

<https://doi.org/10.21603/2074-9414-2025-1-2553>
<https://elibrary.ru/WBNHUK>

Original article
Available online at <https://fppt.ru/en>

Chemical Composition of Essential Oils from *Citrus fortunella* (Kumquat) and Its Mutants



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Received: 25.06.2024

Revised: 30.09.2024

Accepted: 01.10.2024

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Abstract.

Kumquat, known as the little jewel of the citrus family, is a fruit of the plant *Citrus japonica* Thunb. or *Fortunella japonica* Swingle. Kumquat is widely grown for its fruit, but such parts of the plant as leaves and bark are left as waste without being utilized. Therefore, we aimed to determine the chemical composition of essential oils from kumquat fruit and leaves (main species and 6 mutants) grown at the Mersin Alata Horticultural Research Institute.

The fruits and leaves of *Citrus fortunella* trees were collected and their essential oils were obtained by hydrodistillation. Gas chromatography-mass spectrometry (GC-MS) was performed to analyze the components of the essential oils.

Among the essential oil components determined by GC-MS analysis, limonene (69.9–94.4%) was detected at the highest levels in the fruit essential oil, while the leaf essential oil was rich in elemol (13.2–14.8%), β -eudesmol (9.3–11.0%), α -guaiaol (8.5–10.8%), spathulenol (8.1–10.5%), and alismol (6.5–7.9%). Our results showed that essential oil can be produced as a by-product from the leaves and fruits of *C. fortunella* trees.

Kumquat fruit and leaf essential oils contain large amounts of chemical components with potential biological activity, both major and minor. Therefore, they can be used as an herbal resource in different industrial fields such as medicine, perfumery, and cosmetics.

Keywords. Kumquat, *Citrus fortunella*, essential oils, limonene, elemol, β -eudesmol, α -guaiaol, spathulenol, alismol

Funding. The authors thank İnönü University, Turkey for financial support (BAPB – Grant No. TYL-2018-1108).

For citation: Küçükbay F, Büyükkormaz Ç, Özek G, Özek T. Chemical Composition of Essential Oils from *Citrus fortunella* (Kumquat) and Its Mutants. Food Processing: Techniques and Technology. 2025;55(1):29–44. <https://doi.org/10.21603/2074-9414-2025-1-2553>

Химический состав эфирных масел кумквата (*Citrus fortunella*) и мутантов



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Поступила в редакцию: 25.06.2024

Принята после рецензирования: 30.09.2024

Принята к публикации: 01.10.2024

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Аннотация.

Кумкват (*Citrus japonica* или *Fortunella japonica*) – растение семейства цитрусовых, плоды которого широко применяются в пищевой промышленности, однако его листья и кора, как правило, выбрасываются в качестве отходов. Цель данной работы – изучить химический состав эфирных масел, полученных из плодов и листьев кумквата основного вида и шести мутантов (Институт растениеводства «Mersin Alata», Турция).

Эфирные масла плодов и листьев кумквата получали методом гидродистилляции. Компонентный анализ эфирных масел проводили при помощи газовой хромато-масс-спектрометрии (ГХ-МС).

В эфирном масле из плодов самым распространенным компонентом оказался лимонен (69,9–94,4 %); в эфирном масле из листьев – элемол (13,2–14,8 %), β -эудесмол (9,3–11,0 %), α -гвайол (8,5–10,8 %), спатуленол (8,1–10,5 %) и алисмол (6,5–7,9 %). Согласно полученным результатам, эфирное масло может производиться как побочный продукт из листьев и плодов *C. fortunella*.

Эфирные масла плодов и листьев кумквата содержат большое количество химических компонентов с разным уровнем биологической активности, что делает возможным их использование в качестве источника растительного сырья в различных областях промышленности, таких как медицина, парфюмерия или производство косметики.

Ключевые слова. Кумкват, *Citrus fortunella*, эфирные масла, лимонен, элемол, β -эудесмол, α -гвайол, спатуленол, алисмол

Финансирование. Исследование было выполнено при финансовой поддержке университета Инёню, Турция (BAPB, грант № TYL-2018-1108).

Для цитирования: Кючукбай Ф., Бююккормаз Ч., Озек Г., Озек Т. Химический состав эфирных масел кумквата (*Citrus fortunella*) и мутантов. Техника и технология пищевых производств. 2025. Т. 55. № 1. С. 29–44. (На англ.) <https://doi.org/10.21603/2074-9414-2025-1-2553>

Introduction

Herbal medicine has been used for centuries and many traditional cultures still rely on it as their primary form of healthcare. Despite today's prevalence of synthetic and semi-synthetic drugs, there has been a renewed interest in natural medicines due to concerns about the side effects of modern drugs. Many people prefer to use natural remedies as they are believed to be safer and have fewer side effects [1–3].

Herbal medicines are increasingly being used in developed countries as a way to maintain good health and prevent disease, rather than just to treat illness. One of the reasons for this is that herbal medicines often have

multiple indications and can be used to address a wide range of health concerns [2]. However, herbal medicines must still meet certain criteria for safety, effectiveness, and purity to ensure that they do not have negative effects on human health. Additionally, the effects of herbal medicines often result from a combined action of multiple bioactive compounds found in the plant, rather than just one single active ingredient. This makes it more difficult to study the effects of herbal medicines compared to chemical medicines [4, 5]. Since isolated bioactive compounds may not have the same effect as their combination in a plant, whole plant extracts are often preferred in traditional medicine.

