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Research
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The Effect of Different Sowing Dates on the Essential Oil Content and Components of Common Black Cumin (*Nigella sativa* L.) and Damascus Black Cumin (*Nigella damascena* L.)

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Abstract

Objective: This study aimed to investigate the essential oil content and composition of common black cumin and Damascus black cumin (*Nigella damascena* L.) species grown in different sowing dates under Eastern Anatolian ecological conditions, Muş, Türkiye.

Materials and Methods: Seeds of *N. sativa* cv. Çameli was obtained from the Eskişehir Transitional Zone Agricultural Research Institute Directorate. Seeds of *N. damascena* were obtained from the Department of Field Crops, Faculty of Agriculture, Çukurova University. The field experiment was conducted at the Research and Application Field of the Faculty of Applied Sciences, Muş Alparslan University during the 2022 growing season. The experiment followed a split-plot design with three replications, including two species, and four sowing dates.

Results: The essential oil content ranged from 0.18% to 0.54%. Values were similar across the first three sowing dates but decreased markedly at the fourth sowing. Essential oil content was higher in common black cumin compared to Damascus black cumin. Major essential oil of components of common black cumin were 1,3-cyclopentadiene (56.73–71.59%), α-thujene (11.92–18.95%), longifolene (4.29–6.41%), terpinen-4-ol (2.46–5.22%), and β-pinene (2.35–6.40%). In Damascus black cumin β-elemene (49.01–59.24%) and α-selinene (18.56–35.41%) were identified as the dominant constituents.

Conclusion: Essential oil content in of common black cumin decreased with delayed sowing while the highest essential oil content *N. damascena* obteined at the third sowing date (May 25), followed by a sharp decline. The major component 1,3-cyclopentadiene in common black cumin reached its maximum at the first sowing date (April 25), whereas in Damascus black cumin the major component β-elemene was highest at the third sowing date (May 25).

Key words: Black cumin, *Nigella* spp., Sowing date, Essential oil content, Essential oil composition

Adi Çörek otu *(Nigella sativa* L.) ve Şam Çörek otu (*Nigella damascena* L.) Türlerinde Farklı Ekim Zamanı Uygulamalarının Uçucu Yağ Oranı ve Bileşenleri Üzerine Etkisi

Öz

Amaç: Bu çalışma Doğu Anadolu ekolojik koşullarında farklı ekim zamanlarında yetiştirilen *N. sativa* türü Çameli çörek otu çeşidi ve Şam çörek otu (*N. damascena*) türünün uçucu yağ oran ve bileşenlerini araştırmak amacı ile yürütülmüştür.

Materyal ve Yöntem: Araştırmada kullanılan *N. sativa* Çameli çeşidi Eskişehir Geçit Kuşağı Tarımsal Araştırma Enstitüsü Müdürlüğü'nden temin edilmiştir. *N. damascena* türü ise Çukurova Üniversitesi Ziraat Fakültesi Tarla Bitkileri Bölümü'nden alınmıştır. Araştırmada tarla denemesi Muş Alparslan Üniversitesi, Uygulamalı Bilimler Fakültesi, Araştırma ve Uygulama Arazisinde 2022 üretim sezonunda yürütülmüştür. Deneme 2 farklı türde 4 farklı ekim zamanında bölünmüş parseller deneme desenine göre 3 tekrarlamalı olarak yürütülmüştür.

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Bulgular: Uçucu yağ oranı % 0.18-0.54 arasında değişmiştir. İlk üç ekim zamanında uçucu yağ oranı açısından benzer sonuçlar elde edilmiş, ancak dördüncü ekim zamanında belirgin bir düşüş kaydedilmiştir. Adi çörek otunda uçucu yağ oranı Şam çörek oyuna göre daha yüksek bulunmuştur. Adi çörek otunda uçucu yağ ana bileşenleri, 1,3-cyclopentadiene % 56.73 - 71.59, α-thujene % 11.92 – 18.95, longifolene % 4.29 – 6.41, terpinen-4-ol 2.46 – 5.22 %, β-pinene % 2.35 – 6.40, Şam çörek otunda uçucu yağ ana bileşenleri, β-elemene % 49.01 - 59.24, α-selinene % 18.56 – 35.41 olarak saptanmıştır.

