

Variability of Essential Oil Content and Composition of Different Iranian Fennel (*Foeniculum vulgare* Mill.) Accessions in Relation to Some Morphological and Climatic Factors

M. Rahimmalek^{1*}, H. Maghsoudi¹, M. R. Sabzalian¹, and A. Ghasemi Pirbalouti²

ABSTRACT

Fennel (*Foeniculum vulgare* Mill.) is an industrial medicinal plant with different pharmaceutical and food applications. In this study, the leaf essential oil composition of 12 Iranian accessions of fennel collected from different geographical regions was assessed. The essential oil yield of fennel leaves ranged from 0.65% (Varamin accession) to 2.03% (Tabriz accession). *Trans*-anethole, fenchone and limonene were highly abundant in all of the examined oils. *Trans*-anethole ranged from 41.19% in Shiravan to 56.6% in Shiraz accessions and had negative correlation with most of the constituents. According to the major compounds, two chemotypes were defined in which group 1 was considered as the high *trans*-anethole (> 50%) and group 2 was a high limonene group. The correlation of essential oil yield and *trans*-anethol with climatic conditions and some morphological characters were also assessed. Higher temperatures and essential oil yield had negative Pearson correlation ($r = -0.371$), while *trans*-anethol and high temperature showed positive correlation ($r = 0.459$) in fennel. Furthermore, the studied accessions had different flowering time and height. The early flowering and dwarf accessions had higher essential oil yield, while the late flowering ones had higher *trans*-anethol in their leaves.

Keywords: Climatic condition, Fenchone, Limonene, Temperature, *Trans*-anethole.

INTRODUCTION

Fennel (*Foeniculum vulgare* Mill.) is one of the most widespread medicinal plants of the Apiaceae family. It is a perennial herb that grows in many parts of the world (Raal *et al.*, 2011). The leaves and seeds of fennel are used in many culinary traditions (Ehsanipour *et al.*, 2012). Mature fennel fruits and essential oil are used as flavoring agents in food products such as liqueurs, bread, pickles, pastries, and cheese (Zoubiri *et al.*, 2014). They are also used as constituent in cosmetic and pharmaceutical products

(Telci *et al.*, 2009). Fennel stimulates appetite and aids digestion. It is also used in kidney stones, menopausal problems, nausea and obesity (Zahid *et al.*, 2009; Ehsanipour *et al.*, 2012).

There are high morphological and photochemical variations among and within wild and cultivated fennels (Shahat *et al.*, 2012). Different fennel populations have different fruit size, odor, taste, quality, and yield potential. There are several reports showing chemical composition of fennel (Cosge *et al.*, 2008; Chowdhury *et al.*, 2009; Zoubiri *et al.*, 2014; Raal *et al.*, 2011). However, plants

¹ Department of Agronomy and Plant Breeding, College of Agriculture, Isfahan University of Technology, Isfahan 84156 83111, Islamic Republic of Iran.

* Corresponding author, email: mrahimmalek@cc.iut.ac.ir

² Department of Medicinal Plants, Researches Center of Medicinal Plants and Ethno-veterinary, Shahrekord Branch, Islamic Azad University, Shahrekord, Islamic Republic of Iran.



essential oil yield and their components are highly affected by genetic and environmental factors as well as sampling (Ozcan and Chalchat, 2006; Rahimmalek et al., 2009). To run a breeding program addressed to improve essential oil, it is important to assess the amount of essential oil in natural fennel populations.

There are many reports about essential oil variation in Iranian medicinal plants (Askari et al., 2009; Rahimmalek et al., 2009; Ayoughi et al. 2011; Goudarzi et al., 2011). Researchers have not covered the presence of high essential oil variation in fennel plants collected from various parts of different countries. Raal et al. (2011) assessed the essential oil variations in commercial fennel fruits obtained from retail pharmacies in Estonia, Norway, Austria, and Moldova and from a spice shop in Turkey. Shahat et al. (2012) compared the chemical composition of wild and cultivated fennel populations in Egypt. Chowdhury et al. (2009) studied the essential oil composition of seeds and leaves of cultivated fennels in Bangladesh. Chemical composition and larvicidal activity of Algerian fennel seed essential oil was also assessed by Zoubiri et al. (2014). Cosge et al. (2008) reported the composition of essential oil in 20 sweet fennel lines that originated from Turkey. In Iran, most of the researches on the subject were conducted using a limited geographical area (Bamoniri et al., 2009; Ehsanipour et al., 2012) and some studies were focused on essential oil changes during different plant developmental stages (Yamini et al., 2002; Saharkhiz and Tarakezme, 2011). However, there are no comprehensive researches in assessing essential oil variation among and within fennel populations collected from various Iranian regions.