In recent years, there has been an increased interest in natural food and nutritional supplements to promote good health. Tropical and subtropical plants are believed to have protective effects on health due to their unique chemical structures and essential oils. Kumquat, a tropical fruit, is a member of the same family as citrus fruits, but is classified in a different genus. Kumquat has a unique flavor, with a sour flesh and sweet peel, due to the presence of flavonoids and terpenoids. This flavor has a characteristic citrus taste [6, 7]. Like other citrus fruits, kumquat is a rich source of vitamin C, which helps to strengthen the immune system. Additionally, the terpenoids and flavonoids found in the peel of the fruit make it beneficial to consume the peel as well. Kumquat is also used in a variety of culinary applications, such as jams, marmalades, liquors, confectionery, pickles, and dried products. The fruit is known for its unique sweet-sour flavor and its versatility in cooking [8–10]. The essential oil obtained from the bark of the kumquat tree is also used in perfumery, pharmacy, and food industry. The oil has a distinct citrus aroma, which makes it a common ingredient in a variety of products such as perfumes, cleaning agents, and food flavorings. Kumquat essential oil is also applied in traditional medicine for its multiple therapeutic properties, including antioxidant, antifungal, anti-proliferation, and anti-inflammatory effects [10–12].

Kumquat was first described in China in 118 BC. Its name is thought to have originated from the Cantonese language, where it is literally translated as “golden orange,” or “golden tangerine,” meaning “golden luck”. The name “kumquat” is a combination of two words, “gold” and “good luck” (or “quat” in Cantonese), reflecting a belief that this fruit brings good luck and prosperity. The fruit was also considered as a symbol of wealth and prosperity due to its golden color and round shape. Kumquat is known by a variety of names in different languages, such as cumquat, jinju, and kinkan. In Türkiye, it is also known as “golden orange”.

The chemistry of essential oils of *Citrus fortunella* and its mutants studies the chemical compounds present in the essential oils of kumquat fruits and leaves, as well as any variations in these compounds that may occur in different kumquat cultivars or mutants. This research is important for several reasons. Firstly, understanding the composition and quality of kumquat essential oils is important for the food and beverage, cosmetics, and pharmaceuticals industries. Secondly, this research can provide insights into the genetic and environmental factors that influence the chemical composition of kumquat essential oils. Thirdly, it can become a basis for evaluating various kumquat by-products. Noteworthy, while some kumquat mutants have been found to have different chemical contents in their essential oils, further research is needed to fully understand the potential therapeutic benefits of these compounds and to confirm their effectiveness in preventing disease.

Kumquat, a tropical fruit that has both a delicious taste and a rich nutritional profile, is becoming increasingly popular in Türkiye. The cities of Antalya, Alanya, Anamur, Mersin, and Rize, which have hot climates, are especially well-suited for growing kumquats. There have been significant investments in kumquat production in these areas and people’s awareness about the fruit is also increasing. More and more people are consuming this healthy and delicious fruit. Despite its small size and delicate appearance, kumquat’s popularity in Türkiye is expected to rise. However, there is still a lack of research on the chemical content of kumquat and its by-products, which makes it difficult to provide accurate information about the nutritional profile and potential health benefits of this fruit. This also limits the awareness of kumquat’s potential uses. Therefore, more research is needed to explore the potential benefits of this fruit and increase the awareness of its potential as a medicinal plant.

With the increased production of kumquat in Türkiye, it is important to explore ways to make use of the chemical contents in its fruit and by-products. Producers should focus not only on cultivating kumquat fruit for consumption, but also on using its by-products, especially those considered as waste, as high value-added compounds. Yet, scientific literature lacks data on the essential oils from kumquat fruits and leaves grown in Turkey. Therefore, we aimed to determine the components of essential oils obtained by hydrodistillation from the fruit and leaves of *C. fortunella* rootstock species and 6 mutants in the Mersin region. Future studies should focus on isolating and producing active substances that may have financial value, and creating a roadmap to bring these products to market according to their properties. This will help to increase the competitiveness of kumquat in the market and create new opportunities for its use in different industries.

Study objects and methods

Plant materials. The *Citrus Fortunella* fruit and leaf samples used in the study were obtained from the Mersin Alata Horticultural Research Institute, the Ministry of Agriculture and Forestry, in January and November 2018. We studied the samples from the rootstock species (EP (Old Parcel); EP.4, EP.29, EP.31) and six mutants (YP (New Parcel); YP.117, YP.141, YP.188) (Fig. 1). The leaf samples were dried at room temperature and stored in a dry and cool environment. The fresh fruits were frozen, sliced into 2 mm thickness, and dried in a lyophilizer for 72 h. This method of preservation and drying ensured that the samples were in the best condition for the analyses.

Essential oil extraction procedure. Kumquat essential oils were obtained by hydrodistillation. For this, 20 g of dried plant material was mixed with 250 mL of distilled water and placed in a 500 mL round bottom flask. The mixture was distilled for three hours using



Figure 1. *Citrus Fortunella* fruit and leaf samples

Рисунок 1. Образцы плодов и листьев кумквата (*Citrus Fortunella*)

a Clevenger-type apparatus. The process was repeated three times in parallel for each plant material. After distillation, the essential oil was collected in a capillary tube and then transferred to a separating funnel. A small amount of *n*-hexane was added to the mixture and the essential oil was separated from the water. The oil was then transferred to a tared dark vial and stored at 4°C until analysis.

