



## Phytochemical Evaluation and Chemotypic Variations in Improved Cultivars and a Native Landrace of Chamomile

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### ABSTRACT

Chamomile (*Matricaria chamomilla* L.) is a widely recognized medicinal plant, valued for its essential oil (EO) and flavonoid content, which contribute to its therapeutic properties. The demand for high-quality chamomile has led to the development and cultivation of improved cultivars worldwide. However, the adaptation and yield of these cultivars under different climatic conditions, such as those found in Iran, remain underexplored. This investigation was conducted to evaluate the quality of improved cultivars of chamomile cultivated in the climatic conditions of Iran and compare them with a native landrace. Two cultivars, 'Zloty Lan' and 'Lutea' were sourced from Germany and cultivated alongside a native landrace in Abadeh city, Fars province, Iran. Agronomic practices, including irrigation and weeding, were standardized across all test groups. Fully open flowers were harvested for analysis. The study focused on essential oil content and composition, as well as total flavonoid content. Both improved cultivars met European Pharmacopoeia standards for EO content, with 'Zloty Lan' exhibiting nearly double the EO content of the other samples. The native landrace was rich in  $\alpha$ -Bisabolol oxide A (34.5%) and Chamazulene (25.6%), while 'Zloty Lan' and 'Lutea' also demonstrated significantly higher flavonoid content (87% and 68%, respectively). The results suggest that the climatic conditions of southern Iran, particularly Abadeh, are conducive to cultivating chamomile flowers that meet international quality standards (ISO 19332). The findings highlight the potential of this region for producing high-quality chamomile, with the 'Zloty Lan' cultivar showing superior performance in essential oil and flavonoid content.

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### 1. Introduction

Iran, a country with 11 of the 13 global climate classifications, boasts a remarkably diverse flora, including a wide variety of medicinal plants such as chamomile (*Matricaria chamomilla* L.). The presence of these species in the wild suggests strong potential for their commercial cultivation, which could be economically beneficial. However, to optimize such cultivation, preliminary studies are necessary to identify suitable cultivars that can thrive in specific climatic conditions.

The importance of selecting the right cultivar for maximizing yield and ensuring the optimal quality of

biologically active compounds has been emphasized in previous studies (Saebi *et al.*, 2024). Chamomile is particularly valued in traditional medicine and the pharmaceutical industry for its essential oil (EO) and flavonoid content, which are known for their anti-inflammatory, antispasmodic, and wound-healing properties (Singh *et al.*, 2011). The therapeutic effects of chamomile are largely attributed to its rich flavonoid and EO profiles. Numerous flavonoids, including Apigenin and its derivatives, Quercetin, Luteolin, and others, have been identified in chamomile (Das, 2014). Additionally, its EO contains significant compounds such as chamazulene,  $\alpha$ -bisabolol, and  $\alpha$ -bisabolol

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oxides A and B (Azizi, 2007; Ebadi et al., 2009). Over 50 compounds have been identified in chamomile EO from various regions, underscoring its chemical diversity (Orav et al., 2010).

To meet the growing demand from food, cosmetics, and pharmaceutical industries for chamomile extracts and EOs, researchers have developed and introduced various cultivars. For example, the Polish tetraploid cultivar 'Złoty Łan' was introduced in 1972, significantly enhancing the availability of uniform raw materials for pharmaceutical use (Seidler-Lozykowska, 2006). Similarly, the 'Lutea' cultivar, introduced in Slovakia in 1995, is well-suited for dry climates and produces EO with 1.0-1.2% content in the drug, containing 24% chamazulene and 45-50%  $\alpha$ -bisabolol (Oravec et al., 2006). Numerous other cultivars have been commercially introduced in various countries, each adapted to specific ecological and climatic conditions (Otto et al., 2017). To produce high-quality raw materials for the herbal pharmaceutical industry, it is essential to evaluate different cultivars of medicinal plants, considering both imported varieties and local landraces. This approach helps assess the impact of climate on the active ingredients of these plants (Azizi, 2007). Given the favorable ecological conditions in southern Iran for chamomile cultivation and the presence of native landraces, this study compares the phytochemical characteristics of two improved cultivars, 'Złoty Łan' and 'Lutea' with a local landrace.

## 2. Materials and methods

This research was conducted in 2015. Field cultivation was carried out in Abadeh city, Fars province, Iran (Latitude: 31° 09' 38.88" N, Longitude: 52° 39' 2.16" E, Elevation above sea level: 1890 m). The laboratory studies were conducted at Tarbiat Modares University and the Medicinal Plants and Drugs Research Institute at Shahid Beheshti University, Tehran, Iran.

### 2.1. Chemical and reagents

Water was obtained from an ultra-pure water purification system (Water Purification System - New Human Power). Methanol (HPLC grade,  $\geq 99.8\%$ ), Sodium sulphate (anhydrous  $\text{Na}_2\text{SO}_4$ ) and Sodium hydroxide (99%) were purchased from Merck (Germany). Aluminum chloride (anhydrous, reagent grade,  $\geq 99.8\%$ ) and Quercetin ( $\geq 95\%$ ) were purchased

from Sigma Aldrich. Sodium nitrite (reagent grade,  $>98\%$ ) was purchased from Fluka.