Sonuç: Uçucu yağ oranı değerleri Adi çörek otunda ekim zamanının gecikmesi ile düşüş göstermiştir. Şam çörek otunda ise üçüncü ekim zamanında (25 Mayıs) en yüksek uçucu yağ oranlarına ulaşılmıştır, daha sonraki ekimde ise sert bir düşüş göstermiştir. Adi çörek otunda ana bileşen 1,3-cyclopentadiene en yüksek değer birinci ekim zamanında (25 Nisan), Şam çörek otunda ise ana bileşen β -elemene en yüksek üçüncü ekim zamanında (25 Mayıs) tespit edilmiştir.

Anahtar Kelimler: Çörek otu, Ekim zamanı, *Nigella* spp., Uçucu yağ oranı, Uçucu yağ bileşenleri

Introduction

Black cumin L. grows naturally in North Africa, Southern Europe, the Middle East, and Southwest Asia, and is used for various industrial and medicinal purposes (Helvacıoğlu et al., 2021; Topcagic et al., 2017; Shahbazi et al., 2017). It is widely cultivated, especially in India, Pakistan, Iran, Syria, Türkiye, Ethiopia, Oman, and Saudi Arabia (Sharma et al., 2019; Khare, 2004). The seeds are currently attracting significant interest in the food, cosmetic, and pharmaceutical industries (Benazzouz-Smail et al., 2023).

Türkiye has a rich genetic diversity of *Nigella* species. While there are 22 known species worldwide, 19 taxa occur in Türkiye. Among these, *Nigella lancifolia* Hub.-Mor., *Nigella arvensis* L. var. *anatolica* M. Zohary and *Nigella arvensis* L. var. *oblanceolata* P.H. Davis are endemic, while the other remaining 16 taxa are not (Bakış et al., 2011).

Black cumin seeds are rich in carbohydrates (40.0%), fat (28.5%), and protein (26.7%) (Niu et al., 2020), and can be used as an energy booster and dietary supplement (Benazzouz-Smail et al., 2023). Black cumin seeds contain significant amounts of calcium, potassium, phosphorus, magnesium, sodium, copper, zinc, iron, and manganese (Helvacıoğlu et al., 2021). Black cumin seed oil plays an important role in human nutrition and health (Albakry et al., 2022).

Scientific studies on *Nigella* species worldwide have generally focused on *Nigella sativa*, with very little research on *N. damascena* and other species (Korablova et al. 2021; Shanaida et al. 2024).

Recent scientific studies have shown that *N. sativa* seeds possess antimicrobial properties (Iqbal et al., 2017; Korablova et al., 2021), antihistamine (Gholamnezhad et al., 2019), anti-cancer activity (Butt et al., 2019; Majdalawieh and Fayyad, 2016), anticonvulsant (Noor et al., 2012), anti-inflammatory (Kokoska, 2011; Kökdil et al., 2005; Korablova et al., 2021), antidiabetic (Rani et al., 2018), and antihypertensive properties (Shahbazi et al., 2022; Korablova et al., 2021), diuretic and antioxidant (Toma et al., 2015; Korablova et al., 2021), has demonstrated immunomodulatory, hepatoprotective, neuroprotective, cholesterol-lowering, and wound-healing effects (Usama, et al., 2011; Islam et al., 2021; Ahmad et al., 2021; Korablova et al.2021). The biological properties mentioned are related to the richness of various phytochemicals, primarily phenolic compounds, terpenes, essential oils, saponins, and alkaloids (Shahbazi et al., 2022; Toma et al., 2015; D'Antuono et al., 2002; Benazzouz-Smail et al., 2023)

No clinical studies have yet been conducted on *Nigella damascena* L. (love flower). *N. damascena* is primarily known as an ornamental plant and is cultivated less frequently for its medicinal properties. (Salehi et al., 2021; Shanaida et al. 2024).