Essential oil yield and their components are highly affected by genetic and climatic factors (Rahimmalek et al., 2009). Thus, study of chemical composition of essential oil in relation to environmental factors might provide information on what

determines chemical polymorphism. In fennel oil, the major constituent is *trans*-anethole that might be variable in different stages of development (Gross et al., 2002; Koeduka et al., 2009). In Iran, there are no comparative reports regarding essential oil variation in leaves. Furthermore, study of *trans*-anethole variation and introduction of high *trans*-anethole chemotype or high limonene one can be valuable for industrial purposes.

The aims of this study were: (1) to categorize the populations according to essential oil yield and composition; (2) to regroup the chemotypes of Iranian fennels based on their major compounds in leaves, and (3) to assess the relationships between variations of essential oil yield and composition with some environmental factors and morphological characters of populations.

MATERIALS AND METHODS

Plant Materials

Seeds of 12 fennel accessions were collected from different geographical regions of Iran, and were sown in similar condition in Randomized Complete Block Design (RCBD) with three replicates (Table 1). Geographic coordinates, elevation, and mean air temperatures of those regions were determined using the data of the nearest meteorological stations (Table 1).

Essential Oil Isolation

Young leaves of fennel accessions were collected and dried in shade. For each hydro-distillation turn, 50-60 g of samples was used. The round-bottom flask of Clevenger-type apparatus was used to extract essential oils. Four hundred ml distilled water was added and boiled for 5 hours. Then, the essential oil was collected in a container. The essential oil content was

Table 1. Collection site information, essential oil yield, plant height, and flowering time of fennel populations analyzed in the present work.^a

Site no.	Collection site	City, Province	Latitude	Longitude	Elevation (masl)	T _{max} (°C)	T _{min} (°C)	T _{average} (°C)	Oil yield%	Plant height (cm)	Flowering time
1	Shiraz, Fars		29 39 N	52 35 E	1486	27.8	10.7	19.2	0.91 ^{bc} ±0.07	173.35 ^a ±6.0	Late
2	Hamedan, Hamedan		36 46 N	48 34 E	1900	20.8	3.6	12.9	0.86 ^{bc} ±0.17	175.00 ^a ±2.9	Very early
3	Kerman, Kerman		30 3 N	57 1 E	1800	26.8	7.5	17.1	0.85 ^{bc} ±0.08	133.00 ^c ±2.9	Late
4	Isfahan, Isfahan		32 65 N	51 67 E	1570	23.4	7.8	18	0.65 ^c ±0.13	115.00 ^{bc} ±4.4	Early
5	Bushehr, Bushehr		28 92 N	50 83 E	5	32.2	21.1	26.7	0.84 ^{bc} ±0.07	134.00 ^c ±7.2	Late
6	Paveh, Kermanshah		35 03 N	46 27 E	1530	21.3	6.3	15.5	1.00 ^{bc} ±0.10	86.00 ^b ±2.9	Very early
7	Tabriz, Azarbaijan Sharghi		38 05 N	46 28 E	1366	19.6	7.5	13.5	2.03 ^a ±0.05	75.00 ^a ±2.9	Very early
8	Mashhad, Khorasan Razavi		36 19 N	59 37 E	979	22.9	7.7	15.3	0.83 ^{bc} ±0.04	155.00 ^b ±7.2	Late
9	Tehran, Tehran		35 7 N	51 4 E	1190	22.1	11.4	16.8	0.93 ^{bc} ±0.08	131.65 ^c ±7.3	Medium
10	Kashan, Isfahan		33 96 N	51 15 E	982	26.4	9.1	20	1.14 ^b ±0.10	123.34 ^{cd} ±2.9	Medium
11	Shiravan, Khorasan Shomali		37 27 N	57 54 E	1160	20.9	13.9	16.9	1.05 ^{bc} ±0.09	151.65 ^b ±2.8	Medium
12	Varamin, Tehran		35 19 N	51 39 E	918	21.6	11.3	14.2	0.81 ^{bc} ±0.04	100.00 ^d ±2.9	Very early

^a The superscripts are the mean comparison based on LSD analysis. T_{max}: Maximum temperature (°C); T_{min}: Minimum temperature (°C); T_{average}: Average daily temperature.

determined on the basis of dry matter in three replicates.