Gas chromatography-mass spectrometry. Gas chromatography-mass spectrometry (GC-MS) analysis was performed with an Agilent 5975 GC-MSD system (Agilent Technologies, Santa Clara, CA, USA). An Agilent Innowax FSC column (60 m×0.25 mm, 0.25 µm film thickness) was used with He as a carrier gas (0.8 mL/min). The GC oven temperature was kept at 60°C for 10 min, increased to 220°C at a rate of 4°C/min, kept constant at 220°C for 10 min, and then increased to 240°C at a rate of 1°C/min. The split ratio was adjusted to 40:1, and the injector temperature was 250°C. MS spectra were monitored at 70 eV, with a mass range of 35 to 450 m/z.

GC analysis was performed on an Agilent 6890N GC system. To obtain the same elution order as with GC-MS, the line was split for the flame ionization detector (FID) and the MS detector, and a single injection was performed using the same column and appropriate operational conditions. The FID temperature was 300°C. The essential oil components were identified by comparing their mass spectra with those in the Wiley GC/MS Library (Wiley, NY, USA) and Adams Library. A C₈–C₄₀ *n*-alkane standard solution was used to spike the samples to determine relative retention indices. Relative percentage amounts of the separated compounds were calculated by analyzing the FID chromatograms.

Results and discussion

The essential oil components of kumquat fruit and leaves were analyzed by comparing the mass spectra of each component to the mass spectra of the original samples from the Adams Library and literature (Table 1). The main component of kumquat fruit essential oil is limonene. Limonene is a terpene compound that is found

in high concentrations in many citrus fruits and is responsible for their characteristic citrus smell. The percentage of limonene in kumquat fruit essential oil can vary depending on the variety of kumquat, the method of extraction, and the growing conditions. It was reported that the percentage of limonene in kumquat fruit essential oil ranges from 69.9–94.4%.

The composition of essential oils in the *Citrus fortunella* leaves is different from that in the fruit samples. The main components of kumquat leaf essential oils are elemol (13.2–14.8%), β-eudesmol (9.3–11.0%), α-guaiol (8.5–10.8%), spathulenol (8.1–10.5%), and alismol (6.5–7.9%). These compounds are terpenoids that are typically found in high concentrations in essential oils of leaves and twigs of many plants. They are known for their medicinal properties such as anti-inflammatory, anti-cancerous, and anti-bacterial properties [13].

For our study, kumquat fruits and leaves were harvested at the Mersin Alata Horticultural Research Institute, the Ministry of Agriculture and Forestry, in January and November 2018. The essential oils were obtained by applying the hydrodistillation technique, which is commonly used to extract essential oils from plant materials. The chemical contents and composition of the essential oils were determined using GC and GC-MS analysis. Numerous components were identified in both the fruit and leaf essential oils of kumquat.

Limonene was the main component in the essential oil of kumquat fruit. Our results were in line with previous studies [14–16]. Limonene is a terpene compound that is found in high concentrations in essential oils of citrus fruits and many other plant species, including kumquat fruit. Limonene gives citrus fruits their characteristic citrus smell.

Limonene is a cyclic molecule (C₁₀H₁₆) that can exist in two symmetrical forms, D-Limonene and L-Limonene. The D-form is found in higher concentrations in the essential oil of citrus fruits [15], while the L-form is found in the essential oil of mint and other plants. Limonene has many applications as a flavoring agent in the food industry and perfumery, as well as in cleaning products. It is also used as an intermediate in the production of other compounds such as carvone. Limonene has been reported to have some medicinal properties such as anti-inflammatory and anti-cancerous effects [17–24]. Limonene is also a sweetening agent listed as Generally Recognized as Safe (GRAS) by the US Food and Drug Administration [25].

In addition to being used for aromatizing purposes in the cosmetics industry, limonene is also used in drug production to facilitate the *in vitro* and *in vivo* percutaneous passage of drugs [26–29]. Roberto *et al.* observed a greater antioxidant effect in lymphocytes exposed to 1–1000 µg/mL limonene and H₂O₂ at lower concentrations (10–50 µg/mL) than at higher concentrations (100–1000 µg/mL). Furthermore, the radical scavenging effect was similarly greater at lower concentrations

Table 1. Essential oil contents in *Citrus Fortunella* fruit and leaves
Таблица 1. Содержание эфирных масел в плодах и листьях кумквата (*Citrus Fortunella*)

