Determination of Volatile Components of *Citrus* Flowers and Leaves Growing in Hatay, Türkiye

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Species belonging to the genus *Citrus* are produced and traded in large amounts around the world. In addition to the consumption of citrus fruits as food due to their high vitamin C content, their use in many areas has become widespread with the development of the plant-based products industry. In this study, the amount and structure of volatile components of leaves and flowers of 5 different citrus species (*Citrus aurantium*, *Citrus limon*, *Citrus paradisi*, *Citrus reticulata*, and *Citrus sinensis*) were determined using the solid phase micro extraction. Monoterpene hydrocarbons and their oxygenated derivatives were identified as the most abundant chemical component groups. Limonene was the dominant compound in *Citrus limon* flowers (36.5%), leaves (22.5%) and *Citrus paradisi* flowers (22.4%). Linalool, and sabinene were the other major components. Linalool was determined at 50.5% in flowers and 73.3% in leaves of *Citrus aurantium*. Moreover, sabinene had a high amount in *Citrus sinensis* flowers (19.7%), leaves (24.7%), and in *Citrus paradisi* (27.4%) leaves. Apart from these dominant components, γ-terpinene (13.9%) and *p*-cymene (25.4%) were detected in *Citrus reticulata* flowers and leaves in an important amount. It was seen that the leaves and flowers of *Citrus* species gathered from Hatay province were an important source of limonene and linalool compounds.

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INTRODUCTION

Plants are used by humans for construction, food, and ornamental needs, as well as a source of fragrance. Fragrance stimulates instinctive behaviors, such as hormonal changes, choosing a partner, and aggression, allowing behavior in accordance with the environment. Many studies show that people tend to spend more time in pleasantly scented environments (Teller *et al*. 2012; Fidan 2018). When people perceive a scent that they do not like and may harm them, they tend to move away, and they tend to move closer to a scent that they like and that makes them feel safe. Based on this behavioral tendency, aromatherapy has emerged as a form of complementary medicine, aiming to reduce stress and induce a feeling of calmness by stimulating the olfactory system (Kheirkhah *et al.* 2012; Namazi *et al.* 2014). Plants are the sources of fragrance that can be perceived most by humans in nature. While the fragrance of plants can be used in healing gardens, there are many studies on the use of volatile oils obtained from plants in aromatherapy (Saidi *et al*. 2018; Kara *et al*. 2023). The fragrance or odor of plant can be understood and identified by the contents of plant volatile chemical compounds (Correa *et al*. 2010). Volatile or essential oils are fragrant secondary metabolites that are oily mixtures obtained from aromatic plants or herbal drugs, which have a unique smell, taste, color, appearance and low boiling point can be found in liquid form at room temperature. The amount and quality of volatile oil varies depending on the organ of the plant, as well as the time and method of extraction (Bora *et al*. 2020). Plant volatile oil can be extracted using a variety of techniques, the most common ones being solvent extraction, supercritical fluid extraction, and liquid-phase microwave-assisted process extraction (Eikani *et al*. 1999; Zancan *et al*. 2002; Alfaro *et al*. 2004; Kabouche *et al.* 2007). These procedures take a lot of time and can result in the loss of some volatile chemicals, poor extraction efficiency, and hazardous solvent residues. For this reason, it is preferred to develop simple, fast, and solvent-free methods for the analysis of volatile components. Headspace solid-phase microextraction (HS-SPME) combined with GC-MS is a relatively new sampling, concentration technique, which is simple, sensitive, and is a solvent-free method for the extraction of plant volatile components. It is also suitable for identifying the chemical composition of volatile components even at lower temperatures. (Cao *et al*. 2006; Zahradníčková and Bouman 2006; Hashemi *et al.* 2009; Gholivand *et al*. 2013; Özderin *et. al.* 2016; Dönmez and Salman 2017).

