et al.*, 2009)
39	Eugenol	0.43	3.46	1360	1359(Zhao *et al.*, 2009)
40	Decanoic acid	0.87		1378	1380(Pino *et al.*, 2005)
41	Methyl eugenol		0.99	1407	1410(Pino *et al.*, 2005)
42	*β*-(*E*)-Caryophyllene		0.36	1420	1418(Pino *et al.*, 2005)
43	Geranylacetone	0.42	3.70	1455	1453(Pino *et al.*, 2005)
44	2,6,10-Trimethyldecane	9.98		1464	1465(Andriamaharavo 2014)
45	Phenethylisothiocyanate		0.68	1469	1472(Andriamaharavo 2014)
46	Dehydro-*β*-ionone	0.33		1486	1485(Zhao *et al.*, 2006)
47	*β*-Ionone	0.63	2.87	1489	1488(Pino *et al.*, 2005)
48	2-Tridecanone	0.13		1496	1497(Kallio*et al.*, 2006)
49	Pentadecane[c]		0.35	1500	1500(Adams 2007)
50	Dodecanoic acid	1.84		1576	1580(Pino *et al.*, 2005)
51	Caryophyllene oxide		1.39	1582	1581(Pino *et al.*, 2005)

52	Hexadecane[c]		0.56	1601	1600(Adams 2007)
53	Carotol		0.35	1608	1612(Andriamaharavo 2014)
54	2-Pentadecanone	0.22		1690	1689(Pino *et al*., 2005)
55	Heptadecane[c]		0.68	1700	1700(Adams 2007)
56	Pentadecanal		0.63	1714	1711(Pino *et al*., 2005)
57	Tetradecanoic acid	1.20		1776	1780(Pino *et al*., 2005)
58	Hexahydrofarnesyl acetone	16.27	26.94	1849	1846(Zhao *et al*., 2009)
59	2-Methylpropyl butyl phthalate	0.68	1.31	1887	1892(Sarikurkcu *et al*., 2008)
60	Hexadecanoic acid	5.53		1972	1968(Andriamaharavo 2014)
61	Eicosane[c]		0.78	2001	2000(Adams 2007)
62	1-Octadecanol		0.68	2084	2081(Kukic *et al*., 2006)
63	Heneicosane[c]	0.42	1.31	2101	2100(Adams 2007)
64	Phytol	3.12	2.23	2115	2114(Andriamaharavo 2014)
65	2-Methyl-(*Z,Z*)-3,13-octadecadienol	2.64		2145	MS
66	Docosane[c]	0.16	1.23	2200	2200(Adams 2007)
67	Tricosane[c]	0.50	1.07	2301	2300(Adams 2007)
68	Tetracosane[c]		0.72	2400	2400(Adams 2007)
69	Pentacosane[c]	1.76	0.59	2501	2500(Adams 2007)
70	Hexacosane[c]		0.28	2600	2600(Adams 2007)
71	Heptacosane[c]	4.20	0.88	2701	2700(Adams 2007)
72	Nonacosane[c]	5.14	1.79	2902	2900(Adams 2007)
	Total percentages (%)	***86.64***	***92.98***		

APs: Aerial parts(leaf + stem) of *A. alpia,,* Exp. RI: Experiment Retention Index, Lit. RI: Literature Retention Index
[a]Percentages obtained by FID peak-area normalization.
[b]Retention index calculated from retention times relative to n-alkanes (C_6-C_{32}) on the non-polar HP-5MS column.
[c]Included as authentic compound

Table 2. The chemical class distribution in the essential oils of *A. Alpina.*

Constituents	Flower oil		APs oil	
	% Area	*NC[a]*	*% Area*	*NC[a]*
Terpene derivatives	38.58	15	46.76	15
Hydrocarbones	16.10	9	11.74	13
Aldehydes or ketones	14.34	12	14.34	11
Fatty acids	9.44	4		
Alcohols	4.29	5	8.76	6
Others	3.89	6	11.38	8
Total number of compounds		**51**		**53**

