KSÜ Tarım ve Doğa Derg 25 (Ek Sayı 1): 11-21, 2022
KSU J. Agric Nat 25 (Suppl 1): 11-21, 2022
https://doi.org/10.18016/ksutarimdoga.vi.1001837

Determination of Antimicrobial Activity of *Nasturtium officinale* and Its Content of Volatile Organic Compounds and Fatty Acids

Leyla ERCAN [1], Mehmet DOĞRU [2]
[1]Mardın Artuklu Unıversıty Central Research Laboratory, 47000 Mardin, Türkiye, [2] Dicle University, Faculty of Ziya Gökalp Education, Diyarbakır, 21280, Türkiye
[1]https://orcid.org/0000-0002-6570-8128, [2]https://orcid.org/0000-0002-2287-2913
✉: leylaercan@artuklu.edu.tr

ABSTRACT

Due to the side effects of antibiotics used in the treatment of diseases caused by pathogenic bacteria, and antibiotic resistance that develops due to the misuse of antibiotics, scientists have turned to the search for alternative antimicrobial compounds. Plants and antimicrobial compounds in plants are widely researched because they are natural and have been a familiar resource in the field of complementary medicine for centuries. In this study, antimicrobial activities of the methanol and water extracts of *Nasturtium officinale* prepared at different concentrations were investigated on gram-positive bacteria, gram-negative bacteria, and fungi by the disc diffusion method. In addition, volatile organic compound and fatty acidcontent of the plant were determined. For this purpose, fatty acids were determined by converting them to methyl esters in GC-FID (gas chromatography flame ionization detector), volatile compounds were determined by SPME (Solid-phase microextraction) method in GC-MS (gas chromatography-mass spectrometry). In addition, the amounts of volatile components in different parts of the plant were shown comparatively within the scope of the research. According to the results obtained; it was revealed that *Nasturtium officinale* has an antimicrobial effect on *Bacillus megaterium*, *Escherichia coli*, *Candida albicans*, *Pseudomonas aeruginosa*, *Bacillus Spizizenii*, *Klebsiella pneumoniae*, *Staphylococcus aureus* bacteria. The plant showed a stronger antimicrobial effect, especially on *P. aeruginosa*, *C. Albicans, and E. coli*. It has also been determined that *Nasturtium officinale* has important essential fatty acids as well as many volatile components. In the analyzes made, it was determined that the main volatile component of *Nasturtium officinale* was alpha-Terpinolene.

Biochemistry

Research Article

Article History
Received : 30.09.2021
Accepted : 14.01.2022

Keywords
Antimicrobial
Volatile compounds
Nasturtium officinale
Alpha terpinolene
Fatty acids

Nasturtium officinale'nin Antimikrobiyal Aktivitesinin ve İçeriğindeki Uçucu Organik Bileşikler ve Yağ Asitlerinin Belirlenmesi

ÖZET

Patojen bakterilerin neden olduğu hastalıkların tedavisinde kullanılan antibiyotiklerin gerek yan etkileri, gerekse yanlış kullanımına bağlı olarak gelişen antibiyotik direnci nedeniyle bilim insanları alternatif antimikrobiyal bileşik arayışına yönelmiştir. Doğal olması ve yüzyıllardır tamamlayıcı tıp alanında tanıdık bir kaynak olması sebebiyle bitkiler ve bitkilerdeki antimikrobiyal bileşenler yaygın bir şekilde araştırılmaktadır. Bu çalışmada, *Nasturtium officinale*'nin farklı konsantrasyonlarda hazırlanan metanol ve su ekstraktlarının gram-pozitif bakteriler, gram-negatif bakteriler ve mantar üzerindeki antimikrobiyal aktivitesi disk difüzyon yöntemiyle araştırıldı. Bunun yanısıra bitkinin içeriğindeki uçucu organik bileşikler ve yağ asitleri tespit edildi. Bu amaçla GC-FID'de (gaz kromatografisi alev iyonizasyon dedektörü) yağ asitleri metil esterlere dönüştürülerek belirlenirken, GC-MS'de (gaz

Biyokimya

Araştırma Makalesi

Makale Tarihçesi
Geliş Tarihi : 30.09.2021
Kabul Tarihi : 14.01.2022

Anahtar Kelimeler
Antimikrobiyal
Uçucu bileşenler
Nasturtium officinale
Alpha terpinolene
Yağ asitleri

kromatografisi - kütle spektrometrisi) SPME (Katı fazlı mikroekstraksiyon) yöntemi ile uçucu bileşenleri belirlendi. Ayrıca araştırma kapsamında bitkinin farklı kısımlarındaki uçucu bileşenler karşılaştırmalı olarak ele alındı. Elde edilen sonuçlara göre; *Nasturtium officinale*'nin *Bacillus megaterium, Escherichia coli, Candida albicans, Pseudomonas aeruginosa, Bacillus Spizizenii, Klebsiella pneumoniae, Staphylococcus aureus* bakterileri üzerinde antimikrobiyal etkisi olduğu ortaya çıktı. Bitki özellikle *P. aeruginosa*, *C. Albicans ve E. Coli* üzerinde daha güçlü antimikrobiyal etki göstermiştir. Ayrıca *Nasturtium officinale*'nin önemli esansiyel yağ asitlerinin yanısıra çok sayıda uçucu bileşenlere de sahip olduğu tespit edildi. Yapılan analizlerde *Nasturtium officinale*'nin ana uçucu bileşeninin alfa-Terpinolen olduğu belirlendi.

Atıf Şekli: Ercan L, Doğru M 2022. *Nasturtium officinale*'nin Antimikrobiyal Aktivitesinin ve İçeriğindeki Uçucu Organik Bileşikler ve Yağ Asitlerinin Belirlenmesi. KSÜ Tarım ve Doğa Derg 25 (Ek Sayı 1): 11-21. https://doi.org/10.18016/ksutarimdoga.vi.1001837
To Cite : Ercan L, Doğru M 2022. Determination of Antimicrobial Activity of *Nasturtium officinale* and Its Content of Volatile Organic Compounds and Fatty Acids. KSU J. Agric Nat 25 (Suppl 1): 11-21. https://doi.org/10.18016/ksutarimdoga.vi.1001837

INTRODUCTION

N. officinale, a member of the *Brassicaceae* family, is a rare aquatic plant with thin and fibrous roots, and oval baby leaves (Daniel, 2009). *N. officinale* is considered a valuable traditional medicinal plant due to its many health-beneficial components such as vitamins B, C, and E, pro-vitamin A, folic acid, carotenoids, glucosinolates (Gonçalves et al., 2009; Pourhassan-Moghaddam et al, 2014). It is known that watercress (*N. officinale*) leaves are used in anti-inflammatory, diuretic, expectorant, hypoglycemic, antihypertensive, urinary tract infections, and cardiovascular diseases (Amiri, 2012; Shahani et al., 2016).