The sesquiterpene beta-elemene component of *N. damascena* exhibits anti-tumor effects by inhibiting glycolysis in cancer cells (Xie Q et al., 2020; Dong et al., 2024) and demonstrates anti-mycobacterial properties. It has been determined that beta-elemene, the main component of *N. damascena* essential oil, exhibits antimicrobial effects against Mycobacterium tuberculosis (Sieniawska et al., 2018). Researchers have suggested that *N. damascena* essential oil, along with damascenone and beta-elemene isolated from it, exhibits multifactorial mechanisms of immunomodulatory activity (Sieniawska et al., 2019). The content of bioactive compounds varies among different varieties and chemotypes within the same species and has a significant effect on the medicinal properties of plants (Shanaida et al., 2024).

It has been stated that N. damascena L. seeds are richer in phenolic compounds than *N. sativa* L. seeds.

However, this difference was quantitative rather than qualitative (Benazzouz-Smail et al., 2023).

Bioactive compounds found in *N. sativa* seeds, such as thymoquinone, α-hederin, and nigellidin, have been reported to show promise in combating COVID-19 (Islam et al., 2021; Khazdair et al., 2021; Korablova et al., 2021).

This study was conducted in the province of Muş, which has Türkiye's third largest plain (165,000 ha). Muş province has very high agricultural potential. The population engaged in agriculture in the province is 65 percent. With the construction of the Alparslan 1 and later the Alparslan 2 dams, the transition to irrigated agriculture in the province has accelerated. Sugar beet cultivation is predominantly practiced as a summer crop under irrigated conditions. The number of alternative crops is limited. It is necessary to increase the number of crops that can generate high income per unit area and enrich the crop rotation. This study aims to investigate the best optimal sowing date for achieving high-quality seed production of both black cumin species that can adapt to the Eastern Anatolian ecological conditions, Muş, Türkiye.

Materials and Methods

Plant Materials

The *N. sativa* cv. Çameli variety used in the study was obtained from the Directorate of the Eskişehir Transitional Zone Agricultural Research Institute. It was the first *N. sativa* variety registered in Türkiye in 2014. The Çameli variety has a plant height of 40-70 cm, a fixed oil content of 25%, and a seed yield of 140-220 kg/da in dry conditions and 160-260 kg/da in irrigated conditions. Seeds of *N. damescena* were supplied by the Department of Field Crops, Faculty of Agriculture, Çukurova University.

Methods

Experimental Design and Site Conditions

The field experiment was conducted at the Research and Application Field of the Faculty of Applied Sciences, Muş Alparslan University during the 2022 production season. The soil at the trial site has a loamy texture with a pH of 6.61, an organic matter content of 2.21% and available phosphorus (P_2O_5) of 2.20 kg/ha. The climatic characteristics during the period from April to November in which the trial was carried out were an average temperature of 11.05°C, rainfall of 395.8 mm and an average relative humidity of 58.7%. The precipitation during this period was lower than the long-term average of 758.9 mm, the average temperature was 1.7 °C higher than the long-term average and the relative humidity was similar. A total of 32 mm of precipitation fell in April, 91.6 mm in May and 16 mm in June, while no precipitation fell in July and August.

The experiment followed a split-plot design with three replications. In the experiment, the sowing dates were in the main plots, while the species were in the sub-plots. The main plots consisted of four sowing dates: April 25, May 10, May 25, and June 9, 2022. The subplots consisted of the two species (*N. sativa* and *N. damascena*). Harvests were performed on August 23, September 1, September 20, and September 30 in 2022, respectively.

Essential Oil Extraction and Analysis

In this study, the essential oil components were determined in the Central Laboratory for Application and Research at the University of Bingöl. First, 100 g of each of the ground black cumin seed samples were weighed separately and the essential oils were extracted using a Neo-Clavenger device. The essential oil components were analysed using a Gas chromatography-mass spectrometry (GC-MS) device.