Citrus, belonging to Rutaceae, are the important fragrant plants used in aromatherapy and folk medicine (Barrajón-Catalán *et al.* 2011; Loizzo *et al.* 2013). *Citrus* fruits are rich in bioactive chemicals, especially vitamin C, which has several health benefits. They also have a highly well-defined nutritional value (Bora *et al.* 2020). Fruit peels, leaves, and flowers are used as food supplements or tea. In addition to being used in traditional folk medicine of *Citrus* species (Nazami *et al.* 2014), the volatile oils obtained from their fruits, leaves, and flowers are used in the treatment of Alzheimer's disease, anxiety (Loizzo *et al.* 2013), antioxidant, anticancer, antimicrobial, antiseptic, antifungal, antiviral, diuretic, and constipation treatments (Asmah *et al*. 2020; Jeffrey *et al*. 2020; Indriyani *et al*. 2023). In cosmetics, it is used as soap, perfume, skin whitening and cleansing materials, and room fresheners (Palazzolo *et al.* 2013; Bora *et al.* 2020). *Citrus* fruit peels and pruning residues are also used in different industries (Okla *et al.* 2019; Başol and Dönmez 2022; Dönmez and Başol 2022; Dönmez *et al.* 2022). The origin of citrus species is China, India, and southeast Asia (Wu *et al.* 2018; Mahato *et al.* 2019). The cultivation of citrus varieties in the world is concentrated between 40° N and 40° S latitudes (Palazzolo *et al.* 2013). *Citrus* production, usually seen in the Mediterranean Region of Türkiye, ranks 7th in the world (Aslan *et al.* 2020; Atlı and Şahin 2021; Nationmaster 2023).

It was aimed to identify the structure and amount of the leaves and flowers of volatile components of 5 *Citrus* (*Citrus aurantium*, *Citrus limon, Citrus paradisi, Citrus reticulata*, and *Citrus sinensis*) species grown and produced in Hatay-Türkiye, which has Mediterranean climate characteristics.

EXPERIMENTAL

Citrus leaf and flower samples were obtained from Hatay-Dörtyol, where there is an important center of *Citrus* production. In the district where Mediterranean climate conditions are observed, the average annual temperature is 19 °C, the average annual precipitation is 960 to 1000 mm, and the relative humidity is 48%. Leaf and flower samples of bitter orange (*Citrus aurantium*), lemon (*Citrus limon*), grapefruit (*Citrus paradisi*), mandarin (*Citrus reticulata*), and orange (*Citrus sinensis*) were taken and transferred to the laboratory. Leaf and flower samples collected in April 2023 were stored in the freezer until analysis. To avoid differences, especially in terms of chemical analyses, during the sampling phase, sample trees were determined from points close to each other and sampling was performed simultaneously.

Analysis of Volatile Components

Volatile components of orange, mandarin, grapefruit, bitter orange, and lemon leaves and flower samples were determined by HS-SPME (head space-solid phase micro extraction) combined with GC-MS (Shimadzu Corporation, QP 2010 plus, Kyoto, Japan) HS-SPME has predominantly been used to extract volatiles and semi-volatile compounds of flowers and leaves, (Lin *et al.* 2010; Cheong *et al.* 2011; González-Mas *et al.* 2019). This method involves introducing the plant material into a septum-capped vial, allowing it to acclimate, and then exposing a pre-conditioned fiber to the headspace of the vial for a few minutes at ambient temperature or at a higher temperature. About 2 g of leaf and flower samples of each species were placed in a 10 mL vial and kept at 60 °C for 30 min. In this way, the volatile components in the samples were released in the vial. Absorption was achieved by inserting a 75-µm Carboxen/polydimethylsiloxane (CAR/PDMS)-coated fiber needle into the headspace in the vial. Then, desorption was performed by inserting the fiber needle taken from the vial into the GC-MS injection block, and thus the structure of volatile components were determined.

Identification of Components

A Shimadzu QP 2010 plus GC-MS equipment (Shimadzu Corporation, Kyoto, Japan) was used to identify volatile components. The temperature of the injection block, where the fiber needle was immersed, was 250 °C, and the column used was Restek Rx-5 Sil MS (30 m × 0.25 mm × 0.25 µm film thickness; Restek, Bellefonte, PA, USA). The oven temperature program was such that after waiting at 40 ℃ for 2 min, it reaches 250 ℃ with an increase of 4 ℃/min. The detector temperature was 250 ℃ and helium (1.61 mL/min.) was used as carrier gas. Wiley, NIST, and FFNSC spectral libraries were used to identify volatile components.

Statistical Analyses

Using SPSS software (ver. 22.0, SPSS Inc., Cary, NC, USA), the amount of volatile components was statistically analyzed. One-way analysis of variance (ANOVA) was used to determine the significant difference for multiple comparison and Duncan's multiple range test (with a 95% confidence level) was used to confirm the results.