Plants that have been used in the treatment of various diseases for centuries are still recommended in the treatment of many diseases, especially as a supplement. Due to the high cost of drugs used in the treatment of diseases, as well as the risk of unwanted side effects, researchers around the world are trying to identify effective drugs with minimal side effects to overcome the difficulties associated with the unreliability of modern pharmaceuticals (Adokoh et al., 2019; Abd Rashid et al., 2021). Recently, ways to eliminate many bacteria that cause various infections and food poisoning without using antibiotics are being investigated (Şengün and Öztürk, 2018).

The use of herbal products in the industry, health, textile, food, cosmetics, and the increasing trend towards natural products have led to the examination of plants from all aspects (Varlı et al., 2020). For this purpose, plants and their bioactive components, fatty acids, volatile components, and their effects have begun to be investigated more (Varlı et al., 2020). Some volatile compounds obtained from aromatic plants are used in medicine and pharmacology as antimicrobial, anti-inflammatory, antioxidant, expectorant, analgesic, and in the treatment of many ailments and are also effective in defense against herbivores and pathogens (Pichersky et al., 2006; Maffei et al., 2011; Yip et al., 2019). It has been shown in many studies that these compounds, which are synthesized by plants to attract pollinators and fight pests, have many beneficial effects such as anticarcinogen, anti-inflammatory, antidiabetes and neuroprotective, as well as being used as fragrance and food additives (Maffei et al., 2011; Pichersky et al., 2006). ; Vieira et al., 2018; Yip et al., 2019).

On the other hand, fatty acids are also among the important compounds contained in plants. It is also known that essential fatty acids, which cannot be synthesized in the body of humans and other mammals and must be taken from outside, play a key role in the prevention of many diseases such as heart attack, cardiovascular diseases, depression, migraine, arthritis, diabetes, high cholesterol, blood pressure, allergies, and cancer (Santos et al., 2017; Wassell et al., 2010). The essential fatty acids α-linolenic acid and linoleic acid are required for the synthesis of various molecules that affect vital functions (Das, 2006). According to previous research reports, it has been shown that polyunsaturated fats such as omega-3 play an important role in the prevention of coronary heart disease and some cancers (Pretorius and Schőnfeldt, 2021). Linolenic acid, one of the polyunsaturated fatty acids, has anticancer, antiosteoporotic, antioxidant, anti-inflammatory, and coronary protective properties (Santos et al., 2017; Martins et al., 2018). In addition to the many benefits of essential oils, many negative situations arise in the lack of these essential oils. It is important to reveal the volatile compounds and fatty acids contained in plants to find out their valuable aspects. Therefore, in this study, the volatile components, fatty acids, and antimicrobial effects of *N. officinale* were investigated.

MATERIAL and METHOD

Plant material

The *N. officinale* used in this study was collected from the natural environment in Kayseri, Turkey, and the plant samples were authenticated by Prof. Dr. Hasan AKAN. It is stored in herbarium number 6363 at Harran University.

Chemicals

Nutrient agar medium, broth medium (Condalab brand), amikacın (30µg), ampicillin-sulbactam (20 µg), rifampin (5 µg), and erythromycin (15 µg) were purchased from Bioanalyse. Methanol, DMSO (dimethylsulfoxide), and KOH were purchased from Sigma- Aldrich.

Tested Microorganisms

Tested Microorganisms; *Klebsiella aerogenes* ATCC 13048, *Pseudomonas aeruginosa* ATCC 9027, *Bacillus megaterium* ATCC 14581, *Escherichia coli* ATCC 11229, *Klebsiella pneumoniae* ATCC 13883, *Staphylococcus aureus* ATCC 25923, *Candida albicans* ATCC 10231, *Bacillus subtilis subsp. spizizenii* ATCC 6633, strains are purchased from Microbiologics.

Preparation and Analysis of Plant Extracts for Antimicrobial Activity Determination

To investigate the antimicrobial effects of *N. officinale*, two separate extractions were prepared with distilled water and methanol. Water extract was prepared by adding 200 mL of distilled water to 20 grams of plant samples, and methanol extract was prepared by adding 200 mL of methanol to 20 grams of plant samples. After filtration through filter paper, the water extract from the solutions was lyophilized while the methanol extract was evaporated. To determine the antimicrobial effect of the plant, solutions were prepared of lyophilized water extract and evaporated methanol extract with dimethyl sulfoxide (DMSO) at different concentrations (20, 40, 60 mg mL). The Disc Diffusion method was used to determine antimicrobial activity (Wayne, 1997). Microorganisms were incubated at 37°C in nutrient broth (NB) until 0.5 McFarland (1.5x108 Kob mL) turbidity occurred. Turbidity control was performed in a spectrophotometer at a wavelength of 625 nm with a between absorbance of 0.08-0.10. 100 µl of prepared test microorganisms were taken and inoculated in nutrient agar solid plates. Then, the solutions of the lyophilized water extracts and the evaporated methanol extracts prepared were absorbed into sterile discs in the inoculated plate. For *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus, Bacillus megaterium, Klebsiella pneumoniae, Bacillus spizenii,* and *Klebsiella aerogenes* bacteria 24 hours of incubation at 37°C were measured and for *Candida albicans* incubation at 30°C, inhibition was measured after 48 hours of incubation. The same procedure was repeated for the positive control erythromycin (15 µg), rifampin (5 µg), amikacin (30 µg), ampicillin-sulbactam (SAM) (20 µg), and the negative control (DMSO).

Determination of Volatile Components Using SPME-GCMS

The solid-phase microextraction (SPME) method is a simple method that allows direct volatile component analysis without extraction of the plant (Panighel and Flamini, 2014). 0.6 grams of *N. officinale* and leaf and stem parts of *N. officinale* were weighed each to determine the volatile organic compound content by the SPME (Solid Phase Micro Extraction) method in Gas Chromatography-Mass Spectrometry (GC-MS). Volatile organic compounds were determined by the SHIMADZU QP2020 brand GC-MS device. The column used in the analysis is DB-HEAVYMAX (60 m x 0.25 mm x 0.25 µm). The injection temperature is 250°C and the injection mode is split, the total flow rate is 1.05 mL min. The incubation temperature was set to 40°C. The SPME fiber used is 100 µm PDMS (polydimethylsiloxane).

Determination of Fatty Acid Methyl Ester Using GC-FID

To determination of fatty acid methyl ester in gas chromatography flame ionization detector (GC-FID), 20 grams of *N. officinale* plant was taken and 200 mL of hexane was added and mixed with a magnetic stirrer at room temperature for 12 hours. The solution was then filtered through a filter paper and the solvent was evaporated at 35°C in the evaporator until the final volume of the solution was 20 mL. It was taken from this extract 0.1 mL and mixed with 10 mL hexane and 0.5 mL of 2N KOH prepared in methanol was added and kept in the dark for 1-2 hours. Fatty acid methyl ester analysis was performed with SHIMADZU QP2020 brand GC-FID (Gas Chromatography-Flame Ionization Detector). Fatty acids were analyzed by converting them into methyl ester derivatives in the determination of fatty acids in GC-FID branded SHIMADZU QP2020. Rtx-2330 RESTEK (60 m x 0.25 mm x 0.2 µm) column was used for analysis. Injection temperature was set to 250°C, injection mode Split, pressure 100 kPa, Split Ratio 100. The injection volume was set to 1 µL (AOAC, 2012).