The essential oil content (%) and essential oil components (%) were analysed in the study.

Methods Used in Oil Analyses and Component Analyses

Velp scientifica ser 148 solvent extractor device analysis conditions: Solvent Used: Hexane, Plate Temperature: 130 °C, Immersion Time: 120 min, Washing Time: 60 min, Recovery Time: 60 min.

Sample analysis: The crucible was brought to a constant weight at 105 °C and the initial weighing was performed. The sample was weighed and placed in the device's cartridge. 80 ml of hexane was added to the crucible. The sample was processed in the device using the hexane method. After the analysis was completed, the final weighing of the crucible was performed and the result was recorded.

GC-MS/Fid fatty acid analysis report: First, 100 g of each sample of ground black cumin seeds was weighed separately and the volatile oils were extracted using a Neo-Clavenger device.

GC-MS liquid sample preparation: This procedure was performed according to IUPAC method 2.301. 100 microlitres of volatile oil sample was placed in a capped tube. Ten millilitres of hexane were added and vortexed. One hundred microlitres of 2 NKOH (methanolic) were added and vortexed for 30 seconds.

The mixture was centrifuged at 4500 rpm for 10 minutes, and 1.5 ml was taken from the upper layer into a vial for subsequent analysis by GC-MS.

Method used for analysing volatile oil components with a GC-MS device: An Agilent 78904 model GC, 5975C model MS and FID detector were used simultaneously in the analysis. Column J&W 122-7061 specifications: 250 °C: 60 m x 250 µm x 0.15 µm. Injection volume 1 µm and split 10:1 mode selected. Column flow rate 1 mL (carrier gas helium) inlet pressure: 20.83 psi. Total flow: 34 ml/min. Chromatographic conditions: Starting at 50 °C, it waits for 1 minute and reaches 200 °C at a rate of 25 °C/min, then reaches 230 °C at a rate of 3 °C/min and waits for 15 minutes. The total analysis time is 32 minutes. MS results were identified by comparison with the Wiley and NIST libraries stored in the device's memory.

This article is from Irfan ÖZDEMİR's master's thesis entitled "Investigation of the yield and quality characteristics of common black cumin (*Nigella sativa* L.) and Damascus black cumin (*Nigella damascena* L.) at different sowing dates under Muş conditions" at Muş Alparslan University.

Results and Discussion

Essential oil content (%)

As shown in Table 1 and Figure 1, the essential oil content varied between 0.18% and 0.54% depending on the interaction between sowing date and species. The lowest essential oil content was 0.18% in *N. damascena* at the first sowing date, while the highest essential oil content was 0.54% in common black cumin at the first sowing date. The average essential oil content was similar in the 1st, 2nd and 3rd sowings, but was lower in the 4th sowing. In terms of species, the essential oil content was higher in common black cumin. Regarding the date of sowing, the highest essential oil content was obtained in the first sowing (25 April) of common black cumin and in the third sowing (25 May) of Damascus black cumin. According to the results of the tests, the essential oil content is close to the values reported by some researchers (Burits and Bucar, 2000; D'antuono et al., 2002; Özel et al., 2009; Akgören, 2011; Margout et al., 2013; Çamlica and Yaldız, 2019; Anonymous, 2020; Bayati et al., 2020; Keser and Gedik, 2021) and lower than the values reported by some researchers (Ertuğrul, 1986; Geren et al., 1997; Burits and Bucar, 2000; Ashraf et al., 2006; Tektaş, 2015; Kızılyıldırım, 2019; Aysabar, 2021), while some researchers considered them high (Ertuğrul, 1986; Geren et al., 1997; Burits and Bucar, 2000; Ashraf et al., 2006; Tektaş, 2015; Kızılyıldırım, 2019; Aysabar, 2021). Some researchers have reported that the volatile oil content in plants depends on their genetic structure, harvest date, cultivation conditions, and abiotic stresses (Muzik et al., 1989; Stutte, 2006; Rezaei et al., 2019).