NC[a]: Number of compounds

α-Pinene, carvacrol, eugenol, geranylacetone, *β*-ionone, and hexahydrofarnesyl acetone were some of the common terpenes in all parts of the *A. alpina*. When compared to literature results of essential oils of *Arabis* species *(A. purpurea* and *A. cypria)* (Polatoğlu *et al*., 2017). *β*-(*E*)-caryophyllene, geranyl acetone, *β*-ionone, caryophyllene oxide, hexahydrofarnesyl acetone, phytol, 1-octanol, 1-nonanol, 1-octadecanol, nonanal, (*E,E*)-2,4-decadienal, pentadecanal, hexadecane, heptadecane, heneicosane, docosane, tetracosane, pentacosane, heptacosane and nonacosane were similarly detected in *A. alpina* oils. The major components of essential oils of *A. purpurea* and *A. cypria* have been reported as hexahydrofarnesyl acetone, nonacosane, heptacosane and phytol. These compounds were also detected in considerable amounts in oils of *A. alpina* in the present research. However, some differences within the chemical composition of essential oils from *Arabis* species were identified, and it is probably related not only to the species difference but also to environmental factors, time of collection of the plant and climatic conditions.

In this work, essential oils of flower and APs (leaf + stem) of *A. alpina* were studied separately. This situation enlightened us for the information about sulfur containing components of the plant, particularly. In the composition of the investigated APs oils, we encountered some isothiocyanates. Thus, sulfur-containing compounds such as 4-isothiocyanato-1-butene, 2,4,5-trimethyl thiazole, 2,5-thiophenedicarboxaldehyde and phenethyl isothiocyanate were only detected in aerial parts (leaf + stem) at various amounts. Isothiocyanates are the hydrolysis products of glucosinolates (Vaughn

and Berhow, 2005). It has been reported in the literature that some sulfur containing glycosinates are obtained from *Arabis* species (Kjaer, 1960; Kjaer and A. Schuster, 1972; Daxenbichler *et al*., 1991; Hasapis *et al*., 1981). GC-MS analysis of seed *A. turrita* L. showed the presence of long-chain olefinic isothiocyanates along with other long-chain thio functionalized glucosinolate breakdown products (Blaževi*ćet al*., 2015). Isothiocyanates have been shown to possess high bactericidal activity against various food pathogens and food spoilage microorganisms (Luciano and Holley, 2009). In the literature survey has reported that isothiocyanates can lower the incidences of different cancers (Dinkova-Kostova and Kostov, 2012).

According to literature *A. purpurea* and *A. cypria* had very low essential oil yields (<0.01%v/w) (Polatoğlu *et al*., 2017). A Clevenger apparatus was used in the current work as in theirs. Likewise, yields of the essential oils of flower and APs were found low (0.04 and 0.03%v/w, respectively).