Statistical Analysis

All measurements were carried out in triplicate and the results were presented as mean values ± SD (standard deviations). Statistical analyzes between groups of antimicrobial analyzes were revealed by one-way ANOVA. $p<0.05$ was considered significant.

RESULTS and DISCUSSION

Antimicrobial Activity Results

Antimicrobial Efficacy of Methanol Extracts

Antimicrobial activity of the plant on gram-positive bacteria (*S. aureus, B. subtilis subsp. Spizizenii, B. megaterium*), gram-negative bacteria (*E. coli, K. pneumoniae, P. aeruginosa, K. Aerogenesis*), and fungi (*C. albicans)* have been examined. Negative control (DMSO) showed no antibacterial effect. The methanol extracts showed an antimicrobial effect on *K. pneumoniae, B. megaterium, E. coli, C. albicans, P. aeruginosa, B. Spizizenii, S. aureus* bacteria. Methanol extracts antimicrobial activity results (inhibition zone diameters) are given in Table 1.

Antimicrobial Efficacy of Water Extracts

Negative control (DMSO) did not show an antibacterial effect. The antimicrobial activity results (inhibition zone diameters) of the water extracts of *N. officinale* are given in Table 2.

Table 1. Antimicrobial activity results of the methanol extracts (mm)
Çizelge 1. Metanol ekstraktlarının antimikrobiyal aktivite sonuçları (mm)

Methanol extracts	Amikacin (AK-30)	Ampicillin -sulbactam (SAM- 20)	Rifampin (RD-5)	Erythromycin (E-15)	20 mg mL methanol extract	40 mg mL methanol extract	60 mg mL methanol extract
P. aeruginosa	18.02±0.01	15.23±0.01	10.9±0.03	12.61±0.03	9.39±0.07	10.63±0.03	11.69±0.05
B. megaterium	19.05±0.15	26.42±0.32	22.78±0.17	25.36±0.33	8.29±0.03	8.29±0.01	8.8±0.04
C. albicans	21.79±0.45	27.43±0.69	17.00±0.41	14.58±0.04	7.54±0.01	8.35±0.07	8.75±0.05
K. pneumoniae	19.01±0.07	17.30±0.21	12.52±0.03	16.41±0.09	-	7.67±0.03	7.86±0.09
E. coli	14.95±0.08	21.8 ±1.24	9.82±0.01	12.77±0.05	7.74±0.05	9.39±0.05	11.44±0.12
K. aerogenes	17.06±0.03	15.25±0.12	9.37±0.07	18.34±0.12	-	-	-
B. spizizenii	24.34±0.42	25.42±0.21	19.37±0.15	26.87±1.17	6.84±0.07	6.95±0.05	7.31±0.04
S. aureus	12.74±0.02	27.8±1.45	22.34±0.45	19.27±0.03	-	6.42±0.04	6.48±0.02

(-): No inhibition, ± SD (standard deviation)
(-): İnhibisyon yok, ± SD (standart sapma)

Table 2. Antimicrobial activity results of water extracts (mm)
Çizelge 2. Su ekstraktlarının antimikrobiyal aktivite sonuçları (mm)

Water extracts	Amikacin (AK-30)	Ampicillin -sulbactam (SAM- 20)	Rifampin (RD-5)	Erythromycin (E-15)	20 mg mL water extract	40 mg mL water extract	60 mg mL water extract
P. aeruginosa	20.75±0.12	16.7±0.07	10.9±0.08	12.61±0.07	-	9.16±0.04	10.41±0.07
B. megaterium	18.7±0.04	26.99±0.45	22.7±0.53	24.63±0.71	8.06±0.06	8.8±0.03	9.71±0.02
C. albicans	25.26±0.32	20.97±0.06	17.74±0.75	15.18±1.69	7.41±0.04	8.05±0.04	9.31±0.12
K. pneumoniae	20.12±0.09	16.12±0.05	11.84±0.01	15.42±0.26	-	-	-
E. coli	17.69±0.01	19.35±0.01	10.21±0.05	15.49±0.05	-	7.51±0.05	8.81±0.09
K. aerogenes	18.17±0.05	12.87±0.02	9.62±0.09	17.78±0.35	-	-	-
B. spizizenii	19.96±0.21	20.93±0.21	19.37±0.01	28.64±1.24	6.62±0.04	7.49±0.03	8.29±0.07
S. aureus	13.08±0.06	28.72±0.06	23.71±0.04	18.43±0.52	-	-	-

(-): No inhibition, ± SD (standard deviation)
(-): İnhibisyon yok, ± SD (standart sapma)

According to the results given in Table 2, the water extract of the plant has an antimicrobial effect on *B. megaterium, E. coli, C. albicans, P. aeruginosa, B. spizizenii* bacteria. However, the water extract started to inhibit *E. coli, P. aeruginosa* bacteria at a concentration of 40 mg mL. When the results are examined, it is seen that the methanol extracts of the plant have an antimicrobial effect on *K. pneumoniae, B. megaterium, E. coli, C. albicans, P. aeruginosa, B. spizizenii* bacteria. When the inhibition diameters and percent inhibition amounts were examined, water extracts of *N. officinale* showed more antimicrobial effect than methanol extracts on gram-positive bacteria except for *S. aureus*. Methanol extracts, on the other hand, showed more antimicrobial effect on gram-negative bacteria than water extracts, and also inhibited *S. aureus*, a gram-positive bacteria. While the water extracts of the plant did not inhibit *K. pneumoniae, S. aureus* bacteria, it began to inhibit the methanol extracts at a concentration of 40 mg mL. Comparison of antimicrobial activity results of methanol and water extracts of *N. Officinale* is given in Figure 1.