GC/MS Analysis

The model of GC used in this study was Hewlett-Packard 6890. GC/MS analysis was carried out on a gas chromatography fitted with a fused silica HP-5MS capillary column (30 m×0.25 mm×0.25 μm). The oven temperature was programmed from 60°C to 280°C at 4 °C min⁻¹. Helium was used as carrier gas at a flow rate of 2 ml/min. The chromatography was coupled to a Hewlett-Packard 6890 mass selective detector. The MS operating parameters were ionization voltage, 70eV; ion source temperature, 200°C.

Identification of Components

The constituents of the volatile oils were identified by comparing their retention indices to n-alkanes (C9-C18) and their mass spectral fragmentation pattern with those reported in the literature (Adams, 2001), and stored in the MS library (Wiley, 275).

Statistical Analysis

Cluster analysis was performed using Ward's minimum variance by SPSS ver. 11 software. Calculation of correlations among the major compounds was also performed using SPSS ver11. The morphological analysis was done by SAS ver. 8 using one way ANOVA analysis option.

RESULTS AND DISCUSSION

Essential Oil Yield

The yield of essential oil of fennel leaves (mean of three replicates) ranged from 0.65% (Varamin accession) to 2.03%



(Tabriz accession) (Table 1). Most of the previous studies were focused on essential oil of fennel seeds; however there are few reports on the essential oil yield of leaves. Saharkhiz and Tarakezme (2011) studied the variation of essential oil yield of one Iranian fennel accession (Shiraz) during three phenological stages; reporting that essential oil content of fruits ranged from 1.31 to 1.26%, while in the present research, Shiraz accession showed an essential oil yield of leaves of only 0.91%. Ehsanipour *et al.* (2012) reported the essential oil yield of four Iranian fennel leaves which ranged from 1.2 to 1.64%, similar to the present results. Figueredo *et al.* (2012) reported that essential oil yield and composition of Turkish fennels is affected by year of harvest, with an oil yield ranging from 1.3 to 3.09% in different years.

Essential Oil Composition

Essential oil components of the twelve fennel accessions were determined. All compounds, except the components with trace amounts, are shown in Table 2. Results showed the presence of high chemical polymorphism among Iranian fennel accessions. The main constituents were *trans*-anethole with a concentration ranging from 41.19 (Shiravan) to 56.61% (Shiraz), the *cis*-anethole ranged from 0.21% (Kashan) to 4.18% (Varamin), fenchone ranged from 1.7 (Shiravan) to 10.23% (Kashan), limonene ranged from 11.5 (Mashhad) to 31.7% (Paveh), α -Phellandrene ranged from 0 (Isfahan) to 13.34% (Kashan), α -pinene ranged from 0.61 (Shiraz) to 16.89% (Shiravan), and β -ocymene (Z) ranged from 1.24 (Mashhad) to 5.9% (Varamin).

Anethole was the most abundant compound in fennel accessions. Shahat *et al.* (2012) assessed the essential oil variation of the aerial parts of cultivated and wild fennel (*Foeniculum vulgare* Mill) in Egypt. In their studies, cultivated plants showed higher percentages of pinenes, fenchone, estragol,

myrcene, and camphene, while the wild ones showed much higher level of limonene. Roby *et al.* (2012) also compared the essential oil and phenolic compounds of fennel and chamomile in Egypt. They reported 56.4% *trans*-anethole, 8.26% fenchone and 4.2% limonene in Egyptian sample. Napoli *et al.* (2010) compared the seed essential oil of Italian fennels reporting a *cis*-anethole concentration ranging from 0.1 to 0.36%; while in our study this compound varied from 0.21 to 4.18%. Raal *et al.* (2010) compared the essential oil compounds of fennel fruits collected from pharmacies in different countries. The main compound was *trans*-anethole ranging from 34.8 to 82%, while in Iranian fennel leaves, it ranged from 41.19 to 56.61% and Algerian fennels possessed more than 72% *trans*-anethole in their seeds (Zoubiri *et al.*, 2010).