RRI	Compound	EP4 - Leaf extracts	EP29- Leaf extracts	EP31- Leaf extracts	YP.117- Leaf extracts	YP.141- Leaf extracts	YP.188- Leaf extracts	EP.4- Extracts from lyophilized fruit	EP.29- Extracts from lyophilized fruit	EP.31- Extracts from lyophilized fruit	YP.117 - Extracts from lyophilized fruit	YP.141- Extracts from lyophilized fruit	YP.188- Extracts from lyophilized fruit	YP.117- Extracts from fresh fruit	Kumquat- Leaf	Kumquat- Lyophilized
900	Nonane	GS780A	GS780B	GS780C	GS780D	GS780E	GS780F	GS780G	GS780H	GS780I	GS780K	GS780L	GS780M	GS780N	GS780P	GS780R
931	2-Propanol										t	0.1	t	t	1.2	
965	1-Methylcyclohexa- 1,3-diene									t	t					
1000	Decane												t			
1024	2-Butanol														0.9	
1032	α -Pinene							0.3	0.3	0.4	0.4	0.4	0.4	0.2		0.3
1035	α -Thujene											t				
1048	2-Methyl-3-buten- 2-ol	0.2								t	t		t	0.1		t
1076	Camphene									t	t		t			
1093	Hexanal									t	t			t		
1100	Undecane									t	t			t		
1118	β -Pinene									t	t	t	t	t		t
1132	Sabinene									t	0.1	t	t	t		t
1159	δ -3-Carene												t			
1174	Myrcene							1.3	1.7	1.7	1.8	1.8	1.8	1.1		1.6
1176	α -Phellandrene											t	t			
1188	α -Terpinene												t			t
1203	Limonene					4		82.3	94	93.8	94.4	91.4	92.3	69	0.2	92.5
1218	β -Phellandrene							0.1	0.2	0.2	0.2	0.3	0.3	0.1		0.2
1223	<i>p</i> -Mentha-1,3,6- triene									t			t	t		t
1224	<i>o</i> -Mentha-1(7),5,8- triene								t		t	t				
1225	(Z)-3-Hexenal	0.6		0.1									t	t	0.4	
1253	<i>trans</i> - Anhydrolinalool oxide									t	t		t			t
1255	γ -Terpinene										t	t	t			t
1266	(E)- β -Ocimene										t		t			
1280	<i>p</i> -Cymene							0.1	t	0.1	t	t	t	t		0.1
1290	Terpinolene									t	t	t	t	t		t
1300	Tridecane										t					

Continuation of Table 1

RRI	Compound	EP4 - Leaf extracts	EP29- Leaf extracts	EP31- Leaf extracts	YP.117- Leaf extracts	YP.141- Leaf extracts	YP.188- Leaf extracts	EP4- Extracts from lyophilized fruit	EP29- Extracts from lyophilized fruit	EP31- Extracts from lyophilized fruit	YP.117 - Extracts from lyophilized fruit	YP.141- Extracts from lyophilized fruit	YP.188- Extracts from lyophilized fruit	YP.117- Extracts from fresh fruit	Kumquat- Leaf	Kumquat- Lyophilized
		GS780A	GS780B	GS780C	GS780D	GS780E	GS780F	GS780G	GS780H	GS780J	GS780K	GS780L	GS780M	GS780N	GS780P	GS780R
1280	<i>p</i> -Cymene							0.1	t	0.1	t	t	t	t		0.1
1290	Terpinolene									t	t	t	t	t		t
1300	Tridecane										t					
1319	(<i>E</i>)-2,6-Dimethyl- 1,3,7-nonatriene									t	t					
1320	Hydroxyacetone									t				t		
1327	3-Methyl-2-butenol									t	t					
1329	Pinol									t	t	0.1	t	t		t
1360	Hexanol	0.2	0.3	0.1										t	0.3	
1385	Heptyl acetate										t					
1400	Nonanal												t			
1400	Tetradecane										t		t			
1408	1,3,8- <i>p</i> -Menthatriene										t		t			
1410	4,8-Dimethyl-1,3,7- nonatriene									t	t			t		
1412	(<i>E</i>)-2-Hexenol	0.1												t		
1429	Perillen									t	t			t		
1443	2,5- Dimethylstyrene												0.1			
1446	2,6-Dimethyl- 1,3(<i>E</i>),5(<i>Z</i>),7- octatetraene												t			
1450	<i>trans</i> -Linalool oxide (Furanoid)											0.1				
1452	<i>α</i> , <i>p</i> -Dimethylstyrene							0.1	0.1	0.1	0.1			0.1		0.2
1458	<i>cis</i> -1,2-Limonene epoxide							0.3	t	0.2	0.1	t		0.2		0.1
1466	<i>α</i> -Cubebene										t			t		
1468	<i>trans</i> -1,2-Limonene epoxide							0.1		0.1	0.1	t		0.1		0.1
1479	<i>δ</i> -Elemene	0.2	0.3	0.4	0.5		0.4	0.1						0.2	0.2	
1483	Octyl acetate								0.1	0.1	0.1	0.1	0.1			0.1
1493	<i>α</i> -Ylangene	t	t	0.1	0.1		0.1				t			t		
1495	Bicycloelemene		t		0.1		t						t			
1497	<i>α</i> -Copaene	0.1	t	0.1	0.1		0.1			t	t	t	t	t		t

