



ADVANCES IN  
MEDICINAL AND  
AROMATIC PLANTS:  
*From Phytochemistry to  
Sustainable Use*

EDITOR  
Assoc. Prof. Dr. Gülen ÖZYAZICI



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**Advances in Medicinal and Aromatic Plants: From  
Phytochemistry to Sustainable Use**

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## CONTENTS

<b>PREFACE</b>	<b>1</b>
----------------	----------

### **CHAPTER 1**

<b>Chemical Composition of Safflower (<i>Carthamus tinctorius</i> L.) Petals and Their Use in the Food Industry</b> Prof. Dr. Belgin COŞGE ŞENKAL .....	<b>3</b>
----------------------------------------------------------------------------------------------------------------------------------------------------------------	----------

### **CHAPTER 2**

<b>Changes in the Aroma Composition of Different Plant Parts in Stevia (<i>Stevia rebaudiana</i> Bertoni) Depending on Various Drying Methods</b> Dr. Aysel Özcan AYKUTLU Assoc. Prof. Dr. Emine YURTERİ .....	<b>17</b>
------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------	-----------

### **CHAPTER 3**

<b>Effects of Heavy Metal Stress on Secondary Metabolite Production in Pot Marigold (<i>Calendula officinalis</i> L.) Under in vitro Conditions</b> Assoc. Prof. Dr. Emine YURTERİ Sümeyye Beyza BUÇAN .....	<b>33</b>
----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------	-----------

### **CHAPTER 4**

<b>A Look at the Importance and Future of Micro Green Medicinal Plants</b> Assoc. Prof. Dr. Emine YURTERİ Dr. Aysel Özcan AYKUTLU .....	<b>57</b>
---------------------------------------------------------------------------------------------------------------------------------------------------	-----------

### **CHAPTER 5**

<b>Botanical Characteristics, Medicinal Effects and Uses of Rosehip (<i>Rosa canina</i>) Plant</b> Dr. Haydar KÜPLEMEZ .....	<b>89</b>
-------------------------------------------------------------------------------------------------------------------------------------	-----------

### **CHAPTER 6**

<b>Distribution Areas and Medicinal Effects of Thymus Species in the Eastern Black Sea Region of Türkiye</b> Dr. Haydar KÜPLEMEZ .....	<b>104</b>
-----------------------------------------------------------------------------------------------------------------------------------------------	------------

**CHAPTER 7**

---

***Berberis sp.:* An Important Medicinal Plant in Global Traditional  
Medicine**

Assoc. Prof. Dr. Amir RAHİMİ

PhD Student. Samira MORADZADEH

Assoc. Prof. Dr. Gülen ÖZYAZICI ..... 122

---

**CHAPTER 8**

---

**Introducing Drought-Resistant Medicinal Plants**

Assoc. Prof. Dr. Amir RAHİMİ

PhD Student. Shiva AFSHARNIA ..... 150

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## PREFACE

Medicinal and aromatic plants synthesize a wide range of bioactive secondary metabolites—such as essential oils, alkaloids, phenolics, and terpenoids—that form the basis of traditional medicine systems and modern pharmaceutical, cosmetic, and food industries. In recent decades, growing global interest in natural products, coupled with concerns over synthetic chemicals and environmental sustainability, has renewed scientific and commercial attention toward medicinal and aromatic plants.

The increasing challenges posed by climate change, land degradation, and water scarcity have further emphasized the importance of resilient plant species capable of thriving under marginal conditions. Many medicinal and aromatic plants are naturally adapted to arid and semi-arid environments, displaying unique morphological, physiological, and biochemical strategies that allow them to tolerate drought and other abiotic stresses while maintaining valuable phytochemical profiles. These characteristics make them promising candidates for sustainable agriculture, particularly in regions where conventional crop production is increasingly constrained.

This book aims to provide a comprehensive and interdisciplinary overview of medicinal and aromatic plants, integrating botanical, agronomic, physiological, phytochemical, and socio-economic perspectives. The chapters address key topics including plant diversity and taxonomy, bioactive compounds and their medicinal relevance, cultivation and management practices, stress tolerance mechanisms, quality and yield relationships, and emerging approaches for sustainable production. Special emphasis is placed on species adapted to dry and semi-dry regions, reflecting the growing need for climate-resilient cropping systems.

The content of this book is intended for a broad audience, researchers, extension specialists, and professionals working in agriculture, pharmacognosy, plant sciences, and natural product industries. It is our hope that this book will serve not only as a reference source but also as an inspiration for future research and innovation in the field of medicinal and aromatic plants, fostering collaboration between science, industry, and traditional knowledge systems..

Best regards

Assoc. Prof. Dr. Gülen ÖZYAZICI



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**Chemical Composition of Safflower  
(*Carthamus tinctorius* L.) Petals and Their Use in  
the Food Industry**

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## INTRODUCTION

Safflower (*Carthamus tinctorius* L., Family: Asteracea or Compositae) is an important oilseed crop that has been cultivated since ancient times (Emongor, 2010; Li et al., 2023). There are approximately 25 species of the *Carthamus* genus distributed worldwide, of which only *C. tinctorius* is cultivated (Arslan, 2018). Safflower is cultivated primarily for its seeds, which are used for edible oil, bird food, or for its flowers, which are used as a dye source and for medicinal purposes (Labdelli et al., 2019; Li et al., 2023).

Safflower seeds contain between 35-48% oil and approximately 20% protein. Safflower oils are rich in unsaturated fatty acids (C18H34O2-oleic acid and C18H32O2-linoleic acid) (Assefa et al., 2021; Kurt et al., 2025). This makes them a popular choice among consumers conscious of healthier eating. Furthermore, the growing global demand for organic farming and organic products is increasing the popularity of safflower production. There is significant interest in developing safflower-based products, particularly in the food and cosmetics industries.

Safflower is a temperate zone plant that grows in arid and semiarid regions of the world (Zemour et al., 2021). India, one of the countries with the largest safflower cultivation area in the world, is used for oil production and is also used in traditional medicine and as a spice. In Mexico, one of the countries with the largest commercial production of safflower, its flowers are used for natural dye production and cooking oil production (Erbaş and Mutlucan, 2023).

In the USA, safflower is used for high-quality cooking oils and paints, while in China, safflower is widely used in the pharmaceutical and cosmetic sectors, both as a source of oil and as an important plant used in traditional Chinese medicine (Emongor, 2010; Cheng et al., 2024). Ethiopia, a leading center for safflower production in Africa, is used for traditional food and oil production (Kemale and Hailu, 2019).

Kazakhstan, with its extensive agricultural lands, has significant safflower cultivation potential and is used for oil production and feed (Didorenko et al., 2025). Safflower cultivation is also practiced in countries such as Australia, Canada, Russia, Argentina, Iran, Afghanistan, and Pakistan. Türkiye, which ranks 7th among safflower producers globally, has seen a steady increase in safflower cultivation in recent years (Anonymous, 2025). Production is being supported to increase agricultural diversity, meet biofuel production, and meet cooking oil needs. In recent years, climate change, drought, and the increasing demand for biofuels have increased the importance of safflower cultivation. Türkiye, Kazakhstan, and some African countries are expanding their cultivation areas thanks to this plant's drought resistance.

While safflower is traditionally known as an oilseed crop, its petals of flowers also contain important bioactive compounds. The natural pigments, phenolic compounds, and flavonoids derived from the petals are valued in the food industry as additives, colorants, and functional ingredients (Adamska and Biernacka, 2021). Due to the potential health risks associated with synthetic additives, interest in natural and sustainable sources is increasing. In this context, safflower flowers offer a valuable alternative.

The primary objective of this study was to examine the potential use of safflower petals in the food industry based on scientific evidence. The bioactive compounds found in safflower petals, their significance, and the potential use of safflower flowers in the food industry were evaluated.

## **1. THE FLOWERS OF SAFFLOWER**

The flowers are in a small sepal at the end of the main stem and side branches. The sepal diameter is 2-5 cm. The number of sepals per plant ranges from 5-50. The sepal contains between 20-180 flowers, which varies depending on the variety and environmental conditions.

The sepal is surrounded by green bracts. The outermost row of bracts has sharp spines at the tips in spiny varieties. Inside the bracts, the sepals and petals are fused, forming a tube. Flower colors range from whitish yellow to orange-yellow and orange red (Figure 1). Safflower flowers have five stamens and one pistil. Flowers bloom in the sepals on the first side branches of the safflower plant, followed by secondary branches. Therefore, the flowering period of a plant can range from 10-40 days. Flowering progresses from the outside of the sepal to the center, and flowering is complete within 3-5 days (Dajue and Mündel, 1996; Emongor, 2010; Sharma et al., 2022).



**Figure 1:** Safflower flowers and petals.

## **2. CHEMICAL COMPOSITION OF PETALS**

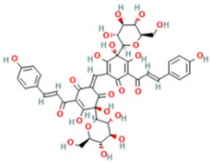
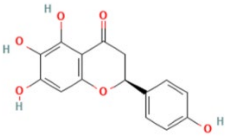
Safflower petals are quite rich in chemical composition (Adamska and Biernacka, 2021). They contain macro and microelements. The total element content of the petals was determined to be 3507.01 mg/100 g, and they are rich in K (2040 mg/100 g) and Ca (680 mg/100 g). Safflower petals contain protein, oil, and crude fiber (Barashovets and Popova, 2016; Kim et al., 2000).

# Advances in Medicinal and Aromatic Plants: From Phytochemistry to Sustainable Use

## Chapter 1

Safflower petals contain two important color pigments: carthamin and carthamidin. Basic information about these pigments (PubChem, 2025) is presented in Table 1.

**Table 1:** Basic information about color pigments in safflower petals

Compounds	Molecular Formula	Structure	Uses
Carthamin	$C_{43}H_{42}O_{22}$		It is the most important pigment in safflower flowers. It is a red color, is insoluble in water, and is soluble in alcohol and alkaline solutions. It is used as a natural red colorant in the food industry.
Carthamidin	$C_{15}H_{12}O_6$		It is a yellow, water-soluble pigment used primarily in baked goods and beverages.

Petals contain numerous phenolic compounds. Phenolic compounds are divided into two groups: phenolic acids and flavonoids. Most identified phenolic substances are natural flavonoids. Flavonoids are polyphenolic antioxidants found naturally in herbal teas, fruits, and vegetables. Phenolic acids found in foods influence color, aroma, odor, bitterness, astringency, and oxidative stability. Phenolic acids have gained significant importance in recent years due to their potential to protect against fatal diseases, particularly cancer and coronary heart disease.

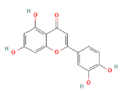
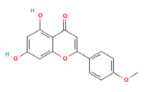
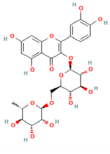
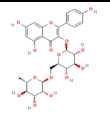
Safflower flower petals contain high amounts of flavonoids, particularly apigenin and luteolin derivatives. Flavonoids are widely distributed throughout the plant kingdom. They constitute the largest

# *Advances in Medicinal and Aromatic Plants: From Phytochemistry to Sustainable Use*

## *Chapter 1*

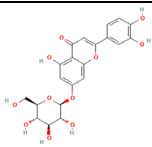
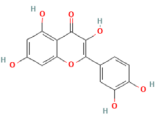
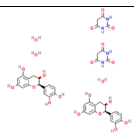

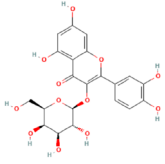
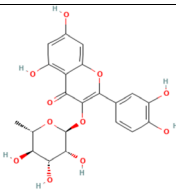
group of polyphenols and are thought to be responsible for the color and flavor of many fruits and vegetables. A variety of biological activities, including antioxidants, anti-inflammatory, estrogenic, antimicrobial, and antitumor properties, have been reported for flavonoids (Hussien, 2017). Quercetin, the primary constituent of safflower flowers, is a flavonoid-type dye (Shirwaikar et al., 2010). The structure and biological activities/effects of some important flavonoids in safflower petals are given in Table 2 (PubChem, 2025).

**Table 2:** Some flavonoids in safflower petals and their biological effect potential

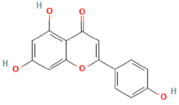
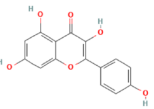
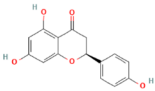
Compounds	Molecular Formula	Structure	Biological Activity/Effect
Luteolin [5280445] <sup>1</sup>	C <sub>15</sub> H <sub>10</sub> O <sub>6</sub>		antioxidant anti-inflammatory antitumor
Acacetin [5280442]	C <sub>16</sub> H <sub>12</sub> O <sub>5</sub>		anticonvulsant effect
Rutoside syn: Rutin [5280805]	C <sub>27</sub> H <sub>30</sub> O <sub>16</sub>		antioxidant
Nictoflorin Kaempferol-3-O- rutinoside [5318767]	syn: C <sub>27</sub> H <sub>30</sub> O <sub>15</sub>		antioxidant

# Advances in Medicinal and Aromatic Plants: From Phytochemistry to Sustainable Use Chapter 1

**Table 2:** Some flavonoids in safflower petals and their biological effect potential (Cont.)

Cinaroside Luteolin 7-glucoside [5280637]	syn: C <sub>21</sub> H <sub>20</sub> O <sub>11</sub>		antioxidant
Quercetin [5280343]	C <sub>15</sub> H <sub>10</sub> O <sub>7</sub>		antibacterial agent antioxidant protein kinase inhibitor antineoplastic agent
Epicatechin [168346974]	C <sub>38</sub> H <sub>42</sub> N <sub>4</sub> O <sub>21</sub>		
Resorcinol [5054]	C <sub>6</sub> H <sub>6</sub> O <sub>2</sub>		antiseptic disinfectant antithyroid
Quercetin-3-galactoside syn: Hyperin, Hyperoside [5281643]	C <sub>21</sub> H <sub>20</sub> O <sub>12</sub>		hepatoprotective activity
Quercetin-3-rhamnoside syn: Quercitrin [5280459]	C <sub>21</sub> H <sub>20</sub> O <sub>11</sub>		antioxidant antileishmanial agent

**Table 2:** Some flavonoids in safflower petals and their biological effect potential (Cont.)

Apigenin [5280443]	$C_{15}H_{10}O_5$		Inducing autophagy in leukemia cells antineoplastic agent
Kaempferol [5280863]	$C_{15}H_{10}O_6$		antioxidant anticancer antibacterial anti-aging
Naringenin [439246]	$C_{15}H_{12}O_5$		antineoplastic agent

<sup>1</sup> PubChem CID

Safflower petals have high medicinal value due to their chemical composition. Safflower flowers are an important medicinal ingredient used in prescriptions for cardiovascular health problems, cerebrovascular diseases affecting blood vessels and blood flow to the brain, and gynecological diseases (Zhou et al., 2009). Safflower petals contain an essential oil ranging from 1.2% to 1.6% (v/w, fresh weight basis). The essential oil obtained by water distillation is yellowish in color. Heptacosane, nonanoic acid, undecanoic acid, dec-2-en-1-ol, 2-pentadecanone, 1-hydroxy-3-propyl-5-(4-methyl-pentene)-2-methylbenzene, 2,5,5-trimethyl-3-propyl, tetrahydro 1-naphthol, benzaldehyde, caryophyllene oxide and lauric acid were identified as important components in the essential oil (Ziarati et al., 2012; Turgumbayeva et al., 2018).

### 3. USE OF SAFFLOWER FLOWER PETALS IN THE FOOD INDUSTRY

While traditionally safflower known as an oilseed crop, its flowers contain numerous important bioactive compounds with potential applications in various sectors. Natural pigments, phenolic compounds, and flavonoids derived from the flowers are used in the food industry as additives, colorants, and functional ingredients. Due to the potential health risks of synthetic additives, interest in natural and sustainable resources is increasing. In this context, safflower flowers offer a remarkable alternative (Barashovets and Popova, 2016; Adamska and Biernacka, 2021; Gomashe et al., 2021). The uses of safflower flower petals in the food industry are summarized in Table 3.

**3:** The uses and importance of safflower flower petals in the food industry

Uses	Function / Importance	Sample Areas	Application
Natural colorant	An alternative to synthetic dyes, a healthy and natural pigment source	Dessert, baked goods, yogurt, ice cream, drinks	
Functional food additive	It gives functional properties to foods with antioxidants and phenolic compounds.	Healthy functional drinks	snacks,
Herbal tea and beverage production	It is used in teas due to its aromatic properties and health benefits.	Safflower tea, blend teas	herbal
Spices and flavor additives	It gives light aroma and color to dishes.	Soups, sauces, meat dishes	
Saffron alternative	Low-cost natural colorant	Rice dishes (pilaw, etc.), desserts, dishes	traditional

Red and yellow pigments obtained from safflower flowers are used as dyes. Safflower petals contain approximately 30% yellow pigment. The proportion of red pigment (0.80%) is considerably lower than that of red pigment (Machewad et al. 2012). Both pigments are used as natural and safe colorants in foods.

Safflower flowers have been added to various food products (rice, sauces, soups, meat dishes, bread, pickles, etc.) as a spice to add flavor, aroma, and color since ancient times. Dried safflower flower petals are added directly to dishes. They are particularly used to impart a yellow color to rice pilaw (Ekin, 2005). It was first cultivated because of the yellow or yellow-red pigments in its petals. This pigment is used to add color and flavor to dishes, especially soups. Due to the high cost of saffron, safflower flowers are considered "false saffron" and offer a more economical color source (Erbaş and Mutlucan, 2023). Today, red pigments are widely used in the food industry to color foods. The red pigment, carthamin, has low water solubility. Therefore, it is particularly useful in chocolate production. Because the yellow pigment, carthamidin, is easily soluble in water, it is used in the production of fruit juices, juice mixes, energy drinks, soda, jelly, and candies. It adds color and enhances the visual quality of dairy products (yogurt, cheese, and milk desserts), baked goods, and confectionery (Popescu et al., 2022; Wu et al., 2024).

Safflower petals are also consumed as herbal tea. Safflower petal tea has a unique flavor, color, and aroma. It is used in herbal tea formulations as a color, flavor, and functional additive (Adamska and Biernacka, 2021).

Safflower petals are rich in phenolic compounds, flavonoids, and antioxidants. These compounds provide antioxidant properties to food, slowing lipid oxidation and extending shelf life. They neutralize free radicals and support the immune system, benefiting human health. Therefore, they are a natural resource suitable for the development of functional foods.

In recent years, consumer interest in natural, clean-label products has increased. Safflower flowers, with their high potential for domestic production, are an economical and environmentally friendly additive that can meet this demand. Thus, they provide added value to domestic agriculture and contribute to the natural-focused development of the food industry.

The importance of using safflower flowers as food goes beyond simply providing color and aroma. They are also noteworthy for their functional compounds, health benefits, and their ability to offer a natural and sustainable alternative. Therefore, safflower petals are an important crop resource that will continue to be utilized in the food industry as a natural additive, sweetener, and functional ingredient.

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# *Advances in Medicinal and Aromatic Plants: From Phytochemistry to Sustainable Use*

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# *Advances in Medicinal and Aromatic Plants: From Phytochemistry to Sustainable Use Chapter 1*

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**Changes in the Aroma Composition of Different  
Plant Parts in Stevia (*Stevia rebaudiana* Bertoni)  
Depending on Various Drying Methods**

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## INTRODUCTION

Stevia (*Stevia rebaudiana* Bertoni), a perennial plant belonging to the Asteraceae family, is native to South America and has been used as a natural sweetener for centuries, particularly in Paraguay and Brazil (Gasmalla et al., 2017). Today, this species, which is successfully cultivated in Asia, Europe, and North America, has gained increasing economic importance worldwide due to the steviol glycosides found in its leaves, which are approximately 200-300 times sweeter than sucrose, have low calories, and do not cause a glycemic response (Kinghorn, 2002; Hossain et al., 2017). The European Food Safety Authority (EFSA) and the World Health Organization have also reported that high-purity stevia extracts are safe for human consumption (Hossain et al., 2017). Stevia-specific bioactive compounds, obtained by drying stevia leaves, are used both in pharmacology and as a food additive (Lemus-Mondaca et al., 2012). Stevia leaves, which have high sweetness potential without being high in calories, have been used by the Guarani people for centuries. The steviol glycosides stevioside and rebaudioside A are 150–300 times sweeter than sucrose (Markovic et al., 2008). The main chemical components contained in stevia leaves are diterpene steviol glycosides, phenolic compounds, flavonoids, alkaloids, chlorophylls, fatty acids, and various volatile oil fractions. Extracts obtained from leaves are reported to be rich in flavonoids, alkaloids, chlorophylls, hydroxycinnamic acids (such as caffeic and chlorogenic acids), oligosaccharides, free sugars, amino acids, lipids, and minerals (Gasmalla et al., 2017).

Studies on the volatile compounds of stevia indicate that monoterpenes, sesquiterpenes, and oxygenated volatile compounds are predominant in the plant's leaves. The essential oils found in stevia leaves are particularly rich in mono- and sesquiterpenes, and components such as  $\alpha$ -cadinol, caryophyllene oxide,  $\beta$ -guaiene, and spathulenol have been identified as the main components of the plant's essential oil (Markovic et al., 2008). Stevia attracts attention not only for its sweetening

properties but also for its therapeutic potential. Various studies have demonstrated that leaf extracts exhibit antihypertensive, antihyperglycemic, anti-inflammatory, antimicrobial, and antitumor effects (Muanda et al., 2011). A significant portion of these biological activities stems from the phenolic compounds and volatile oil components in the leaves. Studies have shown that *Stevia* leaves contain important biologically active fractions that possess not only sweetening properties but also antioxidant and phytotoxic properties (Nuryandani et al., 2024).

The preservation of the chemical composition of plant material is directly related to the drying method, one of the most critical stages in post-harvest applications. Drying inhibits the growth of microorganisms and prevents certain biochemical events that could alter the organoleptic properties of plants (Hossain et al., 2010). Although drying is necessary to prevent microbial growth and ensure storage stability, high temperatures and long drying times can lead to losses in the color, aroma, volatile oil composition, and biologically active components of plants (Diaz-Maroto et al., 2002; Hossain et al., 2017). The selection of drying technology is important for preserving bioactive plants in stevia plants (Roohinejad et al., 2025). Therefore, the drying method chosen plays a decisive role in preserving volatile components. The aroma composition of the leaves is a decisive quality criterion in the industrial use of the stevia plant. However, the aroma profile is significantly affected by post-harvest processes, particularly drying methods. The drying process can alter the amount and composition of volatile compounds. While high temperatures can cause oxidation and loss of volatile components, low-temperature methods are more suitable for preserving aroma. Although there are numerous studies in the literature on the drying of medicinal and aromatic plants, studies examining how the volatile compound profiles of stevia leaves and different plant parts change depending on drying conditions are quite limited. Previous studies have determined that different drying processes significantly alter the volatile compound composition in stevia leaves, and it has been reported that microwave

drying in particular leads to a more compact and homogeneous structure (Gasmalla et al., 2017). Similarly, freeze-drying has been noted as the most effective approach for preserving both volatile compounds and phenolic content, while hot-air drying has been reported to cause losses of volatile compounds (Periche et al., 2016).

Drying methods not only affect the amount of volatile components but also cause oxidation or esterification in the components and can alter the qualitative structure of the volatile fraction due to exposure to various thermal reactions (Diaz-Maroto et al., 2003; Periche et al., 2016). In this context, the sensitivity of monoterpenes to temperature increases is a key factor explaining why freeze-drying provides higher protection. The volatile compound profile varies across different anatomical parts of the Stevia plant (leaves, stems, flowers, etc.). While the leaf portion offers a richer volatile profile, various studies have indicated that flowers may contain more diverse but lower concentrations of compounds (Marković et al., 2002; Muanda et al., 2011). Therefore, evaluating the effects of drying methods separately for different parts of the plant is a critical requirement for industrial applications.

