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ARAŞTIRMA MAKALESI / RESEARCH ARTICLE

CHEMICAL PROFILE AND ANTIMICROBIAL ACTIVITY OF IMPORTANT HONEY PLANTS *THYMUS NUMMULARIUS* M. BIEB. AND *VACCINIUM MYRTILLUS* L.

Önemli Bal Bitkileri *Thymus nummularius* M. Bieb. ve *Vaccinium myrtillus* L.'nin Kimyasal Profili ve Antimikrobiyal Aktivitesi

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ABSTRACT

The aim of this study was to determine the chemical and antimicrobial activity of two taxa which are important nectar and pollen plants for honey bees. *Thymus nummularius* M. Bieb. and *Vaccinium myrtillus* L. which produced by local people of Anzer. For this purpose, chemical compounds scanned by GC–MS system and antimicrobial activities were tested against major oral pathogens including *Porphyromonas gingivalis* ATCC 33277, *Streptococcus salivarius* DSM 13084, *Streptococcus mitis*, *Streptococcus mutans* and *Candida albicans* ATCC 90028 by agar well diffusion and broth microdilution. Aldehydes, alcohols, carboxylic acids and esters, ketones, terpenes, fatty acids and esters, acetic acids and esters and other chemical components were detected in different percentages in both *T. nummularius*. and *V. myrtillus* plants. A total of 30 chemical components were detected in *T. nummularius*. while 26 chemical components were found in *V. myrtillus*. Carvacrol, lauryl acetate, and thymol were detected 20.34%, 6.31% and 3.36%, respectively at high levels in *T. nummularius*. On the other hand, diethyl succinate, hexanoic acid, lauryl acetate and benzoic acid were determined 12.68%, 11.29%, 10.89% and 10.16% respectively predominantly in *V. myrtillus*. As a result of antimicrobial activity of two taxa, *V. myrtillus*. exhibited more potent antimicrobial activity when compared to *T. nummularius*. Antimicrobial activity of *V. myrtillus* against viridans streptococci and *C. albicans* was promising and maybe used for the treatment of cariogenic microorganisms or oral Candidiasis in future studies.

Key words: Antimicrobial activity, Chemical compounds, Honey plants, *Thymus nummularius*, *Vaccinium myrtillus*

ÖZ

Bu çalışmanın amacı, Anzer yöresi halkı tarafından üretilen ve bal arıları için önemli nektar ile polen kaynağı bitkiler olan *Thymus nummularius* M. Bieb. ve *Vaccinium myrtillus* L. adlı iki taksonun kimyasal ve antimikrobiyal aktivitesini belirlemektir. Bu amaçla, kimyasal bileşikler GC-MS sistemi ile

taranmış ve antimikrobiyal aktiviteleri *Porphyromonas gingivalis* ATCC 33277, *Streptococcus salivarius* DSM 13084, *Streptococcus mitis*, *Streptococcus mutans* ve *Candida albicans* ATCC 90028 dahil olmak üzere başlıca oral patojenlere karşı agar kuyu difüzyon ve sıvı mikrodilüsyon ile test edilmiştir. Aldehitler, alkoller, karboksilik asitler ve esterler, ketonlar, terpenler, yağ asitleri ve esterler, asetik asitler ve esterler ve diğer kimyasal bileşenler hem *T. nummularius* hem de *V. myrtillus* bitkilerinde farklı yüzdelerde tespit edilmiştir. *T. nummularius*'ta toplam 30 kimyasal bileşen tespit edilirken, *V. myrtillus*'ta 26 kimyasal bileşen tespit edilmiştir. Karvakrol, lauril asetat ve timol *T. nummularius*'ta sırasıyla %20,34, %6,31 ve %3,36 oranında yüksek seviyelerde tespit edilmiştir. Öte yandan, dietil süksinat, hekzanoik asit, lauril asetat ve benzoik asit *V. myrtillus*'ta baskın olarak sırasıyla %12,68, %11,29, %10,89 ve %10,16 oranlarında tespit edilmiştir. İki taksonun antimikrobiyal aktivitesinin bir sonucu olarak, *V. myrtillus* bitkisi *T. nummularius* ile karşılaştırıldığında daha güçlü antimikrobiyal aktivite göstermiştir. *V. myrtillus*'un özellikle viridans streptokoklara ve *C. albicans*'a karşı antimikrobiyal aktivitesinin olması ileriki çalışmalarda karyojenik mikroorganizmaların veya oral Kandidiyazis'in tedavisinde kullanılabilme potansiyeli olduğunu göstermektedir.