### 2.2. Plant materials

The seeds of 'Złoty Łan' and 'Lutea' cultivars were purchased from Pharmasaat GMBH, Germany and cultivated together with a native landrace in Abadeh city, Fars province, Iran. At the beginning of spring, the seeds were planted in the same conditions in 9 square meters plots of field with three replications and all the agronomic practices such as irrigation intervals (every 7 days), weeding and fertilization were the same for them. The flowers were harvested 85 days after seed planting when they were fully opened (ray or ligulate florets were completely horizontal).

### 2.3. Essential oil extraction

For determination of the EO content, 50 g of shade-dried flower of each chamomile cultivar were hydro-distilled in Clevenger apparatus in 500 mL water for three hours. The EO was dehumidified with anhydrous  $\text{Na}_2\text{SO}_4$  and the amount of EO was determined volumetrically (v/w).

### 2.4. Identification of chemical composition of the essential oils with GC-MS

For this purpose, analysis was performed on an Agilent GC-MS system (Agilent Technologies-5975C-MS, 7890A-GC) equipped with an HP-5MS capillary column (0.25 mm  $\times$  30 m, 0.25- $\mu\text{m}$  film thickness, Agilent) in Medicinal Plants and Drugs Research Institute, Shahid Beheshti University, Tehran, Iran. The operating conditions were as follows: the oven temperature was programmed from 60 to 210°C at the rate of 3°C/min then increased to 240°C at the rate of 20°C/min and the final temperature was kept for 8.5 min; run time was 60 min; ion-source temperature was 230°C; interface line and Injector temperature was 280°C; Split ratio was 1:50; injection volume was 1  $\mu\text{L}$ ; carrier gas was helium at flow rate of 1ml/min; mass range was 50-480 and the electron ionization energy was 70 eV. The identification of the EO components were confirmed by comparison of their KI and their mass spectra with Adams, NIST and Wiley libraries.

### 2.5. Plant extraction and total flavonoid analysis

One gram of each chamomile flower sample was combined with 5 mL of 70% methanol in water and

subjected to extraction for 24 hours in a shaker set at 200 rpm, with the samples wrapped in aluminum foil. The extract solution samples were centrifuged for 15 minutes at 6000 rpm and the clear extract on the sample was stored in a freezer at -20 for further analysis.

For determination of total flavonoids, 300 microliters of 5% sodium nitrate solution (5% NaNO<sub>2</sub>) were added to one milliliter of the prepared extract, 5 minutes later, 600 microliters of 10% Aluminum chloride (AlCl<sub>3</sub>) solution were added (yellow color), 6 minutes later 2 ml of sodium (NaOH) 1 M was added (production of peach color) and the volume was brought to 10 ml by adding distilled water. The obtained solution was vortexed and kept in the dark for 40 minutes. Then, the absorbance of the solution was read by an Agilent-UV-Vis- Cary 100 device at a wavelength of 410 nm against the blank solution (one milliliter of methanol along with other substances) (Shraim et al., 2021). The total flavonoids in the extract samples were quantified using a calibration curve prepared from the flavonoid quercetin as a standard in concentrations of 25, 50, 100, 200 and 400 ppm at wavelengths of 410 nm.

### 2.6. Data analysis

The experiments were conducted on three treatments, comprising two improved cultivars ('Złoty Łan' and 'Lutea') and a native landrace, arranged in a randomized complete block design with three replications. The analysis of variance and mean comparisons of data were performed using Duncan's multiple range test at 5% significance level, and the statistical analysis of the results was done by SAS 9.1 software.

## 3. Results and discussion

### 3.1. Essential oil content

The EO content of different chamomile samples showed that native chamomile contained 11.1 ml/kg of dry weight. EO in imported varieties of 'Lutea' and 'Złoty Łan' was 10.4 and 22.7 ml/kg, respectively (Table 1). 'Złoty Łan' had the highest amount of EO with 22.7 ml/kg. The 'Lutea' cultivar and the native chamomile cultivar had almost the same amount of EO. According to the European Pharmacopoeia standard, chamomile should contain at least 4 ml/kg dry weight. All three chamomile cultivars cultivated in this study

have met the desired standard of the British Pharmacopoeia in terms of EO content.

**Table 1. EO content of chamomile samples.**

Sample	EO (%)
Native landrace	1
'Lutea'	0.9
'Złoty Łan'	2
European pharmacopoeia	Minimum 0.4

### 3.2. Essential oil composition

The gas chromatography with mass spectrometer (GC-MS) analysis of EOs identified more than 40 mono and sesquiterpene compounds from 3 cultivated chamomile varieties (Table 2).