RESULTS AND DISCUSSION

The analysis of volatile components of bitter orange, lemon, grapefruit, mandarin, and orange flowers and leaves was completed *via* SPME, and the results are shown in Table 1. When the analysis results of the volatile components of the flowers were examined, 65 components were detected in *C. aurantium*, 66 in *C. limon*, 69 in *C. paradisi*, 58 in *C. reticulata*, and 65 in *C. sinensis*. In addition, while 46 volatile compounds were identified in *C. aurantium* leaf samples, 68 volatile compounds were detected in *C. limon*, 62 in *C. paradisi*, and 60 volatile compounds in *C. reticulata* and *C. sinensis*.

To the best of the author's knowledge, this was the first time that volatile components grown in Hatay region, which has Mediterranean climatic characteristics, were determined. It is known that terpenes are abundant in the volatile oils of citrus plants (Kohzaki *et al*. 2009); therefore, the SPME method is usually used to determine the mono- and sesquiterpenes. The Citrus volatile oil is mainly composed of monoterpene and sesquiterpene hydrocarbons, and also by their oxygenated derivatives, aliphatic aldehydes, alcohols, and esters (Dugo and Giacomo 2002; Tranchida *et al*. 2012; Vieira *et al*. 2018). After determining the structure of the volatile components, they were classified as hydrocarbons, carbonylic compounds, monoterpene hydrocarbons, oxygenated monoterpenes, sesquiterpene hydrocarbons, oxygenated sesquiterpenes, fatty acids and derivatives, and others. The role of terpenes, especially monoterpenes, in plant defenses against herbivores and plant pathogens or as attractants for pollinators was predicted (Luo *et al.* 2004; Kohzaki *et al.* 2009). Monoterpene hydrocarbons in Citrus samples was determined to be significantly highest in *Citrus reticulata* leaves and then in *Citrus lemon* flowers. The highest amount of oxygenated monoterpenes was detected in *Citrus aurantium* leaves and flowers (Fig. 1).

While linalool (50.5%), linalyl acetate (12.2%), limonene (7.1%), 2-hexenal (4.9%), and β-pinene (4.4 %) were seen as the dominant components in bitter orange flowers, limonene (36.5 %), β-pinene (11.5%), E-citral (6.6%), β-ocimene (5.8%), and caryophyllene (4.7%) were detected at a higher rate in lemon flowers. Moreover, in grapefruit flowers, limonene (22.4%), sabinene (14.7%), linalool (11.2%), β-ocimene (8.6%), 2-hexanal (5.3%) were found in the highest amount. In mandarin flower samples, γ-terpinene (13.96 %), limonene (12.86 %), *p*-cymene (11.2%), 2-hexanal (6.9%), and linalool (6.5%) were determined as the major components. In addition, sabinene (19.7%), linalool (18.6%), limonene (10.9%), β-ocimene (7.2%), and β-myrcene (4.41%) were found having highest amount in orange flowers.

When the volatile components of leaf samples were examined, it was seen that bitter orange leaves contained the highest amount of linalool (73.3%), 2-hexanal (8.2%), linalyl acetate (3.3%), β-pinene (2.8%), and β-ocimene (2%). The dominant components detected in lemon leaves were limonene (22.5%), E-citral (18.7%), β-pinene (13.8%), Z-citral (11.5%), and caryophyllene (5%). While sabinene (27.4%), limonene (15.8%), 2-hexanal (7.7%), β-ocimene (7.6%), linalool (6%) were detected as the most abundant components in grapefruit leaves, *p*-cymene (25.4%), γ-terpinene (14.6%), limonene (9.5%), β-pinene (8.1%), and 2-hexanal (5.3%) were the major components in mandarin leaves. Moreover, sabinene (24.7%), linalool (10.4%), δ-3-carene (6.4%), β-myrcene (5.5%), and limonene (5.3%) were detected as the most abundant volatile component in orange leaves.

The structure and amount of volatile components in *Citrus* flowers and leaves are not always the same. Depending on the species, origin differences, climate, and genetic variations, such monoterpenes as linalool, β-myrcene or β-citronellol, for flowers (Jabalpurwala *et al.* 2009; Sarrou *et al.* 2013; Da Silva *et al*. 2017; Dosoky and Setzer 2018), or sabinene, for leaves (Tomi *et al.* 2008), may be the most dominant compounds (González-Mas *et al.* 2019). Tisserand and Young (2014) determined limonene as the major component with the range of 65.3% to 95.9% in different orange species, mandarin, and grapefruit; however, De Pasquale *et al*. (2006) and Ammar *et al*. (2012) found linalyl acetate and linalool as dominant components in bitter orange.