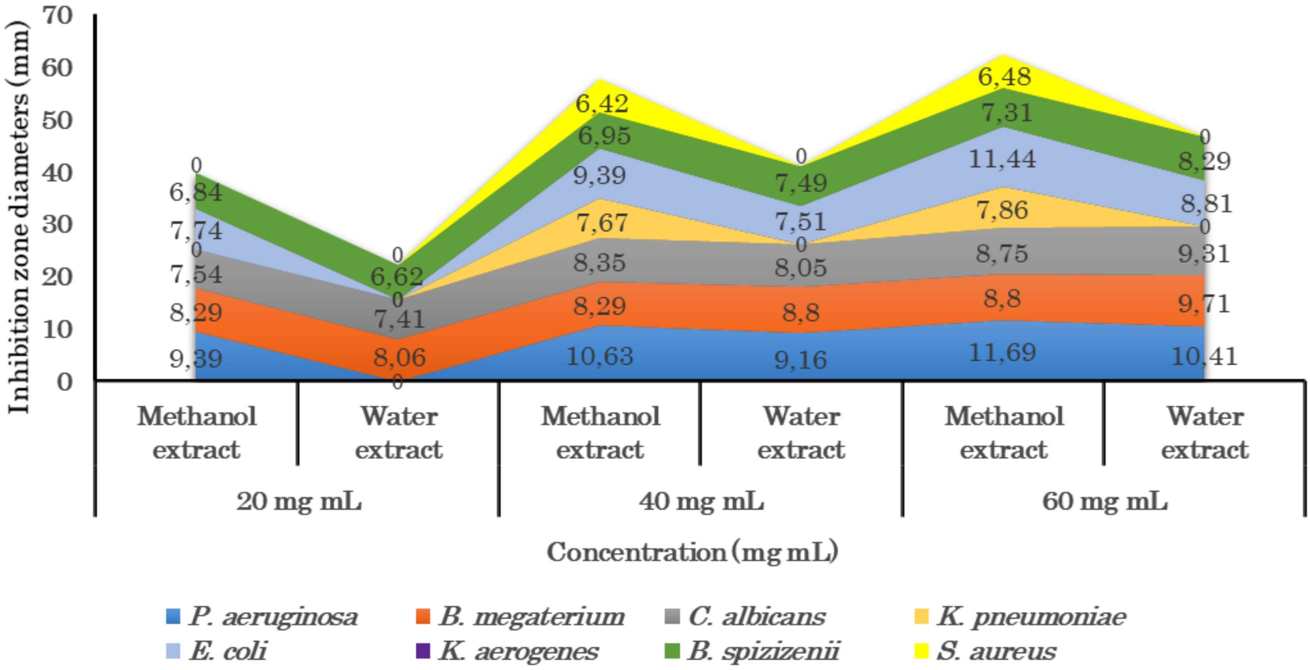

Figure1. Comparison of antimicrobial activities of methanol and water extracts of N. Officinale
Şekil 1. N. Officinale'nin metanol ve su ekstraktlarının antimikrobiyal aktivitelerinin karşılaştırılması

According to these results, methanol extracts have more antimicrobial effects because they inhibit more bacteria. Components that dissolve better in alcohol may have been effective on this result. From this point of view, it can be said that components with antimicrobial properties have a more apolar structure. In addition, methanol extracts at a concentration of 60 mg mL showed a stronger antimicrobial effect on *P. aeruginosa* and *E. coli* bacteria than Rifampin antibiotic. As a result, *N. officinale* showed an antimicrobial effect on *B. megaterium, E. coli, P. aeruginosa, B. Spizizenii, K. pneumoniae, S. aureus* bacteria, and *C. albicans*. The plant showed a better antimicrobial effect especially against *C. albicans, P. aeruginosa*, and *E. coli.* In a study on *Escherichia coli, Salmonella typhimurium*, *Staphylococcus aureus,* and *Listeria monocytogenes* bacteria, it was reported that aqueous and alcoholic extracts of *N. officinale* were more effective on gram positives and did not inhibit gram-negative bacteria (Derhami et al., 2017). In experiments, it was found that *N. officinale* was also effective on *K. pneumoniae, E. coli P. aeruginosa.* The difference between these results may be due to the time the plant was collected, the place where it was collected, as well as the possibility that the plant samples may lose some of their components during drying.

Volatile Organic Component Determination Results

The volatile organic compounds of *N. officinale* were determined and shown in Table 3.

According to these results, it was determined that *N. officinale* is rich in volatile components and contains many terpenes and terpenoid volatile components. It has been observed that most of these volatile components are composed of monoterpenes and *N. officinale* contains alpha-terpinolene (54.46%), which is a monoterpene, as the main component. It contains many monoterpenes such as D-limonene, gamma-terpinene, beta-phellandrene other than alpha-terpinolene, and many sesquiterpenes such as cadina-1(2), 4-diene, caryophyllene, β-sesquiphellandrene. Monoterpenes are among the volatile organic components of many medicinal plants. Very few volatile components were detected in the previous volatile component analysis of *N. Officinale* (Amiri, 2012). The reason for this may be that the plant loses its volatile components depending on the collection time or storage conditions. Because the plant samples used in this study are freshly dried samples.

Many monoterpenes have antimicrobial, antioxidant, anti-inflammatory, and anti-carcinogenic effects. In addition, it has been shown in previous studies that monoterpenes such as limonene prevent breast, lung and other cancers and monoterpenes are effective in cancer treatment (Gould, 1997). It has been reported that β-myrcene, a monoterpene, acts like estrogen activity, which is particularly important for women and also exhibits cardiotonic and diuretic properties (Chappell, 1995; Koziol et al., 2014; Kweka, 2009). β-Myrcene also has antibacterial properties on *Staphylococcus aureus, Escherichia coli, Salmonella enterica* as well as some plant pathogenic bacteria

Table 3. Volatile organic component analysis results of the *N. officinale*
Çizelge 3. N. officinale'nin uçucu organik bileşen analiz sonuçları