Continuation of Table 1

RRI	Compound	EP4 - Leaf extracts	EP29- Leaf extracts	EP31- Leaf extracts	YP117- Leaf extracts	YP141- Leaf extracts	YP188- Leaf extracts	EP4- Extracts from lyophilized fruit	EP29- Extracts from lyophilized fruit	EP31- Extracts from lyophilized fruit	YP117 - Extracts from lyophilized fruit	YP141- Extracts from lyophilized fruit	YP188- Extracts from lyophilized fruit	YP117- Extracts from fresh fruit	Kumquat- Leaf	Kumquat- Lyophilized
1500	Pentadecane				GS780D	GS780E	GS780F	GS780G	GS780H	GS780J	GS780K	GS780L	GS780M	GS780N	GS780P	GS780R
1506	Decanal															
1528	α -Bourbonene	t	t	t	t		t	t			t	0.1	0.1	t		0.1
1532	Camphor									t	t		t			
1535	β -Bourbonene	0.1	0.2	0.2	0.3		0.4	0.1			t		t	0.1	0.1	
1549	β -Cubebene	0.1			0.1			t		t	t		t	t		
1550	<i>cis</i> -Dihydro- α -terpineol	t														
1553	Linalool							0.2	0.2	0.1	0.1	0.1	0.2	0.2		0.1
35	8,9-Limonene epoxide I									t	t			t		t
1562	Octanol									t	t		t	t		
1568	1-Methyl-4- acetylcyclohex-1- ene*							t		t	t		t	t		t
1572	8,9-Limonene epoxide II							t		t	t			t		t
1589	β -Ylangene				0.2						t		t			t
1600	Hexadecane												t			
1600	β -Elemene	1.2	2.4	2.2	3	1.1	3	0.4	t	t	t	t	t	0.7	1.2	t
1610	Calarene (= β -gurjunene)									t				t		
1608	β -Copaene	0.1	t		t					t	t			t		t
1611	Terpinen-4-ol							t	0.1	0.1	0.1	0.1	0.1	0.1		0.1
1612	β -Caryophyllene	1.1	2.4	2.3	3.1	1.6	3.3	0.2					t	0.3	1.1	
1616	Hotrienol									t	t		t			t
1617	6,9-Guaiadiene		t													
1624	<i>trans</i> - Dihydrocarvone									t	t					
1628	Aromadendrene				0.2											
1637	<i>p</i> -Menth-1-en-9-al									t		t				0.1
1638	β -Cyclocitral															
1639	<i>trans-p</i> -Mentha-2,8- dien-1-ol							0.1		0.1	0.1		t	0.1		0.1
1641	<i>cis</i> - β -Terpineol							0.1	0.1	0.1	t	0.1	0.1	0.2		

Continuation of Table 1

RRI	Compound	EP4 - Leaf extracts	EP29- Leaf extracts	EP31- Leaf extracts	YP.117- Leaf extracts	YP.141- Leaf extracts	YP.188- Leaf extracts	EP4- Extracts from lyophilized fruit	EP29- Extracts from lyophilized fruit	EP31- Extracts from lyophilized fruit	YP.117 - Extracts from lyophilized fruit	YP.141- Extracts from lyophilized fruit	YP.188- Extracts from lyophilized fruit	YP.117- Extracts from fresh fruit	Kumquat- Leaf	Kumquat- Lyophilized
1650	γ -Elemene	0.4	0.6	0.5	0.8	0.6	1.1	t		t	t		t	0.1	0.3	t
1661	Alloaromadendrene			0.1												
1667	β -Guaiene	0.5														
1668	Citronellyl acetate									t	t	t	t			t
1669	Safranal									t	t	t	t			
1678	<i>cis-p</i> -Mentha-2,8- dien-1-ol							0.1	t	0.1	0.1	t	t	0.1		0.1
1687	α -Humulene	0.5	0.9	0.8	1.1	0.6	1.1	0.1	t		t	t	t	0.2	0.5	
1688	Selina-4,11-diene (=4,11-Eudes- madiene)															
1690	Cryptone									t	t					t
1694	Drima-7,9(11)-diene			0.1	0.1			t						t		
1695	(<i>E</i>)- β -Farnesene							t	t							
1696	γ -Terpinyl acetate											t				
1700	Heptadecane											t	t			
1700	<i>p</i> -Mentha-1,8-dien- 4-ol (= <i>Limonen-4-ol</i>)									t	t		t	t		t
1704	γ -Murolene								t							t
1705	Zizanene		0.9	0.9	1.3	0.9	1.3	t						0.2	0.9	
1706	α -Terpineol							0.8	0.2	0.4	0.2	0.5	0.6	0.5		0.2
1707	δ -Selinene		0.2		0.2											
1708	Ledene							t		t	t	t				t
1715	Geranyl formate									t			t			t
1715	γ -Terpineol												t			
1722	Dodecanal									t			t			
1726	Germaene D	1.2	0.7	0.8	1.2	0.7	0.8	0.1	0.5	0.2	0.2	0.6	0.6	0.1	1.5	0.4
1737	(<i>Z,E</i>)- α -Farnesene									t						
1738	<i>p</i> -Mentha-1,5-dien- 8-ol												t			
1740	α -Murolene	0.9		0.1	0.2			t		t	t	t	t	t		t
1740	Valencene		t		t							0.1	t	t		
1741	<i>cis</i> - α -Bisabolene									t						
1742	β -Selinene	0.9	1.5	1.5	1.6	0.7	1.9	0.4			t		t	0.6	0.8	t