Table 1. The Amount and the Composition of Volatile Components of *Citrus* Leaves and Flowers (%)

Components	LRI	Flower					Leaf				
		C.au.	C.li.	C.pa.	C.re.	C.si.	C.au.	C.li.	C.pa.	C.re.	C.si.
Hydrocarbons											
Dodecane	1200	0.07	0.04	0.15	0.21	0.08	-	0.02	-	-	-
Tridecane	1320	0.06	0.14	0.10	0.21	0.08	0.01	0.02	0.03	-	0.03
Tetradecane	1400	-		0.12	-	-	0.02	0.04	0.06	-	-
Pentadecane	1520	-	-	-	-	0.07	-	-	-	-	-
Hexadecane	1600	-	-	0.05	-	-	-	-	-	-	-
2-Methylhexadecane	1684	-	-	-	-	0.14	-	-	-	-	-
3-Heptadecene	1697	0.04	-	0.04	-	1.22	-	-	-	-	0.21
Heptadecane	1700	0.04	-	-	-	0.38	-	-	-	-	0.06
Carbonylic compounds											
E-2-Pentanal	696	0.10	0.04	0.15	0.05	0.09	0.28	0.07	0.33	0.12	0.09
E-2-Hexenal	850	4.98	3.35	5.37	6.94	4.25	8.28	1.42	7.73	5.38	4.52
Z-4-Heptanal	919	-	-	0.02	-	-	0.06	-	0.13	0.05	-
2,4-Hexadienal	924	0.02	-	-	-	-	0.28	-	-	0.05	-
E-2-Heptenal	956	0.03	-	0.06	-	-	0.02	0.01	0.03	-	-
Benzaldehyde	964	0.36	0.05	0.40	0.61	0.47	0.06	0.03	0.27	0.11	0.14
Octanal	1020	0.10	0.14	0.11	0.16	-	-	0.11	0.09	-	-
2,4-Heptadienal	1028	0.03	0.05	-	-	0.04	0.75	0.13	0.91	0.41	0.40
Nonanal	1123	-	0.54	0.55	0.69	0.62	-	0.40	0.27	0.15	-
Decanal	1208	0.06	0.09	0.18	0.17	0.09	0.03	0.09	0.31	0.18	0.23
6-Octenal	1251	-	0.29	-	-	-	-	0.63	-	-	0.11
Z-Citral	1258	-	3.74	-	-	-	-	11.50	-	-	2.44
Cuminaldehyde	1260	0.06	0.10	0.06	0.14	0,10	-	-	-	-	0.10
Geraniol	1275	-	-	-	-	-	-	0.48	-	-	-
E-Citral	1286	0.17	6.62	0.33	-	-	0.02	18.75	0.14	0.08	4.13
Perillaldehyde	1293	-	0.10	-	-	-	-	-	-	-	-
Undecanal	1327	-	0.06	-	-	-	-	0.11	-	-	-
Tetradecanal	1736	0.04	0.04	0.04	-	0.05	-	-	-	-	-
Monoterpene hydrocarbons											
α-Thujene	927	0.09	0.22	1.30	0.92	2.00	0.05	0.20	2.84	2.42	2.86
α-Pinene	933	0.30	1.13	0.78	1.77	0.94	0.19	1.47	1.13	2.35	1.50
Camphene	953	0.03	0.09	-	-	-	0.01	0.13	0.05	0.05	-
β-Pinene	978	4.41	11.55	2.12	4.79	1.60	2.80	13.86	3.12	8.15	1.93
β-Myrcene	991	1.44	1.59	4.02	4.30	4.41	1.40	1.42	5.97	2.59	5.58
Pseudolimonen	1019	-	-	-	-	-	-	-	0.06	-	0.05
Phellandrene	1022	-	-	0.19	0.10	0.44	-	-	0.29	0.16	0.68
Δ-3-Carene	1024	0.05	0.06	-	0.06	1.55	-	0.30	0.06	-	6.46
α-Terpinene	1041	0.11	0.18	0.81	0.58	1.20	0.10	0.11	1.54	0.87	1.47
p-Cymene	1046	0.12	0.74	0.51	11.21	0.55	0.06	0.33	1.03	25.44	0.89
Limonene	1048	7.19	36.54	22.43	12.86	10.91	1.12	22.53	15.85	9.56	5.32
E-Ocimene	1054	0.33	0.93	0.36	0.24	0.27	0.25	0.40	0.54	0.11	0.22
Z-β-Ocimene	1065	4.07	5.88	8.63	5.05	7.20	2.05	2.07	7.61	2.64	4.75
γ-Terpinene	1076	0.18	2.20	1.52	13.96	2.39	0.09	0.38	2.66	14.61	2.52
α-Terpinolen	1102	0.08	0.25	0.41	1.11	0.82	0.05	0.22	0.68	1.75	1.44
Alloocimene	1158	0.20	0.17	0.53	0.72	0.33	0.13	0.19	0.49	0.46	0.22
Citronella	1172	-	0.35	0.18	-	0.08	-	2.99	0.18	-	5.22
Fenchene	1237	-	-	0.06	-	0.06	-	-	-	-	-
Verbenol	1256	-	-	0.20	0.21	-	-	0.48	0.08	-	-
Oxygenated monoterpenes											
Sabinene	972	1.47	2.55	14.75	2.34	19.79	0.80	4.15	27.46	1.70	24.74
1,8-Cineole	1048	0.12	1.26	-	1.25	-	-	0.70	-	0.04	-
Z-Sabinene hydrate	1089	-	0.05	0.22	-	0.39	-	0.05	0.23	-	0.37
Linalool	1119	50.55	1.69	11.20	6.50	18.62	73.38	0.94	6.07	2.56	10.49
1,2-Dimethyl-3-vinyl-1,4-cyclohexadiene	1139	-	-	0.08	-	0.08	-	-	0.07	-	-
p-Mentha-1,5,8-triene	1148	0.22	0.34	0.50	0.23	0.32	0.15	0.11	0.44	0.14	0.30