In this context, understanding how the aroma components in different parts of the stevia plant (leaves, stems, flowers) change depending on the drying method used is of great importance both for maintaining product quality and for determining the most suitable technological processes for processing raw materials for functional food, cosmetic, and pharmaceutical applications. The sensitivity of Stevia's volatile components to processing conditions necessitates a detailed examination of the effects of drying methods on aroma stability. This study examines how the aroma component composition in different parts of the stevia plant changes under various drying methods, based on current scientific findings, and comprehensively evaluates the role of drying processes on aromatic quality. In this regard, the effects of drying techniques on both the qualitative (component diversity) and quantitative (component concentration) properties of volatile compounds have been

evaluated comparatively. Additionally, by determining the aromatic responses of different plant parts to drying processes, the aim is to establish a scientific basis for preserving or enhancing the targeted aroma profile in the industrial processing of the stevia plant.

## **1. MATERIALS VE METHODS**

### **1.1. Plant Material**

The research was conducted at the Experimental Field of the Faculty of Agriculture, Recep Tayyip Erdoğan University (Figure 1). Different plant parts of *Stevia rebaudiana* Bertoni were used as material in the research. After harvesting the stevia plant, the flowers, stems, and leaves were separated. The samples were then dried using two different drying techniques: in an oven and in a lyophilizer (Figure 2).



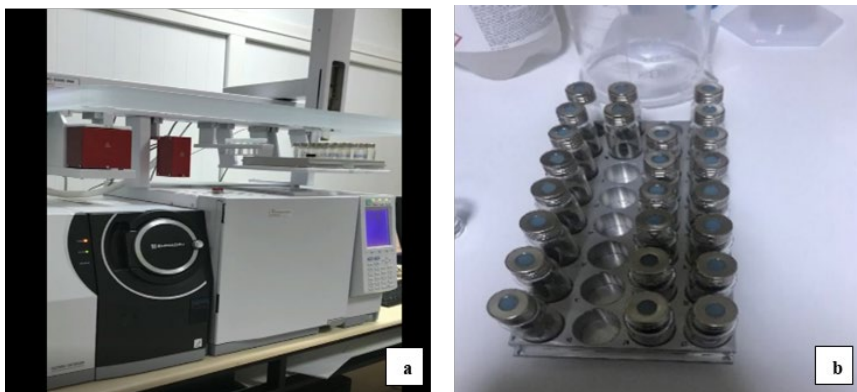
**Figure 1:** Images of the *Stevia rebaudiana* Bertoni plant cultivated in the test area



**Figure 2:** Images related to the drying of Stevia plant samples using a lyophilizer

### **1.2. Sample Preparation and Aroma Components Analysis**

Samples of the collected stevia plant parts were dried in an oven at 40°C for 24 hours, then ground and passed through 2 mm pore size laboratory sieves to prepare them for analysis. Analysis was performed using a GC-MS Device (Shimadzu, Japan) equipped with SPME (Figure 3.a). Ground plant samples weighing 0.2 g were then placed into a 10 mL vial and sealed with a silicone-rubber septum lid (Figure 3.b).



**Figure 3:** Images of the Gas Chromatography Mass Spectrometer (GCMS) device (a) and samples prepared for analysis (b)

Manufacturer guidelines were followed to precondition the fiber. The fiber was left in the headspace at equilibrium for one minute at room temperature. After sampling, the fiber was placed into the needle and then moved to the injection port of the GC-MS device. The column was a CP 5MS (30 m x 0.25 mm i.d., film thickness 0.25  $\mu\text{m}$ ). The oven temperature was set to isothermal at 220  $^{\circ}\text{C}$  for 20 minutes after being scheduled to rise from 40  $^{\circ}\text{C}$  to 240  $^{\circ}\text{C}$  at 2  $^{\circ}\text{C}/\text{min}$ . Helium was used as the carrier gas at a constant flow rate of 1 mL/min. The FFNSC 1.2 databases were used to determine the constituents of essential oils (Seyis et al., 2022).

## **2. RESULTS AND DISCUSSION**

Within the scope of this study, data on the amounts of aroma components in the leaves, flowers, and stems of the stevia plant, depending on different drying methods, are presented in Table 1. The effects of oven drying and freeze-drying (lyophilization) methods applied to samples from different parts of the Stevia (*Stevia rebaudiana* Bertoni) plant (flower, stem, and leaf) on aroma components were determined using a GC-MS device. The results obtained revealed that the volatile component composition showed significant differences depending on both the plant part and the drying method applied.

**Table 1.** Change in aroma components in stevia plant parts based on drying methods (%)

Chemical Group	R.T.*	R.I.**	Component	O.D. Flower	O.D. Stem	O.D. Leaf	L.D. Flower	L.D. Stem	L.D. Leaf
Monoterpene Hydrocarbons	9.151	933	$\alpha$ -Pinene				0,54		
	10.740	975	$\beta$ -Pinene	2,33		1,52	10,39	3,34	1,51
	<b>TOPLAM</b>			<b>2,33</b>		<b>1,52</b>	<b>10,93</b>	<b>3,34</b>	<b>1,51</b>
Oxygenated Monoterpenes	15.563	1100	Linalool	0,37		0,71	1,07		
	15.569	1212	Linalyl formate					0,57	1,19
	19.018	1198	$\alpha$ -Terpineol			0,18			
	19.029	1306	$\alpha$ -Terpinyl formate					0,41	0,36
	<b>TOPLAM</b>			<b>0,37</b>		<b>0,89</b>	<b>1,07</b>	<b>0,98</b>	<b>1,55</b>
Sesquiterpene Hydrocarbons	26.051	1382	$\beta$ -Bourbonene			0,79			0,64
	26.210	1375	$\alpha$ -Copaene			0,28			
	26.275	1390	$\beta$ -Elemene	0,56	0,53	0,27		0,47	
	26.894	1406	$\alpha$ -Gurjunene	0,28	0,62	0,18	0,33	1,57	
	27.263	1418	$\beta$ -Caryophyllene	28	8,59	26,39	32,6	26,05	23,2
	27.745	1432	$\alpha$ -trans-Bergamotene	7,33	3,12	7,93	8,99	10,01	7,79
	28.379	1452	(E)- $\beta$ -Farnesene	22	31,37	21,53	13,16	15,57	23,23
	29.238	1480	Germaacrene D	6,11	7,53	5		5,41	5,41
	29.259	1512	$\gamma$ -Cadinene	1,07	1,51	1,47	4,58	2,55	1,6
	30.570	1518	$\delta$ -Cadinene	1,92	2,48	2,23		2,22	2,11
29.417	1487	$\beta$ -Selinene		0,6					

**Table 1.** (Continued)

	29.656	1496	$\alpha$ -Zingiberene	1				
	29.755	1491	Viridiflorene	11,65	5,51			
	29.665	1387	7-epi-Sesquithujene			1,09	1,28	0,67
	29.770	1557	Germacrene B	5,84		7,55		
	29.761	1479	$\beta$ -Chamigrene		0,5		3,39	6,24
	29.844	1497	$\alpha$ -Muurolene	0,34	0,23			
<b>Sesquiterpene</b>	30.578	1423	$\beta$ -Cedrene			1,62		
<b>Hydrocarbons</b>	30.814	1459	$\alpha$ -Patchoulene	0,21				
	31.767	1508	$\beta$ -Bisabolene	2,56	2,24	4,11	5,79	5,61
	33.100	1458	Aromadendrene	9,67		0,55	0,52	0,76
	34.477	1480	$\alpha$ -Curcumene	0,3	0,81	0,48	0,94	0,93
	35.502	1459	Alloaromadendrene	0,45	0,33	0,79		
	34.584	1492	Valencene			0,52		
	<b>TOPLAM</b>			<b>80,67</b>	<b>76,11</b>	<b>75,82</b>	<b>76,04</b>	<b>75,49</b>
	31.755	1562	Nerolidol	3,11		3,26		
	32.115	1688	$\alpha$ -Bisabolol			0,3		0,58
<b>Oxygenated</b>	32.295	1576	Spathulenol	4,15	11,52	5,73	3,01	9,27
<b>Sesquiterpenes</b>	32.395	1610	Cedrol	0,7	1,34	1,09		
	32.480	1587	Caryophyllene oxide	2,25	4,54	2,64	5,93	5,59

**Table 1.** (Continued)

	33.220	1632	$\gamma$ -Eudesmol									0,8
	33.537	1688	(Z)- $\alpha$ -trans-Bergamotol	1,05	0,19	1,28	0,39					
	33.906	1624	epi- $\gamma$ -Eudesmol	0,27	1,97							2,38
<b>Oxygenated Sesquiterpenes</b>	34.173	1641	epi- $\alpha$ -Muuroiolol	1,01	1,74	1,23	0,96					
	34.462	1676	$\alpha$ -Santalol			0,62						
	35.700	1760	cis-Lanceol	0,46	5,52	0,29	0,58					1,7
	43.778	1594	Vinidiflorol	1,68	0,29	0,27	1,11					1,33
			<b>TOPLAM</b>	<b>14,68</b>	<b>22,57</b>	<b>18,31</b>	<b>8,99</b>	<b>19,53</b>	<b>16,51</b>			
	10.930	978	Vinyl amyl carbinol				0,66					0,32
	15.745	1104	Butyrate (2-methylbutyl)	0,35								
	15.751	1042	2-methylbutyl butyrate			1,06	0,76					0,75
	20.766	1243	3-methyl hexyl butanoate				0,29					
<b>Alcohols, Ketones, Aldehydes, Furans</b>	32.600	1573	Tridec-2(E)-enal			0,28						
	39.775	1841	Phytone	0,19			0,65					
	43.766	2016	Civetone	0,55		0,56						
	34.084	1647	Amyl cinnamaldehyde	0,88		0,76						
	45.763	1884	Cetyl alcohol				0,61					0,69
			<b>TOPLAM</b>	<b>1,97</b>	<b>0,28</b>	<b>2,38</b>	<b>2,97</b>	<b>0,33</b>	<b>1,76</b>			
<b>Alkanes, Alkenes, Alkynes, Arenes</b>	32.810	1500	Pentadecane			0,32						
	32.819	1600	Hexadecane									0,34
	45.748	2100	Heneicosane			0,17						
			<b>TOPLAM</b>			<b>0,32</b>	<b>0,17</b>	<b>0,34</b>	<b>100,01</b>	<b>100</b>	<b>100</b>	<b>99,9</b>
			<b>GENEL TOPLAM</b>	<b>100,02</b>	<b>100</b>	<b>99,09</b>	<b>100</b>	<b>100</b>	<b>100,01</b>	<b>100</b>	<b>100</b>	<b>99,9</b>

According to the analysis results, it was determined that the fraction richest in terms of total volatile compound number and compound concentration was generally found in leaf samples. The major compounds detected in all plant parts, although varying depending on the drying method, were  $\beta$ -Caryophyllene,  $\beta$ -Farnesene,  $\alpha$ -trans-Bergamotene, and Spathulenol. At the same time, among the chemical groups, sesquiterpene hydrocarbons have the highest amount, followed by oxygenated sesquiterpenes. An increase in component diversity has been observed in samples where freeze-drying was applied to the flower and stem parts of the plant. However, it has been observed that the concentrations of these components vary depending on the drying method. When drying flower and stem samples in an oven, the amounts of  $\beta$ -pinene,  $\alpha$ -trans-Bergamotene,  $\beta$ -Caryophyllene,  $\beta$ -Bisabolene, and Caryophyllene oxide decreased compared to lyophilization, while the amounts of compounds such as Spathulenol,  $\beta$ -Farnesene, and Germacrene D increased. In flower samples dried in an oven, the fact that most components are either found at very low levels or are completely lost indicates that the volatile components in flower tissue are more sensitive to heat. In leaf samples, when freeze-drying (lyophilization) was performed instead of drying in an oven, the amount of  $\beta$ -caryophyllene decreased, but the amounts of important compounds such as  $\beta$ -farnesene,  $\beta$ -chamigrene,  $\beta$ -bisabolene, spathulenol, and caryophyllene oxide increased. In general, freeze-drying has been found to be more effective in preserving both the quality and quantity of volatile aroma components.

The findings of this study reveal that the aroma components in *Stevia rebaudiana* vary significantly depending on both the plant part and the drying method applied. The results of the study are largely consistent with previous studies conducted on stevia and other medicinal aromatic plants.

The study determined that leaf samples have a richer and more concentrated volatile compound profile compared to flower and stem

tissues. This situation is consistent with studies in the literature that define Stevia leaves as the primary source of volatile oils and aroma components. Stevia leaves have been reported to be rich in monoterpenes, sesquiterpenes, and oxygenated terpenes; it has been emphasized that the leaf tissue is the most valuable part of the plant in terms of aromatic quality (Muanda et al., 2011). Tursun et al. (2021) reported that terpenes were detected in the highest concentration in the total volume of aromatic substances as a result of their research. Similarly, caryophyllene oxide, spathulenol, and various terpene derivatives have been reported to be predominant in stevia leaf essential oils (Markovic et al., 2008). In this study, the components identified in the leaves, such as  $\beta$ -pinene, spathulenol,  $\beta$ -farnesene, and  $\beta$ -caryophyllene, demonstrate that the aromatic character of stevia leaves is consistent with the literature.

Freeze-drying has attracted attention in the samples applied, as it preserves volatile components better and yields a more balanced profile in terms of component distribution. This finding is consistent with the results of a study conducted on Stevia leaves (Periche et al., 2016). In the study in question, it was stated that freeze-drying is the most effective method for preserving both phenolic compounds and aroma components, while hot air and oven drying cause losses of volatile compounds. Similarly, Gasmalla et al. (2017) reported that thermal processes such as microwave and hot air drying cause the breakdown of volatile compounds, while drying methods performed at low temperatures better preserve aromatic quality. Compared to other drying methods, vacuum drying has been reported to preserve vitamin (vitamin C and E), polyunsaturated fatty acid, and stevioside content. (Lemus-Mondaca et al., 2016). The reduction or complete loss of some volatile components in oven-dried samples can be explained by the thermal degradation and volatilization mechanisms frequently emphasized in the literature. It has been reported that exposure to high temperatures in aromatic plants leads to the oxidation of monoterpenes and ester hydrolysis, resulting in significant losses in the aroma profile (Diaz-Maroto et al., 2003). In this

study, the decrease in volatile compound diversity following oven drying, particularly in flower and stem tissues, indicates that these tissues are more sensitive to heat.

The limited diversity of volatile compounds in flower samples compared to leaves, but the preservation of some monoterpenes through freeze-drying, suggests that the aromatic potential of *Stevia* flowers is largely dependent on processing conditions. Zygadlo et al. (1997) reported that volatile oil content varies depending on the anatomical part of the plant and that flower tissues generally offer a more delicate volatile profile. The findings of this study support the aforementioned assessments specifically regarding *Stevia*. The low content of volatile compounds in the leaf tissue is consistent with the view in the literature that *Stevia* leaves are generally of secondary importance in terms of aromatic quality. However, the fact that some volatile components can be preserved even in the stem tissue after freeze-drying indicates that this method offers a potential advantage not only for leaves but also for other plant parts. This situation demonstrates that the drying method can modulate the aromatic responsiveness specific to plant tissue.

Overall, the results obtained in this study indicate that low temperature, reduction of oxidative stress, and preservation of cellular structure without damage are critically important for the preservation of aroma components in *Stevia* plants. These findings strongly parallel previous studies emphasizing that freeze-drying should be preferred for quality-focused applications in aromatic and medicinal plants. In conclusion, this study expands the limited information in the literature by detailing the aromatic responses of different parts of the *Stevia* plant to drying methods, providing important scientific contributions, particularly to industrial processing, quality standardization, and target aroma profile development studies.

### 3. CONCLUSIONS

The findings obtained in this study clearly demonstrate that the aroma composition of *Stevia rebaudiana* is strongly dependent on both the plant part and the drying method applied. Leaves are the richest part of the Stevia plant in terms of aroma components, while flowers have a more limited but selective aroma profile, and the stem tissue has been found to have a relatively low volatile component content. When drying methods are compared, freeze-drying (lyophilization) has emerged as a more successful method than oven drying in preserving Stevia's natural aroma profile.

It has been determined that heat-sensitive compounds, particularly monoterpenes and oxygenated terpenes, decompose or evaporate during oven drying; however, these compounds are largely preserved by freeze-drying. This situation demonstrates that minimizing low temperatures and oxidative stress is critical for aroma stability.

The results obtained indicate that the drying method is a strategic decision for processing stevia in a way that preserves its aromatic quality in the food, functional product, and cosmetics industries. Especially in applications where high aromatic quality is targeted, it is recommended to process the leaf tissue using the freeze-drying method. Furthermore, the fact that different parts of the plant have different aromatic reactivity indicates that the plant part and drying method must be evaluated together according to the intended industrial use. This study addresses drying-based changes in the aroma components of the stevia plant using a holistic approach, filling an important gap in the literature and providing a scientific basis for future quality optimization and process development studies.

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**Effects of Heavy Metal Stress on Secondary  
Metabolite Production in Pot Marigold  
(*Calendula officinalis* L.) Under  
in vitro Conditions**

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## INTRODUCTION

Medicinal and aromatic plants, which are widely used in the pharmaceutical, food, cosmetic, traditional medicine and spice industries, represent valuable natural resources. Throughout history, humans have explored different parts of plants (roots, leaves, flowers, and fruits), observed their healing effects on health and transmitted this knowledge across generations. It is reported that the earliest written records on herbal therapy date back to the Mesopotamian civilization around 5000 BC and that more than 250 herbal drugs were used for medicinal purposes during this period. Today, medicinal plants have considerable economic and therapeutic value in many fields, including herbal teas, pharmaceutical products, cosmetic formulations and spices (Demirezer, 2010).

Pot marigold (*Calendula officinalis* L.), commonly used today for medicinal and cosmetic purposes, is an annual herbaceous plant belonging to the genus *Calendula* within the Asteraceae family, typically reaching 30–60 cm in height (Ashwlayan et al., 2018) (Figure 1). Native to Central and Southern Europe, Western Asia and America, this species has long been cultivated and utilized in various regions. In Türkiye, it is known by several local names, including “aynısefa”, “tıbbi nergis” and “portakal nergisi” (Baytop, 1984; Deniz et al., 2010).



**Figure 1.** General appearance of *Calendula officinalis* L. (Pot marigold) plant  
Source: Original Photo, S. Beyza BUÇAN

The rich chemical composition of *Calendula officinalis* enables its use in many fields, including medicine, pharmacy, the food industry and the cosmetic industry. This plant contains numerous biologically active constituents and its main chemical groups include terpenoids, flavonoids, coumarins, quinones, essential oils, carotenoids and amino acids (Ashwlayan et al., 2018). In traditional and complementary medicine, these constituents are widely used for the management of various health conditions, such as the common cold, wound healing, febrile illnesses, insect bites, diaper rash, gingival disorders, hemorrhoids, stomach cramps, constipation and hypertension. In addition, extracts obtained from *C. officinalis* are incorporated into the formulations of numerous pharmaceutical and cosmetic products, including tea preparations, shampoos, tinctures, soaps, ointments, creams and lotions (Bruneton, 1995).

To survive and adapt to environmental conditions, plants synthesize not only primary metabolites but also secondary metabolites, which constitute the basis of their defense mechanisms. Secondary

metabolites are specialized compounds that do not directly participate in growth and development processes; instead, they play critical roles in plant adaptation to abiotic stress factors (e.g., drought, salinity, heavy metal contamination and UV radiation) as well as biotic challenges such as pathogen attacks and herbivory (Selmar, 2013; Isah, 2019). Under stress conditions, these compounds activate the plant defense system, confer protective effects against environmental pressures and enhance survival capacity (Selmar, 2013; Isah, 2019).

In recent years, increasing urbanization and industrialization have led to a marked rise in environmental pollution. Among these pollutants, heavy metals exert toxic effects in both terrestrial and aquatic ecosystems and have become major sources of biological stress for all living organisms, including plants. Heavy metal compounds such as silver nitrate ( $\text{AgNO}_3$ ) induce oxidative stress in plant tissues by triggering the production of reactive oxygen species (ROS); this disrupts physiological homeostasis and causes substantial changes in secondary metabolite biosynthesis (Al-Oubaidi et al., 2014).

In this context, investigating the secondary metabolite responses of *Calendula officinalis* L. under heavy metal-induced stress is of great importance for understanding the plant's biological resilience and metabolic adaptive capacity. Moreover, such studies provide new insights into how environmental stressors regulate the production of plant-derived bioactive constituents, thereby establishing a scientific basis for phytocosmetic, phytopharmaceutical and biotechnological applications (Yurteri and Buçan, 2025).

## **1. MATERIALS VE METHODS**

### **1.1. Plant Material**

Seeds of *Calendula officinalis* L. (pot marigold) were used as the plant material. The seeds were obtained from the Atatürk Horticultural Central Research Institute (Yalova, Türkiye).

The seeds were established in vitro under sterile conditions and aseptic plantlets obtained after germination were multiplied using tissue culture techniques to generate the experimental material. Stress treatments were then applied to the propagated in vitro plant material according to the experimental design and post-stress samples were prepared for analyses aimed at determining volatile and phenolic constituents. All stages of the tissue culture procedures and stress applications were carried out under sterile conditions in the Tissue Culture Laboratory and the tissue culture room of the Faculty of Agriculture, Recep Tayyip Erdoğan University.

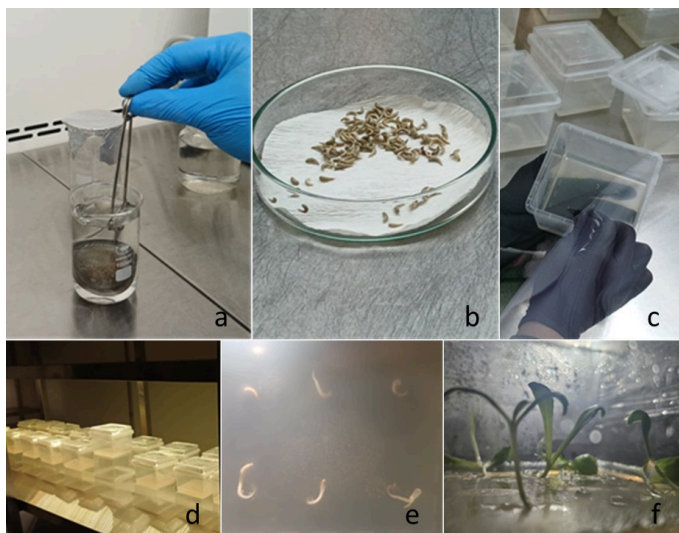
### **1.2. Method**

#### **1.2.1. Seed Sterilization, Germination and Stress Application**

The ridged and irregular surface structure of *Calendula officinalis* (pot marigold) seeds made the sterilization procedure challenging. Several approaches were tested to achieve effective surface sterilization; however, due to the characteristic morphological and structural properties of the seeds, certain difficulties were encountered in both the sterilization and germination stages. Therefore, the sterilization procedure was performed on seeds separated from the seed coat.

Based on preliminary trials, the following protocol yielded satisfactory results: seeds removed from the seed coat were treated with absolute ethanol for 40–50 s and then rinsed three times with sterile distilled water to eliminate residual alcohol. Subsequently, the seeds were immersed for 5 min in a diluted household bleach solution containing 10% sodium hypochlorite (NaOCl) prepared at a final

concentration of 6%. Finally, a last rinse was performed to ensure complete removal of chemical residues, after which the seeds were transferred onto a Petri dish lined with sterile drying paper and prepared for sowing (Figure 2a,b).



**Figure 2.** The procedural steps are as follows: seed sterilization (a,b), inoculation onto the culture medium (c), transfer to the tissue culture growth room (d) and the germination and growth period (e,f)

Following completion of surface sterilization, the seeds were inoculated onto MS (Murashige and Skoog, 1962) medium to promote germination and seedling development. The culture medium was prepared to contain 30 g/L sucrose and 0.7% (w/v) agar and the pH was adjusted to 5.7. All medium components were mixed prior to autoclaving and sterilized at 121 °C for 20 min.

After inoculation (Figure 2c) and transfer to the tissue culture room (Figure 2d), the seeds began to germinate within approximately 4–5 days (Figure 2e) and reached the seedling stage within 10 days (Figure 2f). Healthy seedlings were selected for the experimental treatments and

individually transferred to stress media containing different doses of silver nitrate ( $\text{AgNO}_3$ ).