No biological activity data on antimicrobial or antioxidant properties of essential oils of *Arabis* species in the literature was found. Essential oils are active against most of the microorganisms, including gram positive bacteria such as *Bacillus subtilis* depending on amount and type of terpene derivatives and phenolic compounds (Bakhtiary *et al.*, 2018). Essential oils of *A. alpina* exhibited different inhibition levels against selected eight-gram negative bacteria, four-gram positive bacteria and five fungi (Table 3). According to antimicrobial activity results, the inhibition zone increased with increased concentration of *A. alpina* oils. Four different concentrations (50,100, 500 and 1000 µg/mL) were studied in this work. No antimicrobial activity was observed at 50 µg/mL concentrations. On the other hand, at 1000, 500 and 100 µg/mL concentrations, samples, especially flower oil, exhibited moderate inhibition activity against the bacteria. Bacterial inhibition by essential oils of *A. alpina* was stronger than the inhibition of fungi. The essential oil of flowers showed antibacterial activity against all of the gram-positive bacteria (*B. cereus, B. subtilis, L. monocytogenes* and *S. Aureus)* and the gram-negative bacteria *E. coli* O157:H7at 1000 µg/mL concentration. It also showed activity against *B. cereus* and *B. subtilis* and *E. coli* O157:H7 at 500 µg/mL concentration. Flower oil was effective against *B. cereus* and *B. subtilis* even at100 µg/mL. However, Aps oil was effective only for one-gram negative bacteria (*E. coli* O157:H7) at 1000 and 500 µg/mL. Results indicate that there is better antimicrobial activity for the essential oil of *A. alpina* flower than that of APs. Balpinar reported that methanol and ethanol extracts of *A. alpina* L. subsp. *brevifolia* exhibited antimicrobial activity against *S. typhimurium* (Balpinar, 2018). In that study extracts had no activity against *Bacillus subtilis, Staphylococcus aureus, Salmonella Typhimurium, Enterococcus faecalis, Escherichia coli, Listeria monocytogenes,* and the fungus *Candida albicans*. The essential oils in our study showed no activity against *S. typhimurium* and all tested fungi at all concentrations. There may be differences in antimicrobial effects between related species of the same genus. This is most likely due to the different chemical contents of essential oils and extracts.

Brassica vegetables exhibit biological activities like antibacterial, anticancer activity, antiviral and for the innate immune response system (Shankar *et al.,* 2019). Our study showed that the essential oil of *A. alpina* flower has a moderate antibacterial activity against all tested gram positive bacteria and also essential oils have significant antimicrobial activity against *E. coli*. As known, gram positive bacteria are capable of causing serious and sometimes fatal infections in newborn infants (Mhairi, 2015). *E. coli* O157:H7 is among well-known foodborne pathogens that can cause severe diseases (Campion *et al.*, 2017). Probably, the essential oils of *A. alpina* may find uses as food preservative or food additive. In the proceeding studies, the antimicrobial activity of the flower oil of *A. alpina* should be investigated towards other microorganisms. The fact that these essential oils have an inhibitory effect on both gram-negative and gram-positive bacteria can be used as a source in the search for alternative drugs to replace existing antibiotics.

The antioxidant activities of essential oils (flower and APs) were investigated by using $DPPH^{\bullet}$, $ABTS^{\bullet+}$and Folin-Ciocalteu assays (Table 4). For the determination of total phenolic content (TPC) gallic acid curve with an equation ($y = 0.0059x + 0.0093$ R^2=0.9997) was used (Figure 1). According to this equation, the amount of total phenolic contents of flower and APs essential oils were found 485.60±7.28 and 140.00±3.24 mg GAE/g, respectively. The TPC capacity of flower oil was higher than APs oil.

The total antioxidant activities of essential oils were determined by DPPH method. Trolox and Vitamin C were used as standard antioxidants in %DPPH scavenging tests. %DPPH scavenging value of Trolox and Vitamin C was found 97.23±0.92 and 97.41±0.98%, respectively, at 200 µg/mL concentration. $DPPH^{\bullet}$ scavenging activities of flower and APs essential oils were measured as 49.85±1.22 and 23.20±0.76%, respectively. Balpinar determined the highest DPPH scavenging activity (76.3%) in the flower-fruit-seed ethanol extract of *A. alpina* L. subsp. *brevifolia* (Balpinar, 2018). Due to both the use of a different species, difference of composition of flower-fruit-seed and difference in concentrations of samples and DPPH solution, %DPPH scavenging results of *A. alpina* L. subsp. *brevifolia* could be measured higher than the present results. In all commonly used antioxidant assay

methods, the antioxidant activity of the flower essential oil was found to be higher than APs oil.