p-Mentha-1(7),8-dien-2-ol	1180	-	0.10	-	-	-	-	0.30	-	-	-
endo-Borneol	1184	-	-	-	-	-	-	-	-	-	0.04
4-Terpineol	1188	0.04	0.32	0.58	0.18	0.64	-	-	0.21	-	0.37
α-Terpineol	1198	0.15	1.07	0.74	0.67	0.97	0.04	0.28	0.20	0.04	0.34
Carveol	1257	0.08	-	-	-	0.05	-	-	-	0.03	-
Carvone	1262	0.03	0.07	0.07	0.07	-	-	-	-	-	-
Linalyl acetate	1269	12.23	-	0.27	0.22	0.53	3.39	-	0.03	-	0.09
Bornyl acetate	1302	0.06	-	-	-	-	-	-	-	-	-
Thymol	1303	-	-	-	-	-	-	-	-	0.14	-
Carvacrol	1320	-	-	-	-	-	-	-	-	0.45	-
β-terpinyl acetate	1365	0.10	-	0.18	-	-	-	-	0.07	-	-
Citronellyl acetate	1369	-	-	-	-	-	-	0.10	-	-	0.05
Sesquiterpene hydrocarbons											
Bicycloelemene	1351	-	-	-	-	-	0.03	-	-	-	-
Δ-Elemene	1354	0.04	-	-	0.10	-	0.02	-	-	-	-
α-Cubebene	1366	-	-	-	0.13	-	-	-	-	0.06	-
α-Copaene	1375	-	-	0.11	0.33	0.05	-	-	0.09	0.14	0.03
β-Elemene	1389	0.06	0.18	2.02	4.78	1.49	-	0.26	2.57	4.67	3.06
E-Jasmone	1394	0.07	-	0.07	0.32	0.18	-	-	-	0.04	-
7-epi-Sesquithujene	1422	-	0.12	0.18	-	0.14	-	0.06	0.09	-	0.12
Caryophyllene	1439	1.06	4.74	2.44	2.79	1.63	0.79	5.08	1.81	3.06	1.43
β-Cubebene	1449	-	-	-	-	-	-	-	0.07	-	0.06
α-Bergamotene	1453	0.05	1.74	-	0.05	-	-	0.93	-	-	-
β-Farnesene	1472	0.44	0.58	4.90	0.13	2.22	0.69	0.14	2.10	-	1.45
α-Humulene	1475	0.40	0.41	-	0.72	-	0.47	0.37	0.39	0.81	0.43
α-Amorphene	1489	-	-	0.05	0.05	-	-	-	-	0.07	0.04
β-Himachalene	1496	-		0.05	-	0.04	-	-	-	0.04	0.05
Curcumene	1498	-	0.04	0.13	-	0.08	-	-	0.06	-	-
Germacrene-D	1500	0.08	-	-	0.14	-	-	-	-	0.05	-
β-Chamigrene	1504	-	-	-	-	-	-	-	-	0.03	-
β-Selinene	1508	-	-	0.39	0.60	0.33	-	-	0.20	0.63	0.32
Viridiflorene	1511	-	-	-	-	-	0.02	0.01	-	-	-
Valencene	1513	-	-	-	0.07	-	-	-	-	0.07	-
Bicyclogermacrene	1516	0.22	0.59	-	-	-	0.23	0.21	-	-	-
α-Selinene	1516	-	-	0.42	0.90	0.31	-	-	0.31	0.90	0.44
α-Farnesene	1525	0.08	0.12	0.24	1.79	0.81	-	0.04	-	0.91	0.25
E-Caryophyllene	1526	-	-	-	0.14	-	-	-	-	-	0.10
β-Bisabolene	1529	0.03	2.41	0.25	-	0.19	-	1.28	1.65	0.10	-
ɣ-Cadinene	1533	-	-	-	0.04	-	0.02	-	-	0.04	-
α-Bisabolene	1561	-	0.06	-	-	-	-	0.02	-	-	-
β-Sesquiphellandrene	1577	0.03	-	0.19	-	0.11	-	-	0.10	-	0.12
β-Santalene	1779	-	0.08	-	-	-	-	0.04	-	-	-
Oxygenated sesquiterpenes											
Hexahydrofarnesol	1371	-	-	-	-	-	-	-	-	0.05	-
Neryl acetate	1372	0.13	0.08	-	-	-	0.10	0.93	-	-	-
Aromadendrene	1456	-	0.07	-	-	-	0.04	0.03	-	0.04	-
Alloaromadendrene	1468	-	0.13	-	-	-	-	0.02	-	0.03	-
Levomenol	1521	-	-	-	-	-	-	0.13	-	-	-
Farnesol	1582	2.18	1.25	2.92	-	3.21	0.11	0.04	0.16	-	0.15
Spathulenol	1585	-	-	-	-	-	-	0.03	-	-	-
Caryophyllene oxide	1587	-	-	-	-	-	-	0.08	-	-	-
β-Sinensal	1699	-	-	-	-	0.05	-	-	-	-	0.16
Fatty acids and derivatives											
Z-3-Hexenyl acetate	1023	-	-	-	-	-	0.17	-	-	-	-
Hexyl acetate	1031	0.11	-	-	-	-	-	-	-	-	-
Hexyl hexoate	1193	0.04	-	0.08	0.11	0.05	-	-	-	-	-
Ethyl caprylate	1200	0.07	-	-	0.20	-	-	-	-	-	-
Methyl geranate	1340	-	-	-	-	-	-	0.10	-	-	0.32
Hexylhexanoate	1390	-	-	0.06	-	-	-	-	-	-	-
Others											