Anahtar Kelimeler: Antimikrobiyal aktivite, Kimyasal bileşenler, Ballı bitkiler, *Thymus nummularius*, *Vaccinium myrtillus*

GENİŞLETİLMİŞ ÖZET

Amaç: Bu çalışmanın amacı, Anzer yöresi halkı tarafından üretilen ve bal arıları için önemli nektar ile polen kaynağı bitkiler olan *Thymus nummularius* M. Bieb. ve *Vaccinium myrtillus* L. adlı iki taksonun kimyasal ve antimikrobiyal aktivitesini belirlemektir. Bu amaçla Anzer Bölgesi'nden toplanan *Thymus nummularius* M. Bieb. (Kekik, Anzer çayı, Anuk) ve *Vaccinium myrtillus* L. (Yaban mersini, Maviyemiş, Likapa) bitkilerinin kimyasal analizleri ve oral patojenlere karşı antimikrobiyal aktiviteleri belirlenmiştir.

Gereç ve yöntem: *Thymus nummularius* M. Bieb. ve *Vaccinium myrtillus* L. bitkilerinin ekstraktları methanol ile yapılarak Gaz Kromatografisi ve Kütle Spektrometresi (GC-MS) cihazında kimyasal bileşenleri tespit edilmiştir. Ayrıca antimikrobiyal aktiviteleri *Porphyromonas gingivalis* ATCC 33277, *Streptococcus salivarius* DSM 13084, *Streptococcus mitis* (klinik izolat), *Streptococcus mutans* (klinik izolat) ve *Candida albicans* ATCC 90028 dahil olmak üzere başlıca oral patojenlere karşı agar kuyu difüzyon ve sıvı mikrodilüsyon yöntemi ile test edilmiştir.

Bulgular ve tartışma: GC-MS cihazında yapılan çalışma sonucunda hem *T. nummularius* bitkisinde hem de *V. myrtillus* bitkilerinde farklı yüzdelerde aldehitler, alkoller, karboksilik asitler ve esterler, ketonlar, terpenler, yağ asitleri ve esterler, asetik asitler ve esterler ve diğer kimyasal bileşenler tespit edilmiştir (Tablo 1). *T. nummularius*'ta toplam 30 kimyasal bileşen tespit edilirken, *V. myrtillus*'ta 26 kimyasal bileşen tespit edilmiştir. Karvakrol, lauril asetat ve timol *T. nummularius*'ta sırasıyla %20,34, %6,31 ve %3,36 oranında yüksek seviyelerde tespit edilmiştir (Tablo 1). Ertas vd. (2015) *T. nummularius*'te timol (%60,38) ve terpinil-asetat (%10,49), Gerçek vd. (2022) ise timol (%38,91), linalool (%13,12) ve geraniol (%6,51) bulmuşlardır. Bizim çalışmamızda ise bu çalışmalardan farklı olarak yüksek oranda karvakrol tespit ettik. Karvakrol ve timolün kekik için önemli belirteç bileşenler olduğu belirtilmiştir. Küçükbay vd. (2014) tarafından karvakrol ve timolün yüksek antioksidan aktiviteye sahip olduğu bildirilmiştir. Ayrıca Tepe vd. (2011) karvakrolün antifungal, timolün ise antiseptik etkiye sahip olduğunu söylemişlerdir. Ayrıca, Shah vd. (2020) lauril asetatın antioksidan özellikte olduğunu bildirmişlerdir. Diğer yandan, dietil süksinat, hekzanoik asit, lauril asetat ve benzoik asit *V. myrtillus*'ta baskın olarak sırasıyla %12,68, %11,29, %10,89 ve %10,16 oranlarında tespit edilmiştir (Tablo 1). Aranega-Bou vd. (2014) hekzanoik asidin antifungal aktivitesini bildirmiştir. Ayrıca, Özkök vd. (2016) tarafından benzoik asidin balgam söktürücü, analjezik ve antiseptik aktivitelere sahip olduğu bildirilmiştir. Bununla birlikte, Elkıran ve Avşar (2020) Türkiye'nin Sinop ilinden toplanan *V. myrtillus*'ta 1,8-sineol (%38,6), α-pinen (%21), linalool (%19,5), α-terpineol (%5,8) bulmuştur. Çalışmamızda farklı bileşenler bulunmuş ve bu durum kimyasal içeriğin bölgelere göre değişkenlik olabileceğini göstermiştir.