Table 3. Antimicrobial activity results (in mm) of the essential oils of *A. alpine.*

Gram negative Bacteria	Flower oil			APs*oil			Streptomsin sulphate 10 µg/mL	Nistasine 30 µg/mL
	1000 µg/mL	500 µg/mL	100 µg/mL	1000 µg/mL	500 µg/mL	100 µg/mL		
Aeromonas hydrophila ATCC 7965	-	-	-	-	-	-	17.11±0.05	NT
Enterobacter cloacea ATCC 13047	-	-	-	-	-	-	-	NT
Escherichia coli ATCC 11230	-	-	-	7.30±0.12	4.28±0.08	-	7.08±0.05	NT
Escherichia coli O157:H7 ATCC 33150	6.20±0.05	5.24±0.05	-	-	-	-	15.20±0.05	NT
Klebsiella pneumoniae ATCC 13883	-	-	-	-	-	-	16.31±0.05	NT
Proteus vulgaris ATCC 13319	-	-	-	-	-	-	14.06±0.05	NT
Pseudomonas aeruginosa ATCC 17853	-	-	-	-	-	-	17.26±0.05	NT
Salmonella typhimurium ATCC 14028	-	-	-	-	-	-	18.24±0.05	NT
Gram positive Bacteria								
Bacillus cereus ATCC 33019	8.13±0.05	5.30±0.05	4.08±0.05	-	-	-	16.02±0.05	NT
Bacillus subtilis ATCC 6633	7.10±0.05	6.39±0.05	4.00±0.05	5.49±0.10	-	-	19.35±0.05	NT
Listeria monocytogenes ATCC 7644	5.55±0.05	-	-	-	-	-	19.26±0.05	NT
Staphylococcus aureus ATCC 25923	4.80±0.05	-	-	-	-	-	12.14±0.05	NT
Fungi								
Saccharomyces cerevisiae BC 5461	-	-	-	-	-	-	NT	18.22±0.05
Candida albicans ATCC 1223	-	-	-	-	-	-	NT	12.19±0.05
Aspergillus niger	-	-	-	-	-	-	NT	14.34±0.05
Aspergillus flavus	-	-	-	-	-	-	NT	11.32±0.05
Penicillum	-	-	-	-	-	-	NT	12.25±0.05

(-): no activity.* Aerial parts(APs) of *Arabis alpina,* NT: Not tested

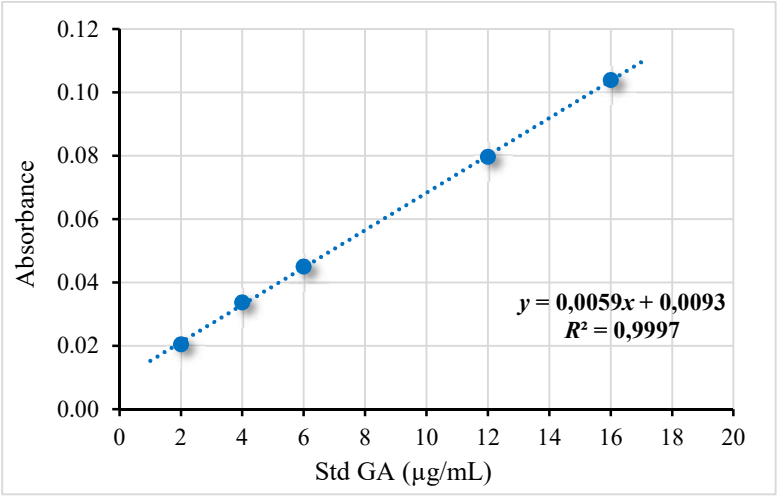

Figure 1. Gallic acid curve used for total phenolic content determination

DPPH$^{\bullet}$ scavenging activities of flower and APs essential oils were measured as 102.46±9.54 and 47.68±1.23 µM Trolox equivalent, respectively. ABTS$^{\bullet+}$scavenging activity of flower and APs essential oils were found 166.43±12.05 and 78.67±5.12µM Trolox equivalent, respectively. Thus, ABTS$^{\bullet+}$scavenging activity values were higher than DPPH$^{\bullet}$ scavenging activity values. According to literature sources, there is a strong correlation between isothiocyanate and phenolic compound contents in essential oils and DPPH and ABTS (Fusari, *et al.,* 2020). Isothiocyanates and phenolic compounds in essential oils may be the cause of the antioxidant effect.