3-Pentenone	677	0.23	0.06	0.09	0.19	0.07	0.10	0.02	0.15	0.16	-
Z-2-Pentenol	767	-	-	-	-	-	-	-	0.06	0.03	-
E-2-Hexen-1-ol	885	-	0.04	0.03	-	-	-	-	-	-	-
n-Hexanol	887	-	0.04	0.13	-	-	-	-	0.12	-	-
6-Methyl-5-hepten-2-one	986	0.18	0.19	0.27	0,16	0.03	0.05	0.14	0.21	0.19	0.18
Benzene-acetaldehyde	1060	1.17	0.11	1.13	1.69	1.04	1.05	0.03	0.15	1.29	0.06
E-2-nonen-1-ol	1092	-	-	-	-	-	-	-	0.05	0.05	-
α-*p*-Dimethylstyrene	1107	-	-	-	2.80	0.15	-	0,51	0.06	1.62	0.14
Neryl nitrile	1132	-	-	0.09	-	0.10	-	0.01	0.05	-	-
Benzeneacetonitrile	1155	0.07	-	0.97	1.07	0.15	0.12	-	-	0.94	-
Nerol	1248	-	0.79	-	-	-	-	1.86	-	-	-
Indole	1311	1.53	0.54	0.43	1.48	1.06	-	0.09	0.11	0.47	0.18
Methyl anthranilate	1359	1.59	0.79	1.86	0.70	2.56	-	-	0.18	0.66	0.51
Dihydropseudoionone	1467	0.07	0.04	0.19	-	0.07	-	-	-	-	-
Octyl ether	1985	0.26	0.08	-	-	0.06	0.12	0.02	-	0.12	0.04

C.au.: *Citrus aurantium*, C.li.: *Citrus limon*, C.pa.: *Citrus paradisi*, C.re.: *Citrus reticulata*, C.si.: *Citrus sinensis*. Different letters indicate significant differences according to Duncan analysis using SPSS software (p = 0.05). LRI: Linear Retention Indices (Rx-5 Sil column).