Ayrca, *T. nummularius* ve *V. myrtillus*'un başlıca oral patojenlere karşı antimikrobiyal aktivitesini agar

kuyusu difüzyonu (Tablo 2, Şekil 2) ve et suyu mikroseyreltme testi (Tablo 3) ile araştırılmıştır. Sonuçlarımıza göre, *V. myrtillus*, test edilen tüm mikroorganizmalara karşı *T. nummularius*'tan daha etkili çıkmıştır. Bununla birlikte, *V. myrtillus, C. albicans*'a karşı güçlü bir antimikrobiyal aktivite sergilemiştir. *V. myrtillus*'un *C. albicans*'a karşı ümit verici antimikrobiyal aktivitesi, nispeten daha yüksek karboksilik asitlere ve esterlere (%23,59), özellikle benzoik aside atfedilebilir. Benzoik asidin öncelikle mantarlara karşı antimikrobiyal aktiviteye sahip olduğu gösterilmiştir (Teodoro vd. 2015).

Sonuç: Bu çalışmada, kimyasal bileşik analizleri her iki bitki türünün de yüksek antioksidan bileşenlere sahip olduğunu göstermiştir. *V. myrtillus*, *T. nummularius* ile karşılaştırıldığında daha güçlü antimikrobiyal aktivite göstermiştir. *V. myrtillus*'un viridans streptokoklarına ve *C. albicans*'a karşı antimikrobiyal aktivitesi ümit vericidir ve gelecekteki çalışmalarda karyojenik mikroorganizmaların veya oral Kandidiyazis'in tedavisinde kullanılabilir.

INTRODUCTION

Local people and traditional healers have long history in using of plants to prevent or cure bacterial infections. Nowadays, in many parts of the world, 70 -95% of people use plants as a primary form of medicine, and many countries have integrated traditional plant-based medicines (Willis 2017). Plants are the main source of a wide variety of secondary metabolites, such as alkaloids, flavonoids, tannins, and terpenoids, which have been determined in vitro to have antimicrobial properties (Cowan 1999).

Thymus L. (Lamiaceae) are well-known genera of the family that is used as folk medicine (Ozen and Demirtas 2015). This species belongs to the group of plants commonly called “kekik” in Turkish (Baytop 1999). For centuries, these taxa have traditionally been used to flavour foods and treatment of various diseases due to the high percentage of their essential oils (Baytop 1999, Lukas et al. 2010, Sezik et al. 1992). *Thymus* species have been used traditionally for bronchitis, coughs, asthma, rheumatism, colic, diarrhoea, and arteriosclerosis (Tammar et al. 2018).

Thymus nummularius M. Bieb. (synonym *Thymus pseudopulegioides* Klokov & Des. Shots.) is distributed widely in the Caucasian area (Güner 2012). It is known as “Anzer tea” (Rize) and “Anuk” (Trabzon) by local people (Günaydın et al. 2017) and also honey bees collect nectar and pollen from this plant. Especially this plant’s antimicrobial bioactive compounds “thymol” and “carvacrol” was found in the honey. Thymol is an important acaricide and is used in the fight against the bee parasite *Varroa destructor*. Therefore, this type of thyme containing thymol is also an important plant in terms of use in bee health (Demirezen 2019, Turkun 2016).

T. nummularius is a perennial shrub, growing wild in the Black Sea Region of Turkiye. Its herbal parts are consumed as tea, condiments, and herbal remedies for gastrointestinal disorders. It is also known that extracts of these plant materials involve bioactive compounds that can be helpful to health such as diuretics, circulation regulators, and sedatives. There have been restricted studies assessing the chemical constituents and biological activity of *T. nummularius* (Baser et al. 1999).

In previous studies (Baser et al. 1999, Gül et al. 2022, Sunar et al. 2009), various *Thymus* species (including *T. nummularius*) were evaluated for their chemical profiles and biological activity. Results revealed significant differences at intra- and interspecific levels. However, a survey of the literature confirmed that *T. nummularius* antibacterial activity was evaluated by Gül et al. 2022.