**Table 2. EO compositions of three cultivated chamomile varieties.**

No.	Compounds	RI	'Lutea'	'Złoty Łan'	Native landrace
1	Artemisia triene	923.5	0.98	-	-
2	Sabinene	970.4	0.10	-	0.70
3	6-methyl-5-Hepten-2-one	983.7	0.28	0.04	0.03
4	p-Cymene	1022	0.52	0.10	0.22
5	1,8-Cineole	1028	0.61	0.10	0.38
6	(Z)-β-Ocimene	1034	0.16	0.14	0.23
7	(E)-β-Ocimene	1044	0.73	0.79	1.26
8	g-Terpinene	1055	0.50	0.20	0.44
9	Artemisia ketone	1057	1.35	0.82	1.47
10	Artemisia alcohol	1080	0.53	0.37	0.38
11	Linalool	1097	0.40	0.11	0.10
12	Borneol	1162	0.30	-	-
13	Artemisyl acetate	1169	-	0.20	-
14	α-Terpineol	1187	0.20	0.07	0.15
15	Methyl chavicol	1195	1.05	0.89	0.28
16	Carvone	1240	0.34	0.11	0.04
17	Thymol	1289	0.30	0.78	0.33
18	Carvacrol	1299	0.25	0.54	0.68
19	delta-Elemene	1333	0.06	0.20	0.07
20	Decanoic acid	1366	0.23	-	-
21	β-Elemene	1388	0.11	0.18	0.21
22	(E)-Caryophyllene	1415	0.12	0.13	0.16
23	(E)-β-Farnesene	1454	4.41	3.87	3.39
24	dehydro-Sesquicineole	1465	0.31	0.20	0.06
25	Germacrene D	1476	-	0.76	0.93
26	gamma-Murolene	1476	0.56	-	-
27	(+)-α-Curcumene	1479	0.23	-	-
28	Bicyclgermacrene	1491	0.63	1.43	0.58
29	(E, E)-α-Farnesene	1499	0.87	0.24	0.14
30	β-Bisabolene	1505	3.17	-	-
31	d-Cadinene	1519	0.33	0.17	0.14
32	(E)-γ-Bisabolene	1539	3.07	-	-
33	(E)-Nerolidol	1560	1.33	1.80	0.99
34	Spathulenol	1572	0.95	0.77	0.58
35	α-Bisabolol oxide B	1651	18.20	24.83	6.56
36	Bisabolene oxide A	1679	7.13	12.40	10.97
37	α-Bisabolol	1681	16.94	-	-
38	Chamazulene	1725	18.34	23.84	25.66
39	α-Bisabolol oxide A	1741	9.04	15.98	34.49
40	En-yn-dicycloether	1871	2.44	4.93	6.20
Total			97.42	97.21	97.93

In the 'Lutea' cultivar, 39 compounds represented more than 97% of the EO. The main ingredients of 'Lutea' EO have included Chamazulene (18.3%)  $\alpha$ -Bisabolol oxide B (18.2%),  $\alpha$ -Bisabolene oxide A (17.1%),  $\alpha$ -Bisabolol (16.9%),  $\alpha$ -Bisabolol oxide A (9%), (E)- $\beta$ -Farnesene (4.4%),  $\beta$ -Bisabolene (3.2%), (E)- $\gamma$ -Bisabolene (3%) and En-yn-dicycloether (2.4%). In the EO of 'Zloty Lan', 33 compounds have been identified, which represented more than 97% of the EO. Its main ingredients were included (E)- $\beta$ -Farnesene (3.8%),  $\alpha$ -Bisabolol oxide B (24.8%),  $\alpha$ -Bisabolene oxide A (12.4%), Chamazulene (23.8%),  $\alpha$ -Bisabolol oxide A (16%) and En-in-dicycloether (4.9%).

In the EO of native landrace, 32 compounds have been identified, which represented more than 97% of the EO. The main components of the EO of this variety were as follows:  $\alpha$ -Bisabolol oxide A (34.5%), Chamazulene (25.6%),  $\alpha$ -Bisabolol oxide B (6.5%),  $\alpha$ -Bisabolene oxide A (11%), En-in-dicycloether (6.2%) and (E)- $\beta$ -Farnesene (3.4%). The main EO components of these samples belonged to various chemical classes, including Oxygenated monoterpenes (Alcohols, Ethers, Ketones, Esters, and Acids), Monoterpene hydrocarbons, Sesquiterpenes hydrocarbons and phenolic terpenoids.

Oxygenated monoterpenes included: Alcohols): Yomogi alcohol, Artemisia alcohol, Linalool, Borneol,  $\alpha$ -Terpineol, (E)-Nerolidol, Spathulenol,  $\alpha$ -Bisabolol,  $\alpha$ -Bisabolol oxide B,  $\alpha$ -Bisabolol oxide A. Ethers): 1,8-Cineole, dihydro-Sesquicineole, En-yn-dicycloether. Ketones): 6-methyl-5-Hepten-2-one, Artemisia ketone, Carvone, Bisabolene oxide A. Esters): Artemisyl acetate. Acids): Decanoic acid.

Monoterpene hydrocarbons included: Artemisia triene, Sabinene, p-Cymene, (Z)- $\beta$ -Ocimene, (E)- $\beta$ -Ocimene, and g-Terpinene.

Sesquiterpenes hydrocarbons included: delta-Elementene,  $\beta$ -Elementene, (E)-Caryophyllene, (E)- $\beta$ -Farnesene, Germacrene D, gama-Muurolene, (+)- $\alpha$ -Curcumene, Bicyclgermacrene, (E, E)- $\alpha$ -Farnesene,  $\beta$ -Bisabolene, d-Cadinene, (E)- $\gamma$ -Bisabolene, Chamazulene.

Phenolic terpenoids included: Methyl chavicol, Thymol, and Carvacrol. Chemical classes of EO components of the samples were according to Fig. 1. Oxygenated monoterpenes and sesquiterpenes hydrocarbons were the largest groups of EO compounds in these samples.

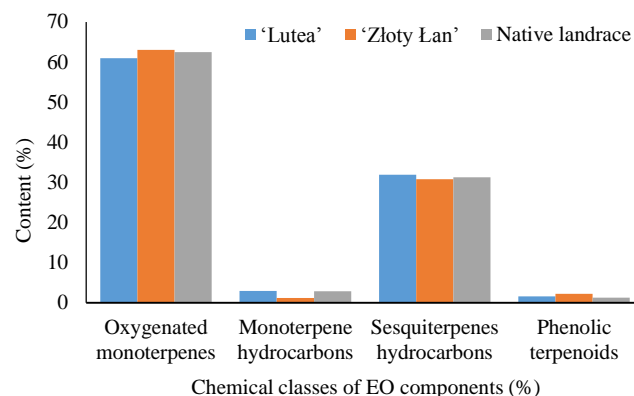


Figure 1. Chemical classes of EO components (%) of chamomile varieties.