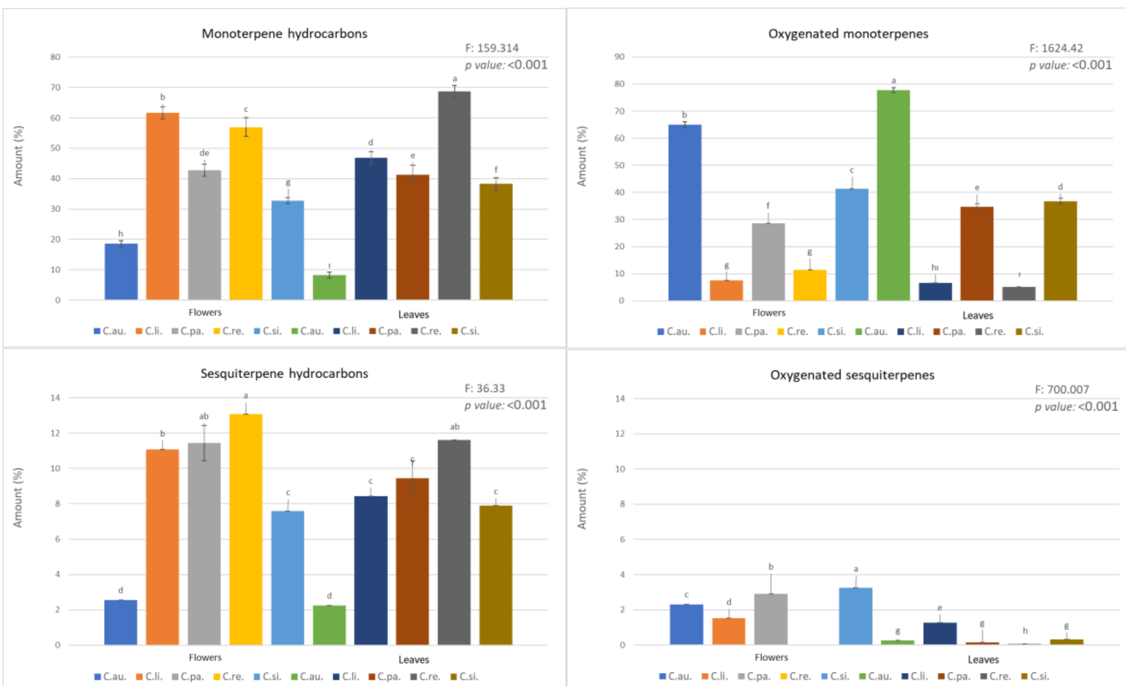

Fig. 1. Comparison of terpenes in *Citrus* species C.au.: *Citrus aurantium*, C.li.: *Citrus limon*, C.pa.: *Citrus paradisi*, C.re.: *Citrus reticulata*, C.si.: *Citrus sinensis*; p value: Significance level of the differences between species calculated by Duncan's Multiple Range Test; a, b, c: Represents a statistically significant difference among *Citrus* species.

Limonene is a widely distributed terpene found in nature and a primary component of many *Citrus* volatile oils (Fig. 2). Limonene is a colorless liquid and has a pleasant lemon-like odor (Hirota *et al*. 2010). This makes it widely used as a flavor and fragrance additive in common food items, as well as in the cosmetic industry as soaps, perfumes, and shampoos (Jongedijk *et al*. 2016; Vieira *et al*. 2018). When the amounts of limonene in the flowers and leaves of the 5 *Citrus* species were examined, values were between 1.1% and

36.5%. The flowers followed by the leaves of *Citrus limon* statistically have the highest amount. Flowers and leaves of *Citrus aurantium* had the lowest limonene amount.