Vaccinium myrtillus L. (Yaban mersini) from Ericaceae family known as Yaban mersini, Maviyemiş and Likapa in black see regions of Turkiye (Baytop 1999, Güner 2012) and also honey bees collect nectar and pollen from this plant. Four species contain *Vaccinium arctostaphylos* L., *Vaccinium myrtillus* L., *Vaccinium vitisidea* L., *Vaccinium uliginosum* L. grown naturally in Turkiye. *V. myrtillus* is a perennial shrub which grows in coniferous forests. This taxon blooms from April through June have edible spheroidal fruit of blue/black colour with many seeds (Davis 1982).

Using the fruit of this plant in different way such as the decoction of dried fruits has a long history in the human diet. Bilberry commercially is observed as fresh, frozen, and dried berries, in addition to inside the shape of preserves, jams, juices, and liquid or powdered concentrates as meal supplements (Chu et al. 2011).

Vaccinium contains a variety of phenolic compounds, including flavonols, tannins, ellagitannins, and phenolic acids but mostly known

as a rich source of anthocyanins (Seeram 2008, Upton 2001). Anthocyanins in the last years have gained research attention due to their numerous functions and applications in mind diet and neuroprotective effects (Chu et al. 2011). These polyphenolics are responsible their blue/black colour and high antioxidant content, and they are believed to be the key bioactive responsible for the many reported health benefits of bilberry (Chu et al. 2011, Upton 2001,). Although most of the researche has been concentrated on the antioxidant properties of anthocyanins other biological activities of them involve cell-signalling pathways, gene expression, DNA repair, and anticancer effects, as well as antimicrobial and antineoplastic effects (Karakaş et al. 2022, Kowalczyk et al. 2003, Seeram 2008, Zafra-Stone et al. 2007).

Some the berry fruit and their phenolics, have been pronounced to reveal antimicrobial results in opposition to human pathogens. The results of numerous berries in opposition to numerous organisms are different. This suggested different taxa extracts may have high potential as antimicrobials (Puupponen-Pimià et al. 2005).

Anzer Region is an important region where Turkiye's aromatic plants are found, and Anzer honey produced here is a world-famous honey. For this reason, the properties of the plants found here are also important for human health. This research attempted to elucidate the chemical and antimicrobial activity of two taxa *T. nummularius* M. Bieb. and *V. myrtillus* L. which are used by local people of Anzer. For this purpose, chemical compounds scanned by GC–MS system and antimicrobial activities were determined.

MATERIAL AND METHODS

Collection of plant material

The research area Anzer Region is placed in the borders of İkizdere district of the Rize province. Bayburt and Trabzon provinces in the west, Erzurum province inside the south, Kalkandere and Rize vital districts inside the north, Çayeli and Çamlıhemşin districts inside the east (Erata et al. 2021) (Figure 1).

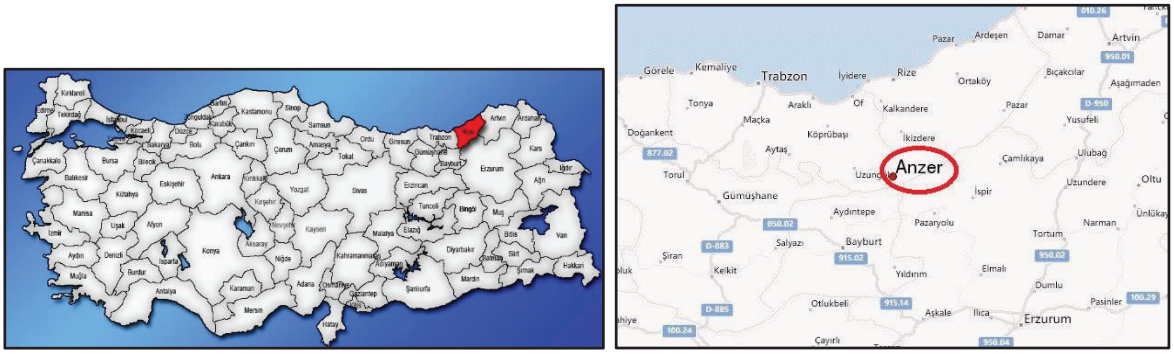

Figure 1. Rize province and Anzer region

The fresh plant material of the *T. nummularius* M. Bieb. and *V. myrtillus* L. were collected from the Anzer Region which is the north-east region of Turkiye. The plants were identified based on the Flora of Turkiye and East Aegean Island (Davis 1982).