Fennel Chemotypes According to Major Compounds

Cluster analysis was done to distinguish possible groups among the accessions. The dendrogram was produced based on major components including *trans*-anethole, limonene, α -Pinene, *cis*-anethole, fenchone and β -Ocimene Z. Figure 1 presents the corresponding dendrogram using Ward method. The cluster analysis allows subdividing the 12 accessions in two major groups, one of which divided into two sub-groups. The classification was highly affected by *trans*-anethole as the major compound. Group 1a included Bushehr, Tabriz, Shiraz, and Kerman accessions. The major compounds of this group were *trans*-anethole (50.31-56.6%), limonene (20.61-22.74%), and fenchone (2.45-6.75%). Group 1b consisted of Hamedan, Isfahan, Varamin and Paveh. The major constituents were *trans*-anethole (44.29-48.79%), limonene (23.06-31.7%), and fenchone (2.87-5.03%). Finally, Tehran, Mashhad, Kashan, and Shiravan were classified in group 2 and *trans*-anethole (41.19-54.83%), limonene (11.5-16.01%), and fenchone (1.7-10.23%)

Table 2. Chemical composition of essential oils of 12 fennel accessions collected from different geographical regions of Iran.^a

Compounds	RI ^b	Shiraz	Hamadan	Kerman	Isfahan	Bushehr	Paveh	Tabriz	Mashhad	Tehran	Kashan	Shiravan	Varamin
α -Pinene	937	0.61±0.02	6.82±0.1	0.51±0.01	3.97±0.02	7.31±0.15	0.69±0.05	6.08±0.11	8.48±0.14	5.28±0.12	1.25±0.06	16.89±0.2	1.66±0.07
Camphene	951	0.13	0.17	0.15	0.14	0.11	0.11	0.16	0.15	0.14	0.24	0.15	0.1
Sabinene	974		0.15		0.08	0.14		0.12	0.2	0.16	0.1	0.46	0.13
β -Pinene	979	0.05	0.77		0.47	0.89	0.06	0.71	1.2	0.67	0.1	2.52	
β -Myrcene	993	0.62	1.03	0.64	0.98	1.06	0.7	1.04	1.11	1.04	1.44	2.24	0.66
α -Phellandrene	1007	0.07	0.61		0.61	3.83	1.26	1.28	1.7	8.4	13.34	9.35	0.71
p-Cymene	1025		3.41	0.24		1.32		1.38	3.62	1.49	1.6	1.61	1.18
Limonene	1032	22.74±0.3	26.61±0.25	20.61±0.22	25.74±0.27	21.5±0.22	31.7±0.4	21.2±0.15	11.5±0.17	13.62±0.13	15.04±0.18	16.01±0.17	23.06±0.23
β -Ocimene Z	1038	3.16±0.03	1.93±0.02	1.64±0.01	2.96±0.02	2.03±0.01	2.42±0.03	1.75±0.01	1.24±0.01	2.43±0.03	2.85±0.04	2.13±0.03	5.9±0.04
β -Ocimene E	1046	0.17	0.08	0.07	0.21	0.13	0.13	0.09		0.15	0.17	0.14	0.12
γ -Terpinene	1057	0.06			0.07	0.06				0.11	0.08	0.08	
Fenchone	1089	4.52±0.04	4.33±0.03	6.75±0.05	4.84±0.03	2.45±0.02	2.87±0.03	5.55±0.03	5.92±0.04	4.95±0.03	10.23±0.05	1.7±0.02	5.03±0.04
α -Fenchyl acetate	1216	1.51	1.38	2.01	2.4	1.11	1.63	1.04	0.56	1.01	2.1	0.21	1.66
β -Fenchyl acetate	1230	3	1.45	3.02	2.72	1.55	1.11	1.74	1.16	1.23	0.97	0.74	4.26
Cis-Anethole	1251	0.77±0.01	2.79±0.02	2.41±0.02	0.47±0.01	0.64±0.01	2.32±0.02	1.42±0.02	2.54±0.03	0.33±0.01	0.21±0.01	0.4±0.01	4.18±0.03
Trans-anethole	1285	56.61±1.5	44.29±0.59	54.32±0.77	48.79±1.6	51.21±3.2	46.29±0.37	50.31±0.12	54.83±1.3	53.58±0.48	46.51±0.77	41.19±1.6	46.05±0.63
α -Copaene	1369			0.09	0.08	0.08	0.12	0.11	0.51	0.11		0.13	
β -Farnesene	1451	0.11		0.13	0.13	0.09	0.2	0.27		0.1		0.12	
Germacrene-D	1474	0.01	0.15	0.17	0.31	0.34	0.36	0.41	0.11	0.42	0.25	0.71	0.17
Total		94.14	95.97	92.76	94.97	95.85	91.97	94.66	94.83	95.22	96.48	96.78	94.87