### 3.3. Total flavonoid content

Total flavonoid analysis of cultivated chamomile varieties was performed and the results are compared in Fig. 2. The comparison showed that the samples of the 'Zloty Lan' and 'Lutea' cultivars had more flavonoid content compared to the native sample, so these two cultivars had 87% and 68% more flavonoid content, respectively.

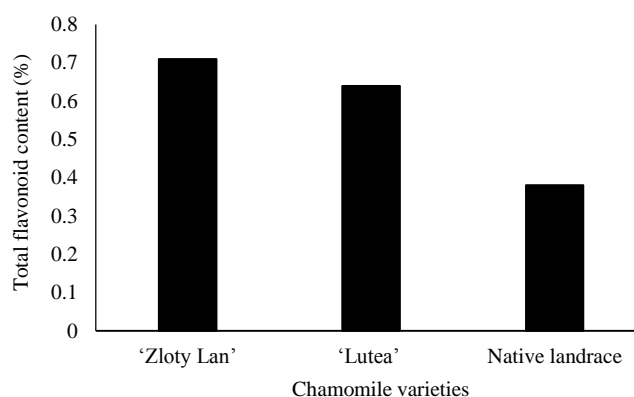


Figure 2. Total flavonoid content (%) of three chamomile varieties.

The first documented study on variations in essential oil (EO) content in chamomile dates back to 1958 when Debska in Poland reported that dried chamomile flowers contained between 0.46% and 0.67% EO. Since then, numerous studies have corroborated these findings, demonstrating variations in EO content across different chamomile samples. For instance, a study analyzing seven chamomile varieties sourced from Argentina, Hungary, Egypt, Germany, Czechoslovakia, the Netherlands, and Yugoslavia—all cultivated in Ozzano, Bologna—reported the following EO contents: Argentina (0.36%), Hungary (0.31%), Egypt (0.22%), Germany (0.22%), Czechoslovakia (0.29%), Netherlands (0.22%), and Yugoslavia (0.39%) (Das, 2014). Additionally, Jalali et al. (2008)

reported EO yields in Iranian chamomile samples ranging from 0.68% to 2.85%. In comparison, the chamomile samples cultivated in the present study demonstrated higher EO yields, ranging from 0.94% to 2.0%, which exceeds many of the previously reported values. This suggests that the environmental conditions and cultivation practices employed in this study may contribute to enhanced EO production.

Pharmacopoeia standards and various scientific studies have classified chamomile EOs into distinct chemotypes based on their dominant compounds. The most common chemotypes include those characterized by  $\alpha$ -bisabolol oxide B,  $\alpha$ -bisabolol oxide A,  $\alpha$ -bisabolol, bisabolene oxide A, and chamazulene. The EOs of these chemotypes are typically blue, due to the presence of chamazulene. However, there also exists a chamazulene-free chemotype, which is distinguished by its yellow color (Das, 2014).

According to European Pharmacopoeia, there are two types of chamomiles EO, including EO rich in  $\alpha$ -bisabolol oxides and EO rich in  $\alpha$ -bisabolol, for which the European Pharmacopoeia standard is according to the Table 3.

**Table 3. Europe Pharmacopoeia standard**

Compounds	Chamomile oil rich in $\alpha$ -bisabolol oxides (%)	Chamomile oil rich in $\alpha$ -bisabolol (%)
$\alpha$ -Bisabolol oxides	29-81	-
$\alpha$ -Bisabolol	-	10-65
Chamazulene	$\leq 1$	$\leq 1$
Total of $\alpha$ -Bisabolol oxides and $\alpha$ -Bisabolol	-	$\leq 20$

According to Table 3, the EOs of native landrace chamomile in this study and 'Złoty Łan' samples were rich in  $\alpha$ -bisabolol oxides and the EO of 'Lutea' sample was one of the EOs rich in  $\alpha$ -bisabolol, and the composition of all three EOs is in accordance with the British Pharmacopoeia standard. Schilcher (1973) classified chamomile EO into four types of chemotypes based on its most abundant compounds: Type A:  $\alpha$ -bisabolol oxide B >  $\alpha$ -bisabolol oxide A >  $\alpha$ -bisabolol, chamazulene, Type B:  $\alpha$ -bisabolol oxide A >  $\alpha$ -bisabolol oxide B >  $\alpha$ -bisabolol, chamazulene, Type C:  $\alpha$ -bisabolol >  $\alpha$ -bisabolol oxide B >  $\alpha$ -bisabolol oxide A, chamazulene, and Type D:  $\alpha$ -bisabolol oxide B  $\equiv$   $\alpha$ -bisabolol oxide A  $\equiv$   $\alpha$ -bisabolol, chamazulene.