Data on the essential oil contents of common black cumin and *N. damascena* at different sowing dates under Muş conditions are provided in Table 1. The data on the essential oil content of common black cumin (*N. sativa* L.) and Damascus black cumin (*N. damascena* L.) at different sowing dates under Muş conditions are shown in Figure 1.

Essential oil composition

The essential oil components of *N. sativa* according to different sowing dates are given in Table 2 and Figure 2. As shown in Table 2 and Figure 2, the essential oil components in common black cumin seeds were highest at the first sowing date, with 1,3-cyclopentadiene at 71.59%, α-thujene at 11.92%, longifolene at 4.29%, terpinen-4-ol 2.46%, β-pinene 2.35%, and at the second sowing date.

Table 1. Fixed and essential oil values in *N. sativa* and *N. damascena* according to sowing date

Parameter	***N. sativa***				***N. damascena***			
	Sowing dates				**Sowing dates**			
	1st	**2nd**	**3rd**	**4th**	**1st**	**2nd**	**3rd**	**4th**
Essential oil content (%)	0.54	0.39	0.28	0.23	0.18	0.33	0.45	0.22
Mean	0.36				0.30			

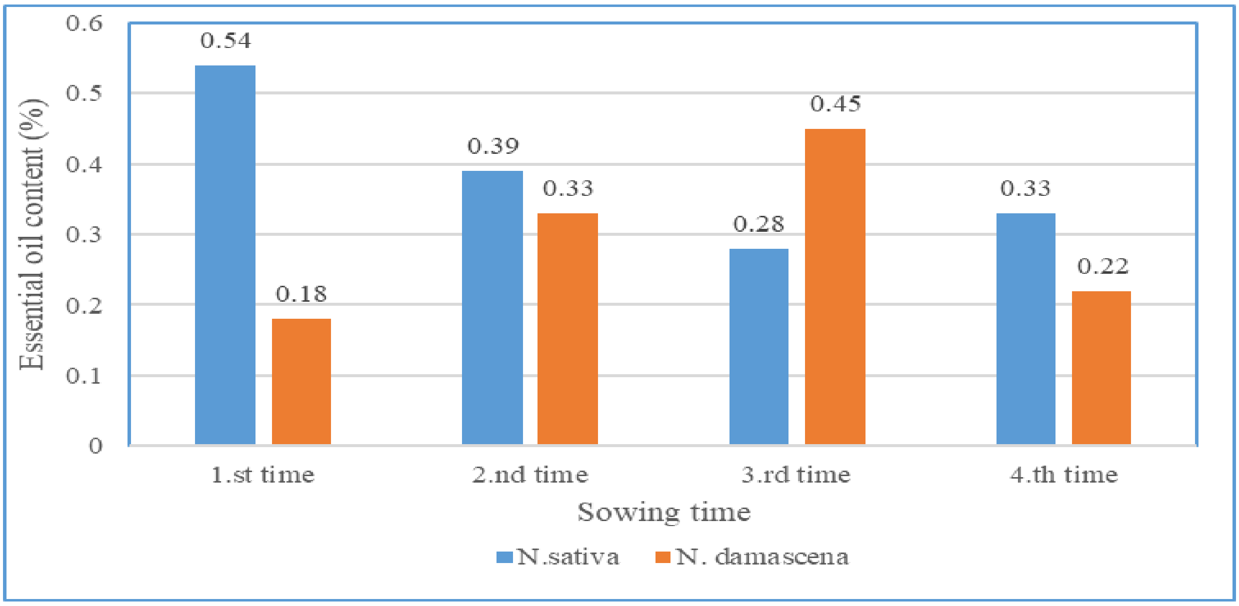

Figure 1. Essential oil content (%) in *N. sativa* and *N. damascena* according to sowing dates

They were highest with 1,3-cyclopentadiene 65.35%, α-thujene 17.17%, longifolene 5.94%, β-pinene 4.1%, terpinen-4-ol 3.94%. The third sowing date showed the highest contents of 1,3-cyclopentadiene 62.93%, α-thujene 13.49%, longifolene 6.41%, terpinen-4-ol 5.22%, β-pinene 6.1%, and the fourth sowing date had the highest content of 1, 3-cyclopentadiene 56.73%, α-thujene 18.95%, β-pinene 6.4%, longifolene 5.11%, terpinen-4-ol 4.63%, limonene 3.07%.