Continuation of Table 1

RRI	Compound	EP4 - Leaf extracts	EP29- Leaf extracts	EP31- Leaf extracts	YP117- Leaf extracts	YP141- Leaf extracts	YP188- Leaf extracts	EP4- Extracts from lyophilized fruit	EP29- Extracts from lyophilized fruit	EP31- Extracts from lyophilized fruit	YP117 - Extracts from lyophilized fruit	YP141- Extracts from lyophilized fruit	YP188- Extracts from lyophilized fruit	YP117- Extracts from fresh fruit	Kumquat- Leaf	Kumquat- Lyophilized
		GS780A	GS780B	GS780C	GS780D	GS780E	GS780F	GS780G	GS780H	GS780J	GS780K	GS780L	GS780M	GS780N	GS780P	GS780R
1743	<i>α</i> -Cadinene										t					
1744	<i>α</i> -Selinene	0.2	0.3	t	0.2			t		t			t	0.1		t
1747	<i>trans</i> -Carvyl acetate												t			t
1751	Carvone							0.6			0.1			0.5		
1755	Bicyclogermacrene	0.2	0.9	0.5	0.8	0.5			0.1		t	t	0.1		0.2	t
1758	(<i>E,E</i>)- <i>α</i> -Farnesene											t	t			
1765	Geranyl acetate							t	0.5	0.4		0.4	0.3			0.5
1773	<i>δ</i> -Cadinene	0.3	0.5	0.4	0.5	0.3	0.6	t	t	t	t	0.1	0.1	t	0.4	0.1
1776	<i>γ</i> -Cadinene	0.2	0.1	0.1	0.2		0.2	t		t	t	t	t	0.1	0.1	t
1779	Bourbon-11-ene	t														
1782	<i>cis</i> -Carvyl acetate									t	t					
1796	Selina-3,7(11)-diene															
1796	Cabreuva oxide-V										t					
1797	<i>p</i> -Methyl acetophenone							t		t	t		t	t		t
1799	Cadina-1,4-diene (=Cubenene)												t			
1807	Perilla aldehyde							0.1	0.1	t	0.1	t	0.1	0.1		0.1
1808	Cabreuva oxide-VI										t	t	t			
1811	<i>trans</i> - <i>p</i> -Mentha- 1(7),8-dien-2-ol							t								
1824	Cabreuva oxide-VI									t						
1825	Geranyl propionate									t	0.3	t	t			t
1845	<i>trans</i> -Carveol							0.4	0.2	0.3	0.2	0.2	0.1	0.4	0.5	0.3
1853	<i>cis</i> -Calamenene				0.4		t	0.1		t	t					t
1853	Itallicene ether				t											
1854	Germacrene-B	0.4	0.6	0.4	0.2		0.4									
1857	Geraniol								t	t	t	t	t	t		t
1864	<i>p</i> -Cymen-8-ol									t	t	t	t			t
1865	Isopiperitenone							0.1			t					
1871	<i>p</i> -Mentha-1,8-dien- 10-yl acetate							t	t	t	t	t	t			t
1882	<i>cis</i> -Carveol							0.2	0.1	0.1	t	0.1	0.1	0.2		0.1

Continuation of Table 1

RRI	Compound	EP4 - Leaf extracts	EP29- Leaf extracts	EP31- Leaf extracts	YP.117- Leaf extracts	YP.141- Leaf extracts	YP.188- Leaf extracts	EP4- Extracts from lyophilized fruit	EP29- Extracts from lyophilized fruit	EP31- Extracts from lyophilized fruit	YP.117 - Extracts from lyophilized fruit	YP.141- Extracts from lyophilized fruit	YP.188- Extracts from lyophilized fruit	YP.117- Extracts from fresh fruit	Kumquat- Leaf	Kumquat- Lyophilized
		GS780A	GS780B	GS780C	GS780D	GS780E	GS780F	GS780G	GS780H	GS780J	GS780K	GS780L	GS780M	GS780N	GS780P	GS780R
1896	<i>cis-p</i> -Mentha- l(7),8-diene-2-ol									t	t					
1900	<i>epi</i> -Cubebol			0.1											0.4	
1907	<i>α</i> -Agarofuran			0.1										0.1		
1916	Perilla acetate									t	t	t				t
1941	<i>α</i> -Calacorene	0.2	0.2	0.2	0.3		0.3			t	t		t	t		t
1945	1,5-Epoxy-salvial- (4)14-ene	0.9	0.8	1.3	1.2	0.7	1.2	0.1			t			0.1	0.7	
1953	Palustrol												t			
1957	Cubebol				0.2			t								
1981	<i>cis</i> -Guai-6-en-10-ol	1.9			0.1		2.5								1.7	
1984	<i>γ</i> -Calacorene						0.1			t	t			t		
2000	Eicosane												t			
2001	Isocaryophyllene oxide		2.2					0.1						0.2		
2008	Caryophyllene oxide	3	4.4	4.3	3.7	4.5	4.4	0.7						1.2	3.4	
2008	<i>p</i> -Mentha-1,8-dien- 10-ol									t						
2028	Junenol								0.1	0.1	t	0.1	0.1			0.1
2029	Perilla alcohol									t	t		t			t
2037	Salvial-4(14)-en- 1-one	0.5	0.2	0.3	0.3		0.2	t						0.1	0.7	
2050	(<i>E</i>)-Nerolidol	4	4.9	4	3.9	5.9	4	0.4	0.1	t	t	t	t	0.9	4.2	
2057	Ledol				0.1						t	t	t			
2071	Humulene epoxide-II	0.6	0.8	0.8	0.7	0.7	0.7	0.2						0.3	0.7	
2074	Cubeban-11-ol									t	t		t			t
2080	Cubenol												t			t
2081	1,10-di- <i>epi</i> -Cubenol	0.3														
2096	Elemol	14.8	13.4	14.8	14.1	13.2	13.8	1.4						3	14.6	
2097	Eurolargone B	0.8	0.8	1.1	1											
2097	Salviadienol	0.7		t	0.4									0.2	0.8	
2098	Globulol							0.1		t	t			0.1		t
2100	Henicosane									t	t	t	t			t
2104	Viridiflorol					0.8	0.8	0.1		t	t	t	t	0.2	0.8	t