Compounds *Bileşikler*	RI	%	Molecular formula	Classification *Sınıflandırma*
2-Isobutyl-4,4-dimethyl-1,3-dioxane	549	0.05	$C_9H_{19}O_2$	acetal
Furan, 2,3-dihydro-	730	0.57	C_4H_6O	enol ether
Ethanol	921	0.15	C_2H_5OH	alcohol
(1R)-2,6,6-Trimethylbicyclo[3.1.1]hept-2-ene (1R-α-Pinene)	1062	0.57	$C_{10}H_{16}$	monoterpene
alpha-Thujene	1068	0.03	$C_{10}H_{16}$	monoterpene
Bicyclo[3.1.1]heptane, 6,6-dimethyl-2-methyl- (beta pinene)	1136	0.18	$C_{10}H_{16}$	monoterpene
l-Phellandrene	1152	0.62	$C_{10}H_{16}$	monoterpene
beta-Myrcene	1154	1.64	$C_{10}H_{16}$	monoterpene
D-Limonene	1189	6.43	$C_{10}H_{16}$	monoterpene
Cyclohexane, 1-methylene-4-(1-methylethenyl (p-Mentha-1(7),8-diene)	1193	0.05	$C_{10}H_{16}$	monoterpene
beta-Phellandrene	1198	6.69	$C_{10}H_{16}$	monoterpene
cis-Ocimene	1233	0.29	$C_{10}H_{16}$	monoterpene
gamma-Terpinene	1242	5.28	$C_{10}H_{16}$	monoterpene
Benzene, methyl(1-methylethyl)- (cymene)	1268	1.23	$C_{10}H_{14}$	monoterpene
alpha-Terpinolene	1283	54.46	$C_{10}H_{16}$	monoterpene
Bicyclo[2.2.1]hept-2-ene, 1,7,7-trimethyl- (Bornylene)	1287	0.07	$C_{10}H_{16}$	monoterpenoid
Octanal	1290	0.09	$C_8H_{16}O$	aldehyde
Tetradecane	1400	0.04	$C_{14}H_{30}$	alkan
3,8-dimethylene-1-cyclooctene	1432	0.21	$C_{10}H_{16}$	monoterpene
Benzene, 1-methyl-4-(1-methylethenyl)- (p-Cymenene)	1451	0.66	$C_{10}H_{12}$	monoterpene alkylbenzene
Epoxyterpinolene	1480	0.69	$C_{10}H_{16}O$	monoterpenic ether
1-Hexanol, 2-ethyl-	1491	0.06	$C_8H_{18}O$	alcohol
Copaene	1500	0.08	$C_{15}H_{24}$	sesquiterpene
Benzaldehyde, 2,5-bis[(trimethylsilyl)oxy]-	1521	0.06	$C_{13}H_{22}O_3Si_2$	aldehit
3,5-Octadien-2-one	1527	0.06	$C_8H_{12}O$	oxygenated hydrocarbon
Benzaldehyde	1538	0.04	C_6H_5CHO	aldehyde
Cyclohexane, 2-ethenyl-1,1-dimethyl-3-methylene	1560	0.10	$C_{10}{}^{13}CH_{16}D_2$	cyclic hydrocarbon
Ethanone, 1-(6,6-dimethylbicyclo[3.1.0]hex-2 en	1562	0.08	$C_{10}H_{14}O$	ketone
beta-Cyclocitral	1565	0.10	$C_{10}H_{16}O$	monoterpenoid
alpha-Bergamotene	1597	0.16	$C_{15}H_{24}$	sesquiterpene
Heneicosane	1600	0.02	$C_{21}H_{44}$	alkan
Caryophyllene	1614	3.21	$C_{15}H_{24}$	sesquiterpene
Cyclohexanol, 2,6-dimethyl-	1622	0.04	$C_8H_{16}O$	alcohol
Naphthalene, 1,2,3,4,4a,7-hexahydro-1,6-dimethyl(Cadina-1(2),4-diene, cis)	1625	6.86	$C_{15}H_{24}$	sesquiterpene
16-Methyl-heptadecane-1,2-diol,trimethylsilyl ether	1640	0.12	$C_{24}H_{54}O_2Si_2$	alcohol
Farnesol	1673	0.24	$C_{15}H_{26}O$	sesquiterpenoid
cis-thujan-10-oic acid methyl ester	1686	0.15	$C_{10}H_{16}O_2$	sesquiterpenoid
alpha-Humulene	1690	0.14	$C_{15}H_{24}$	sesquiterpene
1,8-menthadien-4-ol	1696	1.07	$C_{10}H_{16}O$	monoterpenoid
1H-Cyclopenta[1,3]cyclopropa[1,2]benzene, o (beta cubebene)	1714	0.28	$C_{15}H_{24}$	sesquiterpenoid
Germacrene-D	1730	0.11	$C_{15}H_{24}$	sesquiterpene
beta-Bisabolene	1740	0.16	$C_{15}H_{24}$	sesquiterpene
3,5-Nonadien-7-yn-2-ol, (E,E)-	1777	0.06	$C_9H_{12}O$	alcohol

Cyclohexene,3-(1,5-dimethyl-4-hexenyl)-6-methylene(β-Sesquiphellandrene)	1785	1.95	$C_{15}H_{24}$	sesquiterpene
Ethanone, 1-(4-methylphenyl)-	1795	0.05	$C_9H_{10}O$	alkyl-phenylketone
Bicyclo[3.1.1]hept-2-en-6-ol,2,7,7-trimethyl-(Chrysanthenol, cis)	1814	0.16	$C_{10}H_{16}O$	monoterpenoid
Benzenemethanol, .alpha,alpha,4-trimethyl-(p-Cymen-8-ol)	1858	0.92	$C_{10}H_{14}O$	oxygenated monoterpene
5,9-Undecadien-2-one, 6,10-dimethyl-, (Z)-(cis-Geranylacetone)	1866	0.08	$C_{13}H_{22}O$	monoterpene ketone
Naphthalene, 1,2,3,4-tetrahydro-1,6-dimethyl-(cis calamenene)	1876	0.07	$C_{15}H_{22}$	sesquiterpene
Propanoic acid, 2-methyl-, 1-(1,1-dimethylethyl ester	1891	0.23	$C_8H_{16}O_2$	ester
Neophytadiene	1927	0.15	$C_{20}H_{38}$	sesquiterpenoid
5-Isopropenyl-2-methyl-7-oxabicyclo[4.1.0]heptan-2 ol	1948	0.15	$C_{10}H_{16}O_2$	alcohol
3-Buten-2-one, 4-(2,6,6-trimethyl-1-cyclohexe (β-Ionone)	1958	0.06	$C_{13}H_{20}O$	sesquiterpenoid
1-Nonadecanamine, N,N-dimethyl-	1993	0.15	$C_{21}H_{45}N$	amine
Diphenyl ether	2039	0.09	$(C_6H_5)_2O$	ether
5,7-Octadien-3-ol, 2,4,4,7-tetramethyl-	2044	0.07	$C_{12}H_{22}O$	monoterpenoid
Benzenepropanenitrile	2063	0.33	C_9H_9N	nitrile
Squalene	2153	1.55	$C_{30}H_{50}$	triterpene
1H-Purin-6-amine, [(2-fluorophenyl)methyl]-	2159	0.05	$C_{12}H_{10}FN_5$	amine
2-Cyclohexen-1-one,2-methyl-5-(1-methylethyl)(Carvotanacetone)	2203	0.65	$C_{10}H_{16}O$	menthane monoterpenoid
Cinnamaldehyde, alpha - pentyl-	2255	0.09	$C_{14}H_{18}O$	phenylpropanoid

RI: Retention index, %: Percentage of volatile component in total volatile components (w/w)
RI: Alıkonma indeksi, %: Uçucu bileşenin, toplam uçucu bileşen içindeki yüzdesi (w/w)

(Abdel Rasoul et al., 2012; Wang et al., 2019; Połeć et al., 2020). Squalene is a triterpene that is a precursor to the biosynthesis of cholesterol and other steroids. It has been reported to be anticarcinogenic and reduce tumor growth (Reddy and Couvreur, 2009). β-Sesquiphellandrene has been reported to have anti-proliferative effects in leukemia, multiple myeloma, and colorectal cancer cells (Denyer et al., 1994; Tyagi et al., 2015; Siripoltangman and Chickos, 2019). β-Caryophyllene is a sesquiterpene with anti-inflammatory, anti-spasmodic, antimicrobial effects as well as beneficial effects such as curing asthma and inhibiting hypersensitive immune reaction. (Kim et al., 1998; Cho et al., 2007; Galdino et al., 2012; Dahham et al., 2015; Yoo and Jwa, 2019).