Extraction procedure of plant material

Aerial parts of the plants were air-dried in shade condition and powdered. Aerial parts of *T. nummularius* were extracted by methanol. Methanol extracts were prepared by the maceration method and 10 g of cured drug were extracted in 100 mL of solvent (10% m/v) for 24 h with occasional shaking. At the end of the extraction, the solution was filtered, the process was repeated two times, and the filtrates were combined and collected. Finally, it was evaporated to dryness under low pressure.

The liquid segment becomes separated from the stable residue with the aid of using filtering via Whatman No four filter paper and the organic solvent is eliminated with a rotary evaporator (Büchi

Rotavapor, Büchi Labortechnik AG, Flawil, Switzerland). The extracts were lyophilized.

Chemical profile analysis

The chemical profiles of *T. nummularius* and *V. myrtillus* were determined by Gas Chromatography-Mass Spectrometry (GC–MS) analysis. For this analysis, Agilent 6890N GC system coupled with a mass selective detector MS5973 was used. Lyophilized plant extracts were dissolved in methanol and 1 µl of plant extract was injected into the GC-MS system to screen the chemical compounds. For the GC-MS experimental conditions Temiz et al. (2011) method followed. According to this, a DB 5MS capillary column (30 m x 0.25 mm x 0.25 µm) changed into used and the flow rate of mobile phase (He) changed into a set at 0.7 mL/min. In the gas chromatography part, the temperature changed into stored at 50 °C for 1 min. After this period, the temperature changed extended to 150 °C with a 10 °C/min heating ramp after which stored at 150 °C for two min. Finally, the temperature changed extended to 280 °C with a 20°C/min heating ramp after which stored at 280 °C for 30 min. Chemical compounds have been diagnosed via way of means of pc seek the use of reference The Wiley Registry/NIST Mass Spectral Library, which's to be had inside the records acquisition machine of GC-MS.

Antimicrobial activity analysis

Microorganisms and culture conditions: Antimicrobial activity of two taxa *T. nummularius*, and *V. myrtillus* were tested against main oral pathogens including *Porphyromonas gingivalis* ATCC 33277, *Streptococcus salivarius* DSM 13084, *Streptococcus mitis* (clinical isolate) and *Streptococcus mutans* (clinical isolate) and *Candida albicans* ATCC 90028 by agar well diffusion and broth microdilution. *P. gingivalis* was cultured on Brucella Agar (5% sheep blood agar supplemented with vitamin K1 and hemin) under anaerobic conditions at 37 °C for 5 days. Black-pigmented colonies were confirmed after the incubation period. Viridans streptococci were cultured on blood agar (5% CO2, 95% humidified air, at 37 °C) and *C. albicans* was cultured on Sabouraud Dextrose agar (SDA) at 37 °C overnight.

Agar well diffusion assay: Plant extracts were dissolved in dimethyl sulphoxide (DMSO) prior to antimicrobial activity screening. Agar well diffusion assay was performed by harvesting cells from fresh cultures and a standard suspension became organized with the aid of adjusting the turbidity of the suspension to suit the 0.5 McFarland ($1.5X10^8$ cfu/mL). Five-millimeter diameter wells were prepared in Mueller-Hinton Agar (MHA) with 2% glucose for *C. albicans*, MHA with 5% sheep blood for viridans streptococci and Brucella agar for *P. gingivalis*, and the bacterial suspensions were inoculated into corresponding media. After the incubation period, the inhibition zones were measured and means and standard deviations were calculated.

Broth microdilution assay: The minimum inhibitory concentrations (MIC) of the extracts were determined using broth microdilution assay in sterile 96-well microplates. Briefly, two-fold serial dilutions of the extracts were prepared ranging from 1024 to 0.5 µg/mL using Brain Heart Infusion (BHI) medium for viridans streptococci, BHI medium supplemented with vitamin K1 (0.1 µg/mL) and hemin (5 µg/mL) for *P. gingivalis* and Roswell Park Memorial Institute(RPMI) 1640 Medium for *C. albicans*. The inoculum containing 10^6 CFU/mL of each bacterium was added to each well. After the incubation period bacterial growth was evaluated by observing the presence of turbidity. MIC was defined as the lowest concentration of the samples with no bacterial growth.

RESULTS

Chemical compounds of *T. nummularius*, and *V. myrtillus* were given in Table 1.