Continuation of Table 1

RRI	Compound	EP4 - Leaf extracts	EP29- Leaf extracts	EP31- Leaf extracts	YP117- Leaf extracts	YP141- Leaf extracts	YP188- Leaf extracts	EP4- Extracts from lyophilized fruit	EP29- Extracts from lyophilized fruit	EP31- Extracts from lyophilized fruit	YP117 - Extracts from lyophilized fruit	YP141- Extracts from lyophilized fruit	YP188- Extracts from lyophilized fruit	YP117- Extracts from fresh fruit	Kumquat- Leaf	Kumquat- Lyophilized
2115	4-Hydroxy-4-methyl-cyclohex-2-enone	GS780A	GS780B	GS780C	GS780D	GS780E	GS780F	GS780G	GS780H	GS780J	GS780K	GS780L	GS780M	GS780N	GS780P	GS780R
2127	10- <i>epi</i> - γ -Eudesmol		1.0	1.2	0.9	1.0	1.2	0.1		t	t		t	0.2		
2144	Rosifolol	0.1			0.2					t	t	t	t			t
2144	Spathulenol	9.7	9.4	9.2	8.1	10.4	8.6	1.0	0.1	t	t	t	t	2.3	10.5	0.1
2156	α -Bisabolol oxide B									t	t		t			t
2185	γ -Eudesmol	1.9	1.4	1.5	1.6	1.7	1.6	0.3	0.1	0.1	t	0.1	0.1	0.5	1.4	0.1
2187	T-Cadinol	2.2	2.5	2.4	2.3	2.6	2.5	0.3						0.6		
2190	Eremoligenol	5	5	4.7	4.6	5.4	4.8	0.4		t	t		t	0.7	5.3	t
2192	α -Guaiol	9.4	9.9	9.5	8.5	10.8	9.6		0.1	t	t	t	t	2.5	10.02	
2200	Docosane											0.1	t			t
2209	T-Murolol							0.1		t	t	0.1	0.1			0.1
2226	Methyl hexadecanoate (=methyl palmitate)										t	t	t			t
2237	Valerianol	2.3	2.5	2.3	2.2	2.4	2.3	0.3		t	t			0.5	2.2	t
2250	α -Eudesmol	4.0	4.3	3.8	3.8	4.8	4.2	0.3		t	t	t	t	0.7	3.7	t
2255	α -Cadinol				1.7			0.2	0.1	0.1	t	0.1	0.1			0.1
2256	Torilenol	0.8													0.7	
	(=1-hydroxy-6,8-cyclo-4(14)-eudesmene)															
2257	β -Eudesmol	10.2	10.8	10	9.3	11	10.6	1.6		t	t	t	t	3.3	9.9	t
2289	Alismol (=Guaiol-6,10(14)-diene-4 β -ol)	6.8	6.8	6.5	6.6	7.9	6.8	0.8		t	t	t	t	1.6	7.0	t
2273	Selin-11-en-4 α -ol									t	t	t				t
2298	Decanoic acid											t	t			
2300	Tricosane							0.1	0.1	t	t	0.2	0.1	0.1		0.1
2320	Juniper camphor												t			
2368	Eudesma-4(15),7-diene-1 β -ol	1.2	0.5	0.7	0.6		0.5	0.1						0.3	1.4	t

End of Table 1

RRI	Compound	EP4 - Leaf extracts	EP29- Leaf extracts	EP31- Leaf extracts	YP117- Leaf extracts	YP141- Leaf extracts	YP188- Leaf extracts	EP4- Extracts from lyophilized fruit	EP29- Extracts from lyophilized fruit	EP31- Extracts from lyophilized fruit	YP117 - Extracts from lyophilized fruit	YP141- Extracts from lyophilized fruit	YP188- Extracts from lyophilized fruit	YP117- Extracts from fresh fruit	Kumquat- Leaf	Kumquat- Lyophilized
2392	Caryophylla- 2(12),6-dien-5 β -ol (=Caryophyllenol II)	GS780A	GS780B	GS780C 0.4	GS780D	GS780E	GS780F	GS780G	GS780H	GS780J	GS780K	GS780L	GS780M	GS780N	GS780P	GS780R
2400	Tetracosane									t	t	0.3	0.1			t
2456	(Z)-9-Methyl octadecanoate (=Methyl oleate)										t	t				t
2462	Eudesma-4(15),7- diene-1-ol isomer I	1	0.4	0.7	0.6											
2500	Pentacosane									t	t	0.4	0.1	0.1		0.1
2503	Dodecanoic acid									t	t		t	t		t
2509	(Z,Z)-9,12-Methyl octadecadienoate (=Methyl linoleate)									t	t		t			t
2583	Methyl linolenate									t	t		t			t
2600	Hexacosane										t	0.3	0.1			
2604	α -Costol				0.1											
2606	β -Costol				0.2											
2622	Phytol			0.2	0.2									0.1		
2630	epi-Nootkatol*	0.2														
2637	2 α -Hydroxy- amorpha-4,7(11)- diene*	0.2														
2670	Tetradecanoic acid (=Myristic acid)													t		
2694	14-Hydroxy- calamenene													t		
2700	Heptacosane									t	t	0.3	0.1	t		
2800	Octacosane											0.2				
		92.4	95	91.8	94	94.2	96.3	97.5	99.2	98.9	98.8	98.9	98.4	95.9	90.92	98.2

Note: *Tentative identification from Adams.

Примечание: *Предварительные показатели взяты из библиотеки данных Adams.

(10–50 µg/mL) than at higher concentrations (100–1000 µg/mL) [30]. This suggests that limonene has antioxidant properties and may be able to protect cells from oxidative stress caused by hydrogen peroxide. However, it is important to note that more studies are needed to fully understand the mechanisms of antioxidant activity of limonene and its potential therapeutic applications.