Linalool is one of the principal components of many *Citrus* volatile oils (Kamatou and Viljoen 2008). Because linalool is an unsaturated monoterpene alcohol with a particular odor, it can be found in both non-cosmetic and decorative cosmetic products, as well as in fine fragrances (Ford *et al.* 2000; Letizia *et al.* 2003). There are significant differences in the amount of linalool in *Citrus* species (Fig. 2). While the amount of linalool was significantly highest in bitter orange (*Citrus aurantium*) flowers and leaves (50.5% and 73.3%), it was lowest in *Citrus limon* flowers and leaves (1.6% and 2.5%). Linalool, the dominant compound of *Citrus aurantium* grown in Iran (40.6%) found by Rahimi *et al.* (2014) and France (40%) (Jeannot *et al.* 2005), gave similar results for current results. In contrast, the amount of linalool, analyzed by Değirmenci and Erkurt (2020), was determined as 15.7%, the samples growing in Cyprus were lower compared to Hatay samples.

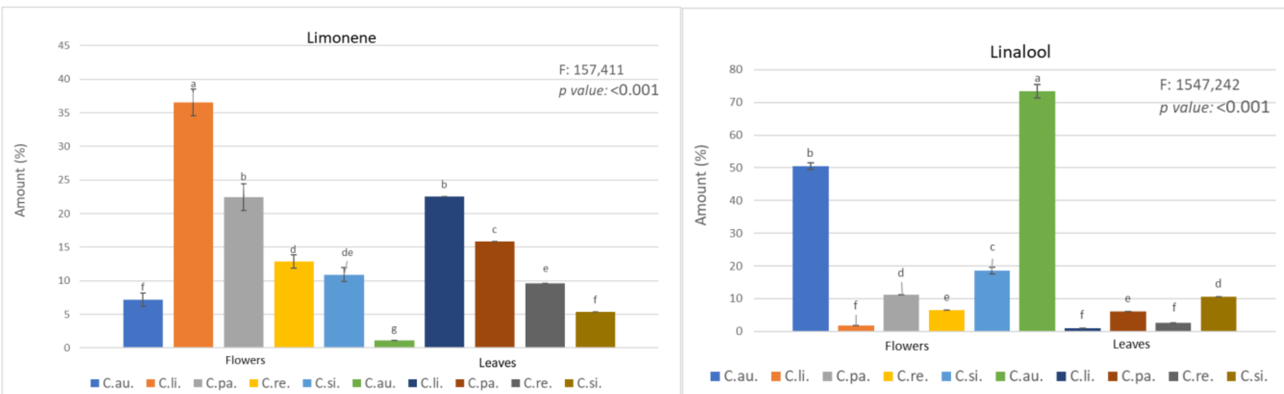

Fig. 2. Comparison of limonene and linalool in *Citrus* species C.au.: *Citrus aurantium*, C.li.: *Citrus limon*, C.pa.: *Citrus paradisi*, C.re.: *Citrus reticulata*, C.si.: *Citrus sinensis*; p value: Significance level of the differences between species calculated by Duncan's Multiple Range Test; a, b, c: Represents a statistically significant difference among *Citrus* species.

CONCLUSIONS

Citrus species are commonly grown in Mediterranean climate conditions. Hatay is the center of *Citrus* production in the Eastern Mediterranean in Türkiye. With this study, the structure and contents of volatile components of 5 *Citrus* species grown in this region were determined for the first time. The following are the conclusions from this study:

1. Monoterpene hydrocarbons and oxygenated derivatives are the most abundant component groups in *Citrus* species. *Citrus reticulata* leaves and *Citrus limon* flowers had the highest amount of total monoterpenes.
2. Limonene is a bioactive compound whose fragrance, antibacterial, and antifungal properties have been demonstrated by previous studies. *Citrus limon* grown in the Hatay region statistically had the highest limonene amount compared to other *Citrus* species. However, *Citrus paradisi* flowers also had a high amount of limonene.

3. Although *Citrus aurantium* flowers and leaves had the lowest amount in terms of total monoterpenes, oxygenated monoterpenes had the highest amount. The most important reason for this is that linalool stands out as the dominant component in flowers and leaves.
4. In terms of the amount of linalool, *Citrus aurantium* flowers and leaves have the highest amount compared to other *Citrus* species and the same species growing in different regions. Linalool is a widely used component in the cosmetics and cleaning industry. With this study, it was determined that *Citrus aurantium* grown in the Hatay region is a rich source of linalool.

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