As seen in Table 1, aldehydes, alcohols, carboxylic acids and esters, ketones, terpenes, fatty acids and esters, acetic acids and esters and other chemical components were detected in different percentages in both *T. nummularius*, and *V. myrtillus* plants. A total of 30 chemical components were detected in *T. nummularius*. while 26 chemical components were found in *V. myrtillus*. While the terpenes group was found most in *T. nummularius*. with 27.13%, carboxylic acids and esters were found most in *V. myrtillus* with 23.59%.

Also, present study investigated the antimicrobial activity of *T. nummularius*. and *V. myrtillus* against major oral pathogens by agar well diffusion (Table 2, Figure 2, Figure 3) and broth microdilution assay (Table 3).

Table 1. Chemical compounds of *T. nummularius*. and *V. myrtillus*

Chemical compounds	*Thymus nummularius* M.Bieb.	*Vaccinium myrtillus* L.
Aldehydes (%)		
trans, trans-2, 4-Hexadienal	1.40	-
Benzaldehyde	1.22	-
Phenylacetaldehyde	-	0.19
Phenylacetaldehyde dimethyl acetal	0.30	-
Cis-6-Nonenal	2.45	-
Total	***5.37***	***0.19***
Alcohols (%)		
3-Octanol	0.83	0.42
2-Pentanol	0.92	-
Isobutyl alcohol	1.16	-
Alpha-Terpineol	0.53	-
Isopropyl alcohol	-	0.37
Total	***3.44***	***0.79***
Carboxylic acids and esters (%)		
Hexanoic acid	-	11.29
Benzoic acid	4.09	10.16
Propionic acid	0.54	-
Pyruvic acid	1.81	-
2-Methyl-2-pentanoic acid	-	0.10
4-Methylpentanoic acid	0.29	1.13
3-Hexenoic acid	-	0.91
Total	***6.73***	***23.59***
Ketones (%)		
Homofuronol	3.20	8.91
6-Methyl-3,5-heptadien-2-one	1.24	-
Methyl-2-pyrrolyl ketone	1.14	-
Total	***5.58***	***8.91***
Terpenes (%)		
Linaloloxide	0.51	0.19
Carvacrol	20.34	-
Thymol	3.36	-
Citronellol	1.97	-
Alpha-terpinene	0.95	-
Isoborneol	-	0.53
Total	***27.13***	***0.72***
Fatty acids and esters (%)		
Myristic acid	-	0.95
Stearic acid	-	5.11
Decanoic acid	2.08	0.62
Octanoic acid	2.81	-
Palmitic acid	2.45	-
Total	***7.34***	***6.68***
Acetic acids and esters (%)		
Lauryl acetate	6.31	10.89
Isopropyl acetate	-	0.45
n-Propyl acetate	-	1.48
Hexyl acetate	0.53	0.73
n-Butyl acetate	-	0.30
Total	***6.84***	***13.85***

Chemical compounds	*Thymus nummularius* M.Bieb.	*Vaccinium myrtillus* L.
Others (%)		
Quinoline	0.77	1.14
Ethyl-p-anisate	0.45	-
Acetanisole	3.68	-
Methyl benzoate	-	0.81
Isobutyl propionate	-	0.58
4-Methylacetophenone	1.11	-
Phenol	3.02	-
Dimethyl anthranilate	-	0.26
Neroloxide	-	0.47
2-Methoxy-3-methylpyrazine	-	0.89
Diethyl succinate	-	12.68
Total	***9.03***	***16.83***

Table 2. Agar well diffusion assay indicating inhibition zones (mm) of *Thymus nummularius* and *Vaccinium myrtillus* on various oral pathogens

	Thymus nummularius M.Bieb (51.2 µg/mL)	*Vaccinium myrtillus* L. (51.2 µg/mL)
S. mutans (clinical isolate)	9.00±0	11.33±0.58
S. mitis (clinical isolate)	9.00±0	14.33±0.58
S. salivarius DSM 13084	No zone	11.33±0.58
P. gingivalis ATCC 33277	8.00±0	9.33±0.58
C. albicans ATCC 90028	No zone	18.67±0.58