Choi studied the essential oil of *Fortunella japonica* Swingle peel using GC and GC-MS analysis [31]. In the study, myrcene was found to be the second dominant component in the essential oil, with a concentration of 1.84%. This value is in agreement with myrcene concentrations in our study (1.10–1.80%). Myrcene is a terpene compound that is commonly found in essential oils of many plants and is known for its sedative and anti-inflammatory properties [31]. As seen in Table 1, the main components of leaf essential oils of kumquat are elemol (13.2–14.8%), β -eudesmol (9.3–11.0%), α -guaiol (8.5–10.8%), spathulenol (8.1–10.5%) and alismol (6.5–7.9%). These are terpenoids that are typically found in high concentrations in essential oils of leaves and twigs of many plants. It is important to note that the essential oil of kumquat leaf has its own unique composition and properties, different from those of the fruit essential oil.

Elemol is a terpenoid compound that is found in the leaves of certain plants. It has been found to have insecticidal and antitermite properties, i.e., it can be used to kill or repel insects and termites [32]. This makes elemol a potential candidate for use in pesticides and other insect control products. The essential oil of kumquat leaves contains 13.2–14.8% of elemol. However, the yield of essential oil from kumquat leaves is relatively low compared to the yield from other plant parts. Therefore, for kumquat leaves to become a valuable commercial source of elemol, efforts have to be made to increase the yield of leaf essential oil.

β -Eudesmol, the second major component of kumquat leaf essential oil, has been found to have anti-cancerous properties. Studies have shown that it can inhibit the formation and development of various types of cancer, including breast cancer and leukemia. However, more research is needed to fully understand the anti-cancerous properties of β -eudesmol and its potential in cancer prevention. In addition, essential oils are not recommended for internal use without proper dilution and professional guidance, as they can be toxic if taken in large doses or used improperly [33].

Guaiol is a hydroxyl sesquiterpene that is found in the essential oils of many medicinal plants, including kumquat leaves. It has a guaiane skeleton and is a key metabolite in the biogenesis of many guaiane natural products. Guaiol has a long history of use as a natural remedy. It is known for its antimicrobial, antifungal, antioxidant, antibacterial, antitumorous, anti-inflammatory, and insecticidal properties [34–37].

Spathulenol is a tricyclic sesquiterpenoid found in the essential oils of many plant species, including kumquat

leaves. It has an aromadendrane carbon skeleton and is known for its strong aroma. Although research on the biological activity of spathulenol is limited, it has been reported to exhibit antimicrobial, anti-inflammatory, antioxidant, and insecticidal properties [38–43].

Alismol is a sesquiterpenoid compound that is found in the essential oil of kumquat leaves. In our study, it was detected at levels between 6.5% and 7.9% in the essential oils of kumquat leaves. Alismol was first isolated from the rhizomes of *Alisma orientale* by Oshima *et al.* in 1983 [44]. Some studies have shown that sesquiterpenoids isolated from the rhizomes of *A. orientale* (a species of ginger) and various parts of the Lauraceae family exhibit enhanced biological activity [45–49].

Thus, we analyzed the essential oils obtained from the fruits and leaves of *C. fortunella* species collected in the Mersin region. The chemical composition of these essential oils has not been previously reported in literature. We determined the chemical content of the essential oils and identified a variety of compounds. A limited literature review showed that these chemicals have potential uses in the prevention of various diseases. However, more research is needed to fully understand the potential therapeutic benefits of these compounds and to confirm their effectiveness in preventing disease. Our results can be used not only to provide information about the nutritional value of kumquat when consumed as fresh fruit, but also to form a basis for evaluating various kumquat by-products. This may include the use of kumquat essential oil in food and beverage, cosmetics, and pharmaceuticals industries. Further research will provide a deeper understanding of the potential benefits of kumquat for human health and a more comprehensive use of kumquat resources.

Conclusion

In this study, we determined the chemical components of the essential oils obtained from *Citrus fortunella* fruits and leaves (Mersin region, Türkiye) by GC-MS. We found that the kumquat fruit essential oil was rich in limonene, while the leaf essential oil contained large amounts of elemol, β -eudesmol, α -guaiol, spathulenol, and alismol. Thanks to these beneficial volatile oil components, kumquat is as valuable as other citrus fruits in a variety of applications for medical, pharmaceutical, aromatic, and other purposes. However, since the safety of chemical preservatives in the food, cosmetic, and agricultural industries has been questioned, further studies are needed to evaluate kumquat fruit and leaf essential oils as alternatives for use in various industries.

Contribution

The authors were equally involved in writing this article and are equally responsible for its content. Fatümetüzzeşra Küçükbay designed the study and managed the data collection process. Çağrı Büyükkorkmaz, Gülmira Özek, and Temel Özek analyzed and interpreted

ted the data. All the authors prepared the article for publication and reviewed the draft of the article. All the authors read and approved the article.

Conflict of interest

The authors declare no conflict of interest regarding this work.

Acknowledgments

The authors thank İnönü University, Turkey for financial support (BAPB – Grant No. TYL-2018-1108).

Критерии авторства

Ф. Кючукбай – концепция исследования, руководство сбором данных, написание статьи. Ч. Бююк-

коркмаз, Г. Озек и Т. Озек – анализ и интерпретация данных, написание статьи. Авторы в равной степени участвовали в написании статьи и несут ответственность за ее содержание. Все авторы одобрили финальную версию статьи.

Конфликт интересов

Авторы заявили об отсутствии потенциальных конфликтов интересов в отношении исследования, авторства и / или публикации данной статьи.

Благодарности

Авторы выражают благодарность университету Инёню, Турция, за финансирование проекта (BAPB, грант № TYL-2018-1108).

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