Silver nitrate ( $\text{AgNO}_3$ ) was used to induce heavy metal stress. First, an  $\text{AgNO}_3$  stock solution was prepared at a concentration of 1 mg/mL. For this purpose, a pre-weighed amount of  $\text{AgNO}_3$  was placed in an Erlenmeyer flask, sterile distilled water ( $\text{dH}_2\text{O}$ ) was added and the compound was completely dissolved. The stock solution was then sterilized by passing it through a sterile syringe filter, transferred into sterile Falcon tubes and stored at  $-20\text{ }^\circ\text{C}$  for subsequent addition to the culture medium after autoclaving. Because  $\text{AgNO}_3$  is heat-sensitive, it was not added prior to autoclaving; instead, it was aseptically supplemented to the medium after sterilization.

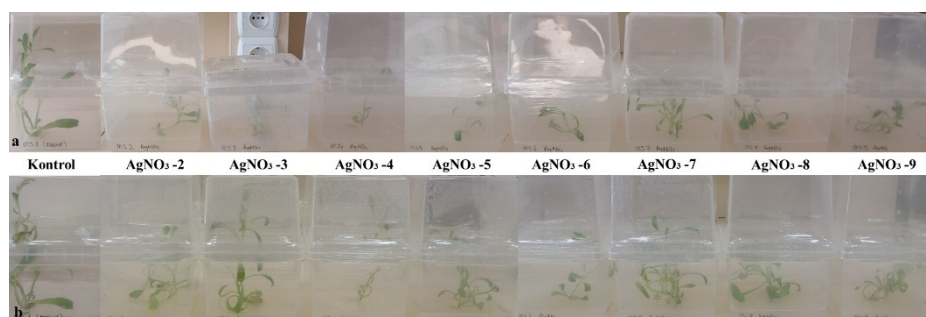
To establish heavy metal stress, media containing  $\text{AgNO}_3$  at different concentrations and a fixed 2,4-D concentration were prepared and an unstressed control group was also included (Table 1).

**Table 1.** Composition of control and heavy metal ( $\text{AgNO}_3$ ) stress media used in tissue culture

Medium	Culture Medium (4.4 gr)	Sakkaroz (gr/L)	$\text{AgNO}_3$ (mg/mL)	2,4-D (mg/mL)	Agar (%)	pH
(Kontrol)	MS	30	-	-	0,7	5,7
2	MS	30	0,1	0,5	0,7	5,7
3	MS	30	0,1	1	0,7	5,7
4	MS	30	0,5	0,5	0,7	5,7
5	MS	30	0,5	1	0,7	5,7
6	MS	30	1	0,5	0,7	5,7
7	MS	30	1	1	0,7	5,7
8	MS	30	1,5	0,5	0,7	5,7
9	MS	30	1,5	1	0,7	5,7

Each treatment was conducted with three replicates, with one plant used per replicate. After the seedlings were transferred to the stress media, they were exposed to stress in a growth chamber for

approximately 8 weeks (2 months). Throughout this period, plant growth rate, morphological characteristics and changes in leaf coloration were monitored regularly, while identical environmental conditions were maintained across all groups (Figure 3a,b).



**Figure 3.** Plants transferred to the control medium and heavy metal stress media containing different AgNO<sub>3</sub> concentrations (a) and their development over a 2-month period (b)

### 1.2.2. Preparation of Samples

At the end of the stress period (2 months), the plants were carefully removed from the culture medium and agar residues remaining on the root and stem surfaces were rinsed off with sterile distilled water. Each sample was coded and labeled, then dried in an oven at 37 °C until a constant weight was reached. The dried samples were ground in a sterile mortar to obtain a homogeneous powder and stored at +4 °C until analysis.

### 1.2.3. Chemical Analysis

To determine the volatile and phenolic constituents, qualitative analyses were performed using GC–MS (gas chromatography–mass spectrometry) and HPLC (high-performance liquid chromatography).

### 1.2.3.1. GC–MS Analysis

From the dried plant samples, 0.20 g was weighed into GC tubes and one drop of hexane was added to each tube. The samples were analyzed at the Plant Analysis Laboratory of the Faculty of Agriculture, Recep Tayyip Erdoğan University, using a Shimadzu GC–MS (GCMS-2010 Plus) system (Figure 4a,b). The determination of volatile constituents was performed by modifying the SPME (solid-phase microextraction) method described by Yurteri et al. (2021).



**Figure 4.** Preparation of samples for GC–MS analysis (a) and GC–MS measurement/analysis (b)

### 1.2.3.2. HPLC Analysis

Quantitative determination of phenolic and flavonoid constituents was performed by HPLC. For each sample, 0.1 g was placed into Falcon tubes, 10 mL of methanol was added and the mixture was extracted in a 40 °C water bath for 1 h. After extraction, the samples were centrifuged at 4000 rpm for 7 min and the supernatant was filtered through a 0.45 µm syringe filter and transferred into HPLC vials. The prepared vials were placed into the autosampler tray and the samples were then injected and analyzed (Figure 5a,b,c).

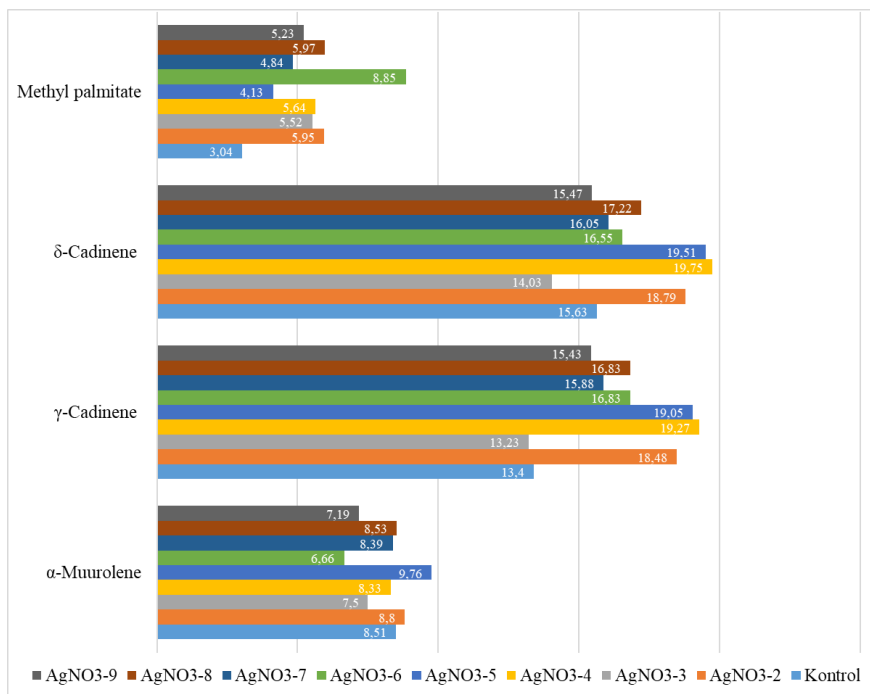


**Figure 5.** Following extraction for HPLC analysis, centrifugation of the samples (a), transfer into vials (b) and HPLC measurement/analysis (c)

## 2. RESULTS AND DISCUSSION

In this study, the effects of heavy metal stress induced by silver nitrate ( $\text{AgNO}_3$ ) at different concentrations on secondary metabolite production in *Calendula officinalis* L. (pot marigold) were investigated at both the volatile and phenolic levels. GC–MS and HPLC analyses demonstrated that increasing  $\text{AgNO}_3$  concentrations promoted the accumulation of phenolic metabolites in *C. officinalis* L., while causing pronounced fluctuations in the composition of volatile constituents.

As a result of gas chromatography–mass spectrometry (GC–MS) analysis, a total of 60 different volatile compounds were identified (Table 2). The predominant compounds were  $\delta$ -cadinene (19.75%),  $\gamma$ -cadinene (19.27%),  $\alpha$ -muurolene (9.76%) and methyl palmitate (8.85%) (Figure 6).



**Figure 6.** Major volatile compounds highlighted in the GC–MS analysis under heavy metal ( $\text{AgNO}_3$ ) stress

Depending on the increasing  $\text{AgNO}_3$  concentrations, dose-specific fluctuating changes were observed in the proportions of sesquiterpene compounds (particularly  $\delta$ -cadinene,  $\gamma$ -cadinene and  $\alpha$ -muurolene). In contrast, marked increases were recorded in compounds such as methyl palmitate, daucol and cedrol in parallel with the  $\text{AgNO}_3$  dose. These findings indicate that  $\text{AgNO}_3$ -induced stress substantially reshapes the volatile profile, particularly through changes in constituents such as methyl palmitate, cedrol and daucal.

**Table 2.** Volatile compounds detected in plants under heavy metal (AgNO<sub>3</sub>) stress using GC-MS analysis (%)

RI *	Compound Name	Control	AgNO <sub>3</sub> -2	AgNO <sub>3</sub> -3	AgNO <sub>3</sub> -4	AgNO <sub>3</sub> -5	AgNO <sub>3</sub> -6	AgNO <sub>3</sub> -7	AgNO <sub>3</sub> -8	AgNO <sub>3</sub> -9
1308	Myrtenyl acetate	—	—	0.29	—	—	—	—	—	—
1345	$\alpha$ -Cubebene	1.65	—	1.57	0.98	—	0.77	1.59	1.64	0.95
1371	$\alpha$ -Copaene	3.94	4.2	4.26	3.46	5.38	2.79	4.8	5.02	3.4
1393	Tetradecane	—	—	—	—	—	—	—	0.31	—
1405	$\alpha$ -Gurjunene	0.43	0.42	0.98	0.32	0.57	—	0.58	0.43	—
1415	$\beta$ -Caryophyllene	1.98	1.06	3.51	—	1.43	0.62	1.3	1.55	0.99
1419	Methyl eugenol	—	—	0.25	—	—	—	—	—	—
1442	$\beta$ -Cedrene	1.3	1.55	1.5	1.14	1.84	0.96	1.63	1.61	1.1
1446	$\alpha$ -Patchoulene	1.35	0.77	0.81	0.59	0.95	0.45	0.86	0.85	0.53
1449	$\alpha$ -Humulene	4.44	2.78	6.94	0.94	3.39	1.86	3.42	3.43	2.71
1451	(E)- $\beta$ -Farnesene	—	—	—	—	—	—	—	0.98	—
1457	$\alpha$ -Himachalene	0.68	—	—	—	—	—	—	0.95	—
1459	10 $\beta$ H-Cadina-1(6),4-diene	2.33	3.73	3.33	3.01	4.21	1.96	3.85	3.01	2.78
1472	$\alpha$ -Amorphene	5.92	6.33	6.84	5.48	7.01	4.06	6.05	6.33	4.95
1477	Germacrene D	—	0.73	2.59	—	1.14	2.89	1.01	0.83	0.5

**Table 2.** (Continued)

1482	Bicyclogermacrene	0.64	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
1483	Viridiflorene	—	—	1.57	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
1485	Lauryl alcohol	—	—	0.61	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0.44
1490	$\alpha$ -Zingiberene	1.92	—	2.4	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
1483	(E)- $\beta$ -Ionone	—	1.08	—	0.59	1.58	0.95	1.49	0.93	0.3	—	—	—	—	—	—	—	—	—
1496	$\alpha$ -Muurolene	8.51	8.8	7.5	8.33	9.76	6.66	8.39	8.53	7.19	—	—	—	—	—	—	—	—	—
1504	$\beta$ -Bisabolene	0.55	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0.56
1511	$\gamma$ -Cadinene	13.4	18.48	13.23	19.27	19.05	16.83	15.88	16.83	15.43	—	—	—	—	—	—	—	—	—
1521	$\delta$ -Cadinene	15.63	18.79	14.03	19.75	19.51	16.55	16.05	17.22	15.47	—	—	—	—	—	—	—	—	—
1526	Tridecyl aldehyde	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0.23
1556	(E)-Tridec-2-enal	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0.42
1557	Spathulenol	—	0.26	—	—	0.46	—	—	—	—	—	—	—	—	—	—	—	—	—
1563	$\gamma$ -Undecalactone	—	0.24	0.26	—	0.34	0.41	0.35	—	—	—	—	—	—	—	—	—	—	—
1565	Germacrene B	—	—	0.45	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
1550	Nerolidol	0.3	0.22	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	0.33
1591	Hexadecane	0.94	0.67	0.76	1.07	1.14	1.89	1.72	2	1.46	—	—	—	—	—	—	—	—	—

**Table 2.** (Continued)

1531	Citronellyl butyrate	—	—	—	0.49	—	—	0.62	—	—	—
1606	Cedrol	3.48	3.13	3	5.81	4.13	5.19	3.52	2.26	5.85	—
1634	Daucol	4.95	3.84	4.32	6.96	5.47	—	3.64	2.12	7.92	—
1643	Cedryl methyl ether	—	—	—	—	—	—	—	—	0.43	—
1646	Hedione	—	0.25	—	—	0.43	—	0.41	—	—	—
1647	(E)-Citronellyl tiglate	—	—	—	—	—	0.73	—	—	0.22	—
1649	$\beta$ -Eudesmol	—	0.37	0.4	0.45	0.53	—	—	—	—	—
1649	$\alpha$ -Bisabolol oxide B	—	—	—	—	—	0.75	—	—	—	—
1637	$\alpha$ -epi-Muurolol	3.32	2.62	2.67	2.25	—	2.01	3.06	1.84	1.65	—
1655	Methyl dihydrojasmonate	—	—	—	0.52	—	—	—	—	—	—
1639	Octyl furan-2-carboxylate	—	0.27	—	1.26	1.52	1.65	1.36	1.27	1.27	—
1684	Myristyl alcohol	—	—	—	—	—	0.49	—	—	—	—
1691	Heptadecane	0.81	0.46	0.34	0.56	0.41	1.56	0.44	0.46	—	—
1736	Cis,cis-Farnesol	—	—	—	—	—	7.33	4.1	—	—	—
1750	Farnesal	0.35	3.29	1.73	4.09	3.39	—	0.51	2.18	1.49	—
1762	Octyl caprylate	—	—	—	—	0.34	—	—	—	0.45	—

**Table 2.** (Continued)

1790	Octadecane	0.79	0.22	—	—	—	0.76	—	—	0.49
1817	Isopropyl myristate	2.09	0.46	0.62	0.52	0.32	0.61	0.38	0.77	0.81
1830	Neophytadiene	10.71	5.76	6.54	4.39	—	6.75	5.73	6.73	14.42
1837	Phytone	1.07	1.47	0.9	1.68	1.16	2.07	1.88	1.54	1.54
1848	6-Acetyl-1,1,2,4,4,7-hexamethyl-tetralin	1.88	—	—	—	0.38	—	0.39	1.06	—
1889	Nonadecane	0.38	—	—	—	—	0.99	—	—	—
1916	Methyl palmitate	3.04	5.95	5.52	5.64	4.13	8.85	4.84	5.97	5.23
1956	Isophytol	—	—	—	—	—	—	—	—	0.25
1985	Ethyl palmitate	—	0.22	—	—	—	—	—	—	—
2014	Isopropyl palmitate	0.4	—	—	—	—	—	—	—	—
2257	Tributyl citrate acetate	—	—	—	0.47	—	—	—	—	—
2271	5 $\alpha$ -Androst-16-en-3-one	—	—	0.28	—	—	—	—	—	—
2386	Tetracosane	0.82	—	—	—	—	—	—	—	—

\* Kovats Retention Index (RI)

High-performance liquid chromatography (HPLC) analyses were conducted to evaluate how increasing AgNO<sub>3</sub> stress intensity (from AgNO<sub>3</sub>-1 to AgNO<sub>3</sub>-9) affected the accumulation of phenolic constituents. In this context, five major phenolic compounds (gallic acid, catechin, chlorogenic acid, caffeic acid and quercetin) were detected (Table 3). Marked changes in the levels of these compounds were observed depending on the AgNO<sub>3</sub> treatment.

In the control group, chlorogenic acid (51.3 mg/g) and catechin (46.4 mg/g) were identified as the predominant compounds, whereas caffeic acid was detected at a moderate level (6.9 mg/g). Quercetin was present at a low level (0.4 mg/g) and gallic acid remained at trace level (Tr.).

Under AgNO<sub>3</sub> stress, the amounts of chlorogenic acid and catechin were generally suppressed compared with the control. Chlorogenic acid decreased to 0.6 to 17 mg/g in the stress groups and did not approach the control level at any stress stage. Similarly, catechin concentrations ranged from Tr. to 16.3 mg/g and remained markedly below the control, not exceeding approximately one-third of the control level even at the highest values. These findings indicate that increasing AgNO<sub>3</sub> stress severity suppresses the accumulation of chlorogenic acid and catechin and suggest that phenolic metabolism may have been redirected toward the formation of alternative phenolic products.

In contrast, quercetin exhibited the most pronounced stress-associated response. Compared with the control, quercetin levels

increased substantially under AgNO<sub>3</sub> treatments, rising to 1.8 to 37.1 mg/g, with the highest value recorded at the highest stress level (AgNO<sub>3</sub>-9: 37.1 mg/g). Caffeic acid displayed a more variable pattern; although it generally remained below the control (0.2 to 4.1 mg/g), partial recoveries were observed at some stages (AgNO<sub>3</sub>-8: 3.9 mg/g). Another notable compound, gallic catechin, was present at trace level in the control but became quantitatively detectable at certain stress intensities (AgNO<sub>3</sub>-5: 7.4 mg/g).

**Table 3.** Components detected by HPLC analysis in plants exposed to heavy metal (AgNO<sub>3</sub>) stress (mg/g)

Compound Name	Galocatechine	Chlorogenic Acid	Catechine	Caffeic Acid	Quercitrin
Kontrol	Tr.	51.3	46.4	6.9	0.4
AgNO <sub>3</sub> -2	5.5	0.6	Tr.	1.7	2.6
AgNO <sub>3</sub> -3	1.3	10.7	16.3	4.1	29.9
AgNO <sub>3</sub> -4	Tr.	Tr.	Tr.	0.4	Tr.
AgNO <sub>3</sub> -5	7.4	Tr.	13.6	2	1.8
AgNO <sub>3</sub> -6	Tr.	Tr.	Tr.	0.2	15.3
AgNO <sub>3</sub> -7	1.8	17	3.7	3.4	20
AgNO <sub>3</sub> -8	2.7	4.6	4.9	3.9	25.4
AgNO <sub>3</sub> -9	2.4	12	5.4	2.7	37.1

\* Tr.: traces

When these results are considered collectively, AgNO<sub>3</sub>-induced heavy metal stress applied under in vitro conditions affected phenolic

accumulation and altered the quantitative distribution of the phenolic profile. As stress intensity increased, certain compounds (chlorogenic acid and catechin) were markedly suppressed, whereas a strong increase was observed in flavonol derivatives such as quercitrin.

In our study, the prominence of  $\delta$ -cadinene (19.75%) and  $\gamma$ -cadinene (19.27%) among the major constituents of the volatile profile indicates that sesquiterpenes may constitute core components of the volatile composition in *Calendula officinalis*. This finding is consistent at the level of predominant compounds with the report of Gazim et al. (2008), who identified  $\delta$ -cadinene (22.5%, 22.1% and 18.4%, respectively) and  $\gamma$ -cadinene (8.9%, 25.4% and 24.9%, respectively) as major constituents in the volatile profiles of flower samples obtained using different extraction approaches (SD, HS-SPME and HS-CF). However, a direct quantitative match should not be expected because that study was based on flower material, whereas our study analyzed samples obtained from plants grown under tissue culture conditions. Plant organ and developmental stage, the influence of in vitro conditions on metabolic fluxes and the extraction approach are key factors that can substantially affect the relative abundances of major constituents. Nevertheless, the recurrence of  $\delta$ -cadinene and  $\gamma$ -cadinene as prominent constituents in both studies suggests that these compounds may represent a reproducible chemical backbone of the *C. officinalis* volatile profile across different materials and conditions.

Okoh et al. (2008) reported that sesquiterpenoids can be predominant in essential oils obtained by hydrodistillation from fresh material of *C. officinalis* leaves and flowers, whereas the composition may change markedly after drying. In that study,  $\delta$ -cadinene was reported among the major constituents in both fresh and dried leaves (11.08% and 9.0%) as well as in flowers (13.1%) (Okoh et al., 2008). In our study, samples were derived from tissue culture conditions and volatile constituents were determined using an SPME-based approach; therefore, it is plausible that the relative abundances do not directly coincide with those reported in hydrodistillation-based studies.

Sabir et al. (2015) reported that hot-water extracts obtained from the flowers and leaves of *C. officinalis* exhibit strong antioxidant potential and that luteolin, caffeic acid, apigenin, rosmarinic acid and kaempferol were shared major constituents in the HPLC profiles of both flowers and leaves (Sabir et al., 2015). In our study, the detection of caffeic acid supports the view that this compound may be among the recurring phenolics of the species. The lack of prominence of other major flavonoids in our profile may be attributed to differences in extraction conditions, the type of plant material and the directing effects of stress conditions on metabolic flux.

Overall, our findings demonstrate that heavy metal stress induced by  $\text{AgNO}_3$  under in vitro conditions reorganized the secondary metabolite profile of *Calendula officinalis* at both the volatile (GC-MS) and phenolic (HPLC) levels. Most previous studies have been conducted

on plant material grown under natural or field conditions and have often focused on different organs such as flowers and leaves. In contrast, our study was performed using samples derived from tissue culture conditions, which limits direct quantitative comparisons. Moreover, the use of an SPME-based approach for volatile analysis and methanolic extraction followed by HPLC for phenolic profiling should be considered, as differences in solvents and analytical methods can affect the relative abundances of major constituents when compared with studies employing alternative methodologies.

It has been emphasized that changes in chemical profiles are associated with many parameters, including abiotic and biotic factors as well as post-harvest processing, extraction methods and storage conditions (Fokou et al., 2020). Therefore, although our results show similarities with the literature at the level of predominant constituents, a one-to-one correspondence in relative percentages is not expected due to the originality of the experimental setup and methodological differences. Accordingly, our findings contribute to the literature by demonstrating that heavy metal stress can modify metabolite distribution in *C. officinalis* under in vitro conditions. However, because studies using an identical experimental design are limited, the observed agreement is better interpreted at the profile level and in terms of dominant constituents, rather than as a strict quantitative match.

### 3. CONCLUSION

Increasing urbanization and industrialization in recent decades have contributed to a substantial rise in environmental pollution. Among these pollutants, heavy metals exert toxic effects in both terrestrial and aquatic ecosystems and have become major sources of biological stress for all living organisms, including plants. Heavy metal compounds such as silver nitrate ( $\text{AgNO}_3$ ) induce oxidative stress in plant tissues by triggering the production of reactive oxygen species (ROS). This process disrupts physiological homeostasis and can lead to significant alterations in secondary metabolite biosynthesis (Al-Oubaidi et al., 2014).

In the present study, the effects of  $\text{AgNO}_3$  application on the secondary metabolite profile of *Calendula officinalis* L. were evaluated using both GC–MS and HPLC analyses. The results indicate that silver ions markedly reprogram metabolic activity in the plant. In the GC–MS analyses, dose-dependent increases and decreases were observed in the relative abundances of certain constituents, including  $\delta$ -cadinene,  $\gamma$ -cadinene and  $\alpha$ -muurolene. In contrast, the levels of methyl palmitate, cedrol and daucol increased under  $\text{AgNO}_3$  treatments.

Consistently, the HPLC results revealed pronounced differences in phenolic constituents. With increasing stress intensity, some compounds (chlorogenic acid and catechin) were strongly suppressed, whereas a pronounced increase in flavonol derivatives such as quercitrin was observed. This pattern suggests that, under stress conditions, phenolic metabolism may shift toward compounds associated with protective and

antioxidant responses. Accordingly, AgNO<sub>3</sub> application appears to influence not only the overall level of the phenolic profile in *C. officinalis* L. but also its composition. These findings indicate that stress intensity should be considered a key variable shaping the balance among phenolic constituents.

Overall, the results suggest that *Calendula officinalis* L. develops biochemical adaptation to AgNO<sub>3</sub>-induced heavy metal stress and that this adaptation is accompanied by a reorganization of both volatile and phenolic constituents.