In this study, the EO sample of native landrace contained  $\alpha$ -Bisabolol oxide A (34.50%),  $\alpha$ -Bisabolol oxide B (6.56%) and Chamazulene (25.66%), which is

the Type B chemotype in Lawrence's classification (Lawrence, 1994). 'Lutea' cultivar EO contained  $\alpha$ -Bisabolol oxide B (18.20%),  $\alpha$ -Bisabolol oxide A (9.04%),  $\alpha$ -Bisabolol (16.94%) and Chamazulene (18.34%), which is in Type A chemotype. 'Złoty Łan' chamomile EO contained  $\alpha$ -Bisabolol oxide B (24.83%),  $\alpha$ -Bisabolol oxide A (15.98%) and Chamazulene (23.84%), which is classified as Type A chemotype. Another study defined chamomile chemotypes in three groups: 1) absence of chamazulene and rich in bisabolene oxide, 2) rich in  $\alpha$ -bisabolol oxides and chamazulene, 3) rich in  $\alpha$ -bisabolol and chamazulene. The third type is shown as the most appropriate type (Das, 2014). In another study, the EO components of seven varieties of chamomile from various countries (cultivated in Bologna) were as follows (Das, 2014):

Argentina:  $\alpha$ -Bisabolol oxide A (11.5%),  $\alpha$ -Bisabolol oxide B (50.4%),  $\alpha$ -Bisabolol (6.8%), Chamazulene (10.3%) and  $\alpha$ -Bisabolene oxide A (1.7%), Hungary:  $\alpha$ -Bisabolol oxide A (35.0%),  $\alpha$ -Bisabolol oxide B (8.4%),  $\alpha$ -Bisabolol (5.9%), Chamazulene (19.7%) and  $\alpha$ -Bisabolene oxide A (5.8%), Egypt:  $\alpha$ -Bisabolol oxide A (53.6%),  $\alpha$ -Bisabolol oxide B (9.5%),  $\alpha$ -Bisabolol (2.9%), Chamazulene (2.7%) and  $\alpha$ -Bisabolene oxide A (8.5%), Germany:  $\alpha$ -Bisabolol oxide A (44.7%),  $\alpha$ -Bisabolol oxide B (13.5%),  $\alpha$ -Bisabolol (1.6%), Chamazulene (7.6%) and  $\alpha$ -Bisabolene oxide A (12.0%), Czechoslovakia:  $\alpha$ -Bisabolol oxide A (2.5%),  $\alpha$ -Bisabolol oxide B (3.9%),  $\alpha$ -Bisabolol (58.5%), Chamazulene (17.5%) and  $\alpha$ -Bisabolene oxide A (0.4%), Holland:  $\alpha$ -Bisabolol oxide A (51.6%),  $\alpha$ -Bisabolol oxide B (10.7%),  $\alpha$ -Bisabolol (2.8%), Chamazulene (3.5%) and  $\alpha$ -Bisabolene oxide A (8.5%), and Yugoslavia:  $\alpha$ -Bisabolol oxide A (28.6%),  $\alpha$ -Bisabolol oxide B (21.3%),  $\alpha$ -Bisabolol (1.7%), Chamazulene (15.8%) and  $\alpha$ -Bisabolene oxide A (12.8%).

Raal et al. (2003) conducted a comprehensive comparative study of chamomile ecotypes from Estonia, Hungary, France, the United Kingdom, and Belgium, grown in Estonia. The study revealed significant variations in the essential oil (EO) compositions among these samples. Notably, the Hungarian chamomile exhibited the highest concentration of  $\alpha$ -bisabolol (24%), while the British sample had the highest levels of  $\alpha$ -bisabolol oxide B

(25%) and chamazulene (14%). The Estonian, Belgian, and French samples were particularly rich in  $\alpha$ -bisabolol oxide A, with concentrations ranging from 43% to 55%. In Latvia, a study by Mežaka et al. (2020) focused on the 'Lutea' and 'Zloty Lan' cultivars, both of which were found to be rich in  $\alpha$ -bisabolol oxide A. Similarly, Nurzyńska-Weirdak (2011) compared the EO components of the 'Zloty Lan' cultivar with a wild-type population in Poland, reporting that the 'Zloty Lan' cultivar had high levels of chamazulene, while the wild population was dominated by  $\alpha$ -bisabolol oxide A. Neither sample contained  $\alpha$ -bisabolol.

In Greece, Tsivelika et al. (2018) evaluated five commercial chamomile cultivars, including 'Banatska', 'Bodegold', 'Lutea', 'Zloty Lan', and 'Goral', along with two Indigenous varieties. Their findings indicated that 'Lutea', 'Zloty Lan', and 'Goral' had higher EO yields (0.6% to 0.7%) compared to other samples. Specifically, the 'Lutea' cultivar was rich in  $\alpha$ -bisabolol (40.3%), making it a chemotype rich in  $\alpha$ -bisabolol, while 'Zloty Lan' was abundant in  $\alpha$ -bisabolol oxide B. Acimovic et al. (2018) analyzed EO content in three tetraploid chamomile cultivars ('Zloty Lan', 'Manzana', and 'Lutea') grown in Serbia. The EO yields ranged from 0.43% to 0.48%, with the main EO compositions varying among the cultivars. For instance, 'Zloty Lan' was characterized by high levels of *trans*- $\beta$ -farnesene (39.0%),  $\alpha$ -bisabolol oxides B (11.9%) and A (9.6%), while 'Manzana' had a high  $\alpha$ -bisabolol content (34.8%).