Table 4. Values of antioxidant activity of *Arabis alpine.*

	Flower	**Aerial Parts**	**Ascorbic Acid**	**Trolox**
TPC (Total Phenolic Content) (mg GAE/g)	485.60^{a}±7.28	140.00^{b}±3.24	-	-
DPPH (Inhibition %)	49.85^{a}±1.22%	23.20^{b}±0.76%	97.4^{c}±0.98%	97.23^{c}±0.92%
DPPH (μmol trolox/g)	102.46^{a}±9.54	47.68^{b}±1.23	-	-
ABTS (μmol trolox/g)	166.43^{a}±12.05	78.67^{b}±5.12	-	-

Means ±standard deviations. Different letters (a-c) on the same lines are significantly different at the 5% level ($P < 0.05$).

The evaluation of the antioxidant capacity of essential oils is complex due to the diversity of oxidants and possible different mechanisms. There is not single test that right reflects the antioxidant capacity of the samples. Therefore, different tests have been used for a complete assessment of antioxidant capacity.

According to results, the essential oil of flower had moderate antioxidant activity. Due to its moderate phenolic content and antioxidant capacity, the essential oil of flower can be used in daily diet or in functional foods. We could not find any reports on the antioxidant properties of the *Arabis* species. Therefore, the results have not been compared with any data. This report is the first study in the literature on antioxidant properties of essential oil of any *Arabis* species.

This study may contribute to future researches on antimicrobial and antioxidant properties of the similar plants (especially *Arabis* species). Hexahydrofarnesyl acetone was the main component of both essential oils. This component demonstrated antimicrobial effect, allopathic and pest control potential (Balogun *et al*. 2017). The allopathic and pest control potential of volatile oils should be investigated individually. Probably, the essential oils and various extracts of *A. alpina* and component compounds may find uses as pesticide and allopathic medicine. The essential oils obtained from *A. alpina* have various isothiocyanates. As known isothiocyanates can lower the incidences of different cancers (Dinkova-Kostova and Kostov, 2012). In further studies, anti-cancer tests can be performed for essential oils and extracts.

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REFERENCES

Abelleira, A., J.P. Mansilla, V. Padilla, I. Hita, C. Cabaleiro, E. Bertolini, A. Olmos and F.J. Legorburu (2010). First report of Arabis mosaic on Grapevine in Spain. Pl. Dis. 94(5): 635-635.

Adams, R. (2007). Identification of essential oil components by gas chromatography/mass spectrometry, 4 th Ed. Allured Publishing Corporation, Illinois (USA).

Andriamaharavo, N.R. (2014). Retention Data. NIST Mass Spectrometry Data Center. from:https://webbook.nist.gov/cgi/cbook.cgi?Source=2014AND%2319410M&Mask=200015.06.2018.

Agbor, G.A., J.A. Vinson and P.E. Donnelly (2014). Folin-Ciocalteau Reagent for Polyphenolic Assay. Int. J. Food Sci. Nut. and Diet. 3(8): 147-156.

Bakhtiary, F., H.R. Sayevand, A.M. Khaneghah, A.G. Haslberger and H. Hosseini (2018). Antibacterial Efficacy of Essential Oils and Sodium Nitrite in Vacuum processed Beef Fillet. App. Food. Biotech. 5(1): 1-10.

Ballabh, B. and O.P. Chaurasia (2009). Medicinal plants of cold desert Ladakha used in the treatment of stomach disorders. In. J. Trad. Know. 8(2): 185-190.