In conclusion, the data demonstrate that environmental stress factors play an important role in directing plant secondary metabolite production. Understanding stress-driven metabolic alterations provides important scientific contributions by elucidating biological resilience mechanisms in plants and by supporting the identification of new active constituents that may be utilized in phytocosmetic and phytopharmaceutical applications.

#### **4. ACKNOWLEDGMENTS**

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## **A Look at the Importance and Future of Micro Green Medicinal Plants**

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## **INTRODUCTION**

Medicinal and aromatic plants are considered a strategic biological resource due to their multifaceted functions throughout human history, including nutrition, providing scent and flavor, protection and especially traditional medicinal use (Acibuca and Budak, 2018; Temel et al., 2018).

Today, medicinal and aromatic plants are important not only in the field of health, but also in terms of agricultural production, rural development and the export economy, as they are the raw material for a wide range of products such as phytotherapy, functional foods, food supplements, herbal teas, cosmetics, perfumery, natural preservatives and aromatherapy (Marshall, 2011; TAGEM, 2021). This importance is further highlighted by the conservation of genetic resources in economically valuable species and their evaluation based on breeding/biotechnology; the role of genetic diversity in the production–quality–sustainability axis is emphasized (Yurteri et al., 2022).

Natural products are attracting increasing global interest due to safety concerns regarding synthetic inputs and the growing emphasis on a “preventive health” approach, which together are driving higher demand for products derived from medicinal and aromatic plants (Temel et al., 2018). This trend has also provided a basis for addressing traditional and complementary medicine practices within a more systematic framework. The World Health Organization’s traditional medicine strategy aims to strengthen the integration of medicinal and aromatic-plant-based products into health systems by encouraging countries to develop policies on product safety, quality, efficacy and sustainable sourcing (WHO, 2013). Within this context, medicinal and aromatic plants stand out as a strategic resource in the “functional food” paradigm, owing to their phytochemical diversity, potential biological activities and their association with innovative production systems.

In recent years, microgreens edible forms of plants harvested at an early developmental stage (at the cotyledon stage with the emergence of the first true leaves) have rapidly become a growing research area in both food science and phytotherapy/phytopharmaceutical studies, owing to their high nutrient density, sensory appeal and compatibility with controlled-environment agriculture (CEA). Because the chemical profile of microgreens can be strongly modulated by variables such as species/cultivar, light spectrum, nutrient solution, stress/elicitor applications and harvest timing, they also open a biotechnological pathway for the “target-compound-oriented” production of medicinal and aromatic plants (Seth et al., 2025).

The health effects of medicinal and aromatic plants largely stem from the structural and functional diversity of the secondary metabolites they accumulate. These compounds can generally be classified into phenolics (phenolic acids and flavonoids), terpenoids/essential oil constituents, alkaloids, tannins, coumarins and other specific metabolite groups. The biological actions of these phytochemicals may arise through the simultaneous contribution of multiple mechanisms, including the reduction of oxidative stress and support of antioxidant defenses, regulation of inflammatory responses, inhibition of microbial growth and modulation of cellular signaling pathways (El-Saadony et al., 2025). In particular, flavonoids constitute a broad class of bioactive compounds characterized by prominent antioxidant and anti-inflammatory properties and they are frequently discussed in research areas related to cardiometabolic processes and cancer biology (Roy et al., 2022).

This framework suggests that secondary-metabolite-rich medicinal and aromatic species may be valorized not only as mature plant material but also at early developmental stages. Indeed, microgreens are edible

seedlings of vegetable and aromatic/medicinal species, typically harvested within 7–21 days and are increasingly highlighted within the functional food paradigm due to their nutrient density and sensory attributes. In this context, the microgreen form of medicinal and aromatic plants that are rich in phenolics and essential oil constituents may offer an innovative avenue for target-compound-oriented production, quality standardization and value-added product development, supported by their short production cycle (Seth et al., 2025; Turner et al., 2020).

However, because medicinal and aromatic plants often exhibit a multi-component activity potential rather than a “single-compound” effect, clinically meaningful biological activity cannot be explained solely by the concentration of constituents. The manifestation of bioactivity is strongly shaped by factors such as bioaccessibility/bioavailability, the food matrix, post-digestive transformations (metabolism) and consumption patterns. Therefore, in evaluating microgreens, considering an integrated “composition + bioaccessibility” approach can support a more realistic interpretation of potential biological effects. For instance, a study on Brassicaceae microgreens assessed both the content of minerals and antioxidant bioactive compounds and their *in vitro* post-digestion bioaccessible fractions in broccoli, curly kale, red mustard and radish microgreens from different genotypes; the findings showed that interpreting “potential biological effects” based on the fraction generated after digestion can be strengthened (de la Fuente et al., 2019).

## **1. A LOOK AT THE UNIQUE VALUE OF MICROGREENS PRODUCTION IN TERMS OF MEDICINAL AND AROMATIC PLANTS**

Medicinal and aromatic plants are regarded as plant-based resources with multi-component bioactivity potential in health-related processes, largely because they are rich in secondary metabolite groups such as phenolics, terpenoids/essential oil constituents and alkaloids. Microgreens are edible seedlings harvested shortly after germination and have gained prominence within the functional food paradigm due to their intense sensory attributes combined with fresh consumption and their short production cycle (Turner et al., 2020; Seth et al., 2025).

The distinctive value of medicinal and aromatic-plant microgreens lies not merely in claims of “high content,” but in their ability to serve as a platform for generating reproducible quality profiles under controlled production conditions. By managing parameters such as light, temperature, growing substrate and nutrient regimes, phytochemical accumulation can be directed, enabling target-compound-oriented standardization and product development. In particular, positioning medicinal and aromatic species as microgreens often referred to as “micro herbs” (freshly consumed microgreen forms of medicinal and aromatic plants) offers an innovative approach that integrates sensory quality with phytochemical richness within a single product (Figure 1, Table 1) (Turner et al., 2020; Falcinelli and Benincasa, 2025). Accordingly, it has been shown that species/genotype selection in aromatic microgreens can markedly alter nutritional and phytochemical profiles (Giordano et al., 2022). Nevertheless, to interpret potential biological effects more realistically, it is recommended that compositional data be evaluated together with post-digestion bioaccessible fractions; bioaccessibility-based assessments in



# Advances in Medicinal and Aromatic Plants: From Phytochemistry to Sustainable Use Chapter 4

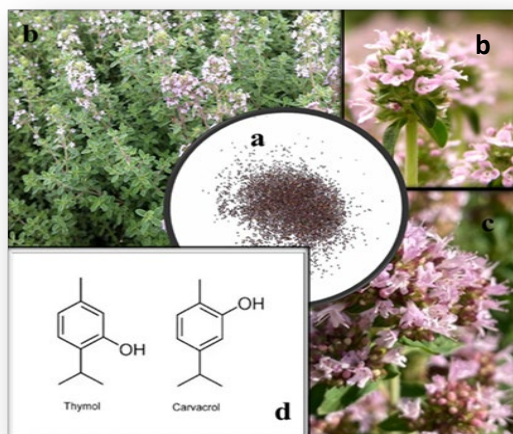
**Table 1:** Medical and aromatic plants evaluated for their micro-herb potential: main active ingredient groups and usage perspective

Medical and aromatic plant species	Main active ingredients	Micro green potential	Kaynak
<i>Origanum spp.</i>	Essential oil monoterpenes: carvacrol, thymol	Strong candidate for “functional fresh product/micro-herbs” lines due to its aromatic profile and essential oil constituents	(Leyva-López et al., 2017).
<i>Salvia officinalis L.</i>	Phenolic diterpenes and phenolic acids: camosic acid, carnosol, rosmarinic acid	Suitable model species for standardization via controlled-environment management of the phenolic profile	(Hrebień-Filińska et al., 2025).
<i>Sideritis spp.</i>	Rich secondary metabolite profile, especially flavonoids and phenylpropanoid derivatives	Controlled microgreen production could reduce dependence on wild harvesting of endemic taxa; suitable for functional/phytotherapeutic input	( Anonymous., 2025 ; Żyżelewicz et al., 2020).
<i>Foeniculum vulgare Mill.</i>	Major essential oil constituent: trans-anethole; also estragole, fenchone	Fast growth + distinctive aroma → strong candidate as a culinary microgreen	(Badgujar et al., 2014).
<i>Ocimum basilicum L.</i>	Polyphenols (flavonoids, phenolic acids) and related antioxidant compounds	Phenolic profile (particularly rosmarinic acid) can be directed via light spectrum/LED ratios and environmental settings, enabling target-compound-oriented production	(Lobiuc et al., 2017).
<b>Brassicaceae mikro yeşilleri</b>	Glucosinolates → isothiocyanates + polyphenols, vitamin C	Among microgreens, one of the strongest evidence bases; a “reference” product line	(de la Fuente et al., 2019).
<i>Petroselinum crispum</i>	Flavonoids: apigenin and luteolin derivatives (glycosides)	Phenolic/flavonoid profile can be modulated at the microgreen stage; suitable for a fresh functional product line	(Subaş et al., 2024).
<i>Coriandrum sativum</i>	Polyphenols (flavonoids, phenolic acids) and related antioxidant compounds	Promising antioxidant/phytochemical density in microgreen form; functional fresh product and extract input	(Mahleyuddin et al., 2021).

## 2. MICRO GREEN MEDICINAL PLANTS: PROMINENT SPECIES AND ACTIVE INGREDIENTS

### 2.1. *Origanum spp.*

Turkey's flora exhibits high diversity in terms of species and taxa within the genus *Origanum*; this diversity not only makes the country an important center for the genus's genetic resources, but also positions it among the leading countries in the production and valorization of commercial "oregano/thyme" (Gürbüz et al., 2011). In this context, *Origanum* species have a wide range of applications in medicinal and pharmaceutical research, particularly due to the bioactive constituents present in their essential oil fraction (Figure 2., Table 1.).



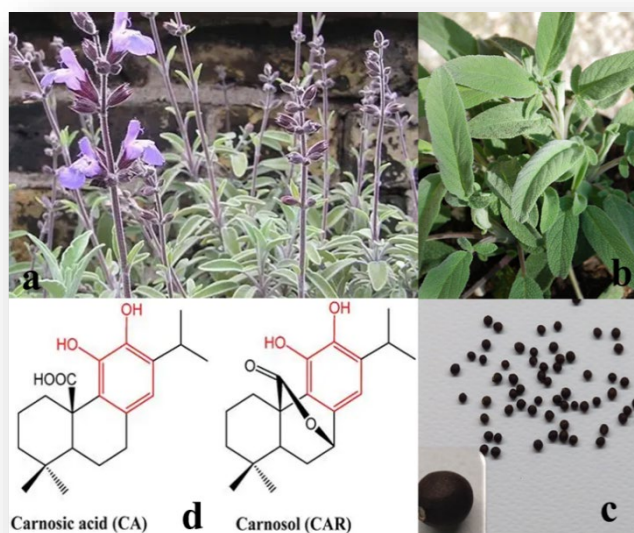
**Figure 2:** *Origanum spp.* general appearance (b), seed (a), flower (c), chemical formula of active ingredient (d)

Among the major components of the essential oil, monoterpene phenols such as carvacrol and thymol are strongly emphasized in the literature for their biological activities, which are associated with antimicrobial effects, limitation of oxidative stress, and regulation of inflammatory processes. (Bakkali et al., 2008; Marchese et al., 2016).

Within the context of microgreen production, *Origanum* species represent a noteworthy candidate group under the “micro herbs” concept, which enables aromatic/medicinal plants to be utilized as fresh products at early developmental stages (Falcinelli et al., 2025). During this phase, rapid growth and intense metabolic activity may create a critical production window in which phenolic compounds and aroma constituents associated with essential oils can be more effectively targeted. Indeed, it has been reported in *Origanum vulgare* that phenological stages lead to significant changes in essential oil yield and composition, indicating that the biosynthesis of aroma-related constituents is sensitive to developmental progression (Chauhan et al., 2013). This overall biosynthetic plasticity suggests that *Origanum* microgreens can be rationally explored for the development of functional fresh products that combine strong sensory appeal with phytochemical value. Moreover, since microgreens have been shown to provide variable yet often rich profiles of phytonutrients as “next-generation” fresh produce, the scientific basis for evaluating *Origanum* microgreens at the intersection of functional foods and phytotherapy is further strengthened (Xiao et al., 2012).

## 2.2. *Salvia officinalis* L.

Medicinal sage (*Salvia officinalis* L.) is an important medicinal and aromatic species that is cultivated in different regions of Türkiye and has a broad range of traditional uses (Elmas, 2021). In terms of its phytochemical profile, it has been reported to exhibit a composition rich in phenolic diterpenes (carnosic acid and carnosol) as well as phenolic acids (rosmarinic acid) (Lu and Foo, 2002). These compounds have been associated with multiple biological activities most notably strong antioxidant capacity, along with modulation of inflammatory responses and neuroprotective effects which are also emphasized in recent pharmacological evaluations of *S. Officinalis* (Figure 3., Table 1.) (Ghorbani and Esmailzadeh, 2017).

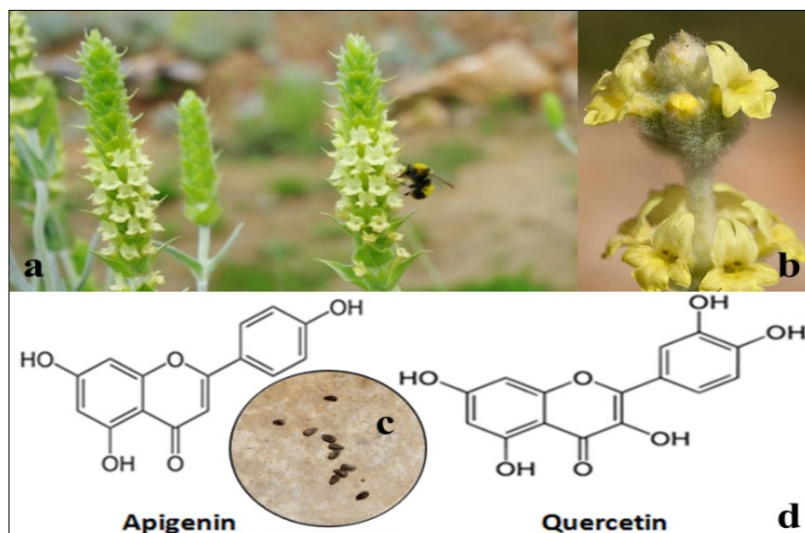


**Figure 3:** *Salvia officinalis* general appearance (a), leaf (b), seed (c) and active ingredient chemical formula (d)

In microgreen production systems, the controlled management of environmental factors particularly through light spectrum manipulation and light-elicitation applications can significantly influence the biosynthesis and accumulation dynamics of phenolic compounds (Teliban et al., 2025). In parallel, studies on in vitro shoot cultures of *Salvia* species have reported that LED spectral composition can markedly alter both polyphenol accumulation and the expression levels of genes involved in the biosynthetic pathways associated with these metabolites (Grzegorzczak-Karolak et al., 2025). Taken together, these findings suggest a strong scientific rationale for spectrum-based cultivation strategies in sage microgreens, supporting the optimization of phenolic acid profiles and the development of production protocols aimed at standardizing target bioactive compounds.

### **2.3. *Sideritis* spp.**

Türkiye's flora displays remarkable diversity in terms of *Sideritis* species and taxa; the fact that many taxa are endemic further underscores the genus as an important resource both for genetic diversity and for medicinal aromatic use. In Türkiye, infusions of *Sideritis* species are widely consumed in folk medicine, and traditional use reports indicate their use for a range of complaints, particularly upper respiratory symptoms such as the common cold, as well as gastrointestinal disorders (Figure 4., Table 1.) (Güvenc et al., 2005). From a phytochemical perspective, the major compound groups highlighted in the biological effects of the *Sideritis* genus include flavonoids, phenolic glycosides, and essential oil constituents and it is emphasized that species-specific differences in these profiles are associated with pharmacological activities (González-Burgos et al., 2011).



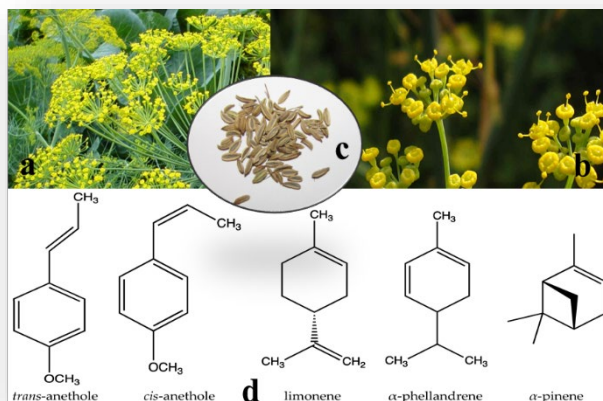
**Figure 4:** General appearance of *Sideritis spp.* (a), flower (b), seed (c) and chemical formula of the active ingredient (d)

The presence of numerous endemic taxa within the genus *Sideritis* in Türkiye increases the strategic importance of this group in terms of both biodiversity and medicinal aromatic use. However, harvesting mountain tea species particularly during the flowering period and before seed set may limit the regenerative capacity of natural populations, raising concerns about a potential decline risk for certain taxa (Baydar and Baydar, 2025). To alleviate this pressure, controlled production approaches that reduce dependence on wild collection represent a complementary solution pathway. Microgreen production, with its short cultivation cycle, traceable process management in closed or semi-closed systems and “functional product” potential suitable for fresh consumption, may enable the more sustainable utilization of high-value medicinal and aromatic plants such as *Sideritis* (Turner et al., 2020; Seth et al., 2025). In addition, European Union–level assessments regarding

Sideritis herb are important because they define the scope of traditional uses and the safety framework for this plant, thereby providing a scientific basis for standardized product development processes (Anonymous., 2025).

#### 2.4. *Foeniculum vulgare* Mill.

Fennel (*Foeniculum vulgare* Mill.) is a classical medicinal and aromatic plant with widespread traditional use, particularly for gastrointestinal complaints. Its pharmacological relevance largely stems from the rich chemical profile of its essential oil fraction, in which trans-anethole is reported as the major constituent, while volatile compounds such as estragole and fenchone are also considered characteristic components. In line with this compositional profile, fennel preparations are commonly associated with carminative and digestion-supporting uses and biological activities such as antispasmodic effects and mild antimicrobial activity have been reported in the literature ( Figure 5, Table 1) (Rather et al., 2016)

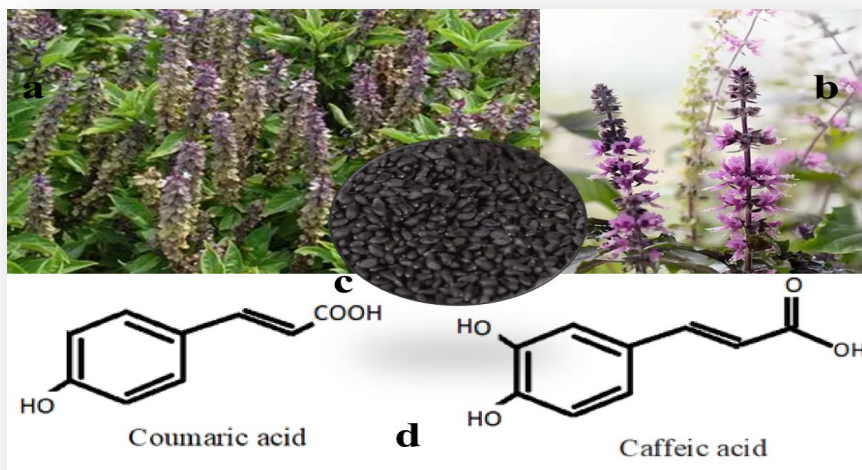


**Figure 5:** *Foeniculum vulgare* Mill. general appearance (a), flower (b), seed (c) and chemical formula of the active ingredient (d)

Fennel (*Foeniculum vulgare* Mill.) is among the aromatic species that can be considered for microgreen product portfolios due to its characteristic anise-like aroma and the sensory contribution it provides in fresh consumption. Indeed, studies aiming to identify aroma-active volatile compounds in fennel microgreens indicate that the microgreen form can exhibit a distinctive volatile profile and offers a suitable basis for product development in terms of sensory quality (Liu et al., 2024). Moreover, because microgreens are consumed as “minimally processed” fresh products, they can be more readily positioned within a functional food framework focused on dietary intake of plant bioactives; microgreens have also been emphasized as “next-generation” fresh produce with potential benefits linked to their vitamin and phytonutrient content (Xiao et al., 2012).

### **2.5. *Ocimum basilicum* L.**

Basil (*Ocimum basilicum* L.) is an important medicinal and aromatic species due to its high culinary value and its established role in traditional practices. From a phytochemical perspective, basil leaves have been reported to contain a range of phenolic compounds and flavonoids, notably rosmarinic acid and caffeic acid derivatives and this profile can significantly influence antioxidant capacity in relation to phenolic composition (Figure 6, Table 1) (Kwee and Niemeyer, 2011). In addition, rosmarinic acid one of the dominant phenolics in basil has been associated with antioxidant and anti-inflammatory effects across different biological models; therefore, it is emphasized that basil’s potential biological activities can be explained, at least in part, through these phenolic constituents (Nadeem et al., 2019).

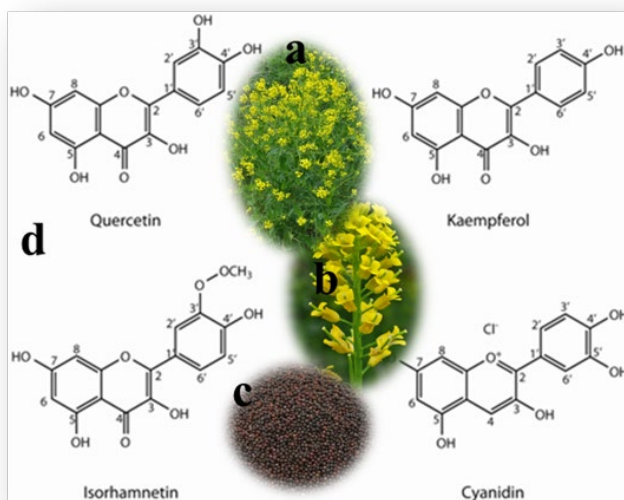


**Figure 6:** *Ocimum basilicum* L. general appearance (a), flower (b), seed (c) and chemical formula of the active ingredient (d)

Studies on basil microgreens indicate that, under controlled cultivation conditions, the combined consideration of targeted light spectrum modulation and fertilization practices can significantly enhance the accumulation of phenylpropanoid-pathway-related phenolics, particularly rosmarinic acid (Teliban et al., 2025). In addition, experimental evidence showing that adjusting red to blue LED ratios can lead to marked increases in total phenolics and selected phenolic acids (including rosmarinic acid) has positioned basil as a frequently used “model species” among medicinal and aromatic plant microgreens for metabolite optimization based on environmental elicitation (Lobiuc et al., 2017).

## 2.6. Brassicaceae Family Microgreens

The Brassicaceae family is regarded as one of the most extensively studied plant groups in the microgreen literature, with some of the strongest evidence bases in terms of both nutritional density and bioactive constituents (Dereje et al., 2023). The distinctive phytochemical signature of Brassicaceae microgreens is defined by glucosinolates and the isothiocyanates formed through myrosinase mediated hydrolysis of these compounds (Figure 7, Table 1). In particular, sulforaphanederived from glucoraphanin is among the best known isothiocyanates within the “indirect antioxidant” paradigm, as it has been linked to the regulation of enzyme systems involved in cellular defense and detoxification (Fahey et al., 2001).