^a Values are the percent of constituents in total oil of 12 accessions, ^b The data were sorted based on the retention index (RI) of components.

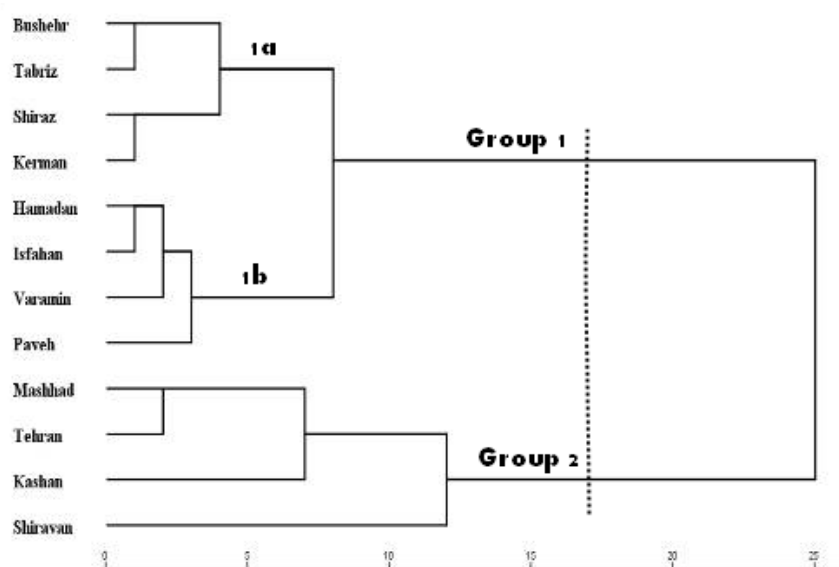


Figure 1. Grouping of 12 fennel accessions according to their major essential oil composition using Ward's minimum variance.

were the major compounds of this group. Thus, group 1a was considered as the high *trans*-anethole (> 50%), while group 1b was the high limonene group (Figure 1).

Correlations analysis (Table 3) showed that the highest correlation coefficient was between β -myrcene and α -pinene ($r=+0.834$). Other significant positive correlations were between camphene and fenchone ($r= 0.736$), β -myrcene and α -phellandrene ($r= 0.718$) and germacrene-D and β -myrcene ($r= 0.73$). The highest negative correlations were between α -fenchyl acetate and α -pinene ($r= -0.811$) and limonene and *cis*-anetole ($r= -0.601$). *Trans*-anethole had negative correlations with most of the constituents. For instance, the correlation with limonene was $r= -0.272$, while in some cases, positive correlations were also observed such as *trans*-anethole with fenchone ($r= 0.24$; Table 3).

***Trans*-anethol Changes in Fennel**

The composition of medicinal plant can highly be affected by their secretory tissue condition and developmental stage (Gross *et al.*, 2002). *Trans*-anethole is derived from

the general phenylpropanoid pathway, through which lignin, flavonoids and other phenylalanine-derived compounds are produced (Gross *et al.*, 2002). *Trans*-anethole could be highly affected by the stage of development. In vegetative phase, young leaves contain the highest levels of *trans*-anethole and also have the highest chavicol and t-anol/iso Eugenol O-methyltransferase activity (the enzyme related in *trans*-anethole synthesis) levels. Production of *trans*-anethole begins early during leaf development. Koeduka *et al.* (2009) showed the biosynthesis of *trans*-anethole in Anise (*Pimpinella anisum*), a plant from Apiaceae family. In their report, the highest level of *trans*-anethole was found in developing fruits (seeds and pods), followed closely by flowers, while *trans*-anethole in young levels were much lower and undetected in roots. In present study, young leaves were used for essential oil extraction. The reduced levels of O-methyl transferases activity could be partially due to a diminished density of the oil accumulating structures in older leaves (Gross *et al.*, 2002). Previous researches showed that many plants synthesize and accumulate high levels of volatiles, including

Table 3. Pearson Correlation of major components belonging to different fennel populations.