The components of the essential oils and their contents varied considerably depending on the date of sowing in the study. While the major component in our study was 1,3-cyclopentadiene, Moretti et al., (2004) and Rchid et al., (2004) identified p-cymene. Many researchers have reported that the number and ratio of components in essential oils depend on variables such as variety, species, climate, soil, fertilization, and cultivation techniques (Burits and Bucar, 2000; D'antuono et al., 2002; Moretti et al., 2004; Benckiser and Schnell, 2006; Wajs et al., 2009; Toma et al., 2010; Güllü and Gülcan, 2013; Heshmati and Namazi, 2015; Rioba et al., 2015; Seyyedi et al., 2015; Sieniawska, 2018).

Table 2. Essential oil components (%) in *N. sativa* according to different sowing dates

Serial Number	Components	RT	*Nigella sativa* sowing date 1st	2nd	3rd	4th
1	α-Thujene	7.074	11.92	17.17	13.49	18.95
2	β-Pinene	7.593	2.35	4.10	6.10	6.40
3	Limonene	8.112	0.33	1.15	1.57	3.07
4	1,3-Cyclopentadiene	8.350	71.59	65.35	62.93	56.73
5	Terpinen-4-Ol	10.146	2.46	3.94	5.22	4.63
6	Longifolene	10.851	4.29	5.94	6.41	5.11
7	9,17-Octadecadienal	11.879	1.16	0.22	0.00	0.00
8	Musk Ambrette	12.190	2.47	0.00	0.00	0.00
9	Carvacrol	15.282	0.92	0.47	0.52	0.17
10	1-Cyclohexene	9.938	0.00	0.00	2.66	0.00
	Total		97.49	98.34	98.90	95.06