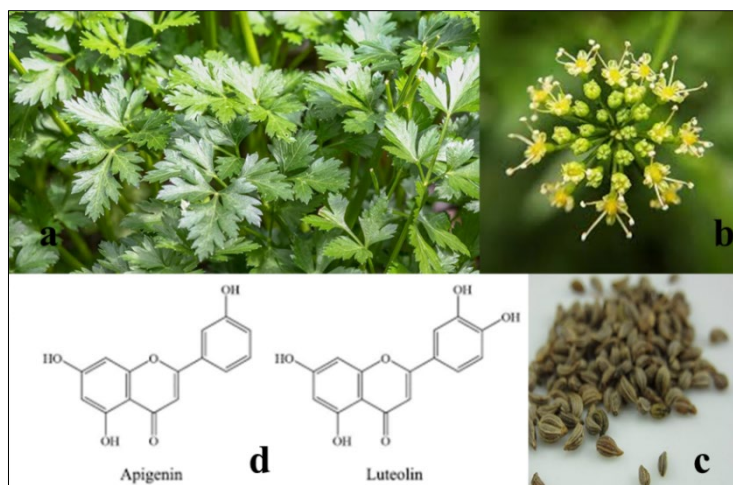


**Figure 7:** General appearance of the plant *Brassica oleracea* L. (a), flower (b), seed (c) and chemical formula of the active ingredient (d)

Studies on Brassicaceae microgreens indicate that certain bioactive compounds can be transferred into bioaccessible fractions following simulated digestion. For instance, in broccoli, curly kale, mustard and radish microgreens, antioxidant-related constituents such as ascorbic acid, soluble polyphenols and total isothiocyanates have been reported to remain bioaccessible to specific extents after gastrointestinal digestion and fractions associated with total antioxidant capacity can also be assessed. These findings suggest that the health related potential of microgreens can be substantiated not only by their compositional levels, but also by their post digestion accessibility (de la Fuente et al., 2019).

### **2.7. *Petroselinum crispum* L.**

Parsley (*Petroselinum crispum*) is widely consumed in daily diets in Türkiye and is also considered a functional plant resource due to its phytochemical profile. In particular, apigenin and luteolin derivatives are prominent within its flavonoid fraction and the literature discusses that these compounds may confer cardioprotective potential through certain cardiovascular related mechanisms, in addition to biological effects associated with antioxidant capacity (Figure 8, Table 1) (Farzaei et al., 2013).



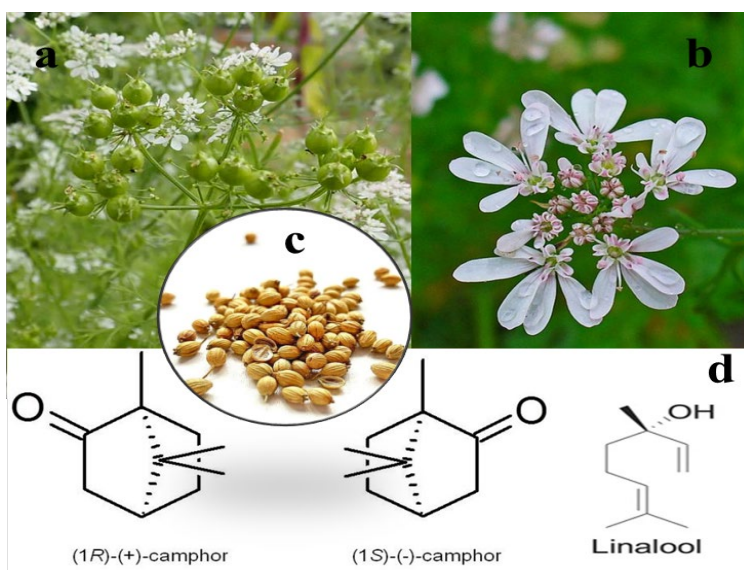
**Figure 8:** *Petroselinum crispum* L. general appearance (a), flower (b), seed (c) and chemical formula of the active ingredient (d)

*Petroselinum crispum* microgreens are among the candidate “micro herb” products that can be consumed fresh and minimally processed and are evaluated in terms of their vitamin and phytonutrient profiles. Microgreens have generally been shown to provide meaningful levels of constituents such as total ascorbic acid (vitamin C) and carotenoids; accordingly, the microgreen form has been highlighted as a notable option within the functional fresh produce approach (Xiao et al., 2012). Specifically for parsley, it has been reported that compound distribution may vary with ontogenetic stage: while the microgreen stage can present a strong profile in terms of carotenoids, more advanced harvest stages (baby greens) may show increases in indicators associated with hydrophilic antioxidants, such as total ascorbic acid and total polyphenols (El-Nakhel et al., 2021). These findings indicate that positioning parsley as a “functional” product including the choice between microgreen and baby green stages should be determined

according to the targeted compound group and the intended health-related attributes (Table 1.).

### 2.8. *Coriandrum sativum*

Coriander (*Coriandrum sativum* L.) is a medicinal and aromatic species rich in both phenolic compounds and essential oil constituents, and it has a broad research scope with respect to antioxidant activity, gastrointestinal-related functions and potential effects linked to metabolic processes (Laribi et al., 2015). In the microgreen form, the “intense phytochemical profile” concept together with a short production cycle and fresh consumption practices makes coriander a particularly promising candidate for functional food and nutraceutical product development (Bhaswant et al., 2023).



**Figure 9:** *Coriandrum sativum* L. general appearance (a), flower (b), seed (c) and chemical formula of the active ingredient (d)

Moreover, studies conducted under controlled conditions have reported that coriander microgreens can exhibit a substantial composition of bioactive constituents such as phenolic compounds and carotenoids, with accumulation dynamics that are sensitive to cultivation parameters (Figure 9, Table 1. )(Kyriacou et al., 2020).

### **3. GENERAL NUTRIENT/PHYTOCHEMICAL CONCENTRATION IN MICROGREENS AND HEALTH CORRELATION**

Microgreens have been shown to exhibit a “dense” profile in terms of nutrients and phytochemical compounds, as demonstrated by comprehensive reviews and studies evaluated together with production parameters such as the growing environment, light management, harvest timing and fertilization strategies. In this context, microgreens are considered within the framework of the “functional fresh product” approach, as they are able to provide vitamins, minerals and various secondary metabolites alongside a high antioxidant capacity. In particular, it has been reported that bioactive components such as phenolic compounds, carotenoids, ascorbic acid and glucosinolates can be present at higher levels at the microgreen stage in some species compared with more advanced developmental stages (Bhaswant et al., 2023).

These phytochemical groups are discussed in the literature as being able to contribute, at a theoretical level, to biological mechanisms associated with chronic disease risk processes, such as the limitation of oxidative stress and the modulation of inflammatory responses. Nevertheless, it is especially emphasized that a substantial proportion of the available findings are derived from in vitro approaches and animal

models and that well designed, controlled human intervention studies are required in order to more clearly elucidate the actual effects of microgreens on human health (Bhaswant et al., 2023; Seth et al., 2025).

#### **4. FUTURE PERSPECTIVE: CONTROLLED ENVIRONMENT AGRICULTURE, TARGETED COMPOUND PRODUCTION AND SAFETY**

The short production cycle of microgreens, their minimal space requirements and their rapid responsiveness to environmental variables render them one of the crop groups most compatible with Controlled Environment Agriculture (CEA) systems. Within the CEA framework, parameters such as LED light spectrum and intensity, photoperiod, nutrient solution composition and elicitation treatments can be integrated into the production process as adjustable “control knobs” thereby making the deliberate steering of phytochemical output in microgreens an increasingly attainable objective (Seth et al., 2025). This framework offers a novel production paradigm that enables medicinal and aromatic plants to be evaluated not merely as agricultural raw materials, but from a “designed phytochemistry” perspective targeting specific bioactive compound(s). Indeed, it has been reported that, in basil (*Ocimum basilicum*) microgreens, feeding strategies combined with light modulation can lead to significant increases in total phenolics and in particular, in target compounds such as rosmarinic acid and that increases in gene expression associated with the phenylpropanoid pathway may also be observed (Teliban et al., 2025).

Recent reviews synthesizing the role of LED “recipes” in shaping the nutraceutical and phytochemical profiles of microgreens further

demonstrate that the designed production approach represents a scientifically maturing line of research (Sobhanan et al., 2025).

Nevertheless, the potential for microgreens to gain a stronger position among medicinal and aromatic plant products in the future depends not only on success in “target compound enhancement”, but equally on the systematic management of safety and quality assurance. Because microgreens are predominantly consumed raw and minimally processed, risks associated with seed, water and substrate borne contamination; the attachment and proliferation of pathogens within production systems; and recontamination along the post harvest chain become critical considerations. In this context, pathogens such as *Salmonella enterica*, *Escherichia coli* and *Listeria monocytogenes* are identified as major hazards in sprout and microgreen production, with contamination sources clustering particularly around seeds, the growing environment and water (Xavier et al., 2025).

Therefore, although CEA systems offer advantages in terms of “controllability,” the translation of this advantage into food safety depends on the verifiable implementation of good agricultural and good hygiene practices, seed sanitation, water quality management, surface disinfection protocols and validated critical control points throughout harvesting, packaging and storage (Xavier et al., 2025). In particular, for microgreen medicinal and aromatic plant products marketed with a “medicinal plant” claim, traceability and standardization encompassing batch to batch chemical consistency and microbiological compliance become increasingly decisive prerequisites for market entry.

From a regulatory and evidence based perspective, while the global use of traditional and complementary medicine products continues to expand, the need for regulatory frameworks centered on quality, safety and efficacy has long been emphasized. The WHO Traditional Medicine

Strategy (2014–2023) clearly highlights the necessity of regulatory and monitoring mechanisms at the levels of products, practices and practitioners to ensure safe and effective use (World Health Organization, 2013).

Similarly, the WHO global report published in 2019 underscores the importance of strengthening national policy and implementation capacities in the field of traditional and complementary medicine through data-driven approaches (World Health Organization, 2019). The emphasis placed on risk-based regulatory approaches and appropriate standards in the recently released draft of the "2025–2034 Global Traditional Medicine Strategy" further demonstrates that this need remains current. When applied to microgreen medicinal and aromatic plant products, this framework clarifies the key determinants for the future: (i) substantiation of health claims with scientific evidence, (ii) definition of chemical "fingerprints" and quality criteria (e.g., HPLC/LC-MS/GC-MS-based targeted and untargeted profiling), (iii) regular monitoring of heavy metals, pesticide residues and production inputs (water and substrate), (iv) establishment of microbiological limits and verification testing and (v) where feasible, strengthening efficacy discussions through human intervention studies (World Health Organization, 2019; Xavier et al., 2025).

In conclusion, while microgreens possess strong potential to serve as one of the most suitable platforms for "designed phytochemistry" production within CEA systems, the sustainable translation of this potential into value depends on a standardization approach implemented in parallel with safety assurance and evidence-based regulatory frameworks (Seth et al., 2025).

The short production cycle of microgreens, their minimal space requirements, and their rapid responsiveness to environmental variables

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## **CONCLUSION**

In conclusion, medicinal and aromatic plants evaluated in microgreen form represent a promising platform for functional and phytotherapeutic product development, owing to their short production cycle, adaptability to controlled cultivation systems and suitability for fresh consumption. Their comparatively high phenolic content and diverse antioxidant secondary metabolites frequently confer a phytochemical profile that is superior to that of many vegetable-derived microgreens, supporting their added functional value.

However, the realization of this potential depends not solely on compositional richness, but on the establishment of standardized and controllable production systems capable of delivering reproducible quality profiles, alongside robust considerations of bioaccessibility, matrix effects and metabolic fate. Accordingly, the sustainable scientific and commercial advancement of medicinal and aromatic plant microgreens will rely on integrated frameworks that combine targeted phytochemical optimization with validated quality, safety and evidence-based efficacy criteria.

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## **Botanical Characteristics, Medicinal Effects and Uses of Rosehip (*Rosa canina*) Plant**

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## INTRODUCTION

Food has been required for survival and reproduction since the beginning of humanity. Until roughly ten thousand years ago, people relied entirely on gathering and hunting for sustenance. However, they eventually settled down and shifted away from gathering. Even now, modern humans retain the tradition of collecting for specific foods, which they received from their forefathers. Rosehip is among the most significant of these foods. Rosehip is valuable due to its diversity in consumption, including marmalade, jam, juice, and herbal tea, as well as its high vitamin C content (Güneş & Şen, 2001).

Rosehip (*Rosa* spp.) is beneficial to health because of its numerous bioactive chemicals. Because of its high antioxidant content, it is consumed as tea, particularly during the winter months, to treat ailments like as the flu and the common cold. Rosehip seed oil is high in fatty acids and used to cure skin damage. This oil possesses antibacterial, antioxidant, and anti-inflammatory effects and is often extracted using conventional processes such as cold pressing. Rosehip is also well-known for its medicinal and fragrant properties. Medicinal and aromatic plants have been passed down through generations as part of humanity's millennia-long history (Macit, 2024). Medicinal and aromatic herbs, which were widely employed in practically every community until the late nineteenth century, received insufficient attention in the early twentieth century due to scientific and technological advancements and socio-cultural shifts. The capacity to synthesis organic molecules, particularly in the early 1940s, resulted in the development and widespread use of synthetic medications. The economic and societal implications of World War II increased interest in synthetic pharmaceuticals while decreasing the usage of medicinal and aromatic plants (Craker et al., 2003).

Since the 1980s, increased awareness of the metabolic side effects of synthetic medications, combined with enhanced health information, has fueled interest in drugs derived from medicinal and aromatic plants,

particularly in industrialized countries. This circumstance has accelerated research and studies on medicinal and aromatic plants in recent years, allowing previously gained knowledge to be compiled in scientific papers and incorporated into the literature over time (Başer, 1998).

The *Rosa* genus has 130 species, 25 of which are found in Türkiye (Cairns, 2001; Kutbay & Kilinc, 1996; Ercisli, 2004). They are usually perennial and shrub-like. The rosehip (*Rosa canina*) species is widespread in Türkiye. It is known by names such as wild rose, rose apple, dog rose, and dog rose among the people. *Rosa canina* has a thorny structure and can be used as a rootstock for roses with crown heights ranging from one to three meters (Hartmann & Kester, 1974; Türkoglu, 1990). Rosehip syrup was used in Europe during World War II to prevent vitamin C insufficiency caused by problems with citrus fruit importation (Haas, 1995). Rosehip, which has grown in popularity in recent years, is a plant sought after in the pharmaceutical and food industries due to its mineral and vitamin content (Didin et al., 1996). It has the highest vitamin C content of any fruit or vegetable (300-4000 mg/100 g) (Ercisli, 2007) and contains minerals, carotenoids, tocopherol, bioflavonoids, fruit acids, tannins, pectins, amino acids, and essential oils (Cinar & Colakoglu, 2005). It is an important raw material in the pharmaceutical and food industries in some European countries such as Switzerland, Germany and Finland. It is also used in vitamin enrichment processes in the processing of other vegetables and fruits (Keskioglu, 1989; Kaack & Kuhn, 1991).

## 1. TAXONOMY AND DISTRIBUTION

*R. canina* (rosehip) is a significant plant species of the genus *Rosa*, which is part of the Rosaceae (Rose family), which includes over 100 genera. It is an erect, broad-leaved shrub that grows to a height of 1-2 meters (Ghazghazi et al., 2010; Tolekova et al., 2020).

**Kingdom :** Plantae

**Subkingdom :** Tracheobionta

**Division :** Magnoliophyta

**Class :** Magnoliopsida

**Subclass :** Rosidae

**Order :** Rosales

**Family :** Rosaceae

**Subfamily :** Rosoideae

**Genus :** *Rosa*

**Species :** *Rosa canina* L.

Rosaceae is a large family with roughly 2.950 recognized taxa from 100 genera of commercial and ecological value. The family name derives from the *Rosa* genus. The Rosaceae family consists primarily of herbaceous, shrubby, and tree-like plants. Most species are deciduous, while a few are evergreen. The genera *Sorbus*, *Alchemilla Crataegus*, *Rubus*, *Cotoneaster* and *Prunus* (plum, peach, cherry, apricot and almond) have the most species, totaling around 200. They are found all around the planet, but the Northern Hemisphere has the most diversity.

Rosaceae is one of the most commonly chosen plant families in ethnobotanical research (Franco et al., 2013; Garcia-Oliveira et al., 2020). The Rosaceae family includes woody or herbaceous plants that are sometimes branching. The leaves are simple or compound, usually grouped in many rows, spirally or alternately, and rarely opposite; auricles are frequently present at the base of the leaves, fused to the base of the petiole. Sepals are 4-5 in number, occasionally an epicalyx, and can be free. Petals are 4-5 in number, free, and contain numerous stamens. Flowers are actinomorphic, hermaphrodite (rarely monoecious), epicalyx, or perigynous. Hypanthium is often present.

Carpels are abundant and free. Fruit is a nut, fleshy or dry achene, juicy with seeds (drupe) or follicular. They frequently yield aggregation fruits. Endospermless seeds contain nutrients concentrated in cotyledons (Davis et al., 1972; Akkemik, 2018). Insects are typically responsible for pollination. Many organisms reproduce vegetatively. Wood's anatomy is rudimentary. Davis (1965), Blumenthal (1988) and Gruenwald et al., (2000) have all reported that flowers can be sterile.

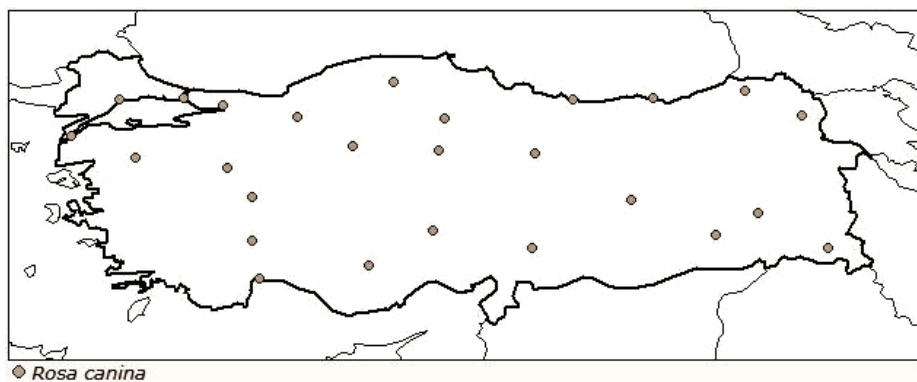
*Rosa canina* L. (Figure 1) is a 1-3 m tall shrub that grows upright and rarely climbs. The leaves are alternately oriented, dull green in color, and include 5-7 (3) leaflets. The leaflets are elliptical, narrow to widely ovate, 1.2-4.5 x 0.9-2.5 cm, often pointy at the tip, seldom blunt or rounded, with a pointed-wedge-shaped base that is rounded to blunt. The leaves are glabrous on both sides, or finely hairy on the underside; the midrib is glabrous or hairy, rarely stalked, glandular, and bears tiny spines. The stipules are 1.3-2.2 x 0.2-0.6 cm, narrow, long, and sometimes wide. The stipules are conspicuous, and the edges are whole and glandular. The lateral branches are normally arching, drooping downward, and flexible; the thorns are 3-7 mm long, tiny, and delicate on juvenile branches, but stiff, downward-curving, and uniform on older and major trunks. Thorns that resemble needles or hairs are absent. The flowers are glabrous or covered with sticky glandular hairs, solitary, and bractless, with a 1.5-2.7 cm long blackish, glandular, or glabrous stalk. Sepals are lanceolate and narrowly long-lobed at the apex, measuring 1.2-2.5 x 0.3-0.4 cm. The outer sepals are pinnatifid, widely lanceolate or narrowly, glandular-toothed lobed or entire, glabrous to softly densely hairy, and seldom glandular at the posterior end. Petals are typically curled backwards and fall off soon after flowering. Petals up to 3 cm long, white to pale pink, seldom dark pink, 1.5-2 x 1.7-2.2 cm, pointy, whitish cream or cream. Style is short and fuzzy, stigma-head is extremely broad, styles are free, typically glabrous or somewhat hairy, long, seldom finely hairy, stigma-head is more or less loose, cone or spherical, disc is broad and cone-shaped, and the mouth is narrow. Hypanthium is spherical or oval, typically glabrous, but infrequently

stalked glandular, orange-red to blackish-red when mature (Nilsson, 1972; Davis et al., 1972; Kültür, 1998).



**Figure 1:** (a) *R. canina* flowers and (b) ripe pseudo fruits of *R. canina* (Okatan et al., 2019).

Rosehip is widely distributed almost all across the world. It grows natively in a wide range of regions, including Western and Central Asia, Russia, Europe and Africa as well as North America and the Middle East. In Türkiye (Figure 2), the distribution areas of the rosehip plant are reported as Çatalca-Kocaeli, Ergene and Southern Marmara Regions, Upper Sakarya, Central and Upper Kızılırmak Regions, Inner Western Anatolia Region, Black Sea Region, Eastern Anatolia Region, Tigris Region and Mediterranean Region (Güner, 2012).



**Figure 2:** Geographic distribution of *Rosa canina* in Türkiye (TUBIVES)

## **2. CHEMICAL CONSTITUENTS OF *R. canina***

*R. canina* L. (red rosehip), a prominent member of the widely recognized *Rosa* L. genus, is regarded as a highly valuable plant-based food source due to its phytochemical properties and biological potential, attributed to its bioactive constituents including vitamin C, phenolic compounds, tocopherols, sugars, carotenoids, organic acids and essential fatty acids (Ercişli, 2007; Barros et al., 2011; Demir et al., 2014; Nadpal et al., 2016).

### **2.1. Fruit (hip) Components**

Rosehip fruit includes vitamin C, phenolics, carotenoids (e.g., lycopene, lutein/zeaxanthin), tocopherols, tannins, pectins and organic acids. USDA research showed that 100 g of wild rosehip contains 426 mg of vitamin C, 6800 µg of lycopene, and 2001 µg of lutein+zeaxanthin (Fan et al., 2014).

### **2.2. Seed and Oil Fraction**

The seed fraction contains high levels of fatty acids, including linoleic and  $\alpha$ -linolenic acid which have been linked to COX enzymes involved in inflammation (Fan et al., 2014; Jäger et al., 2008).

### **2.3. GOPO (galactolipid)**

GOPO, found in rosehip, is an anti-inflammatory galactolipid that prevents human peripheral neutrophil chemotaxis (Larsen et al., 2003; Fan et al., 2014). However, it is important to note that the amount of GOPO in rosehip powder may be modest and that the overall rheumatological activity cannot be explained only by GOPO; the impact is related to the synergy of other components (Fan et al., 2014).

Additionally, the table below summarizes the prominent components, reported effects and types of evidence in *R. canina* (Table 1).

**Table 1:** Selected bioactive constituents of *Rosa canina*, proposed mechanisms of action, and levels of evidence

<b>Bioactive constituent</b>	<b>Plant part</b>	<b>Proposed mechanism(s) of action</b>	<b>Level of evidence</b>	<b>Reference(s)</b>
Ascorbic acid (Vitamin C)	Fruit	Free radical scavenging; inhibition of lipid peroxidation	In vitro, review	Roman et al., 2013; Fan et al., 2014
Phenolic compounds	Fruit, leaves	Modulation of inflammatory mediators; antioxidant activity	In vitro, animal studies	Chrubasik et al., 2008; Pazhouhi et al., 2020
Carotenoids (e.g., lycopene, $\beta$ -carotene)	Fruit	Cellular protection against oxidative damage	In vitro	Fan et al., 2014
Galactolipid (GOPO)	Fruit	Inhibition of neutrophil chemotaxis; suppression of inflammatory response	In vitro	Larsen et al., 2003
Galactolipid (GOPO)	Fruit	Reduction of pain and stiffness in osteoarthritis	Randomized controlled trials (RCTs)	Winther et al., 2005; Chrubasik et al., 2008
Linoleic acid	Seed	Cyclooxygenase (COX-1 and COX-2) inhibition	In vitro	Jäger et al., 2007; Jäger et al., 2008
$\alpha$ -Linolenic acid	Seed	Anti-inflammatory modulation	In vitro	Jäger et al., 2008
trans-Tiliroside	Seed	Regulation of lipid metabolism; anti-obesity effect	Animal studies	Ninomiya et al., 2007
Standardized rose hip powder	Fruit + seed	Reduction of pain and stiffness in osteoarthritis	Randomized controlled trials (RCTs)	Winther et al., 2005
Rose hip preparations	Fruit	Improvement in functional disability scores in rheumatoid arthritis	Randomized controlled trials (RCTs)	Willich et al., 2010

### **3. PHARMACOLOGICAL ACTIVITIES OF *R. canina***

#### **3.1. Antioxidant Effect**

The antioxidant capacity of *R. canina* preparations is explained by the contribution of different fractions, primarily polyphenols and vitamin C. A systematic review reports that various extracts (including phenolic/ascorbate/lipophilic fractions) showed activity in different antioxidant tests, with the phenolic fraction having a significant share (Chrubasik et al., 2008).