N. officinale contains large amounts of alpha-terpinolene. Terpinolene, a flavored ingredient, is also known to have an anti-fungal function (Hammer et al., 2004). On the other hand, it has been suggested that these monoterpenes suppress NF-κB (Nuclear Factor kappa B) activity and that terpinolene and α-phellandrene may contribute to the treatment of wounds by reducing inflammation and oxidative stress (Scherer et al., 2019). It has also been proven that terpinolene is non-genotoxic and exhibits a wide variety of properties such as antioxidant, antiproliferative, anticancer, antifungal, and larvicide (Dorman et al., 2000; Hammer et al., 2004; Conti et al., 2012; Harada et al., 2012). All these findings reinforce that terpinolene is a good and safe natural antioxidant as well as a potential anticancer agent (Aydın et al., 2013). In the light of all this information, many useful components were determined in *N. Officinale* by volatile component analysis.

In addition, the results of the volatile component analysis of the leaf and stem of *N. officinale* are given in Table 4 comparatively.

When the results regarding the volatile components of the leaves and stems of *N. officinale* given in Table 4 are examined, it is seen that the leaf part of the plant is richer in volatile components than the stem part. Alpha-terpinolene is the main volatile component of both the leaf and stem of the plant. On the other hand, compounds such as D-limonene, gamma-terpinene, p-cymenene, cymene, benzenepropanenitrile, squalene, beta-phellandrene were found more in the stem part.

Fatty Acid Methyl Ester Analysis Results

Analysis results of fatty acid methyl ester of *N. officinale* are given in Table 5.

The total unsaturated fatty acid ratio of the plant is 58.75%, and the total saturated fatty acid ratio is 41.25%. The plant contains alpha-linolenic acid, arachidic acid, linolenic acid, elaidic acid and palmitic acid. It has been observed that *N. officinale* can be an important nutrient in daily nutrition because it is rich in alpha-linolenic acid (omega 3), an essential fatty acid with important functions for the human body (Santos et al., 2017; Martins et al., 2018). It also contains linolenic acid, elaidic acid, and palmitic acid. The plant stands as a precious food with this content.

Table 4. Volatile component analysis results of leaves and stems of *N. officinale*
Çizelge 4. N. officinale'nin yaprak ve gövdesinin uçucu bileşen analiz sonuçları

	Compounds *Bileşikler*	Leaf Analysis % *Yaprak analizi %*	Stem Analysis % *Gövde analizi %*
1	Furan, 2,3-dihydro-	0.5	0.43
2	Borane-methyl sulfide complex	0.05	-
3	Acetone	0.07	0.10
4	N-Methylene-tert-butylamine	0.05	-
5	Butanal, 2-methyl-	0.02	-
6	Pentanal	0.1	0.03
7	Octane, 3,5-dimethyl-	0.03	-
8	1R-α-pinene	0.69	0.66
9	Alpha-Thujene	0.03	0.02
10	Beta pinene	2.94	2.88
11	Hexanal	0.06	0.03
12	Alpha terpinene	0.11	0.10
13	D-Limonene	7.2	7.49
14	p-Mentha-1(7),8-diene	0.05	0.05
15	Beta-Phellandrene	9.3	10.8
16	2-Hexenal, (E)-	0.09	-
17	cis-Ocimene	0.37	-
18	Gamma-Terpinene	7.85	10.19
19	Cymene	1.6	1.81
20	Alpha-terpinolene	51.78	49.67
21	Octanal	0.04	0.07
22	1,5-Cyclooctadiene, 3-t-butyl-	0.57	-
23	2-Methylisoborneol	-	0.49
24	1,3,8-p-Menthatriene	-	0.21
25	3,8-dimethylene-1-cyclooctene	0.17	0.20
26	p-Cymenene	0.78	1.06
27	Epoxyterpinolene	0.57	0.77
28	Copaene	0.1	-
29	3,5-Octadien-2-one	0.09	-
30	Benzaldehyde	0.11	-
31	Cyclohexane, 2-ethenyl-1,1-dimethyl-3-methy	0.09	0.10
32	2-Isopropylidene-3-methylhexa-3,5-dienal	-	0.06
33	Ethanone, 1-(6,6-dimethylbicyclo[3.1.0]hex-2	0.05	-
34	Bicyclo[3.1.1]hept-2-ene, 2,6-dimethyl-6-(4-m	0.14	0.11
35	Hexadecane	0.02	-
36	Caryophyllene	3.3	2.57
37	Cyclohexanol, 2,6-dimethyl-	0.14	0.04
38	Cadina-1(2),4-diene, cis	4.48	3.28
39	beta-Cyclocitral	0.09	-
40	Cyclohexane, 1-ethenyl-1-methyl-2-(1-methyl	0.26	-
41	cis-beta-Farnesene	0.16	0.11
42	cis-thujan-10-oıc acıd methyl est	0.11	0.24
43	alpha-Humulene	0.15	0.11
44	1,8-menthadien-4-ol	0.76	1.62
45	Beta cubebene	0.18	0.16
46	Benzoic acid, 4-methyl-, 2-hydroxy-2-phenyl -	-	0.12
47	Cyclohexene, 3-(1,5-dimethyl-4-hexenyl)-6-m -	-	0.69
48	Ethanone, 1-(3-methylphenyl)--	-	0.06
49	Benzenemethanol, .alpha.,.alpha.,4-trimethyl--	-	1.26

50	1s,cis-calamenene	0.05	0.05
51	Neophytadiene	0.42	0.05
52	5-Isopropenyl-2-methyl-7-oxabicyclo[4.1.0]he	0.27	0.28
53	Benzenepropanenitrile -	-	0.10
54	Cyclooctanone	0.06	0.07
55	Squalene	-	1.56
56	3,5-Octadien-2-ol	-	0.05
57	Germacrene-d	0.13	-
58	beta-Bisabolene	0.15	-
59	alpha-Farnesene	0.05	-
60	Benzenemethanol, .alpha.,4-dimethyl-	0.08	-
61	beta-Sesquiphellandrene	2.09	-
62	Ethanone, 1-(4-methylphenyl)-	0.06	-
63	Selina-3,7(11)-diene	0.04	-
64	Chrysanthenol, cis	0.16	0.25
65	Benzenemethanol, alpha, alpha, 4-trimethyl-	0.96	-
66	3-Buten-2-one, 4-(2,6,6-trimethyl-1-cyclohexe	0.09	-
67	Diphenyl ether	0.08	-
68	5,7-Octadien-3-ol, 2,4,4,7-tetramethyl-, (E)-	0.06	-

%: Percentage of volatile component in total volatile components (w/w)
%: Uçucu bileşenin, toplam uçucu bileşen içindeki yüzdesi (w/w)

Table 5. Analysis results of fatty acid methyl ester of *N. officinale*
Çizelge 5. N. officinale'nin yağ asidi metil ester analiz sonuçları

Peak	R.T	Fatty acid	Concentration	Units	Area
1	18.482	Palmitic Acid C16:0	14.422	%	435
2	22.814	Elaidic Acid C18:1n9t	11.890	%	358
3	24.818	Linolenic Acid C18:2n6t	16.368	%	493
4	25.685	Arachidic Acid C20:0	26.827	%	808
5	26.336	Alpha-Linolenic Acid C18: 3n-3	30.493	%	919
				Total	3013

R.T.: Retention time, Total: Total peak area, %: Percentage of the component in total peak area (w/w)
R.T.: Alıkonma zamanı, Total : Toplam pik alanı, %: Bileşenin, toplam pik alanındaki yüzdesi (w/w)

CONCLUSION

In the light of all these results, it has been determined that the *N. officinale* is an important plant that contains essential fatty acids and many volatile organic compounds and also has antimicrobial effects. Methanol extract of *N. officinale* showed more antimicrobial effect than water extract. In the volatile component analysis results, in which we determined the volatile components of different parts of the plant, it was determined that *N. officinale* contains important components such as alpha-terpinolene, β-Myrcene, β-Caryophyllene, Squalene, β-Sesquiphellandrene. In addition, according to the results of the fatty acid analysis, it was determined that it contains important fatty acids such as omega-3 in its structure. As a result, it has been shown that the plant is a species that can be used in new applications with both its antimicrobial activity and the important components it contains.