In Poland, Wesolowska et al. (2015) reported the EO composition of the 'Zloty Lan' cultivar, highlighting the presence of  $\alpha$ -bisabolol oxide B (16.92%–18.29%),  $\alpha$ -bisabolol oxide A (16.33%–17.84%), and chamazulene (14.92%–15.42%). Šalamon (2004) conducted a comparative study of 42 wild chamomile populations from eastern Slovakia, finding that the 'Goral' cultivar was rich in  $\alpha$ -bisabolol (30%), while the 'Lutea' cultivar had the highest content of  $\alpha$ -bisabolol (48%). Jalali et al. (2008) studied the quality of EOs from various chamomile samples in Iran, identifying key compounds such as  $\alpha$ -bisabolol oxide A (3.3%–15.4%), chamazulene (2.6%–10.6%), and bisabolene oxide A (63.4%–92.4%). Furthermore, Šalamon et al. (2010) reported significant chemotypic diversity among 20 native chamomile populations in Iran, with some populations rich in  $\alpha$ -bisabolol (Type C) and others in  $\alpha$ -bisabolol oxide A (Type B). Homami et al. (2016)

analyzed the EO composition of native chamomile from Isfahan, Iran, identifying  $\alpha$ -bisabolol oxide A (62.1%) as the dominant component. Ghanavati et al. (2010) also studied native chamomile samples from central and southern Iran, classifying them under Type B and Type C chemotypes based on their EO composition (Table 4).

**Table 4. Lawrence types of chamomile EO.**

Compounds	Type A	Type B	Type C	Type D
$\alpha$ -Bisabolol oxide B	22.43-58.85%	5.27-8.79%	4.37-15.41%	10.23-24.2%
$\alpha$ -Bisabolol oxide A	4.74-15.68%	31.07-52.25%	2.13-18.5%	9.62-25.83%
$\alpha$ -Bisabolol	4.37-15.41%	8.81-12.92%	24.18-77.21%	8.49-19.58%
Chamazulene	2.7-17.69%	5.4-7.95%	1.45-14.9%	1.91-7.89%

The results summarized in Table 5 demonstrate that the EO composition of chamomile is highly influenced by the climatic conditions of the growing regions. Commercial cultivars such as 'Zloty Lan' and 'Lutea' exhibited different EO profiles depending on the region of cultivation. In contrast, chamomile EO samples from Iran were predominantly rich in  $\alpha$ -bisabolol oxide A and  $\alpha$ -bisabolol (Types B and C), with no reports of  $\alpha$ -bisabolol oxide B-rich EO (Type A).

**Table 5. Comparison of chemotype of 'Zloty Lan', 'Lutea' and native samples of Iran in different studies.**

No.	Location	Seed origin	Chemotype
1	Latvia	'Lutea'	Type B
2	Greece	'Lutea'	Type C
3	Slovakia	'Lutea'	Type C
4	Poland	'Zloty Lan'	Type A
5	Latvia	'Zloty Lan'	Type B
6	Greece	'Zloty Lan'	Type A
7	Iran	Native	Type C
8	Iran	Native	Type B

According to the ISO 19332 standard, chamomile EO is classified into Hungarian and Egyptian types based on its composition. Table 6 compares the composition of chamomile EO in the ISO 19332 standard with the three cultivated samples from this study. The native chamomile variety from this study aligns with the Hungarian type in terms of  $\alpha$ -bisabolol oxide A and B content, but it contains higher levels of bisabolene oxide A and chamazulene, and lower levels of  $\alpha$ -bisabolol, with no detectable *trans*- $\beta$ -farnesene. The 'Lutea' cultivar's EO is also similar to the Hungarian type, though with elevated bisabolene oxide

A and reduced *trans*- $\beta$ -farnesene. For the 'Zloty Lan' EO, only  $\alpha$ -bisabolol oxide A was within the ISO standard range, with other components exceeding the standard levels.

**Table 6. Comparison of chamomile EO composition in ISO 19332 standard and cultivated varieties in this study.**

Component	Egyptian type (%)	Hungarian type (%)	Native landrace (%)	'Lutea' (%)	'Zloty Lan' (%)
<i>trans</i> - $\beta$ -Farnesene	15-35	20-51	3.39	4.41	3.87
$\alpha$ -Bisabolol oxide B	2-8	2-21	6.56	18.20	24.83
Bisabolene oxide A	2-6.5	1-4	10.97	7.13	12.40
$\alpha$ -Bisabolol	1-10	15-40	-	16.94	-
Chamazulene	2-5	5-22	25.66	18.34	23.84
$\alpha$ -Bisabolol oxide A	35-50	2-27	10.97	7.13	12.40

In terms of flavonoid content, [Tsivelika et al. \(2021\)](#) assessed the total flavonoid content in 'Banatska', 'Goral', and 'Lutea' cultivars, reporting 0.53%, 0.57%, and 0.51% of dry weight, respectively, while native landraces in Greece had higher flavonoid content (0.61% and 0.74%). [Haghi et al. \(2014\)](#) reported a flavonoid content of 0.82% as quercetin in native chamomile from Kashan, Iran, while [El Abdali et al. \(2018\)](#) found a higher flavonoid content (1.1% as quercetin) in a Moroccan chamomile sample. [Dehghani Mashkani et al. \(2011\)](#) reported 0.37%–0.44% total flavonoid content as hyperoside in cultivated chamomile from Karaj, Iran. [Golzadeh et al. \(2012\)](#) observed a flavonoid range of 0.3%–0.6% in different cultivated chamomile samples. In this study, the total flavonoid content of the 'Zloty Lan' and 'Lutea' cultivars was 0.71% and 0.64% quercetin, respectively, which exceeded that of the native sample (0.38%). A review of various studies indicates that total flavonoid content in chamomile samples ranges from 0.3% to 1.1%, aligning with the findings of this research.