#### **3.2. Anti-inflammatory effect and joint diseases**

The inhibitory effect of GOPO on neutrophil chemotaxis is considered one of the main mechanisms of rosehip's anti-inflammatory profile (Larsen et al., 2003). Furthermore, it has been reported that rosehip extracts show inhibition of COX-1 and COX-2; some studies have shown that the efficacy is more pronounced in organic solvent extracts (Jäger et al., 2007).

#### **3.3. Osteoarthritis (OA)**

The most debated clinical evidence in OA is from randomized, double-blind, placebo-controlled studies with standardized rosehip powder. Winther et al., (2005) reported that rosehip powder may have beneficial effects on symptoms in patients with hip/knee osteoarthritis.

#### **3.4. Rheumatoid Arthritis (RA)**

In a randomized controlled trial by Willich et al. (2010), improvement in functional patient scores such as HAQ-DI was reported with a standard rosehip preparation (Willich et al., 2010; Fan et al., 2014).

#### **3.5. Metabolic effects (anti-obesity, etc.)**

It has been reported that trans-Tiliroside can exert effects at the preclinical level through pathways related to body weight/visceral fat

and lipid metabolism (Ninomiya et al., 2007). The review sources also report different experimental evidence regarding the anti-obesity/anti-hyperlipidemic potential of rosehip, but emphasize that further studies are needed to clarify clinical significance on an indication basis (Chrubasik et al., 2008; Pazhouhi et al., 2020).

### **3.6. Urinary System and Infections**

Seifi et al. (2018), reported a randomized placebo-controlled (triple-blind) study evaluating the effect of rosehip use on the incidence of UTIs during the puerperium period after cesarean section.

## **4. AREAS OF USE**

### **4.1. Food and Functional Food Applications**

Rosehip is widely used in products such as tea, syrup, jam/jelly, and juice; it can also be considered as an ingredient in probiotic beverages and various food formulations (Pazhouhi et al., 2020).

### **4.2. Phytotherapeutic Products and Supplements**

Standardized rosehip powder preparations (fruit peel + seed fractions) have been the subject of clinical research, particularly in the context of OA/RA symptom management (Winther et al., 2005; Willich et al., 2010).

### **4.3. Cosmetic/Topical Use (oil)**

Seed oil is being investigated for skin applications (e.g., barrier support, appearance improvement claims) due to its fatty acid profile and antioxidant components; this area has more heterogeneous evidence, and product standardization appears critical (Chrubasik et al., 2008).

## 5. CONCLUSION

In conclusion, *Rosa canina* is a biologically rich medicinal plant with considerable potential as a functional food and complementary therapeutic agent. The accumulated evidence clearly demonstrates strong antioxidant and anti-inflammatory properties, supported by robust experimental data and moderate clinical evidence, particularly in the context of osteoarthritis. These effects appear to result from the synergistic action of multiple bioactive constituents rather than from a single active principle.

Clinical findings suggest that standardized rose hip preparations can provide meaningful, albeit moderate, symptomatic relief in inflammatory and degenerative joint diseases. However, the current body of evidence does not support the use of *R. canina* as a stand-alone treatment. Instead, it should be regarded as a supportive intervention integrated into broader, evidence-based treatment strategies.

Future research should prioritize the development of standardized *R. canina* preparations, the elucidation of precise molecular mechanisms underlying its pharmacological effects, and the conduct of large-scale, long-term randomized controlled trials. Such studies are essential to establish optimal dosing regimens, assess long-term safety, and clarify the clinical relevance of emerging indications, including metabolic and immunological disorders.

Overall, *R. canina* represents a compelling example of a traditional medicinal plant whose historical use is increasingly supported by modern scientific research. With appropriate standardization and rigorous clinical evaluation, rose hip may play an increasingly important role in the fields of functional nutrition and evidence-based phytotherapy.

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**Distribution Areas and Medicinal  
Effects of Thymus Species in  
the Eastern Black Sea Region of Türkiye**

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## INTRODUCTION

Medicinal and fragrant plants have long served as raw ingredients in spices, medicines, and cosmetics. People have moved to cheaper, more easily accessible, and solution-oriented therapeutic drugs as a result of reasons such as global population growth, rising diseases, changing living situations, and personal preferences. Naturally, this has advanced the synthetic medicine industry and increased investment in these sectors. However, because of the adverse effects they create on living beings, people have begun to look for compounds that are biologically and naturally compatible. These chemicals have numerous negative consequences, including increasing prices, a lack of effective therapies, and the development of resistance (Bayram et al., 2010).

Furthermore, medicinal plants are employed in nutritional supplements and diets, including herbal teas, spices, and seasonings. They are utilized in body care, fragrance, and cosmetics, as well as pesticides and polishing. These plants' medicinal leaves are derived from one or more of their parts (root, rhizome, stem, tuber or woody structure, bark, leaf, flower, fruit, seed or herb) (Yurteri, 2018). Medicinal aromatic plants have numerous essential secondary metabolites. These secondary metabolites have numerous applications (Ceylan, 1997).

Türkiye has a rich diversity of plant species. The main reason for this diversity is that it is bordered by the continents of Asia and Europe, surrounded by seas on three sides, and geographically encompasses the characteristics of three major phytogeographic regions (European-Siberian, Irano-Turanian, and Mediterranean), each with its own climatic and soil characteristics. This diversity is essential for medicinal and aromatic plants. Turkey's medicinal and aromatic plant diversity necessitates research into their protection, development, commercialization, and distribution (Duke, 1991; Büyükgebiz et al., 2008).

One-third (3.649) of Turkey's 11.707 species, subspecies, variations, and hybrids are indigenous plants (Arslan et al., 2015). In this sense, the Eastern Black Sea region is very significant. 222 of the 2.239 plant species found in the area are indigenous. In terms of species diversity, the Black Sea region ranks third in Türkiye. According to Davis (1982), aromatic and medicinal plants make up 70% of the species found in the area. In terms of plant species, the Eastern Black Sea Region is significant. It is crucial to concentrate on the plants in the area since they contain species with significant medicinal and commercial significance, particularly those in the Lamiaceae family (Küplemez, 2025).

The Lamiaceae are a well-known family of commercially valued plants, shrubs, and trees. These range from wood trees like *Tetoma grandis*, found in Asia's tropical rainforests, to spice, fragrant, and medicinal genera in the Mediterranean region such as *Salvia*, *Thymus* L., *Mentha* L., *Origanum* L., *Rosmarinus* L., *Lavandula* L., *Sideritis* L. and *Satureja* L. They grow natively in warm and temperate regions of the world, particularly around the Mediterranean. They are represented by only a few species in colder climates. Many genera in the family, including *Ajuga* L., *Salvia* L., *Phlomis* L., *Nepeta* L., *Coleus* L. and *Clerodendrum* L. are used ornamentally. Members of the family are an essential plant category for beekeeping because they are rich in nectar (Tanker and Tanker, 1990).

## **1. GEOGRAPHICAL DISTRIBUTION OF *THYMUS* SPECIES IN THE EASTERN BLACK SEA REGION**

The floristic and ethnobotanical data compiled within the scope of this study demonstrate that the Eastern Black Sea Region possesses a remarkable structure in terms of both species' diversity and ecological differentiation of the genus *Thymus*. The sharp gradients in climatic conditions and altitude over relatively short distances from the coastal

zone toward the interior parts of the region play a decisive role in shaping the distribution patterns of *Thymus* taxa (Table 1).

Floristic investigations conducted in the provinces of Trabzon and Rize reveal a pronounced prevalence of subspecies and varieties belonging to the *Thymus praecox* species complex. This pattern indicates that members of this group are well adapted to humid, cool, and high-altitude mountainous environments (Eminağaoğlu & Anşın, 2006; Korkmaz & Engin, 2019). In particular, records reported from the Anzer, Ayder, and Ceymakçur plateaus in Rize Province clearly demonstrate that *T. praecox* constitutes a dominant element of the alpine belt in this area (Başer et al., 2002).

In a study conducted by Küplemez (2025), *Thymus* samples collected from different provinces of the Eastern Black Sea Region were floristically identified and their essential oil compositions were analyzed using the GC-MS method. As a result of evaluating samples obtained from plateau and mountainous areas within the borders of Rize province, it was revealed that *Thymus praecox*, *Thymus nummularius*, and *Thymus kotschyanus* species have a natural distribution in Rize province (Küplemez, 2025).



**Figure 1.** *T. praecox* ssp. *grossheimii* var. *grossheimii* is found in the Anzer plateau (Küplemez, 2025).

**Table 1.** *Thymus* species by province in the Eastern Black Sea region

Province	<i>Thymus</i> taxon	Habitat / Altitude	Reference
Trabzon	<i>Thymus praecox</i> subsp. <i>skorpilii</i> var. <i>skorpilii</i>	Subalpine–alpine meadows, rocky slopes	Eminağaoğlu & Anşin (2006)
Trabzon	<i>Thymus praecox</i> subsp. <i>grossheimii</i> var. <i>grossheimii</i>	Alpine belt	Eminağaoğlu & Anşin (2006)
Trabzon	<i>Thymus pubescens</i>	Stony hillsides	Eminağaoğlu & Anşin (2006)
Trabzon	<i>Thymus pseudopulegioides</i>	Open areas, mountain foothills	Eminağaoğlu & Anşin (2006)
Rize	<i>Thymus praecox</i> (various subspecies)	Alpine and subalpine plateaus	Başer et al. (2002); Küplemez (2025)
Rize	<i>Thymus nummularius</i>	Subalpine–alpine grasslands	Korkmaz & Engin (2019); Küplemez (2025)
Rize	<i>Thymus kotschyanus</i>	Rocky slopes, relatively dry microhabitats	Küplemez (2025)
Rize	<i>Thymus longicaulis</i> subsp. <i>longicaulis</i>	Roadsides, grasslands	Akan et al. (2014)

According to the findings of the study, the high-altitude plateaus and alpine zone of Rize, in particular, offer suitable ecological conditions for *Thymus praecox*. It was stated that *T. praecox* is commonly observed in these areas, characterized by humid and cool climates, and constitutes a large portion of the samples subjected to chemical analysis. This situation supports the species' ability to adapt to the Colchic-Euxine zone of the Eastern Black Sea (Küplemez, 2025).

Furthermore, it has been stated that *Thymus nummularius* was among the samples collected from Rize province within the scope of this study, and that the species is particularly distributed in subalpine and alpine meadows. The study emphasizes that *T. nummularius* can be found together with *T. praecox* in the high mountain ecosystems of Rize, but exhibits significant differences in terms of habitat preferences and chemical profiles (Küplemez, 2025).

The study also reports that another important taxon identified as being distributed in Rize province is *Thymus kotschyanus*. It is stated that this species is mostly found in rocky, relatively drier microhabitats and on sloping hillsides; and that these ecological conditions affect its essential oil composition. Küplemez (2025), states that the presence of *T. kotschyanus* in the Rize flora indicates that the province is not only under the influence of the humid Black Sea, but also harbors micro-scale terrestrial and semi-arid habitats (Table 1).

## 2. MEDICINAL EFFECTS AND CHEMICAL CONSTITUENTS OF *THYMUS* SPECIES

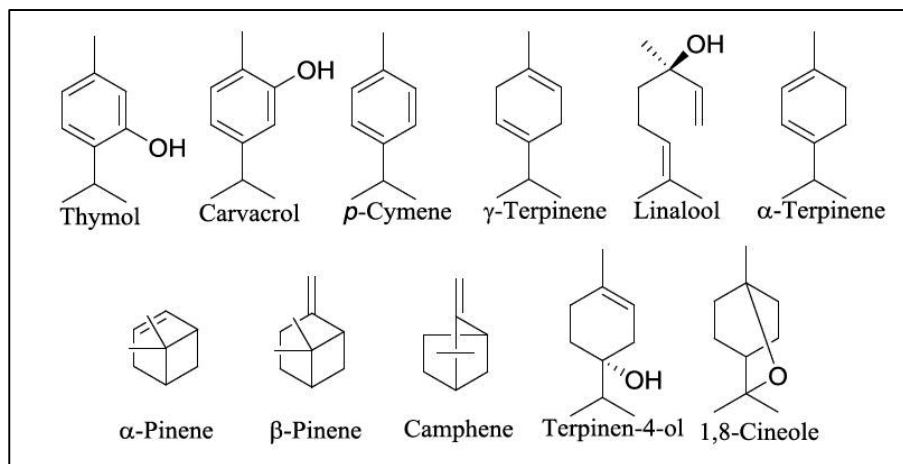
*Thymus* is a plant genus in the Lamiaceae family that produces essential oils. It contains multiple species and subspecies. Many species are found throughout the Mediterranean phytogeographic region (Cosentino et al., 1999). *Thymus* essential oils are utilized in the food sector as flavorings in a variety of meals, beverages, and confectionary, as well as preservatives due to their antibacterial qualities. In the perfumery industry, they are used to fragrance soaps and lotions. *Thymus* essential oils are utilized as nutritional supplements due to their antioxidant characteristics. *Thymus* essential oils possess antiseptic, antibacterial, antifungal, antispasmodic, antitussive, expectorant, and analgesic characteristics, making them useful in medicine and pharmacology (Cosentino et al., 1999; Hedhili et al., 2002; Rasooli & Mirmostafa, 2002; Rasooli et al., 2006; Kabouche et al., 2005).

In Türkiye, species from the *Thymus* genus are referred to as thyme or stone thyme (Başer et al., 2002; Bağcı & Başer, 2005). Aside from being used to add flavor to meals, it is also used in traditional medicine to treat diabetes, regulate blood circulation, as a bile enhancer, anthelmintic, cold relief, asthma breath reliever, and hunger stimulant (Baytop, 1999; Tümen et al., 1995; Başer et al., 1991; Işık et al., 1995). The essential oils of 162 species in the genus *Thymus* have been studied worldwide. These studies resulted in the identification of 360 distinct components in *Thymus* species' essential oils. 75% of these components are terpenes. Monoterpenes account for 43% of the terpenes, whereas sesquiterpenes make up 32%. The percentage of components other than terpenoids is minimal. Chemical analyses have gotten easier as analytical techniques such as GC and GC-MS have advanced, and the composition of chemical substances may now be determined with 95% accuracy (Stahl-Biskup, 2002a). According to studies, *Thymus* species contain roughly 270 terpenes, one or more of which are prominent. The most important terpenes include thymol, carvacrol, linalool, p-cymene, geraniol, and borneol. *Thymus* species are the plant kingdom's most abundant source of monoterpenoid phenols (Stahl-Biskup, 2002b).

Terpenes, terpene alcohols, volatile oils derived from terpenes, esters, and phenolic derivatives are typically the main subjects of research on *Thymus* species. Among these, two well-known additions that contribute to the concentration of commercial interest are thymol and carvacrol, which do not provide a safety risk in accordance with the World Health Organization's (WHO) food additive standard. Less often cited in recent studies are other non-volatile organic substances such as flavonoids, simple phenylpropanoids, lignans, tannins, organic acids, terpenoids, and phytosterols. According to pharmacological studies, chemicals and extracts from *Thymus* species have a variety of properties both in vitro and in vivo, such as antimicrobial, antioxidant, antitumor, anti-inflammatory, analgesic, antispasmodic, antitussive, carminative, antihypertensive, anti-diabetic, and anthelmintic properties (Li et al., 2019).

Additionally, *Thymus* species have been thoroughly investigated as substitutes for commercial synthetic compounds in recent years. On the one hand, *Thymus* essential oils have been successfully added to packaging materials because they are rich sources of active antimicrobial and/or antioxidant compounds that help prolong the shelf life and enhance the quality of fresh foods (Zare et al., 2019; Dairi et al., 2019).

*Thymus* species are a significant source of monoterpenoid chemicals in medicinal and aromatic plants. While about 270 terpenes have been identified in these species, 52 of them are classed as terpenes, with 34 of them considered key volatile components for the *Thymus* genus. Thymol, carvacrol, linalool, p-cymene,  $\gamma$ -terpinene, borneole, 1,8-cineole, and geraniol are the key monoterpenes discovered in *Thymus* species (Figure 2).



**Figure 2.** Chemical structure of some important monoterpenes found in *Thymus* species (Li et al. 2019).

Monoterpenes such as  $\gamma$ -terpinene, p-cymene, thymol, and carvacrol are closely linked in biogenetic processes. *Thymus* species have significant levels of  $\gamma$ -terpinene and p-cymene, both monoterpene hydrocarbons, which are linked to the occurrence of thymol and carvacrol. This can be explained by the fact that some components are

derived from their progenitors.  $\gamma$ -terpinene and p-cymene are precursors of thymol and carvacrol in *Thymus* and *Origanum* species (Cosentino et al., 1999; Stahl-Biskup, 2002a; Ultee et al., 2002).

In addition to essential oils, thyme plants are also rich in phenolic compounds. In particular, flavonoids (e.g., luteolin, apigenin derivatives), phenolic acids (e.g., rosmarinic acid), triterpenes, and tannins are other important phytochemicals detected in thyme species (Jarić et al., 2015). A study conducted on *T. praecox* samples collected from the Kastamonu region reported that the plant had high amounts of total phenolic and flavonoid compounds in various polar extracts, and that the presence of flavonoids such as quercetin, luteolin, and apigenin was confirmed in HPLC analyses (Şener et al., 2021). The same study also stated that minerals such as calcium, potassium, and iron were detected in the leaves and stems of *T. praecox*.

Yurteri (2023), in a study, analyzed cultured *T. praecox* subspecies collected from the highlands of Rize using GC/MS and HPLC. The range of secondary metabolite amounts (mg/g DW) of the flora and cultured samples was found to be: 32.817–225.635 and 28.3–226.3 for isoquercitrin, 926.803–1543.241 and 890.3–1462.3 for rosmarinic acid, 139.44–287.894 and 129.9–312.2 for thymol, and 38.619–121.424 and 26.3–112.9 for galocatechin. Volatile components were also found to be: The concentrations were found to be 5.9–45.42% for carvacrol, 7.75–48.49% for thymol, 2.64–24.65% for  $\alpha$ -terpinyl acetate, 1.59–15.97% for linalool, 0.00–12.47% for carvone, 0.00–12.98% for linalyl acetate, 0.38–9.49% for  $\gamma$ -terpinene, 0.14–9.22% for isoborneol, 1.22–8.69% for  $\alpha$ -himachalene, 0.42–6.27% for menthone, 0.78–5.95% for (Z)-jasmone, and 0.3–2.86% for trans-sabinene hydrate.

Similarly, the presence of phenolic components such as rosmarinic acid, caffeic acid derivatives, and flavonoid glycosides has been reported by other researchers for *T. nummularius* (Ertas et al., 2015). These phenolic compounds significantly contribute to the antioxidant capacity of thyme extracts. Rosmarinic acid, in particular, is a potent antioxidant,

frequently found in thyme and similar plants of the Lamiaceae family, and is also known for its anti-inflammatory properties. In summary, when the thyme species of the Eastern Black Sea are chemically analyzed, it is understood that their essential oils contain terpenes such as thymol, carvacrol, borneol, 1,8-cineol, and myrtenyl acetate; and their phenolic extracts contain gallic acid, isoquercitrin (quercetin derivative), various flavonoids, and phenolic acids (Küplemez, 2025; Şener et al., 2021).

This rich chemical content is the fundamental element explaining the multifaceted biological effects of thyme species (Table 2).

Another noteworthy aspect of thyme species is their antioxidant capacity. Their ability to scavenge free radicals and inhibit lipid peroxidation makes them valuable as natural preservatives in the food industry. For example, *T. praecox* extracts have been found to have a high total antioxidant capacity, and it has been suggested that thyme extracts can be used instead of artificial antioxidants to extend the shelf life of fatty foods (Şener et al., 2021). It is folklorically known that the local people in the Eastern Black Sea highlands add thyme to butter to preserve it, and that this delays the butter from becoming rancid (this practice is scientifically consistent with the antioxidant properties of thyme). A report prepared by DOKAP also states that *T. nummularius* can be used as a natural antioxidant in food preservation (DOKAP, 2020).

**Table 2.** Summary of the Biological and Pharmacological Effects of *Thymus* Species Native to the Eastern Black Sea Region

Thymus Species	Biological / Pharmacological Effects	References
<i>T. nummularius</i> (Plateau thyme)	– Essential oil rich in <b>thymol</b> and <b>carvacrol</b> ; strong <b>antiseptic and antimicrobial</b> activity, especially against Gram-positive bacteria (Bektaş et al., 2016; Jarić et al., 2015).– <b>Antiviral</b> : inhibits certain viruses such as HSV-1 <i>in vitro</i> (Bektaş et al., 2016).–	Bektaş et al., (2016); Özkök et al., (2025);
<i>T. praecox</i> (Anzer thyme)	– <b>Antibacterial</b> : extracts inhibit <i>Staphylococcus aureus</i> (8–12 mm inhibition zones); especially effective against Gram-positive strains (Şener et al., 2021).– <b>Antioxidant</b> : high total phenolic content; strong DPPH and H <sub>2</sub> O <sub>2</sub> scavenging activity.– <b>Antimicrobial</b> : potential natural antimicrobial for complementary therapy.– <b>Anti-inflammatory potential</b> suggested due to high rosmarinic acid levels, though direct studies are limited.	Şener et al. (2021); Jarić et al. (2015)
<i>T. kotschyanus</i>	– <b>Anti-inflammatory</b> : reduced fecal calprotectin levels by 55% in ulcerative colitis patients in a clinical trial (Vazirian et al., 2022).– <b>Antidepressant</b> : reduced depression-like behavior in mice in behavioral tests (Doosti et al., 2018).– <b>Antitumor</b> : showed dose-dependent cytotoxicity against A549 (lung) and HeLa (cervical) cancer cell lines <i>in vitro</i> .– <b>Antimicrobial</b> : effective against certain bacteria and fungi (e.g., <i>E. coli</i> , <i>Candida</i> spp.).	Vazirian et al. (2022); Doosti et al. (2018); Bakhtiarian et al. (2011)
<i>T. serpyllum</i> (Wild thyme)	– <b>Antimicrobial and antifungal</b> : broad-spectrum activity against <i>S. aureus</i> , <i>E. coli</i> , and <i>Candida</i> spp. (Jarić et al., 2015).– <b>Antioxidant</b> : high antioxidant content and free radical scavenging capacity.– <b>Cytotoxic / Anticancer potential</b> : some evidence of activity against cancer cell lines, though further research is needed.– <b>Traditional uses</b> : widely used as expectorant, antitussive, and antispasmodic in folk medicine.	Jarić et al. (2015); Tanker & İlisulu (1981)

There is a growing body of research in the literature on the anti-inflammatory and other pharmacological effects of thyme. Studies, particularly on *Thymus kotschyanus*, have shown that this species may possess anti-inflammatory and immune-modulating effects. A preclinical study conducted in Iran reported that *T. kotschyanus* extract reduced edema and inflammation in experimental animal models (Bakhtiarian et al., 2011). Subsequently, a clinical study by Vazirian et al., (2022) evaluated the efficacy of *T. kotschyanus* extract as an adjunct therapy in patients with ulcerative colitis.

## 5. CONCLUSION

*Thymus* (thyme) species growing in the Eastern Black Sea Region have historically found widespread use as folk medicine and exhibit a rich biological activity profile that has attracted the attention of modern science. This comprehensive study examines the phytochemical properties and medicinal effects of the region's main thyme species, *T. nummularius*, *T. praecox*, *T. kotschyanus* and *T. serpyllum* based on a literature review.