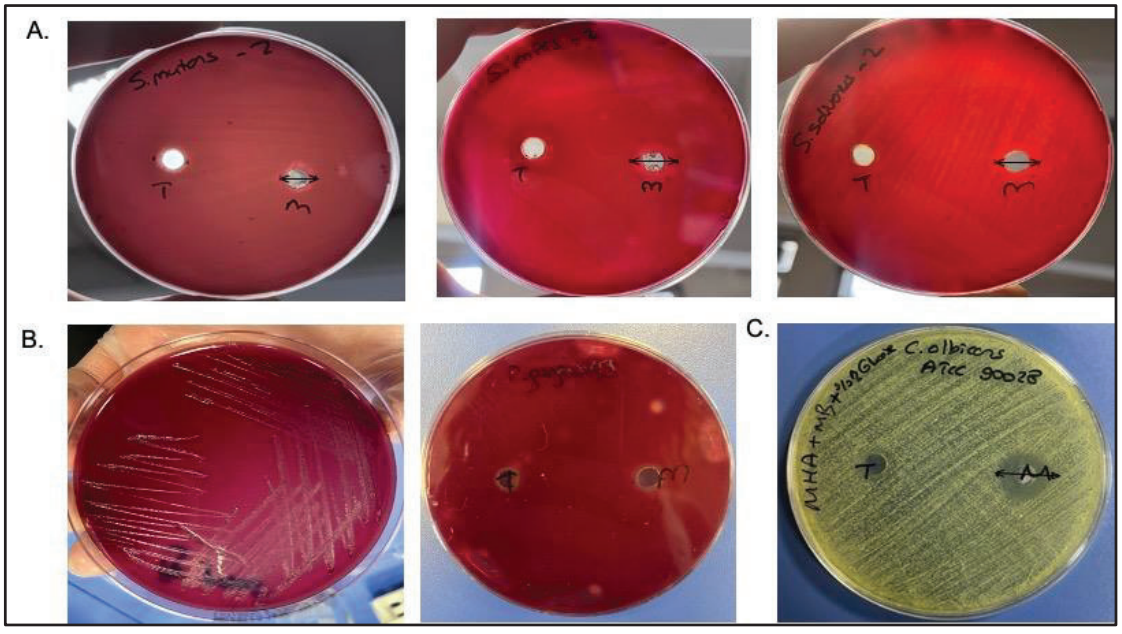

Figure 2. Agar well diffusion results for *V. myrtillus* L. and *T. nummularius* M. Bieb against tested oral microorganisms. A) Inhibition zones for viridans streptococci. B) Black pigmented colonies of *P. gingivalis* ATCC 33277 on Brucella Agar and agar well diffusion results. C) Agar well diffusion results for *C. albicans* ATCC 90028.

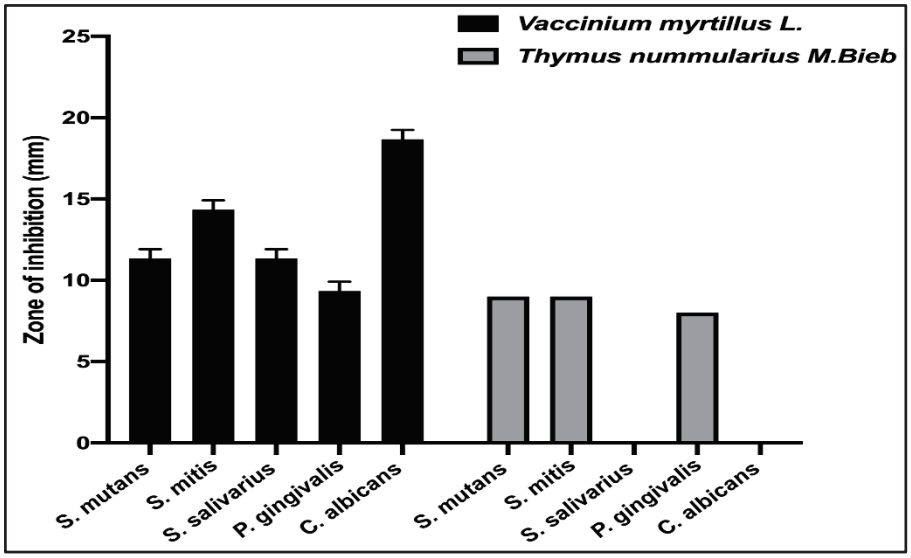

Figure 3. Inhibition zones for *V. myrtillus* L. and *T. nummularius* M. Bieb against tested oral microorganisms.