Compounds	α -pinene	Camphene	β -myrcene	α -phellandrene	limonene	β -ocimene <i>cis</i>	Cis- fenchyl acetate	Fenchyl acetate <i>trans</i>	Cis - anethole	Trans - anethole	Germacrene-D
α -pinene	1										
Camphene	0.026	1									
β -myrcene	0.834**	0.427	1								
α -phellandrene	0.317	0.598*	0.718**	1							
limonene	-0.405	-0.420	-0.495	-0.597*	1						
β -ocimene <i>cis</i>	-0.375	-0.312	-0.274	-0.060	0.239	1					
fenchone	-0.507	0.736**	-0.148	0.284	-0.371	0.050	1				
Cis- fenchyl acetate	-0.811**	0.153	-0.553	-0.204	0.483	0.364	0.498	1			
Fenchyl acetate <i>trans</i>	-0.532	-0.436	-0.639*	-0.591*	0.320	0.688*	0.094	0.505	1		
Cis -anethole	-0.254	-0.360	-0.490	-0.601*	0.322	0.340	-0.011	0.064	0.477	1	
Trans -anethole	-0.399	-0.161	-0.561	-0.327	-0.272	-0.233	0.240	0.072	0.253	-0.096	1
Germacrene-D	0.662*	-0.003	0.729**	0.518	-0.142	-0.192	-0.437	-0.449	-0.519	-0.450	-0.545

phenylpropanes, in their vegetative tissues for defense (Koeduka *et al.*, 2009). These compounds mostly accumulate in trichomes localized in leaf surface. This kind of accumulation is mostly reported in Apiaceae plants such as anise (Koeduka *et al.*, 2009).

Another probable reason that might increase *trans*-anethole in fennel is the increase in the oil duct area, as observed in transversal cross sections in early stages of fennel development. Upon maturation, oleoresin is further accumulated due to increased duct volume as a result of duct elongation. Higher fruit development can lead to the lack of further metabolism and minimal volatilization, as indicated by apparent high lignification of the cells lining the oil ducts (Gross *et al.*, 2006). This phenomenon can also be observed in the leaves. Furthermore, other modifications such as suberification or serification in leaves epidermis can affect the trichome frequency and oil ducts as the secretory tissues.

Correlation of Environmental Factors, Morphological Characters, Essential Oil Yield, and Composition

The correlation of essential oil yield and *trans*-anethole content with climatic data, plant height, and flowering date were calculated. Negative correlation was obtained between essential oil yield and T_{max} ($r = -0.371$; Figure 2). It suggests that increase in temperature lead to decrease of essential oil yield in most cases. In the present study, the highest essential oil yield (2.03%) was obtained in Tabriz accession. The collection site of this sample had the lowest temperature compared with the others. On the other hand, a positive correlation was observed between *trans*-anethole and T_{max} ($r = 0.459$; Figure 3). Thus, it might be suggested that cultivation of fennel in higher temperatures can increase *trans*-anethole content. Essential oil yield had negative significant correlation with plant height ($r = -0.499$) (Figure 4) and flowering date ($r = -0.33$). The dwarf fennel accessions had a compact leaf canopy with higher leaf essential oil yield. *Trans*-anethol

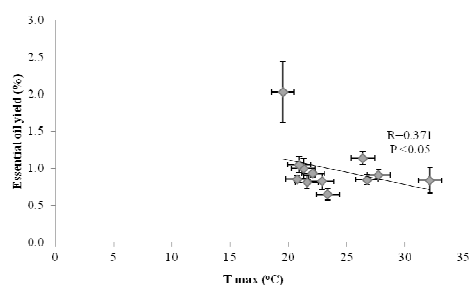


Figure 2. The effect of higher temperatures on essential oil yield of fennel.