The thyme species in the region contain high levels of volatile oils (thymol, carvacrol, etc.) and phenolic compounds (flavonoids, rosmarinic acid, etc.). It was found that *T. nummularius* is particularly rich in thymol/carvacrol, while *T. praecox* and *T. kotschyanus* exhibit different terpene and ester components. This chemical diversity is the source of the different pharmacological effects of the species. Furthermore, strong antimicrobial (against bacteria and fungi) and antioxidant effects have been reported in all thyme species. Examples of these effects include the inhibition of pathogens such as *S. aureus* and the neutralization of free radicals by *T. praecox* extracts, and the broad-spectrum antiseptic properties of *T. nummularius* oil. These findings suggest that thyme can be a natural agent for infectious diseases and food preservation. Regarding anti-inflammatory and other pharmacological

effects, studies on *T. kotschyanus* have revealed that thyme extracts possess anti-inflammatory potential (e.g., reducing symptoms in ulcerative colitis).

Species like *T. serpyllum* are also valuable in complementary medicine for their antioxidant support and respiratory tract soothing effects. The uses of thyme species in folk medicine (for colds, coughs, digestive problems, infections, etc.), which have been used in the region for centuries, have been largely confirmed by modern research. This situation creates a bridge between traditional and modern medicine and once again emphasizes the importance of medicinal plants growing in the region. Sustainable harvesting and cultivation methods should be developed for the region's thyme species, and standardized extract and oil production should be encouraged. Future studies should focus on elucidating the mechanisms of action of thyme components, optimal dosages, and application methods. In particular, clinical research should be increased to comprehensively evaluate the efficacy and safety of thyme extracts in human health; possible side effects and interactions should be identified. In conclusion, *Thymus* species growing in the Eastern Black Sea Region both make valuable contributions to our country's phytotherapy literature and create an opportunity for the regional economy. Protecting this endemic and natural richness and supporting it with scientific studies will pave the way for the development of new natural medicine raw materials and healthy food additives in the future. Continuing research conducted with interdisciplinary approaches (botany, chemistry, pharmacology, clinical sciences) is of great importance to fully reveal the potential of thyme species.

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***Berberis sp.: An Important Medicinal Plant in  
Global Traditional Medicine***

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## INTRODUCTION

With the rapidly increasing demand for natural products and research in this area, interest in medicinal plants is growing, and the bioactive compounds of these plants, historically used in folk medicine, are playing a significant role in modern medicine. The World Health Organization (WHO), in its Global Traditional Medicine Strategy report covering the period 2025–2034, provides an international framework for supporting the use of medicinal plants in safe, effective, and community-centered healthcare (Anonymous, 2024a). Furthermore, recent WHO reports indicate that traditional medicine practices are used by a significant portion of the world's population for the treatment of diseases, and that the integration of these practices into modern healthcare systems can have positive impacts on public health (Anonymous, 2024b). Secondary metabolites found in medicinal plants, such as alkaloids, phenolic compounds, flavonoids, and terpenoids, exhibit antimicrobial, antioxidant, and anti-inflammatory effects, offering valuable biological properties for both folk medicine and scientific research (Pan et al., 2013).

In this context, species belonging to the genus *Berberis* stand out among the important medicinal plants due to their rich chemical components and broad pharmacological effects. *Berberis* species, especially those containing numerous isoquinoline alkaloids, primarily berberine, and possessing antioxidant and antimicrobial activities, have long been of interest in both traditional and modern medicine. Their wide distribution, adaptability to different ecological conditions, and phytotherapeutic potential make the *Berberis* genus noteworthy in both scientific research and pharmaceutical applications. Taxonomically, *Berberis* L. belongs to the Berberidaceae family (Mozaffarian, 2013). The barberry plant is a thorny shrub with triangular-shaped thorns. It grows in mountainous regions and on slopes near water sources. Its fruits are predominantly seedless and fresh when harvested. The stem resembles that of jasmine but is larger and more slender, exhibiting a

yellowish-white coloration throughout. The fruit forms elongated clusters and is firm when unripe. Upon maturation in summer, it transitions from red to purple-black. The ripe fruit is spherical, with the red variety being considered of superior quality (Khorasani Shirazi, 2008; Bayat et al., 2025).

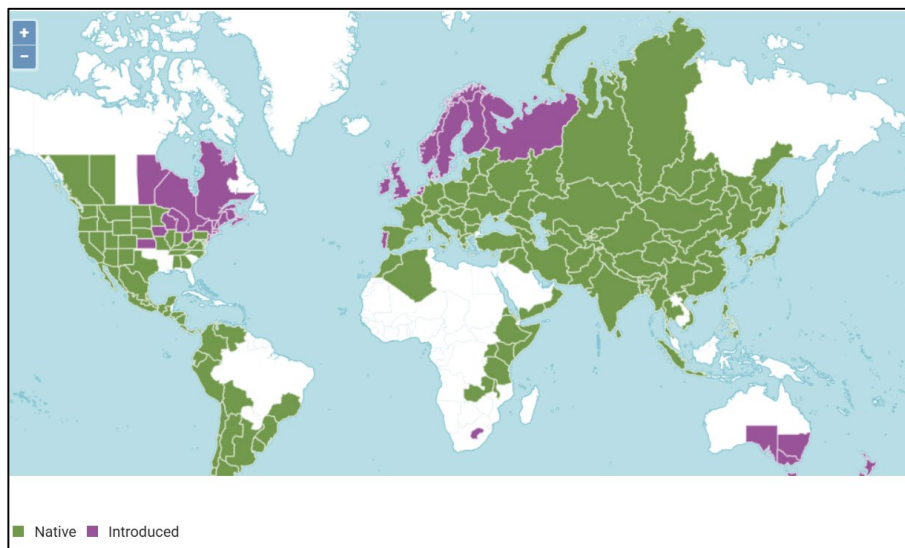
## **1. BOTANICAL CHARACTERISTICS AND GEOGRAPHICAL DISTRIBUTION OF BERBERIS SPECIES**

The medicinal significance of barberry (*Berberis* spp.) stems from bioactive compounds present in multiple plant parts, including roots, bark, leaves, and fruits (Gholizadeh Moghaddam et al., 2017). *Berberis* is known as "Zereshk" in Iran and "Ambarbaris" in TPM (Bayat et al., 2025). The barberry plant is referred to by different vernacular names across various languages and regions. In Arabic and Persian, it is known as Huzuz, while in English it is called Indian barberry. The plant is referred to as Rasaut in Urdu and Kashmiri. In Tamil, it is known as Mullukala or Usikkala, whereas in Punjabi it is called Chitra, Simlu, or Kasmal. In Hindi, the commonly used names are Chitra and Darhald. In Malayalam, it is referred to as Maradarisina or Maramanjal. In Bengali, Marathi, and Hindi, the name Darhaldi/Darhald is widely used. Additionally, in Sanskrit, the plant is traditionally known as Darvi, Kata, or Daruharidra (Wani et al., 2024).

The genus *Berberis* has numerous species worldwide; global taxonomic databases and phylogenetic studies indicate a wide distribution across many continents (Anonymous, 2025, Figure 1). *Berberis* exhibits a broad native distribution, encompassing temperate and subtropical zones across Asia, Europe, Africa, North America, and South America (Rahimi-Madiseh et al., 2017; Tavakoli et al., 2021). The richest areas for *Berberis* species are Asia; Central Asia, the Iran-Anatolia belt, and the Himalayan region, in particular, show high species diversity. China and the Himalayan belt are among the main centers

where the species diversity of the genus is concentrated. In addition, numerous local species have been reported in the Himalayan foothills of India (Tiwari et al., 2012). In Europe, some species (e.g., *Berberis vulgaris*) occur naturally in forest edges, scrublands, and mountainous areas. While a limited number of *Berberis* species also have a natural distribution in North America, endemic species are concentrated in South America, particularly in the Andes Mountains (from Venezuela to Argentina/Chile). Many endemic species have been reported in Chile and Argentina (Landrum, 1999). In North Africa (Mediterranean region; Morocco, Algeria, Tunisia, etc.), there are a limited number of *Berberis* species; in general, the *Berberis* genus is more common in temperate/mountainous regions rather than tropical Africa (Rashmi et al., 2008).

The barberry plant is a thorny shrub with triangular-shaped thorns. It grows in mountainous regions and on slopes near water sources. Its fruits are predominantly seedless and fresh when harvested. The stem resembles that of jasmine but is larger and more slender, exhibiting a yellowish-white coloration throughout. The fruit forms elongated clusters and is firm when unripe. Upon maturation in summer, it transitions from red to purple-black. The ripe fruit is spherical, with the red variety being considered of superior quality (Khorasani Shirazi, 2008; Bayat et al., 2025).



**Figure 1:** Distribution of *Berberis* species in the world.

Iran is both a center for natural diversity and an economically significant production of the *Berberis* genus. Several species—*B. crataegina*, *B. khorasanica*, *B. orthobotrys*, *B. vulgaris*, and *B. thunbergii* var. *atropurpurea*—*B. integerrima* are distributed in Iran (Zarei et al., 2015; (Azadbakht & Ghahremaninejad, 2023). In Iran, Southern Khorasan is the primary producer of barberries (*Berberis* spp.), with over 97% of its cultivation occurring in Ghaenat County, Southern Khorasan Province. This region accounts for 95% of Iran's total barberry fruit production. While various *Berberis* species are cultivated worldwide for specific purposes, barberries are particularly renowned and extensively utilized in culinary applications (Rahimi-Madiseh et al., 2017). Based on studies (Azadi, 2009), the biodiversity of this genus in Iran is remarkable, with approximately six native species identified in the country. Photographs present various *Berberis vulgaris* tree specimens native to Iran (Motalebipour & Pirestani, 2025) (Figure 2).



**Figure 2:** Morphological variants of *Berberis vulgaris*

These species grow in mountainous ecosystems and arid regions, playing a vital role in soil conservation and ecological balance. Iran, with approximately 11,000 hectares under cultivation, accounts for about 95% of the total global production of seedless barberry fruit (*Berberis vulgaris* L.) (Alemardan et al., 2013).

Barberry grows as a thorny shrub, typically reaching heights of 1 to 3 meters, and possesses brittle branches. In favorable environments, its height may reach up to 6 meters. The plant is monoecious and has bisexual flowers. The leaves of seedless barberry are smooth in some cultivars and serrated in others. The seedless barberry plant is thorny, and its thorns develop from modified leaves, causing the branches to widen at their base where the thorns are attached (Mehdizadeh & Nazerii, 2016). Analysis of barberry fruit morphology revealed average measurements of 7.69 mm for length and 3.32 mm for width (Akbulut et al., 2009). Research documented the maximum length of barberry fruit as 11.9 mm in a separate investigation (Talebi et al., 2020). Research indicates barberry thorn length in northern Iran ranges from 9.6 to 39.6

mm (Talebi et al., 2020). Reported seed counts per fruit in *Berberis vulgaris* L. vary between 1.36 and 1.54 (Kremer et al., 2012). Khodabandeh et al. (2022) investigated morphological parameters in 16 barberry genotypes indigenous to Iran, measuring panicle length, fruits per panicle, 1000-seed weight, fruit flesh composition, and color indices in both fresh and dry fruit. Their analysis revealed statistically significant differences across all evaluated traits.

The irregular nature of fruit production—where high-yield years alternate with low-yield years—creates a detrimental economic impact due to inconsistent annual returns (Khayyat et al., 2018a). Kramer (2012) reports that barberry yield correlates positively and significantly with yield components and physiological characteristics like chlorophyll content. Increased leaf chlorophyll amplifies photosynthetic activity and primary metabolite generation, driving improvements in growth metrics such as height and root yield. Consequently, higher root yield escalates secondary metabolite production, elevating both qualitative yield and total product yield. Rezaei et al. (2020) investigated morphological characteristics of seedless barberry across different regions. Their findings revealed significant variations in both quantitative parameters (1000-seed weight, titratable acidity) and qualitative descriptors (color, size, and shape). Researchers examined altitude-dependent shifts in fruit maturity indicators and growth patterns of seedless barberry during the harvest period. Statistically significant differences were observed in fruit number, fresh/dry weight, pH, titratable acidity, TSS, anthocyanin, and total phenols across the harvest season under varying meteorological conditions (Khayyat et al., 2018b). The researchers posited that divergences in qualitative and quantitative traits among ecotypes could be attributed to environmental heterogeneity across growth sites, encompassing climatic factors (temperature, humidity), edaphic conditions, and geographical attributes (Farrokhi et al., 2021). Multiple morphotypes of *Berberis vulgaris*, identified in Iran, appear in these photographs (Motalebipour & Pirestani, 2025) (Figure 3).

Seedy barberry (*B. integririma* L.) produces yellow flowers, with fruits reaching full development in September and attaining complete black coloration in autumn (Sarraf et al., 2019). These small-seeded (two to three per fruit), highly juicy fruits are rich in anthocyanins and antioxidants (Alemardan et al., 2013).



**Figure 3:** Phenotypic diversity in *Berberis vulgaris* fruits

The species thrives in semi-arid and humid highland habitats, with its Iranian distribution concentrated in northern and northeastern regions, including the highlands of Azerbaijan and Kerman (Tavakoli Kaghaz et al., 2021). The morphological, phenological, cytogenetic, and phytochemical profiles of seed-bearing barberry trees demonstrate plasticity in response to different environmental habitats (Iman et al., 2023). The cultivation of barberry is highly suitable for arid and semi-arid regions facing water scarcity. Since water constitutes the primary limiting factor for crop production, establishing precise water requirement metrics for barberry is indispensable (Rezaei, 2019). According to Champion and Seth (1968), most species within the *Berberis* genus exhibit shade tolerance, drought resistance, and thrive across diverse environments including open areas, woodlands, and wetlands.

Owing to lower temperatures in northwestern and northeastern Iran versus central/southern regions, barberry cultivation requires less irrigation to attain field capacity. This results from reduced moisture potential in cooler climates, which decreases the water volume needed for saturation (Rezaei & Falakh Qalheri, 2022). Characterizing the physical properties of agricultural products has consistently garnered significant attention, serving as a fundamental basis for designing harvesting machinery, developing conveying and grading systems, and optimizing processing equipment. Furthermore, these properties are essential for breeding programs aimed at developing high-yielding varieties or cultivars with specialized fruit characteristics (Nazarpour et al. 2011). It was also reported that certain physical properties of barberry specifically geometric mean diameter, thousand-seed mass, and bulk density are influenced by variations in fruit moisture content (Velayati et al., 2011). Scientific information on the physical properties of barberry varieties is crucial for optimally designing storage, transportation, and packaging equipment, as researchers have documented such properties across various agricultural products (Velayati et al., 2011). Several

*Berberis species* occurring in the Indian Himalayas are shown in the images (Figure 4) (Belwal et al., 2020).



**Figure 4:** Some *Berberis species* of the Indian Himalayan Region (IHR): (A) *B. aristata* DC., (B) *B. asiatica* Roxb. Ex DC., (C) *B. jaeschkeana* C.K. Schneid., (D) *B. lycium* Royle, (E) *B. pseudumbellata* R. Parker, (F) *B. thomsoniana* Schneid.

## 2. GENETIC DIVERSITY AND RESEARCH

Differences in morphological characteristics among barberry genotypes are attributed to genetic makeup and environmental

conditions. Studies of barberry morphology in Pakistan demonstrate that genotype and cultivation site are key determinants of growth habit, yield, and fruit nutritional quality (Ahmed et al., 2013). Research on 25 barberry genotypes demonstrated that seedless genotypes display less variation in fruit morphological traits than seeded counterparts, resulting from seedlessness (Alizadeh and Hassanpour, 2017). Wild barberry species, due to their significant diversity, represent a rich genetic resource for selecting superior cultivars for breeding programs. The existence of diversity within plant populations is of particular importance as raw material for breeding. Studying genetic diversity is crucial for germplasm evaluation, developing breeding programs, investigating species evolutionary trends, classification, and addressing numerous other issues (Mezzetti, et al., 2016). Numerous studies on barberry (*Berberis* spp.) have primarily been conducted with agricultural objectives, focusing on population selection and investigating the genetic diversity of this plant using various molecular markers. The results demonstrate extensive diversity among species within this genus. A major finding from these results indicates a very weak correlation between the variations observed in molecular markers and morphological traits. Ultimately, significant genetic diversity among populations is consistently reported (Bottini et al., 1999; Heydari et al., 2008; Safamanesh et al., 2017; Maleki-Meighani et al., 2025). Significant variations among barberry populations studied for their nutritional, medicinal, and agronomic value were observed, including quantitative and qualitative fruit characteristics, anthocyanin pigments, and physiological fruit traits (Alizadeh & Hassanpoor, 2017; Rezaei et al., 2018; Tatari et al., 2019; Khoshandam et al., 2023). Biosystematic investigation of this plant in Khorasan resulted in the identification of three genus species, two documented for the first time in Khorasan (Sodagar et al., 2012). Concurrently, taxonomic studies worldwide have described species behaviors and provided taxonomic delineations, demonstrating diverse levels of intra-genus and inter-species variation (Khan et al., 2015; Ibrahimov et al., 2020; Kremer et al., 2020). Studies conducted on various barberry species (*Berberis* spp.) in different

regions of the world indicate that mutation and natural interspecific hybridization processes have likely led to the creation of biodiversity and the emergence of numerous and diverse cultivars in this plant (Bottini et al., 2000; Ebadi et al., 2010). Research has demonstrated that barberry populations originating from northeastern Iran exhibit substantial morphological variation (Talebi et al., 2020). Analysis of 30 morphological traits across 96 seeded barberry samples collected in the Jasb region (Markazi province, Iran) revealed substantial diversity among the evaluated genotypes (Maleki-Meighani et al., 2025). Furthermore, numerous other studies investigating the quantitative and qualitative characteristics of barberry populations in various regions of the country have demonstrated extensive genetic diversity (Rezaei et al., 2020; Parvane et al., 2020).

### **3. TRADITIONAL AND ETHNOBOTANICAL USES BERBERIS SPECIES**

The medicinal uses of *Berberis vulgaris* date back over 3000 years in Traditional Chinese Medicine and over 2500 years in several other countries (Fallah Huseini et al., 2010). By the 7th century AD, barberry was widely recognized as an essential element in traditional medical practices across the Middle East (Ebrahimi-Mamaghani et al., 2009; Motalebipour & Pirestani, 2025). The primary consumption of barberries is in fresh or dried form, and they are also commonly employed in juice production (Farhadi Chitgar et al., 2017). Meeting substantial demand, Iran produces approximately 22,000 tons annually of fresh, edible, seedless barberries. These fruits are frequently utilized in cooking and jam production (Aghbashlo et al., 2008). The processing of these fruits on a large scale generates various derivatives, including beverages, syrups, candies, and confectionery products, which are especially favored in Iran. The leaves and fruits of barberry (*Berberis vulgaris*) are also processed into teas and used as food flavorings. Renowned for its nutritional properties, barberry has been a cornerstone of herbal medicine

for over three millennia. In traditional medical systems including Persian-Tibb, Indian and Traditional Chinese Medicine, diverse morphological components—fruit, roots, bark, and leaves—are extensively utilized in therapeutic formulations (Birdsall & Kelly, 1997). The flowers of this species are currently in widespread use by Tibetan communities, with Litang County in China's Sichuan Province being a primary region of application (Kang et al., 2016). Ethnobotanical studies highlight *Berberis lycium's* role in diabetes treatment. Ahmed et al. (2004) describe a root bark water extract. Rana et al. (2019) report the use of the whole plant specifically in Chamba district, Himachal Pradesh (West Himalaya, India). Furthermore, Phondani et al. (2010) document the Bhotiya tribal community of India's Central Himalayan region administering roots with water for this condition.

Barberry's medicinal use presents a clear dichotomy: traditional Persian practices involve consuming it as decoctions, infusions, or syrups for treating liver, digestive, and circulatory disorders, based on historical knowledge. In contrast, modern approaches employ scientifically calibrated extracts and capsule supplements, enabling controlled administration and focused therapeutic intervention (Tahmasebi et al., 2019; Och et al., 2024). Traditional use of *Berberis crateagina* DC. dried roots as anti-diabetic agents is documented in Turkey. The standard preparation involves a decoction or infusion of the roots, administered orally at a frequency of once or twice daily for therapeutic purposes (Durmuskahya & Öztürk, 2013). In Urmia, Iran, Bahmani et al. (2014) reported the use of boiling or steaming *Berberis integerrima* Bunge extract by local inhabitants for diabetes treatment.

#### **4. PHYTOCHEMISTRY OF *BERBERIS* SPECIES**

*Berberis vulgaris* contains a spectrum of bioactive compounds, notably the alkaloids berberamine, palmatine, berberine, oxyberberine, columbamine, isocorydine, lambertine, magniflorine, and oxycanthine,

alongside steroidal constituents (lupeol, oleanolic acid, stigmasterol, stigmasterol glucoside) (Sun et al., 2021). Berberine concentrations in the fruit of reddish-brown barberry ranged from 5.2% to 7.7%. Similarly, the root extract was found to contain 0.73 mg/mL of berberine (Yang et al., 2022). Multiple studies identify berberine (BBR) – a quaternary ammonium salt of the benzylisoquinoline alkaloid class – as the most potent bioactive compound derived from *Berberis species* (Dong et al., 2012; Lan et al., 2015; Wang et al., 2017).

The fruit of *Berberis vulgaris* is characterized by a complex array of health-beneficial bioactive compounds alongside essential macronutrients. Analysis of its proximate composition revealed 1.16% fat, 2.00% protein, 16.24% carbohydrate, 0.99% ash, and a moisture content of 79.6% (Chitgar et al., 2016). Reddish-brown barberry fruit was found to contain 75% moisture, along with 0.74% ash, 0.62% fat, 2.62% crude fiber, 0.12% protein, 9.48% total sugars, and 2.63% titratable acidity (expressed as malic acid) (Ardestani et al., 2013). Malic acid, a constituent of *Berberis vulgaris* valued for its exfoliating and skin-rejuvenating effects, is naturally present in the plant (Hanachi & Golkho, 2009). Studies report vitamin A content in reddish-brown barberry between 0.22 and 3.02 IU/kg and vitamin E between 39.19 and 776.86 IU/kg (Sayın & Balcı, 2022). Separate analyses quantify vitamin C at 11.0103 µg/100 g (Hanachi & Golkho, 2009; Naaz & Ali, 2018). Analysis reveals that *B. vulgaris* contains nutritionally significant minerals: calcium (vital for bone health and muscle function) (Cashman, 2002), potassium (regulates blood pressure, promotes cardiovascular health) (Youn & McDonough, 2009), magnesium (supports nerve function, muscle relaxation, energy metabolism) (Faryadi, 2012), iron (essential for oxygen transport and energy levels) (Duck & Connor, 2016), zinc (enhances immune function, wound healing, DNA synthesis) (Rostan et al., 2002), and copper (key for antioxidant defense and collagen production) (Lu et al., 2010). Trace elements, including Ag, Al, As, boron, and Ba, were also identified in Reddish-brown barberry fruit (Akbulut et al., 2009; Yang et al., 2022). This plant, owing to its content

of phenolic and alkaloid compounds (notably berberine), exhibits diverse therapeutic properties (Imanshahidi & Hosseinzadeh et al., 2008; Bhardwaj & Kaushik, 2012; Xu et al., 2021). Chronic diseases associated with oxidative stress may be mitigated by the antioxidant properties of *B. vulgaris*, wherein phenolic compounds play a pivotal role in free radical neutralization and redox balance. The modulation of cell signaling pathways and enhancement of endogenous defense mechanisms are attributed to flavonoids, a major phytochemical group naturally present in fruit (Crozier et al., 2009). The phenolic and flavonoid content of *B. vulgaris*, alongside its pronounced antioxidant activity, has been quantified and verified in recent investigations; population-specific differences arise from genetic and environmental determinants. By revealing substantial biochemical diversity, *B. vulgaris* demonstrates the critical need to preserve wild genetic resources while evaluating ecotypic medicinal value. Such biochemical characterization allows researchers to select elite genotypes for pharmaceutical advancement and functional food design (Yildiz et al., 2014; Garazhian et al., 2020). The strong positive correlation between fruit coloration/fresh weight and antioxidant content indicates that pigment intensity facilitates bioactive compound concentration (Motalebipour & Pirestani, 2025). Consequently, fruit pigmentation serves as a reliable predictor of antioxidant activity (Wang et al., 2022). The findings demonstrated that fruit stem length positively correlated with antioxidant activity. This correlation indicates a potential relationship between fruit structural attributes and secondary metabolite biosynthesis, suggesting that longer stems could promote antioxidant compound accumulation via specific physiological or metabolic pathways (Wang, 2009; Hu et al., 2022).