Balogun, O.S., O.S. Ajayi and A.J. Adeleke (2017). Hexahydrofarnesyl Acetone-Rich Extractives from Hildegardia barteri. J. Herbs, Spices & Medicinal Plant. 23(4): 393-400.

Balpinar, N. (2018). The biological activities of Arabis alpina L. subsp. brevifolia (DC.) Cullen against food pathogens. Open Chemistry. 16(1): 930-936.

Bhoyar, M. S., G.P. Mishra, R. Singh and S.B. Singh (2011). Ethno-botany of Traditional Wild Edible Plants from Cold Arid Desert of Ladakha Potential Source of Winter Vegetables. Indian Forester. 137(8), 1029-1033.

Blažević, I., S. Montaut, G.R. De Nicola and P. Rollin (2015). Long-chain Glucosinolates from Arabis turrita: Enzymatic and Non-enzymatic Degradations. Nat. Prod. Commun.10(6): 1043-1046.

Bouchereau, A., J. Hamelin, I. Lamour, M. Renard and F. Larher (1991). Distribution of sinapine and related compounds in seeds of Brassica and allied genera. Phytochemistry. 30(6): 1873-1881.

Campion, A., R. Morrissey, D. Field, P.D. Cotter, C. Hill and R.P. Ross (2017). Use of enhanced nisin derivatives in combination with food-grade oils or citric acid to control Cronobacter sakazakii and Escherichia coli O157:H7. Food Control. 65: 254-263.

Cullen, J. (1965). Arabis L. In: Davis, P.H. (Ed.), Flora of Turkey and East Aegean Islands, 1, Edinburg University Press. Edinburgh (United Kingdom). 422 p

Daxenbichler, M.E., G.F. Spencer, D.G. Carlson, G.B. Rose, A.M. Brinker and R.G. Powell (1991). Glucosinolate composition of seeds from 297 species of wild plants. Phytochemistry. 30(8): 2623-2638.

Demyttenaere, J.C.R., C. Dagher, R. Verhé and P. Sandra (2002). Flavour analysis of Greek white wine using solid phase microextraction-capillary GC/M*S* in 25th International Symposium on Capillary Chromatography. 1-16.

Dinkova-Kostova, A.T. and R.V. Kostov (2012). Glucosinolates and isothiocyanates in health and disease. Trends in Molecular Medicine. 18(6): 337–347.

Ekici, L. and B. Özaltın (2018). Effects of concentration methods and storage conditions on some bioactive compounds and color of tamarind sorbet: A traditional Turkish beverage. J. Food Meas. Char. 12: 2045-2056.

Fanaro, G.B., R.C. Duarte, A.G. Santillo, M.E.M. Pinto e Silva, E. Purgatto and A.L.C.H. Villavicento (2012). Evaluation of γ-radiation on oolong tea odor volatiles. Rad. Phys. Chem. 81(8): 1152-1156.

Forero, M.D., C.E. Quijano and J.A. Pino (2008). Volatile compounds of Chile pepper (Capsicum annuum L. var. glabriusculum) at two ripening stages. Flav. Fragr. J. 24(1): 25-30.

Fusari, M., M.A. Nazareno, D.A. Locatelli, A. Fontana, V. Baretta and A.B. Camargo (2020). Phytochemical profile and functionality of Brassicaceae species. Food Bioscience. 36: 100606.

González-Burgos, E. and M.P. Gómez-Serranillos (2012). Terpene Compounds in Nature: A Review of Their Potential Antioxidant Activity. Cur. Med. Chem. 19(31): 5319-5341.

Hammami, I., N. Kamoun and A. Rebai (2011). Biocontrol of Botrytis cinerea with essential oil and methanol extract of Viola odorata flowers. Arch. Appl. Sci. Res., 3(5): 44-51.

Hasapis, X., A.J. MacLeod and M. Moreau (1981). Glucosinolates of nine Cruciferae and two Capparaceae species. Phytochemistry. 20(10): 2355-2358.