Table 3. Minimum inhibitory concentrations (µg/ml) of *T. nummularius* and *V. myrtillus* against tested oral pathogens

	***Thymus nummularius* M.Bieb**	***Vaccinium myrtillus* L.**
***S. mutans* (clinical isolate)**	>1024	512
***S. mitis* (clinical isolate)**	1024	256
***S. salivarius* DSM 13084**	>1024	512
***P. gingivalis* ATCC 33277**	>1024	>1024
C. albicans ATCC 90028	1024	128

DISCUSSION

As a result of GC-MS analysis, carvacrol, lauryl acetate, and thymol were detected 20.34%, 6.31% and 3.36% respectively at high levels in *T. nummularius*. On the other hand, Ertas et al. (2015) found thymol (60.38%) and terpinyl-acetate (10.49%) and Gercek et al. (2022) were determined thymol (38.91%), linalool (13.12%) and geraniol (6.51%) in *T. nummularius*. In present study, it found that differently from them carvacrol predominantly. Carvacrol and thymol, are considered important marker components for thyme. It was reported by Küçükbay et al. (2014) that carvacrol and thymol have high antioxidant activity. In addition, Tepe et al. (2011) said that carvacrol has an antifungal effect and thymol has an antiseptic effect. At the same time, thymol can be used against *Varroa destructor*, an important bee parasite. It is also preferred because it does not leave residue in bee products (Demirezen 2019, Turkun 2016). In a study conducted by Demirezen (2019), a total of 36 colonies were studied, with 9 colonies in each group (flumethrin, oxalic acid, thymol and control). In the study results; significant statistical differences were found between the varroa numbers falling according to the spring and autumn periods, according to the days, and according to the days within the same season. According to the percentage change formula, drug efficiencies were found as; flumethrin 85%, oxalic acid 80%, thymol 87% in the autumn period and flumethrin 75%, oxalic acid 68%, thymol 84% in the spring period. The results of this study showed that flumethrin was successful in long-term, oxalic acid and thymol were successful in short-term varroa control. On the other hand, Shah et al. (2020) reported the antioxidant properties of lauryl acetate. Lauryl acetate, also known as dodecyl acetate, has a floral odor and is useful as a perfume additive.

Diethyl succinate, hexanoic acid, lauryl acetate and benzoic acid were determined 12.68%, 11.29%, 10.89% and 10.16% respectively predominantly in *V. myrtillus*. Aranega-Bou et al. (2014) reported antifungal activity of hexanoic acid. Also, it was reported by Özkök et al. (2016) that benzoic acid has an expectorant, analgesic, and antiseptic activity. On the other hand, Elkıran and Avşar (2020) found 1,8-cineole (38.6%), α-pinene (21%), linalool (19.5%), α-terpineol (5.8%) in *V. myrtillus*, which was collected Sinop province of Turkiye. In this present study, it found of different components and it can be related and changed according to regions.

Antimicrobial outcomes of plants and natural products may be through inhibition of bacterial binding (adhesion) to cell walls, direct antimicrobial killing, or by effects that potentiate antibiotics, as evidenced with the aid of using diminished minimal inhibitory concentration (MIC) of antibiotics inside the presence of a plant in comparison to that of the antibiotic alone. Several natural products have been determined to have antimicrobial outcomes (Lee et al. 2006). Cranberry (*V. macrocarpon* Ait.) has effective antiadhesion properties (Dao et al. 2012).

According to antimicrobial results, *V. myrtillus* was more effective than *T. nummularius* against all tested microorganisms. Moreover, *V. myrtillus* exhibited a potent antimicrobial activity against *C. albicans*. The promising antimicrobial activity of *V. myrtillus* against *C. albicans* can be attributed to relatively higher carboxylic acids and esters (23.59%), especially benzoic acid. Benzoic acid has been shown to possess antimicrobial activity primarily against fungi (Teodoro et al. 2015).

Conclusion: In this study, chemical compound analyses showed that both plant species have high antioxidant components. *Vaccinium myrtillus* exhibited more potent antimicrobial activity when compared to *Thymus nummularius*. The antimicrobial activity of *V. myrtillus* against viridans streptococci and *C. albicans* was promising and may be used for the treatment of cariogenic microorganisms or oral Candidiasis in future studies. On the other hand, *T. nummularius* can use against to *Varroa destructor* because of thymol ingredient. At the same time, it is planned to study the contents of monofloral honey produced from these plants in further studies and to introduce these honeys to Turkish beekeeping. Therefore, it is essential to protect these plants.

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