ACKNOWLEDGEMENT

This work was supported by the Dicle University Scientific Research Projects Coordination Unit [With the project no ZGEF.20.001]. We would like to thank Dicle University Scientific Research Projects Coordination Unit for their project support and thank Dicle University Science and Technology Application and Research Center (DUBTAM) for the analyzes. In addition, I am grateful to Prof. Dr. Hasan AKAN for the authentication information of the plant.

Author's Contributions

Author 1: Writing-original draft preparation, data collection, data curation, visualization, analysis, data interpretation. Author 2: Conceptualization, methodology, validation, writing-review, and editing, supervision, provision of analysis tools. All authors have accepted responsibility for the entire content of this manuscript and approved its submission.

Statement of Conflict of Interest

The authors state no conflict of interest.

REFERENCES

AOAC Official Method 2012. 13 Determination of Labeled Fatty Acids.

Abd Rashid N, Abd Halim SAS, Teoh SL, Budin SB, Hussan F, Ridzuan NRA, Jalil NAA 2021. The Role of Natural Antioxidants in Cisplatin-Induced Hepatotoxicity. Biomedicine & Pharmacotherapy 144:112328.

Abdel Rasoul MA, Marei GIK, Abdelgaleil SAM 2012. Evaluation of Antibacterial Properties and Biochemical Effects of Monoterpenes on Plant Pathogenic Bacteria. Afr. J. Microbiol. Res. 6:3667-3672. DOI: 10.5897/AJMR12.118

Adokoh, CK, Asente DB, Acheampong DO, Kotsuchibashi Y, Armah FA, Sirikyi IH, Kimura K, Gmakame E, Rauf SA- 2019. Chemical Profile and In Vivo Toxicity Evaluation of Unripe *Citrus aurantifoli*a Essential Oil. Toxicol. Rep. 6:692-702. https://doi.org/10.1016/j.toxrep.2019.06.020

Amiri, H. 2012. Volatile constituents and antioxidant activity of flowers, stems and leaves of *Nasturtium officinale* R. Br.. Nat. Prod. Res., 26(2):109-115.

Aydın E, Türkez H, Taşdemir Ş 2013. Anticancer and Antioxidant Properties of Terpinolene in Rat Brain Cells. Arh Hig Rada Toksikol 64:415-424 DOI: 10.2478/10004-1254-64-2013-2365

Chappell, J 1995. Biochemistry and molecular biology of the isoprenoid biosynthetic pathway in plants. Annu. Rev. Plant Physiol. Plant. Mol. Biol. 46: 521-547.

Cho JY, Chang HJ, Lee SK, Kim HJ, Hwang JK, Chun HS 2007. Amelioration of Dextran Sulfate Sodium-İnduced Colitis in Mice by Oral Administration of Beta-Caryophyllene, a Sesquiterpene. Life Sciences 80:932-939. DOI: 10.1016/j.lfs.2006.11.038

Conti B, Benelli G, Flamini G, Cioni PL, Profeti R, Ceccarini L, Macchia M, Canale A 2012. Larvicidal and Repellent Activity of *Hyptis suaveolens* (Lamiaceae) Essential Oil Against The Mosquito *Aedes albopictus* Skuse (Diptera: Culicidae). Parasitol Res. 110:2013-21. https://doi.org/10.1007/s00436-011-2730-8

Dahham SS, Tabana YM, Iqbal MA, Ahamed MBK, Ezzat MO, Majid ASA, Majid AMSA 2015. The Anticancer, Antioxidant and Antimicrobial Properties of the Sesquiterpene Beta-Caryophyllene from The Essential Oil of *Aquilaria crassna*. Molecules 20: 11808–11829. doi:10.3390/molecules200711808

Daniel JB 2009. Pacific Northwest Aquatic Invasive Species Profile: *Nasturtium officinale*(Watercress). FSH 423.

Das UN 2006. Essential fatty acids: biochemistry, physiology and pathology. Biotechnology Journal 1(4): 420-439. https://doi.org/10.1002/biot.20060012

Denyer CV, Jackson P, Loakes DM, Ellis MR, Young DA 1994. Isolation of Antirhinoviral Sesquiterpenes from Ginger (*Zingiber officinale*). J. Nat. Prod. 57: 658-662. DOI: 10.1021/np50107a017

Derhami SF, Rad MG, Mahmoudi R (2017). Evaluation of Antibacterial Effects of Aqueous and Alcoholic Extracts of *Nasturtium Officinale* on Some Pathogenic. Medical Laboratory Journal, 10(6): 49-53.

Dorman HJD, Figueiredo AC, Barroso JG, Deans SG 2000. In Vitro Evaluation of Antioxidant Activity of Essential Oils and Their Components. Flavour Fragr J 15: 12-16. DOI: 10.1002/(SICI)1099-1026(200001/02)15:1<12::AID-FFJ858>3.0.CO;2-V

Galdino PM, Nascimento MVM, Florentino IZ, Lino RC, Fajemiroye JO, Chaibub BA, Realino de Paula J, Monteiro de Lima TC, Costa EA 2012. The Anxiolytic-like Effect of an Essential Oil Derived from *Spiranthera odoratissima* A. St. Hil. Leaves and Its Major Component, Betacaryophyllene, in Male Mice. Progress in Neuro-psychopharmacology & Biological Psychiatry 38: 276-284. DOI: 10.1016/j.pnpbp.2012.04.012

Gonçalves EM, Cruz RMS, Abreu M, Brandão TRS, Silva CLM 2009. Biochemical and Colour Changes of Watercress (*Nasturtium officinale* R. Br.) During Freezing and Frozen Storage. J Food Eng. 93(1): 32-39.