İskender, N.Y., N. Yaylı, N. Yıldırım, T.B. Cansu and S. Terzioğlu (2009). The Volatile Constituents of the Flower, Leaf, and Stem of Verbascum wiedemannianum Grown in Turkey. J. Oleo Sci. 58(3): 117-121.

Ito, S., K. Kato and F. Tatsuzawa (2013). Acylated cyaniding 3-sambubioside-5-glucosides in the red-purple flowers of Arabis blepharophylla Hook. &Arn. (Brassicaceae). Biochem. Syst. Ecol. 48: 65-69.

Kala, C.P. (2006). Medicinal plants of the high altitude cold desert in India: diversity, distribution and traditional uses. Int. J. Biodiversity Sci. & Management. 2(1): 43-56.

Kallio, M., M. Jussila, T. Rissanen, P. Anttila, K. Hartonen, A. Reissell, R. Vreuls, M. Adahchour and T. Hyotylainen (2006). Comprehensive two-dimensional gas chromatography coupled to time-of-flight mass spectrometry in the identification of organic compounds in atmospheric aerosols from coniferous forest. J. Chromatogr. A. 1125(2): 234-243.

Kjaer, A. (1960). Naturally derived isothiocyanates(mustard oils) and their parent glucosides. In Fortschritte der Chemieorganischer Naturstoffe/Progress in the Chemistry of Organic Natural products/Progrés Dans la Chimie des Substances Organiques Naturelles. Springer, Vienna (Austria). 122 p

Kjaer, A. and A. Schuster (1972). Glucosinolates in seeds of Arabis hirsute (L.) Scop.: Some new, naturally derived isothiocyanates. Acta Chem. Scand. 26: 8-14.

Koch, M. A., C. Kiefer, D. Ehrich, J. Vogel, C. Brochmann and K. Mummenhoff (2006). Three times out of Asia Minor: The Phylogeography of Arabis alpina L. (Brassicaceae). Mol. Ecol. 15(3): 825-839.

Korkmaz, M., S. Karakuş, S. Selvi and U. Çakılcıoğlu (2016). Traditional knowledge on wild plants in Üzümlü (Erzincan-Turkey), Indian J Tradit Knowle. 15(4): 538-545.

Kukic, J., S. Petrovic, M. Pavlovic, M. Couladis, O. Tzakou and M. Niketic (2006). Composition of essential oil of Stachys alpina L. ssp dinarica Murb. Flav. Fragr. J. 21(3): 539-542.

Luciano, F.B. and R.A. Holley (2009). Enzymatic inhibition by allyl isothiocyanate and factors affecting its antimicrobial action against Escherichia coli O157:H7. Int. J. of Food Microbiology. 131(2-3): 240-245.

Maksimovic, Z.A. and M. Mraovic (2005). Antimicrobial activity of Chenopodium botrys essential oil. Fitoterapia. 76(1): 112-114.

Matlawska, I., M. Sikorska and Z. Kowalewski (1991). Flavonoid compounds in the herb of Arabis caucasica. II. Heterosides derived from kaempferol. Acta Pol Pharm. 48(2): 31-34.

Matlawska, I., M. Sikorska and Z. Kowalewski (1992). Flavonoid compounds in the herb of Arabis caucasica. I. Heterosides derived from quercetin and isorhamnetin. Acta Pol Pharm. 49(3): 45-48.

Mhairi, M.D. (2015). Avery's Neonatology: Pathophysiology and Management of the Newborn. 7 th Ed. Philadelphia (USA). 571 p

Miller, N.J. and C.A. Rice-Evans (1997). Factors influencing the antioxidant activity determined by the ABTS radical cation assay. Free Rad. Res. 26(3): 195-199.

Mutlu, B. (2012). Arabis L., Türkiye Bitkileri Listesi (Damarlı Bitkiler) Nezahat Gökyiğit Botanik Bahçesi ve Flora Araştırmalar Derneği Yayını, İstanbul (Turkey). 256 p.