## 5. PHARMACOLOGICAL ACTIVITIES OF BERBERIS SPECIES

The therapeutic potential of *Berberis species* is tempered by safety considerations. Research indicates possible side effects such as gastrointestinal discomfort and drug interactions, posing particular concerns for pregnant women and patients with chronic illnesses. This underscores the urgent need to develop definitive clinical protocols regarding safe dosing and to elucidate the long-term consequences of consumption (Imanshahidi & Hosseinzadeh, 2008; Mahroozade et al., 2016). This plant is recognized for its efficacy in the prevention and amelioration of Alzheimer's disease symptoms, anticancer activity, seizure control, reduction of nausea, alleviation of inflammation and cutaneous itching, antipyretic effects, compensation for vitamin C deficiency, relief of muscular spasms, enhancement of bile flow, promotion of diuresis, inhibition of fungal infections, hepatoprotection, immunomodulation, sedative effects, and uterine tonicity (Salehi Surmaghi et al., 2008; Sun et al., 2021). Furthermore, barberry's effect on the glycemic index has been demonstrated. It can reduce insulin levels (Safari et al., 2020). For patients with metabolic syndrome (MS), berberine (BBR) therapy effectively managed dysglycemia and dyslipidemia, ameliorated insulin resistance, and suppressed inflammatory responses (Cao and Su, 2019). Kong et al. (2004) suggested berberine (BBR) as a candidate hypolipidemic therapy for mild mixed hyperlipidemia, noting its unique mechanism involving non-statin signaling pathways. The main plant parts utilized as sources for berberine extraction are depicted in Figure 4.



**Figure 4:** Various plant parts of (A) *Berberis asiatica* collected from the Indian Himalayan Region (IHR), including (B) roots, (C) stems, and (D) stem barks. These parts are the major sources for extracting Berberine (yellow color) from *Berberis* species

The documented antioxidant and antibacterial properties of barberry aqueous extract suggest its viability as a candidate for natural food preservation, particularly in extending product shelf-life (Aliakbarlu et al., 2014). The demonstrated biological activities of *Berberis species* solidify their standing as a resource with significant dual-purpose value, meriting attention for both functional food innovation and therapeutic advancement (Bayat et al., 2025).

It can be concluded from the above that barberry is an extremely valuable medicinal plant, playing a vital positive role in human life and deserving of great attention.

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# *Advances in Medicinal and Aromatic Plants: From Phytochemistry to Sustainable Use*

## *Chapter 7*

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# *Advances in Medicinal and Aromatic Plants: From Phytochemistry to Sustainable Use*

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## Introducing Drought-Resistant Medicinal Plants

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## INTRODUCTION

Drought stress is one of the most critical environmental constraints limiting agricultural production worldwide, particularly in arid and semi-arid regions where water scarcity and irregular precipitation prevail. Under these conditions, the cultivation of drought-tolerant crops is essential for ensuring sustainable agricultural systems. Medicinal plants represent a unique group of crops in this context, as many species possess inherent adaptive mechanisms that allow them to survive and produce economically valuable secondary metabolites under limited water availability (Turner, 1986; Farooq et al., 2009). Drought-tolerant medicinal plants are characterized by morphological, physiological, and biochemical traits such as reduced leaf area, deep or extensive root systems, enhanced water-use efficiency, osmotic adjustment, and the accumulation of protective secondary metabolites. These adaptive strategies not only support plant survival under water deficit but may also enhance the concentration of bioactive compounds responsible for medicinal properties, including phenolics, terpenoids, and essential oils (Chaves et al., 2003; Selmar & Kleinwächter, 2013).

Several medicinal plant species cultivated in arid and semi-arid regions exhibit notable drought tolerance. For instance, *Cuminum cyminum* (cumin) is widely grown in dry regions due to its short growth cycle and low water requirement, while *Thymus daenensis* demonstrates strong adaptation to harsh environments and produces essential oils rich in thymol and carvacrol. Similarly, *Crocus sativus* (saffron) is highly valued for its ability to tolerate prolonged summer drought through dormancy and its high economic return under limited irrigation conditions. In addition to native species, several medicinal plants introduced into dry regions, such as *Echinacea purpurea*, have shown moderate adaptability to water-limited environments, although their yield and phytochemical profiles are strongly influenced by drought intensity and timing. In many medicinal plants, drought stress during critical growth stages-particularly flowering and biomass accumulation-

can reduce yield, alter harvest index, and modify secondary metabolite composition (Sinclair et al., 1990; Farooq et al., 2012).

Given the increasing frequency of drought events under climate change scenarios, understanding the adaptive mechanisms and production responses of drought-tolerant medicinal plants is of growing importance. Evaluating these species provides valuable insights for optimizing cultivation practices, improving water-use efficiency, and sustaining both yield and medicinal quality in arid and semi-arid agroecosystems.

### ***Cuminum cyminum* L**

Cumin (*Cuminum cyminum* L.) is an annual plant belonging to the Apiaceae family, widely used in the pharmaceutical, food, and cosmetic industries due to its antioxidant, antibacterial, and antifungal properties. The seeds (fruits) of this plant contain 2 to 5 percent essential oil, of which 40 to 65 percent is cumin aldehyde (Pirzad et al., 2017) (Figure 1). Cumin essential oil has the ability to inhibit both the growth of fungi and the production of aflatoxins (Minoeeian Haghghi & Khosravi, 2014). The essential oil of this plant is effective at controlling storage pests, such as mealybugs, even at low concentrations (Khodadoost et al., 2012).

The economic importance of cumin in arid and semi-arid regions, characterized by water scarcity and low soil fertility, stems from its unique traits. These include its leaf shape, short stature, color, and surface coverage of plant organs; a short growing season; low water requirements; minimal competition with other crops during its growing season; strong economic viability compared to alternative crops; and its potential for export. Given that increasing yield per unit area by enhancing the technical efficiency of producers is the only appropriate way to boost production, optimizing irrigation efficiency to maximize water use is crucial (Rahimian Mashhadi, 1992). This plant is cultivated in Iran across the provinces of Khorasan, Semnan, Yazd, East

Azerbaijan, Isfahan, Sistan and Baluchestan, Kerman, Markazi, and Golestan, using both dryland and irrigated farming methods. Iran holds a significant share of the global production of this crop, ranking first or second worldwide. Green cumin is Iran's second most exported agricultural product after saffron (Kafi, 2002; Forghani & Kiani Abri, 2005; Mohammadi, 2011). The average yield of dryland cumin is up to 400 kg/ha, while the yield of irrigated cumin can reach up to 900 kg/ha (Kafi, 2002; Kafi & Keshmiri, 2011).

In Iran, cumin is primarily cultivated in the arid and semi-arid regions of the eastern, southeastern, and central provinces. Global cumin production amounts to approximately three hundred thousand tons, the area under cumin cultivation in Iran is 42,841 hectares, of which 7 hectares are dedicated to medicinal plants. The yield under dry and irrigated conditions ranges from 500 to 1,500 kilograms per hectare. Iran holds a significant share of the world's cumin production, ranking first or second globally. Cumin is one of the most important and economically valuable medicinal plants, particularly suited for cultivation in the arid and semi-arid regions of Iran, especially under conditions of water scarcity; Its economic significance in these regions, despite water shortages and low soil fertility, is attributed to several characteristics: the shape of its leaves, the compact size of its bushes, the color and surface coverage of its plant organs, a short growing season, low water requirements, minimal overlap with the growing seasons of other crops, strong economic viability compared to alternative crops, and its potential for export (Bahraminejad et al., 2024).



**Figure 1:** *Cuminum cyminum* L. seeds

Iran, with its unique climatic conditions, is home to more than 7,500 plant species—two to three times the vegetation cover of the entire European continent. It is estimated that over 750 of these species have medicinal properties; Despite this potential, the area dedicated to cultivating important medicinal plants in Iran is less than 10,000 hectares. Furthermore, the diversity of species under cultivation is limited to about 40, compared to approximately 200 species cultivated in China; Additionally, the total number of registered herbal medicines in Iran is around 100, representing less than 4% of the medicines available on the market; In contrast, this proportion exceeds 35% in European countries; In 2000, the global sale of medicinal plants exceeded \$10 billion, rising to approximately \$30 billion by 2002. However, Iran's share of this market remained very small, amounting to only \$75 million. In 2001, the total export value of Iranian medicinal plants was about \$75 million, with the most significant products being saffron, cumin, coriander, fennel, and sweet bay; The government of the Islamic Republic of Iran places special emphasis on non-oil exports to reduce dependence on the low-productivity, oil-based economy. Therefore, identifying the comparative advantage of medicinal plants, including cumin, can serve as a valuable tool for decision-making and strategic planning in foreign trade (Forghani et al., 2005).

The short growing season, low water requirements, minimal seasonal overlap, compatibility with other agricultural products, strong economic justification compared to other crops, and export potential all indicate that this plant holds a special place in the cultivation patterns of the dry and semi-dry regions of Khorasan (Rezaei et al., 2020). It is recognized as the most important medicinal plant grown domestically in Iran. This plant has significant economic value, and its export has generated foreign exchange for the country, securing its place in the cultivation patterns of arid and semi-arid regions (Sohrabiyan et al., 2018).

Cumin is one of the world's most valuable medicinal plants and is considered the most important domestic medicinal plant in Iran (Figure 2). Native to Central and South Asia, cumin is cultivated in several countries, including India, Pakistan, Turkey, Iran, Egypt, and Spain. Its significance and the area under cultivation continue to increase each year. In Iran, approximately 90 percent of cumin exports originate from the Khorasan and Semnan provinces. Research indicates that most cumin cultivation in Iran occurs in arid and semi-arid regions. In these areas, cumin often experiences drought stress from the flowering stage through seed setting, which adversely affects grain yield due to moisture deficiency (Safari et al., 2017).



**Figure 2:** General appearance of *C. cyminum*

Cumin (*Cuminum cyminum L.*) is considered one of the valuable medicinal plants in Iran (Omidbeigi, 2011). The most important compounds in the essential oil of this plant include cuminaldehyde, cymene, phellandrene, carvone, cumin alcohol, beta-pinene, and dipentene; Beyond its economic value, medicinal plants are also well-suited to organic cultivation methods; Therefore, the organic production of medicinal plants reduces the likelihood of adverse effects on their medicinal quality (Sakhavi et al., 2017).

Cumin is an important and economically valuable medicinal plant in our country, widely distributed across various regions, especially the mountainous areas of Yazd province. It is also known by other names such as "Kamoon" and originated in Egypt, it is cultivated and semi-cultivated in several other regions. Cumin holds significant medicinal and economic value and is grown in countries including Iran, where it remains the only cultivated variety (Haghiroalsadat et al., 2011).

Cumin is used in the treatment of various conditions due to its anticonvulsant, antiepileptic, stomach tonic, diuretic, antifatulent, and diaphoretic properties. It also helps accelerate the onset of menstruation. Cumin has beneficial effects in both acute and chronic catarrhal diseases, relieves bloating caused by indigestion, and aids in the treatment of abnormal feminine discharge and menstrual irregularities in young women. Additionally, cumin is beneficial for diabetic patients, promotes diuresis, and increases milk supply (Rojhan., 1982 & Trease and Evans., 1996). The essential oil content in the fruit part of cumin is approximately 5%, most of which is composed of paracetamol, alpha- and beta-pinene, cumin alcohol, cumin aldehyde, alpha- and beta-phellandrene, eugenol, perilla aldehyde, alpha-terpineol, and myrcene. Additionally, cumin contains 13% resin, 8% gum and mucilage, 7.7% oil, and 5.15% protein (Steinegger and Hansel., 2013 & Haghiri et al., 2010).

### *Thymus*

*Thymus daenensis* Celak. is an aromatic plant belonging to the mint family (Lamiaceae) and is considered a valuable medicinal plant in traditional medicine (Akbarnia & Mirza, 2008). Thyme is a perennial, multi-stemmed, cushion-shaped plant with a woody appearance and base (Figure 3). The flowering stem is erect, simple, and approximately 1.9 cm long. The leaves measure 1.44–3.1 mm in length and 4.9–5.6 mm in width, varying from linear-lanceolate to narrowly ovate and lanceolate, and are sessile (without petioles). Its flowering season occurs in June and July (Mohkami & Bidarnamani., 2023). In the essential oil of this plant, 49 active compounds have been identified. Among them, carvacrol (6%), thymol (3%), terpinene (4%), beta-bisabolene (1%), para-cymene (4.5%), and carotenoids (1%) are present in the highest concentrations (Sajjadi and Khatamsaz, 2003). Thyme plants are widely used around the world as a beverage (tea), food flavoring, spice, condiment, and herbal medicine; This plant is traditionally used as an anti-flatulent, digestive aid, antispasmodic, antitussive, and expectorant in the food,

pharmaceutical, and cosmetic industries due to its primary active ingredient, thymol; Aqueous, aqueous-alcoholic, and propylene glycol extracts of thyme are also utilized in the formulation of shampoos, creams, and ointments; The plant exhibits antifungal, antiparasitic, and antibacterial properties. Its therapeutic efficacy has been demonstrated in the treatment of asthma, dry coughs, and bronchitis. Various medicinal products, including syrups, lozenges, incense, tablets, and extracts, have been developed from this plant and approved by the Drug Supervision Unit of the Ministry of Health and Medical Education. These products include Tosian syrup, Tosgol drops, Tossion drops, Tim Arta drops, Timex syrup, Thyme syrup, and Bronchotide syrup (Pourmeidani and Mohebbikia., 2024).



**Figure 3:** General appearance of thyme

Thyme is an endemic species of Iran that is genetically diploid ( $2n=32$ ) and is distributed in the high altitudes of the Zagros Mountains and some regions of the Alborz Mountains, This species is rich in

phenolic compounds, especially thymol, and is considered a prime candidate for domestication and industrial cultivation due to its excellent adaptation to various climatic regions of Iran; For the development of varieties with high-quality active ingredients, high essential oil yield, abundant vegetative growth, suitability for mechanized harvesting, uniform germination, and strong adaptation to adverse climatic conditions, thyme is regarded as an important medicinal plant; Owing to its unique pharmacological and biological properties, it has numerous applications in traditional medicine; Additionally, because of its antifungal, antibacterial, antioxidant, antispasmodic, and other activities, it is widely used in the pharmaceutical and food industries (Hadian et al., 2016).

Daenensis thyme (*Thymus daenensis* subsp. *daenensis*) is a native species of Iran that has not yet been domesticated. This plant is found in the northwest, central, and southwestern regions of Iran, demonstrating its high adaptability to diverse soil and climatic conditions. In addition to its adaptability, this species contains higher levels of essential oil and thymol compared to other thyme species. Thyme is an aromatic herb from the mint family (Lamiaceae) known for its anti-flatulent, digestive, antispasmodic, anti-cough, expectorant, antifungal, antibacterial, and antiparasitic properties. Due to its thymol and carvacrol compounds, it is regarded as a valuable and widely used medicinal herb in the pharmaceutical, food, and cosmetic industries. This plant is native to the western Mediterranean region and southern Italy. Distribution in the World and Iran: This plant is native to and exclusively found in Iran (Golestani., 2021). The distribution of this plant in the northwest, central, and southwest regions of Iran indicates its high adaptability to diverse soil and climatic conditions (Golestani., 2020). One of the most important plant families, the Lamiaceae family, belongs to the dicotyledonous group—the largest group of herbaceous plants—within the order Lamiales and the suborder Nepetoideae, and it has a global distribution. The genus *Thymus* comprises approximately 125 species and is one of the most significant genera in this family. *Thymus* is herbaceous

perennial plant characterized by numerous thick stems with opposite, small, ovate leaves, often used as decorative borders. Its flowers resemble those of the Apiaceae family, either solitary or clustered near the leaves (Figure 4). This plant is found in regions such as Kohgiluyeh and Boyer-Ahmad (Kudu Tanda), Fars, Chaharmahal and Bakhtiari, Hamedan, Ilam, Markazi, and in Kurdistan Province, specifically in the counties of Bijar, Kamyaran, and Divandarreh, as well as along the Shudmal and Shudmal Gharbadi rivers (Ashabani et al., 2024).



**Figure 4:** Thyme flowers

### ***Crocus Sativus L.***

From the perspective of geographical and social location, the conditions prevailing in the arid and semi-arid regions of the country—such as drought, water scarcity, low soil fertility, limitations on agricultural mechanization, and reliance on traditional crop production methods—have constrained the growth of agricultural output in these areas. However, among cultivated plants, saffron has demonstrated resilience to water shortages in these regions due to its unique characteristics. Additionally, saffron offers favorable economic returns and maintains relatively stable production under these challenging

conditions; In general, the differentiation of saffron reproductive organs occurs simultaneously with a decrease in soil moisture around the corm and an increase in temperature (Figure 5). Therefore, irrigation and protective tillage during the summer can positively influence the differentiation process of reproductive organs by reducing soil temperature around the corm and maintaining soil moisture, ultimately promoting the formation of flower organs. Although farmers often view the lack of summer irrigation for saffron as an advantage compared to other crops, some researchers have found that summer irrigation and protective tillage can enhance saffron flowering and have recommended their implementation (Feizi et al., 2015).



**Figure 5:** Saffron morphology

**Introduction** As the largest producer of saffron, Iran supplies over 90% of the world's saffron; During the 1397-98 crop year, Iran emerged as the leading global producer of this crop, producing 48.404 tons of saffron on 111,000 hectares of fertile land, with an average yield of 3.620 kg per hectare. Following Iran, the most significant saffron-producing countries, in terms of production, are Morocco, India, Greece, Italy, Spain, and, in recent years, China and Afghanistan; In Iran, during the 2015-2016 crop year, Khorasan Razavi and South Khorasan provinces ranked first and second, respectively, in terms of saffron cultivation area.

Khorasan Razavi province produced 298 tons of saffron, with 87,750 hectares of its land under cultivation. During the same year, South Khorasan province contributed 63 tons. Reason: Corrected grammar, punctuation, and sentence structure for clarity and readability. Improved vocabulary and technical accuracy by specifying (Kalantari et al., 2021).

A comparison of the cultivated area and yield of saffron in Iran and Spain reveals that in Iran, the cultivated area has increased twentyfold over the past 30 years, while the yield has decreased from 15.6 to 3.4 kg/ha. This declining yield trend indicates that saffron production in Iran relies primarily on the expansion of the cultivated area. In Spain, the cultivated area of saffron was reported to be 47,000 hectares in the 1970s, approximately 50 years ago. By 1990, this area had decreased to 2,800 hectares, resulting in a reduction in production from 44 tons per year to six tons per year. It should be noted that current production is now less than one ton per year (Koocheki., 2013).

### *Echinacea purpurea*

Purple coneflower (*Echinacea purpurea*) is a member of the daisy family. It is one of the most popular herbal medicines, with 1 to 4% of the world's population using it annually (Rahimi., 2023).

*Echinacea purpurea* is a significant medicinal plant widely utilized in the pharmaceutical, cosmetic, and health industries; Although this species is not native to Iran, it has recently attracted the attention of agricultural and horticultural researchers and is now cultivated on experimental and commercial farms within the country; This herbaceous perennial features a short rhizome and a straight, somewhat branched root that ranges in color from dark brown to opaque white; In herbal medicine, *Echinacea* is renowned for its immune-stimulating properties and is currently used to prevent and treat the common cold, cough, bronchitis, lung infections, and chronic diseases associated with immune deficiency (Taghiloo et al., 2024) (Figure 6).

Improving crop performance under drought conditions has become one of the most critical challenges in plant breeding; Developing high-yielding cultivars requires selecting individuals both within and between populations that exhibit significant variation in key agronomic traits Iran, with an average annual rainfall of 220 mm, is considered one of the arid and semi-arid regions of the world. In these areas, water shortages result from low and irregular precipitation or insufficient soil water reserves (Afshar et al., 2016).

Due to Echinacea's wide adaptability to various climatic and soil conditions, the extensive cultivation of this important medicinal species has increased worldwide, especially in Iran. However, only a few studies have investigated the behavior of this valuable medicinal plant under harsh and stressful environmental conditions (Zavareh et al., 2015).



**Figure 6:** Echinacea flowers

*Echinacea*, scientifically known as *Echinacea purpurea* L., is a member of the Asteraceae family and is native to North America. Today, it is cultivated in many parts of Europe and Asia, including Iran; the most important medicinal property of this plant is its ability to enhance the immune system's capacity to fight pathogens, which has led to its use as an effective remedy for the prevention and treatment of various illnesses such as colds, flu, and infections; Echinacea products are also used as

blood purifiers, disinfectants, and sedatives (Yousefi et al., 2021). Echinacea, like other members of the chicory family, rarely causes allergic reactions and has not been reported to induce mutagenic effects in toxicological studies (Manjunathaswamy., 2015).

### ***Coriandrum sativum L.***

Coriander, scientifically known as *Coriandrum sativum L.*, is an annual herbaceous plant belonging to the Apiaceae family, originally native to the eastern Mediterranean region; Coriander is cultivated in many parts of Iran, including the provinces of Hamedan, Qazvin, Azerbaijan, Kerman, Kermanshah, Bushehr, Sistan and Baluchestan, and Yazd; Environmental stresses cause a wide range of changes, from gene expression and cellular metabolism to alterations in growth rate and crop yield. Research indicates that the quantitative and qualitative properties of many medicinal plants in the Apiaceae family are significantly affected by water deficit (Gholizadehet al., 2025). Other varieties of coriander are less sensitive to heat and day length, making them better suited for leaf production during the summer. Coriander leaves can be green or purple, depending on the variety. The coriander plant is a dioecious species belonging to the Apiaceae family. It is a member of the coriander genus (Mohebodini and Fathi., 2023) (Figure 7).



**Figure 7:** *Coriandrum sativum* leaves

It is cultivated as a spring crop in many countries and as a winter crop in some Mediterranean and Southeast Asian regions (Behmanesh and Rezaverdinejad., 2023). Although its original origin is attributed to Southwest Asia and the Mediterranean regions, coriander is now found and cultivated worldwide; Coriander seeds contain essential oils rich in beneficial phytonutrients, including carotenoids, geraniol, limonene, borneol, camphor, oleic acid, and linalool; The flavonoids in coriander include quercetin, kaempferol, rhamnetin, and apigenin (Figure 8). Additionally, coriander contains polyphenolic compounds such as caffeic acid and chlorogenic acid. The major component of coriander oil is linalool, which constitutes more than two-thirds of the oil; Coriander has been widely used in traditional medicine to treat digestive disorders, respiratory and urinary system diseases, anxiety and insomnia, allergies, bloody diarrhea, burns, cough, cystitis, dizziness, edema, hay fever, headaches, hemorrhoids, boils, urinary tract infections, and vomiting; Additionally, pharmacological studies have demonstrated its effects in lowering blood sugar and lipid levels, as well as its antihypertensive, anti-inflammatory, anti-anxiety, cognitive-enhancing, antibacterial, and antifungal properties (Amani-Geshnigani and Shabani., 2013).



**Figure 8:** *Coriandrum sativum* seeds

The leaves of the coriander plant are primarily used in nutrition, while its fresh stems are commonly added to salads and soups. The seeds are utilized in the food industry as a seasoning, and also play important

roles in the pharmaceutical, cosmetic, and health industries. Additionally, coriander seed oil is used in both the food and pharmaceutical sectors (Lotfollahi et al., 2021).

Coriander is recommended for the treatment of various diseases, including typhoid fever and other general ailments. The shoots of this plant are rich in active compounds, with 1-decanol being the primary component. It exhibits antifungal, anticancer, anti-aging, and numerous other therapeutic properties (Nadeem et al., 2013).

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