#### 4. Conclusion

The findings of this research demonstrate that the southern region of Iran, specifically Abadeh, holds significant potential for producing high-quality chamomile flowers that meet international standards (ISO 19332). However, to optimize the quality of the essential oil (EO) or flavonoid content based on specific commercial needs, the selection of the appropriate cultivar is crucial. For instance, if the objective is to produce chamomile EO with a high concentration of  $\alpha$ -Bisabolol oxide B (Type A

chemotype), then the 'Zloty Lan' and 'Lutea' cultivars should be favored. Conversely, if EO with a higher  $\alpha$ -Bisabolol oxide A content (Type B chemotype) is required, the cultivation of the native landrace is recommended. Additionally, for higher flavonoid content, the 'Zloty Lan' and 'Lutea' cultivars are more suitable. These results could significantly influence future agricultural practices in regions similar to Abadeh by guiding farmers on which chamomile cultivars to plant based on the desired chemical profile. This could lead to more targeted cultivation strategies, maximizing both yield and quality and thus enhancing the economic viability of chamomile farming in these areas. Commercially, this differentiation allows producers to meet specific market demands for chamomile products with particular EO profiles or flavonoid content, potentially opening up new markets and increasing profitability.

However, several challenges may arise in commercializing these findings and integrating them into existing agricultural practices. There may be logistical and economic challenges in scaling up production, particularly for smallholder farmers who may lack the resources or infrastructure to implement these changes effectively. Ensuring that farmers are educated on the benefits and best practices for cultivating these specific chamomile cultivars will be essential to overcoming these barriers. Collaboration between agricultural researchers, extension services, and local farmers will be key to successfully integrating these findings into commercial practices.

#### Conflict of interests

The authors have no conflict of interest to declare.

#### Ethics approval and consent to participate

No humans or animals were used in the present research. The authors have adhered to ethical standards, including avoiding plagiarism, data fabrication, and double publication.

#### Consent for publications

All co-authors have read and agree with the contents of the manuscript.

#### Availability of data and material

All the data are embedded in the manuscript.

## Authors' contributions

All authors had an equal role in study design, work, statistical analysis and manuscript writing.

## Informed consent

The authors declare not to use any patients in this research.

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## References

- Acimovic M., Stankovic J., Cvetkovic M., Kiproviski B., Todosijevec M. 2018. Essential oil quality of tetraploid chamomile cultivars grown in Serbia. *Journal of Essential Oil Bearing Plants* 21(1): 15-22. <https://doi.org/10.1080/0972060X.2017.1401962>
- Azizi M. 2007. Study of four improved cultivars of *Matricaria chamomilla* L. in climatic condition of Iran. *Iranian Journal of Medicinal and Aromatic Plants Research* 22(4): 386-396. (In Farsi). [https://ijmapr.areeo.ac.ir/article\\_101766.html?lang=en](https://ijmapr.areeo.ac.ir/article_101766.html?lang=en)
- Das M. 2014. Chamomile: Medicinal, biochemical, and agricultural aspects. CRC Press. New York.
- Dehghani Mashkani M.R., Naghdi Badi H., Darzi M.T., Mehrafarin A., Rezazadeh S., Kadkhoda Z. 2011. The effect of biological and chemical fertilizers on quantitative and qualitative yield of Shirazian Baboonch (*Matricaria recutita* L.). *Journal of Medicinal Plants* 10(38): 35-48. (In Farsi). <http://jmp.ir/article-1-209-en.html>
- Ebadi M.T., Azizi M., Omidbaigi R., Hassanzadeh Khayat M. 2009. The effect of sowing date and seeding levels on quantitative and qualitative yield of chamomile (*Matricaria recutita* L.). *Iranian Journal of Medicinal and Aromatic Plants Research* 25(3): 296-308. <https://doi.org/10.1055/s-0031-1282324>
- El abdali Y., Mikou K., Chaouch M., Eloutassi N. 2018. Effect of different drying modes on flavonoid content and antioxidant activity of *Matricaria chamomilla*. *Journal of Applied Science and Environmental Studies* 1(1): 1-7. <https://doi.org/10.48393/IMIST.PRSM/jases-v1i1.15778>
- Ghanavati M., Houshmand S.L., Zainali H., Abrahimpour F. 2010. Chemical composition of the essential oils of *Matricaria recutita* L. belonging to central and south parts of Iran. *Journal of Medicinal Plants* 9(34): 102-108. (In Farsi). <http://dorl.net/dor/20.1001.1.2717204.2010.9.34.10.8>
- Golzadeh H., Mehrafarin A., Naghdi Badi H., Fazeli F., Qaderi A., Zarinpanjeh N. 2012. Effect of bio-stimulators compounds on quantitative and qualitative yield of German chamomile (*Matricaria recutita* L.). *Journal of Medicinal Plants* 11(41): 195-207. (In Farsi). <http://dorl.net/dor/20.1001.1.2717204.2012.11.41.28.4>
- Haghi G., Hatami A., Safaei A., Mehran M. 2014. Analysis of phenolic compounds in *Matricaria chamomilla* and its extracts by UPLC-UV. *Research in Pharmaceutical Sciences* 9(1): 31-37.
- Homami S.S., Jaimand K., Rezaee M.B., Afzalzadeh R. 2016. Comparative studies of different extraction methods of essential oil from *Matricaria recutita* L. in Iran. *Journal of the Chilean Chemical Society* 61(2): 2982-2984. <https://doi.org/10.4067/S0717-97072016000200026>
- Jalali Z., Sefidkon F., Assareh M.H., Attar F. 2008. Comparison of sesquiterpens in the essential oils of *Anthemis hyalina* DC, *Matricaria recutita* L. and *Matricaria aurea* (Loefl.) Schultz-Bip. *Iranian Journal of Medicinal and Aromatic Plants Research* 24(1): 31-37. (In Farsi). [https://ijmapr.areeo.ac.ir/article\\_10018\\_en.html](https://ijmapr.areeo.ac.ir/article_10018_en.html)
- Lawrence B.M. 1994. Progress in essential oils. *Perfumer and Flavorist* 19(6): 57-62
- Mežaka I., Kronberga A., Nakurte I., Taškova I., Jakovels D., Primavera A. 2020. Genetic, chemical and morphological variability of chamomile (*Chamomilla recutita* L.) populations of Latvia. *Industrial Crops and Products* 154: 112614. <https://doi.org/10.1016/j.indcrop.2020.112614>
- Nurzyńska-Wierdak R. 2011. The essential oil of *Chamomilla recutita* (L.) Rausch. cultivated and wild growing in Poland. *Annales Universitatis Mariae Curie-Skłodowska* 24: 199-206
- Orav A., Raal A., Arak E. 2010. Content and composition of the essential oil of *Chamomilla recutita* (L.) Rauschert from some European countries. *Natural Product Research* 24(1): 48-55. <https://doi.org/10.1080/14786410802560690>
- Oravec S.V., Oravec J.V., Oravec V. 2006. Breeding of bisabolol diploid and tetraploid varieties of chamomile in Slovakia. In *I International Symposium on Chamomile Research, Development and Production* 749: 115-120. <https://doi.org/10.1021/bk-2016-1218.ch013>
- Otto L.G., Mondal P., Brassac J., Preiss S., Degenhardt J., He S., Sharbel T.F. 2017. Use of genotyping-by-sequencing to determine the genetic structure in the medicinal plant chamomile, and to identify flowering time and alpha-bisabolol associated SNP-loci by genome-wide association mapping. *BMC Genomics* 18: 1-18. <https://doi.org/10.1186/s12864-017-3991-0>
- Raal A., Arak E., Ivask K. 2003. Comparación de aceites esenciales de *Matricaria recutita* L. de origen diverso. *Ars Pharmaceutica* 44(2): 159-165. (In Spanish). <http://hdl.handle.net/10481/28213>
- Saebi A., Minaei S., Mahdavian A., Ebadi M. 2024. Development and evaluation of a prototype precision harvesting unit for medicinal plants – Tested on *Hyssopus officinalis* L. *International Journal of Horticultural Science and Technology* 11(3): 369-380. <https://doi.org/10.22059/ijhst.2023.358533.636>
- Šalamon I. 2004. The Slovak gene pool of German chamomile (*Matricaria recutita* L.) and comparison in its parameters. *Horticultural Science* 31(2): 70. <https://doi.org/10.17221/3795-HORTSCI>