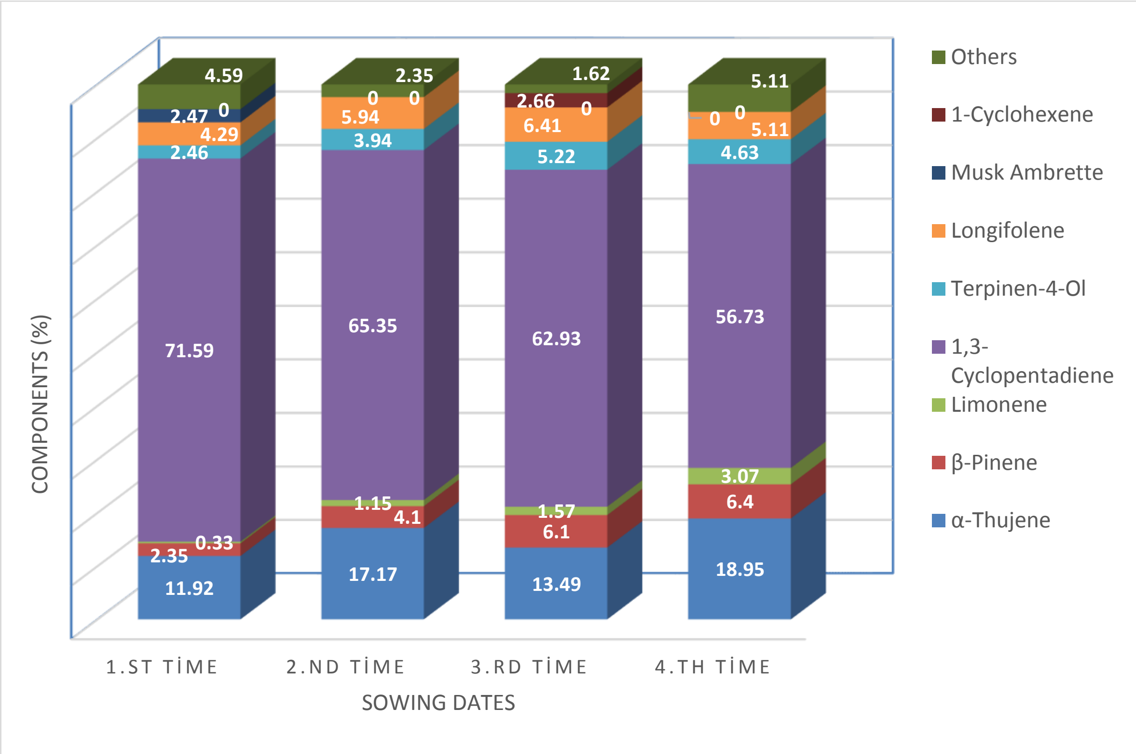

Figure 2. Essential oil components (%) in *N. sativa* according to sowing dates

Seventeen components were found in the first sowing, sixteen in the second sowing, fifteen in the third sowing and twenty-two in the fourth sowing.

The components of Damascus black cumin essential oil according to different sowing dates are given in Table 3 and Figure 3.

Table 3. Essential oil components (%) in *N. damascena* according to different sowing dates

Serial Number	Components	RT	*Nigella damascena* sowing date			
			1st	2nd	3rd	4th
1	β-Elemene	10.634	59.24	55.84	56.98	49.01
2	β-Patchoulene	11.298	1.84	0.00	0.00	0.00
3	α-Selinene	11.749	24.70	18.56	32.79	35.41
4	7 Epi- α-Selinene	12.040	7.15	0.00	0.00	0.00
5	9-Hexadecen	13.954	4.27	0.00	0.00	0.00
6	Selin-11	15.324	1.67	0.97	0.00	2.91
7	γ -Gurjunene	16.434	0.03	12.80	0.30	0.01
8	α-Panasinsen	12.050	0.00	8.82	7.46	8.63
9	9-Hexadecen-1-Ol	13.975	0.00	2.10	0.91	0.00
10	Oxirane, Tridecyl	13.975	0.00	0.00	1.04	0.00
11	Cis-11-Tetradecen-1-Ol	13.970	0.00	0.00	0.00	2.86
Total			98.90	99.09	99.48	98.83