Park, J.Y., H. Kim and H. Lee (2017). Comparative analysis of molecular and physiological traits between perennial Arabis alpina pajares and annual Arabidopsis thaliana Sy-0. Sci Rep. 7(13348): 1-11.

Pérez, R.A., T. Navarro and C. de Lorenzo (2007). HS-SPME analysis of the volatile compounds from spices as a source of flavour in 'Campo Real' table olive preparations. Flavour Fragr. J. 22(4): 265-273.

Pino, J.A., J. Mesa, Y. Muñoz, M.P. Martí and R. Marbot (2005). Volatile components from mango (Mangifera indica L.) cultivars. J. Agric. Food Chem. 53(6): 2213-2223.

Polatoğlu, K., H. Servi, Ö. Özçınar, A. Nalbantsoy and S. Gücel (2017). Essential Oil Composition of Endemic Arabis purpurea Sm. &Arabis cypria Holmboe (Brassicaceae) from Cyprus. J. Oleo Sci. 66(1): 65-70.

Raguso, R.A. and B.A. Roy (1998). 'Floral' scent production by Puccinia rust fungi that mimic flowers. Mol. Ecol. 7(9): 1127-1136.

Sağdıç, O. and M. Özcan (2003). Antibacterial activity of Turkish spice hydrosols. Food Cont. 14(3): 141-143.

Šamec, D., I. Pavlović and B. Salopek-Sondi (2017). White cabbage (Brassica oleracea var. capitata f. alba): Botanical, phytochemical and pharmacological overview. Phytochemistry Reviews. 16, 117–135.

Sarikurkcu, C., B. Tepe, D. Daferera, M. Polissiou and M. Harmandar (2008). Studies on the antioxidant activity of the essential oil and methanol extract of Marrubium globosum subsp. globosum (Lamiaceae) by three different chemical assays. Bioresource Technol. 99(10): 4239-4246.

Saroglou, V., N. Dorizas, Z. Kypriotakis and H.D. Skaltsa (2006). Analysis of the essential oil composition of eight Anthemis species from Greece. J Chromatogr. A. 1104(1-2): 313-322.

Shankar, S., G. Segaran, V.R.S. Dhevi, S. Settu and M. Sathiavelu (2019). Brassicaceae - A Classical Review on Its Pharmacological Activities. Int. J. Pharm. Sci. Rev. Res. 55(1): 107-113.

Singh, S., S.S. Das, G. Singh, M. Perroti, C. Schuff and C.A.N. Catalan (2017). Comparison of chemical composition, antioxidant and antimicrobial potentials of essential oils and oleoresins obtained from seeds of Brassica juncea and sinapis Alba. MOJ Food Process Technol. 4(4): 113-120.

Stevens, P.F. (2011 onwards). Angiosperm Phylogeny Website. Version 14, July 2017 (and more or less continuously updated since).

The Plant List 2018. Version 1.1. Published on the Internet; http://www.theplantlist.org/ (accessed 20th March, 2018).

Vaughn, S.F. and M.A. Berhow (2005). Glucosinolate hydrolysis products from various plant sources: pH effects, isolation, and purification. Industrial Crops and Products 21(2): 193-202.

Zhao, C., Y. Zeng, M. Wan, R. Li, Y. Liang, C. Li, Z. Zeng and F.T. Chau (2009).__Comparative analysis of essential oils from eight herbal medicines with pungent flavor and cool nature by GC-MS and chemometric resolution methods. J. Sep. Sci. 32(4): 660-670.

Zhao, C.X., X.N. Li, Y.Z. Liang, H.Z. Fang, L.F. Huang and F.Q. Guo (2006). Comparative analysis of chemical components of essential oils from different samples of Rhododendron with the help of chemometrics methods. Chemom. Intell. Lab. Syst. 82(1-2): 218-228.