- Šalamon I., Ghanavati M., Abrahimpour F. 2010. Potential of medicinal plant production in Iran and variability of chamomile (*Matricaria recutita* L.) essential oil quality. *Journal of Essential Oil Bearing Plants* 13(5): 638-643. <https://doi.org/10.1080/0972060X.2010.10643874>
- Schilcher H. 1973. Neuere Erkenntnisse bei der Qualitätsbeurteilung von Kamillenblüten bzw. Kamillenöl. 2. Qualitative Beurteilung des ätherischen Öles in flores chamomillae. Aufteilung der Handelskamillen in vier bzw. fünf chemische Typen. *Planta Medica* 23(2): 132-144. (In German). <https://doi.org/10.1055/s-0028-1099424>
- Seidler-Lozykowska K. 2006. Chamomile cultivars and their cultivation in Poland. In I International Symposium on Chamomile Research, Development and Production 749: 111-114. <https://doi.org/10.17660/ActaHortic.2007.749.10>
- Shraim A.M., Ahmed T.A., Rahman M.M., Hijji Y.M. 2021. Determination of total flavonoid content by aluminum chloride assay: A critical evaluation. *LWT* 150: 111932. <https://doi.org/10.1016/j.lwt.2021.111932>
- Singh O., Khanam Z., Misra N., Srivastava M.K. 2011. Chamomile (*Matricaria chamomilla* L.): An overview. *Pharmacognosy Reviews* 5(9): 82. <https://doi.org/10.4103/0973-7847.79103>
- Tsivelika N., Irakli M., Mavromatis A., Chatzopoulou P., Karioti A. 2021. Phenolic profile by HPLC-PDA-MS of Greek chamomile populations and commercial varieties and their antioxidant activity. *Foods* 10(10): 2345. <https://doi.org/10.3390/foods10102345>
- Tsivelika N., Sarrou E., Gusheva K., Pankou C., Koutsos T., Chatzopoulou P., Mavromatis A. 2018. Phenotypic variation of wild Chamomile (*Matricaria chamomilla* L.) populations and their evaluation for medicinally important essential oil. *Biochemical Systematics and Ecology* 80: 21-28. <https://doi.org/10.1016/j.bse.2018.06.001>
- Wesolowska A., Grzeszczuk M., Kulpa D. 2015. Propagation method and distillation apparatus type affect essential oil from different parts of *Matricaria recutita* L. plants. *Journal of Essential Oil Bearing Plants* 18(1): 179-194. <https://doi.org/10.1080/0972060X.2014.895210>

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