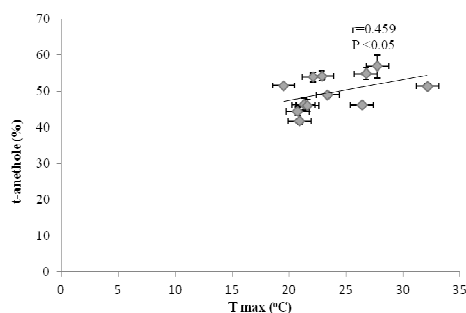


Figure 3. Increase in *t*-anethol with higher temperatures.

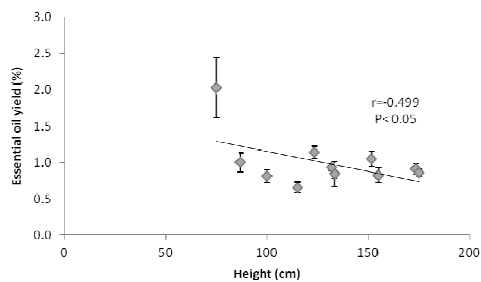


Figure 4. The effect of plant height on essential oil yield of fennel.

content and flowering date had a significant positive correlation ($r = +0.67$). This might be due to longer length of vegetative phase for these genotypes. Thus, the late flowering accessions had enough time to accumulate more *trans*-anethol in their leaves in comparison with the early flowering ones.

CONCLUSIONS

In conclusion, the Iranian fennel accessions have been cultivated and

domesticated in different climatic regions of the country with different phenological development. These accessions have been acclimatized to cultivated area during a period of time and they have gained different characteristics. Among the accessions, Tabriz had the highest essential oil yield and acceptable *trans*-anethole content and it can be introduced as a good candidate for cultivation. Another candidate was from Shiraz, which had the highest *trans*-anethole content with relatively high essential oil yield. In this study, the accessions had different flowering time and height and the early flowering and dwarf accessions had higher essential oil yield, while the late flowering ones had higher *trans*-anethol in their leaves. Finally, it can be concluded that the best harvesting time for fennel leaves is the early stage and the higher temperature might be beneficial in increasing *trans*-anethol, but it could decrease the essential oil yield. Anatomical studies on secretory tissues of different accessions and considering other climatic factors might lead to more insightful results.

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تنوع عملکرد و ترکیبات اسانس نمونه های مختلف جمعیتی رازیانه (*Foeniculum vulgare* Mill.) در ایران در ارتباط با برخی خصوصیات مورفولوژیک و اقلیمی

م. رحیم ملک، ح. مقصودی، م. ر. سبز علیان، ع. قاسمی پیر بلوطی

چکیده

رازیانه یکی از گیاهان مهم صنعتی و دارویی است. در مطالعه حاضر، ترکیبات اسانس برگ ۱۲ نمونه جمعیتی رازیانه متعلق به نواحی مختلف جغرافیایی ایران مورد بررسی قرار گرفت. عملکرد اسانس بین ۰/۶۵٪ (نمونه ورامین) تا ۲/۰۳٪ در نمونه تبریز متغیر بود. ترانس آنتول، فنچون و لیمون ترکیبات اصلی اسانس بودند. ترانس آنتول از ۴۱/۱۹٪ در نمونه شیروان تا ۵۶/۶٪ در نمونه شیراز متغیر بود و با اغلب ترکیبات اسانس همبستگی منفی داشت. بر اساس ترکیبات اصلی سه تیپ شیمیایی در مشخص شد. گروه اول گروه با آنتول بالا (بیشتر از ۵۰٪) و گروه دوم دارای لیمون زیاد بود. در این مطالعه همبستگی عملکرد اسانس، ترانس آنتول، برخی خصوصیات مورفولوژیک و اقلیمی نیز بررسی شد. دمای بالا و عملکرد اسانس همبستگی منفی (۰.۳۷۱-) در حالی که ترانس آنتول و دمای بالا همبستگی مثبت نشان دادند. علاوه بر این نمونه های مورد مطالعه دارای زمان گلدهی و ارتفاع متفاوت بودند. نمونه های زود گلده و کوتاه دارای عملکرد اسانس بالاتر در حالی که نمونه های دیر گلده دارای ترانس آنتول بالاتری بودند.