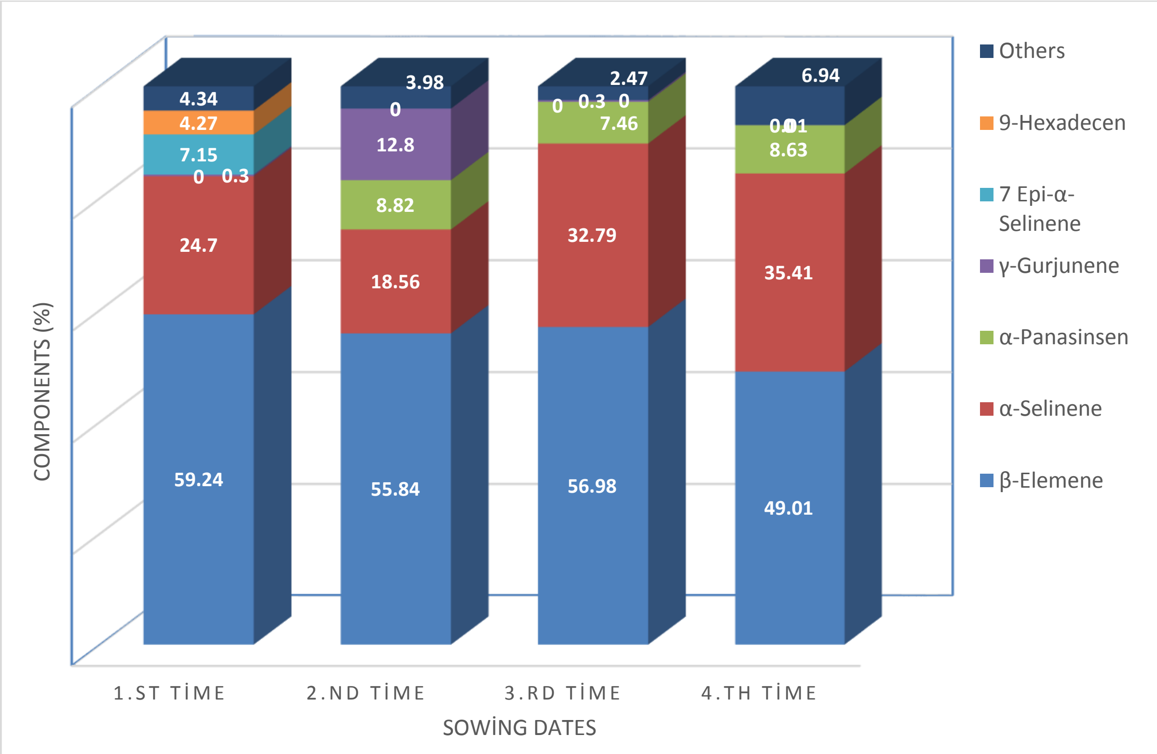

Figure 3. Essential oil components (%) in *N. damascena* according to sowing dates.

As shown in Table 3 and Figure 3, the major essential oil components in Damascus black cumin in the first sowing period are β-elemene 59.24%, α-selinene 24.7%, 7-epi-α-selinene 7.15%, 9-hexadecene 4.27%, in the second sowing date β-elemene 55.84%, α-selinene 18.56%, γ-gurjunene 12.8%, α-panasinene 8.82%, at the third sowing date β-elemene 56.98%, α-selinene 32.79%, α-panasinene 7.46%, and at the fourth sowing date β-elemene 49.01%, α-selinene 35.41%, α-panasinene 8.63%. In the first sowing 13 components were found, in the second sowing 17 components, in the third sowing 18 components and in the fourth sowing 19 components. This is consistent with the results on β-elemene, one of the main components of essential oils (Burits and Bucar, 2000; Moretti et al., 2004; Rchid et al., 2004; Ulusu, 2021; Shanaida et al., 2024), while the ratios of the other components were different. According to many researchers, the components of the essential oil and their relationship to each other vary depending on the variety, species, climate, soil, fertilization and cultivation technique (Burits and Bucar, 2000; D'antuono et al., 2002; Moretti et al., 2004; Benckiser and Schnell, 2006; Wajs et al., 2009; Toma et al., 2010; Güllü and Gülcan, 2013; Heshmati and Namazi, 2015; Rioba et al., 2015; Seyyedi et al., 2015; Sieniawska, 2018).

Conclusion

This study demonstrated that both essential oil content and composition in common black cumin and Damascus black cumin are significantly influenced by sowing date under the Eastern Anatolian ecological conditions Muş, Türkiye. The results are summarised below.

Essential oil content: The values of the essential oil content of common black cumin decreased with delayed sowing dates. For Damascus black cumin, the highest essential oil content was reached at the third sowing (May 25), followed by a strong decrease at the following sowings.

Essential oil components: The major components of the essential oil of common black cumin were identified as 1,3-cyclopentadiene, α-thujene, β-pinene, longifolene, terpinen-4-ol and limonene. The essential oil composition and their contents to each other varied depending on the sowing date in the study. The major essential oil components in Damascus black cumin were identified as β-elemene, α-selinene, α-panasinene, γ-gurjunene, 7-epi-α-selinene and 9-hexadecene. In common black cumin, 1,3-cyclopentadiene was the highest major essential oil component on the first sowing date (April 25), while in Damascus black cumin species, β-elemene was the highest major essential oil component on the third sowing date (May 25).

In this study, which investigated the effects of different sowing dates on the essential oil content and composition of two *Nigella* species under the ecological conditions of Muş, the end of April was found to be the most suitable sowing date for *N. sativa*, while the end of May was found to be the most suitable for *N. damascena*.

Conflict of Interest

The authors have declared no conflict of interest.

Author Contrubiton Statement

The authors have equal contributions.

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