Gould MN 1997. Cancer Chemoprevention and Therapy by Monoterpenes. Environmental Health Perspectives. 105:977-979. DOI: 10.1289/ehp.97105s4977

Hammer KA, Carson CF, Riley TV 2004. Antifungal Effects of *Melaleuca alternifolia* (tea tree) Oil and Its Components on *Candida albicans, Candida glabrata,* and *Saccharomyces cerevisiae.* J Antimicrob Chemother 53:1081-5. https://doi.org/10.1093/jac/dkh243

Harada T, Harada E, Sakamoto R, Ashitani T, Fujita K, Kuroda K 2012. Regio- and Substrate-Specific Oxidative Metabolism of Terpinolene by Cytochrome P450 Monooxygenases in *Cupressus lusitanica* Cultured Cells. Am J Plant Sci 3: 268-75. DOI:10.4236/ajps.2012.32032

Kim HM, Lee EH, Hong SH, Song HJ, Shin MK, Kim SH, Shin TY 1998. Effect of *Syzygium aromaticum* Extract on Immediate Hypersensitivity in Rats. Journal of Ethnopharmacology 60(2): 125-131. https://doi.org/10.1016/S0378-8741(97)00143-8

Koziol A, Stryjewska A, Librowski T, Salat K, Gawel M, Moniczewski A, Lochyński S 2014. An Overview of the Pharmacological Properties and Potential Applications of Natural Monoterpenes. Mini-Reviews in Medicinal Chemistry 14: 1156-1168.

Kweka EJ, Nkya HM, Lyaruu L, Kimaro EE, Mwang'onde BJ, Mahande AM 2009. Efficacy of *Ocimum kilimandscharicum* plant extracts after four years of storage against Anopheles gambiaess. J.Cell Anim. Biol. 3: 171-174. https://doi.org/10.5897/JCAB.9000067

Maffei ME, Gertsch J, Appendino G 2011. Plant Volatiles: Production, Function, and Pharmacology. Nat Prod Rep. 28(8):1359–80. DOI: 10.1039/c1np00021g

Martins RM, Nedel F, Guimarães VBS, Silva AF, Colepicolo P, Pereira CMP, Lund RG 2018. Macroalgae Extracts from Antarctica Have Antimicrobial and Anticancer Potential. Front Microbiol 9:1–10. https://doi.org/10.3389/ fmicb.2018.00412

Panighel A and Flamini R 2014. Applications of Solid-Phase Microextraction and Gas Chromatography/ Mass Spectrometry (SPME-GC/MS) in the Study of Grape and Wine Volatile Compounds. Molecules 19(12): 21291-21309. https://doi.org/ 10.3390/ molecules191221291

Pichersky E, Sharkey TD, Gershenzon J 2006. Plant Volatiles: A Lack of Function or a Lack of Knowledge? Trends Plant Sci. 11(9): 421. DOI: 10.1016/j.tplants.2006.07.007

Połeć K, Broniatowski B, Wydro P, Hąc-Wydro K 2020. The Impact of β-Myrcene–The Main Component of the Hop Essential Oil on the Lipid Films. Journal of Molecular Liquids 308: 113028. https://doi.org/ 10.1016/j.molliq.2020.113028

Pourhassan-Moghaddam M, Zarghami N, Mohsenifar A, Rahmati-Yamchi M, Gholizadeh D, Akbarzadeh A, de la Guardia M, Nejati-Koshki K 2014. Watercress-Based Gold Nanoparticles: Biosynthesis, Mechanism of Formation and Study of Their Biocompatibility in Vitro. Micro Nano Lett. 9(5): 345-50.

Pretorius B, Schőnfeldt HC 2021. Cholesterol, Fatty Acids Profile and the Indices of Atherogenicity and Thrombogenicity of Raw Lamb and Mutton Offal. Food Chemistry 345:128868. https://doi.org/ 10.1016/j.foodchem.2020.128868

Reddy LH, Couvreur P 2009. Squalene: A Natural Triterpene for Use in Disease Management and Therapy. Advanced DrugDelivery Reviews 61(15):1412-1426.

Santos MZ, Colepicolo P, Pupo D, Fujii MT, Pereira CP, Mesko MF 2017. Antarctic Red Macroalgae: a Source of Polyunsaturated Fatty Acids. J Appl Phycol 29: 759–767. https://doi.org/10.1007/s10811-016-1034-x

Scherer MMC, Marques FM., Figueira MM, Peisino MCO, Schmitt EP, Kondratyuk TP, Endringer DC, Scherer R, Fronza M 2019. Wound Healing Activity of Terpinolene and α-Phellandrene by Attenuating Inflammation and Oxidative Stress in Vitro. Journal of Tissue Viability. 28(2): 94-99. https://doi.org/10.1016/j.jtv.2019.02.003

Shahani S, Behzadfar F, Jahani D, Ghasemi M, Shaki F 2017. Antioxidant and Antiinflammatory Effects of *Nasturtium Officinale* Involved in Attenuation of Gentamicin İnduced Nephrotoxicity. Toxicol. Mech. Methods 27(2): 107-114.

Siripoltangman N, Chickos J 2019. Vapor Pressure and Vaporization Enthalpy Studies of the Major Components of Ginger, α-Zingiberene, β-Sesquiphellandrene, and (-) Arcurcumene by Correlation Gas Chromatography. J. Chem. Thermodynamics 138: 107-115.

Şengün IY and Öztürk B 2018. Some Natural Antimicrobials of Plant Origin. Anadolu University Journal of Science and Technology C-Life Sciences and Biotechnology 7(2): 256-276.

Tyagi AK, Prasad S, Yuan W, Li S, Aggarwal BB 2015. Identification of a Novel Compound (β-Sesquiphellandrene) from Turmeric (*Curcuma longa*) with Anticancer Potential: Comparison with Curcumin. Invest New Drugs 33(6): 1175-1186.

Varlı M, Hancı H, Kalafat G 2020. Tıbbi ve Production Potential and Bioavailability of Medicinal and Aromatic Plants. Research Journal of Biomedical and Biotechnology 1: 24-32,

Vieira, AJ, Beserra FP, Souza MC, Totti BM, Rozza AL 2018. Limonene: Aroma of Innovation in Health and Disease. *Chemico-Biological Interactions* 283: 97-106.

Wassell P, Bonwick GA, Smith CJ, Roig EA, Young NG 2010. Towards a Multidisciplinary Approach to Structuring in Reduced Saturated Fat-Based Systems – A review. Int. J. Food Sci. Technol. 45: 642–655 https://doi.org/10.1111/j.1365-2621.2010.02212.x

Wang CY, Chen YW, Hou CY 2019. Antioxidant and Antibacterial Activity of Seven Predominant Terpenoids. Int. J. Food Prop. 22(1): 230-238.

Wayne PA 1997. NCCLS (National Committee for Clinical Laboratory Standards) Performance Standards for Antimicrobial Disk Susceptibility Tests: Approved Standard Enclose -A 7. USA

Yip EC, Tooker JF, Mescher MC, De Moraes CM 2019. Costs of Plant Defense Priming: Exposure to Volatile Cues from a Specialist Herbivore İncreases Short-Term Growth But Reduces Rhizome Production in Tall Goldenrod (*Solidago altissima*). BMC Plant Biol. 19(1): 209. https://doi.org/ 10.1186/s12870-019-1820-0

Yoo HJ, Jwa SK 2019. Efficacy of β-Caryophyllene for Periodontal Disease Related Factors. Archives of Oral Biology 100: 113-118.